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20136, No. **Diochemical Pharmacology of Antiestrogen Action**

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I. Introduction

NONSTEROIDAL antiestrogens have been available as

clinically useful drugs for more than 20 years. One of the

first compounds to enter clinical trial, Clomid [MRL41, compo **first compounds 1. Introduction**
MONSTEROIDAL antiestrogens have been available as
clinically useful drugs for more than 20 years. One of the
first compounds to enter clinical trial, Clomid [MRL41,
a mixture of the E and 1. Introduction
NONSTEROIDAL antiestrogens have been available
clinically useful drugs for more than 20 years. One of t
first compounds to enter clinical trial, Clomid [MRL-
a mixture of the E and Z geometric isomers of 2 NONSTEROIDAL antiestrogens have been available achievation of the clinically useful drugs for more than 20 years. One of the first compounds to enter clinical trial, Clomid [MRL4 a mixture of the E and Z geometric isomers clinically useful drugs for more than 20 years. One of the first compounds to enter clinical trial, Clomid [MRL41, a mixture of the E and Z geometric isomers of 2 chloro-
1-(4 β diethyl aminoethoxyphenyl) 1,2 diphenylet first compounds to enter clinical trial, Clomid [MRL41,
a mixture of the E and Z geometric isomers of 2 chloro-
1-(4 β diethyl aminoethoxyphenyl) 1,2 diphenylethylene]
is successfully used, in short courses of 5 to 10 d a mixture of the E and Z geometric isomers of 2 chloro-

1- $(4\beta$ diethyl aminoethoxyphenyl) 1,2 diphenylethylene]

is successfully used, in short courses of 5 to 10 days, for

the induction of ovulation in subfertile wom 1- $(4\beta$ diethyl aminoethoxyphenyl) 1,2 diphenylethylene]
is successfully used, in short courses of 5 to 10 days, for
the induction of ovulation in subfertile women $(5, 116, 118, 140)$. It should be pointed out, though, is successfully used, in short courses of 5 to 10 days, for v
the induction of ovulation in subfertile women $(5, 116, 118, 140)$. It should be pointed out, though, that this application of antiestrogens is particularly i the induction of ovulation in subfertile women $(5, 116, 118, 140)$. It should be pointed out, though, that this geapplication of antiestrogens is particularly ironic because inthese drugs are potent antifertility agents 118, 140). It should be pointed out, though, that this gapplication of antiestrogens is particularly ironic because in these drugs are potent antifertility agents in laboratory animals and were originally planned to be in application of antiestrogens is particularly ironic because these drugs are potent antifertility agents in laboration
animals and were originally planned to be introduced postcoital contraceptives (83). Nolvadex [tamoxifen these drugs are potent antifertility agents in laboratory
animals and were originally planned to be introduced as
prostcoital contraceptives (83). Nolvadex [tamoxifen, ICI 15
46,474, Z-1(46 dimethylaminoethoxyphenyl) 1,2 d animals and were originally planned to be introduced as postcoital contraceptives (83). Nolvadex [tamoxifen, ICI 46,474, Z-1(4 β dimethylaminoethoxyphenyl) 1,2 diphen-
ylbut-1-ene citrate] is also used in some countries postcoital contraceptives (83). Nolvadex [tamoxifen, ICI 46,474, Z-1(4 β dimethylaminoethoxyphenyl) 1,2 diphen-
ylbut-1-ene citrate] is also used in some countries for the
induction of ovulation (107, 192, 323); however

I. Introduction In 1896, Beatson first demonstrated that some patients
NONSTEROIDAL antiestrogens have been available as with advanced breast cancer will respond to oophorec-
clinically useful drugs for more than 20 years. In 1896, Beatson first demonstrated that some patients In 1896, Beatson first demonstrated that some patie
with advanced breast cancer will respond to oophorec-
tomy (18). This original observation has been adequa In 1896, Beatson first demonstrated that some patients
with advanced breast cancer will respond to oophorec-
tomy (18). This original observation has been adequately
confirmed and it seems that about one-third of patients In 1896, Beatson first demonstrated that some patients
with advanced breast cancer will respond to oophorec-
tomy (18). This original observation has been adequately
confirmed and it seems that about one-third of patients
 In 1896, Beatson first demonstrated that some patient
with advanced breast cancer will respond to oophorec
tomy (18). This original observation has been adequately
confirmed and it seems that about one-third of patient
wit with advanced breast cancer will respond to oophorectomy (18). This original observation has been adequately confirmed and it seems that about one-third of patients with breast cancer will respond to this form of treatment tomy (18). This original observation has been adequately
confirmed and it seems that about one-third of patients
with breast cancer will respond to this form of treatment.
Similarly, about one-third of postmenopausal patie with breast cancer will respond to this form of treatment. will respond to adrenalectomy; however, in this case, it Similarly, about one-third
will respond to adrenalector
is the source of androgens th
gens are converted to estrog
ing enzyme systems (219).
The finding that a recept Il respond to adrenalectomy; however, in this case, it
the source of androgens that is being removed. Andro-
ms are converted to estrogens by peripheral aromatiz-
g enzyme systems (219).
The finding that a receptor molecul is the source of androgens that is being removed. Androgens are converted to estrogens by peripheral aromatizing enzyme systems (219).
The finding that a receptor molecule for estrogen is present in varying concentrations

gens are converted to estrogens by peripheral aromatizing enzyme systems (219).
The finding that a receptor molecule for estrogen is
present in varying concentrations in breast tumors (95,
151, 196, 203, 274) reinforced th ing enzyme systems (219) .
The finding that a receptor molecule for estrogen is
present in varying concentrations in breast tumors $(95, 151, 196, 203, 274)$ reinforced the hypothesis $(100, 145)$
that the determination The finding that a receptor molecule for estrogen
present in varying concentrations in breast tumors (9
151, 196, 203, 274) reinforced the hypothesis (100, 14
that the determination of estrogen binding might b
useful as a present in varying concentrations in breast tumors $(95, 151, 196, 203, 274)$ reinforced the hypothesis $(100, 145)$ that the determination of estrogen binding might be useful as a predictive test to preselect patients fo estrogen receptor assay to determine hormone responSIGM 30RDAN
siveness was evaluated. This world-wide study was care-
fully monitored and the clinical case studies were inde-Journalisty
Siveness was evaluated. This world-wide study was care
fully monitored and the clinical case studies were inde-
pendently assessed and correlated with hormone recepto JORI

siveness was evaluated. This world-wide study was care-

fully monitored and the clinical case studies were inde-

pendently assessed and correlated with hormone receptor

binding determinations. Approximately 60% of siveness was evaluated. This world-wide study was care-
fully monitored and the clinical case studies were inde-
pendently assessed and correlated with hormone receptor
binding determinations. Approximately 60% of patients siveness was evaluated. This world-wide study was care-
fully monitored and the clinical case studies were inde-
pendently assessed and correlated with hormone receptor
binding determinations. Approximately 60% of patients fully monitored and the clinical case studies were inde-
pendently assessed and correlated with hormone receptor
binding determinations. Approximately 60% of patients
with estrogen receptor positive (>10 femtomol/mg of
cyt pendently assessed and correlated with hormone receptor
binding determinations. Approximately 60% of patients and
with estrogen receptor positive (>10 femtomol/mg of 1
cytosol protein) breast tumors responded to endocrine
 binding determinations. Approximately 60% of patients
with estrogen receptor positive $(>10 \text{ femtomol/mg})$
cytosol protein) breast tumors responded to endocrine
therapy, whereas less than 10% of patients with estrogen
receptor with estrogen r
cytosol protein)
therapy, wherea
receptor negativ
tein) responded.
It is generally tosol protein) breast tumors responded to endocrinerapy, whereas less than 10% of patients with estroge ceptor negative tumors (<10 femtomol/mg cytosol prom) responded.
It is generally believed that estrogen can directly s

therapy, whereas less than 10% of patients with estrogen
receptor negative tumors ($\lt 10$ femtomol/mg cytosol pro-
tein) responded.
It is generally believed that estrogen can directly stim-
ulate the growth of breast receptor negative tumors (≤ 10 femtomol/mg cytosol pro-
tein) responded.
It is generally believed that estrogen can directly stim-
ulate the growth of breast cancer; therefore, therapy with
antiestrogens that block th tein) responded.
It is generally believed that estrogen can directly stim-
ulate the growth of breast cancer; therefore, therapy with
antiestrogens that block the binding of estradiol to the
estrogen receptor (ER) provides It is generally believed that estrogen can directly stim-
ulate the growth of breast cancer; therefore, therapy with
antiestrogens that block the binding of estradiol to the
estrogen receptor (ER) provides a logical medica ulate the growth of breast cancer; therefore, therapy we antiestrogens that block the binding of estradiol to estrogen receptor (ER) provides a logical medical proach to prevent estrogen action. This strategy ave the risks antiestrogens that block the binding of estradiol to the strogen receptor (ER) provides a logical medical approach to prevent estrogen action. This strategy avoid the risks of ablative surgery and, in the case of adrenal e trogen receptor (ER) provides a logical medical ap-
oach to prevent estrogen action. This strategy avoids
e risks of ablative surgery and, in the case of adrenal-
tomy, the long-term side effects of steroid replacement.
Th

moach to prevent estrogen action. This strategy avoids
the risks of ablative surgery and, in the case of adrenal-
ectomy, the long-term side effects of steroid replacement.
The nonsteroidal antiestrogens nafoxidine (25, 89 the risks of ablative surgery and, in the case of adrenal-
ectomy, the long-term side effects of steroid replacement.
The nonsteroidal antiestrogens nafoxidine (25, 89,
130), enclomiphene (126, 128), and tamoxifen (62, 314 ectomy, the long-term side effects of steroid replacement.

The nonsteroidal antiestrogens nafoxidine (25, 89,

130), enclomiphene (126, 128), and tamoxifen (62, 314)

were all tested in phase I and phase II clinical tria The nonsteroidal antiestrogens nafoxidine $(25, 89, 130)$, enclomiphene $(126, 128)$, and tamoxifen $(62, 314)$ were all tested in phase I and phase II clinical trials. The objective response rate of each of the antiestro The results of these early clinical trials for the treatment objective response rate of each of the antiestrogens was
similar $(\sim 30\%)$ but only tamoxifen has a low incidence
of side effects $(207, 232)$ and is at present the only
antiestrogen available for the treatment of breast similar $(\sim 30\%)$ but only tamoxifen has a low incidence
of side effects $(207, 232)$ and is at present the only
antiestrogen available for the treatment of breast cancer.
The results of these early clinical trials for th 232). tiestrogen available for the treatment of breast cancer.

he results of these early clinical trials for the treatment

advanced breast cancer have been reviewed (127, 207,

(1

2).

In recent years, attention has focused

The results of these early clinical trials for the treatmen
of advanced breast cancer have been reviewed (127, 207
232).
In recent years, attention has focused upon the use of
tamoxifen as an adjuvant therapy following mas of advanced breast cancer have been reviewed $(127, 207, 232)$.

The recent years, attention has focused upon the use of the drug is

tamoxifen as an adjuvant therapy following mastectomy.

The rationale for this applicat 232).
In recent years, attention has focused upon the use
tamoxifen as an adjuvant therapy following mastectom
The rationale for this application is that the drug
relatively nontoxic compared to cytotoxic chemotherap
and c In recent years, attention has focused upon the use of tamoxifen as an adjuvant therapy following mastectomy.
The rationale for this application is that the drug is
relatively nontoxic compared to cytotoxic chemotherapy,
a tamoxifen as an adjuvant therapy following mastectom
The rationale for this application is that the drug
relatively nontoxic compared to cytotoxic chemotherap
and continuous therapy with antiestrogens might pr
vent the rec The rationale for this application is that the drug is
relatively nontoxic compared to cytotoxic chemotherapy,
and continuous therapy with antiestrogens might pre-
vent the recurrence of hormone-sensitive disease. Labo-
ra relatively nontoxic compared to cytotoxic chemotherapy, on
and continuous therapy with antiestrogens might pre-
vent the recurrence of hormone-sensitive disease. Labo-
ratory studies with carcinogen-induced mammary tumor
m and continuous therapy with antiestrogens might prevent the recurrence of hormone-sensitive disease. Laboratory studies with carcinogen-induced mammary tumor models tend to support this application (156, 160). Two approach vent the recurrence of hormone-sensitive disease. Lal
ratory studies with carcinogen-induced mammary tun
models tend to support this application (156, 160). T
approaches to the clinical evaluation of antihormo
therapy are ratory studies with carcinogen-induced mammary tumor
models tend to support this application (156, 160). Two
approaches to the clinical evaluation of antihormone
therapy are being investigated: either the use of tamoxi-
fe models tend to support this application (156, 160).
approaches to the clinical evaluation of antihoritherapy are being investigated: either the use of tan
fen alone as adjuvant therapy (17, 245, 251, 264
tamoxifen in combi approaches to the clinical evaluation of antihormone
therapy are being investigated: either the use of tamoxi-
fen alone as adjuvant therapy (17, 245, 251, 264) or
tamoxifen in combination with a cytotoxic chemothera-
peut therapy are being investigated: either the use of tamoxi-
fen alone as adjuvant therapy (17, 245, 251, 264) or
tamoxifen in combination with a cytotoxic chemothera-
peutic regimen (98, 99, 138). A preliminary analysis of
t fen alone as adjuvant therapy $(17, 245, 251, 264)$ or
tamoxifen in combination with a cytotoxic chemothera-
peutic regimen $(98, 99, 138)$. A preliminary analysis of
the ongoing combination hormono/chemotherapy trials
is tamoxifen in combination with a cytotoxic chemothera-
peutic regimen (98, 99, 138). A preliminary analysis of
the ongoing combination hormono/chemotherapy trials
is encouraging (98, 99) with a reduction in the recurrence
r peutic regimen (98, 99, 138). A preliminary analysis of
the ongoing combination hormono/chemotherapy trials
is encouraging (98, 99) with a reduction in the recurrence
rate of postmenopausal patients who had tumors with
hig the ongoing combination hormono/chemotherapy trials
is encouraging (98, 99) with a reduction in the recurrence
rate of postmenopausal patients who had tumors with
high levels of estrogen receptor. However, to maintain
the rate of postmenopausal patients who had tumors with
high levels of estrogen receptor. However, to maintain
the gains achieved in the 1- or 2-year treatment trials
(17, 99), studies are underway to continue tamoxifen
thera the gains achieved in the 1- or 2-year treatment trials $(17, 99)$, studies are underway to continue tamoxifen therapy for up to 5 years (311) . Tamoxifen may be a tumoristatic rather than a tumoricidal agent $(242, 293)$ the gains achieved in the 1- or 2-year treatment trials (17, 99), studies are underway to continue tamoxifen therapy for up to 5 years (311). Tamoxifen may be a tumoristatic rather than a tumoricidal agent (242, 293) or s $(17, 99)$, studies are underway to continuatherapy for up to 5 years (311) . Tamoxife tumorstatic rather than a tumoricidal agen so that long treatment regimens should be prevent cells from being recruited to divide. Th erapy for up to 5 years (311). Tamoxifen may be a morstatic rather than a tumoricidal agent (242, 293) that long treatment regimens should be required to event cells from being recruited to divide. The ubiquitous use of t tumorstatic rather than a tumoricidal agent $(242, 293)$ mox
so that long treatment regimens should be required to
prevent cells from being recruited to divide.
The ubiquitous use of tamoxifen in breast cancer ther-
apy h

so that long treatment regimens should be required to prevent cells from being recruited to divide.
The ubiquitous use of tamoxifen in breast cancer ther-
apy has focused attention upon the development of new
antiestrogens prevent cells from being recruited to divide.
The ubiquitous use of tamoxifen in breast cancer the
apy has focused attention upon the development of n
antiestrogens, an area of research that is currently
tracting much inte The ubiquitous use of tamoxifen in breast cancer ther-
apy has focused attention upon the development of new
antiestrogens, an area of research that is currently at-
tracting much interest. Additionally, a better under-
s apy has focused attention upon the development of new
antiestrogens, an area of research that is currently at-
tracting much interest. Additionally, a better under-
standing of antiestrogen action will not only define the

AN
to identify further targets that might be vulnerable to
new drugs. AN
to identify if
new drugs.
There are

I
identify further targets that might be vulnerable to
w drugs.
There are several earlier reviews on the pharmacology
d antitumor activity of antiestrogens (127, 154, 161, to identify further targets that might be vulnerable to
new drugs.
There are several earlier reviews on the pharmacology
and antitumor activity of antiestrogens (127, 154, 161,
178, 294, 297) that the reader might consult the reader might be vulnerable to new drugs.
There are several earlier reviews on the pharmacology
and antitumor activity of antiestrogens (127, 154, 161,
178, 294, 297) that the reader might consult to obtain a
broader un There are several earlier reviews on the pharmacology
and antitumor activity of antiestrogens (127, 154, 161,
178, 294, 297) that the reader might consult to obtain a
broader understanding of this class of drugs. The aim o and antitumor activity of antiestrogens (127, 154, 178, 294, 297) that the reader might consult to obta
broader understanding of this class of drugs. The ai
this review is to present a synthesis of the recent reand ideas o 178, 294, 297) that the reader might consult to obtain a broader understanding of this class of drugs. The aim of this review is to present a synthesis of the recent results and ideas on the biochemical and molecular pharm broader understanding of this class of drugs. The aim of
this review is to present a synthesis of the recent results
and ideas on the biochemical and molecular pharmacol-
ogy of the nonsteroidal antiestrogens. Special cons this review is to present a synthesis of the recent rest and ideas on the biochemical and molecular pharmatogy of the nonsteroidal antiestrogens. Special consistion will be given to metabolism, receptor-media mechanisms of and ideas on the biochemical and molecular pharmacology of the nonsteroidal antiestrogens. Special consideration will be given to metabolism, receptor-mediated mechanisms of action, and structure-activity relationships. Ho ogy of the nonsteroidal antiestrogens. Special consideration will be given to metabolism, receptor-mediated mechanisms of action, and structure-activity relationships. However, it is important to review briefly the develop ation will be given to metabolism, receptor-mediated
mechanisms of action, and structure-activity relation-
ships. However, it is important to review briefly the
development and general pharmacology of antiestrogens
in ord mechanisms of action, and structure-activity is
hips. However, it is important to review bridevelopment and general pharmacology of anties
in order to demonstrate the complexities enco
with a simple definition such as "ant and general pharmacology
demonstrate the complexitive definition such as "antiest"
II. Historical Development
ring studies by Dodds and c

were all tested in phase I and phase II clinical trials. The
objective response rate of each of the antiestrogens was
similar $(\sim 30\%)$ but only tamoxifen has a low incidence
of side effects (207, 232) and is at present t II. Historical Development
The pioneering studies by Dodds and coworkers estabwith a simple definition such as "antiestrogen."

II. Historical Development

The pioneering studies by Dodds and coworkers es

lished the structure-activity relationships for nonste

dal estrogens (69, 70, 72). Diethylsti II. Historical Development
The pioneering studies by Dodds and coworkers estab-
lished the structure-activity relationships for nonsteroi-
dal estrogens (69, 70, 72). Diethylstilbestrol (DES) (fig-
ure 1) was the most pote II. HISTORICAL DEVELOPMENT

The pioneering studies by Dodds and coworkers estab-

lished the structure-activity relationships for nonsteroi-

dal estrogens (69, 70, 72). Diethylstilbestrol (DES) (fig-

ure 1) was the most The pioneering studies by Dodds and coworkers established the structure-activity relationships for nonsteroidal estrogens (69, 70, 72). Diethylstilbestrol (DES) (figure 1) was the most potent compound found (71, 73). Furth lished the structure-activity relationships for nonsteroi-
dal estrogens (69, 70, 72). Diethylstilbestrol (DES) (fig-
ure 1) was the most potent compound found (71, 73).
Further structure-activity studies demonstrated that dal estrogens (69, 70, 72). Diethylstilbestrol (DES) (figure 1) was the most potent compound found (71, 73)
Further structure-activity studies demonstrated thas
substituted triphenylethylenes are also estrogenic (256
but ure 1) was the most potent compound found
Further structure-activity studies demonstra
substituted triphenylethylenes are also estroge
but compared to DES these derivatives tend to
acting (258). $\alpha_i \alpha \text{Di}(p-\text{ethoxyphenyl})-\beta-\text{b$ Further structure-activity studies demonstrated that
substituted triphenylethylenes are also estrogenic (256)
but compared to DES these derivatives tend to be long-
acting (258). $\alpha, aDi(p-ethoxyphenyl)-\beta$ -bromoethylene
(DBE) and substituted triphenylethylenes are also estrogenic (256)
but compared to DES these derivatives tend to be long-
acting (258). α, α Di(*p*-ethoxyphenyl)- β -bromoethylene
(DBE) and trianisylchloroethylene (TACE) are ver but compared to DES these derivatives tend to be long-
acting (258). α, α Di(p-ethoxyphenyl)- β -bromoethylene
(DBE) and trianisylchloroethylene (TACE) are very
long-acting estrogens (257, 307, 308) (figure 1), an effe acting (258). α, α Di(*p*-ethoxypher (DBE) and trianisylchloroethylen
long-acting estrogens (257, 307, 308).
that has been attributed to their a
in body fat (117, 255, 308).
Early attempts to develop estroge. with a simple definition such as "antiestrogen."

II. Historical Development

The pioneering studies by Dodds and coworkers established the structure-activity relationships for nonsteroidal estrogens (69, 70, 72). Diethyl

on physiological antagonism with androgens and proges-

 $\overline{\mathbb{O}}$

ANTIESTROGEN PHARMACOLOGY
togens (79). The first clues that direct antagonism of
estrogen action was possible occurred with the finding ANTIESTROGEN PHA
togens (79). The first clues that direct antagonism of
estrogen action was possible occurred with the finding
that the simultaneous administration of dimethylstilbes-ANTIESTH
togens (79). The first clues that direct antagoniestrogen action was possible occurred with the fi
that the simultaneous administration of dimethyls
trol (DMS) (figure 1) and estradiol into the ovar togens (79). The first clues that direct antagonism
estrogen action was possible occurred with the find
that the simultaneous administration of dimethylstill
trol (DMS) (figure 1) and estradiol into the ovaries
mized mouse togens (79). The first clues that direct antagonism
estrogen action was possible occurred with the find
that the simultaneous administration of dimethylstill
trol (DMS) (figure 1) and estradiol into the ovaries
mized mouse estrogen action was possible occurred with the finding
that the simultaneous administration of dimethylstilbes-
trol (DMS) (figure 1) and estradiol into the ovariecto-
mized mouse vagina prevents the full vagina cornificathat the simultaneous administration of dimethylstilber trol (DMS) (figure 1) and estradiol into the ovariect
mized mouse vagina prevents the full vagina cornificition that is produced with estradiol alone (85, 86).
simil trol (DMS) (figure 1) and estradiol into the ovariecto-
mized mouse vagina prevents the full vagina cornifica-
tion that is produced with estradiol alone (85, 86). A
similar result (49) can be obtained with 3,3', 5,5' tet mized mouse vagina prevents the full vagina cornifica-
tion that is produced with estradiol alone (85, 86). A
similar result (49) can be obtained with 3,3', 5,5' tetra-
methyl α , β -diethylstilbestrol, a compound that ion that is produced with estradiol alone (85, 86).

similar result (49) can be obtained with 3,3', 5,5' tetm

ethyl α,β -diethylstilbestrol, a compound that reduce

the duration of [³H] estradiol binding in vaginal t similar result (49) can be obtained with 3,3', 5,5' te
methyl α , β -diethylstilbestrol, a compound that red
the duration of [³H]estradiol binding in vaginal tis
(50). However, neither of these compounds is an ant
tr

the duration of [³H]estradiol binding in vaginal tissu(50). However, neither of these compounds is an antit trogen when administered systemically (49, 85).
Lerner and coworkers (210) described the pharmalogical propertie (50). However, neither of these compounds is an anties-
trogen when administered systemically (49, 85).
Lerner and coworkers (210) described the pharmaco-
logical properties of the first systemically active, nonste-
roidal trogen when administered systemically (49, 85).

Lerner and coworkers (210) described the pharmace

logical properties of the first systemically active, nonst

roidal antiestrogen, ethamoxytriphetol (MER25) (figui

2). The Lerner and coworkers (210) described the pharmaco-
logical properties of the first systemically active, nonste-
roidal antiestrogen, ethamoxytriphetol (MER25) (figure
2). The compound is virtually devoid of estrogenic acti logical properties of the first systemically active, nonste
roidal antiestrogen, ethamoxytriphetol (MER25) (figure
2). The compound is virtually devoid of estrogenic active
tity in a wide variety of species (mice, rats, ra roidal antiestrogen, ethamoxytriphetol (MER25) (figu
2). The compound is virtually devoid of estrogenic active in a wide variety of species (mice, rats, rabbit
chickens, and monkeys) and is an estrogen antagonia
but with l 2). The compound is virtually devoid of estrogenic activity in a wide variety of species (mice, rats, rabbits, chickens, and monkeys) and is an estrogen antagonist, but with low a potency. An exciting pharmacological in p ity in a wide variety of species (mice, rats, rabbits, chickens, and monkeys) and is an estrogen antagonist, but with low a potency. An exciting pharmacological property of MER25 and the related compound MRL37 (figure 2) i chickens, and monkeys) and is an estrogen antagonist,
but with low a potency. An exciting pharmacological inde
property of MER25 and the related compound MRL37 The
(figure 2) is their antifertility actions in laboratory an but with low a potency. An exciting pharmacologica
property of MER25 and the related compound MRL3'
(figure 2) is their antifertility actions in laboratory ani
mals (14, 44, 282), an observation that stimulated a
search fo property of MER25 and the related compound MRL37 In

(figure 2) is their antifertility actions in laboratory ani-

mals (14, 44, 282), an observation that stimulated a

earch for more potent agents for clinical applicatio (figure 2) is their antifertility actions in laboratory ani-
mals $(14, 44, 282)$, an observation that stimulated a
search for more potent agents for clinical application.
Clomiphene (known originally as chloramiphene and mals $(14, 44, 282)$, an observation that stimulated a search for more potent agents for clinical application.
Clomiphene (known originally as chloramiphene and fMRL41) (132, 283), nafoxidine (U-11,100A) (75), nitromifen search for more potent agents for clinical application. m
Clomiphene (known originally as chloramiphene and f^0
MRL41) (132, 283), nafoxidine (U-11,100A) (75), ni-
tromifene (CI628 or CN-55,945-27) (37), and t Clomiphene (known originally as chloramiphene a
MRL41) (132, 283), nafoxidine (U-11,100A) (75),
tromifene (CI628 or CN-55,945-27) (37), and tamoxii
(ICI 46,474) (122, 123) (figures 2, 3) are all the result
that search, bu $(ICI 46,474)$ $(122, 123)$ (figures 2, 3) are all the result of (ICI 46,474) $(122, 123)$ (figures 2, 3) are all the result of that search, but clinical application as postcoital contraceptives was found to be inappropriate. In the late 1960s enthusiasm for continued research by the p that search, but clinical application as postcoital contraceptives was found to be inappropriate. In the late 1960s enthusiasm for continued research by the pharmaceutical line industry waned. In India, however, structure

C₂H₅
OCH₂CH₂N , Co Ha **OCH2CH2N C2H5 C2H5** ethamoxytriphetol (MER 25) MRL37 **OCH₃** OCH2CH2N OCH2CHOHCH2OH CH₂O **CH** centchroman U-23,469 OCH2CH2N **.OC H2CH2** $\begin{CD} \begin{picture}(100,10) \put(0,0){\line(1,0){15}} \put(10,0){\line(1,0){15}} \put(10,$ **.OCH3** nafoxidine (UII, IOOA) trioxifene (LY133314)
FIG. 2. Nonsteroidal antiestrogens.

FIG. 3. The geometric isomers of substituted triphenylethylenes.

enclomiphene zuclomiphene
FIG. 3. The geometric isomers of substituted triphenylethylenes.
indene and chroman derivatives related to nafoxidine.
The agent, centchroman (figure 2), was developed for FIG. 3. The geometric isomers of substituted triphenylethylenes.

indene and chroman derivatives related to nafoxidine.

The agent, centchroman (figure 2), was developed for

clinical testing as a postcoital contraceptive FIG. 3. The geometric isomers of substituted triphenylethylen
indene and chroman derivatives related to nafoxic
The agent, centchroman (figure 2), was developed
clinical testing as a postcoital contraceptive (175).
The suc dene and chroman derivatives related to nafoxidine
ne agent, centchroman (figure 2), was developed for
inical testing as a postcoital contraceptive (175).
The successful introduction of tamoxifen for the treat-
ent of brea

MRL41) (132, 283), naforidine (U-11,100A) (75), nicationships of antiestrogens. This time the potential ap-
tromifene (CI628 or CN-55,945-27) (37), and tamoxifen plication is as antitumor agents for hormone-dependent
(ICI indene and chroman derivatives related to nafoxidine.
The agent, centchroman (figure 2), was developed for
clinical testing as a postcoital contraceptive (175).
The successful introduction of tamoxifen for the treat-
ment The agent, centchroman (figure 2), was developed fo
clinical testing as a postcoital contraceptive (175).
The successful introduction of tamoxifen for the treat
ment of breast cancer (232, 314) provided the incentive
for a clinical testing as a postcoital contraceptive (175).
The successful introduction of tamoxifen for the treat-
ment of breast cancer (232, 314) provided the incentive
for a renewed investigation of the structure-activity re The successful introduction of tamoxifen for the trement of breast cancer $(232, 314)$ provided the incention for a renewed investigation of the structure-activity lationships of antiestrogens. This time the potential pli for a renewed investigation of the structure-activity relationships of antiestrogens. This time the potential application is as antitumor agents for hormone-dependent disease. Trioxifene (152) (figure 2) is a compound rela lationships of antiestrogens. This time the potential apethylene structure by the introduction of a ketone that plication is as antitumor agents for hormone-dependent
disease. Trioxifene (152) (figure 2) is a compound related
to nafoxidine but diverges from the general triphenyl-
ethylene structure by the introduction of a ketone th disease. Trioxifene (152) (figure 2) is a compound relation and to nafoxidine but diverges from the general tripher ethylene structure by the introduction of a ketone thinks the phenyl ring with the alkylaminoethoxy schain to nafoxidine but diverges from the general triphenylethylene structure by the introduction of a ketone that links the phenyl ring with the alkylaminoethoxy side chain to the rest of the molecule. The general pharmacology ethylene structure by the introduction of a ketone that
links the phenyl ring with the alkylaminoethoxy side
chain to the rest of the molecule. The general pharma-
cology of trioxifene is very similar to that of tamoxifen
 links the phenyl ring with the alkylaminoethoxy side
chain to the rest of the molecule. The general pharma-
cology of trioxifene is very similar to that of tamoxifen
 $(21, 165, 265)$. Phase II clinical trials have shown a chain to the rest of the molecule. The cology of trioxifene is very similar (21, 165, 265). Phase II clinical trials in the treatment of breast cancer (not generally available for therapy. The finding that a metabolite of logy of trioxifene is very similar to that of tamoxi
1, 165, 265). Phase II clinical trials have shown active
the treatment of breast cancer (294) but the dru
t generally available for therapy.
The finding that a metabolit (21, 165, 265). Phase II clinical trials have shown activity
in the treatment of breast cancer (294) but the drug is
not generally available for therapy.
The finding that a metabolite of tamoxifen, 4-hydrox-
ytamoxifen (al Fig. 3. The geometric isomenon of substituted triphenylethylenes.

The agent, central numerality are alted to naforidine.

The agent, central numerality are applicable to the composite of the composite of the composite co

in the treatment of breast cancer (294) but the drug is
not generally available for therapy.
The finding that a metabolite of tamoxifen, 4-hydrox-
ytamoxifen (also called monohydroxytamoxifen or me-
tabolite B) (figure 4), not generally available for therapy.
The finding that a metabolite of tamoxifen, 4-hydrox-
ytamoxifen (also called monohydroxytamoxifen or me-
tabolite B) (figure 4), is a potent antiestrogen in the rat
with a binding affi ytamoxifen (also called monohydroxytamoxifen or me-
tabolite B) (figure 4), is a potent antiestrogen in the rat
with a binding affinity for the ER equivalent to that of
estradiol (159), stimulated a search for compounds wi tabolite B) (figure 4), is a potent antiestrogen in the with a binding affinity for the ER equivalent to that estradiol (159), stimulated a search for compounds we potential use as new research tools and anticanon agents. with a binding affinity for the ER equivalgnt to that of estradiol (159), stimulated a search for compounds with potential use as new research tools and anticancer agents. The hydroxylated metabolites of several antiestrog estradiol (159), stimulated a search for compounds potential use as new research tools and antical agents. The hydroxylated metabolites of several antrogens are now known to have higher potency than the parent compounds (1 potential use as new research tools and anticancer
agents. The hydroxylated metabolites of several anties-
trogens are now known to have higher potency than their
parent compounds (125). The hydroxylated triphenyle-
thylen agents. The hydroxylated metabolites of several anties-
trogens are now known to have higher potency than their
parent compounds (125). The hydroxylated triphenyle-
thylene H-1285 (figure 4) is an interesting derivative
be trogens are now known to have higher potency than their
parent compounds (125) . The hydroxylated triphenyle-
thylene H-1285 (figure 4) is an interesting derivative
because its binding affinity for the ER is reportedly 1 parent compounds (125). The hydroxylated triphenyle-
thylene H-1285 (figure 4) is an interesting derivative
because its binding affinity for the ER is reportedly 10
times higher than that of estradiol (271). These com-
pou thylene H-1285 (figure 4) is an interesting derivative because its binding affinity for the ER is reportedly 10 times higher than that of estradiol (271). These compounds are, however, all partial agonists in rat uterine a because its binding affinity for the ER is reportedly 10 times higher than that of estradiol (271). These compounds are, however, all partial agonists in rat uterine assays in vivo. It can be argued that the ideal antitumo times higher than that of estradiol (271). These com-
pounds are, however, all partial agonists in rat uterine
assays in vivo. It can be argued that the ideal antitumor
agent should have negligible estrogen agonist activit pounds are, however, all partial agonists in rat uterine
assays in vivo. It can be argued that the ideal antitumor
agent should have negligible estrogen agonist activity
and be a potent antagonist with a high affinity for assays in vivo. It can be argued that the ideal antitumor
agent should have negligible estrogen agonist activity
and be a potent antagonist with a high affinity for the
ER. To this end, two novel antiestrogens, LY117018 (2 agent should have negligible estrogen agonist activity
and be a potent antagonist with a high affinity for the
ER. To this end, two novel antiestrogens, LY117018 (21)
and LY156758 (23) (figure 4) have been introduced. Both and be a potent antagonist with a high affinity for the ER. To this end, two novel antiestrogens, LY117018 (21) and LY156758 (23) (figure 4) have been introduced. Both compounds have a high affinity for the ER and low estr ER. To this end, two novel antiestrogens, LY117018 (21)
and LY156758 (23) (figure 4) have been introduced. Both
compounds have a high affinity for the ER and low
estrogenic activity in tests in vivo. Antitumor activity is
 and LY156758 (23) (figure 4) have been introduced. Both compounds have a high affinity for the ER and low estrogenic activity in tests in vivo. Antitumor activity is observed in vivo (59) and in vitro (280), but no clinica estrogenic activity in tests in vivo. Antitumor activity is observed in vivo (59) and in vitro (280), but no clinical trials have been reported that evaluate their efficacy in patients with breast cancer.

FIG. 4. Hydroxylated nonsteroidal antiestrogens with a high binding **affmity for the estrogen receptor.**

LY 117018 keoxifene (LY156758)
FIG. 4. Hydroxylated nonsteroidal antiestrogens with a high leaffinity for the estrogen receptor.
In reviewing the historical development of antiestrogens, it is possible to define two types FIG. 4. Hydroxylated nonsteroidal antiestrogens with a high bind-
ing affinity for the estrogen receptor.
In reviewing the historical development of antiestro-
gens, it is possible to define two types of antiestrogens
that Fig. 4. Hydroxylated nonsterolati antiestrogens with a might
ing affinity for the estrogen receptor.
In reviewing the historical development of antiestr
gens, it is possible to define two types of antiestr
that function th that function through an ER-mediated mechanism.
III. General Classification of Antiestrogens **France Solution School Setter and Text** and the two types of antiestrogens that function through an ER-mediated mechanism.
 A. Estrogens with a Rapid Dissociation Rate from the Estrogen Receptor

Example 12 Follows
Example 12 Follows
Example 2 Extrogen Receptor
Example 2 Follows
Example 2 Follows
Example 2 Follows

III. General Classification of Antiestrogens μ
Estrogens with a Rapid Dissociation Rate from the
introgen Receptor
Dimethylstilbestrol (DMS) was the first example of $\frac{d}{dt}$
is type of antiestrogen. Studies w III. General Classification of Antiestrogens
A. Estrogens with a Rapid Dissociation Rate from the
Estrogen Receptor
Dimethylstilbestrol (DMS) was the first example of diff
this type of antiestrogen. Studies with radiolabel A. Estrogens with a Rapid Dissociation Rate from the

Estrogen Receptor

Dimethylstilbestrol (DMS) was the first example of

this type of antiestrogen. Studies with radiolabeled DMS

have demonstrated an interaction with t Estrogen Receptor

Dimethylstilbestrol (DMS) was the first example of

this type of antiestrogen. Studies with radiolabeled DMS

have demonstrated an interaction with the estrogen re-

ceptor (39); however, continual expos Dimethylstilbestrol (DMS) was the first example of
this type of antiestrogen. Studies with radiolabeled DMS
have demonstrated an interaction with the estrogen re-
ceptor (39); however, continual exposure of target tissues
 this type of antiestrogen. Studies with radiolabeled DMS parameter are the sterogen receptor (39); however, continual exposure of target tissues term to the compound produces a full agonist response (222). with similar sit have demonstrated an interaction with the estrogen reptor (39); however, continual exposure of target tissue
to the compound produces a full agonist response (222
A similar situation occurs with the steroids, estriol an
RU ceptor (39); however, continual exposure of target tissues term
to the compound produces a full agonist response (222). with
A similar situation occurs with the steroids, estriol and the
RU16117 (figure 1). Estriol exhibit to the compound produces a full agonist response (222). \overline{v}
A similar situation occurs with the steroids, estriol and \overline{v}
RU16117 (figure 1). Estriol exhibits a low binding affin-
ity for the ER (194) and in sho A similar situation occurs with the steroids, estriol and
RU16117 (figure 1). Estriol exhibits a low binding affin-
ity for the ER (194) and in short-term tests, produces an
inhibition of estradiol-stimulated increases in RU16117 (figure 1). Estriol exhibits a low binding af
ity for the ER (194) and in short-term tests, produce
inhibition of estradiol-stimulated increases in uter
weight (55). Indeed, estriol is apparently sufficier
"antiest ity for the ER (194) and in short-term tests, produinhibition of estradiol-stimulated increases in uniformation of estradiol-stimulated increases in uniformation (55). Indeed, estriol is apparently sufficient at mammary tu inhibition of estradiol-stimulated increases in uterine
weight (55). Indeed, estriol is apparently sufficiently
"antiestrogenic" to inhibit the induction of hormone-
dependent rat mammary tumors with dimethylbenzan-
thrace weight (55). Indeed, estriol is apparently sufficien
"antiestrogenic" to inhibit the induction of hormor
dependent rat mammary tumors with dimethylbenze
thracene (DMBA) (208). This led Lemon (208) to p
pose that this actio "antiestrogenic" to inhibit the induction of hormone-
dependent rat mammary tumors with dimethylbenzan-
thracene (DMBA) (208). This led Lemon (208) to pro-
pose that this action might be important for the protec-
tion of p dependent rat mammary tumors with dimethylbenzan-
thracene (DMBA) (208). This led Lemon (208) to pro-
pose that this action might be important for the protec-
tion of patients from developing breast cancer. Thus,
high leve thracene (DMBA) (208). This led Lemon (208) to propose that this action might be important for the protection of patients from developing breast cancer. Thus, high levels of circulating estriol would be beneficial. Unfortu pose that this action might be important for the protection of patients from developing breast cancer. Thut high levels of circulating estriol would be beneficial Unfortunately, continual exposure of target tissues that es tion of patients from developing breast cancer. Thu
high levels of circulating estriol would be beneficia
Unfortunately, continual exposure of target tissues
estriol produces full estrogenic effects (218); only inte
mitten high levels of circulating estriol would be beneficed Unfortunately, continual exposure of target tissues estriol produces full estrogenic effects (218); only in mittent administration of estriol produces partial est genic Unfortunately, continual exposure of target tissues to
estriol produces full estrogenic effects (218); only inter-
mittent administration of estriol produces partial estro-
genic action and blocks the effects of estradiol estriol produces full estrogenic effects (218); only inter-
mittent administration of estriol produces partial estro-
genic action and blocks the effects of estradiol adminis-
tered intermittently (53, 55). RU16117 also in mittent administration of estriol produces partial estro-
genic action and blocks the effects of estradiol adminis-
tered intermittently (53, 55). RU16117 also inhibits car-
cinogenesis with DMBA (187) and inhibits the gro genic action and blocks the effects of estradiol administered intermittently (53, 55). RU16117 also inhibits carcinogenesis with DMBA (187) and inhibits the growth of established DMBA-induced tumors (186). However, again R tered intermittently (53, 55). RU16117 also inhibits carcinogenesis with DMBA (187) and inhibits the growth
of established DMBA-induced tumors (186). However,
again RU16117 has a low affinity for the ER and a rapid
dissoci cinogenesis with DMBA (187) and inhibits the growth
of established DMBA-induced tumors (186). However,
again RU16117 has a low affinity for the ER and a rapid
dissociation rate. This property, which contrasts with
the high of established DMBA-induced tumors (186). However,
again RU16117 has a low affinity for the ER and a rapid
dissociation rate. This property, which contrasts with
the high binding affinity of the isomer R2858 (figure 1),
pr again RU16117 has a low affinity for the ER and a rapid
dissociation rate. This property, which contrasts with
the high binding affinity of the isomer R2858 (figure 1),
prompted Bouton and Raynaud (31) to suggest that rapi dissociation rate. This property, which contrasts with
the high binding affinity of the isomer R2858 (figure 1),
prompted Bouton and Raynaud (31) to suggest that rapid
dissociation of a ligand from the receptor could accou the high binding affinity of the isomer R2858 (figure 1),
prompted Bouton and Raynaud (31) to suggest that rapid
dissociation of a ligand from the receptor could account
for antiestrogenic activity. Unfortunately, this pri antiestrogens. dissociation of a ligand from the receptor could account
for antiestrogenic activity. Unfortunately, this principle
cannot be extrapolated to the triphenylethylene-type of
antiestrogens.
B. Triphenylethylene Derivatives

the estrogen triphenylethylene but are distinguished by

a
Nasta strategically substituted side chain, usually an alkyla
inoethoxy group. However, a glyceryl side chain can AN
a strategically substituted side chain, usually an alkylam-
inoethoxy group. However, a glyceryl side chain can be
effective and is present in compounds like U-23,469 AN
a strategically substituted side chain, usually an alkylam-
inoethoxy group. However, a glyceryl side chain can be
effective and is present in compounds like U-23,469
(figure 2). Most of these antiestrogens have a low a (figure 2). Most of these and is determined than the strategically substituted side chain, usually an alkylamined
these and is present in compounds like U-23,469 (figure 2). Most of these antiestrogens have a low affinity
 a strategically substituted side chain, usually an alkylaminoethoxy group. However, a glyceryl side chain can be effective and is present in compounds like U-23,469 (figure 2). Most of these antiestrogens have a low affini incethoxy group. However, a glyceryl side chain can be effective and is present in compounds like U-23,469 (figure 2). Most of these antiestrogens have a low affinity for the ER in vitro, so Korenman (195) suggested that t effective and is present in compounds like U-23,469 (figure 2). Most of these antiestrogens have a low affinity for the ER in vitro, so Korenman (195) suggested that this factor was important to explain their mode of actio (figure 2). Most of these antiestrogens have a low affinit
for the ER in vitro, so Korenman (195) suggested the
this factor was important to explain their mode of action
However, triphenylethylene antiestrogens with a con for the ER in vitro, so Korenman (195) suggested that
this factor was important to explain their mode of action.
However, triphenylethylene antiestrogens with a cor-
rectly positioned phenolic hydroxyl groups have an affin this factor was important to explain their mode of action.
However, triphenylethylene antiestrogens with a correctly positioned phenolic hydroxyl groups have an affinity equivalent to that of estradiol (159). Therefore, th rectly positioned phenolic hydroxyl groups have an affinity equivalent to that of estradiol (159). Therefore, the structure of the drug, rather than its receptor affinity, became of primary importance for future study. The rectly positioned phenolic hydroxyl groups have an affinity equivalent to that of estradiol (159). Therefore, the structure of the drug, rather than its receptor affinity, became of primary importance for future study. The ity equivalent to that of estradiol (159). Therefore, the structure of the drug, rather than its receptor affinity, became of primary importance for future study. The mechanism of action of antiestrogens is believed to be structure of the drug, rather than its receptor affinity, became of primary importance for future study. The mechanism of action of antiestrogens is believed to be related to their structure whereas their potency is relate

IV. General Pharmacology

An antiestrogen is usually identified and classified as to their relative affinity for the ER (211).

IV. General Pharmacology

An antiestrogen is usually identified and classified as

a compound that will inhibit the vaginal cornification

produced by estradiol in ovariectomiz IV. General Pharmacology
An antiestrogen is usually identified and classified as
a compound that will inhibit the vaginal cornification
produced by estradiol in ovariectomized rats or will in-
hibit the increase in uterine HV. GENETAL FRATTRACOLOGY
An antiestrogen is usually identified and classified as
a compound that will inhibit the vaginal cornification
produced by estradiol in ovariectomized rats or will in-
hibit the increase in uterin An antiestrogen is usually identified and classified as
a compound that will inhibit the vaginal cornification
produced by estradiol in ovariectomized rats or will in-
hibit the increase in uterine weight produced by estra a compound that will inhibit the vaginal cornification
produced by estradiol in ovariectomized rats or will in-
hibit the increase in uterine weight produced by estradiol
in immature rats. Typical results from a uterine we produced by estradiol in ovariectomized rats or will in-
hibit the increase in uterine weight produced by estradiol
in immature rats. Typical results from a uterine weight
test are shown in figure 5. Two active compounds o in immature rats. Typical results from a uterine weitest are shown in figure 5. Two active compounds different potencies, tamoxifen and trioxifene, are conserved for their ability to inhibit, in a dose-related mer, the ute test are shown in figure 5. Two active compounds of different potencies, tamoxifen and trioxifene, are compared for their ability to inhibit, in a dose-related manner, the uterotrophic effects of simultaneously administere different potencies, tamoxifen and trioxifene, are compared for their ability to inhibit, in a dose-related manner, the uterotrophic effects of simultaneously administered estradiol. The compound LY126412, trioxifene witho pared for their ab
ner, the uterotrop
tered estradiol.
without an alkyla
the doses tested.

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REVIEY

FIG. 5. The antiestrogenic effect of trioxifene, tamoxifen, and **LY** ¹⁰ **126412** in the 4-day immature rat uterine weight test. Increasing daily
126412 in the 4-day immature rat uterine weight test. Increasing daily
doses of the compounds were administered with a standard dose of Daily Dose of Compounds (μ g)
FIG. 5. The antiestrogenic effect of trioxifene, tamoxifen, and LY
126412 in the 4-day immature rat uterine weight test. Increasing daily
doess of the compounds were administered with a sta cannot be extrapolated to the triphenylethylene-type of
antiestrogens.
B. Triphenylethylene Derivatives
B. Triphenylethylene Derivatives
These compounds all have a structural similarity to
the estrogen triphenylethylene b

PHARMACOLOGICAL REVIEWS

ANTIESTROGEN PHARMACOLOGY 249

ANTIESTROGEN PHARMACOLOGY
The nonsteroidal antiestrogens are competitive inhib- (277). Enclomi
itors of estrogen-induced increases in uterine weight. with antiestrog ANTIESTROGEN
The nonsteroidal antiestrogens are competitive inhib-
itors of estrogen-induced increases in uterine weight.
Increasing doses of estradiol can reverse the antiestro-ANTIESTRO
Increasing doses of estradiol can reverse the antiestic
Increasing doses of estradiol can reverse the anties
genic action of MER25 in the mouse uterus (209) The nonsteroidal antiestrogens are competitive inhib-
itors of estrogen-induced increases in uterine weight. with
Increasing doses of estradiol can reverse the antiestro-
genic action of MER25 in the mouse uterus (209) and The nonsteroidal antiestrogens are competitive in
itors of estrogen-induced increases in uterine wei
Increasing doses of estradiol can reverse the anties
genic action of MER25 in the mouse uterus (209)
the antiestrogenic a itors of estrogen-induced increases in uterine weight.
Increasing doses of estradiol can reverse the antiestro-
genic action of MER25 in the mouse uterus (209) and
the antiestrogenic action of LY117018 and 4-hydroxy-
tamox the antiestrogenic action of LY117018 and 4-hydroxy-
tamoxifen in the immature rat uterus (166). Similar
competition or "rescue" experiments have been per-
formed in vitro. The suppressive effects of an antiestrogenic action of MER25 in the mouse uterus (209)
the antiestrogenic action of LY117018 and 4-hydre
tamoxifen in the immature rat uterus (166). Sim
competition or "rescue" experiments have been
formed in vitro. The suppressi the antiestrogenic action of LY117018 and 4-hydroxy-
tamoxifen in the immature rat uterus (166). Similar by
competition or "rescue" experiments have been per-
formed in vitro. The suppressive effects of an antiestro-
gen tamoxifen in the immature rat uterus (166). Sir
competition or "rescue" experiments have been
formed in vitro. The suppressive effects of an antie
gen on cell growth can be reversed by the addition
excess estradiol to the mpetition or "rescue" experiments have been per-
rmed in vitro. The suppressive effects of an antiestro-
n on cell growth can be reversed by the addition of
cess estradiol to the culture medium (212, 216).
The triphenyleth

formed in vitro. The suppressive effects of an antiestro-
gen on cell growth can be reversed by the addition of
excess estradiol to the culture medium (212, 216).
The triphenylethylene-types of antiestrogens are par-
tial gen on cen growth can be reversed by the addition of positions excess estradiol to the culture medium (212, 216). With The triphenylethylene-types of antiestrogens are partial agonists with regard to inducing the growth o The triphenylethylene-types of antiestrogens are partial agonists with regard to inducing the growth of the
immature rat uterus. However, the uterus is a heteroge-
neous mixture of cell types that respond differentially to tial agonists with regard to inducing the growth c
immature rat uterus. However, the uterus is a hete
neous mixture of cell types that respond differentia
the antiestrogens. The luminal epithelial cells are
stimulated to a immature rat uterus. However, the uterus is a heterogeneous mixture of cell types that respond differentially to the antiestrogens. The luminal epithelial cells are fully stimulated to an increase in size which is indistin minature rat uterus. However, the uterus is a netteroge-
neous mixture of cell types that respond differentially to
the antiestrogens. The luminal epithelial cells are fully
stimulated to an increase in size which is indi the antiestrogens. The luminal epithelial cells are fully stimulated to an increase in size which is indistinguishable from a full estrogen-stimulated increase (47, 52). However, the hypertrophy that occurs as a result of stimulated to an increase in size which is indistinguish-
able from a full estrogen-stimulated increase (47, 52)
However, the hypertrophy that occurs as a result of the
triphenylethylenes is not associated with either an i able from a full estrogen-stimulated increase $(47, 5)$
However, the hypertrophy that occurs as a result of t
triphenylethylenes is not associated with either an increase in [³H]thymidine incorporation (176) or cell d However, the hypertrophy that occurs as a result of t
triphenylethylenes is not associated with either an i
crease in $[^{3}H]$ thymidine incorporation (176) or cell di
sion (68). The stromal and myometrial cells are stim
l triphenylethylenes is not associated with either an crease in [³H]thymidine incorporation (176) or cell dision (68). The stromal and myometrial cells are stime lated to a lesser extent by the triphenylethylene derivives. crease in [³H]thymidine incorporation (176) or cell division (68). The stromal and myometrial cells are stim
lated to a lesser extent by the triphenylethylene derives. It is interesting to note, however, that the antie
t sion (68). The stromal and myometrial cells are stimulated to a lesser extent by the triphenylethylene deriva-
tives. It is interesting to note, however, that the anties-
trogen LY117018 (166), which is only weakly uterolated to a lesser extent by the triphetives. It is interesting to note, howe
trogen LY117018 (166), which is
trophic in the rat, does not stimulat
the size of luminal epithelial cells.
The pharmacology of the so-called res. It is interesting to note, however, that the anties ogen LY117018 (166), which is only weakly uterophic in the rat, does not stimulate a large increase is a size of luminal epithelial cells.
The pharmacology of the so

trogen LY117018 (166), which is only weakly uter
trophic in the rat, does not stimulate a large increase
the size of luminal epithelial cells.
The pharmacology of the so-called antiestrogens, ho
ever, is extremely complex trophic in the rat, does not stimulate a large increase in
the size of luminal epithelial cells.
The pharmacology of the so-called antiestrogens, how-
ever, is extremely complex and, at times, rather incon-
sistent. The dr the size of luminal epithelial cells.

The pharmacology of the so-called antiestrogens, how-

ever, is extremely complex and, at times, rather incon-

sistent. The drugs have different properties in different

estrogen ta ever, is extremely complex and, at times, rather inconsistent. The drugs have different properties in different estrogen target tissues and different species. For this reason it has been difficult to establish a unifying m ever, is extremely complex and, at times, rather inco
sistent. The drugs have different properties in differe
estrogen target tissues and different species. For the
reason it has been difficult to establish a unifying mec
 sistent. The drugs have different properties in different gestrogen target tissues and different species. For this reason it has been difficult to establish a unifying mechanism of action for the compounds under all test estrogen target tissues and different species. For this
reason it has been difficult to establish a unifying mech-
anism of action for the compounds under all test condi-
tions. It is therefore important to establish the reason it has been difficult to establish a unifying mechanism of action for the compounds under all test conditions. It is therefore important to establish the validity of using an "antiestrogen" in a particular system be tions. It is therefore important to establish the validity of using an "antiestrogen" in a particular system before concluding that the effect observed results from an antagonism of estrogen action. ons. It is therefore important to establish the valid
using an "antiestrogen" in a particular system before including that the effect observed results from an a
gonism of estrogen action.
Part of the confusion with the def

of using an "antiestrogen" in a particular system before concluding that the effect observed results from an an tagonism of estrogen action.
Fart of the confusion with the definition and description of antiestrogens has de Part of the confusion with the definition and description of antiestrogens has developed from the early interpretation of the pharmacological properties of the geometric isomers of clomiphene and tamoxifen (figure 3). Part of the confusion with the definition and description of antiestrogens has developed from the early inter-
pretation of the pharmacological properties of the geo-
metric isomers of clomiphene and tamoxifen (figure 3).
 tion of antiestrogens has developed from the early inter-
pretation of the pharmacological properties of the geo-
metric isomers of clomiphene and tamoxifen (figure 3).
Originally the *cis* geometric isomer of clomiphene pretation of the pharmacological properties of the geometric isomers of clomiphene and tamoxifen (figure 3).
Originally the *cis* geometric isomer of clomiphene (isomer B) was shown to have antiestrogenic (67, 244, 284) an Originally the *cis* geometric isomer of clomiphene (isomer

B) was shown to have antiestrogenic $(67, 244, 284)$ and

antitumor (281) properties in the rat, while the *trans*

geometric isomer (isomer A) was primarily substituted triphenylethylene is an antiestrogenic (67, 244, 264) and antitumor (281) properties in the rat, while the *trans* geometric isomer (isomer A) was primarily estrogenic (67, 284). In contrast, tamoxifen, the *t* geometric isomer (isomer A) was primarily estrogenic ^{en}

(67, 284). In contrast, tamoxifen, the *trans* isomer of a m

substituted triphenylethylene is an antiestrogen (121, ⁽¹

169) in the rat with antitumor propertie (67, 284). In contrast, tamoxifen, the *trans* isomer of a
substituted triphenylethylene is an antiestrogen (121, 169) in the rat with antitumor properties (161) whereas
the *cis* isomer ICI47,699 is an estrogen (121, 169 substituted triphenylethylene is an antiestrogen $(121, 169)$ in the rat with antitumor properties (161) whereas with a
the *cis* isomer ICI47,699 is an estrogen $(121, 169)$. The utering
structure of ICI47,699 as the the *cis* isomer ICI47,699 is an estrogen (121, 169). The structure of ICI47,699 as the *cis* isomer was confirmed by x -ray crystallography (189), but in the light of new analytical information, the supposed *cis* and ene cis isomer FOT47,033 is an estrogen (121, 103). The
structure of ICI47,699 as the cis isomer was confirmed
by x-ray crystallography (189), but in the light of new
analytical information, the supposed cis and trans isodesignation was incorrect. Special attention should be the paid to the paid to the mers of clomiphene were renamed enclomiphene and interaction was incorrect. Special attention should be the paid to this particular point b analytical information, the supposed *cis* and *trans* iso-
mers of clomiphene were renamed enclomiphene and int
zuclomiphene, with an acknowledgment that the original I
designation was incorrect. Special attention should mers of clomiphene were renamed enclomiphene and
zuclomiphene, with an acknowledgment that the original
designation was incorrect. Special attention should be
paid to this particular point because an investigator may
inadv

(277). Enclomiphene *(trans* isomer) is a partial agonist EXTERNACOLOGY

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(277). Enclomiphene (*trans* isomer) is a partial agonist

with antiestrogenic activity in the immature rat whereas

zuclomiphene (*cis* isomer) is an estrogen (52, 169). HARMACOLOGY
(277). Enclomiphene *(trans* isomer) is a partial ago
with antiestrogenic activity in the immature rat whe
zuclomiphene *(cis* isomer) is an estrogen (52, 169).
The biological activity of the geometric isomers 77). Enclomiphene (*trans* isomer) is a partial agonist
th antiestrogenic activity in the immature rat whereas
clomiphene (*cis* isomer) is an estrogen (52, 169).
The biological activity of the geometric isomers of 4-
dro

(277). Enclomiphene (*trans* isomer) is a partial agonist
with antiestrogenic activity in the immature rat whereas
zuclomiphene (*cis* isomer) is an estrogen (52, 169).
The biological activity of the geometric isomers of with antiestrogenic activity in the immature rat whereas zuclomiphene (*cis* isomer) is an estrogen $(52, 169)$.
The biological activity of the geometric isomers of 4-
hydroxytamoxifen and CI628 has been evaluated and
bot zuclomiphene (*cis* isomer) is an estrogen (52, 169).
The biological activity of the geometric isomers of 4-
hydroxytamoxifen and CI628 has been evaluated and
both isomers appear to be antiestrogenic (169). The Z
isomers (The biological activity of the geometric isomers of 4-
hydroxytamoxifen and CI628 has been evaluated and
both isomers appear to be antiestrogenic (169). The Z
isomers (related to the *trans* structure of tamoxifen) are
ve mydioxytamoxical and Crozo has been evaluated and
both isomers appear to be antiestrogenic (169). The Z
isomers (related to the *trans* structure of tamoxifen) are
very potent compared with the E isomers, but it is
possibl isomers (related to the *trans* structure of tamoxifen) are
very potent compared with the E isomers, but it is
possible that fractional conversion from a compound with low biological activity to one with high antiestro-
genic potency can cloud the pharmacological assessment
of the $E(cis)$ isomer. The isomers of hydroxylated trivery potent compared with the E isomers, but it is
possible that fractional conversion from a compound
with low biological activity to one with high antiestro-
genic potency can cloud the pharmacological assessment
of the possible that Hacklethal conversion from a compound
with low biological activity to one with high antiestro-
genic potency can cloud the pharmacological assessment
of the E(cis) isomer. The isomers of hydroxylated tri-
phe with low biological activity to one with high anties genic potency can cloud the pharmacological assesss of the $E(cis)$ isomer. The isomers of hydroxylated phenylethylenes are unstable in solution and in a restudy with the genic potency can cloud the pharmacological assessment
of the $E(cis)$ isomer. The isomers of hydroxylated tri-
phenylethylenes are unstable in solution and in a recent
study with the tritium-labeled E isomer of 4-hydroxy of the E(*cis*) isomer. The
phenylethylenes are unsta
study with the tritium-lab
moxifen, conversion to the
culture conditions (182).
Different species have d

Different species have different degrees of agonist and study with the tritium-labeled E isomer of 4-hydroxyta-
moxifen, conversion to the Z isomer occurred under cell
culture conditions (182).
Different species have different degrees of agonist and
antagonist actions to nonste moxiten, conversion to the Z isomer occurred under cell
culture conditions (182).
Different species have different degrees of agonist and
antagonist actions to nonsteroidal antiestrogens in their
target tissues. This parti culture conditions (182).

Different species have different degrees of agonist and

antagonist actions to nonsteroidal antiestrogens in their

target tissues. This particularly important aspect of the

pharmacology of anti Different species have different digrees of agomet antagonist actions to nonsteroidal antiestrogens in the target tissues. This particularly important aspect of pharmacology of antiestrogens will be considered in c junctio pharmacology of antiestrogens will be considered in conjunction with their duration of action and metabolism.
A. Species Differences

extragmism of estrogen action.

Part of the confusion with the definition and description and description for antiestrogens has developed from the early inter-

pretation of antiestrogens has developed from the early inter MER25 is considered to be uniformly antiestrogenical
Species Differences
MER25 is considered to be uniformly antiestrogenic
all species (210) although very slight estrogenic actions Figures. The species and the model of the species (210) although very slight estrogenic actions
have been noted by some workers (60, 327). Compounds A. *Species Differences*
MER25 is considered to be uniformly antiestrogenic
in all species (210) although very slight estrogenic actions
have been noted by some workers (60, 327). Compounds
based upon triphenylethylene are MER25 is considered to be uniformly antiestrog
in all species (210) although very slight estrogenic act
have been noted by some workers $(60, 327)$. Compou
based upon triphenylethylene are more potent anties
gens in the in all species (210) although very slight estrogenic actions
have been noted by some workers $(60, 327)$. Compounds
based upon triphenylethylene are more potent antiestro-
gens in the rat but they are also partial agoni gens in the rat but they are also partial agonists (37, 122). In the mouse, mixed results are reported depending upon the test system and the compound tested. In the immature mouse uterine weight test, tamoxifen is a full based upon triphenylethylene are more potent antiestro-
gens in the rat but they are also partial agonists (37,
122). In the mouse, mixed results are reported depending
upon the test system and the compound tested. In the
 122). In the mouse, mixed results are reported depending
upon the test system and the compound tested. In the
immature mouse uterine weight test, tamoxifen is a full
agonist (306) with no detectable antiestrogenic activit (305). In the model, mixed resents are reported depending
upon the test system and the compound tested. In the
immature mouse uterine weight test, tamoxifen is a full
agonist (306) with no detectable antiestrogenic activit upon the test system and the compound tested. In the immature mouse uterine weight test, tamoxifen is a full agonist (306) with no detectable antiestrogenic activity (305). Similarly, in ovariectomized mice, the systemic a immature mouse uterine weight test, tamoxifen is a full agonist (306) with no detectable antiestrogenic activity (305). Similarly, in ovariectomized mice, the systemic administration of tamoxifen in an Allen-Doisy test res response (305). Similarly, in ovariectomized mice, the systemic administration of tamoxifen in an Allen-Doisy test results in full vaginal cornification (121); however, this response is dose- and time-dependent. A large do definition of tamoxifen in an Allen-Doisy test results in full vaginal cornification (121); however, this response is dose- and time-dependent. A large dose (3 mg) of tamoxifen causes an initial stimulation of the vagina e the animal vegining commission (122), however, the response is dose- and time-dependent. A large dose (3 mg) of tamoxifen causes an initial stimulation of the vagina epithelium, but the cornification is transient and the a to estrogen stimulation for several stimulation of the vagina epithelium, but the cornification is transient and the animals develop a leukocytic smear that is refractory to estrogen stimulation for several weeks $(84, 15$ vagina epithelium, but the cornification is transient and
the animals develop a leukocytic smear that is refractory
to estrogen stimulation for several weeks (84, 153). The
vagina is hypertrophied (223) but the changes are vagina is hypertrophied (223) but the changes are different than those observed during continued estrogen administration (294). In contrast, 3,4-dihydroxytamoxifen (162) and LY117018 (21, 167) are both partial agonists (64, 155). The vagina is hypertrophied (223) but the changes are different than those observed during continued estrogen administration (294). In contrast, 3,4-dihydroxytamoxifen (162) and LY117018 (21, 167) are both parti vagina is hypertrophied (223) but the changes are different than those observed during continued estrogen administration (294). In contrast, 3,4-dihydroxytamoxifen (162) and LY117018 (21, 167) are both partial agonists wit ent than those observed during continued estrogen administration (294). In contrast, 3,4-dihydroxytamoxifen (162) and LY117018 (21, 167) are both partial agonists with antiestrogenic properties in overiectomized mouse uter ministration (294). In contrast, 3,4-dihydroxytamox (162) and LY117018 (21, 167) are both partial agor with antiestrogenic properties in overiectomized moterine weight tests. Both compounds also have estrogenic activity in (162) and LY117018 (21, 167) are both partial agonists with antiestrogenic properties in overiectomized mouse uterine weight tests. Both compounds also have less estrogenic activity in the immature rat and antiestrogenic with undescrigative properties in overlect
solmized intensity understand and the estrogenic activity is retained (21, 159, 166). Therefore, an-
tiestrogens can be designed with high potency and low
intrinsic activity as es estrogenic activity in the in
genic activity is retained (2
tiestrogens can be designed
intrinsic activity as estrogen
In the chick oviduct (20, 2 estrogenic activity in the immature rat and antiestro-
genic activity is retained $(21, 159, 166)$. Therefore, an-
tiestrogens can be designed with high potency and low
intrinsic activity as estrogens.
In the chick oviduc

tiestrogens can be designed with high potency and low
intrinsic activity as estrogens.
In the chick oviduct (20, 291, 295) and liver (41, 200),
the triphenylethylene-based compounds are uniformly
antiestrogenic with virtua tions.

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REVIEW

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B. Duration of Action 0
Duration of Action
Studies with radiolabeled antiestrogens have demo
rated long biological half-lives in animals (102, 1 350

B. Duration of Action

Studies with radiolabeled antiestrogens have demon-

strated long biological half-lives in animals (102, 158,

181) and man (103). The prolonged biological half-lives 18. Duration of Action

Studies with radiolabeled antiestrogens have demon-

strated long biological half-lives in animals (102, 158,

181) and man (103). The prolonged biological half-lives

of triphenylethylene antiestro Studies with radiolabeled antiestrogens have demon-
strated long biological half-lives in animals (102, 158, m
181) and man (103). The prolonged biological half-lives
of triphenylethylene antiestrogens is not surprising s Studies with radiolabeled antiestrogens have demon-
strated long biological half-lives in animals $(102, 158,$ met
181) and man (103) . The prolonged biological half-lives
of triphenylethylene antiestrogens is not surpri (117) (figure 1) have a long duration of action which has
been ascribed to their high solubility in body fat. Al-
the main that has high linearly in body fat. Al-
the main that has high linearly in body fat. Al-
the main 181) and man (103) . The prolonged biological half-lives
of triphenylethylene antiestrogens is not surprising since
the related estrogenic derivatives DBE (257) and TACE
 (117) (figure 1) have a long duration of actio of triphenylethylene antiestrogens is not surprising s
the related estrogenic derivatives DBE (257) and TA
(117) (figure 1) have a long duration of action which
been ascribed to their high solubility in body fat.
though th the related estrogenic derivatives DBE (257) and TACE (117) (figure 1) have a long duration of action which has been ascribed to their high solubility in body fat. Although the high lipophylicity of nonsteroidal antiestrog (117) (figure 1) have a long duration of action which has been ascribed to their high solubility in body fat. A though the high lipophylicity of nonsteroidal antiestrogens might contribute to their long duration of action been ascribed to their high solubility in body fat. Al-
though the high lipophylicity of nonsteroidal antiestro-
gens might contribute to their long duration of action,
the ubiquitous presence of so-called "antiestrogen bi though the high lipophylicity of nonsteroidal antiestro-
gens might contribute to their long duration of action,
the ubiquitous presence of so-called "antiestrogen bind-
ing components," in many (193, 290, 298), if not all gens might contribute to their long duration of action,
the ubiquitous presence of so-called "antiestrogen bind-
ing components," in many (193, 290, 298), if not all,
tissues will also serve to retard metabolism and excre the ubiquitous presence of so-called "antiestrogen bind-
ing components," in many (193, 290, 298), if not all,
tissues will also serve to retard metabolism and excretion.
The binding components have a very high affinity fo ing components," in many (193, 290, 298), if not all,
tissues will also serve to retard metabolism and excretion.
The binding components have a very high affinity for
triphenylethylene antiestrogens with alkylaminoethoxy
 tissues will also serve to retard metabolism and excretion.
The binding components have a very high affinity for
triphenylethylene antiestrogens with alkylaminoethoxy
side chains (290); triphenylethylenes without the side
 triphenylethylene antiestrogens with alkylaminoethoxy
side chains (290); triphenylethylenes without the side
chain do not appear to bind (290). Another contribution
to extended biological activity is the enterohepatic retriphenylethylene antiestrogens with alkylaminoethomoside chains (290); triphenylethylenes without the sichain do not appear to bind (290). Another contribution to extended biological activity is the enterohepatic referent side chains (290); triphenylethylenes without the side
chain do not appear to bind (290). Another contribution
to extended biological activity is the enterohepatic re-
circulation of metabolites. The primary route of excr chain do not appear to bind (290) . Another contribution
to extended biological activity is the enterohepatic re-
circulation of metabolites. The primary route of excre-
tion of the conjugated metabolites of tamoxifen is to extended biological activity is the enterohepatic
circulation of metabolites. The primary route of exc
tion of the conjugated metabolites of tamoxifen is via
bile duct; however, studies in the rat and dog h
demonstrated rculation of metabolites. The primary route of ex-
on of the conjugated metabolites of tamoxifen is via
le duct; however, studies in the rat and dog h
monstrated reabsorption of hydrolyzed metabolite
The hydroxylated metab

bile duct; however, studies in the rat and dog have demonstrated reabsorption of hydrolyzed metabolites.
The hydroxylated metabolite of tamoxifen, 4-hydroxytamoxifen, would be expected to have reduced lipophylicity and an ytamoxifen, would be expected to have reduced lipophyldemonstrated reabsorption of hydrolyzed metabolite
The hydroxylated metabolite of tamoxifen, 4-hyd
ytamoxifen, would be expected to have reduced lipop
icity and an increased sensitivity to metabolic conj
tion. The affinity The hydroxylated metabolite of tamoxifen, 4-hydroxytamoxifen, would be expected to have reduced lipophylicity and an increased sensitivity to metabolic conjugation. The affinity of 4-hydroxytamoxifen for "antiestrogen bind ytamoxifen, would be expected to have reduced lipophylicity and an increased sensitivity to metabolic conjugation. The affinity of 4-hydroxytamoxifen for "antiestrogen binding components" is reduced compared with tamoxifen tion. The affinity of 4-hydroxytamoxifen for "antiestro-
gen binding components" is reduced compared with ta-
moxifen (290). These factors might explain the shorter de-
duration of action of 4-hydroxytamoxifen compared to gen binding components" is reduced compared with ta-
moxifen (290). These factors might explain the shorter
duration of action of 4-hydroxytamoxifen compared to
tamoxifen (155). Similarly, the hydroxylated antiestro-
gen C moxifen (290). The
duration of action
tamoxifen (155). S.
gen CI680M has a
ether CI680 (97).
The polyhydrox Tration of action of 4-hydroxytamoxifen compared to flum
moxifen (155). Similarly, the hydroxylated antiestro-
action CI680M has a short action compared to its methyl ads
her CI680 (97).
The polyhydroxylated compounds LY11

tamoxifen (155). Similarly, the hydroxylated antiestro-
gen CI680M has a short action compared to its methyl
ether CI680 (97).
The polyhydroxylated compounds LY117018 and
LY156758 have a short duration of action in vivo (1 gen CI680M has a short action compared to its methyl
ether CI680 (97).
The polyhydroxylated compounds LY117018 and
LY156758 have a short duration of action in vivo (167;
V. C. Jordan, B. Gosden, and E. M. Cormier, unpublis the CI680 (97). The polyhydroxylated compounds LY117018 and LY156758 have a short duration of action in vivo (167; V.C. Jordan, B. Gosden, and E. M. Cormier, unpublished observation). Although LY117018 has a high affinity The polyhydroxylated compounds LY117018 and
LY156758 have a short duration of action in vivo (167;
V.C. Jordan, B. Gosden, and E. M. Cormier, unpublished
observation). Although LY117018 has a high affinity for
estrogen rec LY156758 have a short duration of action in vivo (167;
V.C. Jordan, B. Gosden, and E. M. Cormier, unpublished
observation). Although LY117018 has a high affinity for
estrogen receptors (24) there is a reduced affinity for
 V. C. Jordan, B. Gosden, and E. M. Cormier, unpublish observation). Although LY117018 has a high affinity featrogen receptors (24) there is a reduced affinity f"antiestrogen binding components" of the rat (290, 326 Clearly tion. "antiestrogen binding components" of the rat (290, 326).
Clearly this will facilitate a rapid metabolism and excretion.
V. Metabolism

The clinical use of antiestrogens has focused much tion.
V. Metabolism
The clinical use of antiestrogens has focused muclettention upon their pharmacokinetics and metabolism.
Two different experimental approaches have been taken V. Metabolism
The clinical use of antiestrogens has focused much
attention upon their pharmacokinetics and metabolism.
Two different experimental approaches have been taken:
(a) the development of specific analytical techn (a) the dinical use of antiestrogens has focused much
attention upon their pharmacokinetics and metabolism.
Two different experimental approaches have been taken:
(a) the development of specific analytical techniques to
st The clinical use of antiestrogens has focused much
attention upon their pharmacokinetics and metabolism.
Two different experimental approaches have been taken:
(a) the development of specific analytical techniques to
study attention upon their pharmacokinetics and metabol.
Two different experimental approaches have been tal
(a) the development of specific analytical technique
study serum levels of tamoxifen and its metabolite
patients; and (Two different experimental approache (a) the development of specific analy study serum levels of tamoxifen and patients; and (b) the synthesis of radi
gens to study metabolism in animals. *A. Metabolises of tamoxife*
 A. Metabolites of Tamoxifen
 A. Metabolites of Tamoxifen
 A. Metabolites of Tamoxifen
 Fromson and coworkers (10

tients; and (b) the synthesis of radiolabeled antiestroms to study metabolism in animals.
 $4-i$
 $3-4$
 $3-4$
 $4-4$

Fromson and coworkers (102, 103) were the first to

scribe the metabolism of tamoxifen in laboratory an gens to study metabolism in animals.

A. Metabolites of Tamoxifen

Fromson and coworkers (102, 103) were the first to

describe the metabolism of tamoxifen in laboratory ani-

mals and women. Initially a range of metabolit A. Metabolites of Tamoxifen
Fromson and coworkers (102, 103) were the first to
describe the metabolism of tamoxifen in laboratory ani-
mals and women. Initially a range of metabolites (A-F)
was characterized in laboratory A. Metabolites of 1 amoxifen
Fromson and coworkers (102, 103) were the first to
describe the metabolism of tamoxifen in laboratory ani-
mals and women. Initially a range of metabolites $(A-F)$
was characterized in laborator Fromson and coworkers (102, 103) were the first to
describe the metabolism of tamoxifen in laboratory ani-
mals and women. Initially a range of metabolites $(A-F)$
was characterized in laboratory animals by the isolation
of

AN
ing thin-layer chromatography (TLC). R_is were com
pared with synthetic standards and the structure of some AN
ing thin-layer chromatography (TLC). R₁8 were com-
pared with synthetic standards and the structure of some
metabolites were confirmed by gas chromatography/ AN
ing thin-layer chromatography (TLC). R_rs were con
pared with synthetic standards and the structure of son
metabolites were confirmed by gas chromatography
mass spectrometry (GC/MS) (metabolites B and E ing thin-layer chromatography (TLC). R_is were com-
pared with synthetic standards and the structure of some
metabolites were confirmed by gas chromatography/
mass spectrometry (GC/MS) (metabolites B and E).
Initially, me ing thin-layer chromatography (TLC). R₁6 were com-
pared with synthetic standards and the structure of some
metabolites were confirmed by gas chromatography/
mass spectrometry (GC/MS) (metabolites B and E).
Initially, me pared with synthetic standards and the structure of some metabolites were confirmed by gas chromatography/
mass spectrometry (GC/MS) (metabolites B and E).
Initially, metabolite B (4-hydroxytamoxifen) was be-
lieved to be metabolites were confirmed by gas chromatograp
mass spectrometry (GC/MS) (metabolites B and
Initially, metabolite B (4-hydroxytamoxifen) was
lieved to be the major metabolite in man (103); howe
there is now adequate eviden mass spectrometry (GC/MS) (metabolites B and E).
Initially, metabolite B (4-hydroxytamoxifen) was be-
lieved to be the major metabolite in man (103); however,
there is now adequate evidence to prove that N-desme-
thyltamo Initially, metabolite B (4-hydroxytamoxifen) was be-
lieved to be the major metabolite in man (103); however,
there is now adequate evidence to prove that N-desme-
thyltamoxifen (known also as metabolite X) is the major
me lieved to be the major metabolite in man (103) ; however,
there is now adequate evidence to prove that N-desme-
thyltamoxifen (known also as metabolite X) is the major
metabolite (3). The misidentification occurred becau there is now adequate evidence to prove that N-desme-
thyltamoxifen (known also as metabolite X) is the major
metabolite (3). The misidentification occurred because
4-hydroxytamoxifen and N-desmethyltamoxifen have
the sam thyltamoxifen (known also as metabolite X) is the major
metabolite (3). The misidentification occurred because
4-hydroxytamoxifen and N-desmethyltamoxifen have
the same R_f value by TLC with the particular solvent
system metabolite (3). The misidentification occurred because 4-hydroxytamoxifen and N-desmethyltamoxifen have the same R_t value by TLC with the particular solvent system used (benzene/triethylamine). The known metabolites of 4-hydroxytamoxifen and N-desmethyltamoxifen have
the same R_f value by TLC with the particular solvent
system used (benzene/triethylamine). The known me-
tabolites of tamoxifen in animals and man are shown in
figure 6. R system used (benzene/triethylamine). The known metabolites of tamoxifen in animals and man are shown in figure 6. Recently, two other metabolites of tamoxifen have been identified in patient sera: metabolite Y (13, 157, 1 tabolites of tamoxifen in animals and man are shown in figure 6. Recently, two other metabolites of tamoxifen have been identified in patient sera: metabolite Y (13, 157, 188) (a deaminated derivative of tamoxifen (figu figure 6. Recently, two other metabolites of tamoxifen
have been identified in patient sera: metabolite Y $(13, 157, 188)$ (a deaminated derivative of tamoxifen (figure
6)) and metabolite Z (188) (the didemethyl derivat figure 6. Recently, two other metabolites of tamoxifen
have been identified in patient sera: metabolite Y (13,
157, 188) (a deaminated derivative of tamoxifen (figure
6)) and metabolite Z (188) (the didemethyl derivative have been identified in patient sera: metabolite 157, 188) (a deaminated derivative of tamoxifen 6)) and metabolite Z (188) (the didemethyl deriv tamoxifen). Kemp and coworkers (188) have su that tamoxifen is first convert 157, 188) (a deaminated derivative of tamoxifen (figure 6)) and metabolite Z (188) (the didemethyl derivative of tamoxifen). Kemp and coworkers (188) have suggested that tamoxifen is first converted to N-desmethyltamoxifen 6)) and metabolite Z (188) (the didemethyl derivative
tamoxifen). Kemp and coworkers (188) have suggest
that tamoxifen is first converted to N-desmethyltam
ifen, then to didesmethyltamoxifen (primary amine), a
finally to m tamoxifen). Kemp and coworkers (188) have suggest
that tamoxifen is first converted to N-desmethyltamo
ifen, then to didesmethyltamoxifen (primary amine), at
finally to metabolite Y (primary alcohol). The mech
nism for thi *B.* Analy to metabolite Y (produce Techniques
B. Analytical Techniques
P. Analytical Techniques
The analytical techniques

The metabolities is (primary alcohol). The mecha-
Sm for this final conversion has not been elucidated.
Analytical Techniques
The analytical techniques that are available to detect
moxifen and its metabolites in patient se the metabolical metabolical and its metabolical series of the analytical rechniques
The analytical techniques
tamoxifen and its metabolites in patient sera are com-
pared in table 1. The TLC and HPLC methods both B. Analytical Techniques
The analytical techniques that are available to detect
tamoxifen and its metabolites in patient sera are com-
pared in table 1. The TLC and HPLC methods both
depend upon the conversion of triphenyl D. Analytical rechtiques
The analytical techniques that are available to detect
tamoxifen and its metabolites in patient sera are com-
pared in table 1. The TLC and HPLC methods both
depend upon the conversion of triphenyl The analytical techniques that are available to detect
tamoxifen and its metabolites in patient sera are com-
pared in table 1. The TLC and HPLC methods both
depend upon the conversion of triphenylethylenes to
fluorescent tamoxifen and its metabolites in patient sera are compared in table 1. The TLC and HPLC methods both depend upon the conversion of triphenylethylenes to fluorescent phenanthrenes for their detection. The reaction, original pared in table 1. The TLC and HPLC methods both
depend upon the conversion of triphenylethylenes to
fluorescent phenanthrenes for their detection. The re-
action, originally described by Mallory et al. (220), was
adapted b depend upon the conversion of triphenylethylenes to
fluorescent phenanthrenes for their detection. The re-
action, originally described by Mallory et al. (220), was
adapted by Sternson and coworkers (229) to identify
tamox

ANTIESTROGEN
TABLE 1
Comparison of the assay methods available to measure the
centration of tamoxifen and its metabolites in biological fluids **Comparison of the assay methods available to measure the

concentration of** *tamoxifen* **and** *its* **metabolites in biological fluids

concentration of** *tamoxifen* **and** *its* **metabolites in biological fluids

constitutive**

Assay Method	Compound Identified (ref.)	Sensitivity (per ml)
TLC*	1. Tamoxifen (4)	2.5 ng
	2. Tamoxifen, N-desmethyltamoxi- fen, metabolite Y and metabolite Z (188)	ND
HPLC	1. Tamoxifen, 4-hydroxytamoxifen (229)	1 ng
	2. Tamoxifen, N-desmethyltamoxi- fen, 4-hydroxytamoxifen (111)	0.1 _{ng}
	3. Tamoxifen, N-desmethyltamoxifen (34) , metabolite Y (34)	$<$ 5 ng
	4. Tamoxifen, N-desmethyltamoxi- fen, 4-hydroxytamoxifen (36)	\leq l ng
GC/MS	1. Tamoxifen (106)	ND
	2. Tamoxifen, 4-hydroxytamoxifen (65)	1 _{ng}
	3. Tamoxifen 4-hydroxytamoxifen	
	N-desmethyltamoxifen (64)	ND

chromatography; GC/MS, gas chromatography/mass spectrometry; * TLC, thin-laye
chromatography; (
ND, not determine

The triphenylethylenes are first extracted with diethylether and converted to phenanthrenes (figure 7) before FIG. 7. The conversion of triphenylethylenes to fluorescent phen-
anthrenes by ultraviolet (UV) activation.
The triphenylethylenes are first extracted with diethyl-
ether and converted to phenanthrenes (figure 7) before
c anthrenes by ultraviolet (UV) activation.
The triphenylethylenes are first extracted with diethylenter and converted to phenanthrenes (figure 7) before $\frac{1}{10}$
chromatography. The methodology has been improved
and modi The triphenylethylenes are first extracted with diethylether and converted to phenanthrenes (figure 7) before chromatography. The methodology has been improves and modified (111) but suffers from no internal standardizatio The triphenylethylenes are first extracted with diethylether and converted to phenanthrenes (figure 7) before chromatography. The methodology has been improved and modified (111) but suffers from no internal stan-dardizati ether and converted to phenanthrenes (figure 7) before chromatography. The methodology has been improve and modified (111) but suffers from no internal standardization, the large amounts of serum used for assage and the po chromatography. The methodology has been improved
and modified (111) but suffers from no internal stan-
dardization, the large amounts of serum used for assay,
and the poor control that is available during the ultra-
viole and modified (111) but suffers from no internal standardization, the large amounts of serum used for assay, and the poor control that is available during the ultraviolet activation reaction. Recent developments have used i dardization, the large amounts of serum used for assay,
and the poor control that is available during the ultra-
violet activation reaction. Recent developments have
used internal standards, postcolumn fluorescent activa-
 and the poor control that is available during the ultraviolet activation reaction. Recent developments have used internal standards, postcolumn fluorescent activation (34), and preliminary purification from interfering sub violet activation reaction. Recent developments have
used internal standards, postcolumn fluorescent activa-
tion (34), and preliminary purification from interfering
substances by using Sep-Pack C₁₈ cartridge (Water As-
 used internal standards, postcolumn fluorescent active tion (34), and preliminary purification from interferiaubstances by using Sep-Pack C_{18} cartridge (Water A soc., Milford, MA) (36). It should also be pointed cha tion (34), and preliminary purification from interfe
substances by using Sep-Pack C_{18} cartridge (Water
soc., Milford, MA) (36). It should also be pointed
that the extraction methodology recommended by Go
der and Stern substances by using Sep-Pack C_{18} cartridge (Water Assoc., Milford, MA) (36). It should also be pointed out that the extraction methodology recommended by Golander and Sternson (111) converts 50% of the 4-hydroxy-tamox soc., Milford, MA) (36) . It should also be pointed out that the extraction methodology recommended by Golander and Sternson (111) converts 50% of the 4-hydroxy-tamoxifen to its phenanthrene derivative. Clearly ether e

HARMACOLOGY
activation is to be used since this will lead to an under-
estimate of 4-hydroxytamoxifen. HARMACOLOGY
activation is to be used since
estimate of 4-hydroxytamoxi **C. Pharmacokinetics**
C. Pharmacokinetics
Each of the assay

estimate of 4-hydroxytamoxifen.

C. Pharmacokinetics

Each of the assay procedures has been used to monitor

the pharmacokinetics of tamoxifen, and in some cases. its metabolites, in patients. Overall, the studies confirm C. Pharmacokinetics
Each of the assay procedures has been used to monitor
the pharmacokinetics of tamoxifen, and in some cases,
its metabolites, in patients. Overall, the studies confirm
that tamoxifen has a long biologica Each of the assay procedures has been used to monitor
the pharmacokinetics of tamoxifen, and in some cases,
its metabolites, in patients. Overall, the studies confirm
that tamoxifen has a long biological half-life and read Each of the assay procedures has been used to monithe pharmacokinetics of tamoxifen, and in some case its metabolites, in patients. Overall, the studies confithat tamoxifen has a long biological half-life and read accumula the pharmacokinetics of tamoxifen, and in some cases,
its metabolites, in patients. Overall, the studies confirm
that tamoxifen has a long biological half-life and readily
accumulates to steady-state levels upon repeated a its metabolites, in patients. Overall, the studies confirm
that tamoxifen has a long biological half-life and readil
accumulates to steady-state levels upon repeated admin-
istration. A single oral dose of 10 mg of tamoxif that tamoxiten has a long biological half-life and readily
accumulates to steady-state levels upon repeated admin-
istration. A single oral dose of 10 mg of tamoxifen pro-
duces peak serum levels of 20 to 30 ng of tamoxife accumulates to steady-state levels upon repeated administration. A single oral dose of 10 mg of tamoxifen produces peak serum levels of 20 to 30 ng of tamoxifen/ml within 3 to 6 hr, but patient variation is very large (92) extration. A single oral dose of 10 mg of tamoxiten produces peak serum levels of 20 to 30 ng of tamoxifen/ml
within 3 to 6 hr, but patient variation is very large (92).
Nevertheless, continuous therapy with either 10 mg duces peak serum levels of 20 to 30 ng of tamoxifen/ml
within 3 to 6 hr, but patient variation is very large (92).
Nevertheless, continuous therapy with either 10 mg bid
(92) or 20 mg bid (240) produces a steady state in s within 3 to 6 hr, but patient variation is very large (92) .
Nevertheless, continuous therapy with either 10 mg bid
 (92) or 20 mg bid (240) produces a steady state in serum
within 4 weeks. The administration of loadi Nevertheless, continuous therapy with either 10 mg bid
(92) or 20 mg bid (240) produces a steady state in serum
within 4 weeks. The administration of loading doses (92,
322) has been recommended to raise the level of drug (92) or 20 mg bid (240) produces a steady state in serum
within 4 weeks. The administration of loading doses (92,
322) has been recommended to raise the level of drug in
the blood rapidly, followed by daily maintenance do 322) has been recommended to raise the level of drug in the blood rapidly, followed by daily maintenance doses of 20 mg. As yet, no therapeutic benefit has been reported for this approach. 2) has been recommended to raise the level of drug is
e blood rapidly, followed by daily maintenance dose
20 mg. As yet, no therapeutic benefit has been reporte
r this approach.
In general, the serum levels of N-desmethylt

the blood rapidly, followed by daily maintenance doses
of 20 mg. As yet, no therapeutic benefit has been reported
for this approach.
In general, the serum levels of N-desmethyltamoxifen
are between 50% and 100% above the l of 20 mg. As yet, no therapeutic benefit has been report
for this approach.
In general, the serum levels of N-desmethyltamoxif
are between 50% and 100% above the levels observ
with tamoxifen, but again the patient-to-patie for this approach.
In general, the serum levels of N-desmethyltamoxifen
are between 50% and 100% above the levels observed
with tamoxifen, but again the patient-to-patient varia-
tion is very great. Patients taking 20 mg o In general, the serum levels of N-desmethyltamoxifen
are between 50% and 100% above the levels observed
with tamoxifen, but again the patient-to-patient varia-
tion is very great. Patients taking 20 mg of tamoxifen
bid ac are between 50% and 100% above the levels observed
with tamoxifen, but again the patient-to-patient varia-
tion is very great. Patients taking 20 mg of tamoxifen
bid achieve a steady state for tamoxifen at 4 weeks and
for with tamoxifen, but again the patient-to-patient variation is very great. Patients taking 20 mg of tamoxifen
bid achieve a steady state for tamoxifen at 4 weeks and
for N-desmethyltamoxifen at 8 weeks. These data have
been tion is very great. Patients taking 20 mg of tand is defined as the for tamoxifen at 4 we for N-desmethyltamoxifen at 8 weeks. These dappend used to calculate approximate biological had of 7 and 14 days for tamoxifen and N bid achieve a steady state for tamoxifen at 4 weeks an
for N-desmethyltamoxifen at 8 weeks. These data have
been used to calculate approximate biological half-live
of 7 and 14 days for tamoxifen and N-desmethyltamox
ifen, for N-desmethyltamoxifen at 8 weeks. These data have
been used to calculate approximate biological half-lives
of 7 and 14 days for tamoxifen and N-desmethyltamox-
ifen, respectively (246). Metabolite Y (157) and 4-hy-
dro been used to calculate approximate biological half-lives
of 7 and 14 days for tamoxifen and N-desmethyltamox-
ifen, respectively (246). Metabolite Y (157) and 4-hy-
droxytamoxifen (64, 65, 91) are both minor metabolites
of of 7 and 14 days for tamoxiten and N-desmethyltamox-
ifen, respectively (246). Metabolite Y (157) and 4-hy-
droxytamoxifen (64, 65, 91) are both minor metabolites
of tamoxifen although it should be borne in mind that 4-
hy ifen, respectively (246) . Metabolite Y (157) and 4-hy-
droxytamoxifen $(64, 65, 91)$ are both minor metabolites
of tamoxifen although it should be borne in mind that 4-
hydroxytamoxifen has a binding affinity for huma droxytamoxifen (64, 65, 91) are both minor metabolites
of tamoxifen although it should be borne in mind that 4-
hydroxytamoxifen has a binding affinity for human
breast tumor estrogen receptors that is about 50 to 100
tim of tamoxiten although it should be borne in mind that 4-
hydroxytamoxifen has a binding affinity for human
breast tumor estrogen receptors that is about 50 to 100
times greater than that of tamoxifen (91). Examples of
the hydroxytamoxifen has a binding affinity for human
breast tumor estrogen receptors that is about 50 to 100
times greater than that of tamoxifen (91). Examples of
the range of blood or serum concentration to be expected
with 2.

TABLE **2** *Comparison* of *the concentration* of *tamoxifen and its* metabolites *in patient* blood *during therapy for breast cancer.*

Dosa	Compound Measured (ref.)	Concentration (nq/ml)		
10 mg bid	Tamoxifen	167 (143–197)*		
	4-hydroxytamoxifen (65)	$3(2-5)$		
10 mg bid	Tamoxifen	113 (77-189)†		
	N-desmethyltamoxifen	204 (163-265)		
	Metabolite Y (157)	$18(5 - 49)$		
20 mg bid	Tamoxifen	300 (270-520)*		
	N-desmethyltamoxifen	462 (210-761)		
	4-hydroxytamoxifen (64)	$7(3-11)$		
20 mg bid	Tamoxifen	310 (164–494)†		
	N-desmethyltamoxifen	481 (300-851)		
	Metabolite Y (188)	49 (22-136)		
20 mg/m^2 bid	Tamoxifen	163 (95-240)		
after loading	N-desmethyltamoxifen	289 (187-325)		
doses	4-hydroxytamoxifen (91)	$10(4-21)$		

t Serum.

ARMACOL

D. Metabolism by Laboratory Animals in Vivo
The first report (102) of the metabolism of ¹⁴C-labeled 2

Metabolism by Laboratory Animals in Vivo

The first report (102) of the metabolism of ¹⁴C-labeled

moxifen in rat, mouse, rhesus monkey, and dog iden-Jones 252

D. Metabolism by Laboratory Animals in Vivo

The first report (102) of the metabolism of ¹⁴C-labelee

tamoxifen in rat, mouse, rhesus monkey, and dog iden

tified conversion to 4-hydroxytamoxifen as a signific D. Metabolism by Laboratory Animals in Vivo
The first report (102) of the metabolism of ¹⁴C-labeled
tamoxifen in rat, mouse, rhesus monkey, and dog iden-
tified conversion to 4-hydroxytamoxifen as a significant
metabolic D. *Metabolism by Laboratory Financias in* $Vt\omega$
The first report (102) of the metabolism of ¹⁴C-labe
tamoxifen in rat, mouse, rhesus monkey, and dog id
tified conversion to 4-hydroxytamoxifen as a signific
metabolic p The first report (102) of the metabolism of ¹⁴C-labeled
tamoxifen in rat, mouse, rhesus monkey, and dog iden-
tified conversion to 4-hydroxytamoxifen as a significant
metabolic pathway in all species. The catechol, me tamoxifen in rat, mouse, rhesus monkey, and dog identified conversion to 4-hydroxytamoxifen as a significant metabolic pathway in all species. The catechol, metabolite D, is present as a glucuronide in the feces of all sp tified conversion to 4-hydroxytamoxifen as a significant
metabolic pathway in all species. The catechol, metabo-
lite D, is present as a glucuronide in the feces of all
species studied and metabolite E, tamoxifen without t metabolic pat
lite D, is pre
species studie
aminoethoxy;
in dog bile.
Recently the e D, is present as a glucuronide in the feces of ecies studied and metabolite E, tamoxifen without thinoethoxyside chain, is found as a minor metabol dog bile.
Recently the use of the tritiated antiestrogens, tamon (30) U-

species studied and metabolite E, tamoxifen without the
aminoethoxyside chain, is found as a minor metabolite
in dog bile.
Recently the use of the tritiated antiestrogens, tamoxime
ifen (30) U-23,469 (304), CI628 (nitromi aminoethoxyside chain, is found as a minor metaboli
in dog bile.
Recently the use of the tritiated antiestrogens, tamo:
ifen (30) U-23,469 (304), CI628 (nitromifene) (181), ar
LN1643 (28), have all confirmed that a primary in dog bile.

Recently the use of the tritiated antiestrogens, tamox-

ifen (30) U-23,469 (304), CI628 (nitromifene) (181), and

LN1643 (28), have all confirmed that a primary meta-

bolic transformation is from the paren Recently the use of the tritiated antiestrogens, tamoximum if the 130 U-23,469 (304), CI628 (nitromifene) (181), and LN1643 (28), have all confirmed that a primary metabolic transformation is from the parent drug to the p ifen (30) U-23,469 (304), CI628 (nitromifene) (181), and
LN1643 (28), have all confirmed that a primary meta-
bolic transformation is from the parent drug to the m
phenolic derivative. The antiestrogens with a methoxy
gro LN1643 (28), have all confirmed that a primary metabolic transformation is from the parent drug to the may
phenolic derivative. The antiestrogens with a methoxy 47.6
group are demethylated [analogous to the conversion o bolic transformation is from the parent drug to the phenolic derivative. The antiestrogens with a methoxy group are demethylated [analogous to the conversion of mestranol to ethinyl estradiol (9)] and those with an unsubst phenolic derivative. The antiestrogens with a methoxy
group are demethylated [analogous to the conversion of
mestranol to ethinyl estradiol (9)] and those with an
unsubstituted ring are *para* hydroxylated. These reac-
E mestranol to ethinyl estradiol (9)] and those with an mestranol to ethinyl estradiol (9)] and those with an unsubstituted ring are *para* hydroxylated. These reactions are illustrated in figure 8 with U-23,469 and LN1643. The conversion of the phenolic derivative increases t unsubstituted ring are *para* hydroxylated. These reactions are illustrated in figure 8 with U-23,469 and LN1643. The conversion of the phenolic derivative increases the affinity of the compound for the estrogen receptor a tions are illustrated in figure 8 with
LN1643. The conversion of the phenolic
creases the affinity of the compound for
eceptor and the hydroxylated metaboli
in the estrogen target tissues (30, 181).
Conflicting results hav N1643. The conversion of the phenolic derivative is
eases the affinity of the compound for the estrog
ceptor and the hydroxylated metabolites concentri
the estrogen target tissues $(30, 181)$.
Conflicting results have bee

creases the affinity of the compound for the estrogen
receptor and the hydroxylated metabolites concentrate
in the estrogen target tissues (30, 181).
Conflicting results have been obtained for the metab-
olism of tamoxifen receptor and the hydroxylated metabolites concentrate
in the estrogen target tissues (30, 181).
Conflicting results have been obtained for the metab-
olism of tamoxifen in the chicken. Borgna and Rochefort
(30) demonstrat in the estrogen target tissues (30, 181).
Conflicting results have been obtained for the metab-
olism of tamoxifen in the chicken. Borgna and Rochefort
(30) demonstrated the conversion of tamoxifen to 4-
hydroxytamoxifen i Conflicting results have been obtained for the metab-
olism of tamoxifen in the chicken. Borgna and Rochefort
(30) demonstrated the conversion of tamoxifen to 4-
hydroxytamoxifen in vivo and in vitro. Indeed the me-
tabol olism of tamoxifen in the chicken. Borgna and Rochefort (30) demonstrated the conversion of tamoxifen to 4-hydroxytamoxifen in vivo and in vitro. Indeed the metabolism of tamoxifen to 4-hydroxytamoxifen by chick liver was hydroxytamoxifen in vivo and in vitro. Indeed the m
tabolism of tamoxifen to 4-hydroxytamoxifen by chi
liver was used to prepare the first sample of [³H]
hydroxytamoxifen for subsequent estrogen receptor stu
ies (29). In (30) demonstrated the conversion of tamoxiren to 4-
hydroxytamoxifen in vivo and in vitro. Indeed the me-
tabolism of tamoxifen to 4-hydroxytamoxifen by chick
liver was used to prepare the first sample of $[^3H]4-$
hydroxy tabolism of tamoxifen to 4-hydroxytamoxifen by ch
liver was used to prepare the first sample of [³H
hydroxytamoxifen for subsequent estrogen receptor st
ies (29). In contrast, Binart and coworkers (20) did a
observe 4-hy liver was used to prepare the first sample of [³H]4-hydroxytamoxifen for subsequent estrogen receptor studies (29). In contrast, Binart and coworkers (20) did not observe 4-hydroxytamoxifen as a metabolite of tamoxifen i hydroxytamoxifen for subsequent estrogen receptor studies (29). In contrast, Binart and coworkers (20) did not posserve 4-hydroxytamoxifen as a metabolite of tamoxifen in serum. It is possible that the technique of ether ies (29). In contrast, Binart and coworkers (20) diobserve 4-hydroxytamoxifen as a metabolite of tan fen in serum. It is possible that the technique of α extraction used by the latter workers to isolate tamometabolites observe 4-hydro
fen in serum. It
extraction used h
metabolites resu
moxifen (111).
Large doses o It is possible that the technique of ether
traction used by the latter workers to isolate tamoxifen
etabolites resulted in the degradation of 4-hydroxyta-
pxifen (111).
Large doses of tamoxifen or enclomiphene have been
e

extraction used by the latter workers to isolate tamoxifem
metabolites resulted in the degradation of 4-hydroxyta-
moxifen (111).
Large doses of tamoxifen or enclomiphene have been
used to demonstrate metabolism to 4-hydro metabolites resulted in the degradation of 4-hydrox
moxifen (111).
Large doses of tamoxifen or enclomiphene have b
used to demonstrate metabolism to 4-hydroxytamox
(32) or 4-hydroxyenclomiphene (269) in rats. Never
less, s moxifen (111).
Large doses of tamoxifen or enclomiphene have been
used to demonstrate metabolism to 4-hydroxytamoxifen
(32) or 4-hydroxyenclomiphene (269) in rats. Neverthe-
less, several different polar metabolic products

labeled tamoxifen have been observed but, as yet, these
have not been characterized. Borgna and Rochefort (30) AN
labeled tamoxifen have been observed but, as yet, these
have not been characterized. Borgna and Rochefort (30)
described a polar metabolite, M₂, which is found to AN
labeled tamoxifen have been observed but, as yet, these
have not been characterized. Borgna and Rochefort (30)
described a polar metabolite, M_2 , which is found to
accumulate in the nuclear compartment of rat uteri labeled tamoxifen have been observed but, as yet, these
have not been characterized. Borgna and Rochefort (30)
described a polar metabolite, M_2 , which is found to
accumulate in the nuclear compartment of rat uteri 24 labeled tamoxifen have been observed but, as yet, these
have not been characterized. Borgna and Rochefort (30)
described a polar metabolite, M_2 , which is found to
accumulate in the nuclear compartment of rat uteri 24
a have not been characterized. Borgna and Rochefort (30) described a polar metabolite, M_2 , which is found to accumulate in the nuclear compartment of rat uteri 24 and 48 hr after the administration of tamoxifen. Furtherm described a polar metabolite, M_2 , which is found to accumulate in the nuclear compartment of rat uteri 24 and 48 hr after the administration of tamoxifen. Furthermore, several polar metabolites of tamoxifen were observ accumulate in the nuclear compartment of rat uteri 24
and 48 hr after the administration of tamoxifen. Fur-
thermore, several polar metabolites of tamoxifen were
observed by Robertson and coworkers (254). The same
group co and 48 hr after the administration of tamoxifen. Fur-
thermore, several polar metabolites of tamoxifen were
observed by Robertson and coworkers (254). The same
group compared the metabolism of tamoxifen and its *cis*
isome thermore, several polar metabolites of tamoxifen were
observed by Robertson and coworkers (254). The same
group compared the metabolism of tamoxifen and its *cis*
isomer, ICI 47,699, in vivo and in vitro. No dramatic
metab observed by Robertson and coworkers (254). The same
group compared the metabolism of tamoxifen and its *cis*
isomer, ICI 47,699, in vivo and in vitro. No dramatic
metabolic differences were observed between the isomers;
ho group compared the metabolism of tamoxifen and its *cis* isomer, ICI 47,699, in vivo and in vitro. No dramatic metabolic differences were observed between the isomers; however, an unknown metabolite was observed in the cy isomer, ICI 47,699, in vivo and in vitro. No dramatic
metabolic differences were observed between the isomers;
however, an unknown metabolite was observed in the
cytosols of ICI 47,699-treated rat uteri. Further study
may metabolic differences were observed between the isome
however, an unknown metabolite was observed in
cytosols of ICI 47,699-treated rat uteri. Further study
may be warranted since the single dose $(5 \mu g)$ of $\frac{1}{47,699}$ cytosols of ICI 47,699-treated rat uteri. Further study may be warranted since the single dose $(5 \mu g)$ of ICI 47,699 selected for the study may have been inappropriate, as it appears to be at the lower end of the dose re cytosols of ICI \cdot
may be warrant
47,699 selected i
priate, as it appe
response curve.
 F Metabolism in *E.* 47,699 selected for the studential and priate, as it appears to be a
response curve.
E. Metabolism in Vitro
There are very few report Fiate, as it appears to be at the lower end of the deponse curve.
 $Metabolism in Vitro$

There are very few reports that describe the metal

m of antiestrogens in vitro. However, the studies the

response curve.

E. Metabolism in Vitro

There are very few reports that describe the metabo-

lism of antiestrogens in vitro. However, the studies that

have been completed are rather interesting and several E. Metabolism in Vitro
There are very few reports that describe the metabo-
lism of antiestrogens in vitro. However, the studies that
have been completed are rather interesting and several
novel metabolites have been ident E. Metabolism in Vitro
There are very few reports that describe the metabo-
lism of antiestrogens in vitro. However, the studies that
have been completed are rather interesting and several
novel metabolites have been ident There are very few reports that describe the metabolism of antiestrogens in vitro. However, the studies that have been completed are rather interesting and several novel metabolites have been identified. Borgna and Rochefo lism of antiestrogens in vitro. However, the studies that
have been completed are rather interesting and several
novel metabolites have been identified. Borgna and Ro-
chefort (30) found that tamoxifen can be converted to have been completed are rather interesting and several
novel metabolites have been identified. Borgna and Ro-
chefort (30) found that tamoxifen can be converted to 4-
hydroxytamoxifen in liver, chicken oviduct, and lamb
ut novel metabolites have been identified. Borgna and Rochefort (30) found that tamoxifen can be converted to 4-
hydroxytamoxifen in liver, chicken oviduct, and lamb
uterus in vitro but rat uterus, dimethylbenz[a]anthra-
cene chefort (30) found that tamoxifen can be converted to 4-
hydroxytamoxifen in liver, chicken oviduct, and lamb
uterus in vitro but rat uterus, dimethylbenz[a]anthra-
cene-induced rat mammary tumor and MCF7 cells do
not tran (134) had shown that $[{}^3H]$ tamoxifen was not metabolized uterus in vitro but rat uterus, dimethylbenz[a]anthra-
cene-induced rat mammary tumor and MCF7 cells do
not transform tamoxifen significantly. An earlier study
(134) had shown that [³H]tamoxifen was not metabolized
by MC cene-induced rat mammary tumor and MCF7 cells do
not transform tamoxifen significantly. An earlier study
(134) had shown that [³H] tamoxifen was not metabolized
by MCF-7 cells but an unknown compound was observed
to appe not transform tamoxifen significantly. An earlier study (134) had shown that [³H]tamoxifen was not metabolized
by MCF-7 cells but an unknown compound was observed
to appear in the media. This may have been a breakdown
pr (134) had shown that $[{}^3H]$ tamoxifen was not metabolize
by MCF-7 cells but an unknown compound was observe
to appear in the media. This may have been a breakdow
product of tamoxifen rather than a metabolite. The
observa by MCF-7 cells but an unknown
to appear in the media. This ma
product of tamoxifen rather
observation illustrates that the
atives are potentially unstable.
Rat liver microsomes conver appear in the media. This may have been a breakdoduct of tamoxifen rather than a metabolite.
Servation illustrates that the triphenylethylene de
ives are potentially unstable.
Rat liver microsomes convert tamoxifen to 4-hy

^{2H}
thigh concentration of unidentified polar metabolites are

^{2H}
consider the N-oxide of tamoxifen (figure 6) has

^{2H}
consider identified (101) as a metabolite of tamoxifen if

⁰⁻
microsomes from phenobarbital-trea product of tamoxifen rather than a metabolite. The
observation illustrates that the triphenylethylene deriv-
atives are potentially unstable.
Rat liver microsomes convert tamoxifen to 4-hydrox-
ytamoxifen and N-desmethylta observation illustrates that the triphenylethylene derivatives are potentially unstable.

Rat liver microsomes convert tamoxifen to 4-hydroxytamoxifen and N-desmethyltamoxifen (101). ICI 47,699 is also apparently hydroxyla atives are potentially unstable.

Rat liver microsomes convert tamoxifen to 4-hydrox-

ytamoxifen and N-desmethyltamoxifen (101). ICI 47,699

is also apparently hydroxylated and demethylated but a

high concentration of un Rat liver microsomes convert tamoxifen to 4-hydrox-
ytamoxifen and N-desmethyltamoxifen (101). ICI 47,699
is also apparently hydroxylated and demethylated but a
high concentration of unidentified polar metabolites are
obse ytamoxifen and N-desmethyltamoxifen (101). ICI 47,699
is also apparently hydroxylated and demethylated but a
high concentration of unidentified polar metabolites are
observed (254). The N-oxide of tamoxifen (figure 6) has
 is also apparently hydroxylated and demethylated but a
high concentration of unidentified polar metabolites are
observed (254). The N-oxide of tamoxifen (figure 6) has
been identified (101) as a metabolite of tamoxifen if
 observed (254). The N-oxide of tamoxifen (figure
been identified (101) as a metabolite of tamox
microsomes from phenobarbital-treated rats are
The N-desmethyl metabolite of tamoxifen was pos
identified as the major metabol been identified (101) as a metabolite of tamoxifen
microsomes from phenobarbital-treated rats are use
The N-desmethyl metabolite of tamoxifen was positive
identified as the major metabolite and 4-hydroxytame
ifen (based up microsomes from phenobarbital-treated rats are u
The N-desmethyl metabolite of tamoxifen was positi
identified as the major metabolite and 4-hydroxytan
ifen (based upon HPLC retention times alone) is con
ered to be a minor The N-desmethyl metabolite of tamoxifen was positively
identified as the major metabolite and 4-hydroxytamox-
ifen (based upon HPLC retention times alone) is consid-
ered to be a minor metabolite. Mass spectral identificaidentified as the major metabolite and 4-hydroxytamoxifen (based upon HPLC retention times alone) is considered to be a minor metabolite. Mass spectral identification of 4-hydroxytamoxifen was not possible, but ether was u ifen (based upon HPLC retention times alone) is converted to be a minor metabolite. Mass spectral identifition of 4-hydroxytamoxifen was not possible, but et was used for extraction so there is a strong possibilithat the 4 ered to be a minor metabolite. Mass spectral identification of 4-hydroxytamoxifen was not possible, but ether was used for extraction so there is a strong possibility that the 4-hydroxytamoxifen is converted to phenan-
thr tion of 4-hydroxytamoxifen was not possible, but ether
was used for extraction so there is a strong possibility
that the 4-hydroxytamoxifen is converted to phenan-
threnes during extraction (111). Tamoxifen N-oxide has
ant was used for extractic
that the 4-hydroxyta
threnes during extract
antiproliferative actio
cells in culture (15).
Although the metal at the 4-hydroxytamoxifen is converted to phenan-
renes during extraction (111). Tamoxifen N-oxide has
ttiproliferative actions against MCF-7 breast cancer
lls in culture (15).
Although the metabolism of [³H]CI628 (nitro

FIG. 8. Metabolic activation of nonsteroidal antiestrogens to phe-

FIG. 8. Metabolic activation of nonsteroidal antiestrogens to phe-

FIG. 8. Metabolic activation of nonsteroidal antiestrogens to phe-

olites (figure 9) threnes during extraction (111). Tamoxifen N-oxide has antiproliferative actions against MCF-7 breast cancer cells in culture (15).

Although the metabolism of $[^{3}H]CI628$ (nitromifene) has been extensively investigated antiproliferative actions against MCF-7 breast cancer
cells in culture (15).
Although the metabolism of $[^{3}H]C1628$ (nitromifene)
has been extensively investigated in vivo (181), recent
reports (266, 268) of the metabol cells in culture (15).

Although the metabolism of $[^{3}H]C1628$ (nitromifene)

has been extensively investigated in vivo (181), recent

reports (266, 268) of the metabolism of ¹⁴C-labeled ni-

tromifene in vitro, in th Although the metabolism of $[^{3}H]C1628$ (nitromifer
has been extensively investigated in vivo (181), rece
reports (266, 268) of the metabolism of ¹⁴C-labeled in
tromifene in vitro, in the presence of rat cecal conten
h reports (266, 268) of the metabolism of 14 C-labeled nihave resulted in the identification of three novel metab-

 $\mathbb O$

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FIG. 9. Metabolism of CI 628 (nitromifene) in vivo and in vitro.

The nitro group of nitromifene to an amino

group followed by enolization and hydrolysis to the ke-FIG. 9. Metabolism of CI 628 (nitromifene) in vivo and in vitro.
reduction of the nitro group of nitromifene to an amino
group followed by enolization and hydrolysis to the ke-
tone. The oxidation of the pyrrolidine ring FIG. 9. Metabolism of CI 628 (nitromifene) in vivo and in vitro.
reduction of the nitro group of nitromifene to an amino
group followed by enolization and hydrolysis to the ke-
tone. The oxidation of the pyrrolidine ring o reduction of the nitro group of nitromifene to an amino
group followed by enolization and hydrolysis to the ke-
tone. The oxidation of the pyrrolidine ring of nitromifene
is probably analogous to the oxidation of nicotine reduction of the nitro group of nitromifene to an amino
group followed by enolization and hydrolysis to the ke-
tone. The oxidation of the pyrrolidine ring of nitromifene
is probably analogous to the oxidation of nicotine group followed by enolization and hydrolysis to the ke-
tone. The oxidation of the pyrrolidine ring of nitromifene
is probably analogous to the oxidation of nicotine to
conitine (139) and tremorine to oxotremorine (45). I tone. The oxidation of the pyrrolidine ring of nitromifene
is probably analogous to the oxidation of nicotine to
conitine (139) and tremorine to oxotremorine (45). It has
been pointed out that the ring-oxidized metabolite is probably analogous to the oxidation of nicotine to conitine (139) and tremorine to oxotremorine (45). It has been pointed out that the ring-oxidized metabolite of nitromifene is less active as an antiestrogen, but more conitine (139) and tremorine to oxotremorine (45). It has
been pointed out that the ring-oxidized metabolite of
nitromifene is less active as an antiestrogen, but more
active as an estrogen than the parent compound (268).
 nitromifene is less active as an antiestrogen, but more active as an estrogen than the parent compound (268). However, the biological activity was assessed in vivo, thereby exposing the molecule to further potential transmitromifene is less active as an antiestrogen, but more
active as an estrogen than the parent compound (268).
However, the biological activity was assessed in vivo,
thereby exposing the molecule to further potential trans-However, the biological activity was assessed in vivo,
thereby exposing the molecule to further potential trans-
formation. The result, may, therefore, not reflect the
intrinsic efficacy of the parent compound at all.
VI. **VALUATE:** The result, may, therefore, not reflect

ficacy of the parent compound at all.
 VI. Models of Estrogen Action

on for the target site (uterus, vagina, pitui

mation. The result, may, therefore, not reflect the

trinsic efficacy of the parent compound at all.

The reason for the target site (uterus, vagina, pituitary

The reason for the target site (uterus, vagina, pituitary

a intrinsic efficacy of the parent compound at all.

VI. Models of Estrogen Action

The reason for the target site (uterus, vagina, pituitary

gland) specificity of estrogens remained obscure until the

synthesis of radiolab VI. Models of Estrogen Action
The reason for the target site (uterus, vagina, pituitary
gland) specificity of estrogens remained obscure until the
synthesis of radiolabeled compounds. The first studies
with ¹⁴C-labeled V1. Models of Estrogen Action
The reason for the target site (uterus, vagina, pituitary
gland) specificity of estrogens remained obscure until the
synthesis of radiolabeled compounds. The first studies
with ¹⁴C-labeled The reason for the target site (uterus, vagina, pituitary gland) specificity of estrogens remained obscure until the synthesis of radiolabeled compounds. The first studies with ¹⁴C-labeled DES in vivo established that i gland) specificity of estrogens remained obscure until the synthesis of radiolabeled compounds. The first studies with ¹⁴C-labeled DES in vivo established that it is rapidly (within 21 hr) excreted via the bile duct (120 synthesis of radiolabeled compounds. The first studies
with ¹⁴C-labeled DES in vivo established that it is rapidly
(within 21 hr) excreted via the bile duct (120, 312), but
no selective binding by uterine tissue was obse with ¹⁴C-labeled DES in vivo established that it is ra
(within 21 hr) excreted via the bile duct (120, 312)
no selective binding by uterine tissue was observed
cause the specific activity of the compound was too
[³H]H (within 21 hr) excreted via the bile duct $(120, 312)$, but
no selective binding by uterine tissue was observed be-
cause the specific activity of the compound was too low.
[³H]Hexestrol, produced by the catalytic triti no selective binding by uterine tissue was observed be
cause the specific activity of the compound was too low
[³H]Hexestrol, produced by the catalytic tritiation/re
duction of DES, by Glascock working with Sir Charle
Do cause the specific activity of the compound was too low.

[³H]Hexestrol, produced by the catalytic tritiation/re-

duction of DES, by Glascock working with Sir Charles (²

Dodds (70), was used to demonstrate the selec [³H]Hexestrol, produced by the catalytic tritiation/re-
duction of DES, by Glascock working with Sir Charles
Dodds (70), was used to demonstrate the selective accu-
weight and sheep (110). However, the
synthesis and ext duction of DES, by Glascock working with Sir Charles Codds (70), was used to demonstrate the selective accumulation of radioactivity by the reproductive organs of dimmature female goats and sheep (110). However, the synth mulation of radioactivity by the reproductive organs of $\frac{1}{2}$ immature female goats and sheep (110). However, the EFsynthesis and extensive study of the distribution of $[{}^{3}H]$ afferentially in the immature rat by J synthesis and extensive study of the distribution of $[^{3}H]$ estradiol in the immature rat by Jensen and Jacobson (147), established the concept of target tissue-specific binding by a physiologically active estrogen. The estradiol in the immature rat by Jensen (147), established the concept of target binding by a physiologically active estrogens.

laid the foundation for all the subsequent ing the mechanism of action of estrogens.

The dif 47), established the concept of target tissue-specific
nding by a physiologically active estrogen. Their work
d the foundation for all the subsequent studies involv-
g the mechanism of action of estrogens.
The differentia

binding by a physiologically active estrogen. Their work
laid the foundation for all the subsequent studies involv-
ing the mechanism of action of estrogens.
The differential centrifugation of homogenates pre-
pared from r laid the foundation for all the subsequent studies inv
ing the mechanism of action of estrogens.
The differential centrifugation of homogenates
pared from rat uteri prebound with $[^3H]$ estradiol in v
demonstrated the subc ing the mechanism of action of estrogens.
The differential centrifugation of homogenates pre-
pared from rat uteri prebound with [³H]estradiol in vivo,
demonstrated the subcellular distribution of radioactiv-
ity in both The differential centrifugation of homogenates pared from rat uteri prebound with [³H]estradiol in videmonstrated the subcellular distribution of radioactive in both the nuclear/myofibrillar fraction and cytation at 105 pared from rat uteri prebound with [³H]estradiol in vivo,
demonstrated the subcellular distribution of radioactiv-
ity in both the nuclear/myofibrillar fraction and cytosol
(the soluble proteins in the supernatant after demonstrated the subcellular distribution of radioactivity in both the nuclear/myofibrillar fraction and cytosol (the soluble proteins in the supernatant after sedimentation at $105,000 \times g$ for 1 hr) (238, 299). These stu

HARMACOLOGY 253
to a discrete protein within the cytosol and nuclear
fractions. The cytosol protein was originally identified HARMACOLOGY 253
to a discrete protein within the cytosol and nuclear
fractions. The cytosol protein was originally identified
as a macromolecule that sediments at 9.5S on sucrose 253
to a discrete protein within the cytosol and nuclear
fractions. The cytosol protein was originally identified
as a macromolecule that sediments at 9.5S on sucrose
density gradients (309), but later studies reclassified to a discrete protein within the cytosol and nuclear
fractions. The cytosol protein was originally identified
as a macromolecule that sediments at 9.5S on sucrose
density gradients (309), but later studies reclassified th to a discrete protein within the cytosol and nuclear
fractions. The cytosol protein was originally identified
as a macromolecule that sediments at 9.5S on sucrose
density gradients (309), but later studies reclassified the fractions. The cytosol protein was originally identified
as a macromolecule that sediments at 9.5S on sucrose
density gradients (309), but later studies reclassified the
sedimentation value at 8S (260). The finding that as a macromolecule that sediments at 9.5S on sucrose
density gradients (309), but later studies reclassified the
sedimentation value at 8S (260). The finding that [³H]
estradiol can specifically bind to the ER in vitro (density gradients (309), but later studies reclassified the sedimentation value at 8S (260). The finding that $[^{3}H]$ estradiol can specifically bind to the ER in vitro (310) was a fundamental observation for all the subs sedimentation value at 8S (260). The finding that $[^{3}H]$ estradiol can specifically bind to the ER in vitro (310) was a fundamental observation for all the subsequent research and application of the methodology to breas tradiol can specifically bind to the ER in vitro (310)
is a fundamental observation for all the subsequent
search and application of the methodology to breast
ncer research.
Although the cytosol ER sediments at approximate

was a fundamental observation for all the subsequences
research and application of the methodology to brea
cancer research.
Although the cytosol ER sediments at approximate
8S, the ER extracted from the nuclear-myofibrilla 8S, the ER extracted from the nuclear-myofibrillar fraction with 0.4 M KCl sediments at 5S on gradients containing KCl (148). The cytosolic estrogen receptor was cancer research.

Although the cytosol ER sediments at approximately

8S, the ER extracted from the nuclear-myofibrillar frac-

tion with 0.4 M KCl sediments at 5S on gradients con-

taining KCl (148). The cytosolic estrog Although the cytosol ER sediments at approximately
8S, the ER extracted from the nuclear-myofibrillar frac-
tion with 0.4 M KCl sediments at 5S on gradients con-
taining KCl (148). The cytosolic estrogen receptor was
found 8S, the ER extracted from the nuclear-myofibrillar fraction with 0.4 M KCl sediments at 5S on gradients containing KCl (148). The cytosolic estrogen receptor was found to be disaggregated by 0.4 M KCl into 4S subunits (87, tion with 0.4 M KCl sediments at 5S on gradients con
taining KCl (148). The cytosolic estrogen receptor was
found to be disaggregated by 0.4 M KCl into 4S subunit
(87, 146). There was, however, some disagreement as to
whe taining KCl (148). The cytosolic estrogen receptor was
found to be disaggregated by 0.4 M KCl into 4S subunits
(87, 146). There was, however, some disagreement as to
whether the subunits of the cytosolic receptor sedi-
men found to be disaggregated by 0.4 M KCl into 4S subunits (87, 146). There was, however, some disagreement as to whether the subunits of the cytosolic receptor sedimented at 5S (197) so that they were similar to the nucle $(87, 146)$. There was, however, some disagreement as to whether the subunits of the cytosolic receptor sedi-
mented at 5S (197) so that they were similar to the
nuclear complex. It should be pointed out though that
the whether the subunits of the cytosolic receptor sedi-
mented at 5S (197) so that they were similar to the
nuclear complex. It should be pointed out though that
the precise sedimentation coefficient of the nuclear re-
cepto mented at 5S (197) so the
nuclear complex. It should
the precise sedimentation c
ceptor is questionable since
value from 4S to 6S (109).
Gorski et al. (112) and J iclear complex. It should be pointed out though to precise sedimentation coefficient of the nuclear
ptor is questionable since it can be isolated at an
lue from 4S to 6S (109).
Gorski et al. (112) and Jensen et al. (149) i

Dodds (70), was used to demonstrate the selective accu-
mulation of radioactivity by the reproductive organs of decrease in the dissociation rate of estradiol from the
immature female goats and sheep (110). However, the E the precise sedimentation coefficient of the nuclear receptor is questionable since it can be isolated at any S
value from 4S to 6S (109).
Gorski et al. (112) and Jensen et al. (149) independ-
ently developed a similar sub ceptor is questionable since it can be isolated at any S
value from 4S to 6S (109).
Gorski et al. (112) and Jensen et al. (149) independ-
ently developed a similar subcellular model to describe
the early events involved in value from 4S to 6S (109).
Gorski et al. (112) and Jensen et al. (149) independ-
ently developed a similar subcellular model to describe
the early events involved in the expression of estrogen
action. Blood-borne estradiol Gorski et al. (112) and Jensen et al. (149) independently developed a similar subcellular model to describe
the early events involved in the expression of estrogen
action. Blood-borne estradiol freely diffuses into all cel ently developed a similar subcellular model to describite early events involved in the expression of estrogenection. Blood-borne estradiol freely diffuses into all cell but is prevented from leaving the cells of an estrog the early events involved in the expression of estrogen
action. Blood-borne estradiol freely diffuses into all cells
but is prevented from leaving the cells of an estrogen
target tissue because the steroids bind, with a h action. Blood-borne estradiol freely diffuses into all cells
but is prevented from leaving the cells of an estrogen
target tissue because the steroids bind, with a high affin-
ity $(K_d = 0.7 \text{ nM})$, to the cytoplasmic ER pr but is prevented from leaving the cells of an estrogen
target tissue because the steroids bind, with a high affin-
ity $(K_d = 0.7 \text{ nM})$, to the cytoplasmic ER protein. The
resulting steroid-receptor complex then moves into target tissue because the steroids bind, with a high affinity $(K_d = 0.7 \text{ nM})$, to the cytoplasmic ER protein. The resulting steroid-receptor complex then moves into the nucleus (translocation) where it is concentrated for resulting steroid-receptor complex then moves into the nucleus (translocation) where it is concentrated for sub-
sequent interaction with nuclear components. To explain
the observation that 5S ER is extracted from uterine resulting steroid-receptor complex then moves into the
nucleus (translocation) where it is concentrated for sub-
sequent interaction with nuclear components. To explain
the observation that 5S ER is extracted from uterine
 nucleus (translocation) where it is concentrated for sub-
sequent interaction with nuclear components. To explain
the observation that 5S ER is extracted from uterine
nuclei after the administration of [³H]estradiol in v sequent interaction with nuclear components. To explain
the observation that 5S ER is extracted from uterine
nuclei after the administration of [³H]estradiol in vivo,
but the 8S cytoplasmic ER is dissociated into 4S unit the observation that 5S ER is extracted from uterin
nuclei after the administration of [³H]estradiol in vivo
but the 8S cytoplasmic ER is dissociated into 4S unit
by KCl in vitro (150), Jensen suggested that there is
tem but the 8S cytoplasmic ER is dissociated into 4S units
by KCl in vitro (150), Jensen suggested that there is a
temperature-dependent "transformation" of the ER com-
plex from 4S to 5S before translocation to the nuclear
co but the 8S cytoplasmic ER is dissociated into 4S uniby KCl in vitro (150), Jensen suggested that there is temperature-dependent "transformation" of the ER corplex from 4S to 5S before translocation to the nucle compartment by KCl in vitro (150), Jensen suggested that there is a
temperature-dependent "transformation" of the ER com-
plex from 4S to 5S before translocation to the nuclear
compartment (33, 148). An extensive study of the trans-
f temperature-dependent "transformation" of the ER com-
plex from 4S to 5S before translocation to the nuclear
compartment (33, 148). An extensive study of the trans-
formation of ER in cytosol has been made by Notides
(239, plex from 4S to 5S before translocation to the nuclear
compartment (33, 148). An extensive study of the trans-
formation of ER in cytosol has been made by Notides
(239, 241, 318). These careful studies correlate the con-
v compartment (33, 148). An extensive study of the transformation of ER in cytosol has been made by Notides (239, 241, 318). These careful studies correlate the conversion of the ER from a 4S to a 5S form in vitro with a dec formation of ER in cytosol has been made by Notides
 $(239, 241, 318)$. These careful studies correlate the con-

version of the ER from a 4S to a 5S form in vitro with a

decrease in the dissociation rate of estradiol fr (239, 241, 318). These careful studies correlate the conversion of the ER from a 4S to a 5S form in vitro with a decrease in the dissociation rate of estradiol from the ER, i.e., an increase in affinity. Recently, this ch version of the ER from a 4S to a 5S form in vitro with a decrease in the dissociation rate of estradiol from the ER, i.e., an increase in affinity. Recently, this change in affinity has been correlated with a positive coop decrease in the dissociation rate of estradiol from the ER, i.e., an increase in affinity. Recently, this change in affinity has been correlated with a positive cooperativity of estradiol interactions with the ER (240); ho vivo. finity has been correlated with a positive cooperativity
estradiol interactions with the ER (240) ; however, it
not known whether this type of interaction occurs in
vo.
Studies on the subsequent interaction of the recept

of estradiol interactions with the ER (240); however, it
is not known whether this type of interaction occurs in
vivo.
Studies on the subsequent interaction of the receptor
complex with sites within the nucleus and the eve is not known whether this type of interaction occurs
vivo.
Studies on the subsequent interaction of the recep
complex with sites within the nucleus and the event
fate of the steroid and the protein are extremely cont
versi vivo.
Studies on the subsequent interaction of the receptor
complex with sites within the nucleus and the eventual
fate of the steroid and the protein are extremely contro-
versial. On the basis of the events observed with Studies on the subsequent interaction of the receptor
complex with sites within the nucleus and the eventual
fate of the steroid and the protein are extremely contro-
versial. On the basis of the events observed with MCF-7 complex with sites within the nucleus and the eventual
fate of the steroid and the protein are extremely contro-
versial. On the basis of the events observed with MCF-7
breast cancer cells in culture, it has been suggested fate of the steroid and the protein are extremely contro
versial. On the basis of the events observed with MCF-
breast cancer cells in culture, it has been suggested tha
nuclear estradiol-ER complexes are almost all destro versial. On the basis of the events observed with MCF-7
breast cancer cells in culture, it has been suggested that
nuclear estradiol-ER complexes are almost all destroyed
or "processed" over a 5-hr period in the continual nuclear estradiol-ER complexes are almost all destroyed
or "processed" over a 5-hr period in the continual pres-
ence of estradiol (135). This event has been implicated
as the signal for the eventual synthesis of progester

254

receptor (137). The site within the nucleus that regulates

these biochemical events is unknown, but an interaction
 $=$ 5 JORD.

1993 These biochemical events is unknown, but an interaction

1994 These biochemical events is unknown, but an interaction

1995 The containing base pairs in

1997 The Containing base pairs in of receptor (137). The site within the nucleus that regulates
these biochemical events is unknown, but an interaction
of receptor complexes with G-C containing base pairs in
the DNA may be involved because actinomycin D, receptor (137). The site within the nucleus that regulates
these biochemical events is unknown, but an interaction
of receptor complexes with G-C containing base pairs in
the DNA may be involved because actinomycin D, in
h of receptor complexes with G-C containing base pairs in the DNA may be involved because actinomycin D, in high concentrations, prevents processing (136). While this hypothesis is extremely attractive, it unfortunately does of receptor complexes with G-C containing base pairs in
the DNA may be involved because actinomycin D, in
high concentrations, prevents processing (136). While
this hypothesis is extremely attractive, it unfortunately
doe

hypothesis. Some reports (214, 215) have challenged the
cytoplasmic site for the "transformation" reaction and
have suggested that translocation of the 4S ER precedes
transformation to the 5S form in the nucleus. Indeed,

Faised to the ER are powerful tools for the detection of

ER without the necessity of steroid binding. The results

have proved to be controversial. A polyclonal antibody,

raised to human breast tumor ER, and tagged with ER without the necessity of steroid binding. The results
have proved to be controversial. A polyclonal antibody,
raised to human breast tumor ER, and tagged with a
fluorescent dye has been used to demonstrate both cy-
top have proved to be controversial. A polyclonal antibody,

raised to human breast tumor ER, and tagged with a

fluorescent dye has been used to demonstrate both cy-

toplasmic and nuclear ER, but more importantly, trans-

l

PHARMACOLOGICAL REVIEWS

aspet

REVIEW CAL HARMACOLOGIO

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ANTIESTROGE
ER-containing GH₃ rat pituitary tumor cells with cyto-
chalasin B. Separation of cells into nucleoplasts and **ER-containing GH₃ rat pituitary tumor cells with cyto-** conchalasin B. Separation of cells into nucleoplasts and In extrapolast on Percoll gradients containing cytochalasin B oliz ANTIESTROGEN PHARMA
ER-containing GH₃ rat pituitary tumor cells with cyto-
comple
chalasin B. Separation of cells into nucleoplasts and In con
cytoplast on Percoll gradients containing cytochalasin B olized
results in mo ER-containing GH_3 rat pituitary tumor cells with cytochalasin B. Separation of cells into nucleoplasts and Incytoplast on Percoll gradients containing cytochalasin B oliveaults in most of the unoccupied receptor being l ER-containing GH_3 rat pituitary tumor cells with cyto-
chalasin B. Separation of cells into nucleoplasts and
cytoplast on Percoll gradients containing cytochalasin B
results in most of the unoccupied receptor being loca chalasin B. Separation of cells into nucleoplasts and cytoplast on Percoll gradients containing cytochalasin B results in most of the unoccupied receptor being located in the nucleoplast faction. Although it is possible th cytoplast on Percoll gradients containing cytochalasin l
results in most of the unoccupied receptor being locate
in the nucleoplast faction. Although it is possible tha
each of these studies is generating a discrete artifa results in most of the unoccupied receptor being located
in the nucleoplast faction. Although it is possible that
each of these studies is generating a discrete artifactual
result, the simplified model 2 of estrogen action in the nucleoplast faction. Although it is
each of these studies is generating a discreed the simplified model 2 of estroger
trated in figure 10 should also be conside
resentative of subcellular events in vivo.
The nonster ch of these studies is generating a discrete artifact
sult, the simplified model 2 of estrogen action ill
atted in figure 10 should also be considered to be r
sentative of subcellular events in vivo.
The nonsteroidal antie

l nuclear membi
(figure 11). This
of any high affir
A *in vivo* result, the simplified model 2 of estrogen action illus-
trated in figure 10 should also be considered to be rep-
resentative of subcellular events in vivo.
The nonsteroidal antiestrogens produce some interest-
ing results trated in figure 10 should also be considered to be rep-
resentative of subcellular events in vivo.
The nonsteroidal antiestrogens produce some interest-
ing results that could be interpreted to support the
simplified mode resentative of subcellular events in vivo.
The nonsteroidal antiestrogens produce some interest-
ing results that could be interpreted to support the
simplified model of estrogen action rather than the tra-
ditional two-st ditional two-step hypothesis. The administration of es-
tradiol, or an antiestrogen with a high affinity for the
ER like 4-hydroxytamoxifen, to immature rats causes an
increase in uterine wet weight and an elevation of pro ditional two-step hypothesis. The administration of estandiol, or an antiestrogen with a high affinity for the effects are associated increase in uterine wet weight and an elevation of progesterone receptor levels (68). Th tradiol, or an antiestrogen with a high affinity for the ER like 4-hydroxytamoxifen, to immature rats causes an increase in uterine wet weight and an elevation of progesterone receptor levels (68). The effects are associat ER like 4-hydroxytamoxifen, to immature rats causes an $(54$
increase in uterine wet weight and an elevation of proset
gesterone receptor levels (68) . The effects are associated $(K$
with a re-compartmentalization of est increase in uterine wet weight and an elevation of progesterone receptor levels (68). The effects are associated with a re-compartmentalization of estrogen receptors from the cytosol to the nucleus and this is believed to gesterone receptor levels (68). The effects are associated (*I* with a re-compartmentalization of estrogen receptors function the cytosol to the nucleus and this is believed to be site a requirement before estrogen-stimula with a re-compartmentalization of estrogen receptors
from the cytosol to the nucleus and this is believed to be
a requirement before estrogen-stimulated effects can oc-
cur (model I). However, antiestrogens with a low affi from the cytosol to the nucleus and this is believed to be
a requirement before estrogen-stimulated effects can oc-
cur (model I). However, antiestrogens with a low affinity
for the ER that are unlikely to be metabolically a requirement before estrogen-stimulated effects can oc-
cur (model I). However, antiestrogens with a low affinity (for the ER that are unlikely to be metabolically activated, al
cause uterine growth, and an increase in p cur (model I). However, antiestrogens with a low affinity
for the ER that are unlikely to be metabolically activated,
cause uterine growth, and an increase in progesterone
receptor but, apparently, do not cause localizatio for the ER that are unlikely to be metabolically activated, cause uterine growth, and an increase in progesterone receptor but, apparently, do not cause localization of receptor complexes in the nucleus compartment (168). cause uterine growth, and an increase in progesterone
receptor but, apparently, do not cause localization of
receptor complexes in the nucleus compartment (168).
One explanation is that the ER has a predominantly
nuclear receptor but, apparently, do not cause localization of receptor complexes in the nucleus compartment (168). A.
One explanation is that the ER has a predominantly muclear location in vivo, but when the cell is disrupted in receptor complexes in the nucleus compartment (168).
One explanation is that the ER has a predominantly
nuclear location in vivo, but when the cell is disrupted
in vitro, the unoccupied receptor "leaks out" of the
damaged One explanation is that the ER has a predominantly
nuclear location in vivo, but when the cell is disrupted
in vitro, the unoccupied receptor "leaks out" of the
damaged nuclear membrane and appears as a cytosolic
protein (nuclear location in vivo, but when the cell is disrupted
in vitro, the unoccupied receptor "leaks out" of the
damaged nuclear membrane and appears as a cytosolic 17
protein (figure 11). This "leakage" is prevented by th

Low Affinity High Affinity
H H Ligand Ligand **H** H P R R R \bullet R 's Activation .HR R' HR^* Nuclea LR⁴ HR^{*} LR' HR* ۱R R R
 B

Unoccupied

B CELL DISRUPTION in vitro

FIG. 11. A functional model for estrogen action. High affinity ligand R \sim R \sim \sim Unoccupied Receptor

Receptor

H) enters the CELL DISRUPTION *in vitro*

H) enters the cell and binds to the estrogen accion. High affinity ligand

H) enters the cell and binds to the estrogen receptor (R) in the nucleus

o produce an activate **B CELL DISRUPTION** in **vitro**
FIG. 11. A functional model for estrogen action. High affinity ligand
H) enters the cell and binds to the estrogen receptor (R) in the nucleus
co produce an activated complex HR^* and estro **cell disruption** is the strongen action. High affinity ligand H) enters the cell and binds to the estrogen receptor (R) in the nucleus to produce an activated complex HR^* and estrogenic responses. During cell disruption FIG. 11. A functional model for estrogen action. High affinity ligand Cy

H) enters the cell and binds to the estrogen receptor (R) in the nucleus is

co produce an activated complex is retained in the nucleus. Low affin complex dissociates and activated complex HR^* and estrogenic responses. During
cell disruption, this complex is retained in the nucleus. Low affinity
ligands (L) enter the cell and bind to nuclear receptor to produce an cell disruption, this complex is retained in this
digands (L) enter the cell and bind to nuclea
activated complex LR*. During homogenize
complex dissociates and unoccupied R, and j
out of the nucleus into the cytosolic fra

EXEMBLE 1986
Complex is retained in the nucleus during cell disruption
In contrast, the low affinity ligands that are not metab IN COLOGY
IN COMPLEX IS retained in the nucleus during cell disruption
In contrast, the low affinity ligands that are not metablized to high affinity compound in vivo can bind to t olized to high affinity compound in vivo can bind to the In contrast, the low affinity ligands that are not metabolized to high affinity compound in vivo can bind to the ER in the nucleus to elicit estrogenic response but their rapid dissociation during homogenization allows the complex is retained in the nucleus during cell disruption.
In contrast, the low affinity ligands that are not metab-
olized to high affinity compound in vivo can bind to the
ER in the nucleus to elicit estrogenic response In contrast, the low affinity ligands that are not metab-
olized to high affinity compound in vivo can bind to the
ER in the nucleus to elicit estrogenic response but their
rapid dissociation during homogenization allows t olized to high affinity compound in vivo can bind to the ER in the nucleus to elicit estrogenic response but their rapid dissociation during homogenization allows the uncocupied receptor to leak out of the nucleus. Thus th ER in the nucleus to elicit estrogenic response but their
rapid dissociation during homogenization allows the un-
occupied receptor to leak out of the nucleus. Thus the
majority of the receptor appears to be in the cytosol rapid dissociation during homogenization
occupied receptor to leak out of the r
majority of the receptor appears to leak
fraction after treatment with these
though estrogenic stimulation occurs.
A further element to be con majority of the receptor appears to be in the cytosol
fraction after treatment with these compounds even
though estrogenic stimulation occurs.
A further element to be considered in this complicated

ing results that could be interpreted to support the picture is the heterogeneity of nuclear estrogen receptors
simplified model of estrogen action rather than the tra-
(54). Two classes have been described: type I, the cl simplified model of estrogen action rather than the tra-
ditional two-step hypothesis. The administration of es-
ical estrogen receptor, and type II which may be found
tradiol, or an antiestrogen with a high affinity for picture is the heterogeneity of nuclear estrogen receptors fraction after treatment with these compounds eve
though estrogenic stimulation occurs.
A further element to be considered in this complicate
picture is the heterogeneity of nuclear estrogen receptor
(54). Two classes have though estrogenic stimulation occurs.

A further element to be considered in this complicated

picture is the heterogeneity of nuclear estrogen receptors

(54). Two classes have been described: type I, the class-

ical est A further element to be considered in this complicate
picture is the heterogeneity of nuclear estrogen receptor
(54). Two classes have been described: type I, the class
ical estrogen receptor, and type II which may be foun picture is the heterogeneity of nuclear estrogen receptors (54). Two classes have been described: type I, the class-
ical estrogen receptor, and type II which may be found
either in cytosol (54, 88) or the nuclear compartm (54). Two classes have been described: type I, the class-
ical estrogen receptor, and type II which may be found
either in cytosol (54, 88) or the nuclear compartment
(54, 88, 224) but whose distribution is unaffected by
 ical estrogen receptor, and type II which may be found
either in cytosol $(54, 88)$ or the nuclear compartment
 $(54, 88, 224)$ but whose distribution is unaffected by
estrogen. The type II sites have low affinity for estr either in cytosol $(54, 88)$ or the nuclear compartment $(54, 88, 224)$ but whose distribution is unaffected by estrogen. The type II sites have low affinity for estradiol $(K_d \ 30 \ nM)$ and appear to have high capacity. Th estrogen. The type II sites have low affinity for estradiol $(K_d 30 \text{ nM})$ and appear to have high capacity. Their function is unknown; however, an increase in type II sites has been suggested to be an intermediate step in $(K_d$ 30 nM) and appear to have high capacity. T
function is unknown; however, an increase in typ
sites has been suggested to be an intermediate step
the mechanism by which estradiol causes uterine gro
(54, 226). Recently, function is unknown;
sites has been sugges
the mechanism by whi
(54, 226). Recently, a
ally identified (225). *A. Studies in Vivo* **Note in the Standard Section**
 VH. Antiestrogen Action

V**H. Antiestrogen Action**

Vivo

(54, 88, 224) but whose distribution is unaffected by
estrogen. The type II sites have low affinity for estrated
io (K_4 30 nM) and appear to have high capacity. Their
function is unknown; however, an increase in type I The dose-related inhibition of estradiol-stimulated uterine weight produced by antiestrogen in the rat (163, VII. Antiestrogen Action

A. Studies in Vivo

The dose-related inhibition of estradiol-stimulated

uterine weight produced by antiestrogen in the rat (163,

174) is correlated with an ability to inhibit the binding 4. Studies in Vivo
The dose-related inhibition of estradiol-stimulated
uterine weight produced by antiestrogen in the rat (163,
174) is correlated with an ability to inhibit the binding
of [³H]estradiol in vivo (172). An The dose-related inhibition of estradiol-stimulaterine weight produced by antiestrogen in the rat 174) is correlated with an ability to inhibit the bind of $[^{3}H]$ estradiol in vivo (172). Antiestrogen adminition followin The dose-related inhibition of estradiol-stimulated
uterine weight produced by antiestrogen in the rat (163,
174) is correlated with an ability to inhibit the binding
of $[^{3}H]$ estradiol in vivo (172). Antiestrogen admin uterine weight produced by antiestrogen in the rat (163,
174) is correlated with an ability to inhibit the binding
of [³H]estradiol in vivo (172). Antiestrogen administra-
tion following the binding of [³H]estradiol by 174) is correlated with an ability to inhibit the binof $[^{3}H]$ estradiol in vivo (172). Antiestrogen administion following the binding of $[^{3}H]$ estradiol by estro-target tissues also rapidly reverses the estrogen binof of [³H]estradiol in vivo (172). Antiestrogen administration following the binding of [³H]estradiol by estrogen target tissues also rapidly reverses the estrogen binding (figure 12). This property of the nonsteroidal an tion following the binding of $[^{3}H]$ estradiol by estrogen target tissues also rapidly reverses the estrogen binding (figure 12). This property of the nonsteroidal antiestrogens, however, does not completely explain anti target tissues also rapidly reverses the estrogen binding (figure 12). This property of the nonsteroidal antiestragens, however, does not completely explain antiestroge action. Antiestrogens, which are estrogens in the mou (figure 12). This property
gens, however, does not c
action. Antiestrogens, wh
also inhibit the binding o
gen target tissues (153).
Rochefort et al. (263) p ns, however, does not completely explain antiestrogen
tion. Antiestrogens, which are estrogens in the mouse,
so inhibit the binding of [³H]estradiol in mouse estro-
n target tissues (153).
Rochefort et al. (263) provided

Unoccupied

Receptor
 \overline{B} CELL DISRUPTION *in vitro*
 \overline{B} FIG. action. Antiestrogens, which are estrogens in the mouse,
also inhibit the binding of $[^{3}H]$ estradiol in mouse estro-
gen target tissues (153).
Rochefort et al. (263) provided the first evidence that
antiestrogen can app also inhibit the binding of [³H]estradiol in mouse estro
gen target tissues (153).
Rochefort et al. (263) provided the first evidence that
antiestrogen can apparently translocate the ER to the
nucleus in vivo. The develo gen target tissues (153).

Rochefort et al. (263) provided the first evidence that

antiestrogen can apparently translocate the ER to the

nucleus in vivo. The development (10) of [³H]estradiol

exchange assay to identif Rochefort et al. (263) provided the first evidence that
antiestrogen can apparently translocate the ER to the
nucleus in vivo. The development (10) of [³H]estradiol
exchange assay to identify filled nuclear ER sites has
 antiestrogen can apparently translocate the ER to the
nucleus in vivo. The development (10) of [³H]estradiol
exchange assay to identify filled nuclear ER sites has
proved to be a powerful technique with which to study
th nucleus in vivo. The development (10) of [³H]estra
exchange assay to identify filled nuclear ER sites
proved to be a powerful technique with which to st
the cellular "distribution" and kinetics of ER in
following antiest exenange assay to identify filled nuclear ER sites has
proved to be a powerful technique with which to study
the cellular "distribution" and kinetics of ER in vivo
following antiestrogen administration. Most antiestro-
ge the cellular "distribution" and kinetics of ER in vivo
following antiestrogen administration. Most antiestro-
gens studied (38, 51, 163, 179) appear to translocate ER
to the nucleus. Large doses of antiestrogens produce a
 following antiestrogen administration. Most antiestrogens studied (38, 51, 163, 179) appear to translocate ER
to the nucleus. Large doses of antiestrogens produce a
long-term depletion of the rat uterine cytoplasmic ER
poo gens studied (38, 51, 163, 179) appear to translocate ER
to the nucleus. Large doses of antiestrogens produce a
long-term depletion of the rat uterine cytoplasmic ER
pool (51, 57). Clark and coworkers (51) first suggested
 to the nucleus. Large doses of antiestrogens produce a long-term depletion of the rat uterine cytoplasmic ER pool (51, 57). Clark and coworkers (51) first suggested that the antiestrogen-ER complex remains in the nucleus f long-term depletion of the rat uterine cytoplasmic ER that the antiestrogen-ER complex remains in the nucleus
for a prolonged period producing a specific inhibition of
cytoplasmic ER resynthesis (57). As a result, the tissue
is refractory to any subsequent estrogenic stimuli. for a prolonged period producing a specific inhibition of cytoplasmic ER resynthesis (57). As a result, the tissue is refractory to any subsequent estrogenic stimuli. This attractive theory gained support (179) but nafoxid cytoplasmic ER resynthesis (57). As a result, the tissue
is refractory to any subsequent estrogenic stimuli. This
attractive theory gained support (179) but nafoxidine
was shown to replenish ER (38) and it now seems clear
 is refractory to any subsequent estrogenic stimuli. This
attractive theory gained support (179) but nafoxidine
was shown to replenish ER (38) and it now seems clear
that the ability of antiestrogens to produce a prolonged
 attractive theory gained support (179) but nafoxidine
was shown to replenish ER (38) and it now seems clear
that the ability of antiestrogens to produce a prolonged
depletion of the cytoplasmic ER pool is a reflection of
t

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aspet

Hours ofter \mathbb{C}^3 H \mathbb{C} Estradiol Injection
Fig. 12. The effect of an injection of 20 μ g LY 117018 (sc in 0.1 ml
of peanut oil) on the levels of radioactivity in immature rat uterus (O),
vagina (Δ) and pit **FIG.** 12. The effect of an injection of 20 μ g LY 117018 (sc in 0.1 ml
of peanut oil) on the levels of radioactivity in immature rat uterus (O), ce
vagina (Δ) and pituitary glands (\square). The LY 117018-treated group FIG. 12. The effect of an injection of 20 μ g ET 117016 (sc in 0.1 m) of peanut oil) on the levels of radioactivity in immature rat uterus (O), vagina (Δ) and pituitary glands (\Box). The LY 117018-treated groups lir vagina (Δ) and pituitary glands (\square). The LY 117018-treate
injected 2 h after an injection of 10 μ Ci (0.04 μ g) [6,7³H]est
0.1 ml of peanut oil. Control groups were injected with peanut
Ten rats per group. Tis 0.1 ml of peanut oil. Control groups were injected with peanut oil alone.
Ten rats per group. Tissue radioactivity was determined by using a
Tricarb tissue oxidizer. Data from Jordan and Gosden (166).
stances, any resynthe

gands from the blood. Evidence can be presented to Fricarb tissue oxidizer. Data from Jordan and Gosden (166). (184
stances, any resynthesized cytoplasmic receptor is trans-
located immediately to the nuclear compartment by li-
has
gands from the blood. Evidence can be pre stances, any resynthesized cytoplasmic receptor is trans-
located immediately to the nuclear compartment by li-
gands from the blood. Evidence can be presented to
demonstrate that prolonged depletion of the cytoplasmic
ER stances, any resynthesized cytoplasmic receptor is trans-
located immediately to the nuclear compartment by li-
gands from the blood. Evidence can be presented to
demonstrate that prolonged depletion of the cytoplasmic
ER located immediately to the nuclear compartment by ligands from the blood. Evidence can be presented the demonstrate that prolonged depletion of the cytoplasmine ER pool is not a primary antiestrogenic mechanism. This effec gands from the blood. Evidence can be presented to
demonstrate that prolonged depletion of the cytoplasmic
ER pool is not a primary antiestrogenic mechanism. This
effect can be duplicated by the administration of long-
act demonstrate that prolonged depletion of the cytoplasmic cep
ER pool is not a primary antiestrogenic mechanism. This rece
effect can be duplicated by the administration of long-
acting estrogens which deplete the cytoplasmi ER pool is not a primary antiestrogenic mechanism. This
effect can be duplicated by the administration of long-
acting estrogens which deplete the cytoplasmic receptor
pool but stimulate full tissue growth $(53, 174, 180)$ effect can be duplicated by the administration of longacting estrogens which deplete the cytoplasmic receptor
pool but stimulate full tissue growth (53, 174, 180). On
the other hand, tamoxifen produces, as might be ex-
late
pected, a depletion of the cytoplasmic ER pool in a pool but stimulate full tissue growth (53, 174, 180). On ifen r
the other hand, tamoxifen produces, as might be ex-
pected, a depletion of the cytoplasmic ER pool in a dose-
related manner (174), but small doses of tamoxif the other hand, tamoxifen produces, as might be ex-
pected, a depletion of the cytoplasmic ER pool in a dose-
related manner (174), but small doses of tamoxifen can
grownhibit estrogen action without dramatically altering pected, a depletion of the cytoplasmic ER pool in a dose-
related manner (174), but small doses of tamoxifen can
inhibit estrogen action without dramatically altering ER
is levels in the cytoplasm (163). A similar result c (198). levels in the cytoplasm (163). A similar result can be obtained with tamoxifen in the ovariectomized rate (198).
Progress in understanding nuclear effects of antiestro-

levels in the cytoplasm (163). A similar result can be
obtained with tamoxifen in the ovariectomized ra
(198). Progress in understanding nuclear effects of antiestro-
gens is retarded because the mechanism whereby estro-
g obtained with tamoxifen in the ovariectomized rate fo

(198). sy

Progress in understanding nuclear effects of antiestro-

logens is retarded because the mechanism whereby estro-

gen stimulated uterine growth is poorly un (198).

Progress in understanding nuclear effects of antiestro-

gens is retarded because the mechanism whereby estro-

gen stimulated uterine growth is poorly understood. The

idea that true growth results from a strong Progress in understanding nuclear effects of antiestigens is retarded because the mechanism whereby estigen stimulated uterine growth is poorly understood. This discussion is that true growth results from a strong associat gens is retarded because the mechanism whereby estro-
gen stimulated uterine growth is poorly understood. The
idea that true growth results from a strong association
(salt resistant) of the ER complex with nuclear "accep-

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JORDAN
complexes are only loosely bound by the nuclear matrix
(16, 163). Studies also indicate (16) that estrogen and AN
complexes are only loosely bound by the nuclear matrix
(16, 163). Studies also indicate (16) that estrogen and
antiestrogen may bind to separate sites in the nucleus. AN
complexes are only loosely bound by the nuclear matrix
(16, 163). Studies also indicate (16) that estrogen and
antiestrogen may bind to separate sites in the nucleus.
In assessing the relevance of these studies, it shou complexes are only loosely bound by the nuclear matrix (16, 163). Studies also indicate (16) that estrogen and antiestrogen may bind to separate sites in the nucleus.
In assessing the relevance of these studies, it should complexes are only loosely bound by the nuclear matrix (16, 163). Studies also indicate (16) that estrogen and antiestrogen may bind to separate sites in the nucleus.
In assessing the relevance of these studies, it should (16, 163). Studies also indicate (16) that estrogen and antiestrogen may bind to separate sites in the nucleus.
In assessing the relevance of these studies, it should be pointed out that the estrogenic and antiestrogenic antiestrogen may bind to separate sites in the nucleus.
In assessing the relevance of these studies, it should be
pointed out that the estrogenic and antiestrogenic geo-
metric isomers of clomiphene can be salt extracted f In assessing the relevance of these studies, it should be
pointed out that the estrogenic and antiestrogenic geo-
metric isomers of clomiphene can be salt extracted from
nuclei to the same extent (270). It is possible thou pointed out that the estrogenic and antiestrogenic geometric isomers of clomiphene can be salt extracted from
nuclei to the same extent (270). It is possible though that
the low affinity ligand dissociates from the complex metric isomers of clomiphene can be salt extracted from
nuclei to the same extent (270). It is possible though that
the low affinity ligand dissociates from the complex and
the unoccupied receptor readily extracts. Thus on nuclei to the same extent (270). It is possible though that
the low affinity ligand dissociates from the complex and
the unoccupied receptor readily extracts. Thus one effect
(as previously suggested in section VI) that ai the low affinity ligand dissociates from the complex and
the unoccupied receptor readily extracts. Thus one effect
(as previously suggested in section VI) that aids nuclear
extraction would be ligand dissociation rather th (as previously suggested in section VI) that aids nuclear extraction would be ligand dissociation rather than the physicochemical properties of the complex. Ruh and Ruh (272) have shown, however, that the high affinity ant (as previously suggested in section VI) that aids nuclear
extraction would be ligand dissociation rather than the
physicochemical properties of the complex. Ruh and Ruh
(272) have shown, however, that the high affinity ant extraction would be ligand dissociation rather than the
physicochemical properties of the complex. Ruh and Ruh
(272) have shown, however, that the high affinity anties-
trogen H1285-estrogen receptor complex is readily exphysicochemical properties of the complex. Ruh and R
(272) have shown, however, that the high affinity antie
trogen H1285-estrogen receptor complex is readily e
tracted from nuclei, indicating that nuclear estrogen a
antie (272) have shown, however, that the high affinit trogen H1285-estrogen receptor complex is retracted from nuclei, indicating that nuclear estrantiestrogen receptor complexes may indeed has ent binding characteristics withi ogen H1285-estrogen receptor complex is readily ex-
acted from nuclei, indicating that nuclear estrogen and
tiestrogen receptor complexes may indeed have differ-
t binding characteristics within the nucleus.
Antiestrogen a

tracted from nuclei, indicating that nuclear estrogen an antiestrogen receptor complexes may indeed have different binding characteristics within the nucleus.
Antiestrogen-action in the uterus is based upon thinhibition of antiestrogen receptor complexes may indeed have different binding characteristics within the nucleus.
Antiestrogen action in the uterus is based upon the inhibition of estrogen-stimulated uterine growth. However, the situa ent binding characteristics within the nucleus.
Antiestrogen action in the uterus is based upon the
inhibition of estrogen-stimulated uterine growth. How-
ever, the situation is complicated by the ability of an-
tiestrogen Antiestrogen action in the uterus is based upon the
inhibition of estrogen-stimulated uterine growth. How-
ever, the situation is complicated by the ability of an-
tiestrogens to initiate progesterone receptor synthesis
(6 inhibition of estrogen-stimulated uterine growth. However, the situation is complicated by the ability of i tiestrogens to initiate progesterone receptor synthe (68, 191, 201) which is considered to be an estrogener res ever, the situation is complicated by the ability of antiestrogens to initiate progesterone receptor synthesis (68, 191, 201) which is considered to be an estrogenic response (96). It is this mixture of estrogenic and anti tiestrogens to initiate progesterone receptor synthesis (68, 191, 201) which is considered to be an estrogenic response (96). It is this mixture of estrogenic and antiestrogenic effects, produced by the triphenylethylene t (68, 191, 201) which is considered to be an ϵ response (96). It is this mixture of estrogenic at trogenic effects, produced by the triphenylethy of antiestrogens in vivo, that makes a unifying for antiestrogen action i *B. Studies in Vitro*
B. Studies in Vitro
B. Studies in Vitro
Much of the research

of antiestrogens in vivo, that makes a unifying theory
for antiestrogen action in vivo rather difficult.
B. Studies in Vitro
Much of the research on the subcellular aspects of
antiestrogen action has focused upon human bre for antiestrogen action in vivo rather difficult.

B. Studies in Vitro

Much of the research on the subcellular aspects of

antiestrogen action has focused upon human breast can-

cer cell lines. The most widely studied, t B. Studies in Vitro
Much of the research on the subcellular aspects of
antiestrogen action has focused upon human breast can-
cer cell lines. The most widely studied, the MCF-7 cell
line, was cultivated from a pleural effu B. Studies in Vitro

Much of the research on the subcellular aspects of

antiestrogen action has focused upon human breast can-

cer cell lines. The most widely studied, the MCF-7 cell

line, was cultivated from a pleural Much of the research on the subcellular aspects of
antiestrogen action has focused upon human breast can-
cer cell lines. The most widely studied, the MCF-7 cell
line, was cultivated from a pleural effusion derived from
a line, was cultivated from a pleural effusion derived from
a patient with breast cancer (288). Estrogen, androgen,
progesterone, and glucocorticoid receptors are all present
in MCF-7 cells (133) and estrogen increases cell cer cell lines. The most widely studied, the MCF-7 cell
line, was cultivated from a pleural effusion derived from
a patient with breast cancer (288). Estrogen, androgen,
progesterone, and glucocorticoid receptors are all p ine, was cultivated from a pieural effusion derived from
a patient with breast cancer (288). Estrogen, androgen,
progesterone, and glucocorticoid receptors are all present
in MCF-7 cells (133) and estrogen increases cell n progesterone, and glucocorticoid receptors are all present
in MCF-7 cells (133) and estrogen increases cell numbers
(184, 217). Following the localization of the estradiol-ER
complex in the nucleus, a rapid destruction or in MCF-7 cells (133) and estrogen increases cell numbers (184, 217). Following the localization of the estradiol-ER complex in the nucleus, a rapid destruction or processing has been described (135, 137) and correlated wit (184, 217). Following the localization of the estradiol-ER complex in the nucleus, a rapid destruction or processing has been described (135, 137) and correlated with the subsequent appearance of cytoplasmic progesterone complex in the nucleus, a rapid destruction or processing
has been described (135, 137) and correlated with the
subsequent appearance of cytoplasmic progesterone re-
ceptor. Of interest is the observation that nafoxidine
r has been described (135, 137) and correlated with
subsequent appearance of cytoplasmic progesterone
ceptor. Of interest is the observation that nafoxid
receptor complexes are not processed and do not prove
the appearance o subsequent appearance of cytoplasmic progesterone
ceptor. Of interest is the observation that nafoxid
receptor complexes are not processed and do not prove
the appearance of progesterone receptor (134). Tamo
fen seems to f ceptor. Of interest is the observation that nafoxidiveceptor complexes are not processed and do not provo
the appearance of progesterone receptor (134). Tamo:
fen seems to fall into an intermediate category. Tamo
ifen rece receptor complexes are not processed and do not prove the appearance of progesterone receptor (134). Tam
fen seems to fall into an intermediate category. Tan
ifen receptor complexes undergo processing and stilate progester the appearance of progesterone receptor (134). Tamoxi-
fen seems to fall into an intermediate category. Tamox-
ifen receptor complexes undergo processing and stimu-
late progesterone receptor appearance at low concentra-
t fen seems to fall into an intermediate category. Tamoxien receptor complexes undergo processing and stimulate progesterone receptor appearance at low concentrations of the antiestrogens, but at high concentrations all grow ifen receptor complexes undergo processing and stimu-
late progesterone receptor appearance at low concentra-
tions of the antiestrogens, but at high concentrations all
growth activities cease (134). The concept of process late progesterone receptor appearance at low concentra-
tions of the antiestrogens, but at high concentrations all
growth activities cease (134). The concept of processing
is further complicated though by the finding (76) tions of the antiestrogens, but at high concentrations all growth activities cease (134). The concept of processing is further complicated though by the finding (76) that low concentrations of antiestrogens with high affin is further complicated though by the finding (76) that low concentrations of antiestrogens with high affinity for the estrogen receptor stimulate progesterone receptor synthesis in MCF-7 cells, whereas antiestrogens wit is further complicate
low concentrations of
or the estrogen recep
synthesis in MCF-7 (
low affinity do not.
The essence of "pl w concentrations of antiestrogens with high affinity
r the estrogen receptor stimulate progesterone receptor
mthesis in MCF-7 cells, whereas antiestrogens with a
w affinity do not.
The essence of "processing" is that the a

for the estrogen receptor stimulate progesterone recepty synthesis in MCF-7 cells, whereas antiestrogens with low affinity do not.
The essence of "processing" is that the antiestro receptor complex is resistant to some, as synthesis in MCF-7 cells, whereas antiestrogens with a low affinity do not.
The essence of "processing" is that the antiestrogen
receptor complex is resistant to some, as yet undeter-
mined, biochemical event that destroys low affinity do not.
The essence of "processing" is that the antiestroge
receptor complex is resistant to some, as yet undete
mined, biochemical event that destroys receptor. The
results in an accumulation of receptor comp The essence of "processing" is that the antiestrogen
receptor complex is resistant to some, as yet undeter-
mined, biochemical event that destroys receptor. This
results in an accumulation of receptor complexes. Link-
ing

ANTIESTROGEN PHA
that antiestrogens inhibit receptor resynthesis, one an
would expect there to be a stagnation of the receptor un ANTIESTROGEN 1
that antiestrogens inhibit receptor resynthesis, one
would expect there to be a stagnation of the receptor
dynamics within the nucleus. Recent studies suggest, ANTIESTROGEN
that antiestrogens inhibit receptor resynthesis, one
would expect there to be a stagnation of the receptor
dynamics within the nucleus. Recent studies suggest,
however, a much more complex situation (78). Nucl that antiestrogens inhibit receptor resynthesis, one would expect there to be a stagnation of the receptor dynamics within the nucleus. Recent studies suggest, however, a much more complex situation (78). Nuclear ER synthe that antiestrogens inhibit receptor resynthesis, or would expect there to be a stagnation of the receptodynamics within the nucleus. Recent studies suggesthowever, a much more complex situation (78). Nuclearly ER synthesis would expect there to be a stagnation of the receptor undynamics within the nucleus. Recent studies suggest, to however, a much more complex situation (78). Nuclear ER synthesis and turnover has been measured in MCF- d 7 dynamics within the nucleus. Recent studies suggest, teron
however, a much more complex situation (78). Nuclear Then
ER synthesis and turnover has been measured in MCF- duce
7 breast cancer cells by a density shift techni however, a much more complex situation (78). Nuclear
ER synthesis and turnover has been measured in MCF-
7 breast cancer cells by a density shift technique. Cells
are incubated in medium supplemented with ¹³C, ¹⁵N,² ER synthesis and turnover has been measured in MCF-
7 breast cancer cells by a density shift technique. Cells via
are incubated in medium supplemented with ^{13}C , ^{15}N , ^{2}H co
amino acids (dense amino acids) and a are incubated in medium supplemented with ${}^{13}C, {}^{16}N, {}^{2}H$ comino acids (dense amino acids) and a shift is monitored the from "old light" (pre-existing) to "new dense" (newly alsynthesized) receptors by velocity sedi amino acids (dense amino acids) and a shift is monitored this from "old light" (pre-existing) to "new dense" (newly also synthesized) receptors by velocity sedimentation on 0.4 ma MKCl 5% to 20% sucrose gradients prepared from "old light" (pre-existing) to "new dense" (newly synthesized) receptors by velocity sedimentation on 0.4 M KCl 5% to 20% sucrose gradients prepared in buffered deuterium oxide. The unoccupied nuclear receptor was fou synthesized) receptors by velocity sedimentation on 0.4
M KCl 5% to 20% sucrose gradients prepared in buffered
deuterium oxide. The unoccupied nuclear receptor was
found to have a half-life of 4.47 \pm 0.26 hr, but nucle M KCl 5% to 20% sucrose gradients prepared in buffered torial deuterium oxide. The unoccupied nuclear receptor was "an found to have a half-life of 4.47 ± 0.26 hr, but nuclear receptors occupied with estradiol, CI628 or deuterium oxide. The unoccupied nuclear receptor was
found to have a half-life of 4.47 ± 0.26 hr, but nuclear
receptors occupied with estradiol, CI628 or nafoxidine
were found to have half-lives of 3.00 ± 0.38 hr, $4.$ found to have a half-life of 4.47 ± 0.26 hr, but nuclear receptors occupied with estradiol, CI628 or nafoxidine of were found to have half-lives of 3.00 ± 0.38 hr, 4.9 ± 0.66 at hr, and 3.43 ± 0.37 hr, respectivel receptors occupied with estradiol, CI628 or nafoxidine of were found to have half-lives of 3.00 ± 0.38 hr, 4.9 ± 0.66 appears, and 3.43 ± 0.37 hr, respectively. Clearly, the receptor the is turned over in the presen were found to have half-li
hr, and 3.43 ± 0.37 hr, reis
turned over in the pilgands and there appear
estrogens or antiestrogen
Antiestrogens produce %, and 3.43 ± 0.37 hr, respectively. Clearly, the receptor tain turned over in the presence or absence of binding shands and there appears to be no distinction between potogens or antiestrogens.
Antiestrogens produce in is turned over in the presence or absence of binding
ligands and there appears to be no distinction between
estrogens or antiestrogens.
Antiestrogens produce inconsistent effects with regard
to progesterone receptor synthe

ligands and there appears to be no distinction betwee
estrogens or antiestrogens.
Antiestrogens produce inconsistent effects with regate
to progesterone receptor synthesis, but several other
intracellular or secreted prote estrogens or antiestrogens.
Antiestrogens produce inconsistent effects with regator progesterone receptor synthesis, but several oth
intracellular or secreted proteins are regulated by estrogens and antiestrogens. Estrogen Antiestrogens produce inconsistent effects with regard to progesterone receptor synthesis, but several other tion
intracellular or secreted proteins are regulated by estro-
gens and antiestrogens. Estrogen stimulates the s to progesterone receptor synthesis, but several other
intracellular or secreted proteins are regulated by estro-
gens and antiestrogens. Estrogen stimulates the secre-
tion of a 52 K dalton protein (originally identified intracellular or secreted proteins are regulated by estro-
gens and antiestrogens. Estrogen stimulates the secre-
tion of a 52 K dalton protein (originally identified as a
46 K dalton protein) from MCF-7 cells (320). Tamox gens and antiestrogens. Estrogen stimulates the secretion of a 52 K dalton protein (originally identified as a 46 K dalton protein) from MCF-7 cells (320). Tamoxifen and 4-hydroxytamoxifen prevent estrogen-stimulated synth tion of a 52 K dalton protein (originally identified a 46 K dalton protein) from MCF-7 cells (320). Tamoxii and 4-hydroxytamoxifen prevent estrogen-stimulates synthesis of the 52 K dalton protein and have no agon activity 46 K dalton protein) from MCF-7 cells (320). Tamoxifen
and 4-hydroxytamoxifen prevent estrogen-stimulated
synthesis of the 52 K dalton protein and have no agonist
activity (321). Similarly, estrogen stimulates the synthe-
 and 4-hydroxytamoxifen prevent estrogen-stimulated
synthesis of the 52 K dalton protein and have no agonist
activity (321). Similarly, estrogen stimulates the synthe-
sis of specific, intracellular, proteins with molecula synthesis of the 52 K dalton protein and have no agonist
activity (321). Similarly, estrogen stimulates the synthe-
sis of specific, intracellular, proteins with molecular
weights of 24 K and 36 K whereas nafoxidine is in activity (321). Similarly, estrogen stimulates the syntais of specific, intracellular, proteins with molect weights of 24 K and 36 K whereas nafoxidine is inaction.
(80). The function of each of the intracellular or secrep weights of 24 K and 36 K whereas nafoxidine is inactive (80). The function of each of the intracellular or secreted proteins is unknown, but research is aimed at establishing a link between their synthesis and estroge proteins is unknown, but research is aimed at establishing a link between their synthesis and estrogen-stimulated cell division.
Tamoxifen and nafoxidine alone produce an inhibitory effect on $\binom{3}{1}$ thymidine incorpora The function of each of the intracellular or secreted

(b). The function of each of the intracellular or secreted

oteins is unknown, but research is aimed at establish-

g a link between their synthesis and estrogen-stim

proteins is unknown, but research is aimed at establish-
ing a link between their synthesis and estrogen-stimu-
lated cell division.
Tamoxifen and nafoxidine alone produce an inhibitory
effect on [³H]thymidine incorporat ing a link between their synthesis and estrogen-stimu-
lated cell division.
Tamoxifen and nafoxidine alone produce an inhibitory
effect on [³H]thymidine incorporation (217) and DNA
polymerase activity (82) as well as cau lated cell division.

Tamoxifen and nafoxidine alone produce an inhibitory

effect on [³H]thymidine incorporation (217) and DNA

polymerase activity (82) as well as causing a reduction

in DNA content of cultures (61) an Tamoxifen and nafoxidine alone produce an inhibitory
effect on $[^3H]$ thymidine incorporation (217) and DNA
polymerase activity (82) as well as causing a reduction
in DNA content of cultures (61) and cell numbers (61,
217) effect on [³H]thymidine incorporation (217) and DNA
polymerase activity (82) as well as causing a reduction
in DNA content of cultures (61) and cell numbers (61,
217). This inhibitory effect on MCF-7 cell growth is not
a polymerase activity (82) as well as causing a reduction
in DNA content of cultures (61) and cell numbers (61,
217). This inhibitory effect on MCF-7 cell growth is not
a simple cytotoxic action of the drug since it can be
r in DNA content of cultures (61) and cell numbers (61, 217). This inhibitory effect on MCF-7 cell growth is not a simple cytotoxic action of the drug since it can be readily reversed by addition of estradiol to the culture 217). This inhibitory effect on MCF-7 cell growth is not
a simple cytotoxic action of the drug since it can be
readily reversed by addition of estradiol to the culture
media (216). Studies of cell cycle kinetics show that a simple cytotoxic action of the drug since it can be readily reversed by addition of estradiol to the culture media (216). Studies of cell cycle kinetics show that at concentrations of 2 to 6 μ M in the culture, tamoxi readily reversed by addition of estradiol to the culture
media (216). Studies of cell cycle kinetics show that at
concentrations of 2 to 6 μ M in the culture, tamoxifen
reduces the proportion of cells in S phase and inc media (216). Studies of cell cycle kinetics show that at concentrations of 2 to 6 μ M in the culture, tamoxifen reduces the proportion of cells in S phase and increases the number of cells in G₁ (242, 293). At 10 μ concentrations of 2 to 6 μ M in the culture, tamoxifen
reduces the proportion of cells in S phase and increases
the number of cells in G₁ (242, 293). At 10 μ M, tamoxifen
causes cell death within 48 hr. Similar inhi reduces the proportion of cells in S phase and increase the number of cells in G_1 (242, 293). At 10 μ M, tamoxicauses cell death within 48 hr. Similar inhibitory effectof tamoxifen on [³H]thymidine incorporation an the number of cells in G_1 (242, 293). At 10 μ M, tamoxifen
causes cell death within 48 hr. Similar inhibitory effects
of tamoxifen on [³H]thymidine incorporation and/or cell
numbers have been described for two othe causes cell death within 48 hr. Similar inhibitory effects
of tamoxifen on [³H]thymidine incorporation and/or cell
numbers have been described for two other estrogen-
responsive cell lines. CG-5 is a variant of MCF-7 whi of tamoxifen on [³H]thymidine incorporation and/or cell
numbers have been described for two other estrogen-
responsive cell lines. CG-5 is a variant of MCF-7 which
is claimed to be highly sensitive to estrogen; tamoxifen numbers have been described for two other estrogen-
responsive cell lines. CG-5 is a variant of MCF-7 which
is claimed to be highly sensitive to estrogen; tamoxifen
not only inhibits the growth of CG-5 cells but also
poten responsive cell lines. CG-5 is a variant of MCF-7 which
is claimed to be highly sensitive to estrogen; tamoxifen
not only inhibits the growth of CG-5 cells but also
potentiates the inhibitory effects of progestins (141). T is claimed to be highly sensitive to estrogen; tamoxifen
not only inhibits the growth of CG-5 cells but also
potentiates the inhibitory effects of progestins (141). The
ZR-75-1 cell line has the advantage that it grows in
 not only inhibits the growth of CG-5 cells but also
potentiates the inhibitory effects of progestins (141). The
ZR-75-1 cell line has the advantage that it grows in
defined media. In the absence of estradiol, tamoxifen
cau ZR-75-1 cell line has the advantage that it grows in defined media. In the absence of estradiol, tamoxifen causes cell death, an effect that can be reversed by estrogen provided it is not later than 48 hr after the

HARMACOLOGY 257
antiestrogen (7, 8). It is also interesting to note that,
unlike MCF-7 cells, tamoxifen has no effect on proges-HARMACOLOGY

antiestrogen (7, 8). It is also interesting to note the

unlike MCF-7 cells, tamoxifen has no effect on prog

terone receptor synthesis in the ZR-75-1 cell line (6). 25'
antiestrogen (7, 8). It is also interesting to note that
unlike MCF-7 cells, tamoxifen has no effect on proges
terone receptor synthesis in the ZR-75-1 cell line (6).
There is reasonable evidence that antiestrogens pro tiestrogen (7, 8). It is also interesting to note that, like MCF-7 cells, tamoxifen has no effect on proges-
rone receptor synthesis in the ZR-75-1 cell line (6).
There is reasonable evidence that antiestrogens pro-
cce th

7 breast cancer cells by a density shift technique. Cells via an estrogen receptor mechanism. However, very high are incubated in medium supplemented with ^{13}C , ^{15}N , ^{2}H concentrations of antiestrogens inhibit c antiestrogen $(7, 8)$. It is also interesting to note that,
unlike MCF-7 cells, tamoxifen has no effect on proges-
terone receptor synthesis in the ZR-75-1 cell line (6) .
There is reasonable evidence that antiestrogens unlike MCF-7 cells, tamoxifen has no effect on proges-
terone receptor synthesis in the ZR-75-1 cell line (6).
There is reasonable evidence that antiestrogens pro-
duce their inhibitory effects on growth in the cell lines
 terone receptor synthesis in the ZR-75-1 cell line (6).
There is reasonable evidence that antiestrogens pro-
duce their inhibitory effects on growth in the cell lines
via an estrogen receptor mechanism. However, very high
 There is reasonable evidence that antiestrogens pro-
duce their inhibitory effects on growth in the cell lines
via an estrogen receptor mechanism. However, very high
concentrations of antiestrogens inhibit cell growth and
 duce their inhibitory effects on growth in the cell lines
via an estrogen receptor mechanism. However, very high
concentrations of antiestrogens inhibit cell growth and
this cannot be "reversed" with estrogen (115). There this cannot be "reversed" with estrogen (115) . There is concentrations of antiestrogens inhibit cell growth and
this cannot be "reversed" with estrogen (115). There is
also some evidence that tamoxifen will inhibit growth of
mammary cancer cells that do not have estrogen recepthis cannot be "reversed" with estrogen (115). There is
also some evidence that tamoxifen will inhibit growth of
mammary cancer cells that do not have estrogen recep-
tors (115). While this effect could be modulated via a also some evidence that tamoxifen will inhibit growth of
mammary cancer cells that do not have estrogen recep-
tors (115). While this effect could be modulated via an
"antiestrogen binding protein" (298), the concentratio mammary cancer cells that do not have estrogen receptors (115). While this effect could be modulated via an "antiestrogen binding protein" (298), the concentrations required to produce an effect ($>7.5 \mu$ M) and the affini tors (115). While this effect could be modulated via "antiestrogen binding protein" (298), the concentration required to produce an effect (>7.5 μ M) and the affine of tamoxifen for the binding site (1 nM) seems to argu "antiestrogen binding protein" (298), the concentrations
required to produce an effect $(>7.5 \mu M)$ and the affinity
of tamoxifen for the binding site (1 nM) seems to argue
against this correlation. In this regard it m against this correlation. In this regard it may be important to consider the recent report by Lam (199) who has tant to consider the recent report by Lam (199) who shown that tamoxifen, in the 1 to 10 μ M range, ipotent inhibitor of calmodulin action. Since calmode has been implicated in the control of cell proliferat (43, 143), tion. motion of california action. Since complicated in the control of cell problems, these observations warrant further
VIII. Radiolabeled Antiestrogen
l nonsteroidal antiestrogens have been

of tamoxifen for the binding site (1 nM) seems to argue

against this correlation. In this regard it may be impor-

tant to consider the recent report by Lam (199) who has

shown that tamoxifen, in the 1 to 10 μ M range S. 143), these observations warrant further investion.

WIII. Radiolabeled Antiestrogens

Several nonsteroidal antiestrogens have been synthed in tritium-labeled form to aid in an understand tion.

VIII. Radiolabeled Antiestrogens

Several nonsteroidal antiestrogens have been synthe-

sized in tritium-labeled form to aid in an understanding

of their metabolism and binding characteristics to ER. VIII. Radiolabeled Antiestrogens
Several nonsteroidal antiestrogens have been synthe-
sized in tritium-labeled form to aid in an understanding
of their metabolism and binding characteristics to ER.
The compounds that are n VIII. Kadiolabeled Antiestrogens
Several nonsteroidal antiestrogens have been synthe-
sized in tritium-labeled form to aid in an understanding
of their metabolism and binding characteristics to ER.
The compounds that are n Several nonsteroidal antiestrogens have been synthe-
sized in tritium-labeled form to aid in an understanding
of their metabolism and binding characteristics to ER.
The compounds that are now commercially available are
ill sized in tritium-labeled form to aid in an understanding
of their metabolism and binding characteristics to ER.
The compounds that are now commercially available are
illustrated in figure 13; however, several compounds hav The compounds that are now commercially available are
illustrated in figure 13; however, several compounds have
been synthesized by individual investigators. In Dr. John
A. Katzenellenbogen's laboratory, tritium-labeled illustrated in figure 13; however, several compounds have *trans* isomers of tamoxifen and 4-hydroxytamoxifen

FIG. 13. Commercially available radiolabeled antiestrogens.

Downloaded from pharmrev.aspetjournals.org at Thammasart University on December 8, 2012

(252), and tamoxifen aziridine (184) were successfully binding of the ligands. The model illustrated in figure 1
synthesized and in Dr. Thomas S. Ruh's laboratory, has been proposed to explain these observations (301). Jo
(252), and tamoxifen aziridine (184) were successfully
synthesized and in Dr. Thomas S. Ruh's laboratory
tritium-labeled H1285 was synthesized (271, 272). Al-10R

(252), and tamoxifen aziridine (184) were successfully

synthesized and in Dr. Thomas S. Ruh's laboratory,

tritium-labeled H1285 was synthesized (271, 272). Al-

though most [³H]antiestrogens have been shown to be (252), and tamoxifen aziridine (184) were successfully bin
synthesized and in Dr. Thomas S. Ruh's laboratory, has
tritium-labeled H1285 was synthesized (271, 272). Al-
though most [³H]antiestrogens have been shown to be (252), and tamoxifen aziridine (184) were successfully synthesized and in Dr. Thomas S. Ruh's laboratory, tritium-labeled H1285 was synthesized (271, 272). Although most [³H]antiestrogens have been shown to be metabolic synthesized and in Dr. Thomas S. Ruh's laboratritium-labeled H1285 was synthesized (271, 272) though most [³H]antiestrogens have been shown metabolically activated to hydroxylated derivative fore binding in target tissue tritium-labeled H1285 was synthesized $(271, 272)$. Although most [³H]antiestrogens have been shown to be site metabolically activated to hydroxylated derivatives be-
fore binding in target tissues $(30, 181)$, [³H]4though most
metabolicall
fore binding
tamoxifen bi
vivo (158).
The studio etabolically activated to hydroxylated derivatives
re binding in target tissues (30, 181), [³H]4-hydro
moxifen binds directly to rat estrogen target tissues
vo (158).
The studies that describe the interaction of tritiu
b fore binding in target tissues $(30, 181)$, $[^{3}H]4$ -hydrotamoxifen binds directly to rat estrogen target tissue
vivo (158) .
The studies that describe the interaction of tritic
labeled antiestrogens with the ER and oth

tamoxifen binds directly to rat estrogen tar,
vivo (158).
The studies that describe the interaction
labeled antiestrogens with the ER and oth
gen receptor binding sites will be reviewed.
A. The Estrogen Receptor vivo (158).

The studies that describe the interaction of tritium-

labeled antiestrogens with the ER and other, nonestro-

gen receptor binding sites will be reviewed.

A. The Estrogen Receptor

The studies with [³H]tam beled antiestrogens with the ER and other, nonestro-
n receptor binding sites will be reviewed.
The Estrogen Receptor
The studies with [³H]tamoxifen (40, 173) and [³H] c
628 (181) demonstrate estrogen-specific binding

gen receptor binding sites will be reviewed.

A. The Estrogen Receptor

The studies with $[^{3}H]$ tamoxifen (40, 173) and $[^{3}H]$

CI628 (181) demonstrate estrogen-specific binding of

antiestrogens to the 8S estrogen rec A. The Estrogen Receptor
The studies with $[^{3}H]$ tamoxifen (40, 173) and $[^{3}H]$
CI628 (181) demonstrate estrogen-specific binding of
antiestrogens to the 8S estrogen receptor from immature
rat uteri. The antiestrogens A. The Estrogen Receptor

The studies with [³H] tamoxifen (40, 173) and [³H] can

CI628 (181) demonstrate estrogen-specific binding of

antiestrogens to the 8S estrogen receptor from immature

rat uteri. The antiestro The studies with [³H]tamoxifen (40, 173) and [³H] cl628 (181) demonstrate estrogen-specific binding of antiestrogens to the 8S estrogen receptor from immature rat uteri. The antiestrogens tamoxifen and Cl628 have a ra CI628 (181) demonstrate estrogen-specific binding of recentiestrogens to the 8S estrogen receptor from immature rat uteri. The antiestrogens tamoxifen and CI628 have a rapid rate of dissociation from the receptor (40, 181 antiestrogens to the 8S estrogen receptor from immature
rat uteri. The antiestrogens tamoxifen and CI628 have a
rapid rate of dissociation from the receptor (40, 181) so
that accurate studies of the interaction of tamoxif rat uteri. The antiestrogens tamoxifen and CI628 have rapid rate of dissociation from the receptor (40, 181) that accurate studies of the interaction of tamoxifen a CI628 with the ER are particularly difficult. Borgna a R rapid rate of dissociation from the receptor (40, 181)
that accurate studies of the interaction of tamoxifen a
CI628 with the ER are particularly difficult. Borgna a
Rochefort (29) prepared the first sample of $[^{3}H]4-h$
 that accurate studies of the interaction of tamoxifen and
CI628 with the ER are particularly difficult. Borgna and
Rochefort (29) prepared the first sample of $[^{3}H]4-hy-$
droxytamoxifen by the metabolic oxidation of $[^{3$ CI628 with the ER are particularly difficult. Borg
Rochefort (29) prepared the first sample of [³H
droxytamoxifen by the metabolic oxidation of [
moxifen in chicken liver slices. This material wa
to confirm the high bind Rochefort (29) prepared the first sample of $[^{3}H]4-hy-$
droxytamoxifen by the metabolic oxidation of $[^{3}H]$ ta-
moxifen in chicken liver slices. This material was used
to confirm the high binding affinity of 4-hydroxyta moxifen in chicken liver slices. This material was used
to confirm the high binding affinity of 4-hydroxytamox-
ifen for the ER and describe the physical properties of
estradiol and 4-hydroxytamoxifen ER complexes (29). A to confirm the high binding affinity of 4-hydroxytamoxifen for the ER and describe the physical properties of estradiol and 4-hydroxytamoxifen ER complexes (29). A similar study has been reported with the compound CI628M (ifen for the ER and describe the physical properties of
estradiol and 4-hydroxytamoxifen ER complexes (29). A
similar study has been reported with the compound
CI628M (183). There are, however, several recent reports
 $(77$ estradiol and 4-hydroxytamoxifen ER complexes (29
similar study has been reported with the compo
CI628M (183). There are, however, several recent rep
(77, 90, 260, 301) that describe specific differences in
physicochemical similar study has been reported with the compoure CI628M (183). There are, however, several recent report (77, 90, 260, 301) that describe specific differences in the physicochemical properties of estradiol and 4-hydrox ta CI628M (183). There are, however, several recent reports

(77, 90, 260, 301) that describe specific differences in the

physicochemical properties of estradiol and 4-hydroxy-

tamoxifen ER complexes. By documenting these (77, 90, 260, 301) that describe specific differences in physicochemical properties of estradiol and 4 -hydro tamoxifen ER complexes. By documenting these difences, it is hoped that changes in the receptor composity ulti physicochemical properties of estradiol and 4-hydroxy-
tamoxifen ER complexes. By documenting these differ-
ences, it is hoped that changes in the receptor complex
might ultimately explain the differences in the pharma-
co moxifen ER complexes. By documenting these differ-
ces, it is hoped that changes in the receptor complex
ight ultimately explain the differences in the pharma-
logy of agonists and antagonists.
Estradiol rapidly dissociate

ences, it is hoped that changes in the receptor complex
might ultimately explain the differences in the pharma-
cology of agonists and antagonists.
Estradiol rapidly dissociates from the untransformed
receptor, whereas the might ultimately explain the differences in the pharma-
cology of agonists and antagonists.
Estradiol rapidly dissociates from the untransformed
receptor, whereas the dissociation is slowed from the
heat transformed recep cology of agonists and antagonists.

Estradiol rapidly dissociates from the untransformed

receptor, whereas the dissociation is slowed from the

heat transformed receptor. Molybdate ions appear to

prevent the transformat Estradiol rapidly dissociates from the untransformed
receptor, whereas the dissociation is slowed from the
heat transformed receptor. Molybdate ions appear to
prevent the transformation of the estradiol-ER complex
and est receptor, whereas the dissociation is slowed from the
heat transformed receptor. Molybdate ions appear to
prevent the transformation of the estradiol-ER complex
and estradiol rapidly dissociates from the receptor in the
pr heat transformed receptor. Molybdate ions appear to
prevent the transformation of the estradiol-ER complex
and estradiol rapidly dissociates from the receptor in the
presence of molybdate. In contrast, 4-hydroxytamoxifen
d prevent the transformation of the estradiol-ER complex
and estradiol rapidly dissociates from the receptor in the
presence of molybdate. In contrast, 4-hydroxytamoxifer
dissociates slowly from both the transformed and un-
 and estradiol rapidly dissociates from the receptor in the presence of molybdate. In contrast, 4-hydroxytamoxifen
dissociates slowly from both the transformed and un-
transformed receptor and is unaffected by molybdate.
Ro presence of molybdate. In contrast, 4-hydroxytamoxifen
dissociates slowly from both the transformed and un-
transformed receptor and is unaffected by molybdate.
Rochefort and Borgna (260) suggest that antiestrogens
produce dissociates slowly from both the transformed and untransformed receptor and is unaffected by molybdate.
Rochefort and Borgna (260) suggest that antiestrogens bethered a change in the ER discretely different than that with transformed receptor and is unaffected by molybdate.
Rochefort and Borgna (260) suggest that antiestrogens
produce a change in the ER discretely different than that
produced by estradiol. A similar conclusion has been
reac Rochefort and Borgna (260) suggest that antiestrogens produce a change in the ER discretely different than that we produced by estradiol. A similar conclusion has been reached as a result of studies to describe the intera produce a change in the ER discretely different than t
produced by estradiol. A similar conclusion has b
reached as a result of studies to describe the interact
of human breast tumor ER with a polyclonal antib
raised to th produced by estradiol. A similar conclusion has be
reached as a result of studies to describe the interaction
of human breast tumor ER with a polyclonal antibor
raised to the calf uterine ER in the goat (113). Preinc
batio reached as a result of studies to describe the interaction of human breast tumor ER with a polyclonal antibody D
raised to the calf uterine ER in the goat (113). Preincu-
bation of antibody with cystolic ER impairs the su of human breast tumor ER with a polyclonal antibody Dl
raised to the calf uterine ER in the goat (113). Preincu-
bation of antibody with cystolic ER impairs the subse-
alt
quent binding of [³H]estradiol and reduces the a raised to the calf uterine ER in the goat (113) . Preincu-
bation of antibody with cystolic ER impairs the subse-
quent binding of $[^{3}H]$ estradiol and reduces the affinity
of the ligand-receptor protein interaction. Ho bation of antibody with cystolic ER impairs the subsequent binding of [³H]estradiol and reduces the affinity of the ligand-receptor protein interaction. However, the binding of 4-hydroxytamoxifen to the receptor is unimquent binding of [³H]estradiol and reduces the affinity of the ligand-receptor protein interaction. However, the binding of 4-hydroxytamoxifen to the receptor is unim-
paired by equivalent concentrations of the antibody. of the ligand-receptor protein interaction. However, the binding of 4-hydroxytamoxifen to the receptor is unim-
paired by equivalent concentrations of the antibody. two Higher concentrations of antibody can, however, sub-
 binding of 4-hydroxytamoxifen to the receptor is unim-
paired by equivalent concentrations of the antibody. twe
Higher concentrations of antibody can, however, sub-
but stantially inhibit antiestrogen binding. In contrast,

binding of the ligands. The model illustrated in figure 14 AN
binding of the ligands. The model illustrated in figure 14
has been proposed to explain these observations (301).
Estradiol initially interacts with the ligand binding

I
Inding of the ligands. The model illustrated in figure 14
Is been proposed to explain these observations (301).
Estradiol initially interacts with the ligand binding
I ee on the receptor which then induces an activation binding of the ligands. The model illustrated in figure 14
has been proposed to explain these observations (301).
Estradiol initially interacts with the ligand binding
site on the receptor which then induces an activation binding of the ligands. The model illustrated in figure 14
has been proposed to explain these observations (301).
Estradiol initially interacts with the ligand binding
site on the receptor which then induces an activation site on the receptor which then induces an activation or transformation reaction that locks the steroid into the receptor complex. The antiestrogen, because of its shape, wedges into the binding site on the receptor and pr Estradiol initially interacts with the ligand binding
site on the receptor which then induces an activation or
transformation reaction that locks the steroid into the
receptor complex. The antiestrogen, because of its shap site on the receptor which then induces an activation or
transformation reaction that locks the steroid into the
receptor complex. The antiestrogen, because of its shape,
wedges into the binding site on the receptor and pr transformation reaction that locks the steroid into the receptor complex. The antiestrogen, because of its shap wedges into the binding site on the receptor and prevent the full range of conformational changes required for receptor complex. The antiestrogen, because of its shape,
wedges into the binding site on the receptor and prevents
the full range of conformational changes required for
receptor activation. The polyclonal antibody may int wedges into the binding site on the receptor and prevents
the full range of conformational changes required for
receptor activation. The polyclonal antibody may inter-
act with the unfilled or resting receptor to prevent t the full range of conformational changes required for
receptor activation. The polyclonal antibody may inter-
act with the unfilled or resting receptor to prevent the
conformational changes that subsequently occur to lock
 receptor activation. The polyclonal antibody may inter-
act with the unfilled or resting receptor to prevent the
conformational changes that subsequently occur to lock
estradiol into the receptor. However, the antiestrogen act with the unfilled or resting receptor to prevent the conformational changes that subsequently occur to lock estradiol into the receptor. However, the antiestrogen can still wedge into the proposed binding site, but bec conformational changes that subsequently occur to loot estradiol into the receptor. However, the antiestroge can still wedge into the proposed binding site, but b cause the ligand has a multipoint attachment to the recepto estradiol into the receptor. However, the antiestrogen can still wedge into the proposed binding site, but be-
cause the ligand has a multipoint attachment to the
receptor, it does not require further conformational
change cause the ligand has a multipoint attachment to the receptor, it does not require further conformational changes to produce high affinity binding. It is suggested that once the conformational changes occur to lock the ster cause the ligand has a multipoint attachment to the receptor, it does not require further conformational changes to produce high affinity binding. It is suggested that once the conformational changes occur to lock the ster receptor, it does n
changes to produce l
that once the confor
steroid into the rece
reverse the process.
Monoclonal antib anges to produce high affinity binding. It is suggested
at once the conformational changes occur to lock the
proid into the receptor then the antibody is unable to
verse the process.
Monoclonal antibodies raised to the ER

Extra different (25) prepared the first sample of $[11]$ +-hy⁻ known to interact at different sites on the protein. Androxytamoxifen by the metabolic oxidation of $[{}^{3}H]$ ta-
moxifen in chicken liver slices. This materi that once the conformational changes occur to lock the
steroid into the receptor then the antibody is unable to
reverse the process.
Monoclonal antibodies raised to the ER (114) are
known to interact at different sites on steroid into the receptor then the antibody is unable to
reverse the process.
Monoclonal antibodies raised to the ER (114) are
known to interact at different sites on the protein. An-
tibody D547 (raised to the extranuclea reverse the process.

Monoclonal antibodies raised to the ER (114) are

known to interact at different sites on the protein. An-

tibody D547 (raised to the extranuclear receptor from

MCF-7 cells), which interacts at a s Monoclonal antibodies raised to the ER (114) are
known to interact at different sites on the protein. An-
tibody D547 (raised to the extranuclear receptor from
MCF-7 cells), which interacts at a site far removed from
the l known to interact at different sites on the protein. Antibody D547 (raised to the extranuclear receptor from MCF-7 cells), which interacts at a site far removed from the ligand binding site, binds equally with estradiol an MCF-7 cells), which interacts at a site far removed from
the ligand binding site, binds equally with estradiol and
4-hydroxytamoxifen receptor complexes from human
breast tumor cytosols (300) . There are no differences
w MCF-7 cells), which interacts at a site far removed from
the ligand binding site, binds equally with estradiol and
4-hydroxytamoxifen receptor complexes from human
breast tumor cytosols (300). There are no differences
whet the ligand binding site, binds equally with estradiol and 4-hydroxytamoxifen receptor complexes from human breast tumor cytosols (300). There are no differences whether D547 is preincubated with the receptor before the lig 4-hydroxytamoxifen receptor complexes from huma-
breast tumor cytosols (300). There are no difference-
whether D547 is preincubated with the receptor befor-
the ligand or incubated with the receptor complex. Thu-
the antib breast tumor cytosols (300). There are no differences whether D547 is preincubated with the receptor before the ligand or incubated with the receptor complex. Thus, the antibody D547 does not discriminate between estrogen the antibody D547 does not discriminate between estro-
gen and antiestrogen receptor complexes. The new re-
search tool will, however, be useful for immunohistothe ligand or incubated with the receptor complex. Thus,
the antibody D547 does not discriminate between estro-
gen and antiestrogen receptor complexes. The new re-
search tool will, however, be useful for immunohisto-
che the antibody D547 does not discriminate between estro-
gen and antiestrogen receptor complexes. The new re-
search tool will, however, be useful for immunohisto-
chemical experiments to study the fate of agonist and
antago gen and antiestrogen receptor complexes. The new research tool will, however, be useful for immunohisto-
chemical experiments to study the fate of agonist and
antagonist receptor complexes in tumor cells. In contrast,
the search tool will, however, be useful for immunohisto-
chemical experiments to study the fate of agonist and
antagonist receptor complexes in tumor cells. In contrast,
the monoclonal antibody B36, developed from the calf
ut chemical experiments to study the fate of agonist and
antagonist receptor complexes in tumor cells. In contrast,
the monoclonal antibody B36, developed from the calf
uterine nuclear ER, can apparently discriminate between
 (262). e monoclonal antibody B36, developed from the cali
erine nuclear ER, can apparently discriminate between
tradiol- or 4-hydroxytamoxifen-receptor complexes
62).
Finally, the interaction of estradiol and antiestrogen-
hydrox

uterine nuclear ER, can apparently discriminate between
estradiol- or 4-hydroxytamoxifen-receptor complexes
(262).
Finally, the interaction of estradiol and antiestrogen-
(4-hydroxytamoxifen or CI628M) ER complexes with
ei estradiol- or 4-hydroxytamoxifen-receptor complexes
(262).
Finally, the interaction of estradiol and antiestrogen-
(4-hydroxytamoxifen or CI628M) ER complexes with
either DNA or polynucleotides has been reported by
several (262).
Finally, the interaction of estradiol and antiestrogen-
(4-hydroxytamoxifen or CI628M) ER complexes with
either DNA or polynucleotides has been reported by
several laboratories. Initial studies showed no differences Finally, the interaction of estradiol and antiestrogen-
(4-hydroxytamoxifen or CI628M) ER complexes with
either DNA or polynucleotides has been reported by
several laboratories. Initial studies showed no differences
betwee (4-hydroxytamoxifen or CI628M) ER complexes with
either DNA or polynucleotides has been reported by
several laboratories. Initial studies showed no differences
between the binding of estradiol or 4-hydroxytamoxifen
with p either DNA or polynucleotides has been reported by
several laboratories. Initial studies showed no differences
between the binding of estradiol or 4-hydroxytamoxifen
with polynucleotide-cellulose columns (236) or sheered
c several laboratories. Initial studies showed no differences
between the binding of estradiol or 4-hydroxytamoxifen
with polynucleotide-cellulose columns (236) or sheered
calf thymus DNA (29). Recently, it has been shown th between the binding of estradiol or 4-hydroxytamoxifen
with polynucleotide-cellulose columns (236) or sheered
calf thymus DNA (29). Recently, it has been shown that
estradiol-ER complexes bind more tightly to calf thymus
D with polynucleotide-cellulose columns (236) or sheered
calf thymus DNA (29). Recently, it has been shown that
estradiol-ER complexes bind more tightly to calf thymus
DNA than antiestrogen receptor complexes (90). There
is calf thymus DNA (29). Recently, it has been shown that
estradiol-ER complexes bind more tightly to calf thymus
DNA than antiestrogen receptor complexes (90). There
is a possibility that this finding indicates that there is estradiol-ER complexes bind more tightly to calf thymus
DNA than antiestrogen receptor complexes (90). There
is a possibility that this finding indicates that there is an
alteration in the charge distribution on the recept NA than antiestrogen receptor complexes (90). There
a possibility that this finding indicates that there is an
teration in the charge distribution on the receptor when
estrogen or antiestrogen is located at the binding sit

paired by equivalent concentrations of the antibody. tween estradiol and antiestrogen-ER complexes. Small, Higher concentrations of antibody can, however, sub-
stantially inhibit antiestrogen binding. In contrast, the duct is a possibility that this finding indicates that there is an alteration in the charge distribution on the receptor when an estrogen or antiestrogen is located at the binding site. The technique of sucrose density gradient alteration in the charge distribution on the receptor when
an estrogen or antiestrogen is located at the binding site.
The technique of sucrose density gradient analysis has
been used to demonstrate sedimentation differenc an estrogen or antiestrogen is located at the binding site.
The technique of sucrose density gradient analysis has
been used to demonstrate sedimentation differences be-
tween estradiol and antiestrogen-ER complexes. Small The technique of sucrose density gradient analysis has
been used to demonstrate sedimentation differences be-
tween estradiol and antiestrogen-ER complexes. Small,
but consistent differences are observed with chick ovi-
du been used to demonstrate sedimentation differences be-
tween estradiol and antiestrogen-ER complexes. Small,
but consistent differences are observed with chick ovi-
duct ER (108) and human breast tumor ER in the
presence o tween estradiol and antiestrogen-ER complexes. Small,
but consistent differences are observed with chick ovi-
duct ER (108) and human breast tumor ER in the
presence of KCl (300) or polyclonal antibodies (301) but
the biol

PHARMACOLOGICAL REVIEWS

aspet

to the ligand-binding site on the estrogen receptor (301).

known. High salt (0.4 to 0.6 M KCl) nuclear extract

from MCF-7 breast cancer cells that have been incubate

with either [³H]estradiol or [³H]4-hydroxytamoxi known. High salt $(0.4 \text{ to } 0.6 \text{ M KCl})$ nuclear extracts
from MCF-7 breast cancer cells that have been incubated
with either [³H]estradiol or [³H]4-hydroxytamoxifen
contain a 4S [³H]estradiol ER complex but a 5S [³ known. High salt (0.4 to 0.6 M KCl) nuclear extracts diffom MCF-7 breast cancer cells that have been incubated high with either [³H]estradiol or [³H]4-hydroxytamoxifen recontain a 4S [³H]estradiol ER complex but a 5 from MCF-7 breast cancer cells that have been incubated
with either [³H]estradiol or [³H]4-hydroxytamoxifen
contain a 4S [³H]estradiol ER complex but a 5S [³H]4-
hydroxytamoxifen estrogen receptor complex (77). Th with either [³H]estradiol or [³H]4-hydroxytamoxifen
contain a 4S [³H]estradiol ER complex but a 5S [³H]4-
hydroxytamoxifen estrogen receptor complex (77). These
original findings have been confirmed (303); however contain a 4S ^{[3}H]estradiol ER complex but a 5S [³H]4-
hydroxytamoxifen estrogen receptor complex (77). These tie
original findings have been confirmed (303); however, re
the results are not identical for all cell line hydroxytamoxifen estrogen receptor complex (77). The original findings have been confirmed (303); howeve
the results are not identical for all cell lines. Similexperiments with GH₃ rat pituitary tumor cells has
shown tha original findings have been confirmed (303) ; however, the results are not identical for all cell lines. Similar experiments with GH_3 rat pituitary tumor cells have shown that the nuclear estrogen and antiestrogen rece shown that the nuclear estrogen and antiestrogen recep-

to the ligand-binding site on the estrogen receptor (301).
 known. High salt (0.4 to 0.6 M KCl) nuclear extracts diol are a mixture of 4S and 5S complexes although the

from MCF-7 breast cancer cells that have been incu the results are not identical for all cell lines. Similar systems may have different amounts of protease enzyme
experiments with GH_3 rat pituitary tumor cells have systems. The unusual, though consistent (77, 237, 303), raised in the goat, on the binding of estradiol and 4-hydroxytamoxifen
diol are a mixture of 4S and 5S complexes although the
heavier complex predominates in antiestrogen-treated raised in the goat, on the binding of estradiol and 4-hydroxytamoxife
diol are a mixture of 4S and 5S complexes although the
heavier complex predominates in antiestrogen-treated
rats. Attardi (12) has suggested that differ diol are a mixture of 4S and 5S complexes although the heavier complex predominates in antiestrogen-treated rats. Attardi (12) has suggested that differences in sedimentation characteristics of nuclear estrogens and an diol are a mixture of 4S and 5S complexes although the heavier complex predominates in antiestrogen-treated rats. Attardi (12) has suggested that differences in sedimentation characteristics of nuclear estrogens and anties diol are a mixture of 4S and 5S complexes although the
heavier complex predominates in antiestrogen-treated
rats. Attardi (12) has suggested that differences in sedi-
mentation characteristics of nuclear estrogens and an-
 heavier complex predominates in antiestrogen-treated rats. Attardi (12) has suggested that differences in sedimentation characteristics of nuclear estrogens and antiestrogen receptor complexes from rat uteri are the result rats. Attardi (12) has suggested that differences in sedi-
mentation characteristics of nuclear estrogens and an-
tiestrogen receptor complexes from rat uteri are the
result of sensitivities to proteases. Thus different ce mentation characteristics of nuclear estrogens and antiestrogen receptor complexes from rat uteri are the result of sensitivities to proteases. Thus different cell systems may have different amounts of protease enzyme syst tiestrogen receptor complexes from rat uteri are the
result of sensitivities to proteases. Thus different cell
systems may have different amounts of protease enzyme
systems. The unusual, though consistent (77, 237, 303),
f result of sensitivities to proteases. Thus different constems may have different amounts of protease enzyn systems. The unusual, though consistent (77, 237, 303 finding of a 4S [³H]estradiol-estrogen receptor complextrac systems may have different amounts of protease enzyme finding of a $4S$ [³H]estradiol-estrogen receptor complex

The synthesis of radiolabeled antiestrogens permitted
attribution of radiolabeled antiestrogens permitted
e study of other proteins that specifically bind antiesest.

B. Antiestrogen Binding Sites

The synthesis of radiolabeled antiestrogens permitt

the study of other proteins that specifcally bind anti

trogen. The first report by Sutherland and Foo (29 B. Antiestrogen Binding Sites

The synthesis of radiolabeled antiestrogens permitted

the study of other proteins that specifically bind anties-

trogen. The first report by Sutherland and Foo (292)

described the interac B. Antiestrogen Binding Sites
The synthesis of radiolabeled antiestrogens permitted
the study of other proteins that specifically bind anties-
trogen. The first report by Sutherland and Foo (292)
described the interaction The synthesis of radiolabeled antiestrogens permitted
the study of other proteins that specifically bind anties-
trogen. The first report by Sutherland and Foo (292)
described the interaction of tritium-labeled tamoxifen
a the study of other proteins that specifically bind anties-
trogen. The first report by Sutherland and Foo (292)
described the interaction of tritium-labeled tamoxifen
and CI628 with rat uterine and chick oviduct cytosol. I trogen. The first report by Sutherland and Foo (292)
described the interaction of tritium-labeled tamoxifen
and CI628 with rat uterine and chick oviduct cytosol. In
the rat uterus, CI628, tamoxifen, and estradiol bound described the interaction of tritium-labeled tamoxifered and CI628 with rat uterine and chick oviduct cytosol. In the rat uterus, CI628, tamoxifen, and estradiol bound to a similar number of saturable binding sites and est and CI628 with rat uterine and chick oviduct cytosol.
the rat uterus, CI628, tamoxifen, and estradiol bound
a similar number of saturable binding sites and estrad
could completely inhibit the binding of $[^{3}H]$ antiestrog the rat uterus, CI628, tamoxifen, and estradiol bound to
a similar number of saturable binding sites and estradiol
could completely inhibit the binding of $[^{3}H]$ antiestrogens
to these sites. In contrast, high affinity, a similar number of saturable binding sites and estradiol
could completely inhibit the binding of $[^{3}H]$ antiestrogens C.
to these sites. In contrast, high affinity, saturable anties-
trogen binding sites in chick ovidu could completely inhibit the binding of $[^{3}H]$ antiestrogens
to these sites. In contrast, high affinity, saturable anties-
trogen binding sites in chick oviduct, present at three
times the concentration of estradiol bind to these sites. In contrast, high affinity, saturable anties-
trogen binding sites in chick oviduct, present at three
times the concentration of estradiol binding sites and
estradiol could only partially inhibit the bindi trogen binding sites in chick oviduct, present at three stimes the concentration of estradiol binding sites and restradiol could only partially inhibit the binding of $[^3H]$ antiestrogens. Subsequent studies (296, 298) id times the concentration of estradiol binding sites a
estradiol could only partially inhibit the binding of [³
antiestrogens. Subsequent studies (296, 298) identifi
antiestrogen binding sites in the cytosols of ER posit
b estradiol could only partially inhibit the binding of $[^{3}H]$ acteristrogens. Subsequent studies (296, 298) identified reantiestrogen binding sites in the cytosols of ER positive troloreast tumors and several estrogen ta antiestrogens. Subsequent studies $(296, 298)$ identified relative states antiestrogen binding sites in the cytosols of ER positive the breast tumors and several estrogen target tissues including immature rat uterus. The antiestrogen binding sites in the cytosols of ER positive tro
breast tumors and several estrogen target tissues includ-
ing immature rat uterus. The antiestrogen binding site as
in rat uterus $(K_d$ approximately 1 nM) is o breast tumors and several estrogen target tissues inc
ing immature rat uterus. The antiestrogen binding
in rat uterus $(K_d$ approximately 1 nM) is only obset
if 90% to 95% of ER is occupied by prior treatment v
estra ing immature rat uterus. The antiestrogen binding site a in rat uterus $(K_d$ approximately 1 nM) is only observed if 90% to 95% of ER is occupied by prior treatment with approximately in vivo (233). The concentrations of a in rat uterus $(K_d$ approximately 1 nM) is only observe
if 90% to 95% of ER is occupied by prior treatment wit
estradiol in vivo (233). The concentrations of antiestre
gen binding sites in rat uterine cytosol fluctuates du the estrous cycle and is more resistant to thermal denagen binding sites in rat uterine cytosol fluctuates during 2
the estrous cycle and is more resistant to thermal dena-
turation than the ER (94). However, some controversy
now surrounds the exact subcellular location, ident the estrous cycle and is more resistant to thermal denaturation than the ER (94). However, some controversy phenow surrounds the exact subcellular location, identification and function of the site. Some laboratories (261) turation than the ER (94). However, some controversy phow surrounds the exact subcellular location, identification and function of the site. Some laboratories (261) thare unable to identify antiestrogen binding sites in now surrounds the exact subcellular location, identication and function of the site. Some laboratories (2) are unable to identify antiestrogen binding sites $100,000 \times g$ supernatants (cytosols) whereas others (119, 164, 1 are unable to identify antiestrogen binding sites in $100,000 \times g$ supernatants (cytosols) whereas others can (119, 164, 193). The target site specificity is also controversial as ER negative tumors (164, 231) and all huma are unable to identify antiestrogen binding sites in $100,000 \times g$ supernatants (cytosols) whereas others can (119, 164, 193). The target site specificity is also controversial as ER negative tumors (164, 231) and all huma $100,000 \times g$ supernatants (cytosols) whereas others c
(119, 164, 193). The target site specificity is also contr
versial as ER negative tumors (164, 231) and all hum
tissues tested have antiestrogen binding sites (193). E (119, 164, 193). The target site specificity is also cont versial as ER negative tumors $(164, 231)$ and all hum tissues tested have antiestrogen binding sites (193) . It tensive studies in the rat (290) have identifie versial as ER negative tumors (164, 231) and all hum
tissues tested have antiestrogen binding sites (193). I
tensive studies in the rat (290) have identified the mic
somal fraction of tissues to contain the highest conce
t tissues tested have antiestrogen binding sites (193). E
tensive studies in the rat (290) have identified the micr
somal fraction of tissues to contain the highest conce
tration of antiestrogen-binding sites. The liver is p tensive studies in the rat (290) have identified the microsomal fraction of tissues to contain the highest concentration of antiestrogen binding sites. The liver is particularly rich in the sites $(290, 325)$. Antiestro somal fraction of tissues to contain the highest concerned tration of antiestrogen binding sites. The liver is particularly rich in the sites (290, 325). Antiestrogen-sensitive binding sites in uterus, vagina and liver hav tration of antiestrogen binding sites. The liver is particularly rich in the sites $(290, 325)$. Antiestrogen-sensitive
binding sites in uterus, vagina and liver have been de-
scribed for the immature rat in vivo (158) . ularly rich in the sites (290, 325). Antiestrogen-sensitive
binding sites in uterus, vagina and liver have been de-
in scribed for the immature rat in vivo (158). Furthermore, site
a triphenylethylene-antiestrogen binding binding sites in uterus, vagina and liver have been α scribed for the immature rat in vivo (158). Furthermo a triphenylethylene-antiestrogen binding site is prese on rat low density lipoprotein (LDL) $(K_d 28 \text{ nM})$ whis scribed for the immature rat in vivo (158). Furthermore, si
a triphenylethylene-antiestrogen binding site is present and
on rat low density lipoprotein (LDL) $(K_d 28 \text{ nM})$ which cos
is distinct from the binding site in li a triphenylethylene-antiestrogen binding site is present and
on rat low density lipoprotein (LDL) $(K_d 28 \text{ nM})$ which cell
is distinct from the binding site in liver (326). An endog-
enous ligand (58) is present in boiled on rat low density lipoprotein (LDL) $(K_d 28 \text{ nM})$ which
is distinct from the binding site in liver (326). An endog-
enous ligand (58) is present in boiled ethanol extracts of
rat liver that prevents the binding of $[^3H]$ is distinct from the binding site in liver (326). An endogenous ligand (58) is present in boiled ethanol extracts of but that prevents the binding of [³H]tamoxifen to of both LDL and liver preparations. The "ligand" has unknown. t liver that prevents the binding of $[^{3}H]$ tamoxifen the LDL and liver preparations. The "ligand" has no yet been characterized, and its physiological role is known.
There is general agreement about the structural specboth LDL and liver preparations. The "ligand" has not as yet been characterized, and its physiological role is unknown.
There is general agreement about the structural specificity of "antiestrogen binding" sites (119, 290,

as yet been characterized, and its physiological role is unknown.
There is general agreement about the structural specificity of "antiestrogen binding" sites (119, 290, 298, 325)
The steroids estradiol, progesterone, testo unknown.
There is general agreement about the structural specificity of "antiestrogen binding" sites (119, 290, 298, 325
The steroids estradiol, progesterone, testosterone, dihy
drotestosterone, or hydrocortisone do not af There is general agreement about the structural spificity of "antiestrogen binding" sites $(119, 290, 298, 32$
The steroids estradiol, progesterone, testosterone, dildrotestosterone, or hydrocortisone do not affect the bi ificity of "antiestrogen binding" sites (119, 290, 298, 325). if
The steroids estradiol, progesterone, testosterone, dihy-
drotestosterone, or hydrocortisone do not affect the bind-
ing of [³H] tamoxifen. Nonpolar anties The steroids estradiol, progesterone, testosterone, dihy-
drotestosterone, or hydrocortisone do not affect the bind-
ing of [³H]tamoxifen. Nonpolar antiestrogens, tamoxi-
fen, CI628, enclomiphene, and nafoxidine, have a drotestosterone, or hydrocortisone do not affect the bind-
ing of [³H]tamoxifen. Nonpolar antiestrogens, tamoxi-
fen, CI628, enclomiphene, and nafoxidine, have a high
affinity for the binding sites, but polar antiestroge fen, CI628, enclomiphene, and nafoxidine, have a high affinity for the binding sites, but polar antiestrogens, 4-hydroxytamoxifen LY 117018 or LY 156758, have a lower affinity (290, 326). Interestingly enough, an "LY 11701

AN
binding component" of rabbit and rat uterine cytosols
has recently been described (313) that appears to be AN
binding component" of rabbit and rat uterine cytosols
has recently been described (313) that appears to be
specific for this particular compound. The relevance of AN
binding component" of rabbit and rat uterine cytosols
has recently been described (313) that appears to be
specific for this particular compound. The relevance of
this observation to the mechanism of antiestrogen action this of rabbit and rat uterine cytosols
has recently been described (313) that appears to be
specific for this particular compound. The relevance of
this observation to the mechanism of antiestrogen action
is unknown. has recently been described (313) that appears to be specific for this particular compound. The relevance of this observation to the mechanism of antiestrogen action is unknown.
The alkylaminoethoxy side chain of tamoxifen is recently been described (313) that appears to be ecific for this particular compound. The relevance of is observation to the mechanism of antiestrogen action unknown.
The alkylaminoethoxy side chain of tamoxifen (315) d

specific for this particular compound. The relevance of
this observation to the mechanism of antiestrogen action
is unknown.
The alkylaminoethoxy side chain of tamoxifen (315)
and enclomiphene (234) is important for high a this observation to the mechanism of antiestrogen action
is unknown.
The alkylaminoethoxy side chain of tamoxifen (315)
and enclomiphene (234) is important for high affinity
binding; minor modifications in length or remova is unknown.
The alkylaminoethoxy side chain of tamoxifen (315)
and enclomiphene (234) is important for high affinity
binding; minor modifications in length or removal are
generally detrimental but substitution of the amino The alkylaminoethoxy side chain of tamoxifen (315)
and enclomiphene (234) is important for high affinity
binding; minor modifications in length or removal are
generally detrimental but substitution of the amino
group in ta and enclomiphene (234) is important for high affin
binding; minor modifications in length or removal a
generally detrimental but substitution of the ami
group in tamoxifen with various unsaturated amine ri
system increa Example, annot accurations in tength of removal a
generally detrimental but substitution of the amingroup in tamoxifen with various unsaturated amine risystem increases affinity for the binding sites (290).
C. Antiestrogen oup in tamoxifen with various unsaturated amine ring
stem increases affinity for the binding sites (290).
Antiestrogen Binding Sites: Biological Function
The ubiquitous distribution of antiestrogen binding
ces (193) tends

system increases affinity for the binding sites (290).
C. Antiestrogen Binding Sites: Biological Function
The ubiquitous distribution of antiestrogen binding
sites (193) tends to argue against their central role in the
reg C. Antiestrogen-Binding Sites: Biological Function
The ubiquitous distribution of antiestrogen-binding
sites (193) tends to argue against their central role in the
regulation of estrogen-dependent events. Antiestrogen
acti C. Antiestrogen Binding Sites: Biological Function
The ubiquitous distribution of antiestrogen binding
sites (193) tends to argue against their central role in the
regulation of estrogen-dependent events. Antiestrogen
acti The ubiquitous distribution of antiestrogen binding
sites (193) tends to argue against their central role in the
regulation of estrogen-dependent events. Antiestrogen
action in vivo (166, 209) and in vitro (61, 216) is gen sites (193) tends to argue against their central role in the regulation of estrogen-dependent events. Antiestrogen action in vivo (166, 209) and in vitro (61, 216) is generally reversible with estradiol. Therefore, if by d regulation of estrogen-dependent events. Antiestrogen
action in vivo (166, 209) and in vitro (61, 216) is generally
reversible with estradiol. Therefore, if by definition, es-
trogen can only compete with antiestrogens for action in vivo (166, 209) and in vitro (61, 216) is a
reversible with estradiol. Therefore, if by definitrogen can only compete with antiestrogens fo
trogen receptor and not antiestrogen binding si
a single mode of action versible with estradiol. Therefore, if by definition, espacen can only compete with antiestrogens for the espacen receptor and not antiestrogen binding sites, then single mode of action appears to be operating.
The estroge

if 90% to 95% of ER is occupied by prior treatment with appear to correlate with relative binding affinities for the estradiol in vivo (233). The concentrations of antiestro-
estrogen receptor and not antiestrogen b trogen can only compete with antiestrogens for the es-
trogen receptor and not antiestrogen binding sites, then
a single mode of action appears to be operating.
The estrogenic and antiestrogenic properties of ligands
appea trogen receptor and not antiestrogen binding sites, then
a single mode of action appears to be operating.
The estrogenic and antiestrogenic properties of ligands
appear to correlate with relative binding affinities for the a single mode of action appears to be operating.
The estrogenic and antiestrogenic properties of ligands
appear to correlate with relative binding affinities for the
estrogen receptor and not antiestrogen binding sites (51 The estrogenic and antiestrogenic properties of ligar
appear to correlate with relative binding affinities for
estrogen receptor and not antiestrogen binding sites (
235). The triphenylethylenes ICI 47,699 (*cis* isomer
ta appear to correlate with relative binding affinities for the estrogen receptor and not antiestrogen binding sites $(51, 235)$. The triphenylethylenes ICI 47,699 (*cis* isomer of tamoxifen) and zuclomiphene (*cis* isomer o tamoxifen) and zuclomiphene (cis isomer of enclomi-235). The triphenylethylenes ICI 47,699 (cis isomer of tamoxifen) and zuclomiphene (cis isomer of enclomiphene) both have high affinity for "antiestrogen binding site" (290) but the compounds are weak estrogens rather tha tamoxifen) and zuclomiphene (cis isomer of enclomi-
phene) both have high affinity for "antiestrogen binding"
site" (290) but the compounds are weak estrogens rather
than antiestrogens. Structural derivatives of clomiphene phene) both have high affinity for "antiestrogen bin
site" (290) but the compounds are weak estrogens ra
than antiestrogens. Structural derivatives of clomipl
(235) and the metabolites of tamoxifen (61, 250) con
the growth site" (290) but the compounds are weak estrogens rather
than antiestrogens. Structural derivatives of clomiphene
(235) and the metabolites of tamoxifen (61, 250) control
the growth of MCF-7 breast cancer cells at concentra (235) and the metabolites of tamoxifen (61, 250) conthe growth of MCF-7 breast cancer cells at concent tions consistent with their relative affinities for estrogen receptor. In particular, the potent antiestrog LY 117018, (235) and the metabolites of tamoxifen $(61, 250)$ control
the growth of MCF-7 breast cancer cells at concentra-
tions consistent with their relative affinities for the
estrogen receptor. In particular, the potent antiest the growth of MCF-7 breast cancer cells at concentra-
tions consistent with their relative affinities for the
estrogen receptor. In particular, the potent antiestrogen,
LY 117018, has a high affinity for the ER and is a po tions consistent with their relative affinities for the estrogen receptor. In particular, the potent antiestrogen, LY 117018, has a high affinity for the ER and is a potent agent for the control of MCF-7 breast cancer cell estrogen recepton
LY 117018, has a
agent for the con
(280) but has a lexite (290, 326).
Breast cancer B 117018, has a high affinity for the ER and is a potent
ent for the control of MCF-7 breast cancer cell growth
80) but has a low affinity for the antiestrogen binding
 B (290, 326).
Breast cancer cell lines have been (280) but has a low affinity for the antiestrogen binding.

agent for the control of MCF-7 breast cancer cell growth (280) but has a low affinity for the antiestrogen binding site (290, 326).
Breast cancer cell lines have been studied extensively in an attempt to correlate levels o site (290, 326).
Breast cancer cell lines have been studied extensive in an attempt to correlate levels of antiestrogen bind
sites with inhibition of cell growth by antiestrogens. F
and coworkers (93) have described a tam Breast cancer cell lines have been studied extensively
in an attempt to correlate levels of antiestrogen binding
sites with inhibition of cell growth by antiestrogens. Faye
and coworkers (93) have described a tamoxifen-re in an attempt to correlate levels of antiestrogen binding
sites with inhibition of cell growth by antiestrogens. Faye
and coworkers (93) have described a tamoxifen-resistant
cell line RT \times 6 derived from MCF-7 that has sites with inhibition of cell growth by antiestrogens. Faye
and coworkers (93) have described a tamoxifen-resistant
cell line RT \times 6 derived from MCF-7 that has ER levels
equivalent to those observed in wild-type MCF-7 and coworkers (93) have described a tamoxifen-resistant
cell line RT \times 6 derived from MCF-7 that has ER levels
equivalent to those observed in wild-type MCF-7 cells
but the tamoxifen-resistant cells contain very low le cell line RT \times 6 derived from MCF-7 that has ER levels
equivalent to those observed in wild-type MCF-7 cells
but the tamoxifen-resistant cells contain very low levels
of antiestrogen binding sites. In contrast, Miller equivalent to those observed in wild-type MCF-7 cells
but the tamoxifen-resistant cells contain very low levels
of antiestrogen binding sites. In contrast, Miller and
Katzenellenbogen (231) have compared three breast can-
 but the tamoxifen-resistant cells contain very low levels
of antiestrogen binding sites. In contrast, Miller and
Katzenellenbogen (231) have compared three breast can-
cer cell lines MCF-7, T47D, and MD-MB-231 that con-
ta of antiestrogen binding sites. In contrast, Miller a Katzenellenbogen (231) have compared three breast corr cell lines MCF-7, T47D, and MD-MB-231 that cotain similar levels of antiestrogen binding sites but hilow, and und Katzenellenbogen (231) have compared three breast cancer cell lines MCF-7, T47D, and MD-MB-231 that contain similar levels of antiestrogen binding sites but high, low, and undetectable levels of ER, respectively. Tamoxifen cer cell lines MCF-7,
tain similar levels of a
low, and undetectable
ifen inhibits the grow
presence of the ER.
Sutherland and cow in similar levels of antiestrogen binding sites but high,
w, and undetectable levels of ER, respectively. Tamox-
n inhibits the growth of the cells depending upon the
esence of the ER.
Sutherland and coworkers (115) have

low, and undetectable levels of ER, respectively. Tamox-
ifen inhibits the growth of the cells depending upon the
presence of the ER.
Sutherland and coworkers (115) have shown that high
concentrations ($>5 \mu$ M) of antiest ifen inhibits the growth of the cells depending upon the
presence of the ER.
Sutherland and coworkers (115) have shown that high
concentrations ($>5 \mu$ M) of antiestrogens that inhibit the
growth of breast cancer cells can presence of the ER.
Sutherland and coworkers (115) have shown that high
concentrations ($>5 \mu$ M) of antiestrogens that inhibit the
growth of breast cancer cells cannot be reversed by
estrogens. It is possible that this mi Sutherland and coworkers (115) have shown that high
concentrations ($>5 \mu M$) of antiestrogens that inhibit the
growth of breast cancer cells cannot be reversed by
estrogens. It is possible that this might represent a
spec concentrations $(>\5mu)$ of antiestrogens that inhibit the growth of breast cancer cells cannot be reversed by estrogens. It is possible that this might represent a specific method of controlling the growth of cells by a no

PHARMACOLOGICAL REVIEWS

PHARMACOLOGICAL REVIEWS

aspet

however, it is unclear why such a high concentration of incorporation (216), DNA increases (82), and the cell antiestrogen (at the limit of solubility of the compounds) cycle $(G_1 \text{ block})$ of estrogen-sensitive cells (242, 2 ANTIESTROGEN
however, it is unclear why such a high concentration of
antiestrogen (at the limit of solubility of the compounds)
is required to activate an antitumor mechanism via a **isomether and in the multipular and inconduct to activate an antitumor mechanism via a** The binding site with a K_d of 1 nM. Blow however, it is unclear why such a
antiestrogen (at the limit of solub
is required to activate an antitu
binding site with a K_d of 1 nM.
Black and Goode (22) have pr wever, it is unclear why such a high concentration of
tiestrogen (at the limit of solubility of the compounds)
required to activate an antitumor mechanism via a
fiding site with a K_d of 1 nM.
Black and Goode (22) have p

antiestrogen (at the limit of solubility of the compounds)
is required to activate an antitumor mechanism via a
binding site with a K_d of 1 nM.
Black and Goode (22) have proposed that tamoxifen
may produce some of its is required to activate an antitumor mechanism via
binding site with a K_d of 1 nM.
Black and Goode (22) have proposed that tamoxit
may produce some of its *estrogenic* effects in the
uterus via the "antiestrogen binding binding site with a K_d of 1 nM. Blood-
Black and Goode (22) have proposed that tamoxifen lipopromay produce some of its *estrogenic* effects in the rat diffuse
uterus via the "antiestrogen binding site." This conclu-
si may produce some of its estrogenic effects in the rat diffuses into all tissues. In an estrogen target tissue, an uterus via the "antiestrogen binding site." This conclu-
sion is based upon the finding that LY 117018 has may produce some of its *estrogenic* effects in the rat duterus via the "antiestrogen binding site." This conclu-
sion is based upon the finding that LY 117018 has low sites affinity for antiestrogen binding sites and a hi uterus via the "antiestrogen binding site." This conclu-
sion is based upon the finding that LY 117018 has low
affinity for antiestrogen binding sites and a high affinity (19
for the ER, but while LY 117018 can inhibit est affinity for antiestrogen binding sites and a high affinity (199). The interaction equilibria that are established
for the ER, but while LY 117018 can inhibit estrogen depend upon the relative binding affinities (RBA) of for the ER, but while LY 117018 can inhibit estrogen for the ER, but while LY 117018 can inhibit estrogen action in the uterus, high doese (1 mg/rat) of LY 117018 are unable to inhibit the uterotropic effect of tamoxifen (1 mg/rat) . However, recent studies $(167, 316)$ de e unable to inhibit the uterotropic effect of tamoxife mg/rat). However, recent studies (167, 316) demorate that LY 117018 can inhibit the uterotropic effect tamoxifen if the correct dosage ratios are used.
Finally, one co

of tamoxifen if the correct dosage ratios are used.
Finally, one could suggest that the antiestrogen binding protein does not have a positive biological function (1 mg/rat). However, recent studies (167, 316) demon-
strate that LY 117018 can inhibit the uterotropic effects for
of tamoxifen if the correct dosage ratios are used.
Finally, one could suggest that the antiestrogen bind strate that LY 117018 can inhibit the uterotropic effects
of tamoxifen if the correct dosage ratios are used.
Finally, one could suggest that the antiestrogen bind-
ing protein does not have a positive biological function
 of tamoxifen if the correct dosage ratios are used.
Finally, one could suggest that the antiestrogen bind-
ing protein does not have a positive biological function
per se, but may have an adverse effect on the expressions Finally, one could suggest that the antiestrogen bind-
ing protein does not have a positive biological function
per se, but may have an adverse effect on the expressions
of the pharmacological actions of tamoxifen. If some ing protein does not have a positive biological function
per se, but may have an adverse effect on the expressions
of the pharmacological actions of tamoxifen. If some
breast tumors have larger concentrations of antiestro of the pharmacological actions of tamoxifen. If some
breast tumors have larger concentrations of antiestrogen
binding sites (or perhaps it is induced during therapy
with tamoxifen) than others, then the drug may prefer-
e of the pharmacological actions of tamoxifen. If some
breast tumors have larger concentrations of antiestrogen
binding sites (or perhaps it is induced during therapy
with tamoxifen) than others, then the drug may prefer-
e breast tumors have larger concentrations of antiestro-
binding sites (or perhaps it is induced during ther-
with tamoxifen) than others, then the drug may pre-
entially bind to the high affinity sites $(K_d \, 1 \, \text{nM})$ rat binding sites (or perhaps it is induced during therapy
with tamoxifen) than others, then the drug may prefer-
entially bind to the high affinity sites $(K_d 1 nM)$ rather
than block the ER $(K_d \approx 80 nM)$. Under these biochem-
 with tamoxifen) than others, then the drug may preferentially bind to the high affinity sites $(K_d 1 nM)$ rather than block the ER $(K_d \approx 80 nM)$. Under these biochemical circumstances, the hormone-dependent tumors might cont entially bind to the high affinity sites $(K_d 1 \text{ nM})$ rather
than block the ER $(K_d \approx 80 \text{ nM})$. Under these biochem-
ical circumstances, the hormone-dependent tumors
might continue to grow in the face of tamoxifen therapy than block the ER $(K_d \approx 80 \text{ nM})$. Under these biochemical circumstances, the hormone-dependent tumors might continue to grow in the face of tamoxifen therapy.
Indeed, treatment of patients that have failed tamoxifen ther ical circumstances, the hormone-dependent tumors
might continue to grow in the face of tamoxifen therapy.
Indeed, treatment of patients that have failed tamoxifen
therapy with aminoglutethimide has resulted in a sub-
stant might continue to grow in the face of tamoxifen therapy.

Indeed, treatment of patients that have failed tamoxifen

therapy with aminoglutethimide has resulted in a sub-

stantial number of second objective responses (124 Indeed, treatment of patients that have failed tame
therapy with aminoglutethimide has resulted in a
stantial number of second objective responses (124,
Aminoglutethimide is considered to be an inhibit
aromatizing enzyme s therapy with aminoglutethimide has resulted in a substantial number of second objective responses (124, 131).
Aminoglutethimide is considered to be an inhibitor of aromatizing enzyme systems that convert androstenedione to stantial number of second objective responses $(124, 131)$.
Aminoglutethimide is considered to be an inhibitor of aromatizing enzyme systems that convert androstenedione to estrone. Hormone-dependent disease is controlled Aminoglutethimide is considered to be an inhibitor of situation aromatizing enzyme systems that convert androstenedi-
one to estrone. Hormone-dependent disease is controlled
by preventing estrogen synthesis rather than pr Aminoglutethimide is considered to be an initiative of sites" (S. D. Lyman and V. C. Jordan, unpublished
aromatizing enzyme systems that convert androstenedi-
one to estrone. Hormone-dependent disease is controlled
by pre one to estrone. Hormone-dependent disease is controlled
by preventing estrogen synthesis rather than preventing
estrogen action in the tumor (275). It may, therefore, be
possible to predict patients that can respond readil by preventing estrogen synthesis rather than preventing
estrogen action in the tumor (275) . It may, therefore, be
possible to predict patients that can respond readily to
tamoxifen therapy by an assessment of "antiestro estrogen action in the tumor (275). It may, therefore, be
possible to predict patients that can respond readily to
tamoxifen therapy by an assessment of "antiestrogen
binding sites" in a tumor: a high level of sites would possible to predict patients that can respond readily to
tamoxifen therapy by an assessment of "antiestrogen
binding sites" in a tumor: a high level of sites would
prevent tamoxifen from blocking estradiol binding to the
 tamoxifen therapy by an assessment of "antiestrogen binding sites" in a tumor: a high level of sites would prevent tamoxifen from blocking estradiol binding to the receptor. In fact, a preliminary report from Bloom and Fis binding sites" in a tumor: a high level of sites wo
prevent tamoxifen from blocking estradiol binding to
receptor. In fact, a preliminary report from Bloom a
Fishman (26) suggests that they may have already
veloped such a prevent tamoxifen from blocking estradiol binding to the receptor. In fact, a preliminary report from Bloom and Fishman (26) suggests that they may have already developed such a test. The response of patients to tamoxifen receptor. In fact, a preliminary report from Bloom and
Fishman (26) suggests that they may have already de-
veloped such a test. The response of patients to tamoxi-
fen was correlated to the ability of tamoxifen to inhibit Fishman (26) suggests that they may have already de
veloped such a test. The response of patients to tamoxi
fen was correlated to the ability of tamoxifen to inhibi
the binding of $[^3H]$ estradiol to tumor ER. Those patien veloped such a test. The response of patients to tamoxi-
fen was correlated to the ability of tamoxifen to inhibit
the binding of $[^{3}H]$ estradiol to tumor ER. Those patients
whose tumor was ER positive with estradiol as fen was correlated to the ability of tamoxifen to inhite binding of $[^3H]$ estradiol to tumor ER. Those patie whose tumor was ER positive with estradiol as a copetitor for $[^3H]$ estradiol binding, but was refractory compet the binding of [³H]estradiol to tumor ER. Those patients
whose tumor was ER positive with estradiol as a com-
petitor for [³H]estradiol binding, but was refractory to
competition with tamoxifen, did not respond to tamo whose tumor was ER positive with estradiol as a competitor for $[^{3}H]$ estradiol binding, but was refractory to competition with tamoxifen, did not respond to tamoxifen therapy. Clearly, future research could establish t

Ix. **Antiestrogenic** Mechanisms: Summary

It is now appropriate to summarize many of the sub-
cellular effects observed with antiestrogens in vitro (figvalidity of this hypothesis.
 IX. Antiestrogenic Mechanisms: Summary

It is now appropriate to summarize many of the sub-

cellular effects observed with antiestrogens in vitro (fig-

ure 15). Although various exceptions IX. Antiestrogenic Mechanisms: Summary
It is now appropriate to summarize many of the sub
cellular effects observed with antiestrogens in vitro (figure 15). Although various exceptions have been men-
tioned previously, ant IX. Antiestrogenic Mechanisms: Summary
It is now appropriate to summarize many of the sub-
cellular effects observed with antiestrogens in vitro (fig-
ure 15). Although various exceptions have been men-
tioned previously, It is now appropriate to summarize many of the sucellular effects observed with antiestrogens in vitro (fure 15). Although various exceptions have been metioned previously, antiestrogens, in general, regulate trogen-stimu cellular effects observed with antiestrogens in vitro (figure 15). Although various exceptions have been mentioned previously, antiestrogens, in general, regulate estrogen-stimulated prolactin synthesis (211, 212), progest ure 15). Although various exceptions have been mentioned previously, antiestrogens, in general, regulate estrogen-stimulated prolactin synthesis (211, 212), progesterone receptor production (76, 134), 24 K, 36 K (80), and

HARMACOLOGY 261
incorporation (216), DNA increases (82), and the cell
cycle (G₁ block) of estrogen-sensitive cells (242, 293). HARMACOLOGY 261
incorporation (216), DNA increases (82), and the cell
cycle (G₁ block) of estrogen-sensitive cells (242, 293).
These effects may be caused by a variety of mechanisms. 26
incorporation (216), DNA increases (82), and the ce
cycle $(G_1 \text{ block})$ of estrogen-sensitive cells (242, 293
These effects may be caused by a variety of mechanisms.
Blood-borne antiestrogen [possibly bound to low densi incorporation (216), DNA increases (82), and the cell
cycle $(G_1 \text{ block})$ of estrogen-sensitive cells (242, 293).
These effects may be caused by a variety of mechanisms.
Blood-borne antiestrogen [possibly bound to low densit incorporation (216), DNA increases (82), and the cell
cycle (G_1 block) of estrogen-sensitive cells (242, 293).
These effects may be caused by a variety of mechanisms.
Blood-borne antiestrogen [possibly bound to low den cycle $(G_1 \text{ block})$ of estrogen-sensitive cells $(242, 293)$.
These effects may be caused by a variety of mechanisms.
Blood-borne antiestrogen [possibly bound to low density
lipoprotein (326)] dissociates from carrier prot These effects may be caused by a variety of mechanisms.
Blood-borne antiestrogen [possibly bound to low density
lipoprotein (326)] dissociates from carrier proteins and
diffuses into all tissues. In an estrogen target tiss Blood-borne antiestrogen [possibly bound to low density
lipoprotein (326)] dissociates from carrier proteins and
diffuses into all tissues. In an estrogen target tissue, an
antiestrogen can bind to ER (29), antiestrogen bi lipoprotein (326)] dissociates from carrier proteins and
diffuses into all tissues. In an estrogen target tissue, an
antiestrogen can bind to ER (29), antiestrogen binding
sites (290, 298, 315), or, as recently reported, c diffuses into all tissues. In an estrogen target tissue, an antiestrogen can bind to ER (29), antiestrogen binding sites (290, 298, 315), or, as recently reported, calmodulin (199). The interaction equilibria that are esta antiestrogen can bind to ER (29), antiestrogen binding
sites (290, 298, 315), or, as recently reported, calmodulin
(199). The interaction equilibria that are established
depend upon the relative binding affinities (RBA) o sites (290, 298, 315), or, as recently reported, calmodulin (199). The interaction equilibria that are established depend upon the relative binding affinities (RBA) of the antiestrogen from the proteins. Tamoxifen has a h (199). The interaction equilibria that are established
depend upon the relative binding affinities (RBA) of the
antiestrogen from the proteins. Tamoxifen has a high
binding affinity for the antiestrogen binding sites (K_d depend upon the relative binding affinities (RBA) of the
antiestrogen from the proteins. Tamoxifen has a high
binding affinity for the antiestrogen binding sites $(K_d 1$
nM, RBA = 100, E_2 RBA = 0), but a low binding aff antiestrogen from the proteins. Tamoxifen has a high
binding affinity for the antiestrogen binding sites $(K_d 1$
nM, RBA = 100, E_2 RBA = 0), but a low binding affinity
for the estrogen receptor $(K_d \simeq 80 \text{ nM}$, RBA = binding affinity for the antiestrogen binding sites $(K_d 1 \text{ nM}, \text{RBA} = 100, \text{E}_2 \text{ RBA} = 0)$, but a low binding affinity for the estrogen receptor $(K_d \simeq 80 \text{ nM}, \text{RBA} = 5, \text{E}_2 \text{RBA} = 100$. In contrast, LY117018 has a nM, RBA = 100, E₂ RBA^{*}= 0), but a low binding affinity
for the estrogen receptor ($K_d \approx 80$ nM, RBA = 5, E₂
RBA = 100). In contrast, LY117018 has a low binding
affinity for the antiestrogen binding site (RBA \approx <1 RBA = 100). In contrast, LY117018 has a low binding
affinity for the antiestrogen binding site (RBA \simeq <1)
but a high affinity for the estrogen receptor (RBA >
100). On the one hand, it is possible that interaction of
 affinity for the antiestrogen binding site $(RBA \approx < 1)$
but a high affinity for the estrogen receptor $(RBA > 100)$. On the one hand, it is possible that interaction of
antiestrogens with either antiestrogen binding sites or
c but a high affinity for the estrogen receptor (RBA > 100). On the one hand, it is possible that interaction of antiestrogens with either antiestrogen binding sites or calmodulin (or both) could affect the cell cycle. On th antiestrogens with either antiestrogen binding sites or calmodulin (or both) could affect the cell cycle. On the other hand, low concentrations of estrogens can reverse the inhibitory effects of antiestrogens in most model antiestrogens with either antiestrogen binding site
calmodulin (or both) could affect the cell cycle. On
other hand, low concentrations of estrogens can rev
the inhibitory effects of antiestrogens in most m
systems, altho tions $(7.5 \times 10^{-6} \text{ M})$ of antiestrogen.
Tamoxifen is a potent inhibitor of calmodulin-mediher hand, low concentrations of estrogens can i
e inhibitory effects of antiestrogens in most
stems, although this is difficult with high concens (7.5 \times 10⁻⁶ M) of antiestrogen.
Tamoxifen is a potent inhibitor of cal

the inhibitory effects of antiestrogens in most model
systems, although this is difficult with high concentra-
tions $(7.5 \times 10^{-6} \text{ M})$ of antiestrogen.
Tamoxifen is a potent inhibitor of calmodulin-medi-
ated phosphodie systems, although this is difficult with high concentr
tions $(7.5 \times 10^{-6} \text{ M})$ of antiestrogen.
Tamoxifen is a potent inhibitor of calmodulin-mec
ated phosphodiesterase (199). The other recognized i
hibitors of calmodul tions $(7.5 \times 10^{-6}$ M) of antiestrogen.
Tamoxifen is a potent inhibitor of calmodulin-mediated phosphodiesterase (199). The other recognized inhibitors of calmodulin are major tranquilizers, e.g., trifluperazine, which i Tamoxifen is a potent inhibitor of calmodulin-mediated phosphodiesterase (199). The other recognized inhibitors of calmodulin are major tranquilizers, e.g., trifluperazine, which incidentally are efficient inhibitors of [ated phosphodiesterase (199). The other recognized in-
hibitors of calmodulin are major tranquilizers, e.g., triflu-
perazine, which incidentally are efficient inhibitors of
[³H]tamoxifen binding to rat liver "antiestrog hibitors of calmodulin are major tranquilizers, e.g., triflu-
perazine, which incidentally are efficient inhibitors of
[³H]tamoxifen binding to rat liver "antiestrogen binding
sites" (S. D. Lyman and V. C. Jordan, unpubl perazine, which incidentally are efficient inhibitors of [³H]tamoxifen binding to rat liver "antiestrogen binding sites" (S. D. Lyman and V. C. Jordan, unpublished observation) and inhibit the colony formation of breast [³H]tamoxifen binding to rat liver "antiestrogen binding sites" (S. D. Lyman and V. C. Jordan, unpublished observation) and inhibit the colony formation of breast cancer cells (317). Calmodulin is believed to be intimate sites" (S. D. Lyman and V. C. Jordan, unpublished
observation) and inhibit the colony formation of breast
cancer cells (317). Calmodulin is believed to be intimately
involved in cell division (43, 324) and inhibitors of c observation) and inhibit the colony is
cancer cells (317). Calmodulin is belie
involved in cell division (43, 324) an
modulin, such as trifluoperazine, will
the G_1 phase of the cell cycle (143).
Trifluoperazine recentl ncer cells (317). Calmodulin is believed to be intimately volved in cell division (43, 324) and inhibitors of cal-
odulin, such as trifluoperazine, will produce a block in
e G_1 phase of the cell cycle (143).
Trifluoper

involved in cell division $(43, 324)$ and inhibitors of calmodulin, such as trifluoperazine, will produce a block in the G_1 phase of the cell cycle (143) .
Trifluoperazine recently has been shown to prevent the bindin modulin, such as trifluoperazine, will produce a block in
the G_1 phase of the cell cycle (143).
Trifluoperazine recently has been shown to prevent
the binding of iodinated epidermal growth factor to neo-
plastic, but n the G_1 phase of the cell cycle (143).
Trifluoperazine recently has been shown to preve
the binding of iodinated epidermal growth factor to ne
plastic, but not normal, cells in culture (27). The
fascinating observations Trifluoperazine recently has been shown to previous the binding of iodinated epidermal growth factor to no plastic, but not normal, cells in culture (27). The fascinating observations may provide alternative meanisms to ex the binding of iodinated epidermal growth factor to neo-
plastic, but not normal, cells in culture (27). These
fascinating observations may provide alternative mech-
anisms to explain the antiproliferative actions of tamox plastic, but not normal, cells in culture (27). These fascinating observations may provide alternative mechanisms to explain the antiproliferative actions of tamoxifen in either ER positive or negative cells in culture (11 fascinating observations may provide alternative mechanisms to explain the antiproliferative actions of tamoxing in either ER positive or negative cells in culture (115). It is, therefore, possible to envision that breast anisms to explain the antiproliferative actions of tame
ifen in either ER positive or negative cells in cult
(115). It is, therefore, possible to envision that bre
tumors that contain a heterogeneous mixture of l
positive ifen in either ER positive or negative cells in cult (115). It is, therefore, possible to envision that bre tumors that contain a heterogeneous mixture of positive and negative cells may be influenced by tame ifen to: (a) (115). It is, therefore, possible to envision that breast tumors that contain a heterogeneous mixture of ER positive and negative cells may be influenced by tamoxien to: (a) control cell-cell communication by modulation o tumors that control
positive and negatifen to: (a) control
tion of the action
mediated events.
Most studies at sitive and negative cells may be influenced by tamomies in to: (a) control cell-cell communication by modular on of the action of growth factors; or (b) control EF ediated events.
Most studies at present have focused upon

ifen to: (a) control cell-cell communication by modulation of the action of growth factors; or (b) control ER-
mediated events.
Most studies at present have focused upon the inter-
action of antiestrogens with the ER: (a) tion of the action of growth factors; or (b) control ER-
mediated events.
Most studies at present have focused upon the inter-
action of antiestrogens with the ER: (a) Antiestrogens
inhibit the binding of [³H]estradiol t mediated events.
Most studies at present have focused upon the inter-
action of antiestrogens with the ER: (a) Antiestrogens
inhibit the binding of $[^{3}H]$ estradiol to the ER (287). $[^{3}H]$
Antiestrogens bind directly to Most studies at present have focused upon the inter-
action of antiestrogens with the ER: (a) Antiestrogens
inhibit the binding of $[^{3}H]$ estradiol to the ER (287). $[^{3}H]$
Antiestrogens bind directly to the ER (29, 40, action of antiestrogens with the ER: (a) Antiestrogens
inhibit the binding of $[^3H]$ estradiol to the ER (287). $[^3H]$
Antiestrogens bind directly to the ER (29, 40, 173, 181).
(b) Studies with radiolabeled estrogens and a inhibit the binding of [³H]estradiol to the ER (287). [³H]
Antiestrogens bind directly to the ER (29, 40, 173, 181).
(b) Studies with radiolabeled estrogens and antiestrogens
demonstrate that the ligands interact with Antiestrogens bind directly to the ER (29, 40, 173, 181).
(b) Studies with radiolabeled estrogens and antiestrogens
demonstrate that the ligands interact with the receptor
in different ways (262, 301). Estrogens and anties demonstrate that the ligands interact with the receptor
in different ways (262, 301). Estrogens and antiestrogens
may have a different method of "activating" receptors
(260). (c) Differences in the size of nuclear estrogen

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Solution Receptor Form
303). (d) Differences in the interaction of estrogen and the
303). (d) Differences in the interaction of estrogen and the
303). (d) Differences in the interaction of estrogen and the
303). (d) Diff FIG. 15. Potential mechanisms of acception receptor complexes have been noted (77, the 303). (d) Differences in the interaction of estrogen and the antiestrogen receptor complexes with DNA have been ties described (90). Th antiestrogen receptor complexes have been noted (77, 303). (d) Differences in the interaction of estrogen and antiestrogen receptor complexes with DNA have been described (90). This may be related to the observation that a antiestrogen receptor complexes have been noted (77, the 303). (d) Differences in the interaction of estrogen and the antiestrogen receptor complexes with DNA have been ties described (90). This may be related to the obser 303). (d) Differences in the interaction of estrogen and the antiestrogen receptor complexes with DNA have been tied described (90). This may be related to the observation in that antiestrogen receptor complexes are more antiestrogen receptor complexes with DNA have been ties
described (90). This may be related to the observation in
that antiestrogen receptor complexes are more easily to
extracted from nuclei by 0.4 M KCl than estradiol-ER described (90). This may be related to the observation im
that antiestrogen receptor complexes are more easily to
extracted from nuclei by 0.4 M KCl than estradiol-ER rat
complexes (270). (e) The concentration of estradiol that antiestrogen receptor complexes are more easily
extracted from nuclei by 0.4 M KCl than estradiol-ER
complexes (270). (e) The concentration of estradiol-ER
complexes extracted with 0.4 M KCl decrease over the
first 6 extracted from nuclei by 0.4 M KCl than estradiol-ER rats a complexes (270). (e) The concentration of estradiol-ER difficit complexes extracted with 0.4 M KCl decrease over the anties first 6 hr of estrogen exposure ("proc complexes (270). (e) The concentration of estradiol-ER
complexes extracted with 0.4 M KCl decrease over the
first 6 hr of estrogen exposure ("processing") whereas
antiestrogen-ER complexes do not (135–137, 303). (f) ER
res complexes extracted with 0.4 M KCl decrease over th
first 6 hr of estrogen exposure ("processing") wherea
antiestrogen-ER complexes do not (135–137, 303). (f) El
resynthesis was believed to be impaired by antiestroge
(57); first 6 hr of estrogen exposure ("processing") whereas matter
antiestrogen-ER complexes do not $(135-137, 303)$. (f) ER
resynthesis was believed to be impaired by antiestrogen pre
 (57) ; however, the replenishment of rec antiestrogen-ER complexes do not (135–137, 303). (f) ER
resynthesis was believed to be impaired by antiestrogen
(57); however, the replenishment of receptor in the pres-
ence of estrogen and antiestrogen has been found to synthesis was believed to be impaired by antiestrogen p
7); however, the replenishment of receptor in the pres-
ce of estrogen and antiestrogen has been found to be
tinilar (78).
The differences in the physicochemical prop

(57); however, the replenishment of receptor in the pres-
ence of estrogen and antiestrogen has been found to be
trip
similar (78). of
The differences in the physicochemical properties of
the estrogen or antiestrogen recep ence of estrogen and antiestrogen has been found to be training (78).

The differences in the physicochemical properties of pickers of the estrogen or antiestrogen receptor complexes may preflect differences in charge dist of
changes in the physicochemical properties of
the estrogen or antiestrogen receptor complexes may
preflect differences in charge distribution or tertiary
changes in protein structure. Valuable insights into the
differenc The differences in the physicochemical properties of
the estrogen or antiestrogen receptor complexes may
reflect differences in charge distribution or tertiary
changes in protein structure. Valuable insights into the
diffe the estrogen or antiestrogen receptor complexes reflect differences in charge distribution or tertichanges in protein structure. Valuable insights into differences in agonist and antagonist receptor comple can be obtained reflect differences in charge distribution or tertiary Nev
changes in protein structure. Valuable insights into the be considerences in agonist and antagonist receptor complexes ture
can be obtained by a study of structur changes in protein structure. Valuable insights into the be differences in agonist and antagonist receptor complexes ture can be obtained by a study of structure activity relation-
ships. Studies in vivo and in vitro will differences in agonist and antican be obtained by a study of ships. Studies in vivo and in develop a hypothetical model an interaction with the ER. can be obtained by a study of structure activity relation-
ships. Studies in vivo and in vitro will be considered to
develop a hypothetical model for antiestrogen action via
an interaction with the ER.
X. Structure-Activit

X. Structure-Activity Relationships (SAR)
The structure-activity relationships studies of anties-
trogens are a natural extrapolation of the work completed an interaction with the ER.
 $X.$ Structure-Activity Relationships (SAR) $\begin{bmatrix} 1 & 0 \\ 0 & 1 \end{bmatrix}$

The structure-activity relationships studies of anties-

trogens are a natural extrapolation of the work completed

by Do Best Manuscript Manuscrit and Technology
The structure-activity relationships studies of anties-
trogens are a natural extrapolation of the work completed pota
by Dodds and Emmens with nonsteroidal estrogens in 6-m
the 193 X. Structure-Activity Relationships (SAR)
The structure-activity relationships studies of anties-
trogens are a natural extrapolation of the work completed
by Dodds and Emmens with nonsteroidal estrogens in
the 1930s, -40s The structure-activity relationships studies of anti-
trogens are a natural extrapolation of the work complet
by Dodds and Emmens with nonsteroidal estrogens
the 1930s, -40s and -50s (69, 83). The structure-activi-
relatio ogens are a natural extrapolation of the work completed

Dodds and Emmens with nonsteroidal estrogens in

e 1930s, -40s and -50s (69, 83). The structure-activity

lationships of estrogens have been reviewed (171).

Tereniu by Dodds and Emmens with nonsteroidal estrogens in
the 1930s, -40s and -50s (69, 83). The structure-activity
relationships of estrogens have been reviewed (171).
Terenius (306) compared the agonist and antagonis
activities

the 1930s, -40s and -50s (69, 83). The structure-activity anti-
relationships of estrogens have been reviewed (171). ami
Terenius (306) compared the agonist and antagonist with
activities of a broad range of antiestrogens

t action of antiestrogens (AE).

the 3-day immature mouse uterine weight test. Howeve

the agonist activity of the triphenylethylene-based an

tiestrogens in the mouse (121, 174) make SAR studie f action of antiestrogens (AE).
the 3-day immature mouse uterine weight test. However,
the agonist activity of the triphenylethylene-based an-
tiestrogens in the mouse (121, 174) make SAR studies
impractical. Most of the e the 3-day immature mouse uterine weight test. However,
the agonist activity of the triphenylethylene-based an-
tiestrogens in the mouse (121, 174) make SAR studies
impractical. Most of the early studies were undertaken
to the agonist activity of the triphenylethylene-based antiestrogens in the mouse (121, 174) make SAR studies
impractical. Most of the early studies were undertaken
to determine the antifertility activity of compounds in
rats tiestrogens in the mouse $(121, 174)$ make SAR studies
impractical. Most of the early studies were undertaken
to determine the antifertility activity of compounds in
rats and mice $(63, 204-206)$. As a result, it is often impractical. Most of the early studies were undertake
to determine the antifertility activity of compounds is
rats and mice (63, 204–206). As a result, it is ofte
difficult to evaluate the impact of the estrogenic an
antie to determine the antifertility activity of compounds
rats and mice (63, 204–206). As a result, it is of
difficult to evaluate the impact of the estrogenic a
antiestrogenic components of a test compound's ph
macology from t ts and mice (63, 204–206). As a result, it is ofterficult to evaluate the impact of the estrogenic antiestrogenic components of a test compound's phaneology from the results of antifertility experiments. With regard to spe

difficult to evaluate the impact of the estrogenic and antiestrogenic components of a test compound's pharmacology from the results of antifertility experiments.
With regard to specific compound groups, the inter-
pretatio antiestrogenic components of a test compound's phar-
macology from the results of antifertility experiments.
With regard to specific compound groups, the inter-
pretation of the early studies are further complicated by
eit macology from the results of antifertility experiments.
With regard to specific compound groups, the inter-
pretation of the early studies are further complicated by
either misidentification (244) of geometric isomers of
t With regard to specific compound groups, the interpretation of the early studies are further complicated leither misidentification (244) of geometric isomers triphenylethylene derivatives or the testing of mixture of geome pretation of the early studies are further complicated by
either misidentification (244) of geometric isomers of
triphenylethylene derivatives or the testing of mixtures
of geometric isomers (63). Since the pharmacological either misidentification (244) or geometric isomers or
triphenylethylene derivatives or the testing of mixtures
of geometric isomers (63). Since the pharmacological
properties are often opposing (52, 121, 169), this ap-
pr of geometric isomers (63). Since the pharmacological
properties are often opposing (52, 121, 169), this ap-
proach to structure-activity analysis is inappropriate.
Nevertheless, with these reservations, several studies wil properties are often opposing (52, 121, 169), this
proach to structure-activity analysis is inappropri
Nevertheless, with these reservations, several studies
be considered to illustrate the important structural
tures of a proach to structure-activity analy
Nevertheless, with these reservation
be considered to illustrate the impo
tures of a compound necessary for
genic activity in vivo and in vitro.
A Studies in Vivo **A. Studies is a compound network of a compound network of a compound network of a compound network of a Studies in Vivo**
A. Studies in Vivo
Simple hydroxylated is res of a compound necessary for it to exert antiestro-
nic activity in vivo and in vitro.
Studies in Vivo
Simple hydroxylated indenes (273, 286), that are su-
rficially related to the structure of DES are potent

peric activity in vivo and in vitro.

A. Studies in Vivo

Simple hydroxylated indenes (273, 286), that are superficially related to the structure of DES are potent

estrogens. The structure-activity relationships of the in A. Studies in Vivo
Simple hydroxylated indenes (273, 286), that are superficially related to the structure of DES are potent
estrogens. The structure-activity relationships of the in-
dene nucleus have been investigated in A. Studies in Vivo

Simple hydroxylated indenes (273, 286), that are su-

perficially related to the structure of DES are potent

estrogens. The structure-activity relationships of the in-

dene nucleus have been investiga Simple hydroxylated indenes (273, 286), that are su-
perficially related to the structure of DES are potent
estrogens. The structure-activity relationships of the in-
dene nucleus have been investigated in the search for
p perficially related to the structure of DES are potent estrogens. The structure-activity relationships of the in-
dene nucleus have been investigated in the search for
potent antifertility relationships (204) (figure 16). estrogens. The structure-activity relationships of the in-
dene nucleus have been investigated in the search for
potent antifertility relationships (204) (figure 16). The
6-methoxy group is an advantage for activity but po dene nucleus have been investigated in the search for
potent antifertility relationships (204) (figure 16). The
6-methoxy group is an advantage for activity but potent
antifertility activity is determined by the substitute potent antifertility relationships (204) (figure 16). The 6-methoxy group is an advantage for activity but potent antifertility activity is determined by the substituted amine ethoxy side chain. Optimal activity is observe 6-methoxy group is an advantage for activity but potent antifertility activity is determined by the substituted amine ethoxy side chain. Optimal activity is observed with the pyrrolidino side chain (IND 1, figure 16) and antifertility activity is determined by the substituted
amine ethoxy side chain. Optimal activity is observed
with the pyrrolidino side chain (IND 1, figure 16) and
other substituted side chains (IND 2, 3, 4) have reduced

aspet

aspet

NAF 5
FIG. 16. The relative antifertility activity of substituted indenes in
the rat. Data adapted from Lednicer et al. (204).
NAF 6
compound with approximately 1% of the activity of IND
I with the pyrrolidino side chain. Fig. 16. The relative antifertility activity of substituted indenes in
the rat. Data adapted from Lednicer et al. (204).
compound with approximately 1% of the activity of IND
1 with the pyrrolidino side chain. In the same Lednicer and composite that the activity of IND
1 with the pyrrolidino side chain. In the same study,
Lednicer and coworkers (204) showed that the 6 phenol
of IND 4 had approximately 5% of the potency of the compound with approximately 1% of the activity of IND
1 with the pyrrolidino side chain. In the same study, hyderlead and coworkers (204) showed that the 6 phenol
of IND 4 had approximately 5% of the potency of the
methox compound with approximately 1% of the activity of IND
1 with the pyrrolidino side chain. In the same study, by
Lednicer and coworkers (204) showed that the 6 phenol al.
6 IND 4 had approximately 5% of the potency of the
me 1 with the pyrrolidino side chain. In the same study, hydrediction and coworkers (204) showed that the 6 phenol al. (2) of IND 4 had approximately 5% of the potency of the methoxy compound. As previously discussed in secti of IND 4 had approximately 5% of the potency of the
methoxy compound. As previously discussed in section binding, respectively. The methyl substitutions reduce
IV B, a hydroxylated derivative might be expected to the numb of IND 4 had approximately 5% of the potency of the methoxy compound. As previously discussed in section bin IV B, a hydroxylated derivative might be expected to the have a shorter duration of action so that larger doses w methoxy compound. As previously discussed in section bin
IV B, a hydroxylated derivative might be expected to the
have a shorter duration of action so that larger doses will add
be required to maintain adequate drug levels IV B, a hydroxylated derivative might be expected to the have a shorter duration of action so that larger doses will add be required to maintain adequate drug levels. Recently a hydroxylated indene derivative (figure 21:2) have a shorter duration of action so that larger doses will
be required to maintain adequate drug levels. Recently a
hydroxylated indene derivative (figure 21:2) has been
shown to possess antitumor activity (278), although be required to maintain adequate drug levels. Recently a
hydroxylated indene derivative (figure 21:2) has been act
shown to possess antitumor activity (278), although it is side
not clear whether the compound is a weak est hydroxylated indene derivative (figure 21:2) has been shown to possess antitumor activity (278), although it is not clear whether the compound is a weak estrogen that a has a short biological half life, or the compound ha activity. t clear whether the compound is a weak estrogen that
s a short biological half life, or the compound has the
ility to induce a receptor complex with low intrinsic of
tivity.
The 3,4-dihydronaphthalenes further exemplify th

has a short biological half life, or the compound has the ability to induce a receptor complex with low intrinsic activity.
The 3,4-dihydronaphthalenes further exemplify the importance of the substituted side chain for opt ability to induce a receptor complex with low intrinsientivity.
The 3,4-dihydronaphthalenes further exemplify the
importance of the substituted side chain for optima
activity (figure 17). Nafoxidine is the most potent comof

The 3,4-dihydronaphthalenes further exemplify the

importance of the substituted side chain for optimal

weativity (figure 17). Nafoxidine is the most potent com-

be pound of the series although the ether oxygen of th The 3,4-dihydronaphthalenes further exemplify the optimation of the substituted side chain for optimal weativity (figure 17). Nafoxidine is the most potent compound of the series although the ether oxygen of the side R cha importance of the substituted side chain for optimal we
activity (figure 17). Nafoxidine is the most potent com-
pound of the series although the ether oxygen of the side Re
chain can be replaced by carbon with very little activity (figure 17). Nafoxidine is the most potent compound of the series although the ether oxygen of the side chain can be replaced by carbon with very little loss of potency. However, decrease in the length of the side pound of the series although the ether oxygen of the side

chain can be replaced by carbon with very little loss of

tarpotency. However, decrease in the length of the side

chain (NAF 1-3) reduces the antiestrogenic poten chain can be replaced by carbon with very little loss of potency. However, decrease in the length of the side chain (NAF 1-3) reduces the antiestrogenic potency and in fact, removal of the side chain (NAF 6) results in the chain (NAF 1-3) reduces the antiestrogenic potency and, trop in fact, removal of the side chain (NAF 6) results in the tage complete loss of antagonist activity. The resulting compounds are estrogens $(204-206)$. These ob in fact, removal of the side chain (NAF 6) results in the
complete loss of antagonist activity. The resulting com-
pounds are estrogens (204--206). These observations led
Lednicer et al. (206) to suggest that a basic grou complete loss of antagonist activity. The resulting compounds are estrogens (204-206). These observations led Theorical Lednicer et al. (206) to suggest that a basic group, at a transference is required to obtain a molecul pounds are estrogens (204–206). These observations led
Lednicer et al. (206) to suggest that a basic group, at a
given position in space is required to obtain a molecule
with estrogen antagonist activity. This point of vie Lednicer et al. (206) to suggest that a basic group, at a trogiven position in space is required to obtain a molecule A with estrogen antagonist activity. This point of view is activity further supported by the observation with estrogen antagonist activity. This point of view is
further supported by the observation that dimethylation
ortho to the aminoethoxyside chain in MER25 (48) and
tamoxifen (1) reduces antiestrogen activity and receptor

FIG. 17. The relative antiestrogenic activity **of** substituted 3,4-dihydronaphthalenes in immature rats. Data adapted from Lednicer et FIG. 17. The relative antiestrogenic activity of substituted 3,4-dihydronaphthalenes in immature rats. Data adapted from Lednicer et al. (205, 206).
binding, respectively. The methyl substitutions reduce the number of posi

hydronaphthalenes in immature rats. Data adapted from Lednicer et al. (205, 206).

binding, respectively. The methyl substitutions reduce

the number of positions in space that the side chain can

adopt. adopt. nding, respectively. The methyl substitutions reduce
e number of positions in space that the side chain can
opt.
The importance of the side chain for the antiestrogen
tivity of tamoxifen has been studied. Removal of the

potency. However, decrease in the length of the side chains (figure 19) was tested for estrogenic and anties-
chain (NAF 1-3) reduces the antiestrogenic potency and, trogenic activity (2). The phenolic hydroxyl is an adva binding, respectively. The methyl substitutions reduce
the number of positions in space that the side chain can
adopt.
The importance of the side chain for the antiestrogen
activity of tamoxifen has been studied. Removal o the number of positions in space that the side chain can
adopt.
The importance of the side chain for the antiestrogen
activity of tamoxifen has been studied. Removal of the
side chain to produce the phenol (metabolite E) d adopt.
The importance of the side chain for the antiestrogen
activity of tamoxifen has been studied. Removal of the
side chain to produce the phenol (metabolite E) destroys
antiestrogen activity and increases estrogenic ac The importance of the side chain for the antiestrogen
activity of tamoxifen has been studied. Removal of the
side chain to produce the phenol (metabolite E) destroys
antiestrogen activity and increases estrogenic activity
 activity of tamoxifen has been studied. Removal of the side chain to produce the phenol (metabolite E) destroys antiestrogen activity and increases estrogenic activity (165). Robertson and coworkers (253) have tested a ser side chain to produce the phenol (metabolite E) destroys
antiestrogen activity and increases estrogenic activity
(165). Robertson and coworkers (253) have tested a series
of compounds related to tamoxifen but with side cha antiestrogen activity and increases estrogenic activity (165). Robertson and coworkers (253) have tested a series of compounds related to tamoxifen but with side chains of differing basicity (figure 18). They concluded tha (165). Robertson and coworkers (253) have tested a series
of compounds related to tamoxifen but with side chains
of differing basicity (figure 18). They concluded that the
optimal interactions of the side chain with the r of compounds related to tamoxifen but with side chains
of differing basicity (figure 18). They concluded that the
optimal interactions of the side chain with the receptor
were unlikely to be ionic but rather hydrogen bondi of differing basicity (figure 18). They concluded that the optimal interactions of the side chain with the receptor were unlikely to be ionic but rather hydrogen bonding between the side chain and amino acids of the recept optimal interactions of the side chain with the receptor
were unlikely to be ionic but rather hydrogen bonding
between the side chain and amino acids of the receptor.
Recently a series of fixed-ring tricyclic derivatives o were unlikely to be ionic but rather hydrogen bondinetween the side chain and amino acids of the recept
Recently a series of fixed-ring tricyclic derivatives
tamoxifen and 4-hydroxytamoxifen with different s
chains (figure between the side chain and amino acids of the recept
Recently a series of fixed-ring tricyclic derivatives
tamoxifen and 4-hydroxytamoxifen with different si
chains (figure 19) was tested for estrogenic and antia
trogenic Recently a series of fixed-ring tricyclic derivatives contained tamoxifen and 4-hydroxytamoxifen with different side chains (figure 19) was tested for estrogenic and anties trogenic activity (2). The phenolic hydroxyl is a chains (figure 19) was tested for estrogenic and antieschains (figure 19) was tested for estrogenic and antitrogenic activity (2). The phenolic hydroxyl is an advetage for binding to the ER but a relatively planar (or gen-containing) tricyclic ring system is a disadvanta The a trogenic activity (2). The phenolic hydroxyl is an a
tage for binding to the ER but a relatively planar
gen-containing) tricyclic ring system is a disadva
The alkylaminoethoxy side chain confers optimal a
trogen activity i tage for binding to the ER but a relatively planar (oxy-
gen-containing) tricyclic ring system is a disadvantage.
The alkylaminoethoxy side chain confers optimal anties-
trogen activity in the hydroxy dibenzo[a,e]cyclo-oct gen-containing) tricyclic ring system is a disadvantage.
The alkylaminoethoxy side chain confers optimal anties-
trogen activity in the hydroxy dibenzo[a,e]cyclo-octenes.
A glyceryl side chain causes a decrease in antagoni The alkylaminoethoxy side chain confers optimal anties-
trogen activity in the hydroxy dibenzo[a,e]cyclo-octenes.
A glyceryl side chain causes a decrease in antagonist
activity, whereas replacement with an allyl side chain trogen activity in the hydroxy dibenzo[a,e]cyclo-octenes.
A glyceryl side chain causes a decrease in antagonist
activity, whereas replacement with an allyl side chain
causes a loss of antiestrogenic activity. This finding activity, whereas replacement with an allyl side chain causes a loss of antiestrogenic activity. This finding is particularly interesting since the compound LN 1643 and LN 2839 with only an ethyl side chain is an antiestro

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REVIEW

activity of tamoxifen. Data adapted from Robertson et al. (253).

$$
R_{1}^{\ast} - OCH_{2}CH_{2}N_{CH_{3}}^{OHS}
$$
\n
$$
- OCH_{2}CHCH_{2}OH
$$
\n
$$
OH
$$
\n
$$
- OCH_{2}CH=CH_{2}
$$
\n
$$
X = 0, S, SCH_{2} or CH_{2}CH_{2}
$$
\n
$$
TPB 4
$$

 $X = 0$, S, SCH₂ or CH₂CH₂
FIG. 19. The structure of fixed ring derivatives of triphenylethylene
with different side chains.

 $X = 0$, S, SCH₂ or CH₂CH₂
FIG. 19. The structure of fixed ring derivatives of triphenylethylene
with different side chains.
with antitumor properties in vivo (74) and in vitro (28).
It is possible that the ether oxy FIG. 19. The structure of fixed ring derivatives of triphenylethylene
with different side chains.
with antitumor properties in vivo (74) and in vitro (28) .
It is possible that the ether oxygen linking the allyl side
c The statement meaning derivatives of diphalyled point
with different side chains.
with antitumor properties in vivo (74) and in vitro (28).
It is possible that the ether oxygen linking the allyl side
chain to the phenyl ri with antitumor properties in vivo (74) and in vitro (28).
It is possible that the ether oxygen linking the allyl side
chain to the phenyl ring flexes the side chain out of the (part
area of interaction on the receptor tha

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AN
prevent estrogen action. Alternatively, the allyl side
chain may be sensitive to cleavage so that an estrogenic AN
prevent estrogen action. Alternatively, the allyl side
chain may be sensitive to cleavage so that an estrogenic
phenol is produced. AN
prevent estrogen a
chain may be sensit
phenol is produced.
The substitution event estrogen action. Alternatively, the allyl side
ain may be sensitive to cleavage so that an estrogenic
eenol is produced.
The substitution of a 4-phenolic hydroxyl in tamoxifen
nfers potent antiestrogen activity and v

prevent estrogen action. Alternatively, the allyl side chain may be sensitive to cleavage so that an estrogeniphenol is produced.
The substitution of a 4-phenolic hydroxyl in tamoxife confers potent antiestrogen activity a chain may be sensitive to cleavage so that an estrogenic
phenol is produced.
The substitution of a 4-phenolic hydroxyl in tamoxifen
confers potent antiestrogen activity and very high bind-
ing affinity for the ER (159). Th phenol is produced.
The substitution of a 4-phenolic hydroxyl in tamoxifen
confers potent antiestrogen activity and very high bind-
ing affinity for the ER (159). The catechol derivative of
tamoxifen (3,4-dihydroxytamoxife The substitution of a 4-phenolic hydroxyl in tamoxif
confers potent antiestrogen activity and very high bin
ing affinity for the ER (159). The catechol derivative
tamoxifen (3,4-dihydroxytamoxifen, figure 20) also h
a high confers potent antiestrogen activity and very high bind-
ing affinity for the ER (159). The catechol derivative of
tamoxifen (3,4-dihydroxytamoxifen, figure 20) also has
a high binding affinity for the ER, but is a weak an ing affinity for the ER (159). The catechol derivative of tamoxifen (3,4-dihydroxytamoxifen, figure 20) also has a high binding affinity for the ER, but is a weak anties-
trogen (159). Of interest though, is the finding th tamoxifen (3,4-dihydroxytamoxifen, figure 20) also has
a high binding affinity for the ER, but is a weak anties-
trogen (159). Of interest though, is the finding that 3,4-
dihydroxytamoxifen has no estrogenic properties in a high binding affinity for the ER, but is a weak anties-
trogen (159). Of interest though, is the finding that 3,4-
dihydroxytamoxifen has no estrogenic properties in the
3-day immature rat uterine weight test (159). Howe dihydroxytamoxifen has no estrogenic properties in the 3-day immature rat uterine weight test (159). However, 4-hydroxytamoxifen and 3-hydroxytamoxifen are partial agonists (267), so it is possible that the dihydroxylated dihydroxytamoxifen has no estrogenic properties in the 3-day immature rat uterine weight test (159). However, 4-hydroxytamoxifen and 3-hydroxytamoxifen are partial agonists (267), so it is possible that the dihydroxylated 3-day immature rat uterine weight test (159) . However,
4-hydroxytamoxifen and 3-hydroxytamoxifen are partial
agonists (267) , so it is possible that the dihydroxylated
metabolite is able to inhibit estradiol from bindi 4-hydroxytamoxifen and 3-hydroxytamoxifen are partial agonists (267) , so it is possible that the dihydroxylated metabolite is able to inhibit estradiol from binding to the receptor, but is short acting because it is met

A $\sqrt{GM_3}$ OCH₂CH₂N CH. R \sim / $R \n\n**R** R₁ R₂ R₃$ Compound R R₁ R₂ R₃

B

Compound	x	x,	x_{2}	Antitumor Activity
triphenylbut-I-ene (TPB)	н	н	н	NS
TPB I	CH3CO n	н	н	Weak
TPB ₂	CH ₃ CO O	CH ₃ CO H		Potent
TPB ₃	н	CH3CO H		Weak
TPB ₄	CH ₃ CO ő	н	CH ₃ CO	Potent
TPB ₅	н	CH3CO	CH₃CO	Weak

TPB 5

H CH₃CO CH₃CO Weak
 $\bigcup_{n=0}^{\infty}$ CH₃CO Weak

FIG. 20. The formula of hydroxylated derivatives of tamoxifen (part

A) and antitumor activity of acetoxy derivatives of triphenylbut-1-ene

(part B). Antitumor

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ANTIESTROGEN PHAR

response. Ruenitz et al. (267) have shown that the 4

position for substitution with a hydroxyl is ideal for both ANTIESTROGEN PHA

position for substitution with a hydroxyl is ideal for both

position for substitution with a hydroxyl is ideal for both

receptor binding and antiestrogenic activity; other sub-ANTIESTROGEN
response. Ruenitz et al. (267) have shown that the 4
position for substitution with a hydroxyl is ideal for both
receptor binding and antiestrogenic activity; other sub-
stituted positions produce compounds wi response. Ruenitz et al. (267) have shown that the 4 position for substitution with a hydroxyl is ideal for both receptor binding and antiestrogenic activity; other substituted positions produce compounds with reduced rece response. Ruenitz et al. (267) have shown that the 4 position for substitution with a hydroxyl is ideal for both receptor binding and antiestrogenic activity; other substituted positions produce compounds with reduced rece position for substitution with a hydroxyl is ideal for both
receptor binding and antiestrogenic activity; other sub-
stituted positions produce compounds with reduced re-
ceptor binding and antiestrogenic activity. A simil receptor binding and antiestrogenic activity; other
stituted positions produce compounds with reduce
ceptor binding and antiestrogenic activity. A si
relationship between estrogen receptor binding an
titumor in vivo can be ceptor binding and antiestrogenic activity. A similar relationship between estrogen receptor binding and antitumor in vivo can be made in a group of acetoxysubstituted derivatives of 1,1,2-triphenylbut-1-ene (figure 20). I ceptor binding and antiestrogenic activity. A similar relationship between estrogen receptor binding and antitumor in vivo can be made in a group of acetoxy substituted derivatives of 1,1,2-triphenylbut-1-ene (figure 20). relationship between estrogen receptor binding and antitumor in vivo can be made in a group of acetoxysubstituted derivatives of 1,1,2-triphenylbut-1-ene (figure 20).
It is an advantage to have an acetoxy group in position titumor in vivo can be made in a group of acetoxysubstituted derivatives of 1,1,2-triphenylbut-1-ene (figure 20).
It is an advantage to have an acetoxy group in position X (figure 20, TBP 2 and 4) (which is equivalent to t tuted derivatives of 1,1,2-triphenylbut-1-ene (figure 20).
It is an advantage to have an acetoxy group in position
X (figure 20, TBP 2 and 4) (which is equivalent to the
4-hydroxy group in 4-hydroxytamoxifen) for receptor
 It is an advantage to have an acetoxy group in position X (figure 20, TBP 2 and 4) (which is equivalent to the 4-hydroxy group in 4-hydroxytamoxifen) for receptor binding. Antitumor activity was assessed against the growth X (figure 20, TBP 2 and 4) (which is equivalent to the 4-hydroxy group in 4-hydroxytamoxifen) for receptor binding. Antitumor activity was assessed against the growth of ER positive human breast tumors in athymic mice (279 4-hydroxy group in 4-hydroxytamoxifen) for receptor
binding. Antitumor activity was assessed against the
growth of ER positive human breast tumors in athymic
mice (279). However, it is difficult to determine, in vivo,
whet binding. Antitumor activity was assessed against the growth of ER positive human breast tumors in athymic mice (279). However, it is difficult to determine, in vivo, whether the action of the compounds to control tumor gro growth of ER positive human breast tumors in athymic
mice (279). However, it is difficult to determine, in vivo,
whether the action of the compounds to control tumor
growth is the result of estrogenic or antiestrogenic act mice (279). However, it is difficult to determine, in vivo,
whether the action of the compounds to control tumor
growth is the result of estrogenic or antiestrogenic action.
The acetyl groups will almost certainly by hydro growth is the result of estrogenic or antiestrogenic action.
The acetyl groups will almost certainly by hydrolyzed in
vivo to present the tumor with potentially estrogenic
triphenylethylenes (171). It is known, however, t The acetyl groups will almost certainly by hydrolyzed in The acetyl groups will almost certainly by hydrolyzed in vivo to present the tumor with potentially estrogenitiphenylethylenes (171). It is known, however, that high dose estrogen therapy can be used to control the growth vivo to present the tumor with potentially estrogenic
triphenylethylenes (171). It is known, however, that high
dose estrogen therapy can be used to control the growth
of breast cancer (130, 142). A related diacetoxy com-
 pound, cyclofenyl (figure 21:5), is a full agonist in rat and mouse uterine weight tests (105), but is able to control the growth of dimethylbenzanthracene-induced rat mammary carcinomata (66). However, deacetylation of breast cancer (130, 142). A related diacetoxy comof breast cancer (130, 142). A related diacetoxy compound, cyclofenyl (figure 21:5), is a full agonist in rat and mouse uterine weight tests (105), but is able to control the growth of dimethylbenzanthracene-induced rat ma pound, cyclofenyl (figure 21:5), is a full agonist in
and mouse uterine weight tests (105), but is ablection of dimethylbenzanthracene-ind
rat mammary carcinomata (66). However, deacetyla
of cyclofenyl and introduction of and mouse uterine weight tests (105), but is able to control the growth of dimethylbenzanthracene-induced rat mammary carcinomata (66). However, deacetylation of cyclofenyl and introduction of a single pyrrolidinoe-thoxy s control the growth of dimethylbenzanthrace
rat mammary carcinomata (66). However, de
of cyclofenyl and introduction of a single p
thoxy side chain (figure 21:6) produces a par
with antietrogenic activity in the rat (105).
 t mammary carcinomata (66). However, deacetylatio
cyclofenyl and introduction of a single pyrrolidino
oxy side chain (figure 21:6) produces a partial agonis
th antietrogenic activity in the rat (105).
The cyclohexane ring of cyclofenyl and introduction of a single pyrrolidinc
thoxy side chain (figure 21:6) produces a partial agoni
with antietrogenic activity in the rat (105).
The cyclohexane ring system in the cyclofenyl mol-
cule indicates

thoxy side chain (figure 21:6) produces a partial agonist
with antietrogenic activity in the rat (105) .
The cyclohexane ring system in the cyclofenyl mole-
cule indicates that only a small spacing group is neces-
sary t with antietrogenic activity in the rat (105).
The cyclohexane ring system in the cyclofenyl mole-
cule indicates that only a small spacing group is neces-
sary to occupy the ER binding site rather than a stilbene-
like sys The cyclohexane ring system in the cyclofenyl mole-
cule indicates that only a small spacing group is neces-
sary to occupy the ER binding site rather than a stilbene-
like system. Indeed, a substituted cyclopropane system cule indicates that only a small spacing group is necessary to occupy the ER binding site rather than a stilbene-
like system. Indeed, a substituted cyclopropane system
will suffice as a skeleton for compounds with high af like system. Indeed, a substituted cyclopropane system
will suffice as a skeleton for compounds with high affinity
for the ER and estrogenic activity (221). An example of
a *trans* substituted compound is ilustrated in fig will suffice as a skeleton for compounds with high affinity
for the ER and estrogenic activity (221) . An example of
a *trans* substituted compound is ilustrated in figure 21:7.
However, of some significance is the findi for the ER and estrogenic activity (221) . An example of a *trans* substituted compound is ilustrated in figure $21:7$. However, of some significance is the finding that the *cis* substituted derivative, known as analog a *trans* substituted compound is ilustrated in figure 21:7.
However, of some significance is the finding that the *cis*
substituted derivative, known as analog II (figure 21:8),
has low estrogenic activity in the mouse ut However, of some significance is the finding that the *cis* substituted derivative, known as analog II (figure 21:8), has low estrogenic activity in the mouse uterine weight test and weak antiestrogenic activity (248) a substituted derivative, known as analog II (figure 21:8),
has low estrogenic activity in the mouse uterine weight
test and weak antiestrogenic activity (248) and antitumor
activity against dimethylbenzathracene-induced tu has low estrogenic activity in the mouse uterine weight test and weak antiestrogenic activity (248) and antitumor activity against dimethylbenzathracene-induced tumors (247). The chlorine atoms are important as spacing gro test and weak antiestrogenic activity (248) and antitumor
activity against dimethylbenzathracene-induced tumors
(247). The chlorine atoms are important as spacing
groups because their removal destroys all biological (es-
t activity against dimethylbenzathracene-induced tumors (247). The chlorine atoms are important as spacing groups because their removal destroys all biological (estrogenic and antiestrogenic) activity. This finding is quite (247). The chlorine atoms are important as spacing $\frac{1}{2}$ groups because their removal destroys all biological (estrogenic and antiestrogenic) activity. This finding is quite significant when the biological properties groups because their removal destroys all biological (es-
trogenic and antiestrogenic) activity. This finding is of
quite significant when the biological properties of other
simple chlorinated compounds are considered. Ch trogenic and antiestrogenic) activity. This findit quite significant when the biological properties of c simple chlorinated compounds are considered. Chlor atoms are probably required as spacing groups in estrogenic insect quite significant when the biological properties of other com
simple chlorinated compounds are considered. Chlorine S
atoms are probably required as spacing groups in the limit
estrogenic insecticide methoxychlor[2,2 bis simple chlorinated compounds are considered. Chlorine
atoms are probably required as spacing groups in the
estrogenic insecticide methoxychlor[2,2 bis $(p$ -methoxy-
phenyl]-1,1,1-trichloroethane] which is demethylated in
v atoms are probably required as spacing groups in the
strogenic insecticide methoxychlor[2,2 bis $(p$ -methoxy
phenyl)-1,1,1-trichloroethane] which is demethylated i
vivo to the bisphenolic compound (35) . This metaboli
act estrogenic insecticide methoxy
phenyl)-1,1,1-trichloroethane]
vivo to the bisphenolic compo
activation is probably necessar
ing in the target tissue (243).
R. Studies in Vitro *B.* Studies is probably not
ing in the target tissue (
B. Studies in Vitro
Structure-activity rela tivation is probably necessary to permit receptor bind
g in the target tissue (243).
Studies in Vitro
Structure-activity relationship studies in vivo are com-
cated by the potential metabolism of the compound

ing in the target tissue (243).

B. Studies in Vitro

Structure-activity relationship studies in vivo are com-

plicated by the potential metabolism of the compound

and the relative pharmacokinetics of the parent com-B. Studies in Vitro
Structure-activity relationship studies in vivo are com-
plicated by the potential metabolism of the compound
and the relative pharmacokinetics of the parent com-
pound and its metabolites. An antiestro B. Studies in Vitro
Structure-activity relationship studies in vivo are com-
plicated by the potential metabolism of the compound
and the relative pharmacokinetics of the parent com-
pound and its metabolites. An antiestro

FIG. 21. Nonsteroidal estrogens and antiestrogens.

tabolized to a mixture of estrogens and antiestrogens (9) (10)
FIG. 21. Nonsteroidal estrogens and antiestrogens.
tabolized to a mixture of estrogens and antiestrogens
which interact in different proportions at single or mul-
tiple receptor sites within a given target tiss FIG. 21. Nonsteroidal estrogens and antiestrogens.
tabolized to a mixture of estrogens and antiestro
which interact in different proportions at single or i
tiple receptor sites within a given target tissue. Struct
activity rion and consider and and antiestrogens
tabolized to a mixture of estrogens and antiestrogens
which interact in different proportions at single or mul-
tiple receptor sites within a given target tissue. Structure-
activity tabolized to a mixture of estrogens and antiestrogens
which interact in different proportions at single or mul-
tiple receptor sites within a given target tissue. Structure-
activity relationship studies in vitro can circu which interact in different protiple receptor sites within a given
tiple receptor sites within a given
activity relationship studies in
of these problems and dissect compound and its metabolites
Studies of antiestrogens in ble receptor sites within a given target tissue. Structure-
tivity relationship studies in vitro can circumvent some
these problems and dissect out the action of the parent
mpound and its metabolites.
Studies of antiestrog activity relationship studies in vitro can circumvent some
of these problems and dissect out the action of the parent
compound and its metabolites.
Studies of antiestrogens in vitro are restricted by the
limited number of

of these problems and dissect out the action of the parent
compound and its metabolites.
Studies of antiestrogens in vitro are restricted by the
limited number of estrogen-responsive test systems.
However, two systems have compound and its metabolites.

Studies of antiestrogens in vitro are restricted l

limited number of estrogen-responsive test sy:

However, two systems have been studied: (a) the g

of MCF-7 breast cancer cells; and (b) es Studies of antiestrogens in vitro are restricted by the
limited number of estrogen-responsive test systems.
However, two systems have been studied: (a) the growth
of MCF-7 breast cancer cells; and (b) estrogen-stimu-
lated limited number of estrogen-r
However, two systems have bee
of MCF-7 breast cancer cells;
lated prolactin synthesis by 1
immature rat pituitary glands.
The MCF-7 breast cancer ce owever, two systems have been studied: (a) the growth
MCF-7 breast cancer cells; and (b) estrogen-stimu-
ted prolactin synthesis by primary cell cultures of
mature rat pituitary glands.
The MCF-7 breast cancer cell system of MCF-7 breast cancer cells; and (b) estrogen-stimu-
lated prolactin synthesis by primary cell cultures of
immature rat pituitary glands.
The MCF-7 breast cancer cell system has been used
to determine the effects of tamox

lated prolactin synthesis by primary cell cultures of immature rat pituitary glands.
The MCF-7 breast cancer cell system has been used
to determine the effects of tamoxifen and its metabolites
on cell proliferation (61). A immature rat pituitary glands.
The MCF-7 breast cancer cell system has been
to determine the effects of tamoxifen and its metabo
on cell proliferation (61). As might be expected from
relative binding affinity for the recep The MCF-7 breast cancer cell system has been used
to determine the effects of tamoxifen and its metabolites
on cell proliferation (61). As might be expected from the
relative binding affinity for the receptor, 4-hydroxytato determine the effects of tamoxifen and its metabolites
on cell proliferation (61). As might be expected from the
relative binding affinity for the receptor, 4-hydroxyta-
moxifen is a more potent inhibitor than tamoxifen

metabolite has a low potency as an antiproliferative agent, whereas a low potency as an antiproliferative metabolite has a low potency as an antiproliferative agent, whereas Reddel and coworkers (250) have demonstrated that concentrations of N-desmethyltamoxife 266
metabolite has a low potency as an antiproliferative agent, whereas Reddel and coworkers (250) have den
onstrated that concentrations of N-desmethyltamoxife
as high as 10 μ M are extremely effective, apparent metabolite has a low potency as an antiproliferative agent, whereas Reddel and coworkers (250) have demonstrated that concentrations of N-desmethyltamoxifen as high as 10 μ M are extremely effective, apparently more so agent, whereas Reddel and coworkers (250) have demonstrated that concentrations of N-desmethyltamoxifen as high as 10 μ M are extremely effective, apparently more so than either tamoxifen or 4-hydroxytamoxifen. The *cis* onstrated that concentrations of N-desmethyltamoxides high as 10 μ M are extremely effective, apparen more so than either tamoxifen or 4-hydroxytamoxife. The *cis* geometric isomer of tamoxifen, ICI 47,699, ineffective as high as 10 μ M are extremely effective, apparently tree more so than either tamoxifen or 4-hydroxytamoxifen. It The *cis* geometric isomer of tamoxifen, ICI 47,699, is essented in effective as an antiproliferative ag more so than either tamoxifen or 4-hydroxytamoxifen.
The cis geometric isomer of tamoxifen, ICI 47,699, is
ineffective as an antiproliferative agent (61) and stimu-
lates MCF-7 cell proliferation (182). However, the cis
g The *cis* geometric isomer of tamoxifen, ICI 47,699, is
ineffective as an antiproliferative agent (61) and stimu-
lates MCF-7 cell proliferation (182). However, the *cis*
geometric isomer of enclomiphene, zuclomiphene, is ineffective as an antiproliferative agent (61) and stimu-
lates MCF-7 cell proliferation (182). However, the cis
geometric isomer of enclomiphene, zuclomiphene, is ap-
parently extremely effective as an antiproliferative lates MCF-7 cell proliferation (182). However, the geometric isomer of enclomiphene, zuclomiphene, is parently extremely effective as an antiproliferative ag at a concentration of 5μ M (235). This observation is interes geometric isomer of enclomiphene, zuclomiphene, is ap-
parently extremely effective as an antiproliferative agent 4-1
at a concentration of 5 μ M (235). This observation is of mo
interest because a similar cytotoxic eff (211). a concentration of $5 \mu M$ (235). This observation is of
terest because a similar cytotoxic effect for zuclomi-
nene has been reported in the prolactin-synthesis assay
11).
The antitumor activity of several clomiphene anal

interest because a similar cytotoxic effect for zuclo
phene has been reported in the prolactin-synthesis as
(211).
The antitumor activity of several clomiphene anal
has been tested: 9,599 (mono de-ethylated enclo
phene), 6 phene has been reported in the prolactin-synthesis assay

(211).

The antitumor activity of several clomiphene analogs

has been tested: 9,599 (mono de-ethylated enclomi-

phene), 6,866 (enclomiphene with an addition of c (211).
The antitumor activity of several clomiphene anal
has been tested: $9,599$ (mono de-ethylated enclo
phene), $6,866$ (enclomiphene with an addition of carl
in the aminoethoxy side chain), and $10,222$ (enclo
phene w The antitumor activity of several clomiphene analogs of
has been tested: 9,599 (mono de-ethylated enclomi-
phene), 6,866 (enclomiphene with an addition of carbon
in the aminoethoxy side chain), and 10,222 (enclomi-
phene has been tested: 9,599 (mono de-ethylated enclomi-
phene), 6,866 (enclomiphene with an addition of carbon to the
in the aminoethoxy side chain), and 10,222 (enclomi-
phene with nitrogen substituted for the ether oxygen of phene), 6,866 (enclomiphene with an addition of carbon to the estrogen receptor (9, 212), and the derivatives of
in the aminoethoxy side chain), and 10,222 (enclomi-
phene with nitrogen substituted for the ether oxygen of in the aminoethoxy side chain), and 10,222 (enclomitation
phene with nitrogen substituted for the ether oxygen of sist
the side chain) (235). At low concentrations (0.25 to 1.0 rec
 μ M), where the growth-inhibitory effe phene with nitrogen substituted for the ether oxygen of
the side chain) (235). At low concentrations (0.25 to 1.0
 μ M), where the growth-inhibitory effects are reversed by
estradiol, the relative antitumor activity (6,8 the side chain) (235). At low concentrations (0.25 to 1.0 recomplement), where the growth-inhibitory effects are reversed by be restration, the relative antitumor activity (6,866 > 10,222 required by a percompleme > 9,599 μ M), where the growth-inhibitory effects are reversed by
estradiol, the relative antitumor activity (6,866 > 10,222 \rightarrow
> enclomiphene > 9,599) was in the same order as their
relative binding affinities for the ER. Th $>$ enclomiphene $>$ 9,599) was in the same order as their relative binding affinities for the ER. There appears to be no correlation of antitumor activity with affinity for the antiestrogen binding sites.

relative binding affinities for the ER. There ap
be no correlation of antitumor activity with aff
the antiestrogen binding sites.
A similar structure-activity study has been unc
with a small series of substituted bromotrip be no correlation of antitumor activity with affinity for
the antiestrogen binding sites.
A similar structure-activity study has been undertaken
with a small series of substituted bromotriphenylethy-
ity relations:
LN 1643 the antiestrogen binding sites.

A similar structure-activity study has been undertaken

with a small series of substituted bromotriphenylethy-

itenes: LN 1643 (figure 8), LN 2299 (cis isomer of LN

1643, figure 21.3), L A similar structure-activity study has been undertal
with a small series of substituted bromotriphenylet
lenes: LN 1643 (figure 8), LN 2299 (*cis* isomer of 1
1643, figure 21.3), LN 2833 (LN 1643 with a secon
1643, figure with a small series of substituted bromotriphenylethy
lenes: LN 1643 (figure 8), LN 2299 (cis isomer of L1
1643, figure 21.3), LN 2839 (the hydroxy metabolite of
1643, figure 21.4), and LN 2833 (LN 1643 with a secono
ary a lenes: LN 1643 (figure 8), LN 2299 (*cis* isomer of LN 1643, figure 21.3), LN 2839 (the hydroxy metabolite of binding 1643, figure 21.4), and LN 2833 (LN 1643 with a secondary alcohol substitution in the ethyl side chain) 1643, figure 21.3), LN 2839 (the hydroxy metabolite of 1643, figure 21.4), and LN 2833 (LN 1643 with a secondery alcohol substitution in the ethyl side chain). Antitium or activity is directly correlated with the binding 1643, figure 21.4), and LN 2833 (LN 1643 with a second-

ary alcohol substitution in the ethyl side chain). Anti-

tumor activity is directly correlated with the binding

affinity for the receptor: 4-hydroxytamoxifen > LN ary alcohol substitution in the ethyl side chain). Anti-
tumor activity is directly correlated with the binding
affinity for the receptor: 4-hydroxytamoxifen > LN 2839
> LN 1643 (28). LN 2299 and 2839 have very low
affini tumor activity is directly correlated with the binding
affinity for the receptor: 4-hydroxytamoxifen > LN 2839
> LN 1643 (28). LN 2299 and 2839 have very low
affinities for the receptor but no antitumor effects have
been r affinity for the receptor: 4-hydroxytamoxifen > LN 2839

> LN 1643 (28). LN 2299 and 2839 have very low

affinities for the receptor but no antitumor effects have

been reported (28). LN 2299 is an estrogen of equivalent
 $>$ LN 1643 (28). LN 2299 and 2839 have very low affinities for the receptor but no antitumor effects have in been reported (28). LN 2299 is an estrogen of equivalent potency to zuclomiphene in the prolactin synthesis ass affinities for the receptor but no antitumor effection reported (28). LN 2299 is an estrogen of equations) potency to zuclomiphene in the prolactin synths, says and LN 2833 is a weak antiestrogen (V. C and M. E. Lieberman, Example 128). LN 2299 is an estrogen of equivalent tency to zuclomiphene in the prolactin synthesis as-
ys and LN 2833 is a weak antiestrogen (V. C. Jordan in the MCF-7 breast cancer cells out that MCF-7 breast cancer cel potency to zuclomiphene in the prolactin synthesis a
says and LN 2833 is a weak antiestrogen (V. C. Jorda
and M. E. Lieberman, unpublished observations).
It is fair to point out that MCF-7 breast cancer cel
in culture have

says and LN 2833 is a weak antiestrogen (V. C. Jordan and M. E. Lieberman, unpublished observations).
It is fair to point out that MCF-7 breast cancer cells in culture have not been extensively used to study structure act It is fair to point out that MCF-7 breast cancer cells
in culture have not been extensively used to study struc-
ture activity relationships; however, LeClercq and co-
workers (202) have recently presented some interesting in culture have not been extensively used to study structure activity relationships; however, LeClercq and co-
workers (202) have recently presented some interesting
SAR data. The bis phenolic dichloroethylene (figure
21:9 ture activity relationships; however, LeClercq and co-
workers (202) have recently presented some interesting
SAR data. The bis phenolic dichloroethylene (figure
21:9) is estrogenic and able to reverse the antitumor
action workers (202) have recently presented some interesting SAR data. The bis phenolic dichloroethylene (figure 21:9) is estrogenic and able to reverse the antitumor action of nafoxidine. As might be predicted, substitution $\$ SAR data. The bis phenolic dichloroethylene (figure 21:9) is estrogenic and able to reverse the antitumor action of nafoxidine. As might be predicted, substitution of one phenolic group with an alkylaminoethyl side chain (21:9) is estrogenic and able to reverse the antitumor
action of nafoxidine. As might be predicted, substitution
of one phenolic group with an alkylaminoethyl side chain
(figure 21:10) reduces estrogenic activity but incre action of nafoxidine. As might be predicted, substitutio
of one phenolic group with an alkylaminoethyl side chai
(figure 21:10) reduces estrogenic activity but increase
antiproliferative properties. The structural similari % of one phenolic group
 $(figure 21:10)$ reduce
 $antiproliferative$ prop

these compounds wit

trated in figure 21.

Overall the MCF-7 gure 21:10) reduces estrogenic activity but increases
tiproliferative properties. The structural similarity of
ese compounds with cyclofenyl and analog II is illus-
ated in figure 21.
Overall the MCF-7 system for assay app

antiproliferative properties. The structural similarity of
these compounds with cyclofenyl and analog II is illus-
trated in figure 21.
Overall the MCF-7 system for assay appears to produce
results very similar to the pro these compounds with cyclofenyl and analog II is illus-
trated in figure 21.
Overall the MCF-7 system for assay appears to produce
results very similar to the prolactin synthesis assay which
indicates that the compounds m trated in figure 21.

Overall the MCF-7 system for ass

results very similar to the prolactin

indicates that the compounds mi

mechanism of action via the ER.

more so than either tamoxifen or 4-hydroxytamoxifen. ture-activity relationships within groups of nonsteroidal
The *cis* geometric isomer of tamoxifen, ICI 47,699, is estrogens and antiestrogens. The antiestrogens, tamoxi-Primary cultures of rat pituitary cells respond to phys-AN
Primary cultures of rat pituitary cells respond to phys-
iological concentrations of estradiol by a specific increase
in prolactin synthesis (213). This model system for es-AN
Primary cultures of rat pituitary cells respond to phys-
iological concentrations of estradiol by a specific increase
in prolactin synthesis (213). This model system for es-
trogen action has been validated for the stud Frimary cultures of rat pituitary cells respond to phy
iological concentrations of estradiol by a specific increa
in prolactin synthesis (213). This model system for e
trogen action has been validated for the study of stru Primary cultures of rat pituitary cells respond to physiological concentrations of estradiol by a specific increase
in prolactin synthesis (213). This model system for es
trogen action has been validated for the study of s iological concentrations of estradiol by a specific incre
in prolactin synthesis (213). This model system for
trogen action has been validated for the study of str
ture-activity relationships within groups of nonsteroi
est trogen action has been validated for the study of structure-activity relationships within groups of nonsteroidal estrogens and artiestrogens. The antiestrogens, tamoxifen and 4-hydroxytamoxifen, inhibit estradiol-stimu-lat lated prolactin synthesis (212). Their potencies are conture-activity relationships within groups of nonsteroidal estrogens and antiestrogens. The antiestrogens, tamoxi-
fen and 4-hydroxytamoxifen, inhibit estradiol-stimu-
lated prolactin synthesis (212). Their potencies are co estrogens and antiestrogens. The antiestrogens, tamoxi-
fen and 4-hydroxytamoxifen, inhibit estradiol-stimu-
lated prolactin synthesis (212). Their potencies are con-
sistent with their relative binding affinities for the fen and 4-hydroxytamoxifen, inhibit estradiol-stimulated prolactin synthesis (212) . Their potencies are con sistent with their relative binding affinities for the ER 4-hydroxytamoxifen is 30 times more potent than tamox lated prolactin synthesis (212). Their potencies are consistent with their relative binding affinities for the ER;
4-hydroxytamoxifen is 30 times more potent than ta-
moxifen. To avoid the possibility that tamoxifen is me sistent with their relative binding affinities for the ER
4-hydroxytamoxifen is 30 times more potent than ta
moxifen. To avoid the possibility that tamoxifen is met
abolically activated to 4-hydroxytamoxifen in vitro, sev
 4-hydroxytamoxifen is 30 times more potent than
moxifen. To avoid the possibility that tamoxifen is m
abolically activated to 4-hydroxytamoxifen in vitro, s
eral para substituted derivatives of tamoxifen (p-meth
p-chloromoxifen. To avoid the possibility that tamoxifen is met-
abolically activated to 4-hydroxytamoxifen in vitro, sev-
eral *para* substituted derivatives of tamoxifen (*p*-methyl-,
p-chloro-, and *p*-fluoro-) that are unli eral para substituted derivatives of tamoxifen (p-methyl-, p -chloro-, and p -fluoro-) that are unlikely to be metabtamoxifen inhibit estradiol-stimulated prolactin syntheolized to 4-hydroxytamoxifen (9) have been tested. The
substitution does not affect the binding of the compounds
to the estrogen receptor (9, 212), and the derivatives of
tamoxifen inhibit estradiol-stimulated prolactin sy substitution does not affect the binding of the compounds
to the estrogen receptor (9, 212), and the derivatives of
tamoxifen inhibit estradiol-stimulated prolactin synthe-
sis consistent with their relative binding affini to the estrogen receptor $(9, 212)$, and the derivatives of tamoxifen inhibit estradiol-stimulated prolactin synthesis consistent with their relative binding affinities for the receptor. Although it is an advantage for ta tamoxifen inhibit estradiol-stimulated prolactin s
sis consistent with their relative binding affinities
receptor. Although it is an advantage for tamox
be metabolized to 4-hydroxytamoxifen, it is clearl
requirement for an s consistent with their relative binding affinities for the ceptor. Although it is an advantage for tamoxifen to metabolized to 4-hydroxytamoxifen, it is clearly not a quirement for antiestrogenic activity (9, 212).
Anties

the antiestrogen binding sites. The system of known estrogens and antiestrogens has been tested in
A similar structure-activity study has been undertaken the system to establish its usefulness for structure-activ-
with a s receptor. Although it is an advantage for tamoxifen to
be metabolized to 4-hydroxytamoxifen, it is clearly not a
requirement for antiestrogenic activity $(9, 212)$.
Antiestrogen action in the pituitary cells has been
show be metabolized to 4-hydroxytamoxifen, it is clearly not a
requirement for antiestrogenic activity (9, 212).
Antiestrogen action in the pituitary cells has been
shown to be both competitive and reversible with the
addition requirement for antiestrogenic activity (9, 212).
Antiestrogen action in the pituitary cells has been
shown to be both competitive and reversible with the
addition of excess estradiol (212). Furthermore, a series
of known Antiestrogen action in the pituitary cells has
shown to be both competitive and reversible wire
addition of excess estradiol (212). Furthermore, a
of known estrogens and antiestrogens has been tee
the system to establish shown to be both competitive and reversible with the addition of excess estradiol (212). Furthermore, a series of known estrogens and antiestrogens has been tested in the system to establish its usefulness for structure-ac addition of excess estradiol (212). Furthermore, a series
of known estrogens and antiestrogens has been tested in
the system to establish its usefulness for structure-activ-
ity relationship studies (211). The biological p of known estrogens and antiestrogens has been tested in
the system to establish its usefulness for structure-activ-
ity relationship studies (211). The biological potency of
the binding ligands is directly related to their the system to establish its usefulness for structure-activ relationship studies (211). The biological potency the binding ligands is directly related to their relationing affinity for the ER. The relative potency estrogen ity relationship studies (211). The biological potency of
the binding ligands is directly related to their relative
binding affinity for the ER. The relative potency of
estrogens to stimulate prolactin synthesis was dieth the binding ligands is directly related to their relative
binding affinity for the ER. The relative potency of
estrogens to stimulate prolactin synthesis was diethyl-
stilbestrol; \equiv estradiol > ICI 77,949 (tamoxifen wi binding affinity for the ER. The relative potency estrogens to stimulate prolactin synthesis was diethy stilbestrol; \equiv estradiol > ICI 77,949 (tamoxifen withoutis dimethyl aminoethane side chain) > ICI 47,699 (cisomer estrogens to stimulate prolactin synthesis was diethyl-
stilbestrol; \equiv estradiol > ICI 77,949 (tamoxifen without
its dimethyl aminoethane side chain) > ICI 47,699 (*cis*
isomer of tamoxifen) \equiv zuclomiphene (*cis* is stilbestrol; \equiv estradiol > ICI 77,949 (tamoxifen without its dimethyl aminoethane side chain) > ICI 47,699 (*cis* isomer of tamoxifen) \equiv zuclomiphene (*cis* isomer ofen-clomiphene). The relative potencies of antiest isomer of tamoxifen) = zuclomiphene (*cis* isomer ofen-
clomiphene). The relative potencies of antiestrogens to
inhibit estradiol-stimulated prolactin synthesis was *trans*
4-hydroxytamoxifen = LY 117018 > trioxifene > en clomiphene). The relative potencies of antiestrogens to

inhibit estradiol-stimulated prolactin synthesis was *trans*

4-hydroxytamoxifen = LY 117018 > trioxifene > enclom-

iphene = tamoxifen. The compound LY 126412 (tri inhibit estradiol-stimulated prolactin synthesis was *trans*
4-hydroxytamoxifen = LY 117018 > trioxifene > enclom-
iphene = tamoxifen. The compound LY 126412 (trioxi-
fene without the side chain) does not interact with ER 4-hydroxytamoxifen \equiv LY 117018 > trioxifene > enclom-
iphene \equiv tamoxifen. The compound LY 126412 (trioxi-
fene without the side chain) does not interact with ER
up to test concentrations of 10^{-6} M or exhibit estr iphene \equiv tamoxifen. The compound LY 126412 (trioxi-
fene without the side chain) does not interact with ER
up to test concentrations of 10^{-6} M or exhibit estrogenic
or antiestrogenic properties with the prolactin sy

FIG. 22. A general ligand model to describe the structural require- ment to control biological activity in vitro.

aspet

occurring. FIG. 23. Hypothetic models to describe the binding of estradiol or 4-hydroxytamoxifen with the ligand binding site

induce a conformational change in the receptor to lock the ligand into the receptor whereas the antiestrog

can induce a conformational change in the receptor to lock the ligand into
occurring.
The prolactin-synthesis assay system is currently C
being used to evaluate the structural requirements for
estrogen and antiestrogen act occurring.

The prolactin-synthesis assay system is currently C .

being used to evaluate the structural requirements for

estrogen and antiestrogen action. The compounds in

figure 20 are proving to be of particular int The prolactin-synthesis assay system is currently C
being used to evaluate the structural requirements for
estrogen and antiestrogen action. The compounds in
figure 20 are proving to be of particular interest. 4-
Hydrox The prolactin-synthesis assay system is curren
being used to evaluate the structural requirements
estrogen and antiestrogen action. The compounds
figure 20 are proving to be of particular interest.
Hydroxytamoxifen is the being used to evaluate the structural requirements for
estrogen and antiestrogen action. The compounds in
figure 20 are proving to be of particular interest. 4-
Hydroxytamoxifen is the optimal substitution for tamox-
ifen; figure 20 are proving to be of particular interest. 4

Hydroxytamoxifen is the optimal substitution for tamoxical content drugs, the substituents that determine potency are

iferent from those that determine pharmacologic a low potency as an antiestrogen in vitro because it is
a low potency as an antiestrogen in vitro because it is
unstable. Nevertheless, much increased antiestrogenic
 $\frac{1}{2}$. The dimensions of the ED binding site are gr Hydroxytamoxiten is the optimal substitution for tamox-
ifen; however, 3,4-hydroxytamoxifen, though it has a
high binding affinity for the estrogen receptor (159), has
a low potency as an antiestrogen in vitro because it i Then; however, 3,4-hydroxytamoxiten, though it has a
high binding affinity for the estrogen receptor (159), has
a low potency as an antiestrogen in vitro because it is
unstable. Nevertheless, much increased antiestrogenic migh binding affinity for the estrogen receptor (159), has
a low potency as an antiestrogen in vitro because it is
unstable. Nevertheless, much increased antiestrogenic
potency is observed if the assay in vitro is conduct a low potency as an antiestrogen in vitro because it
unstable. Nevertheless, much increased antiestroge
potency is observed if the assay in vitro is conducted
media containing ascorbic acid, as an antioxidant a
U-0521, an unstable. Nevertheless, much increased antiestrogenic potency is observed if the assay in vitro is conducted in and media containing ascorbic acid, as an antioxidant and with U-0521, an inhibitor of catechol orthomethyltr media containing ascorbic acid, as an antioxidant and U-0521, an inhibitor of catechol orthomethyltransferase (COMT). The COMT inhibitor, U-0521, is known to stabilize the assay of catechol estrogens in vitro (129). The su edia containing ascorbic acid, as an antioxidant and
 -0.521 , an inhibitor of catechol orthomethyltransferase
 \overline{OMT}). The COMT inhibitor, U-0521, is known to

abilize the assay of catechol estrogens in vitro (129).

U-0521, an inhibitor of catechol orthomethyltransferase
(COMT). The COMT inhibitor, U-0521, is known to
stabilize the assay of catechol estrogens in vitro (129).
The substituted triphenylbut-1-enes are uniformally
estroge (COMT). The COMT inhibitor, U-0521, is known to stabilize the assay of catechol estrogens in vitro (129).
The substituted triphenylbut-1-enes are uniformally estrogenic as long as X_2 remains unoccupied (figure 20).
Wha stabilize the assay of catechol estrogens in vitro (129).
The substituted triphenylbut-1-enes are uniformally
estrogenic as long as X_2 remains unoccupied (figure 20).
What is particularly interesting though, is that *b* The substituted triphenylbut-1-enes are uniformally
estrogenic as long as X_2 remains unoccupied (figure 20)
What is particularly interesting though, is that *bis* sub
stitution at X and X_2 (TPB4) with acetoxy groups estrogenic as long as X_2 remains unoccupied (figure 20).
What is particularly interesting though, is that *bis* substitution at X and X_2 (TPB4) with acetoxy groups produced an antiestrogen. However, the deacetylated What is particularly interesting though, is that *bis* sulstitution at X and X_2 (TPB4) with acetoxy groups produced an antiestrogen. However, the deacetylated con-
pound is a partial agonist with antiestrogenic propert stitution at X and X_2
duced an antiestrogen
pound is a partial ago.
(170). A similar relatio
pound 5, figure 21).
Overall, compounds ced an antiestrogen. However, the deacetylated com
und is a partial agonist with antiestrogenic propertie
70). A similar relationship occurs with cyclofenyl (com
und 5, figure 21).
Overall, compounds can be classified into

pound is a partial agonist with antiestrogenic properties
(170). A similar relationship occurs with cyclofenyl (com-
pound 5, figure 21).
Overall, compounds can be classified into three cate-
gories based upon their struct (170). A similar relationship occurs with cyclorenyl (compound 5, figure 21).

Overall, compounds can be classified into three categories based upon their structure (170) . Antiestrogens

have a side chain extending aw pound 5, figure 21).

Overall, compounds can be classified into three categories based upon their structure (170). Antiestrogens

have a side chain extending away from the binding site.

Partial agonists have a *bis* pheno Overall, compounds can be classified into three cate
gories based upon their structure (170). Antiestrogen
have a side chain extending away from the binding site
Partial agonists have a bis phenolic structure and ago
nists gories based upon their structure (170). Antiestrogens of antiestrogens (tamoxifen) compared to the less
have a side chain extending away from the binding site. rigid triphenylethane (MRL37) and triphenylethanol
Partial ag be described.

the receptor whereas the antiestrogen prevents these changes from
The Antiestrogenic Ligand
When considering the design of an antiestrogen, sev-
al features are dominant (figure 22). However, as with C. The Antiestrogenic Ligand
When considering the design of an antiestrogen, sev-
eral features are dominant (figure 22). However, as with
other drugs, the substituents that determine potency are C. The Antiestrogenic Ligand
When considering the design of an antiestrogen, sev-
eral features are dominant (figure 22). However, as with
other drugs, the substituents that determine potency are
different from those that When considering the design of an antiestrogen, several features are dominant (figure 22). However, as with other drugs, the substituents that determine potency are different from those that determine pharmacological activ tivity. other drugs, the substituents that determine potency are
different from those that determine pharmacological ac-
tivity.
1. There is a broad range of compounds that bind to
the ER and produce an estrogenic response in vivo

Fierent from those that determine pharmacological ac-
ity.
1. There is a broad range of compounds that bind to
e ER and produce an estrogenic response in vivo (171).
2. The dimensions of the ER binding site are specific
d tivity.

1. There is a broad range of compounds that bind to

the ER and produce an estrogenic response in vivo (171).

2. The dimensions of the ER binding site are specific

and precise. DES is a good example of a simple 1. There is a broad range of compounds that bind to
the ER and produce an estrogenic response in vivo (171).
2. The dimensions of the ER binding site are specific
and precise. DES is a good example of a simple compound
wit activity. 2. The dimensions of the ER binding site are specific d precise. DES is a good example of a simple compound th a high affinity for the ER and potent estrogenic tivity.
3. A phenolic hydroxy, equivalent to the C_3 phenol

with a high affinity for the ER and potent estrogenic
activity.
3. A phenolic hydroxy, equivalent to the C_3 phenol of
estradiol, is extremely important for high affinity binding
to the ER. This structural feature permi with a high affinity for the ER and potent estrogenic
activity.
3. A phenolic hydroxy, equivalent to the C_3 phenol of
estradiol, is extremely important for high affinity binding
to the ER. This structural feature permi activity.

3. A phenolic hydroxy, equivalent to the C_3 phenol of

estradiol, is extremely important for high affinity binding

to the ER. This structural feature permits a variety of

"spacing groups" to occupy the rec 3. A phenolic hydrestradiol, is extremely
to the ER. This stru
"spacing groups" to
(phenyl, cyclohexane
4. Alkyl ethers on tradiol, is extremely important for high affinity binding
the ER. This structural feature permits a variety of
pacing groups" to occupy the receptor binding site
henyl, cyclohexane).
4. Alkyl ethers on ring A (figure 22) h

to the ER. This structural feature permits a variety of "spacing groups" to occupy the receptor binding site (phenyl, cyclohexane).

4. Alkyl ethers on ring A (figure 22) have a decreased affinity for the receptor, but an "spacing groups"
(phenyl, cyclohe
4. Alkyl ether
affinity for the
action in vivo.
5. Depending

5. (phenyl, cyclohexane).

4. Alkyl ethers on ring A (figure 22) have a decreased

affinity for the receptor, but an increased duration of

action in vivo.

5. Depending upon the substitutions, triphenyl ethyl-

enes can p affinity for the receptor, but an increased duration
action in vivo.
5. Depending upon the substitutions, triphenyl eth
enes can possess estrogenic or antiestrogenic activi
The increased potency of the triphenylethylene-ty action in vivo.

5. Depending upon the substitutions, triphenyl ethyl-

enes can possess estrogenic or antiestrogenic activity.

The increased potency of the triphenylethylene-type

of antiestrogens (tamoxifen) compared to 5. Depending upon the substitutions, triphenyl eth
enes can possess estrogenic or antiestrogenic activi
The increased potency of the triphenylethylene-ty
of antiestrogens (tamoxifen) compared to the le
rigid triphenylethan The increased potency of the triphenylethylene-type The increased potency of the triphenylethylene-type
of antiestrogens (tamoxifen) compared to the less
rigid triphenylethane (MRL37) and triphenylethanol
(MER25) derivatives is at the cost of increased estrogenic
activity s of antiestrogens

rigid triphenyle

(MER25) deriva

activity since all

partial agonists.

6. Substitutio gid triphenylethane (MRL37) and triphenylethanol
1ER25) derivatives is at the cost of increased estrogenic
tivity since all the triphenyl ethylene derivatives are
rtial agonists.
6. Substitution of spacing groups (phenyl)

PHARMACOLOGI

JORDAN
OH or OCH₃ does not have a major impact upon potency gressively lower affinities for the estrogen receptor than
or pharmacological activity. estradiol, their log dose-response curves in an assay 268
OH or OCH₃ does not have a
or pharmacological activity.
7. Substitutions on pheny

8
H or OCH₃ does not have a major impact upon pote
pharmacological activity.
7. Substitutions on phenyl ring B governs pharm
gical activity. Compounds without substitution are OH or OCH₃ does not have a major impact upon potency
or pharmacological activity.
7. Substitutions on phenyl ring B governs pharmaco-
logical activity. Compounds without substitution are es-
trogens in vivo and in vitro: OH or OCH₃ does not have a major impact upon potency
or pharmacological activity. est
7. Substitutions on phenyl ring B governs pharmaco-
logical activity. Compounds without substitution are es-
dicogens in vivo and in or pharmacological activity.

7. Substitutions on phenyl ring B governs pharmaco-

logical activity. Compounds without substitution are es-

trogens in vivo and in vitro: (a) A para hydroxy on ring

B predicts estrogenic a logical activity. Compounds without substitution are es-
trogens in vivo and in vitro: (a) A *para* hydroxy on ring
B predicts estrogenic activity in vivo but partial agonist
activity in vitro. (b) Extention of an alkylami logical activity. Compounds without substitution are es-
trogens in vivo and in vitro: (a) A para hydroxy on ring
B predicts estrogenic activity in vivo but partial agonist
activity in vitro. (b) Extention of an alkylamino trogens in vivo and in vitro: (a) A para hydroxy on riferenties in vivo but partial agon activity in vitro. (b) Extention of an alkylaminoethor glyceryl side chain on ring B predicts partial agon and antagonist properties B predicts estrogenic activity in vivo but partial agonist mate activity in vitro. (b) Extention of an alkylaminoethoxy prop or glyceryl side chain on ring B predicts partial agonist the and antagonist properties in vivo b activity in vitro. (b) Extention of an alkylaminoethoxy
or glyceryl side chain on ring B predicts partial agonist
and antagonist properties in vivo but complete antago-
nist activity in viro. (c) An acetoxy side chain on r or glyceryl side chain on ring B predicts partial agonist the and antagonist properties in vivo but complete antagonist activity in viro. (c) An acetoxy side chain on ring B predicts agonist activity in vivo (possibly meta and antagonist properties in vivo but complete antagonist activity in vitro. (c) An acetoxy side chain on ring B predicts agonist activity in vivo (possibly metabolic activation to the phenols) but antiestrogenic activity mist activity in vitro. (c) An acetoxy side chain on ring
B predicts agonist activity in vivo (possibly metabolic use
activation to the phenols) but antiestrogenic activity in
int
vitro. (d) An allyloxy side chain on ring B predicts agonist activity in vivo (possibly metabolic activation to the phenols) but antiestrogenic activity in vitro. (d) An allyloxy side chain on ring B reduces antiestrogenic activity in vivo and in vitro compared wi activation to the phenols) but antiestrogenic activity in vitro. (d) An allyloxy side chain on ring B reduces antiestrogenic activity in vivo and in vitro compared with the alkylaminoethoxy side chain. (e) A para ethyl sub the alkylaminoethoxy side chain. (e) A *para* ethyl substitution on ring B predicts antagonist activity in vitro.
D. Application of Drug Receptor Theories
In their simplist form, the current theories of drug Example alkylaminoethoxy side chain. (e) A para ethyl sub-

itution on ring B predicts antagonist activity in vitro.
 $\begin{array}{c} \text{A} \text{polication of Drug Receptor Theories} \\ \text{In their simplest form, the current theories of drug} \\ \text{for each of the entire image} \end{array}$

interaction on ring B predicts antagonist activity in vi

D. Application of Drug Receptor Theories

In their simplist form, the current theories of

interaction with receptors are based upon the fundan

tal studies by Clar D. Application of Drug Receptor Theories
In their simplist form, the current theories of drug
interaction with receptors are based upon the fundamen-
tal studies by Clark (46) and Gaddum (104) who sug-
gested that the resp D. Application of Drug Receptor Theories of drug interaction with receptors are based upon the fundamental studies by Clark (46) and Gaddum (104) who suggested that the response to a drug is proportional to the commun In their simplist form, the current theories of drug
interaction with receptors are based upon the fundamental
studies by Clark (46) and Gaddum (104) who sug-
gested that the response to a drug is proportional to the
 interaction with receptors are based upon the fundamental studies by Clark (46) and Gaddum (104) who suggested that the response to a drug is proportional to the commumber of receptors occupied. However, the occupation wit tal studies by Clark (46) and Gaddum (104) who sug-
gested that the response to a drug is proportional to the
number of receptors occupied. However, the occupation
theory was modified by Stephenson (289) and Ariens and
Sim gested that the response to a drug is proportional to the
number of receptors occupied. However, the occupation
theory was modified by Stephenson (289) and Ariens and
Simonis (11) into two steps: receptor binding (dependen number of receptors occupied. However, the occupation with
theory was modified by Stephenson (289) and Ariens and grad
Simonis (11) into two steps: receptor binding (dependent the
upon the affinity of the drug for the rece theory was modified by Stephenson (289) and Ariens and Simonis (11) into two steps: receptor binding (dependen
upon the affinity of the drug for the receptor) followed
by the production of a response (dependent upon the
ef Simonis (11) into two steps: receptor binding (dependent
upon the affinity of the drug for the receptor) followed
by the production of a response (dependent upon the
efficiency of intrinsic activity of the drug receptor c upon the affinity of the drug for the receptor) followed
by the production of a response (dependent upon the
efficiency of intrinsic activity of the drug receptor com-
plex). Thus, within a series of nonsteroidal estrogen

 $\frac{trans}{100}$ geometric isomers $\frac{cis}{cos}$ geometric isomers

FIG. 24. Hypothetical models for estrogenic and antiestrogenic li-

gands binding to the estrogen receptor. Estradiol-17 β is anchored at a

phenolic site (PS) gands binding to the estrogen receptor. Estradiol-17 β is anchored at a phenolic site (PS) with high affinity binding (HAB). *trans* Monohy-droxytamoxifen has the same high affinity binding but this antiestrogenic ligan phenolic site (PS) with high affinity binding (HAB). trans Monohy-
droxytamoxifen has the same high affinity binding but this antiestro-
genic ligand binds to the receptor site so that the alkylaminoethoxy
side chain can i droxytamoxifen has the same high affinity binding but this antiestro-
genic ligand binds to the receptor site so that the alkylaminoethoxy
ring.
side chain can interact with a hypothetical antiestrogen region (AER) would
 $= C_2H_4$, $R_2 = C1$).

estradiol, their log dose-response curves in an assay AN
gressively lower affinities for the estrogen receptor than
estradiol, their log dose-response curves in an assay
system will be progressively shifted to the right of estra-AN
gressively lower affinities for the estrogen receptor the
estradiol, their log dose-response curves in an asse
system will be progressively shifted to the right of estra-
diol's curve. However, for a group of compounds gressively lower affinities for the estrogen receptor than estradiol, their log dose-response curves in an assay system will be progressively shifted to the right of estradiol's curve. However, for a group of compounds wit gressively lower affinities for the estrogen receptor the estradiol, their log dose-response curves in an asses system will be progressively shifted to the right of estradiol's curve. However, for a group of compounds with estradiol, their log dose-response curves in an assay
system will be progressively shifted to the right of estra-
diol's curve. However, for a group of compounds with
intrinsic activities progressively less than 1.0, the m system will be progressively shifted to the right of estra-
diol's curve. However, for a group of compounds with
intrinsic activities progressively less than 1.0, the maxi-
mal responses in their log dose-response curves w diol's curve. However, for a group of compounds with
intrinsic activities progressively less than 1.0, the maxi-
mal responses in their log dose-response curves will be
progressively lower. These are partial agonists. Howe intrinsic activities progressively less than 1.0, the maximal responses in their log dose-response curves will be progressively lower. These are partial agonists. However, the ideal antagonist would have a high affinity fo mal responses in their log dose-response curves will be
progressively lower. These are partial agonists. However,
the ideal antagonist would have a high affinity for the
estrogen receptor but would have an intrinsic activi progressively lower. These are partial agonists. However, the ideal antagonist would have a high affinity for the estrogen receptor but would have an intrinsic activity of zero. This ideal has been achieved with antiestrog the ideal antagonist would have a high affinity for the estrogen receptor but would have an intrinsic activity of zero. This ideal has been achieved with antiestrogens used in assay systems in vitro; therefore, the recepto estrogen receptor but would have an intrinsic activity of zero. This ideal has been achieved with antiestrogeniused in assay systems in vitro; therefore, the receptointeraction of estradiol or an antiestrogen such as 4 hy zero. This ideal has been achieved with antiestrogens
used in assay systems in vitro; therefore, the receptor
interaction of estradiol or an antiestrogen such as 4-
hydroxytamoxifen can be represented in the hypothetical
 interaction of estradiol or an antiestrogen such as 4-
hydroxytamoxifen can be represented in the hypothetical
scheme in figure 23. Estradiol first interacts via the C_y
phenolic group with a phenolic site on the recepto interaction of estradiol or an antiestrogen such as 4-
hydroxytamoxifen can be represented in the hypothetical
scheme in figure 23. Estradiol first interacts via the C₃-
phenolic group with a phenolic site on the recept hydroxytamoxifen can be represented in the hypothetical
scheme in figure 23. Estradiol first interacts via the C_3 -
phenolic group with a phenolic site on the receptor which
then directs the steroid to the correct posit scheme in figure 23. Estradiol first interacts via the C_3 -
phenolic group with a phenolic site on the receptor which
then directs the steroid to the correct position at the
binding site on the protein. The initial bind then directs the steroid to the correct position at the binding site on the protein. The initial binding step is followed by a change in the tertiary structure of the protein that locks the steroid into the receptor; this then directs the steroid to the correct position at the binding site on the protein. The initial binding step is followed by a change in the tertiary structure of the protein that locks the steroid into the receptor; this binding site on the protein. The initial binding step is
followed by a change in the tertiary structure of the
protein that locks the steroid into the receptor; this
change develops the intrinsic activity of the receptor
c followed by a change in the tertiary structure of the protein that locks the steroid into the receptor; this change develops the intrinsic activity of the receptor complex. The antiestrogen 4-hydroxytamoxifen binds with hi protein that locks the steroid into the receptor; this change develops the intrinsic activity of the receptor complex. The antiestrogen 4-hydroxytamoxifen binds with high affinity via the interaction of the phenolic group change develops the intrinsic activity of the receptor
complex. The antiestrogen 4-hydroxytamoxifen binds
with high affinity via the interaction of the phenolic
group with the phenolic site on the receptor. However,
the t complex. The antiestrogen 4-hydroxytamoxifen binds
with high affinity via the interaction of the phenolic
group with the phenolic site on the receptor. However,
the tertiary changes in the receptor necessary to develop
in with high affinity via the inference with the phenolic site of the tertiary changes in the rece intrinsic activity in the compalkylaminoethoxy side chain.
Based upon structure-activity oup with the phenolic site on the receptor. However,
e tertiary changes in the receptor necessary to develop
trinsic activity in the complex are prevented by the
kylaminoethoxy side chain.
Based upon structure-activity re

the tertiary changes in the receptor necessary to develop
intrinsic activity in the complex are prevented by the
alkylaminoethoxy side chain.
Based upon structure-activity relationship studies, a
hypothetical model of the alkylaminoethoxy side chain.
Based upon structure-activity relationship studies, a
hypothetical model of the ligand interaction with the
estrogen receptor binding site has been developed to
describe the structural features alkylaminoethoxy side chain.

Based upon structure-activity relationship studies, a

hypothetical model of the ligand interaction with the

estrogen receptor binding site has been developed to

describe the structural fea Based upon structure-activity relationship studies, a
hypothetical model of the ligand interaction with the
estrogen receptor binding site has been developed to
describe the structural features necessary to initiate or
to hypothetical model of the ligand interaction with the estrogen receptor binding site has been developed to describe the structural features necessary to initiate or to inhibit prolactin synthesis in vitro (211). Among the estrogen receptor binding site has been developed to describe the structural features necessary to initiate or to inhibit prolactin synthesis in vitro (211). Among the triphenylethylenes, compounds that have *cis* and *tra* describe the structural features necessary to initiate or
to inhibit prolactin synthesis in vitro (211). Among the
triphenylethylenes, compounds that have *cis* and *trans*
geometric isomers are extremely important for the triphenylethylenes, compounds that have *cis* and *trans* geometric isomers are extremely important for the development of a ligand-receptor model because the isomeric molecules encompass estrogenic and antiestrotriphenylethylenes, compounds that have *cis* and *tro* geometric isomers are extremely important for the *v*elopment of a ligand-receptor model because the is meric molecules encompass estrogenic and antiest genic actions geometric isomers are extremely important for the development of a ligand-receptor model because the isomeric molecules encompass estrogenic and antiestrogenic actions. Examples of the *trans* isomers, i.e., tamoxifen and velopment of a ligand-receptor model because the iso-
meric molecules encompass estrogenic and antiestro-
genic actions. Examples of the *trans* isomers, i.e., tamox-
ifen and enclomiphene are antiestrogens with zero in-
 meric molecules encompass estrogenic and antiestro-
genic actions. Examples of the *trans* isomers, i.e., tamox-
ifen and enclomiphene are antiestrogens with zero in-
trinsic activity, whereas the *cis* isomers ICI 47,699 1. In and enclomiphene are antiestrogens with zero in-
insic activity, whereas the *cis* isomers ICI 47,699 and
clomiphene are estrogens with an intrinsic activity of
To describe the interaction of the geometric isomers
th t

gands binding to the estrogen receptor. Estradiol-17\$ is anchored at a
phenyl ring substituted with the p-alkylaminoethoxy side
monethoxy side
monethoxy side
monethoxy side
monethoxy side
monethoxy side
monethoxy side
mon trinsic activity, whereas the *cis* isomers ICI 47,699 are zuclomiphene are estrogens with an intrinsic activity of 1.
To describe the interaction of the geometric isome with the estrogen receptor, the *trans* stilbene-lik To describe the interaction of the geometric isomers
with the estrogen receptor, the *trans* stilbene-like struc-
ture of tamoxifen and enclomiphene could sit loosely at
the binding site with low affinity binding so that t 1.
To describe the interaction of the geometric isomers
with the estrogen receptor, the *trans* stilbene-like struc-
ture of tamoxifen and enclomiphene could sit loosely at
the binding site with low affinity binding so th To describe the interaction of the geometric isomers
with the estrogen receptor, the *trans* stilbene-like struc-
ture of tamoxifen and enclomiphene could sit loosely at
the binding site with low affinity binding so that with the estrogen receptor, the *trans* stilbene-like structure of tamoxifen and enclomiphene could sit loosely at the binding site with low affinity binding so that the phenyl ring substituted with the p -alkylaminoetho ture of tamoxifen and enclomiphene could sit loosely at the binding site with low affinity binding so that the phenyl ring substituted with the p -alkylaminoethoxy side chain is projected away from the binding site (figu the binding site with low affinity binding so that the phenyl ring substituted with the *p*-alkylaminoethoxy side chain is projected away from the binding site (figure 24). The estrogenic ligands, zuclomiphene and ICI 47,6 chain is projected away from the binding site (figure 24).
The estrogenic ligands, zuclomiphene and ICI 47,699,
with their low affinity for the ER, can create a *trans*
stilbene-like structure with the *para* substituted The estrogenic ligands, zuclomiphene and ICI 47,699,
with their low affinity for the ER, can create a *trans*
stilbene-like structure with the *para* substituted phenyl
ring. In this binding state, the aminoethoxy side cha with their low affinity for the ER, can create a *trans* stilbene-like structure with the *para* substituted phenyl ring. In this binding state, the aminoethoxy side chain would lie next to the phenolic site on the recepto stilbene-like structure with the *para* substituted phenyl
ring. In this binding state, the aminoethoxy side chain
would lie next to the phenolic site on the receptor with
a weak interaction through the ether oxygen (figur ring. In this binding state, the aminoethoxy side chain
would lie next to the phenolic site on the receptor with
a weak interaction through the ether oxygen (figure 24).
There would be no interaction of the side chain with would lie next to the phenolic site on the receptor with
a weak interaction through the ether oxygen (figure 24).
There would be no interaction of the side chain with a
hypothetical antiestrogen region of the receptor and,

spet

 $\overline{\mathbb{O}}$

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Downloaded from pharmrev.aspetjournals.org at Thammasart University on December 8, 2012

agonists with the estrogen receptor (ER). The phenol group on the ligand interacts with the phenolic site on the ER (closed triangle) and produce a high affinity interaction if the geometry of the ligand is correct. Estra SCP

FIG. 25. Adaptation of Belleau's macromolecular perturbation theory (19) to describe the interaction of agonists, antagonists, and partial

agonists with the estrogen receptor (ER). The phenol group on the ligand inte SCP
FIG. 25. Adaptation of Belleau's macromolecular perturbation theory (19) to describe the interaction of agonists, antagonists, and partial
agonists with the estrogen receptor (ER). The phenol group on the ligand intera FIG. 25. Adaptation of Belleau's macromolecular perture
agonists with the estrogen receptor (ER). The phenol group of
a high affinity interaction if the geometry of the ligand is
(SCP) whereas 4-hydroxytamoxifen (OHTAM), a a high affinity interaction if the geometry of the ligand is correct. Estra (SCP) whereas 4-hydroxytamoxifen (OHTAM), antagonist, only induces a agonist) produces a mixture of SCP and NSCP in the ER.
changes that are neces (SCP) whereas 4-hydroxytamoxifen (OHTAM), antagonist, only induces a nonspecific conformational perturbation (NSCP). Bisphenol (partial agonist) produces a mixture of SCP and NSCP in the ER.

changes that are necessary to a high affinity interaction if the geometry of the ligand is correct. Estradiol (E_2) , an agonist, induces a specific conformational perturbation (SCP) whereas 4-hydroxytamoxifen (OHTAM), antagonist, only induces a nonsp

mist) produces a mixture of SCP and NSCP in the ER.
anges that are necessary to develop a high intrinsitivity for the complex can occur unimpeded.
The *geminal bis* parahydroxyphenyl compounds (detylated cyclofenyl, bisphe changes that are necessary to develop a high intrinsic
activity for the complex can occur unimpeded.
The *geminal bis parahydroxyphenyl* compounds (de-
acetylated cyclofenyl, bisphenol) that are partial agonists
in vitro a changes that are necessary to develop a high intrivactivity for the complex can occur unimpeded.
The *geminal bis parahydroxyphenyl* compounds acetylated cyclofenyl, bisphenol) that are partial agon
in vitro are particular activity for the complex can occur unimpeded. The *geminal bis parahydroxyphenyl* compounds (de-
acetylated cyclofenyl, bisphenol) that are partial agonists in
in vitro are particularly interesting. Belleau's macromo-
lecu The *geminal bis parahydroxyphenyl compounds (cacetylated cyclofenyl, bisphenol)* that are partial agonis
in vitro are particularly interesting. Belleau's macrom
lecular perturbation theory (19), which was origina
proposed acetylated cyclofenyl, bisphenol) that are partial agonists
in vitro are particularly interesting. Belleau's macromo-
lecular perturbation theory (19), which was originally
proposed to explain agonist, partial agonist, and in vitro are particularly interesting. Belleau's macromo-
lecular perturbation theory (19), which was originally
proposed to explain agonist, partial agonist, and antag-
onist activity of drugs at the muscarinic cholinergi lecular perturbation theory (19), which was originally
proposed to explain agonist, partial agonist, and antag
onist activity of drugs at the muscarinic cholinergic
receptor, may be used to explain partial agonists in term proposed to explain agonist, partial agonist, and antag-
onist activity of drugs at the muscarinic cholinergic
receptor, may be used to explain partial agonists in terms
of the ER model. According to Belleau's hypothesis, onist activity of drugs at the muscarinic cholinergic These hypothetical views of the interaction of the ER receptor, may be used to explain partial agonists in terms with agonists and antagonists may gain some substance o receptor, may be used to explain partial agonists in terms of the ER model. According to Belleau's hypothesis anagonist binds to the receptor and induces a specific conformational perturbation (SCP). An antagonist, or the of the ER model. According to Belleau's hypothesis, the anagonist binds to the receptor and induces a specific reconformational perturbation (SCP). An antagonist, on sythe other hand, binds to the receptor, produces a nonthe other hand, binds to the receptor, produces a non-
specific conformational perturbation (NSCP), but the
complex has zero intrinsic activity. Between these ex-
tremes, a partial agonist binds to the receptor and proconformational perturbation (SCP). An antagonist, on
the other hand, binds to the receptor, produces a non-
specific conformational perturbation (NSCP), but the
complex has zero intrinsic activity. Between these ex-
tremes the other hand, binds to the receptor, produces a non-
specific conformational perturbation (NSCP), but the
complex has zero intrinsic activity. Between these ex-
tremes, a partial agonist binds to the receptor and pro-
du specific conformational perturbation (NSCP), but the with ϵ complex has zero intrinsic activity. Between these ex-
to the tremes, a partial agonist binds to the receptor and pro-
duces an equilibrium mixture of agonist complex has zero intrinsic activity. Between these ex-
tremes, a partial agonist binds to the receptor and pro-
duces an equilibrium mixture of agonist and antagonist de
receptor complexes. Applying these definitions to th tremes, a partial agonist binds to the receptor and pro-
duces an equilibrium mixture of agonist and antagonist de-
receptor complexes. Applying these definitions to the ER sit
(Figure 25), estradiol binds with high affini

a nonspecific conformational perturbation (NSCP). Bisphenol (partial
ligand being locked into the binding site. 4-Hydroxyta-
moxifen (antagonist) wedges into the resting receptor
and only produces a NSCP. Bisphenol (partia ligand being locked into the binding site. 4-Hydroxyta-
moxifen (antagonist) wedges into the resting receptor
and only produces a NSCP. Bisphenol (partial agonist)
interacts at the ligand binding site, but while some of ligand being locked into the binding site. 4-Hydroxyta-moxifen (antagonist) wedges into the resting receptor and only produces a NSCP. Bisphenol (partial agonist) interacts at the ligand binding site, but while some of the ligand being locked into the binding site. 4-Hydroxyta-
moxifen (antagonist) wedges into the resting receptor
and only produces a NSCP. Bisphenol (partial agonist)
interacts at the ligand binding site, but while some of
th moxifen (antagonist) wedges into the resting receptor
and only produces a NSCP. Bisphenol (partial agonist)
interacts at the ligand binding site, but while some of
the receptors can be induced to lock the ligand into the
p and only produces a NS
interacts at the ligand b
the receptors can be inde
protein, other ligand inte
a NSCP in the complex.
These hypothetical vie teracts at the ligand binding site, but while some of

e receptors can be induced to lock the ligand into the

otein, other ligand interactions are only able to induce

NSCP in the complex.

These hypothetical views of th

the receptors can be induced to lock the ligand into the protein, other ligand interactions are only able to induce a NSCP in the complex.
These hypothetical views of the interaction of the ER with agonists and antagonists protein, other ligand interactions are only able to induce
a NSCP in the complex.
These hypothetical views of the interaction of the ER
with agonists and antagonists may gain some substance
through studies of the molecular a NSCP in the complex.
These hypothetical views of the interaction of the ER
with agonists and antagonists may gain some substance
through studies of the molecular biology of the estrogen
receptor. If, in the future, the g These hypothetical views of the interaction of the ER
with agonists and antagonists may gain some substance
through studies of the molecular biology of the estrogen
receptor. If, in the future, the gene that controls ER
sy with agonists and antagonists may gain some substance
through studies of the molecular biology of the estrogen
receptor. If, in the future, the gene that controls ER
synthesis can be cloned and large quantities of receptor through studies of the molecular biology of the estrogen
receptor. If, in the future, the gene that controls ER
synthesis can be cloned and large quantities of receptor
prepared, then biophysical studies of receptor comple receptor. If, in the future, the gene that controls ER
synthesis can be cloned and large quantities of receptor
prepared, then biophysical studies of receptor complexes
with estradiol or 4-hydroxytamoxifen may provide clue synthesis can be cloned and large quantities of receptor
prepared, then biophysical studies of receptor complexes
with estradiol or 4-hydroxytamoxifen may provide clues
to the 3-dimensional folding of the protein. Indeed,
 prepared, then biophysical studies of receptor complexes
with estradiol or 4-hydroxytamoxifen may provide clues
to the 3-dimensional folding of the protein. Indeed,
knowledge of the amino acid sequence of the ER and a
desc with estradiol or 4-hydroxytamoxifen may provide clues
to the 3-dimensional folding of the protein. Indeed,
knowledge of the amino acid sequence of the ER and a
description of peptide fragments near the ligand binding
site to the 3-dimensional folding of the protein. Indeed,
knowledge of the amino acid sequence of the ER and a
description of peptide fragments near the ligand binding
site obtained with alkylating compounds like tamoxifen
azir knowledge of the amino acid sequence of the ER and a
description of peptide fragments near the ligand binding
site obtained with alkylating compounds like tamoxifen
aziridine (184) will permit the development of a precise

370 3081
short term, though, a computer model of the receptor
site is possible by extensive structure-activity relation-270
short term, though, a computer model of the recep
site is possible by extensive structure-activity relati
ship studies in vitro in parallel with receptor bind JORDA!
short term, though, a computer model of the receptor
site is possible by extensive structure-activity relation-
ship studies in vitro in parallel with receptor binding
studies. studies. State is possible by extensive structure-activity relation-

Ship studies in vitro in parallel with receptor binding

studies.

XI. Concluding Comments: Unresolved Issues

The past 5 years have seen many important advances
in the understanding of estrogen and antiestrogen action. Progress has been facilitated by the introduction of ra-XI. Concluding Comments: Unresolved Issues
The past 5 years have seen many important advances
in the understanding of estrogen and antiestrogen action.
Progress has been facilitated by the introduction of ra-
diolabeled an Al. Concluding Comments: Onresolved issues

The past 5 years have seen many important advances

in the understanding of estrogen and antiestrogen action.

Progress has been facilitated by the introduction of ra-

diolabele The past 5 years have seen many important advances
in the understanding of estrogen and antiestrogen action
Progress has been facilitated by the introduction of ra-
diolabeled antiestrogens with a high specific activity an in the understanding of estrogen and antiestrogen action.
Progress has been facilitated by the introduction of ra-
diolabeled antiestrogens with a high specific activity and
high binding affinity for the ER, antibodies (mo Progress has been facilitated by the introduction of ra-
diolabeled antiestrogens with a high specific activity and
high binding affinity for the ER, antibodies (monoclonal
and polyclonal) raised to the ER, and renewed in diolabeled antiestrogens with a high specific activity and high binding affinity for the ER, antibodies (monoclona
and polyclonal) raised to the ER, and renewed interes
in structure-activity relationships with assay system high binding affinity for the ER, antibodies (monoclonal
and polyclonal) raised to the ER, and renewed interest
in structure-activity relationships with assay systems in
vitro. A unifying theory of antiestrogen action is, and polyclonal) raised to the ER, and renewed interest
in structure-activity relationships with assay systems in
vitro. A unifying theory of antiestrogen action is, how-
ever, impractical because there are several unexplai in structure-activity relationships with assay systems in vitro. A unifying theory of antiestrogen action is, how-
ever, impractical because there are several unexplained
observations with antiestrogens that require furthe vitro. A unifying theory of antiestrogen action is, how-
ever, impractical because there are several unexplained
observations with antiestrogens that require further
study: (a) The species differences in the pharmacology
o ever, impractical because there are several unexplained
observations with antiestrogens that require further
study: (a) The species differences in the pharmacology
of antiestrogens is perplexing. While it is possible that
 observations with antiestrogens that require further study: (a) The species differences in the pharmacology of antiestrogens is perplexing. While it is possible that the triphenylethylene-type antiestrogens (tamoxifen) are study: (a) The species differences in the pharmacology
of antiestrogens is perplexing. While it is possible that
the triphenylethylene-type antiestrogens (tamoxifen) are
metabolized to estrogens in rodents, no convincing e of antiestrogens is perplexing. While it is possible that the triphenylethylene-type antiestrogens (tamoxifen) are metabolized to estrogens in rodents, no convincing evidence has been presented to show metabolic difference the triphenylethylene-type antiestrogens (tamoxifen) are
metabolized to estrogens in rodents, no convincing evi-
dence has been presented to show metabolic differences
between chickens and rodents. (b) Most antiestrogens
e metabolized to estrogens in rodents, no convincing evidence has been presented to show metabolic differences between chickens and rodents. (b) Most antiestrogens exhibit agonist or partial agonist actions in vivo, but in v dence has been presented to show metabolic differences
between chickens and rodents. (b) Most antiestrogens $19.$
exhibit agonist or partial agonist actions in vivo, but in
vitro the compounds usually have zero intrinsic between chickens and rodents. (b) Most antiestrogens
exhibit agonist or partial agonist actions in vivo, but in
vitro the compounds usually have zero intrinsic efficacy.
The reason for this is unknown. (c) Tamoxifen binds virto the compounds usually have zero intrinsic efficacy.

The reason for this is unknown. (c) Tamoxifen binds to

The reason for this is unknown. (c) Tamoxifen binds to

Rea. Commun. 91: 812-818, 1979.

The specificity an The reason for this is unknown. (c) Tamoxifen binds to
the so-called "antiestrogen binding site" with precise
structual specificity and high affinity. The binding site
requires definition biochemically and its physiologica the so-called "antiestrogen binding site" with precise e so-called "antiestrogen binding site" with precise 21. BLA

ructual specificity and high affinity. The binding site $\frac{2}{3}$

quires definition biochemically and its physiological 22. BLA

le needs to be established.

structual specificity and high affinity. The binding site
requires definition biochemically and its physiological 22.
role needs to be established.
Finally, it is perhaps naive to believe that a clear view 23.
of the mecha requires definition biochemically and its physiolog
role needs to be established.
Finally, it is perhaps naive to believe that a clear v
of the mechanism of action of antiestrogens can
described when the molecular mechanis role needs to be established.
Finally, it is perhaps naive to believe that a clear view
of the mechanism of action of antiestrogens can be
described when the molecular mechanism of estrogen-
controlled protein synthesis an Finally, it is perhaps naive to believe that a clear view
of the mechanism of action of antiestrogens can be
described when the molecular mechanism of estrogen-
controlled protein synthesis and cell division is as yet
unkn of the mechanism of action of antiestrogens can
described when the molecular mechanism of estrog
controlled protein synthesis and cell division is as
unknown. Antiestrogens will prove to be useful tools
probe estrogen acti described when the molecular mechanism of estrogen-
controlled protein synthesis and cell division is as yet
unknown. Antiestrogens will prove to be useful tools to
probe estrogen action and to provide valuable compara-
ti both estrogens and antiestrogens.
Acknowledgments. Many of the studies described in this review were tive information to establish a molecular mechanism for

tive information to establish a molecular mechanism for
both estrogens and antiestrogens.

Acknowledgments. Many of the studies described in this review were

supported by grants P30-CA-14520 awarded to the Wisconsin Cli Acknowledgments. Many of the studies described in this review were
supported by grants P30-CA-14520 awarded to the Wisconsin Clinical
Cancer Center, PO1-CA-20432, RO1-CA-32713 and grants from ICI
plc (Pharmaceuticals, Wilm 1. ABBoTT, A. C., CLARK, E. R., AND JORDAN, V. C.: The inhibition of contradiol binding to estrogen receptor proteins by a methyl substituted

1. ABBOTT, A. C., CLARK, E. R., AND JORDAN, V. C.: The inhibition of contradiol

REFERENCES

- REFERENCES

1. ABBOTT, A. C., CLARK, E. R., AND JORDAN, V. C.: The inhibition

constradiol binding to estrogen receptor proteins by a methyl substitt

analogue of tamoxifen. J. Endocrinol. 69: 445-446, 1976.

2. ACTON, D.,
- HOTT, A. C., CLARK, E. R., AND JORDAN, V. C.: The inhibition constradiol binding to estrogen receptor proteins by a methyl substitution. J. D. Endocrinol. 69: 445–446, 1976.
Tron, D., HILL, G., AND TAIT, B. S.: Tricyclic t constrained binding to estrogen receptor proteins by a methyl substituted
analogue of tamoxifen. J. Endocrinol. 69: 445-446, 1976.
2. ACTON, D., HILL, G., AND TAIT, B. S.: Tricyclic triarylethylene antiestro-
gens. Dibenzo TAMORIC DEL TRESS. BIOLOGIC INTERNATION, D., HILL, G., AND TAIT, B. S.: Tricyclic triarylethylene are gena: Dibenz (b.f) oxepins, dibenzo (b.f) thispins dibenzo (a,e) cyclend dibenzo (b.f) thiocins. J. Med. Chem. 26: 1131analogue of tamoxifen. J. Endocrinol. 69: 445-446, 1976.

2. Acron, D., HILL, G., AND TAIT, B. S.: Tricyclic triarylethylene antiestro-

gens. Dibens (b.f) oxepins, dibenso (b.f.) thispins dibenso (a.e) cycloctenes

and di gens: Dibens (b.f) oxepins, dibenzo (b.f.) thiepins dibenzo (a,e) cycloctenes
and dibenzo (b.f) thiocins. J. Med. Chem. 26: 1131-1137, 1983.
3. ADAM, H. K., DOUGLAS, E. J., AND KEMP, J. V.: The metabolism of
tamoxifen in h
-
-
- tamoxifen in humans. Biochem. Pharmacol. 27: 145-147, 1979.
4. ADAM, H. K., GAY, M. A., AND MOORE, R. H.: Measurement of tamoxifen
in serum by thin layer densitometry. J. Endocrinol. 84: 35-42, 1980.
5. ADAMS. R., MISHELL, **timorifen in humans. Biochem. Pharmacol. 27: 145-147, 1979.**
 4. ADAM, H. K., GAV, M. A., AND MOORE, R. H.: Measurement of tamoxifen

in serum by thin layer densitometry. J. Endocrinol. 84: 35-42, 1980.
 5. ADAMS, R.,
- regulation of programming D. R., JR, AND ISRAEL, R.: Treatment of refractory anovulation with increased dosage and prolonged duration of cyclic clomiphene citrate. Obstet. Gynecol. **39:** 562-566, 1971.
LEGRA, J. C., KORAT, miphene citrate. Obstet. Gynecol. 39: 562-568, 1971.
6. ALLEGRA, J. C., KORAT, D., Do, H. M. T., AND LIPPMAN, M. E.: The regulation of progesterone receptor by 17β -estradiol and tamoxifen in the ZR-75-1 human breast can
-
- N

8. ALLEGRA, J. C., AND LIPPMAN, M. E.: The effect of 17β estradiol and

tamoxifen on the ZR-75-1 luman breast cancer cell line in defined

medium. Eur. J. Cancer Clin. Oncol. 16: 1007-1015, 1980.

9. ALLEN, K. E., C **AEGEA, J. C., AND LIPPMAN, M. E.: The effect of** 17β **estradiol and
tamoxifen on the ZR-75-1 human breast cancer cell line in defined
medium. Eur. J. Cancer Clin. Oncol. 16: 1007-1015, 1980.
LEN, K. E., CLARK, E. R., AN** relationships. Br. J. Cancer Clin. Oncol. 16: 1079 estradiol and tamoxifen on the ZR-75-1 human breast cancer cell line in defined medium. Eur. J. Cancer Clin. Oncol. 16: 1007-1015, 1980.

9. ALLEN, K. E., CLARK, E. R., AN
-
- LEN, K. E., CLARK, E. R., AND JORDAN, V. C.: Evidence for the metabolic activation of non-steroidal antioestrogens: A study of structure-activity relationships. Br. J. Pharmacol. 71: 83-91, 1980.
J. 10ER8ON, J. M., CLARK, activation of non-steroidal antioestrogens: A study of structure-activity
relationships. Br. J. Pharmacol. 71: 83-91, 1960.
10. ANDERSON, J. M., CLARK, J. H., AND PECK, E. J.. Oestrogen and nuclear
1972.
1972.
11. ARIENS,
-
- rat uterus after *in in statements* and the increase with $\frac{1}{2}$ and $\frac{1}{2}$ 1972.
UENS, E. J., AND SIMONIS, A. M.: A molec
Pharm. Pharmacol. 16: 137–157, 1964.
TARDI, B.: Multiple forms of nuclear estrog
rat uterus after in vitro exchange with [³H]e
Mol. Cell. Endocrinol. **29:** 159–167, 1983.
IN 11. ARIENS, E. J., AND SIMONIS, A. M.: A molecular basis for drug action. J.

22. ATTARDI, B.: Multiple forms of nuclear estrogen receptor in the immature

rat uterus after in vitro exchange with [³H]estradiol or [³H]a
- **the inner in particular in particular serves in the immature rat uterus after in vitro exchange with [³H] estraiol or [³H] antiestrogens. Mol. Cell. Endocrinol. 29: 159-167, 1983.
Mol. Cell. Endocrinol. 29: 159-167, 1** rat uterus after in vitro exchange with [³H] estradiol or [³H] antiestrogens.
Mol. Cell. Endocrinol. 29: 159-167, 1983.
13. BAN, R. R., AND JORDAN, V. C.: Identification of a new metabolite of
tamoxifen in patient seru
-
- macol. 32: 373-375, 1983.

RNES, J. E., AND MEYER, R. K.: Effects of ethamorytriphetol, MRL37

and clomiphene on reproduction in rats. Fertil. Steril. 13: 472–480, 1962.

ATES, D. J., FOSTER, A. B., GRIGGS, L. J., JARMAN, 14. BARNES, J. E., AND MEYER, R. K.: Effects of ethamoxytriphetol, MRL37
and clomiphene on reproduction in rats. Fertil. Steril. 13: 472–480, 1962.
15. BATES, D. J., FOSTER, A. B., GRIGGS, L. J., JARMAN, M., LECLERCQ, G.,

- **EXECUTE:** Antiestrogenic activity of tamoxifen N-oxide. Biochem.

Pharmacol. 31: 2823-2827, 1982.

16. BAUDENDISTEL, L. J., AND RUH, T. S.: Antiestrogen action: Differential

nuclear retention and extractability of the es 16. BAUDENDISTEL, L. J., AND RUH, T. S.: Antiestrogen action: Differential
nuclear retention and extractability of the estrogen receptor. Steroids 28:
223-237, 1976.
17. BAUM, M. AND OTHER MEMBERS OF THE NOLVADEX ADJUVANT
-
- UM, M. AND OTHER MEMBERS OF THE NOLVADEX ADJUVANT TEIAL ORGANISATION: Controlled trial of tamoxifen as adjuvant agent in management of early breast cancer. Lancet 1: 257-261, 1963.
ANTSON, G. T.: On the treatment of inoper 17. BAUM, M. AND OTHER MEMBERS OF THE NOLVADEX ADJUVANT TE:
 ORGANISATION: Controlled trial of tamosfie as adjuvant agent in manuma:
 Exargence of order breaths cannot for operable cases of carcinoma of

mamma: suggest
-
- agement of early breast cancer. Lancet 1: 257–261, 1983.

18. BEATBON, G. T.: On the treatment of inoperable cases of carcinoma of the

mamma: suggestions for a new method of treatment, with illustrative

cases. Lancet 2: mational pertubations of receptors. J. Med. Chem. 7: 776-784, 1964.

20. BINART, N., CATELLI, M. H., GEYNET, C., PURI, V., HAHNEL, R., MESTER,

J., AND BAULIEU, E. E.: Monohydroxytamoxifen: An antiestrogen with

high affin
-
- 122. BLACK, L. J., AND GOOD, R. L.: Uterine biossay of tamoxifen, trioxifene and a new setrogen action), R. L.: Uterine biossay of tamoxifen, trioxifene and a new setrogen antagonist (LY 117018) in rats and mice. Life Sci. and a new estrogen antagonist (LY 117018) in rats and mice. Life Sci.

26: 1453-1458, 1980.

22. BLACK, L. J., JAND GOODE, R. L.: Evidence for biological action of the

antiestrogens LY117018 and tamoxifen by different mec **26:** 1453-1458, 1980.

22. BLACK, L. J., AND GOODE, R. L.: Evidence for biological action of the

antiestrogens LY117018 and tamoxifen by different mechanisms. Endo-

crinology 109: 987-989, 1981.

23. BLACK, L. J., JONE
-
- antiestrogens LY117018 and tamoxifen by different mechanisms. Endo-
crinology 109: 987-989, 1981.
23. BLACK, L. J., JONES, C. D., AND FALCONE, J. F.: Antagoniam of estrogen
action with a new benzothiophene-derived antiestr
- action with a new benzothiophene-derived antiestrogen. Life Sci. 32:

1031-1036, 1963.

24. BLACK, L.J., JoNES, C. D., AND GOODE, R. L.: Differential interaction of

antiestrogens with cytosol estrogen receptors. Mol. Cell 95-103, 1981.
LOOM, H. J. G., AND BOESEN, E.: Antioestrogens in treatment of breast
cancer: Value of nafoxidine in 52 advanced cases. Br. Med. J. 2: 7-10,
1974.
LOOM, N. D., AND FISHMAN. J. H.: Tamoxifen treatment failures
-
-
- 1974.

26. BLOOM, N. D., AND FISHMAN. J. H.: Tamoxifen treatment failures in

hormonally responsive breast cancer. Cancer 51: 1190-1194, 1983.

27. BODINE, P. V., AND TUPPER, J. H.: Calmodulin antagonists decrease the

bin DDINE, P. V., AND TUPPER, J. H.: Calmodulin antagonists decrease the binding of epidermal growth factor to transformed, but not to normal, human fibroblasts. Biochem. J. 218: 629-632, 1984.
DEGNA, J. L., COEY, E., AND ROCH 29. BORONA, J. L., COEXY, E., AND ROCHEFORT, H.: Mode of action of LN1643

(a triphenylbromo-ethylene antiestrogen): Probable mediation by the

estrogen receptor and high affinity metabolite. Biochem. Pharmacol. 31:

3187-(a triphenylbromo-ethylene antiestrogen): Probable mediation by the
strogen receptor and high affinity metabolite. Biochem. Pharmacol. 31
3187-3191, 1982.
DRONA, J. L., AND ROCHEFORT, H.: High affinity binding to the estro
- 3187-3191, 1982.

20. BORGNA, J. L., AND ROCHEFORT, H.: High affinity binding to the estrogen

receptor of [²H]4-hydroxytamoxifen, an active antiestrogen metabolite.

Mol. Cell. Endocrine. **20**: 71-85, 1980.

20. BORGNA,
- receptor of [⁵H]4-hydroxytamoxifen, an active antiestrogen metabolite.
Mol. Cell. Endocrinol. 20: 71-85, 1980.
30. BoRNA, J. L., AND ROCHEFORT, H.: Hydroxylated metabolites of tamoxifen
are formed in vivo and bound to es
- Mol. Cell. Endocrinol. 20: 71-85, 1980.

30. BORGNA, J. L., AND ROCHEPORT, H.: Hydroxylated metabolites of tamoxifen are formed in vivo and bound to estrogen receptor in target tissues. J.

31. Bourron, M. M., AND RAYNAU9,
- Biol. Chem. 256: 859-868, 1981.

UTON, M. M., AND RAYNAUD, J. P.: The relevance of interaction kinetics

in determining biological response to estrogens. Endocrinology 105: 509-

515, 1979.

SWAAN, S. P., LEAKE, A., AND MO in determining biological response to estrogens. Endocrinology 105: 509-
515, 1979.
32. BOWMAN, S. P., LEAKE, A., AND MORRIS, I. D.: Hypothalamic, pituitary
and uterino cytoplasmic and nuclear osatrogen receptors and their tionship to the serum concentration of tamoxifen and its metal
hydroxytamoxifen, in the ovariectomised rat. J. Endocrinol. 94: 1982.
1980.
1982.
ECHER, P. I., NUMATA, M., DESOMBRE, E. R., AND JENSEN
Conversion of uterine 4
-
- hydroxytamoxifen, in the ovariectomised rat. J. Endocrinol. 94: 167-175, 1982.

33. BRECHER, P. I., NUMATA, M., DESOMBRE, E. R., AND JENSEN, E. V.: Conversion of uterine 4S estradiol-receptor complex in a soluble system. F Conversion of uterine 48 estradiol-receptor complex to 58 complex in a
soluble system. Fed. Proc. Am. Soc. Exp. Biol. 29: 249, 1970.
34. BROWN, R. R., BAIN, R. R., AND JORDAN, V. C.: Determination of tamoxifen
and metaboli
-

spet

ARMACOLO

spet

 $\overline{\mathbb{O}}$

PHARM
REV

hydroxyphenyl)-1,1,1-trichloroethane with **rat uterine** estrogen receptor.

- MNTIESTROGEN PH

hydroxyphenyl)-1,1,1-trichloroethane with rat uterine estrogen receptor.

J. Toxicol. Environ. Health 4: 881-893, 1978.

36. CAMAGGI, C. M., STROCCHI, E., AND CANOVA, N.: High performance liquid

chromatog
-
- 36. CAMAGGI, C. M., STROCCHI, E., AND CANOVA, N.: High performance liquid
chromatographic analysis of tamoxifen and major metabolites in human
plasma. J. Chromatogr. 275: 436-442, 1963.
37. CALLANTINE, M. R., HUMPHREY, R. 223-251, 1975.

223-251, AND OCHEFORT, H.: *In vivo* effect of an estrogen antagonist

39. CAPONY, F., AND ROCHEFORT, H.: *In vivo* effect of antiestrogens on localization and replenishment of estradiol receptors. Mol. Cel 38. CAPONY, F., AND ROCHEFORT, H.: *In vivo* effect of antisetrogens on localization and replenishment of estradiol receptors. Mol. Cell. Endocrinol. 3:
223-251, 1975.
39. CAPONY, F., AND ROCHEFORT, H.: *In vitro* and *in*
-
- 40. CAPONY, F., AND ROCHEFORT, H.: High affinity binding of the antiestrogen [²H]tamoxifen to the 8S estradiol receptor. Mol. Cell. Endocrinol. 11:
181-198. 1978. 223–251, 1975.
**PONY, F., AND ROCHEFORT, H.: In vitro and in vivo interactions of [³H] dimethylstilbestrol with the estrogen receptor. Mol. Cell. Endocrinol. 8:
47-64, 1977.
PONY, F., AND ROCHEFORT, H.: High affinity bin**
-
- dimethylatilbestrol with the estrogen receptor. Mol. Cell. Endocrinol. 8:
47-64, 1977.
40. CAPONY, F., AND ROCHEFORT, H.: High affinity binding of the antiestrogen
[²H]tamoxifen to the 8S estradiol receptor. Mol. Cell. interaction of estrogens and antiestrogens in the regulation of apolipoteins β synthesis. Endocrinology 106: 1862-1868, 1981.
42. CARLSON, R. A., AND GORSKI, J.: Characterization of a unique population of unifold estrog RLSON, R. A., AND GORSKI, J.: Characterization of a unique pof unfilled estrogen-binding sites associated with the nuclear friends in the cells in the cells in the cell cycle. Cell 36: 73-81, 1980.
IMEANS, A. R.: Changes i
-
- immature rat uteri. Endocrinology 106: 1776-1785, 1980.
43. CHAFOULEAS, J. G., LAGACE, L., BOULTON, W. E., BOYD, A. E., AND
MEANS, A. R.: Changes in calmodulin and its mRNA accompany re-entry
of quiescent (G₉) cells in t sylethanol. Endocrinology 65: 339-342, 1969.

MEANS, A. R.: Changes in calmodulin and its mRNA accompany re-entry

of quiescent (G_a) cells in the cell cycle. Cell 36: 73-81, 1964.

44. CHANG, M. C.: Degeneration of avea
- of quiescent (G₉) cells in the cell cycle. Cell 36: 73-31, 1984.

44. CHANG, M. C.: Degeneration of over in the rat and rabbit following oral

administration of 1-(p-2-diethylaminoethoxyphenyl)-1-phenyl-2-p-ani-

average
-
- sylethanol. Endocrinology 65: 339-342, 1969.
45. CHO, A., HUSLETT, W. L., AND JENDEN, D. J.: The identification of an
active metabolite of tremorine. Biochem. Biophys. Res. Commun. 5: 276-
279, 1961.
46. CLARK, B. I. The r 279, 1961.

46. CLARK, A. J.: The reaction between acetyl choline and muscle cells. J.

Physiol. (Lond.) 61: 530-546, 1926.

47. CLARK, B. R., DIX, C. J., JORDAN, V. C., PRESTWICH, G. AND SEXTON, S.:

A comparison at the c
-
- Pharmacol. 62: 442P-443P, 1978.

ARK, E. R., AND JORDAN, V. C.: Osstrogenic, anti-osstrogenic and fertility

properties of a series of compounds related to ethamorytriphetol

(MER25). Br. J. Pharmacol. 57: 487-493, 1976.
 48. CLARK, E. R., AND JORDAN, V. C.: Oestrogenic, anti-oestrogenic and fertility
properties of a series of compounds related to ethamoxytriphetol
(MER25). Br. J. Pharmacol. 57: 487–493, 1976.
49. CLARK, E. R., AND MCCRAK MRK, E. R., AND MCCRAKEN, A. M.: The oestrogenic and anti-oestrogenic properties of ring methyl substituted stilboestrols. J. Pharm. Pharmacol.
23: 330-346, 1971. MCCRAKEN, A. M.: Effect of some anti-oestrogenic ring
met
-
- **33: 339-346, 1971.**
50. CLARK, R. R., AND MCCRAKEN, A. M.: Effect of some anti-osatrogenic ring methyl-substituted stillbostrols on the uptake of tritiated osstradiol by the mouse vagina. J. Endormind. 52: 263-267, 1972.
 23: 339-346, 1971.
ARK, E. R., AND MCCRAKEN, A. M.: Effect of some anti-osstrogenic ring
methyl-substituted stillboostrols on the uptake of tritiated costradiol by
the mouse vagina. J. Endocrinol. S2: 283-267, 1972.
KR, methyl-substituted stilbosetrols on the uptake of tritiated oestradiol by
the mouse vagina. J. Endocrinol. 52: 263-267, 1972.
51. CLARK, J. H., ANDERSON, J., AND PECK, E. J.: Estrogen receptor antiestro-
gen complex: Atypi
-
- gen complex: Atypical binding by uterine nuclei and effect on uterine
growth. Steroids 22: 707-718, 1973.
ARK, J. H., AND GUTHRIE, S. C.: The agonistic and antagonistic effects
of elomiphenes cirrate and its isomers. Biol.
- 53. CLARK, J. H., HARDIN, J. W., PADYKULA, H. A., AND CARDASIS, C. A.: Role of estrogen receptor binding and transcriptional activity in the stimulation of hyperestrogenisation and nuclear bodies. Proc. Natl. Acad. Sci. U. U.S.A. 75: 2781-2784, 1978.

54. CLARK, J. H., MARKAVERICH, B., UPCHURCH, S., ERIKSSON, H., HARDIN,

J. W., AND PECK, E. J.: Heterogeneity of estrogen binding aites: Relation-

also to estrogen receptors and estrogen respo **J. W., AND PECK, E. J.: Heterogeneity of estrogen binding sites: Relation-**
ship to estrogen receptors and estrogen responses. Recent Prog. Horm.
Res. 36: 89-134, 1960.
55. CLARK, J. H., PASEBO, Z., AND PECK, E. J.: Nucle
- ahip to estrogen receptors and estrogen responses. Recent Prog. Horm.

Res. 36: 89-134, 1960.

55. CLARK, J. H., PASZBO, Z., AND PECK, E. J.: Nuclear binding and retention

of the receptor estrogen complex: Relation to the
- 56. CLARK, J. H., AND PECK, E. J.: Nuclear retention of receptor-estrogen
-
-
- properties of estriol. Endocrinology 100: 91-96, 1977.

56. CLARK, J. H., AND PECK, E. J.: Nuclear retention of receptor-estrogen

complex and nuclear acceptor sites. Nature (Lond.) 260: 635-637, 1976.

57. CLARK, J. H., P ARK, J. H., WINNEKER, R. C., GUTHRIE, S. C., AND MARKAVERICH, B.
M.: An endogenous ligand for the triphenylethylene antiestrogen binding
site. Endocrinology 113: 1167-1169, 1983.
EMENS, J. A., BENNETT, D. R., BLACK, L. J., 60. CLEMENS, J. A., BENNETT, D. R., BLACK, L. J., AND JONES, C. D.: Estone and Construction in the state of a new antiestrogen, keoxifene (LY 156758) on growth of carcino
induced manumary tumors and on LH and prolactin lev ort of a new antiestrogen, keorifene (LY 156758) on growth of carcinogen-
induced mammary tumors and on LH and prolactin levels. Life Sci. 32:
2969–2875, 1963.
60. CLITHEROE, J. H., AND LEATHAM, J. H.: Effect of estraiol a
-
- 2869–2875, 1983.

60. CLITHEROE, J. H., AND LEATHAM, J. H.: Effect of estradiol and ethamory-

triphetol (MER25) on the mouse uterus. Endocrinology 76: 127–130,

1965.

61. COEXY, E., BORGNA, J. L., AND ROCHEFORT, H.: Tamo
-
- inhibition. Cancer Res. 42: 317-323, 1982.

M.E., M. P., JONES, C. T. A., AND TODD, I. D. H.: A new anti-oestrogenia

agent in late breast cancer. Br. J. Cancer 25: 270-275, 1971.

M.L.INS, D. J., Honses, J. V., AND EMMENS
- 271

64. DANIEL, P., GASKELL, S. J., BISHOP, H., CAMPRELL, C., AND NICHOLSON,

R. I.: Determination of tamoxifen and biologically active metabolites in

human breast tumors and plasma. Eur. J. Cancer Clin. Oncol. 17: 1183-64. DANIEL, P., GASKELL, S. J., BISHOP, H., CAMPEELL, C., AND NICHOLSON,
R. I.: Determination of tamoxifen and biologically active metabolites in
human breest tumors and plasma. Eur. J. Cancer Clin. Oncol. 17: 1183-
65. DA
- from presst tumors and plasma. Eur. J. Cancer Clin. Oncol. 17:
1189, 1961.
NIEL., C. P., GASKELL, S. J., BISHOP, H., AND NICHOLSON,
Determination of tamoxism and an hydroxylated metabolite in prom patients with advanced br mass ipectrometrometry. J. E., G. ASKELL, S. J., BISHOP, H., AND NICHOLSON, R. I.: Determination of tamorifsn and an hydroxylated metabolite in plasma from patients with advanced breast cancer using gas chromatography-
66. Determination of tamoxifen and an hydroxylated metabolite in plasma
from patients with advanced breast cancer using gas chromatography-
mass spectrometry. J. Endocrinol. 83: 401-408, 1979.
66. DEVLEESCHOUWER, N., LECLERCQ,
- INVLEESCHOUWER, N., LECLERCQ, G., DANGUY, A., AND HEUSON, J. C.:
Antitumor effect of cyclofenil (F6066) on DMBA-induced rat mammary
tumors. Eur. J. Cancer 14: 721-723, 1978.
Pirstrao, D. L., SANDERS, F. J., AND Goss, D. A.
- 67. DIPIE'rRo, D. L, SANDERS, F. J., AND Goes, D. A.: Effect of *cia* and *trans* Antitumor effect of cyclofenil (F6066) on DMBA-induced rat mamn
tumors. Eur. J. Cancer 14: 721-723, 1978.
67. DIPIETRO, D. L., SANDERS, F. J., AND G**oss, D. A.: Effect of cis and tracellular**
isomers of clomiphene citrate
- tumora. Eur. J. Cancer 14: 721–723, 1978.
PIETEO, D. L., SANDERS, F. J., AND GOSS, D. A.: Effect of cis and *trans*
isomers of clomiphene citrate on uterine hexokinase activity. Endocrinology 84: 1404–1408, 1969.
x, C. J., nology 84: 1404-1408, 1969.

68. Dr., C. J., AND JORDAN, V. C.: Subcellular effects of monohydroxytamoxifen

in the rat uterus: Steroid receptors and mitosis. J. Endocrinol. 85: 393-404, 1980.

99. DODDS, E. C.: Synthetic
-
-
- activity of certain synthetic compounds. Nature (Lond.) 1137-147, 1949.

70. Donne, E. C., FOLLEV, S. J., GLASCOCK, R. F., AND LAWSON, W.: The

excretion of microgram doses of hexestrol by rabbits and rata. Biochem.

3. 68
- **J. 68: 161-167, 1958.**

DDDS, E. C., GOLDEERO, L., LAWSON, W., AND ROEINSON, R.: Oestrogenic

activity of certain synthetic compounds. Nature (Lond.) 141: 247-248,

1938.

Proc. Royal Soc. B 127: 140-106, 1939.

Proc. Roy extivity of certain synthetic compounds. Nature (Lond.) 141: 247-248,
1938.

72. DODD8, E. C., GOLDBERG, L., LAWSON, W., ROBINSON, SIR R.: Synthetic

costrogenic compounds related to stilbene and diphenylethane. Part I.

P
-
- 72. DODDS, E. C., GOLDEERG, L., LAWSON, W., ROBINSON, SIR R.: Synthetic
cestrogenic compounds related to stilbene and diphenylethane. Part I.
Proc. Royal Soc. B 127: 140-166, 1939.
73. DODDS, E. C., LAWSON, W., AND NOBLE, T3. DODDS, E. C., LAWSON, W., AND NOSLE, R. L.: Biological effects of the synthetic costrogenic substance 4:4' dihydroxy- α : β -disthyl stilbene. Lan-
Cet 1: 1389-1391, 1938.
74. DEOSDOWSKY, M., EDERY, M., GUGGIARI, M., G., AND VIVES, C.: Inhibition of prolactin-induced mammary cancer in C3H (XVII) mice with the *trans* isomer of bromotriphenylethylene.
Cancer Res. 40: 1674-1679, 1980.
UNCAN, G. W., LVSTER, S. C, CLARK, J. J., AND LEDNICE
-
- C., AND VIVES, C.: Inhibition of prolactin-induced mammery cancer in
C., AND VIVES, C.: Inhibition of prolactin-induced mammery cancer in
C3H (XVII) mice with the *trans* isomer of bromotriphenylsthylene.
Cancer Res. 40: 1 76. ECKERT, R. L., AND KATZENELLENBOGEN, B. S.: Effects of estrogens and
antiestrogens on estrogen receptor dynamics and the induction of proges-
terone receptor in MCF-7 human breast cancer cells. Cancer Res. 42:
139-144,
- Chem. 257: 8840-8846, 1982.
78. Eckert, R. L., Mullick, A., Robke, B. A., and Katzenellenbogen, B.
- terone receptor in MCF-7 human breast cancer cells. Cancer Res. 42:

139–14, 1962.

77. Ecker, R. L., AND KATENELLENBOGEN, B. S.: Physical properties of

estrogen receptor complexes in MCF-7 human breast cancer cells. J. B
- 8.: Estrogen receptor synthesis and turnover in MCF-7 breast cancer cells
measured by a density shift technique. Endocrinology 114: 629-637,
1984.
79. EDGREN, R. A., AND CALHOUN, D. W.: Estrogen antagonisms: Inhibition of
 S.: Estrogen receptor synthesis and turnover in MCF-7 breast cancer cells
measured by a density shift technique. Endocrinology 114: 629-637,
1964.
T. EDOREN, R. A., AND CALHOUN, D. W.: Estrogen antagonisms: Inhibition of
e
- and 17-ethyl-19-nortestosterons. Proc. Soc. Exp. Biol. Med. 94: 537-539,
1967.
20. EDWARDS, D. P., ADAMS, D. J., SAVAGE, N., AND MCGUIRE, W. L.:
Estrogen-induced synthesis of specific proteins in human breast cancer
cells.
-
- and antiestrogen on DNA polymerase in human breast cancer cells. Exp. Cell Res. 127: 197-213, 1980.
McGUIRER, W. L.: Subcellular compartmentalization of estrogen receptors
in human breast cancer cells. Exp. Cell Res. 127: 82. EDWARDS, D. P., MURTHY, S. R., AND MCGUIRE, W. L.: Effects of estrogen and antiestrogen on DNA polymerase in human breast cancer. Cancer
Res. 40:1722-1726, 1980.

NR. A. (2012) 26: 45-51, 1970.

83. EMMENS, C. W.: Comp
-
- antifertility activity in mice and rata. J. Reprod. Fert. 26: 175-182, 1971.
-
-
- HMENS, C. W., COX, R. I., AND MARTIN, L.: Oestrogen inhibitors of the stilboestrol series. J. Endocrinol. 18: 372-380, 1958.
Host, T.: Properties of a uterine oestradiol receptor. Biochem. Biophys Ree. Commun. 32: 338-343, tilbostrol series. J. Endocrinol. 18: 372-380, 1958.

87. ERDOS, T.: Properties of a uterine oestradiol receptor. Biochem. Biophys.

Rea. Commun. 82: 338-343, 1968.

88. ERRSSON, H., UPCHURCH, S., HARDIN, J. W., PECK, E. J
- 88. ERIKSSON, H., UPCHURCH, S., HARDIN, J. W., PECK, E. J., AND CLARK, J.
H.: Heterogeneity of estrogen receptors in the cytosol and nuclear frec-
tions of the rat uterus. Biochem. Biophys. Res. Commun. 81: 1-7, 1978.
89. ROPEAN ORGANISATION FOR RESEARCH ON TREATMENT OF CANCER (EORTC). BREAST CANCER GROUP: Clinical trial of naforidine, an osetrogen antagonist in advanced breast cancer. Eur. J. Cancer 8: 387-389, 1972.
1972.
1972. MNS, E., B
- (EORTC). BREAST CANCER GROUP: Clinical trial of naforidine, an ostrogen antagonist in advanced breast cancer. Eur. J. Cancer 8: 387-389, 1972.
90. EVANS, E., BASKEVITCH, P. P., AND ROCHEFORT, H.: Estrogen receptor-
DNA int
- DNA interaction: Difference between activation by estrogen and anties-
trogen. Eur. J. Biochem. 128: 185-191, 1982.
91. FABIAN, C., THLER, L., AND STERNSON, L.: Comparison binding affinities
of tamoxifen, 4-hydroxytamoxife DNA interaction: Difference between activation by estrogen and antiestrogen. Eur. J. Biochem. 128: 185-191, 1982.
BIAN, C., TILEER, L., AND STERNSON, L.: Comparison binding affinities
BIAN, C., TILEER, L., AND STERNSON, L. levels in patients with metastatic breast cancer. The metastatic breast cancer of the metastatic breast cancer
BiAN, C., Tukzne, L., AND STERNSON, L.: Comparison binding affinities
of tamorifen, 4-hydroxytamoxifen and desm 2:381-390,1981.

Downloaded from pharmrev.aspetjournals.org at Thammasart University on December 8, 2012

2012

- **92. FABIAN, C., STERNSON, L., AND BARRETT, M.: Clinical pharmacology of**
92. FABIAN, C., STERNSON, L., AND BARRETT, M.: Clinical pharmacology of
tanoxifen in patients with breast cancer: Comparison of traditional and JORDAN
BIAN, C., STERNSON, L., AND BARRETT, M.: Clinical pharmacology of
tamoxifen in patients with breast cancer: Comparison of traditional and
loading dose schedules. Cancer Treat. Rep. 64: 765–773, 1980.
- JORDA

92. FABIAN, C., STERNSON, L., AND BARRETT, M.: Clinical pharmacology of

tamoxifen in patients with breast cancer: Comparison of traditional and

loading does schedules. Cancer Treat. Rep. 64: 765-773, 1980.

93. FA sites. Proc. Natl. Acad. Sci. U.S.A. 80: 3158-3162, 1983.

93. FAYE, J. C., JOZAN, S., REDEWITH, G., BAULIEU, E. E., AND BAYARD, F.:

Physicochemical and genetic evidence for specific antiestrogen binding

sites. Proc. Nat affinity saturable binding sites. Proc. Natl. Acad. Sci. U.S.A. 80: 3158-3162, 1983.

Physicochemical and genetic evidence for specific antiestrogen binding

ites. Proc. Natl. Acad. Sci. U.S.A. 80: 3158-3162, 1983.

121

9
-
- **94. FAYE, J. C., LASSERRE, B., AND BAYARD, F.: Antiestrogen specific, high affinity saturable binding sites in rat uterine cytosol. Biochem. Biophys. Rea. Commun. 93: 1225-1231, 1960.

95. FEHERTY, P., FARRER-BROWN, AND K Res. Commun. 93: 1225-1231, 1980.**

95. **FEHERTY, P., FARRER-BROWN, AND KELLIE, A. E.: Oestradiol receptors in**

carcinoma and benign disease of the breast: An *in vitro* assay. Br. J.

Cancer 25: 697-710, 1971.

96. FEIL
- Cancer 26: 697-710, 1971.

96. FEIL, P. D., GLASSER, S. R., TOPT, D. O., AND O'MALLEY, B. W.: Progenterone binding in the mouse and rat uterus. Endocrinology 91: 738-746,

1972.

97. FERGUSON, E. R., AND KATZENELLENBOGEN,
- 96. FEIL, P. D., GLASSER, S. R., TOPT, D. O., AND O'MALLEY, B. W.: Progesterone binding in the mouse and rat uterus. Endocrinology 91: 738-746, 1972.

77. FERGUSON, E. R., AND KATEENELLENBOGEN, B. S.: A comparative study o
- ulated uterine growth and effects on estrogen receptor levels. Endocrinology 100: 1242–1251, 1977.
FIFER, B.: REDMOND, C., BROWN, A., WICKERMAN, D. L., WOLMARK, N., ALLEGRA, J., ESCHER, G., LIPPMAN, M., SAVLOV, E., WITTLI nology 100: 1242-1251, 1977.

SHER, B., REDMOND, C., BROWN, A., WICKERMAN, D. L., WOLMARK, N.,

ALLEGRA, J., ESCHER, G., LIPPMAN, M., SAVLOV, E., WITTLIFF, J., AND

FISHER, E. R.: Influence of tumoritien and chemotherapy i ALLEGRA, J., ESCHER, G., LIPPMAN, M., SAVLOV, E., WITTLIFF, J., AND FISHER, E. R.: Influence of tumor estrogen and progesterone receptor levels on the response to tamoxifer and chemotherapy in primary breast cancer. J. Cli
- FISHER, E. R.; Influence of tumor estrogen and progesterone receptor
levels on the response to tamoxifen and chemotherapy in primary breast
cancer. J. Clin. Oncol. 1: 227–241, 1983.
SHER, B., REDMOND, C., BROWNA, D., SACHS levels on the response to tamoxifen and chemotherapy in primary breast
cancer. J. Clin. Oncol. 1: 227–241, 1963.
SHER, B., REDMOND, C., BROWNA, A., WOLMARK, N. WITTLIFF, J., ¹²⁶
FREHRE, R. R. PLOTKIN, D., BOWMAN, D., SAC cancer. J. Clin. Oncol. 1: 227-241, 1983.
 201. PASHER, B., REDMOND, C., BROWN, A., WOLMARK, N. WITTLIFF, J.
 PRELICK, R., AND DESSER, R.: Treatment of primary breast cancer with

chemotherapy and tamoxifen. N. Engl. J FISHER, E. R., PLOTKIN, D., BOWMAN, D., SACHS, S., WOLTER, J., FRELICK, R., AND DESSER, R.: Treatment of primary breest cancer with chemotherapy and tamoxifen. N. Engl. J. Med. 305: 1–6, 1961.
100. FOLCA, P. J., GLASCOCK,
-
- FRELICK, R., AND DESSER, R.: Treatment of primary breast cancer with
chemotherapy and tamoxifen. N. Engl. J. Med. 305: 1-6, 1981.
100. FOLCA, P. J., GLASCOCK, R. F., AND IRVINE, W. T.: Studies with tritium-
labeled beroest Formation of the N-oxide, a new metabolite. Biochem. Pharmacol breast cancer. Lancet 2: 796-797, 1961.

101. FOSTER, A. B., GRIGGS, L. J., JARMAN, M., VAN MAANEN, J. M. S., AND

SCHULTEN, H. -R.: Metabolism of tamoxifen by **SCHULTEN, H. -R.: Metabolism of tamoxifen by rat liver microsomes:**
Formation of the N-oxide, a new metabolite. Biochem. Pharmacol. 29:
1977-1979, 1980.
commoxifen (ICI 46,474) Part I in laboratory animals. Xenobiot
- Formation of the N-oxide, a new metabolite. Biochem. Pharmacol. 29:

1977-1979, 1980.

102. **FROMSON, J. M., PEARSON, S., AND BRAMAH, S.: The metabolism of**

tamoxifen (ICI 46,474) Part I in laboratory animals. Xenobiotica
- tomson, J. M., PEARSON, S., AND BRAMAH, S.: The metabolism of tamoxifen (ICI 46,474) Part I in laboratory animals. Xenobiotica 3: 693-709, 1973.
Tomos ... M., PEARSON, S., AND BRAMAH, S.: The metabolism of tamoxifen (ICI 4 TROMSON, J. M., PEARSON, S., AND BRAMAH, S.: The metabolism of tamoxifen (ICI 46,474) Part II in female patients. Xenobiotica 3: 711-

713, 1973.

104. GADDUM, J. H.: The action of adrenalin and ergotamine on the uterus of
-
- 105. GARG, S., BINDAL, R. D., DURANI, S., AND KAPIL, R. S.: Structure-activity
relationship of estrogens: A study involving cyclofenyl as the model
compound. J. Steroid Biochem. 18: 89-95, 1983.
106. GABKELL, S. J., DANIEL
-
-
- 274-285, 1979. However, 1979. 2003. Comparison between tamoxifen and clomphene therapy in women with anovulation. Arch. Gynaekol. 227:
107. GERHARD, I., AND RUNNEBAUM, B.: Comparison between tamoxifen and clomphene therapy clomiphene therapy in women with anovulation. Arch. Gynaekol. 227:
274–288, 1979.
108. GEYNET, C., SHYAMALA, G., AND BAULIEU, E. E.: Similarities and differences of the binding of estradiol and 4-hydroxytamoratien (an anti 108. GEYNET, C., SHYAMALA, G., AND BAULIEU, E. E.: Similarities and differences of the binding of estradiol and 4-hydroxytamoxifen (an antiestrogen) in the chick oviduct cytosol. Biochem. Biophys. Acta 756: 349–353, 1983.

-
- 1983.

ANNOPOULOS, G., AND GORSKI, J.: Estrogen-binding protein of the rat

uterus. J. Biol. Chem. 246: 2630-2536, 1971.

ASCOCK, R. F., AND HOEKSTRA, W. G.: Selective accumulation of tritium-

labeled hexoestrol by the re **and Sheep. Biol. Chem. 246: 2530-2536, 1971.**
 and Sheep. 246: 2530-2536, 1971.
 110. GLASCOCK, R. F., AND HOEKSTRA, W. G.: Selective accumulation of tritium-labeled herosetrol by the reproductive organs of immature fe
- A8COCK, R. F., AND HOEKSTRA, W. G.: Selective accumulation of tritium-
labeled hexoestrol by the reproductive organs of immature female goats
and sheep. Biochem. J. 72: 673-682, 1969.
DLAND**ER, Y.**, AND STERNSON, L. A.: Pa iabeled hexoestrol by the reproductive organs of immature female goats
and sheep. Biochem. J. 72: 673-682, 1969.
111. GOLANDER, Y., AND STERNSON, L. A.: Paired-ion chromatographic analysis
of tamoniten and two major metabo 111. GOLANDER, Y., AND STERNSON, L. A.: Paired-ion chromatographic analysis
-
- Recent Prog. Horm. Res. 24: 45–72, 1968.
REENE, G. L., CLOSS, L. E., DESOMBRE, E. R., AND JENSEN, E. V.:
Antibodies to estrophilin: Comparison between rabbit and goat antisera. 41-49, 1980.

112. GORSKI, J., TOFT, D., SHYAMALA, G., SMITH, D., AND NOTIDES, A.: Hormono recoptors: Studies on the interaction of estrogen with the uterus.

1410 mone recoptors: Studies to a interaction of estrogen with
- J. Steroid Biochem. 11: 333-341, 1979.

114. GREENE, G. L., FrrCH, F. W., AND JENSEN, E. V.: Monoclonal antibodies

to estrophilin: Probes for the study of estrogen receptors. Proc. Natl.

Aced. Sci. U.S.A. 77: 167-161, 19
- 115. GREEN, G. L., FITCH, F. W., AND JENSEN, E. V.: Monoclonal antibodies
to estrophilin: Probes for the study of estrogen receptors. Proc. Natl.
Acad. Sci. U.S.A. 77: 157-161, 1980.
I.C. GREN, M. D., WHYBOURNE, A. M., TAY to estrophilin: Probes for the study of estrogen receptors. Proc. Natl.
Acad. Sci. U.S.A. 77: 157-161, 1980.
LEEN, M. D., WHYBOURNE, A. M., TAYLOR, I. W., AND SUTHERLAND, R.
L.: Effects of antioestrogens on the growth and Press, **Sydney,** 1981. L.: Effects of antioestrogens on the growth and cell cycle kinetics of
cultured human mammary carcinoma cells. In Nonsteroidal Antioestro-
gens, ed. by R. L. Sutherland and V. C. Jordan, pp. 397–412, Academic
Press, Sydney
- **facts and all after administration.** Am. B., BARFIELD, W. E., JUNGCK, E. C., AND RAY, A. W.: 144.

Induction of ovulation with MRL-41—Preliminary Report. J. Am. Med.

Assoc. 178: 101-104, 1961.

117. GREENBLATT, R. B., AN
-
-

JUNOCK, E. C.: Induction of ovulation. Am. J. Obstet. GynecoL **84: 900-** 912, 1962.

- **119. GULINO, A., AND PASQUALINI, J. It: Heterogeneity of** binding sites **for**
-
- 120. HANAHAN, D. J., DASKALAKIS, E. G., EDWARDS, T., AND DAUBEN, H. J.:
The metabolic pattern of C^{14} -diethylstilbestrol. Endocrinology 53: 163-170, 1953.
170, 1953.
121. HARPER, M. J. K., AND WALPOLE, A. L.: Contrasti The metabolic pattern of C¹⁴-diethylstilbestrol. Endocrinology 53:

170, 1953.

121. HARPER, M. J. K., AND WALPOLE, A. L.: Contrasting endocrine action of cise and *trans* isomers in a series of substituted triphenylethy
- **lena: Effect on implantation and mode of substituted triphenylethylenes.**
Nature (Lond) 212: 87, 1966.
Nature (Lond) 212: 87, 1966.
Nature M.J. K., AND WALPOLE, A. L.: A new derivative of triphenylethy-
Nature. M.J. K., A of cis and *trans* isomers in a series of substituted triphenylethylenes.

132. HARPER, M. J. K., AND WALPOLE, A. L.: A new derivative of triphenylethylene:

123. HARPER, M. J. K., AND WALPOLE, A. L.: Mode of action of ICI
-
- ALLEGRA, J., ROUGHER, G., LIPPMAN, M., SAVLOV, E., WITHER, J., AND ALLEGRA, H. A., LIPPON, A., WHITE, D. S., SANTON, R. J., BOUCHER, A. E.,
ulated uterine growth and effects on estrogen receptor levels. Endocrimental parti lene: Effect on implantation and mode of action in rata. J. Reprod. Fertil.
13: 101-119, 1967.
123. HARPER, M. J. K., AND WALPOLE, A. L.: Mode of action of ICI 46,474 in
preventing implantation in rata. J. Endocrinol. 37: **424. HARVEY, H. A., LIPTON, A., WHITE, D. S., SANTON, R. J., BOUCHER, A. E., SHAFIK, A. S., AND DIXON, R. J.: Cross over comparison of tamoxifen and aminoglutethimide in advanced breast cancer. Cancer Res. Suppl. 42: 345.**
	- and aminoglutethimide in advanced breast cancer. Cancer Res. Suppl.
42: 34518-3453S, 1982.
125. HAYES, J. R., RORKE, E. A., ROBERTSON, D. W., KATZENELLENBOGEN, B.
5., AND KATZENELLENBOGEN, B. S.: Biological potency and ute 8., AND KATEENELLENBOGEN, B. S.: Biological potency and uteriestrogen receptor interactions of the metabolites of the antiestrogens 628 and U23,469. Endocrinology 108: 164-172, 1981.
126. HECKER, R., VEGH, I., AND LEVY, C.
	-
	- **628 and U23,469. Endocrinology 108: 164-172, 1961.**
126. HECKER, E., VEGH, I., AND LEVY, C. M.: Clinical trial of clomiphene in
advanced breast cancer. Eur. J. Cancer 10: 747-749, 1974.
127. HEEL, R. C., BROGDEN, R. N.; S **128. HECKER, E., VEGH, I., AND LEVY, C. M.: Clinical trial of clomiphene in advanced breast cancer. Bur. J. Cancer 10: 747-749, 1974.**

	127. HERL, R. C., BROGDEN, R. N., SPEGHT, T. M., AND AVERY, G. S.: Tamoxime in the tr
	- (NSC-357) in disseminated mammary carcinoma. Cancer Chemotic use in the treatment of breast cancer. Drugs 16: 1-24, 1978.

	128. HERSEY, A. L., GRIFFITHS, C. T., AND KISTNER, R. W.: Clomiphene citrate (NSC-35770) in dissemi
	- potency, 1980–41, 1964.

	Rep. 43: 39–41, 1964.
 Poster interactions and metabolism of catechol estrogens in

	potency, R. M. Wenter, interactions and metabolism of catechol estrogens in

	the immature rat uterus in *vitro* **there is in the immature in the immature in vitro.** Endocrinology 111: 806-903, 1982.
 129. HERSEY, R. M. WEISS, J., AND KATEENELLENBOGEN, B. S.: Estrogenic

	potency, receptor interactions and metabolism of catechology
- 103. FROMSON, J. M., PEARSON, S., AND BRAMAH, S.: The metabolism of the relation of the uterus of the strip of the uterus of the university of the unive potency, receptor interactions and metabolism of catechol estrogens in
the immature rat uterus in vitro. Endocrinology 111: 896-903, 1982.
EUSON, J. C., ENGELSMAN, E., BLANK-VAN DER WIJST, J., MAASE, H.,
DROCHMANS, A., MIC 130. HEUSON, J. C., ENGELSMAN, E., BLANK-VAN DER WIJST, J., MAASE, H., DEOCHMANS, A., MICHEL, J., NOWAKOWSKI, AND GORINS, A.: Comparative trial of nafoxidine and ethinyl osstradiol in advanced breast cancer:
An EORTC study
	- 347, 1981. HOLTKAMP, D. E., GRESLIN, S. C., Roor, C. A., AND LERNER, L. J.: Gonadotropin inhibiting and antifecundity effects of chloramiphene.
2132. HOLDAWAY, D. B., GRESLIN, S. C., Roor, C. A., AND LERNER, L. J.: Gonadot
	- cancer to aminoglutethimide or tamoxifen. Aust. N.Z. J. Surg. 51: 345-347, 1981.

	132. HOLTKAMP, D. E., GRESLIN, S. C., ROOT, C. A., AND LERNER, L. J.: Gonadotropin inhibiting and antifecundity effects of chloramiphene.

	P bLTKAMP, D. E., GRESLIN, S. C., ROOT, C. A., AND LERNER, L. Gonadotropin inhibiting and antifecundity effects of chloramipher
Proc. Soc. Exp. Biol. Med. 1065: 197-201, 1960.
 PROC. Soc. Exp. Exp. District PROCETTER, W. L.
- compound. J. Steroid Biochem. 18: 89-95, 1983.

compound. J. Steroid Biochem. 18: 89-95, 1983.

106. GASKELL, S. J., DANIEL, C. P., AND NICHOLSON, R. I.: Determination of

tamoxifen in rat plasma by gas chromatography-mass Conadotropin inhibiting and antifecundity effects of chloramiphene.
Proc. Soc. Exp. Biol. Med. 106: 197-201, 1960.
133. HORWITZ, K. B., COSTLOW, M. E., AND MCGUIRZ, W. L.: MCF-7: A human
breast cancer cell line with estrog
	- and the with estrogen. androgen. progesterone and glucocorticoid receptors. Steroids 26: 785-795, 1975.

	134. HORWITZ, K. B., KOSEKI, Y., AND McGuIRE, W. L.: Estrogen control of progesterone receptor in human breest cancer receptor in human breast cancer. Role of estradiol and antiestrogen. Endocrinology 103: 1742-1751, 1978.

	136. HoRWITZ, K. B., AND McGumE, W. L.: Estrogen control of progesterone newpra K. B., AND McGumE, W. L.: Actinomyci
	-
	- progessions constructed receptor in human breast cancer. J. Biol. Chem. 253: 2223-2228, 1978.
 processing of estrogen receptor in human breast cancer. J. Biol. Chem. 253: 2223-2228, 1978.
 DRWITZ, K. B., AND MCGUIRZ, W. 137. HORWITZ, K. B., AND McGUIRE, W. L.: Estrogen control of progesterone receptor in human breast cancer. J. Biol. Chem. 253: 2223-2228, 1978.
136. HORWITZ, K. B., AND McGUIRE, W. L.: Actinomycin D prevents nuclear
proces
		-
	- 138. HORWITZ, K. B., AND MCGUIRE, W. L.: Actinomycin D prevents nuclear
processing of estrogen receptor. J. Biol. Chem. 253: 6319-6322, 1978.
137. HORWITZ, K. B., AND MCGUIRE, W. L.: Nuclear mechanisms of estrogen
action. **139. HUGKER, C. A., PEARSON, O. H., AND 38 PARTICIPATING INVESTIGATORS:**
 139. HUBAY, C. A., PEARSON, O. H., AND 38 PARTICIPATING INVESTIGATORS:
 71-82, 1981.
 139. HUCKER, H. B., GILLETTE, J. R., AND BRODIE, B. B.: 138. HUBAY, C. A., PEARSON, O. H., AND 38 PARTICIPATING INVESTIGATORS:
Adjuvant therapy of Stage II breast cancer. Breast Cancer Res. Treat. 1:
77-82, 1981.
139. HUCKER, H. B., GILLETTE, J. R., AND BRODIE, B. B.: Enxymatic
	-
	-
	- Steril. 3. However, H. B., GILLETTE, J. R., AND BRODIE, B. B.: Ensymmetic pathway
for the formation of cotinine, a major metabolite of nicotine in rabbit
liver. J. Pharmacol. Exp. Ther. 129: 94-100, 1960.
140. HUPPERT, L. esteril. Steril. S. C.: Induction of ovulation with clomiphene citrate. Fertil.
Steril. 31: 1-8, 1979.
141. LACOBELLI, S., NATOLI, C., SICA, G., AND GAGGINI, C.: Common and distinct features in the growth inhibitory activi Steril. 31: 1-8, 1979.

	COBELLI, S., NATOLI, C., SICA, G., AND GAGGINI, C.: Common and distinct features in the growth inhibitory activity of MPA and tamoxifen on

	netrogen-sensitive human breest cancer cells. Proceedings estrogen-sensitive human breest cancer cells. Proceedings of the International Symposium on Medroxyprogesterone Acetate, ed. by F. Cavalli, W. L. McGuire, F. Pamuti, A. Pellegrini, and Anna G. Robustellidella, pp. 80-87, E
	- The Muslim in the state of the state cancer. Here, KVOLS, L. K., NICHOLS, W. C., CREAGAN, E. T., HAHN, R. **143.** ITO, KVOLS, L. K., NICHOLS, W. C., CREAGAN, E. T., HAHN, R. G., RUBIN, J., AND FRYTAK, S.: Randomised clinical trial of disthylstilbestrol versus tamoxifen in postmenopausal women with advanced breast cancer. N.
E. **the** growth of solid sarcoma-180. Cancer Lett. 19: 215-220, 1983.
 the growth of solid sarcoma-iSO. Cancer Lett. 19: 215-220, 1983.

	144. JAKESZ, R., AND HIPAKA, H.: Antitumor effect of a calmodulin antagonist on the gr
	-
	- **ingl. J. Med. 304:** 16-21, 1981.
 in the growth of solid sarcoma-180. Cancer Lett. 19: 215-220, 1983.
 it is growth of solid sarcoma-180. Cancer Lett. 19: 215-220, 1983.
 it is strogen receptor. J. Biol. Chem.
 in
	- **268: 11798-11806, 1983.** 145. AND HIDAKA, H.: Antitumor effect of a calmodulin antagonist on the growth of solid sarcoma-180. Cancer Lett. 19: 215-220, 1983.
 216-220, 1983. R., KASID, A., AND LIPPMAN, M. E.: Continuous in the rat does not induce loss of uterine estrogen receptor. J. Biol. Chem.
258: 11798–11806, 1983.
258: NERN, E. V., BLOCK, G. E., SMITH, S., KYSER, K., AND DESOMBRE, E.
R.: Estrogen receptors and breast cancer respo

ARMACOLO

- 146. JENSEN, E. V., HURST, D. J., DESOMBRE, E. R., AND JUNGBLUT, P. W.: simmature rat uterus during the sequential administration of antiosstro-
Sulfhydryl groups and estradiol-receptor interaction. Science 158: 385,
1967. 146. JENSEN, E. V., HURST, D. J., DESOMBER, E. R., AND JUNGELUT, P. W.:

Sulfhydryl groups and estradiol-receptor interaction. Science 158: 385,

1967.

147. JENSEN, E. V., AND JACOBSON, H. I.: Basic guides to the mechanis
-
- 1967.
NSEN, E. V., AND JACOBSON, H. I.: Basic guides to the mechanism of
strogen action. Recent Prog. Horm. Rea. 18: 387-414, 1962.
NSEN, E. V., NUMATA, M., BRECHER, P. I., AND DESOMBRE, E. R.: 174.
Hormone-receptor intera **149. Hormone-receptor interactions as a guide to biochemical mechanism.** In The Biochemistry of Steroid Hormone Action, ed. by R. M. S. Smellie, pp. 133–159, Academic Press, London, 1971.

ENSEN, E. V., SUZUKI, T., KAWASH
- **P. W., AND DESOMBRE, E. R.: A two-step mechanism for the interaction

of estradiol with rat uterus. Proc. Natl. Aced. Sci. U.S.A. 59: 632-638,

1968.**
 1968.
 1968.
 1968.
 1978.
 1988. Henry, R. S. H., ANDERSEN, E. V., SUZUKI, T., KAWASHIMA, T., STUMPP, W. E., JUNGELIUT, 175. KAMBOJ, V. P., SETTY, B. S., CHANDRA, H., ROY, S. K., AND KAR, A. B.:
P. W., AND DESOMBER, E. R.: A two-step mechanism for the int
-
-
- phenyl).1-naphthalenyl][4-[2.(1.pyrrolidinyl)ethoxy]-phenyllmetha-692–698, 1970.

162. JONES, C. D., SUAREZ, T., MASSEY, E. H., BLACK, L. J., AND TINSLEY, F.

C.: Synthesis and antiestrogenic activity of [3,4-dihydro-2-(4-methoxy-phenyl)-1-naphthalenyl][4-[2-(1-pyrrolidinyl)ethoxy]-pheny
-
-
- none, methanesulfane acid salt. J. Med. Chem. 22: 962-966, 1979.

153. JORDAN, V. C.: Prolonged antioestrogenic activity of ICI 46,474 in the

ovariectomized mouse. J. Reprod. Fertil. 52: 251-258, 1975.

154. JORDAN, V. C. EDAN, V. C.: Pharmacology of antiestrogens. In Endocrinology of Cancer III, ed. by D. P. Rose, pp. 129-173, CRC Press, Boca Raton, Florida, 1982. RDAN, V. C., AND ALLEN, K. E.: Evaluation of the antitumour activity of the 156. JORDAN, V. C., AND ALLEN, K. E.; Evaluation of the antitumour activity of the non-steroidal antioestrogen monohydroxytamoxifen in the DMBA-

induced rat mammary carcinoma model. Eur. J. Cancer 16: 239-251, 181. KATERN
-
- the non-steroidal antioestrogen monohydroxytamoxifen in the DMBA-
induced rat mammary carcinoma model. Eur. J. Cancer 16: 239-251,
1960.
1960.
In laboratory animals. Cancer Treat. Rep. 64: 745-759, 1960.
197. JORDAN, V. C. A: Determination and pharmacology of a new hydroxylated metabolite
of tamoxifen observed in patient sera during therapy for advanced breast
cancer. Cancer Res. 43: 1446-1450, 1983.
158. JORDAN, V. C., AND BOWSER-FINN, R. A
-
- tamoxifen by immature rat tissues in vivo. Endocrinology 110: 1281-1291, 1982.
159. JORDAN, V. C., COLLINS, M. M., ROWSBY, L., AND PRESTWICH, G.: A monohydroxylated metabolite of tamoxifen with potent antiosetrogenic activ
- RDAN, V. C., COLLINS, M. M., ROWSBY, L., AND PRESTWICH, G.: A monohydroxylated metabolite of tamoxifen with potent antioestrogenic retivity. J. Endocrinol. 75: 305-316, 1977.
activity. J. Endocrinol. 75: 305-316, N. K. E.: monohydroxylated metabolite of tamoxifen with potent antioestrogenic 184
activity. J. Endocrinol. 75: 305–316, 1977.
 **RDAN, V. C., DIX, C. J., AND ALLEN, K. E.: The effectiveness of long-
term tamoxifen treatment in a lab S. E. S. E. S. E. S. E. Jones, P. S. S. S. AND ALLEN, K. E.: The effectiveness of long-
term tamorifen treatment in a laboratory model for adjuvant hormone
therapy of breast cancer. In Adjuvant Therapy of Cancer, vol. II,**
- therapy of breast cancer. In Adjuvant Therapy of Cancer, vol. 11, ed. by

8. E. Salmon and S. E. Jones, pp. 19-26, Grune and Stratton, New York,

1979.

161. Jonnan, V. C, Drx, C. J., AND ALLEN, K. E.: Effects of antioestr carcinogen-induced rat mammary cancer. In Non-Steroidal Antioestrogens, ed. by R. L. Sutherland and V. C. Jordan, pp. 260-280, Academic Press, Sydney, 1981.

Frees, Sydney, 1981.

186.

186. JORDAN, V. C., DIX, C. J., NAYL
- 162. JORDAN, V. C., DIX, C. J., NAYLOR, K. E., PRESTWICH, G., AND ROWSBY,

L.: Non-steroidal antiestrogens: Their biological effects and potential

mechanisms of action. J. Toxicol. Environ. Health 4: 364-390, 1978.

163.
-
-
- compound with osatrogenic activity. J. Chem. Soc. B: 1-5, 1970.
306, 1982.
External and antibodies actions of the estrogenic activity. J. Chem. Soc. B: 1-5, 1970.
- trioxic in the immature rat uterus: A comparison of hydroxylated antiestrogen action in the **immature rat uterus.** A comparison of hydroxylated antiestrogens with 191. KIECHHOFF, J., GRUNKE, W., HOFFMANN, B., NAGEL, W., AN chain for the estrogenic and antiestrogenic actions of tamoxifen and
trioxifene in the immature rat uterus. Mol. Cell. Endocrinol. 27: 291-
306, 1982.
1980. JORDAN, V. C., AND GOSDEN, B.: Differential antiestrogen action i estative rat uterus: A comparison of hydroxylated antiestrogens with
high affinity for the strogen receptor. J. Steroid Biochem. 19: 1249-
1258, 1983.
show, V. C., AND GosDEN, B.: Inhibition of the uterotropic activity of

- **Endocrinology 108: 463-468, 1983.**
 Endocrinology 12: 1249-1258, 1983.
 ETA JORDAN, V. C., AND GOSDEN, B.: Inhibition of the uterotropic activity of estrogens and antiestrogens by the short acting antiestrogen LY 11701 167. JORDAN, V. C., AND GOSDEN, B.: Inhibition of the uterotropic activity of
estrogens and antiestrogens by the short acting antiestrogen LY 117018.
Endocrinology 113: 463-468, 1963.
168. JORDAN, V. C., GOSDEN, B., AND TA
-
-
- Endocrine Society Proceeding Abstract 1049, San Antonio, 1963.

169. JORDAN, V. C., HALDEMANN, B., AND ALLEN, K. E.: Geometric isomers of substituted triphenylethylenes and antiestrogen action. Endocrinology

106: 1353-136
- agonists, partial agonists and antagonists. Pharmacologist 25: Abstract 198. KORENMAN, S. G., AND DUKES, B. A.: Specific estrogen binding by the cytoplasm of human breast carcinoma. J. Clin. Endocrinol. 30: 639–645,
171. J
-

- 273

immature rat uterus during the sequential administration of antioestro-

gena. Br. J. Pharmacol. 65: 167-173, 1979.

173. JORDAN, V. C., AND PRESTWICH, G.: Binding of [²H]tamoxifen in rat uterine

cytosols: A compar immature rat uterus during the sequential administration of antioestro-
gens. Br. J. Pharmacol. 65: 167-173, 1979.
173. JORDAN, V. C., AND PRESTWICH, G.: Binding of [²H] tamoxifen in rat uterine
cytosols: A comparison of
- effect of non-steroidal antioestrogen and non-steroidal oestrogens and provides: A comparison of swinging bucket and vertical tube rotor sucrose
density gradient analysis. Mol. Cell. Endocrinol. 8: 179–188, 1977.
RDAN, V. density gradient analysis. Mol. Cell. Endocrinol. 8: 179-188, 1977.

174. JORDAN, V. C., ROWSBY, L., DIX, C. J., AND PRESTWICH, G.: Does-related

effect of non-steroidal antioestrogens and non-steroidal oestrogens on the
 effect of non-steroidal antioestrogens and non-steroidal cestrogens on the measurement of cytoplasmic centrogen receptors in the rat and mouse uterus. J. Endocrinol. T8: 71-81, 1978.
AMBOJ, V. P., SETTY, B. S., CHANDRA, H.
- measurement of cytoplasmic osstrogen receptors in the rat and mouse
uterus. J. Endocrinol. 78: 71-81, 1978.
175. KAMBOJ, V. P., SETTY, B. S., CHANDRA, H., Roy, S. K., AND KAR, A. B.:
Biological profile of centerbrowan-A ne
- 175. KAMBOJ, V. P., SETTY, B. S., CHANDRA, H., ROY, S. K., AND KAR, A. B.:
Biological profile of centchroman—A new post-coital contraceptive. Ind.
J. Exp. biol. 15: 1144-1146, 1977.
176. KANG, Y. H., ANDERSON, W. A., AND For receptor receptors and **receptor** receptors receptors receptors and extreme for phology and growth by estradiol-17 β and estrogen antagonist.

J. Cell. Biol. 64: 682-691, 1975.

177. KASSES, J. A., AND GORSKI, J.: Es
-
- uterine morphology and growth by estradiol-17 β and estrogen antagonist.
J. Cell. Biol. 64: 682-691, 1975.
ASSISS, J.A., AND GORSKI, J.: Estrogen receptor replenishment—Evidence
for receptor recycling. J. Biol. Chem. 25 **antiestrogen action in reproductive tissues** and tumors. Receptor receptor recycling. J. Biol. Chem. 256: 7378-7382, 1981.
 TEENELLENBOGEN, B. S., AND KATEENELENBOGEN, J. A.: REG. 2018.
 TATEE, T., T8AI, T. L. S., AND 179. KATZENELLENBOGEN, B. S., BHAKOO, H. S., FERGUSON, E. R., LAN, N. C., TATER, T., TSAI, T. L. S., AND KATZENELLENBOGEN, J. A.: Estrogen and antiestrogen action in reproductive tissues and tumors. Recent Prog. 160 and th
-
- antiestrogen action in reproductive tissues and tumors. Recent Prog.
Horm. Res. 35: 259-300, 1979.
T. R. KATENELLENBOGEN, B. S., AND FERGUSON, E. R.: Antiestrogen action in
the uterus: Biological ineffectiveness of nuclear the uterus: Biological inducery
energy 97: 1-12, 1975.

antiestrogen. Endocrinology 97: 1-12, 1975.

180. KATENELENBOGEN, B.S., FERGUSON, E. R., AND LAN, N.C.: Fundamental differences in the action of estrogen and antiestr
- comparison between compounds with similar durations of action. Endo-
crinology 100: 1252-1259, 1977.
TEENELLENBOGEN, B. S., KATEENELLENBOGEN, J. A., FERGUSON, E. R.,
NEAUTHAMMER, N.: Antiestrogen interaction with uterine e crinology 100: 1252-1259, 1977.

181. KATZENELLENBOGEN, B. S., KATZENELLENBOGEN, J. A., FERGUSON, E. R.,

AND KEAUTHAMMER, N.: Antiestrogen interaction with uterine estrogen

receptors: Studies with a radiolabeled anti-est
- AND KRAUTHAMMER, N.: Antiestrogen interaction with uterine estrogen receptors: Studies with a radiolabeled anti-estrogen (CI 628). J. Biol. Chem. 253: 697-707, 1978.
 Chem. 253: 697-707, 1978.
 TEENELLENBOGEN, B. S., NO Chem. 253: 697-707, 1978.

NTERNELLENBOGEN, B. S., NORMAN, M. J., ECKERT, R. L., PELTE, S. W., ND. MANOEL, W. F.: Bioactivities, estrogen receptor interactions and plasminogen activator-inducing activities of tamoxifen and
- moxifen isomers in MCF-7 human breast cancer cells. Cancer Res. 44:

112-119, 1964.
 PARTERNELLENBOGEN, B. S., PAVLIK, E. J., ROBERTSON, D. W., AND KATE-
 ENELLENBOGEN, J. A.: Interaction of high affinity antiestrogen USI. KATZENELLENBOGEN, B. S., PAVLIK, E. J., ROBERTSON, D. W., AND KATZENELLENBOGEN, J. A.: Interaction of high affinity antiestrogen (α -[4-pyrrolidinoethoxy]phenyl-4-hydroxy- α' -nitrosostilbene CI 628M) with uterine **ENELLENBOGEN, J. A.: Interaction of high affinity antiestrogen (a-{4-pyrrolidinoethoxy]phenyl-4-hydroxy-a'-nitrosostillene CI 628M) with uterine estrogen receptor. J. Biol. Chem. 2564: 2908-2915, 1981.
ITZENELLENBOGEN, J.**
- pyrrolidinosthoxy]phenyl-4-hydroxy- α' -nitrosostilbene CI 628M) with
uterine estrogen receptor. J. Biol. Chem. 256: 2908–2915, 1961.
TXENELLENBOGEN, J. A., CARLSON, K. E., HEIMAN, D. F., ROBERTSON,
D. W., WEI, L. L., AND uterine estrogen receptor. J. Biol. Chem. 256: 2908-2915, 1981.

184. KATZENELLENBOGEN, J. A., CARLSON, K. E., HEIMAN, D. F., ROBERTS

D. W., WEI, L. L., AND KATZENELLENBOGEN, B. S.: Efficient and his

selective covalent l
- phenylj-attack of the antiestrogen receptor with [⁹H] tamoxifen

azirkine. J. Biol. Chem. 258: 3487-3495, 1983.

TEENELLENBOGEN, J. A., TATEE, T., AND ROBERT8ON, D. W.: Prepara-

tion of tritium-labeled 4-bydroxy-a-[p-(2 186. KELLY, P. A., ASSELIN, J., CARON, M. G., LABRIE, F., AND RAYNAUD, J.

186. KELLY, P. A., ASSELIN, J., CARON, M. G., LABRIE, F., AND RAYNAUD, J.

286. KELLY, P. A., ASSELIN, J., CARON, M. G., LABRIE, F., AND RAYNAUD, J
- growth of DMBA-induced mammary tumors. **J. Natured Maximum 18:** 865-679, 1961.

186. KELLY, P. A., ASSELIN, J., CARON, M. G., LABRIE, F., AND RAYNAUD, J. P.: Potent inhibitory effect of a new antiestrogen (RU 16117) on the **F.: Potent inhibitory effect of a new antiestrogen (RU 16117) on the growth of DMBA-induced mammary tumors. J. Natl. Cancer Inst. 58:
823-623, 1977.
ELIX, P. A., ASSELIN, J., CARON, M. G., RAYNAUD, J. P., AND LABRIE, LIX,**
- mechanisms of action. J. Toxicol. Environ. Health 4: 364-390, 1978.

187. KELLY, P. A., ASSELIN, J., CARON, M. G., RAYNAUD, J. P., AND LABRIT, U. (10 methods on F.: High inhibitory activity of a new antiestrogen, RU 16117 growth of DMBA-induced mammary tumors. J. Natl. Cancer Inst. 58:
623-628, 1977.
187. KELLY, P. A., ASSELIN, J., CARON, M. G., RAYNAUD, J. P., AND LABRIE,
F.: High inhibitory activity of a new antiestrogen, RU 16117 (11*a* F.: High inhibitory activity of a new antiestrogen, RU 16117 (11 α methoxy
ethinyl estradiol) on the development of dimethylbenz(a)anthracene-
induced mammary tumors. Cancer Res. 37: 76-81, 1977.
188. KEMP, J. V., ADAM,
	-
	- mide of 1-p-(2-dimethylaminoethoxyphenyl)-1,2,cis diphenylbut-1-ene, and biological activity of tamoxifen metabolites in human serum. Bio-
chem. Pharmacol. 32: 2045-2052, 1983.
189. KILBOURN, B. T., AND OWSTON, P. G.: Crys and biological activity of tamoxifen metabolites in human serum. Bio-
chem. Pharmacol. 32: 2045–2052, 1983.
189. KILBOURN, B. T., AND OWSTON, P. G.: Crystal structure of the hydrobro-
mide of 1-p-(2-dimenthylaminosthoxyphe 189. KILBOURN, B. T., AND OWSTON, P. G.: Crystal structure of the hydrobromide of 1-p-(2-dimethylaminoethoxyphenyl)-1,2,cis diphenylbut-1-ene, a compound with oestrogenic activity. J. Chem. Soc. B: 1-5, 1970.
190. KING, W.
	-
	- **Estrogen agonistic and antagonistic action of eight non-steroidal antiestrogens on progestin receptor or induction in rat pituitary gland and uterus. Brain Res. 289: 38-384, 1983.** roother and the multiple and the multiple method. Nature (Lond.) 307: 745-747, 1984.
191. KIRCHHOFF, J., GRUNKE, W., HOFFMANN, B., NAGEL, W., AND GHRAF, R.:
Estrogen agonistic and antagonistic action of eight non-steroidal **193. KLOPPER, A., AND HALL, M.: New synthetic agent for the induction of ovulation: Preliminary trials in women. Br. Med. J. 1: 152-154, 1971.

	193. KLOPPER, A., AND HALL, M.: New synthetic agent for the induction of ovul**
	-
	-
	-
	- ovulation: Preliminary trials in women. Br. Med. J. 1: 152-154, 1971.

	193. KoN, O. L.: An antiestrogen-binding protein in human tissues. J. Bio.

	194. KoRENMAN, S. G.: Comparative binding affinity of estrogens and its rel **ing** 193. Kon, O. L.: An antiestrogen-binding protein in human tissues. J. Biol.
Chem. 258: 3173-3177, 1983.
194. KORENMAN, S. G.: Comparative binding affinity of estrogens and its relation
to estrogenic potency. Steroids 196. KORENMAN, S. G.; Relation between estrogen inhibiting activity and binding to cytosol of rabbit and human uterus. Endocrinology 87: 1119-1923, 1970.
 A.: Specific estrogen binding by the cytoplasm of human breast ca
	- 1970.

	1970.

	1970.

	1970.

	1970.
 **KORENMAN, S. G., AND DUKES, B. A.: Specific estrogen binding by t

	cytoplesm of human breast carcinoma. J. Clin. Endocrinol. 30: 639-6

	1971. KORENMAN, S. G., AND RAO, B. R.: Reversible**
	- estrogen of human breast carcinoma. J. Clin. Endocrinol. 30: 639–645, 1970.
eytoplasm of human breast carcinoma. J. Clin. Endocrinol. 30: 639–645, 1970.
pracNMAN, S. G., AND RAO, B. R.: Reversible disaggregation of the cyt
- 198. Koseki, Y., ZAVA, D. T., CHAMNESS, G. C., AND McGuina, W. L.: Estrogen
receptor translocation and replenishment by the antiestrogen tamoxifen.
Endocrinology 101: 1104–1110, 1977.
- FORD.

199. KOSEKI, Y., ZAVA, D. T., CHAMNESS, G. C., AND MCGUIRE, W. L.: Estrogen

199. LAM, P. H.Y.: Tamoxifen is a calmodulin antagonist in the activation of

199. LAM, P. H.Y.: Tamoxifen is a calmodulin antagonist in t 220. LAM, P. H-Y.: Tamoxifen is a calmodulin antagonist in the activation of
c. AM, P. H-Y.: Tamoxifen is a calmodulin antagonist in the activation of
1964.
1964.
220. LAXIER, C. B., CAPONY, F., AND WILLIAMS, D. L.: Anti
- chick **liver** Effects **on** oestrogen receptors **and** oestrogen-inducedproteins. **Inc. Property Constraints and Constraints Constraints Comm. 118: 27-32, 1984.**
 ISBN 4.5182, C. B., CAPONY, F., AND WILLIAMS, D. L.: Antioestrogen action in chick liver. Effects on osstrogens, ed. by R. L. Sutherland and 201. LATIER, C. B., CAPONY, F., AND WILLIAMS, D. L.: Antioestrogen action in chick liver. Effects on osatrogen receptors and osatrogen-induced proteins on chick liver. Effects on osatrogen receptors and osatrogen-induced p receptor formation by estrogen receptors and osstrogen-induced proteins.
In Non-steroidal antiosetrogens, ed. by R. L. Sutherland and V. C. Jordan,
pp. 215–230, Academic Press, Sydney, 1981.
ANTT, W. C. HEN, J. J., AND ALL
- pp. 215-230, Academic Press, Sydney, 1981.

201. LEAVITT, W. W., CHEN, J. J., AND ALLEN, T. C.: Regulation of progesterone

230. receptor formation by estrogen action. Ann. N.Y. Acad. Sci. 286: 210-

225, 1977.

202. LECLE
-
- TAGNON, H. J.: Guidelines in
the design of new antiestrogens and cytotoric-linked estrogens for the
treatment of breast cancer. J. Steroid. Biochem. 19: 75-85, 1983.
203. LECLERCQ, G., HEUSON, J. C., SCHOENFELD, R., MATTHE **CLERCQ, G., HEUSON, J. C., SCHOENFELD, R., MATTHEIEM, W. H., AND TAGNON, H. J.: Estrogen receptors in human breast cancer. Eur. J. Cancer 9: 665–673, 1973.
DVNCSER, D., BABCOCK, J. C., MARLATT, P. E., LYSTER, S. C., AND D**
-
- diphenylindenes. J. Med. Chem. 8: 52-57, 1973.
204. LEDNICER, D., BABCOCK, J. C., MARLATT, P. E., LYSTER, S. C., AND
DUNCAN, G. W.: Mammalian antifertility agents. I. Derivatives of 2,3-
205. LEDNICIER, D., LYSTER, S. C., DUNCAN, G. W.: Raimmalian antifertility agents. I. Derivatives or 2,5-
2005. LEDNIGHORES. J. Med. Chem. 8: 52-57, 1965.
2005. LEDNICER, D., LYSTER, S. C., ASPERGREN, B. D., AND DUNCAN, G. W.:
Mammalian antifertility agents Mammalian antifertility agents. III. 1-Aryl-2-phenyl-1,2,3,4-tetrahydro-1-naphthols, 1-aryl-2-phenyl-3,4-dihydronaphthalene and their derivatives. J. Med. Chem. 9: 172–176, 1966.
 agents. D., Lysrues, 3. C., and DUNCAN,
- 206. LEDNICER, D., LYSTER, S. C., AND DUNCAN, G. W.: Mammalian antifertility
agents. IV. Basic 3,4-dihydronsphthalenes and 1,2,3,4-tetrahydro-1-naph-
thols. J. Med. Chem. 10: 78-66, 1967.
207. LEGHA, S. S., AND CARTER, S.
-
-
- 207. LEGHA, S. S., AND CARTER, S. K.: Antiestrogens in the treatment of breast
cancer. Cancer Treat. Rev. 3: 205-216, 1976.
208. LEMON, H. M.: Estriol prevention of mammary carcinoma induced by 7,12-
dimethylbenzanthracene
- Academic Press, Statemic Press, J. F., AND THOMPSON, C. R.: A non-steroidal estrogen antagonist 1-(p-2-diethylaminoethoxyphenyl)-1-phenyl-2-p-
mothoxyphenylothanol. Endocrinology 63: 295-318, 1958.
211. LERERMAN, M. E., GO
-
- estrogen antagonist 1-(p-2-diethylaminoethoxyphemyl)-1-phenyl-2-p-
estrogen antagonist 1-(p-2-diethylaminoethoxyphemyl)-1-phenyl-2-p-
mothoxyphemylethanol. Endocrinology 63: 295-318, 1958.
211. LIEBERMAN, M. E., GORSKI, J. model to describe the regulation of prolactin synthesis by antiestrogen
 in vitro. **J.** Biol. Chem. 258: 4741-4745, 1983.
 **EBERMAN, M. E., JORDAN, V. C., FRITSCIO, M. SANTOS, M. A., AN

GORBEI, J.: Direct and reversible 212.** LIEBERMAN, M. E., JORDAN, V. C., FRITSCH, M., SANTOS, M. A., AND GORSKI, J.: Direct and reversible inhibition of estradiol-stimulated prolactin synthesis by antiestrogen *in vitro.* J. Biol. Chem. 258: 4734-4740, 19
- **213.** LIEBERMAN, M. E., MAURER, R. A., AND GORSKI, J.: Estrogen control of

prolactin synthesis *in vitro*. Proc. Natl. Acad. Sci. U.S.A. 75: 5946-5949,
 1977.
 214. LINKIE, D. M.: Estrogen receptors in different targ
-
- 1978.

1978. 214. LINKIR, D. M.: Estrogen receptors in different target tissues: Similarities of

form-dissimilarities of transformation. Endocrinology 101: 1862-1870,

1977.

1977. D. M., AND SIFTRRI, P. K.: A re-examinat
-
-
- 218. LIPPMAN, M. E., MONACO, M. E., AND BOLAN, G.: Effects of estrone, estradiol and estriol on hormone responsive human breast cancer in long-term tissue culture. Cancer Res. 37: 1901-1907, 1977.
-
- 219. LONGCOPE, C., PRATT, J. H., SCHNEIDER, S. H., AND FINEBERG, S. E.:
Aromatization of androgens by muscle and adipose tissue in vivo. J. Clin.
Endocrinol. Metab. 46: 146-152, 1978.
220. MALLORY, F. B., WOOD, C. S., AND
- 221. MAGARIAN, R. A., **AND BENJAMIN, E. J.: Synthesis of cyclopropyl** analogs **of** stilbene **and** stilbenediol as possible antiestrogens. J. Pharm. Sci. 64: 1626-1632, 1975.

221. MAGARIAN, R. A., AND BENJAMIN, E. J.: Synthesis of cyclopropyl analog of stilbene. Roc. 86: 3094-3102, 1964.

221. MAGARIAN, R. A., AND BENJAMIN, E. J.: Synthesis of cyclopropyl analog of stilbene an 13. MAGARIAN, R. A., AND BENJAMIN, E. J.: Synthesis of cyclopropyl analogs of stilbene and stilbenediol as possible antiestrogens. J. Pharm. Sci. 64:
1626–1632, 1975.
222. MARTIN, L.: Dimethylstilbestrol and 16-oxo-estradi
-
- 1626–1632, 1975.

222. MARTIN, L.: Dimethylstilbestrol and 16-oxo-estradiol: Antiestrogens or es-

trogens. Steroids 13: 1–10, 1969.

223. MARTIN, L., AND MIDDLETON, E.: Prolonged oestrogenic and mitogenic

activity of tam
-
- 216. 125-129, 1978.

224. MARKAVERICH, B. M., AND CLARK, J. H.: Two binding aites for estradiol in

224. MARKAVERICH, B. M., AND CLARK, J. H.: Two binding aites for estradiol in

225. MARKAVERICH, B. M., ROBERTS, R. R., FI rat uterine nuclei: Relationship to uterotropic response. Endocrinology

105: 1458-1462, 1979.

225. MARKAVERICH, B. M., ROBERTS, R. R., FINNEY, R. W., AND CLARK, J. M.:

Preliminary characterization of an endogenous inhib
-

binding site **for estradiol in** estrogen action. **J.** Steroid **Biochem. 14:** 125- **132,** 1981. dexamethasone antagonism of uterine growth: A role for a second nuclear
binding site for estradiol in estrogen action. J. Steroid Biochem. 14: 125-
132, 1961.
227. MARIDAN, P. M.: Towards a new model for the mechanism
of a

-
-
- 132, 1981.

227. MARTIN, P. M., SHERIDAN, P. M.: Towards a new model for the mechanism

of action of steroids. J. Steroid Biochem. 16: 215–229, 1982.

228. MCGUIRE, W. L., CARBONE, P. P., AND VOLLMER, E. P.: Estrogen Recep Plastic agent, in plasma. Clin. Chem. 24: 1518-1524, 1978.

229. MENDENHALL, D. W., KOBAYASHI, H., SHIH, F. M. L., STERNSON, L. A., HiGOCHI, T., AND KATAZENELLENBOGEN, an antino-plastic agent, in plasma. Clin. Chem. 24: 15
- receptor transformation in MCF-7 breast cancer cells. The HIR, P. M. L., STERNSON, L. A., HIGUCHI, T., AND FABIAN, C.: Clinical analysis of tamorifen, an antineoplastic agent, in plasma. Clin. Chem. 24: 1518-1524, 1978.
 plastic agent, in plasma. Clin. Chem. 24: 1518-1524, 1978.
230. MILLER, M. A., GREENE, G. L., AND KATZENELLENBOGEN, B. S.: Estrogen receptor transformation in MCF-7 breast cancer cells: Characterization by immunchemical an
- receptor transformation in MCF-7 breast cancer cells: Characterization
by immunochemical and sedimentation analyses. Endocrinology 114:
296-298, 1984.
201. Mil.ER, M. A., AND KATZENELLENBOGEN, B. S.: Characterization and
q 233. MURPHY, L. C., AND SUTHERLAND, R. L.: A high-affinity binding site for the antiostrogens, tamorifen in advanced breast cancer. Cancer Treat. Rev. 5: 131-141, 1978.

233. MURPHY, L. C., AND SUTHERLAND, R. L.: A high-af
-
- **the antionestrogens, tamoxifen** and **CI** 628, in immature rate in and **CI** 628, in immature rate rate from the estrogens, tamoxifen and CI 628, in immature rat uterine cytosol which is distinct from the estrogen receptor. 233. MURPHY, L. C., AND SUTHERLAND, R. L.: A high-affinity binding site for
the antioestrogens, tamoxifen and CI 628, in immature rat uterine cytosol
which is distinct from the estrogen receptor. J. Endocrinol. 91: 155-161 which is distinct from the estrogen receptor. J. Endocrinol. 91: 155-161,
- 100: 1353-1360, 1981. 235. MURPHY, **L C., AND SUTHERLAND, R. L: Antitumor** activity **of clomiphene**
- band **chain of clomiphene influences affinity for a specific antioestrogen**
binding site in MCF-7 cell cytosol. Biochem. Biophys. Res. Commun.
100: 1353-1360, 1981.
analogs *in vitro*: Relationship to affinity for the estr 235. MURPHY, L. C., AND SUTHERLAND, R. L.: Antitumor activity of clomiphene analogs in vitro: Relationship to affinity for the estrogen-receptor and another high affinity antiestrogen-binding site. J. Clin. Endocrinol. Met analogs in vitro: Relationship to affinity for the estrogen receptor and another high affinity antiestrogen-binding site. J. Clin. Endocrinol. Metab. 57: 373-379, 1983.
URPHY, L. C., AND SUTHERLAND, R. L.: The interaction
- **another high affinity antiest:**
tab. 57: 373–379, 1983.
URPHY, L. C., AND SUTHERLA
antiestrogen-receptor comple
112: 707–714, 1983.
WATA, H., BRONZERT, D., AN
- dal Antioestrogens, ed. by R. L. Sutherland and V. C. Jordan, pp. 1-16, 237. NAWATA, H., BRONZERT, D., AND LIPPMAN, M. E.: Isolation and character-
Academic Press, Sydney, 1981.

ERNER, L. J., HOLTHAN, M. E.: L. J., HOLTHA tab. 57: 373-379, 1963.

236. MURPHY, L. C., AND SUTHERLAND, R. L.: The interaction of estrogen- and

antiestrogen-receptor complexes with polynucleotides. Endocrinology

112: 707-714, 1963.

237. NAWATA, H., BRONERRT, D.,
	-
	- ixation of a tamoxifen-resistant cell line derived from MCF-7 human
breast cancer cells. J. Biol. Chem. 256: 5016-5021, 1981.
238. NOTEBOOM, W. D., AND GORSKI, J.: Stereospecific binding of estrogens in
the rat uterus. Arc
	- breast cancer cells. J. Biol. Chem. 256: 5016-5021, 1981.

	238. NOTEBOOM, W. D., AND GORSKI, J.: Stereospecific binding of estrogens in

	the rat uterus. Arch. Biochem. Biophys. 111: 559-568, 1965.

	239. NOTIDES, A. C., HAM 240. Northes, A. C., **LERNER, N., AND HAMILTON, D. E.: Positive cooperativity**

	of the estrogen receptor. Proc. Natl. Acad. Sci. U.S.A. 78: 4926-4930,

	1961.

	241. Northes, A. C., AND NIELSEN, S.: The molecular mechanism o
	-
	- 1961.

	1961. Normes, A. C., AND NIELSEN, S.: The molecular mechanism of the in vitro

	1961. Normes, A. C., AND NIELSEN, S.: The molecular mechanism of the in vitro

	48 to 58 transformation of the uterine estrogen receptor.
	-
- antiestrogens on hormone responsive human breast
216. LIPPMAN, M. E., AND BOLAN, G.: Oestrogen-responsive human breast
216. LIPPMAN, M. E., AND BOLAN, G.: Oestrogen-responsive human breast
216. LIPPMAN, M. E., AND BOLAN, G Results in early G₁ phase. Cancer Res. 43: 3583-3585, 1983.

243. OUSTERHOUT, J., STRUCK, R. F., AND NELSON, J. A.: Estrogenic activities

of methoxychlor metabolites. Biochem. Pharmacol. 30: 2369-2871, 1961.

244. PALOR
	-
	- 244. PALOPOLI, F. P., FEII., V. J., ALLEN, R. F., HOLTKAMP, D. E., AND
RICHARDSON, JR., A.: Substituted aminoalkoxytriarylhaloethylenes. J.
Med. Chem. 10: 84-86, 1967.
245. PALSHOF, T.: Adjuvant endocrine therapy in the ma term Nolvadex therapy, correlated Cancer, Suppl. 7, pp. 65-75, 1961.
 TERBON, J. S., SETTATREE, R. S., ADAM, H. K., AND KEMP, J. V.:

	Serum concentrations of tamoxifen and major metabolites during long-
 INCOLUMENT ROWA Serum concentrations of tamoxifen and major metabolites during long-
term Nolvadex therapy, correlated with clinical response. In Breast
Cancer-Experimental and Clinical Aspects, ed. by H. T. Mouridsen and
T. Palabof, pp.
	-
	- activity of the non-steroidal antiestrogens analog

	II and tamorifen in 7,12-dimethylbenz[a]enthracene-induced rat mam-

	mary tumors. Cancer Lett. 15: 261-269, 1982.

	248. PENTO, J. M., MAGARIAN, R. A., WRIGHT, R. J., KING **248.** PENTO, J. M., MAGARIAN, R. A., WRIGHT, R. J., KING, M. M., AND BENJAMIN, E. J.: Nonsteroidal estrogens and antiestrogens: Biological activity of cyclopropyl analogs of stilbene and stilbenediol. J. Pharm. Sci. 70: 3
	- activity of cyclopropyl analogs of stilbene and stilbenediol. J. Pharm. Sci.
70: 399–403, 1981.
AM, S., RECHARDSON, G. S., BRADLEY, F., MACHAUGHLIN, D., SUN, L.,
FRANKEL, F., AND COHEN, J. L. Translocation of cytoplasmic **FRANKEL, F., AND COHEN, J. L. Translocation of cytoplasmic estrogen receptors to the nucleus: Immuno-histochemical demonstration utilizing rabbit antibodies to estrogen receptors of mammary carcinomas. Breast Cancer Res.** FRANKEL, F., AND COHEN, J. L. Translocation of cytoplasmic estrogen
receptors to the nucleus: Immuno-histochemical demonstration utilizing
rabbit antibodies to estrogen receptors of mammary carcinomas. Breast
Cancer Res. T
	- 250. REDDEL, R. R., MURPHY, L. C., AND SUTHERLAND, R. L.: Effects of
biologically active metabolites of tamoxifen on the proliferation kinetics
of MCF-7 human breast cancer cells in vitro. Cancer Res. 43: 4618-4624,
1983.

	-

ARMACOLO

spet

 $\overline{\mathbb{O}}$

- **ANTIESTROGEN PHAR**
252. Robertson, D. W., AND KATZENELLENBOGEN, J. A.: Synthesis of the E
and Z isomers of the antisetrogen tamoxifen and its metabolite, hy**and** *Z* isomers of the antiestrogen tamoxifen and its metabolite, hy-

droxytamoxifen, in tritium-labeled form. J. Org. Chem. 47: 2387-2393,

1982.
- droxytamoxifen, in tritium-labeled form. J. Org. Chem. 47: 2387-2393,
1962.
253. ROBERTBON, D. W., KATEENELLENBOGEN, J. A., HAYES, J. R., AND KATEENELLENBOGEN, B. S.: Antiestrogen besicity-activity relationships: A compari
- amino)etholyn-1911-1918(aminomiren Noivadex) having antered backity. J. Med. Chem. 26: 167-171, 1982.
254. ROBERTSON, D. W., KATZENELLENBOGEN, J. A., LONG, D. J., ROBKE, E.
A., AND KATZENELLENBOGEN, B. S.: Tamoxifen anties A., AND KATZENELLENBOGEN, B. S.: Tamoxifen antiestrogens. A comparison of the activity, pharmacokinetics and metabolic activation of the body. J. Pharm. Exp. J. Pharm. IS: J. Phenylbromo ethylene) in the body. J. Pharm. Ex
- 79: 340-345, 1943.
256. ROBSON, J. M., AND SCHONBERG, A.: Oestrous reactions, including mating, cis and trans isomers of tamoxifen. J. Steroid Biochem. 16: 1-13, 1962.

255. ROBSON, J. M., AND ANSARI, M. Y.: The fate of DBE ($\alpha_r a$ -di-(p-ethoxy-

phenyl) β -phenylbromo ethylene) in the body. J. Pharm. Exp. Therap.

-
-
- prolonged action **when given orally. Nature (Lond.)** 150: 22-23, 1942.
-
- tor activation by oestrogen and antioestrogen. Nature (Lond.) **292:** 257-
- 260. ROCHEFORT, H., AND BORGNA, J. L.: Differences between osstrogen receptor activation by osstrogen and antioestrogen. Nature (Lond.) 292: 257-269. 1981.

260. ROCHEFORT, H., AND BORGNA, J. L.: Differences between osstro tor activation by osstrogen and antiosstrogen. Nature (Lond.) 292: 257-

261. ROCHEFORT, H., BORGNA, J. L., COEY, E., VIGNON, F., AND WESTLEY,

B.: Mechanism of action of tamorifen and metabolites in MCF-7 human

breast ca
-
- erland and V. C. Jordan, pp. 355-364, Academic Press, Sydney, 1961.

262. ROCHEFORT, H., BORGNA, J. L., AND EVANS, E.: Cellular and molecular

mechanism of action of antiestrogens. J. Steroid Biochem. 19: 69-74,

1963.

26
- 263. ROCHEFORT, H., LIGNON, F., AND CAPONY, F.: Formation of estrogen
muclear receptor in uterus: Effects of androgen, estrone and nafoxidine.
Biochem. Biophys. Res. Commun. 47: 662-670, 1972.
264. ROSE, C., THORPE, S. M., H., AND ANDERSON, K. W.: Antiestrogen treatment of postmenopausal
women with primary high risk breast cancer. Breast Cancer Res. Treat.
8: 77-84, 1963.
ssg. D. P., Fischure, A. H., AND JORDAN, V. C.: Activity of the antios
- 266. Rosz, D. P., FISCHER, A. H., AND JORDAN, V. C.: Activity of the antiose-
trogen trioxifene against N-nitrosomethylures-induced rat mammary car-
chiarylacetophenone metabolites of the antiestrogen nitromiphene (CI 628)
- **in the presence of rat cecal contents.** Life Sci. 33: 1051-1056, 1983.
 266. RUENTTZ, P. C., AND BAGLEY, J. R.: Formation of benzophenane and α , distributed to the presence of rat cecal contents. Life Sci. 33: 1051-1
- ENITE, P. C., AND BAGLEY, J. R.: Formation of benzophenane and α ,*a*-diarylacetophenone metabolites of the antiestrogen nitromiphene (CI 628) in the presence of rat cecal contents. Life Sci. 33: 1051-1056, 1983. Interv diarylacetophenone metabolites of the antiestrogen nitromiphene (CI 628)
in the presence of rat cecal contents. Life Sci. 33: 1051-1056, 1983.
267. RUENITZ, P. C., BAGLEY, J. R., AND MOKLER, C. M.: Estrogenic and
antiestro
- 1997. RUENITZ, P. C., BAGLEY, J. R., AND MOKLER, C. M.: Estrogenic and 299. SUTHERLAND, R. L., AND JORDAN, V. C.: NOBEGODER PRESS, Sydemic antiestrogenic ectivity of monophenolic analogues of tamoxifen (Z)-2-[p-10010000000 Chem. 25: 1056-1060, 1982.

ENITZ, P. C., BAGLEY, J. R., AND MOKLER, C. M.: Estrinding and estogenic/antisestrogenic effects of two new

initromiphene, 2-[p-[nitro-1-(4-methoxyphenyl)-2-phenylvi

N-ethylpyrrolidine. J. Med 269. RUENTTZ, P. C., BAGLEY, J. R., AND MOKLER, C. M.: Estrogen receptor binding and estrogenic/antiestrogenic effects of two new metabolites of nitromiphene, 2-[p-[nitro-1-(4-methoxyphenyl)-2-phenylvinyl]phenoxy]-

Nethyb
- activity of clomiphene metabolites. Biochem. 2013. And Chem. 2013. The method is a set of the metabolites. Biochem. Pharmacol. 32: 2941-2947,

270. RUH, T. S., AND BAUDENDISTEL, L. J.: Different nuclear binding sites for a
-
- clomiphene in the rat. Estrogen receptor affinity and antiestrogenic
activity of clomiphene metabolites. Biochem. Pharmacol. 32: 2941-2947,
1983.
270. RUH, T. S., AND BAUDENDISTEL, L. J.: Different nuclear binding sites fo 426, 1977.

271. RUH, T. S., KEENE, J. L., AND ROSS, P.: Estrogen receptor binding parameters of the high affinity antiestrogen [⁹H]H1285. In Hormone Antagonistic, ed. by M. K. Agarwal, pp. 163-176, Walter de Gruyter and
- **lertify A. Physiol Chem. 274:** 39-47, 1946.
 left Chem. 273. SALEX. Physiol. Chem. 274: 39-47, 1946.
 left's Z. Physiol. Chem. 274: 39-47, 1946.
 left's Z. Physiol. Chem. 274: 39-47, 1946.
 left's Z. Physiol. Chem
- 273. SAL**ZER, W. Uber neue synthetische hochwirksame östrogene. Hoppe-Sey-ler's Z. Physiol. Chem. 274: 39–47, 1946.**
- **cancer patients.** Acts **PathoL MicrobioL Scand. (A) 74:** 301-302, 1968.
- 273. SALEER, W. Uber neue synthetische hochwirksame östrogene. Hoppe-Sey-
ler's Z. Physiol. Chem. 274: 39-47, 1946.
274. SANDER, S.: The in viro uptake of cestradiol in biopsies from 25 breast
274. SANDER, S.: The in viro as treatment of breast carcinoma: Pharmacological and clinical studies
with aromatase inhibitors. In Clinics in Oncology, ed. by B. J. A. Furr,
pp. 77-130, W. B. Saunders Company, Philadelphia, 1982.
276. SARFF, M., AND G
- **10: 2557-2563, 1971. 278. SARFF, M., AND GORSKI, J.: Control of estrogen binding protein concentration under beasl conditions after estrogen administration. Biochemistry
10: 2557-2563, 1971.
2977. SASON, S., AND NOTIDES, A. C.: The inhibition**
- I. Biochemistry into the basel conditions after estrogen administration. Biochemistry 10: 2557-2563, 1971.

277. SASSON, S., AND NOTIDES, A. C.: The inhibition of the estrogen receptor's positive cooperative [*FI*]estradio
-

- and Z isomers of the antiestrogen tamoxifen and its metabolite, hy-

droxytamoxifen, in tritium-labeled form. J. Org. Chem. 47: 2387-2393, 279. SCHNEIDER, M. R., VON ANGERER, E., SCHOENENBERGER, H., MICHEL, R.

263. ROBERT HARMACOLOGY 275
toxy-2(3'-acetoxyphenyl)-3-ethyl-1-methyl-2-indene. Eur. J. Med. Chem.
17: 245-248, 1982.
279. SCHNEIDER, M. R., von ANGERER, E., SCHOENENBERGER, H., MICHEL, R.
TH., AND FORTMEYER, H. P. 1,1,2-Triphenyl but toxy-2(3'-acetoxyphenyl)-3-ethyl-1-methyl-2-indene. Eur. J. Med. Chem.
17: 245–248, 1982.
HNEIDER, M. R., VON ANGERER, E., SCHOENENBERGER, H., MICHEL, R.
TH., AND FORTMEYER, H. P. 1,1,2-Triphenyl but-1-enes: Relationalip
b 279. SCHNEIDER, M. R., VON ANGERER, E., SCHOENENBERGER, H., MICHEL, R.
TH., AND FORTMEYER, H. P. 1,1,2-Triphenyl but-1-enes: Relationship
between structure, estradiol receptor affinity and mammary tumor in-
inibiting prope
	-
	- and its isomers upon dimethylbenz[a]anthracene-induced rat tumors.

	231. SCHULER, K. D., HASELMEIER, B., AND HOLEEL, F.: The influence of clomid

	231. SCHULER, K. D., HASELMEIER, B., AND HOLEEL, F.: The influence of clomid 281. SCHULEE, K. D., HASELMEIER, B., AND HOLEEL, F.: The influence of clone and its isomers upon dimethylbenz[a]anthracene-induced rat tumo Acta. Endocr. Copenh. (Suppl.) 138: 236. 1989.
228. SEGAL, J. S., AND NELSON, W. O
	-
	-
- 257. Rosson, J. M., AND SCHONBERG, A.: Oestrous reactions, including mating.

264. SELF, L. W., HOLTKAMP, D. E., AND KUHN, W. L.: Pituitary-gonad related

266. Rosson, J. M., AND SCHONBERG, A.: Oestrous reactions, includin and its isomers upon dimethylbenz[a]anthracene-induced rat tumors.
Acta. Endocr. Copenh. (Suppl.) 138: 236, 1969.
282. SEGAL, J. S., AND NELSON, W. O.: An orally active compound with antifer-
tility effects in rata. Proc. 283. SEGAL, J. S., AND NELSON, W. O.: Antifertility action of chloramiphene.
Anat. Rec. 139: 273, 1961.
284. SELF, L. W., HOLTKAMP, D. E., AND KUHN, W. L.: Pituitary-gonad related
effects of isomers of clomiphene citrate.
	- (effects of isomers of clomiphene citrate. Fed. Proc. Fed. Am. Soc. Exp.

	Biol. 26: 534, 1967.

	285. SHERIDAN, P. J., ANSELMO, V. C., BUCHANAN, J. M., AND MARTIN, P. M.:

	Equilibrium: The intracellular distribution of ster
	-
- 257. ROBSON, J. M., AND SCHONBERG, A.: A new synthetic cestrogen with

prolonged action when given orally. Nature (Lond.) 150: 22-23, 1942.

258. ROSSN, J. M., SCHONBERG, A., AND RAHIMS. Duration of the condensity. H., AND 285. SHERIDAN, P. J., ANSELMO, V. C., BUCHANAN, J. M., AND MARTIN, P. M.:

Equilibrium: The intracellular distribution of steroid receptors. Nature

(Lond.) 282: 579-582, 1979.

286. SILVERMAN, M., AND BOGERT, M. T.: The 267. SKIDMORE, J. R., WALFOLE, A. L., AND WOODBURN, J.: Effect of some
triphenylsthylenes on osstradiol binding *in vitro* to macromolecules from
uterus and anterior pituitary. J. Endocrinol. 52: 299-296, 1972.
289. SOULE,
	- 11: 379-392, 1956. 290. **SUDOR A., ALBERT, S., AND BRENNAN, M.: A**
human cell line from a pleural effusion derived from a breast carcinoma.
J. Natl. Cancer Inst. 51: 1409-1413, 1973.
299. STRHENBOOR, R. P.: A modification
	-
	- **J. Natl. Cancer Inst. 51: 1409-1413, 1973.**

	EPHENSON, R. P.: A modification of receptor theory. Br. J. Pharmact

	11: 379-392, 1966.

	DO, K., MONSMA, F. J., AND KATEENELLENBOGEN, B. S.: Antiestrogen

	binding sites distinc EPHENSON, R. P.: A modification of receptor theory. Br. J. Pharmacol.
11: 379–392, 1956.
Do, K., MoNSMA, F. J., AND KATZENELLENBOGEN, B. S.: Antiestrogen
binding sites distinct from the estrogen receptor: Subcellular local 11: 379-392, 1966.
 11: 379-392, 1966.

	290. SUDO, K., MONSMA, F. J., AND KATERNELLENDOGEN, B. S.: Antisetrogen binding sites distinct from the estrogen receptor: Subcellular localization, ligand specificity and distribu
	- binding sites distinct from the estrogen receptor: Subcellular localization, ligand specificity and distribution in tissues of the rat. Endocrinology 112: 425-434, 1983.
Triphenylethylene derivatives and their inter-
activ 112: 425-434, 1983.

	291. SUTHERLAND, R. L.: Estrogen antagonists in chick oviduct: Antagonist

	activity of eight synthetic triphenylethylene derivatives and their inter-

	actions with cytoplasmic and nuclear estrogen rece activity of eight synthetic triphenylethylene derivatives and their interactions with cytoplasmic and nuclear estrogen receptors. Endocrinology
109: 2061–2068, 1981.
Tri-REALAND, R. L., AND FOO, M. S.: Differential binding
	- actions with cytoplasmic and nuclear estrogen receptors. Endocrinology

	109: 2061–2068, 1981.

	292. SUTHERLAND, R. L., AND FOO, M. S.: Differential binding of antiestrogens

	by rat uterine and chick oviduct cytosol. Bioche
	- 298. Surveye and anterior pitulitary. **J. C.** C. C. The builders of anterior state of the builders deviation of the source of the state of the 292. SUTHERLAND, R. L., AND FOO, M. S.: Differential binding of antiestrogens
by rat uterine and chick oviduct cytosol. Biochem. Biophys. Res. Com-
mun. 91: 183-191, 1979.
293. SUTHERLAND, R. L., GREEN, M. D., HALL, R. E.,
	- 294. SUTHERLAND, R. L., AND JORDAN, V. C.: Nonsteroidal antioestrogens:

	Molecular pharmacology and antitumour actions. Academic Press, Syd-

	295. SUTHERLAND, R. L., MESTER, J., AND BAULIEU, E. E.: Tamoxifen is a

	potent "
	- 434-435, **1977.** ney, 1981.

	295. SUTHERLAND, R. L., MESTER, J., AND BAULIEU, E. E.: Tamoxifen is a

	potent "pure" antioestrogen in the chick oviduct. Nature (Lond.) 267:

	434-435, 1977.

	296. SUTHERLAND, R. L., AND MURPHY, L. C.: The bind
	-
	-
	- 434-435, 1977.

	296. SUTHERLAND, R. L., AND MURPHY, L. C.: The binding of tamoxifen to

	human mammary carcinoma cytosol. Eur. J. Cancer 16: 1141-1148, 1980.

	297. SUTHERLAND, R. L., AND MURPHY, L. C.: Mechanisms of oestrog onism by nonsteroidal antioestrogens. Mol. Cell. Endocrinol. **25:** 5-23,
1982.
298. SUTHERLAND, R. L., MURPHY, L. C., FOO, M. S., GREEN, M. D., WHY-
BOURNE, A. M., AND KROZOWSKI, Z. S.: High affinity antioestrogen
binding 298. SUTHERLAND, R. L., MURPHY, L. C., FOO, M. S., GREEN, M. D., WHY-BOURNE, A. M., AND KROZOWSKI, Z. S.: High affinity antioestrogen binding site distinct from the ocetrogen receptor. Nature (Lond.) 288:

	279-275.176.1980
	-
	- Eerlin, 1982.

	272. Ruts, T. S., AND JORDAN, DESOMBRE, R. R., GREENE, G. L., JENSEN, E. V., AND JORDAN,

	27. R., AND RUH, M. F.: The agonistic and antagonistic properties of V. C.: Interaction of [²H]estradiol- and [²H synthesis. Proc. Natl. Acad. Sci. U.S.A. 52: 1059-1066, 1964.

	300. TATE, A. C., DESOMBRE, E. R., GREENE, G. L., JENSEN, E. V., AND JORDAN,

	V. C.: Interaction of [⁷H]estradiol- and [⁷H]monohydroxytamoxifen-se-

	trogen
		- complexes with a monoclonal antibody. Breast Cancer
Res. Treat. 3: 267–277, 1983.
TE, A. C., GREENE, G. L., DESOMBRE, E. R., JENSEN, E. V., AND JORDAN,
V. C.: Differences between estrogen- and antisestrogen-setrogen recept Res. Treat. 3: 267-277, 1983.

		301. TATE, A. C., GREENE, G. L., DESOMBRE, E. R., JENSEN, E. V., AND JORDAN,

		V. C.: Differences between estrogen- and antiestrogen-estrogen receptor

		complexes from human breast tumors ident
		- requires the estrogen receptor. Cancer Res. 44: 1012-1018, 1984.

		302. TATE, A. C., LIEBERMAN, M. E., AND JORDAN, V. C.: The inhibition of

		prolactin synthesis in GH3 rat pituitary tumor cells by monohydroxyta-

		morifen is OHTAM)- and [H3 rat pituitary tumor cells by monohydroxyte-moxifen is associated with changes in the properties of the estrogen receptor. J. Steroid Biochem. 20: 391-395, 1984.

		The properties of the estrogen receptor comp
		- **W., C., C., C., AND** CONSTANT (B.) and [²H] estratiol (E₂) -estrogen receptor complexes in the MCF-7 breast cancer and GH₃ pituitary tumor cell lines. Mol. Cell. Endocrinol. 36: 211–219, 1984.
T. T.E., T., C
		- **304. TAm, T.,** CARLSON, K. E., KATZENELLENBOGEN, **J. A., ROBERTsON, D.**

spet

 $\overline{\mathbb{O}}$

and synthesis of a biologically important metabolite. J. Med. Chem. 22:
1509–1517, 1979.
305. TERENIUS, L.: Two modes of interaction between oestrogen and antioestro-

-
- JORDAN

and synthesis of a biologically important metabolite. J. Med. Chem. 22:

1509–1517, 1979.

305. TERNIUS, L.: Two modes of interaction between oestrogen and antioestro-

317

306. TERENIUS, L.: Structure-activity re ENGONET SUPPLEM THE SUPPLEM SUPPLEM THE SUPPLEM SUPPLEM SUPPLEM SUPPLEM SUPPLEM THAT A RESERVIUS, L.: Structure-activity relationships of antioestrogens with restroken with 17 *H* oestrogen tri-p-anisylch-control Suppl. **6** 108. TERENIUS, L.: Structure-activity relationships of antioestrogens with regard
to interaction with 17 β -oestradiol in the mouse uterus and vagina. Acta
Endocrinol Suppl. 66: 431-447, 1971.
307. THOMPSON, C. R., AND WE
-
- **Endocrinol Suppl. 66: 431–447, 1971.**
 Endocrinol Suppl. 66: 431–447, 1971.
 IOMPSON, C. R., AND WERNER, H. W.: Studies of estrogen tri-p-anisylch-
 IOMPSON, C. R., AND WERNER, H. W.: Fat storage of an estrogen in

- 309. THOMPSON, C. R., AND WERNER, H. W.: Fat storage of an estrogen in women following orally administered tri-p-anisylchloroethylene. Proc. 320. V

Soc. Exp. Biol. Med. 84: 491-492, 1953.

309. Torr, D., AND GoRskI, J.: A Soc. Erp. Biol. Med. 84: 491-492, 1953.

309. Torr, D., AND GORSKI, J.: A receptor molecule for estrogen, isolation from

rat uterus and preliminary characterization. Proc. Natl. Acad. Sci. U.S.A.

55: 1574-1581, 1966.

20 Studies using a cell-free system. Proc. Natl. Acad. Sci. U.S.A. 57: 1740-581, 1966.

Torr, D., AND GORSKI, J.: A receptor molecule for estrogens:

310. Torr, D., SHYAMALA, G., AND GORSKI, J.: A receptor molecule for estrog
- 55: 1574–1581, 1966.

310. Torr, D., SHYAMALA, G., AND GORSKI, J.: A receptor molecule for estrogens:

Studies using a cell-free system. Proc. Natl. Acad. Sci. U.S.A. 57: 1740–

1743, 1967.

1743, 1967.

1743, 1867.
 Pilo
- car Rae. Treat., 4: 297-302, 1984.

car Rea. Treat., 4: 297-302, 1984.

SI2. TWOMBLY, G. H., AND SCHOENEWALDT, E. F.: Tissue localization and

excretion routes of radioactive diethylstilbestrol. Cancer 4: 298-302, 1981.

-
- 311. TORMEY, D. C., AND JORDAN, V. C.: Long-term adjuvant therapy in node
positive breast cancer—A metabolic and pilot clinical study. Breast Cancer
free. Treat., 4: 297-302, 1984.
312. TWOMBLY, G. H., AND SCHOENEWALDT, E **WOMBLY, G. H., AND SCHOENEWALDT, E. F.: Tissue localization and excretion routes of radioactive disthylstilbestrol. Cancer 4: 296-302, 1951**
NOSBREE, T. R., OLSEN, M. R., TATE, A. C., JORDAN, V. C., AND
MOUSLER, G. C.: An **EXECUTE:** 143-149, 1984.
 **313. VAN OOSBRES, T. R., OLSEN, M. R., TATE, A. C., JORDAN, V. C., AND

MUELLER, G. C.: Antiometrical antiestrogen binds as a specific uterine

protein in addition to estrogen receptor proteins.** MUELLER, G. C.: An immobilized antiestrogen binds as a specific uterine protein in addition to estrogen receptor proteins. Mol. Cell. Endocrinol. 35: 143-149, 1984.
314. War. H. W. C.: Antiosstrogen therapy for breast canc
-
- **315. WATRELAND IN SUTHERLAND, ISSNET IN MARPHY, L. C. Antioestrogen therapy for breast cancer—A trial of the MARD, H. W. C.: Antioestrogen therapy for breast cancer—A trial of theorities at two does levels. Br. Med. J. 1:** carcinoma cells. Demonstration of high affinity and narrow specificity for transmitters, C. K. W., MURPHY, L. C. AND SUTHERLAND, R. L.: Microsomal binding sites for nonstatroidal articostroman cells. Demonstration of high **4229,** 1984. carcinoma cells. Demonstration of high affinity and narrow specificity for effects of clomiphene, MER25 and CN-56,945-27 on the rat uterus and
basic ether derivatives of triphenylethylene. J. Biol. Chem. 259: 4223-
4229, 1
-

the biological effects of tamoxifen and a new antiestrogen (LY 117018) on the immature rat uterus. **J. Endocrinol. 99:** 447-453, 1983.

- 317. We biological effects of tamoxifen and a new antiestrogen (LY 117018)
on the immature rat uterus. J. Endocrinol. 99: 447–453, 1983.
317. WEI, J. W., HICKIE, R. A., AND KLAESSEN, D. J.: Inhibition of human
breast cance the biological effects of tamoxifen and a new antiestrogen (LY 11'
on the immature rat uterus. J. Endocrinol. 99: 447–453, 1963.
EI, J. W., HICKIE, R. A., AND KLAESSEN, D. J.: Inhibition of ht
breast cancer colony formatio
- the biological effects of tamoxifen and a new antiestrogen (LY 117018)
on the immature rat uterus. J. Endocrinol. 99: 447-453, 1983.
317. WEI, J. W., HICKIE, R. A., AND KLAESSEN, D. J.: Inhibition of human
breast cancer co beast cancer colony formation by anticalmodulin agents: Trifluorpera-
ine, W-7 and W-13. Cancer Chemother. Pharmacol. 11: 86-90, 1983.
318. WEICHMAN, B. M., AND NOTIDES, A. C.: Estradiol-binding kinetics of the
activated a
-
- 318. WEIGHMAN, B. M., AND NOTIDES, A. C.: Estradiol-binding kinetics of the
activated and nonactivated estrogen receptor. J. Biol. Chem. 252: 8856-
8862, 1977.
19. WELSHONS, W. V., LIEBERMAN, M. E., AND GORSKI, J.: Nuclear 919. WELSHONS, W. V., LIEBERMAN, M. E., AND GORSKI, J.: Nuclear localisation of unoccupied oestrogen receptors. Nature (Lond.) 307: 747-749, 1984.
320. WESTLEY, B. R., AND ROCHEFORT, H.: Estradiol-induced proteins in the M
-
- 320. WESTLEY, B. R., AND ROCHEPORT, H.: Estrediol-induced proteins in the MCF-7 human breast cancer cell line. Biochem. Biophys. Res. Commun.
90: 410-416, 1979.
321. WESTLEY, B., AND ROCHEPORT, H.: A secreted glycoproteincancer. Was a care of the mother cancer cell lines. Cell 20: 353-362, 1980.

322. WILKINSON, P. M., RIBIERO, G. G., ADAM, H. K., KEMP, J. V., AND PATTERSON, J. S.: Tamoxifen (Nolvadex) therapy—rationale for loading

doe fo PATTERSON, J. S.: Tamoxifen (Nolvadex) therapy—rationale for loading
dose followed by maintenance dose for patients with metastatic breast
cancer. Cancer Chemother. Pharmacol. 10: 33-35, 1982.
323. WILLIAMSON, J. G., AND E
-
- LILIAMSON, J. G., AND ELLIS, J. P.: The induction of ovulation by
tamoxifen. J. Obstet. Gynaecol. Br. Commonw. 80: 844-847, 1973.
ILLINGHAM, M. C., WEHLAND, J., KLEE, C. B., RICHERT, N. O., RUTH-
ERFORD, A. V., AND PASTAN, 445-461, 1984. **WINNER, R. C., AND PASTAN, I. H.:** Ultrastructural immunocytochemic localization of calmodulin in cultured cells. J. Histochem. Cytochemic localization of calmodulin in cultured cells. J. Histochem. Cytoche ERFORD, A. V., AND PASTAN, I. H.: Ultrastructural immunocytochemical
localization of calmodulin in cultured cells. J. Histochem. Cytochem. 31:
1445–461, 1984.
TRIVERER, R. C., AND CLARK, J. H.: Estrogenic stimulation of th
- **1910-1915,** 1983. 326. WINNEKER, R. C., AND CLARK, J. H.: Estrogenic stimulation of the anties-
trogen specific binding site in rat uterus and liver. Endocrinology 112:
1910–1915, 1983.
WINNEKER, R. C., GUTHRIE, S. C., AND CLARK, J. H.: Cha
-
- Section and interest in the consideration of a triphenylethylene-antisetrogen-binding site on rat serum low density li
-

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