Enantioselective synthesis of ferrocene analogs of hexestrol and estradiol; recognition towards estradiol receptors

M. Gruselle and B. Malezieux

Laboratoire de Chimie Organométallique, URA 403-CNRS, Ecole Nationale Supérieure de Chimie, 11 rue P. et M. Curie, 75231 Paris Cedex 05 (France)

V.I. Sokolov and L.L. Troitskaya

Institute of Organoelements Compounds (INEOS), Russian Academy of Sciences 28, Vavilov Street, 117813 Moscow (Russian Federation)

(Received January 10, 1994)

Abstract

We describe herein the synthesis of the first non-steroidal ferrocene analogs of estradiol, in which the five-membered ring (D) is replaced by a cyclopentadienyl ligand. The control of the three chiral elements for 3-((2-hydroxymethyl)ferrocenyl)-4-(4-hydroxyphenyl)hexane (**4a**,**4b**) and 3-((2-methyl)ferrocenyl)-4-(4-hydroxyphenyl)-hexane: (**5a**,**5b**) was achieved by asymmetric cyclopalladation of *N*,*N*-dimethylaminomethylferrocene. The relative configurations were unambiguously determined from X-ray structure analyses on the racemic materials at each key step of the synthesis. All the ferrocene derivatives synthesized are recognized by the estradiol receptors. The best results are obtained for molecules possessing the same ethyl group disposition as that found in *meso*-hexestrol.

Key words: Enantioselective synthesis; Ferrocene; Synthetic estrogens

Introduction

The great potential of organometallic complexes as biologically active compounds has not been sufficiently used up to the present [1]. However, many metal compounds are non-toxic and some of them play an important role in biomolecules such as hemoglobins cytochromes, vitamin B_{12}, metallo-enzymes etc.

Synthetic compounds can exhibit biological activity only if they are able to be bound to specific receptors for natural metabolites. They have to be recognized by them due to specific groups existing in the synthetic molecules which imitate the natural substrates.

Two main strategies can be used.

First, a natural substance can be modified by introducing an organometallic moiety; the affinity for specific receptors has to be maintained in the resulting hybrid molecule. This approach can be illustrated by the method developed by Jaouen and co-workers [2] in the case of steroid hormones, see Fig. 1. These compounds have been employed as tracers in hormone receptor measurement based on the IR absorption properties of the metal–carbonyl groups. This concept, introduced as an alternative to the usual radioactive procedure, has been recently extended and confirmed to be suitable for immunologic assays [3].

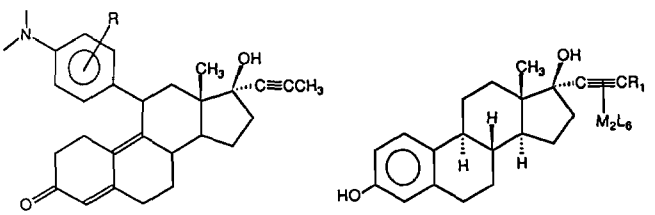


Fig. 1.

Secondly, a purely synthetic way of building organometallic analogs of natural derivatives is possible. This approach has been used by Sokolov *et al.* who succeeded in synthesizing ferrocene analogs of prostaglandins in which the five-membered ligand behaves as a latent form of the cyclopentane ring [4], Fig. 2.

Following these concepts we decided to prepare ferrocene analogs of estradiol, the structure of which

SSDI 0020-1693(94)03893-S

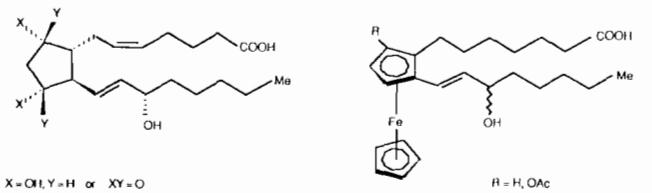


Fig. 2. General formulation of prostaglandins and their ferrocenic analogs.

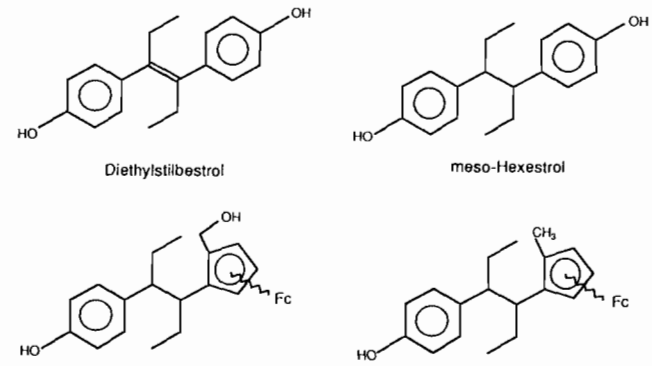


Fig. 3.

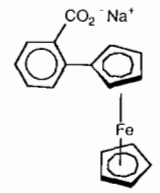


Fig. 4. Ferroceron: *o*-ferrocenyl benzoic acid.

can be compared to that of hexestrol or diethylstilbestrol, Fig. 3.

The choice of ferrocene as the organometallic core was made for the following reasons:

(i) Ferrocene has a well established and widely developed chemistry including chiral stereochemistry which is very important for preparing enantiomeric compounds [5].

(ii) More than 30 years of studies have shown that ferrocenes may be generally considered as non-toxic. A drug against iron deficiency anemia, which has been used for many years in Russia under the trade name 'Ferroceron', is the sodium salt of *o*-ferrocenylbenzoic acid, Fig. 4. Another interesting feature in therapy is the increased activity induced by the introduction of a ferrocenyl group [6]. Metabolism of ferrocenes is known as a xenobiotic process involving demetallation.

(iii) Ferricinium salts exhibit antitumor activity; they appear to be a step in the metabolism of ferrocene compounds [7]. Thus, it seems to be possible to conceive specific drugs with potential antitumoral properties and rather limited secondary effects.

The 17β-ferrocenyl estradiol has recently been prepared as both a neutral molecule and a ferricinium salt, Fig. 5. This molecule tested with specific receptors of estradiol turned out to be recognizable (*RBA* 6%) [8]. Therapeutic effects have not yet been performed.

We describe herein the synthesis of structural analogs of estradiol in which, like in the prostaglandin example, the five-membered ring D is replaced by a cyclopentadienyl ligand of ferrocene (Fig. 6).

Synthetic strategy

Asymmetric cyclopalladation of dimethylaminomethylferrocene (Scheme 1) suggests an easy access to the optically active 2-lithiated derivative with an absolute configuration of the chiral plane [9]. Moreover, the amino group allows the introduction of an hydroxyl group by way of nucleophilic substitution.

We first planned to built our target molecule through condensation of the cyclopalladated compound with 2-aryl-propanoic acid chloride previously synthesized [10] but the cyclopalladated compound in question did not react with this acid chloride as described in the literature for aryl compounds [11] (Scheme 2). However, the organolithium compound generated from the cyclopalladated compounds via the iodide derivative reacts with ketones in a normal way.

Thus, use of a chiral substituted hexane-3-one allowed us to perform the key-step of coupling two moieties affording the ferrocenic amino-alcohols. Three elements of chirality had to be controlled in constructing the framework of the steroid analog. Scheme 3 points out the main role in stereochemistry of the following reactions:

- enantioselective cyclopalladation
- attack of the ketone by the lithiated *N,N*-dimethylaminoferrocene
- replacement of the hydroxyl group by a hydrogen atom

Asymmetric cyclopalladation has been widely reported in the literature [9]; this procedure turned out to be the most appropriate way to generate the ferrocenyl unit with the expected absolute configuration. The lithiated ferrocenyl compound possesses a plane of chirality while the ketonic function will provide a chiral center. During the reaction a new asymmetric carbon C_3 is formed. Therefore, in the absence of stereospecificity, four isomers distinguishable by NMR spectroscopy would be expected. In fact, only two diastereomers were obtained in roughly equal amounts and isolated by chromatography on silica gel. This result can be ascribed to a complete, 100%, stereospecificity of this reaction, induced by one element of chirality. Relative configurations of these diastereomeric amino-alcohols were determined by X-ray diffraction analysis for the racemic series, and provided evidence that the

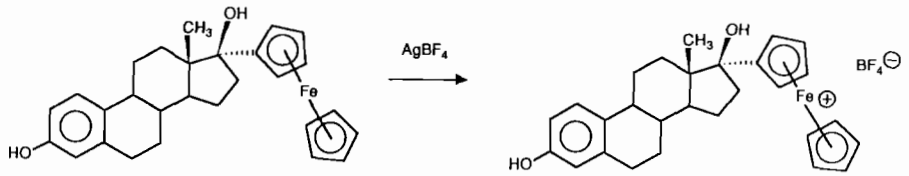


Fig. 5. 17α-Ferrocenyl estradiol and its derived ferricinium salt.

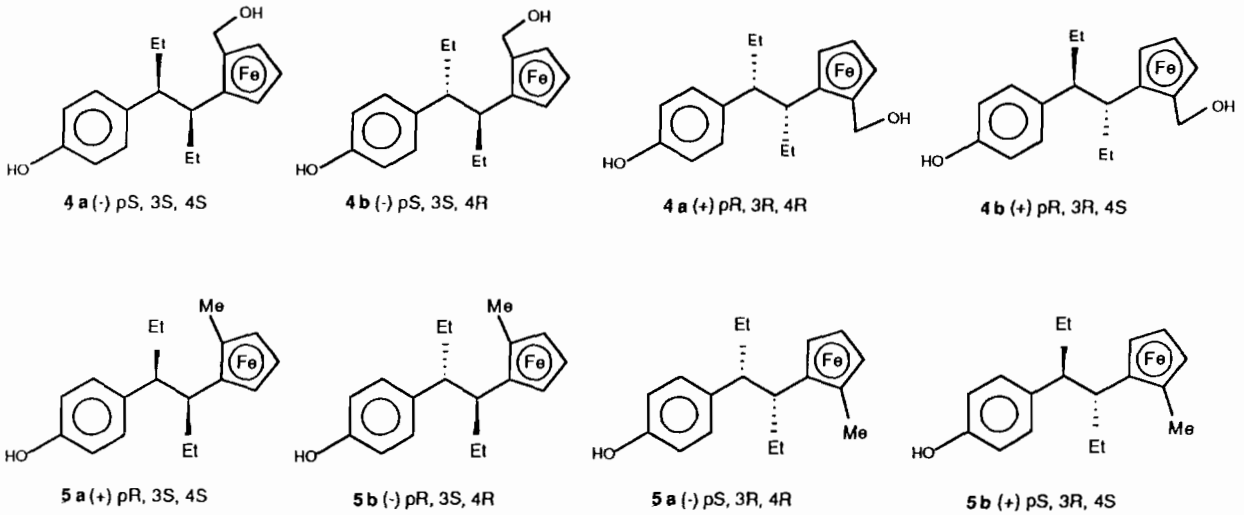
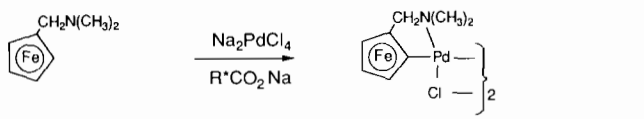
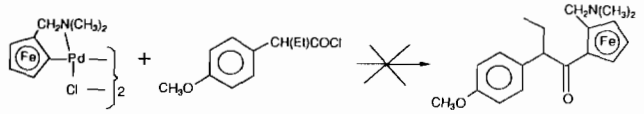


Fig. 6.



Scheme 1.

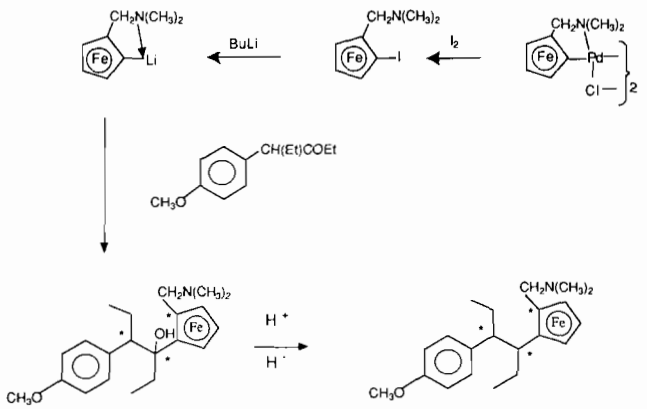


Scheme 2.

relative configurations are respectively: (pR*,3R*,4S*)-**1b** for the more polar isomer and (pR*,3S*,4R*)-**1a** for the other one [12]. The respective ORTEP plots are presented in Fig. 7.

According to Felkin's rules [13], when the reaction proceeds via a 'reactant-like' transition state, the chiral center at the α position adjacent to carbonyl group C_3, appears to be a much more efficient inductor to influence the configuration of the new chiral center C_4, than the plane of chirality of lithiated *N,N*-dimethylamino-methylferrocene. From the known configuration of the starting ferrocene moiety, the absolute configurations of the pair of diastereomers formed (Fig. 8) can be determined unambiguously.

The formal replacement of the hydroxyl function by a hydrogen atom gives, in both cases, the correct relative



Scheme 3. Synthesis of the ferrocenyl amino-alcohols by reduction of the ketone by the ferrocene lithiated compound followed by ionic reduction with retention or inversion.

configuration for C_3 and C_4 as that found in *meso*-hexestrol. Starting from amino-alcohols in racemic and optically active series the challenge is to maintain the configuration of C_3 through the reduction of the hydroxyl function.

The classical procedure described by Kursanov *et al.* [14] with trialkylsilane in acidic medium led in our case exclusively to elimination products.

The reduction of our amino-alcohols following a procedure of ionic hydrogenation reported by Gribble *et al.* [15] turned out to be a suitable method. In this

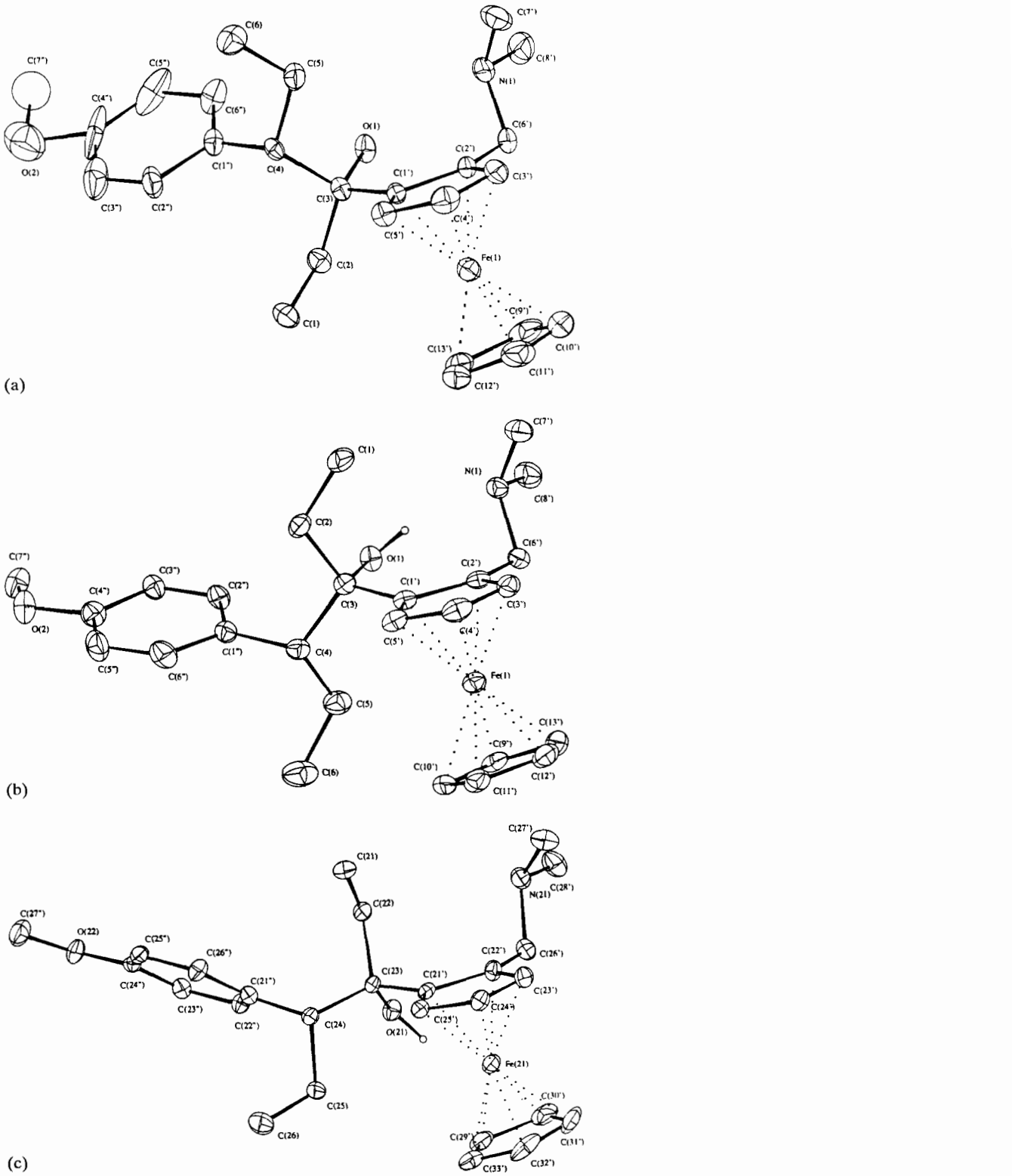


Fig. 7. ORTEP plots of: (a) (p*R*,3*R*,4*S*)-**1b**; (b) (p*R*,3*S*,4*R*)-**1a** conformer 1 and (c) conformer 2.

strategy, extended by Nicholas and Siegel to $Co_2(CO)_6$–propargylic alcohol complexes [16] (in which sodium borohydride is used as reducing agent and trifluoro acetic acid as protonating reagent) elimination is largely suppressed (Scheme 4). Starting from the amino-alcohols with the relative configurations (p*R**,3*S**,4*R**)-**1a** and (p*R**,3*R**,4*S**)-**1b** we obtained the reduced products as (p*S**,3*S**,4*S**)-**2a** and

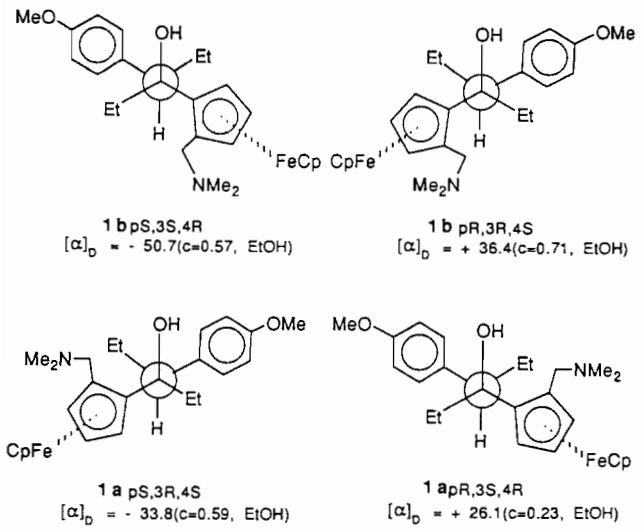
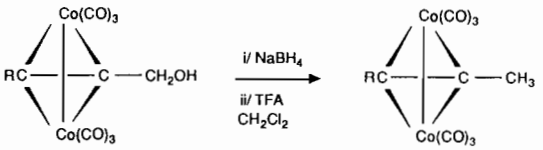


Fig. 8. Pair of enantiomers obtained by action of the racemic *N,N*-dimethylaminomethylferrocenyl lithium on the ketone.



Scheme 4.

(p*S**,3*S**,4*R**)-**2b**, respectively. Then **2a** and **2b** are treated with BBr_3 to deprotect the phenol group giving **3a** and **3b**. These relative configurations were determined by X-ray diffraction analysis [17] onto the racemic quaternary ammonium iodide salts derived from **3a** or **3b** [18] by action of methyliodide: *rac*-**3a**-MeI, *rac*-**3b**-MeI. The ORTEP plots of the structures are reported in Fig. 9.

As can be seen, the stereochemical course of the ionic hydrogenation is drastically different for the two diastereomeric amino-alcohols. The reaction would be expected to give the product (p*S**,3*S**,4*R**)-**2b** starting from (p*R**,3*R**,4*S**)-**1b**, i.e. 'normal' retention for C_3. However, we observed a total inversion for the other diastereomer (p*R**,3*S**,4*R**)-**1a** leading to (p*S**,3*S**,4*S**)-**2a**.

As a matter of fact, the α-hydroxyl chiral compounds belonging to ferrocene [19] or benchrotrene series [20], reacted under ionic hydrogenation conditions with retention of configuration (Fig. 10). This behavior can be explained by the metal stabilization of the carbocations.

However, few exceptions are known for chromium carbonyl complexes in which steric strains determine the geometry of the carbenium ion formed [20]. To our knowledge, only one example of inversion in the ferrocene series has been reported [8b]. In our case, this unexpected result is related to the different conformations for the diastereomers in solution which are caused by the respective nature of the hydrogen bonds formed.

A confirmation of this hypothesis was obtained by the results of the elimination reactions performed with TFA in $CHCl_3$ which always led to a mixture of two tertiary and one quaternary olefins either *trans*-**6** and (*cis*+*trans*)-**7** or *cis*-**6** and (*cis*+*trans*)-**8**, starting from the amino-alcohol **1a** or **1b**, respectively (Scheme 5). Nevertheless, it is noteworthy that for elimination in the case of (p*R**,3*R**,4*S**)-**1b** part of the unreacted starting material is always recuperated. This can be explained by the reluctance to ionization.

With regard to the ionization step process outlined above, the *anti*-elimination of the hydrogen atom from C_4 leads to the *cis* or *trans* quaternary olefin, depending on the amino-alcohol chosen (Fig. 11).

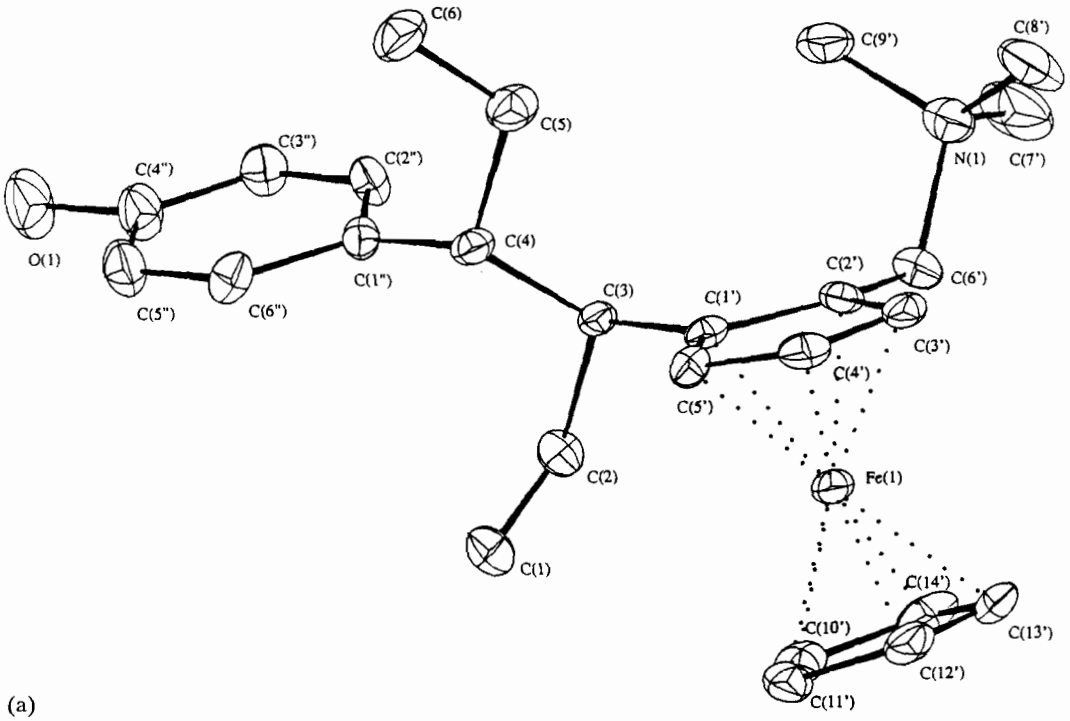
Further deprotection steps to obtain our target molecules from reduced products do not affect the stereochemistry. The targeted diols **4a** and **4b** have been prepared after demethylation by BBr_3 [18]. The dimethyl amino function could be substituted by an acetate group using an excess of acetic anhydride at 100 °C for 20 h [21]. The expected diols were obtained in good yields after hydrolysis. The reduced products **5a** and **5b** can be prepared from the acetates or from the quaternary ammonium salts by ionic hydrogenation according to the procedure of Nicholas (Scheme 6).

Relative binding affinity of the diols and subsequent reduction products towards estradiol receptors

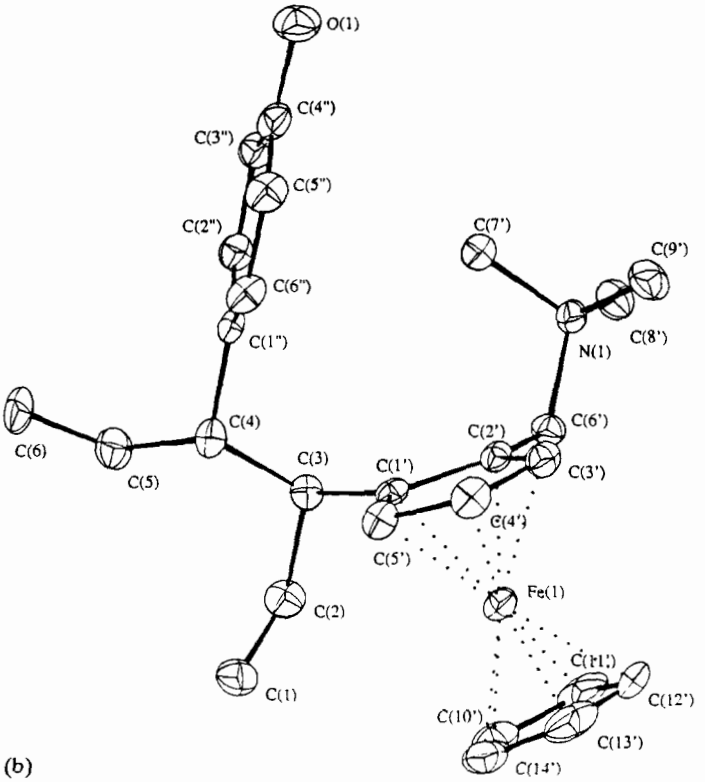
The relative binding affinity (*RBA*) is a quantifiable measure of the ability of a molecule to attach itself to the specific receptor sites. In this study the *RBA* of estradiol itself is assigned a value of 100%. Diols **4a**,**4b** and reduced products **5a**,**5b** enter in the class of non-steroidal hormones; they do not result from a transformation of a natural hormone by grafting an organometallic moiety. Under these conditions the *RBA* values can be attributed to the inherent activity of the synthetic compounds. The results for **4a**,**4b** and **5a**,**5b** are reported in Table 1.

These results show that all the compounds synthesized are recognized by estradiol receptors. The low values observed 0.16 to 1.07% do not exclude them from further biological investigations. For example, a non-steroidal hormone, Tamoxifen (Fig. 12), used in the hormonal treatment of breast cancer [22] exhibits an *RBA* value of 1% [23].

Furthermore, it is possible to correlate these low values with regard to those of estradiol derivatives. It is well-established that the hydroxyl functions in position 3 and 17β play an important role in the binding process,



(a)



(b)

Fig. 9. ORTEP plots of: (a) (p*S*,3*S*,4*R*)-**3b**-MeI; (b) (p*S*,3*S*,4*S*)-**3a**-MeI.

since the *RBA* values fall to 3% for the estradiol without phenolic group and to 3.4% when the 17β hydroxyl group is absent [22]. The configuration of the steroidal skeleton plays an important role itself and does not serve exclusively as a spacer between the two hydroxyl groups. This is illustrated by the enantiomer of estradiol, which has exactly the same distance between the two hydroxyl groups, and where the *RBA* value was found as 4% [22]. In our series of diols due to the stereochemistry of the reduction of starting amino-alcohols

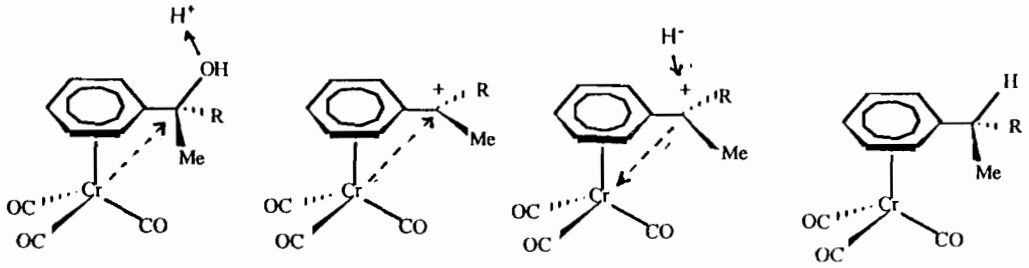
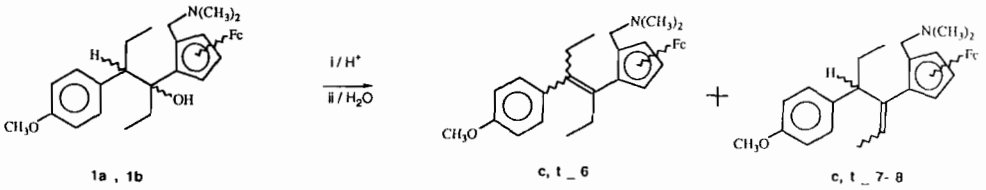


Fig. 10.



Scheme 5.

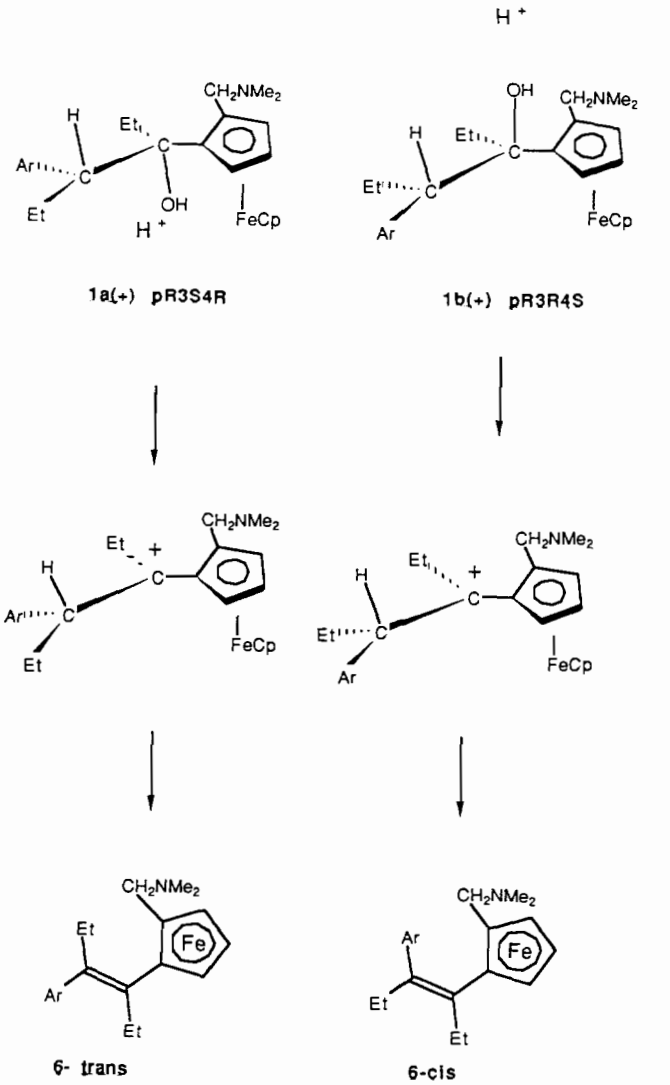
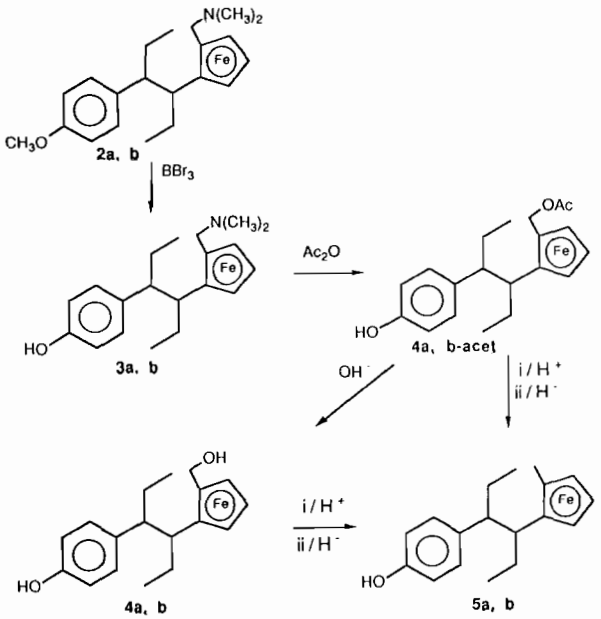


Fig. 11.



Scheme 6.

we found for (−)-**4b** the absolute configuration (p*S*,3*S*,4*R*) which corresponds to that of the enantiomeric image of estradiol (Fig. 13).

Under these conditions the comparison should be more appropriate with the enantiomer of natural estradiol. For (+)-**4b**, compared to natural estradiol, this diol differs only in the hydroxyl position on the five-membered ring. This situation is not so far from an estradiol with an aromatic ring complexed in the very disfavorable β-position in which *RBA* drastically falls to 1.67% [24]. For the series (+),(−)-**5a,5b**, the results could be related to estradiol without a hydroxyl function in the 17β-position. In this case the *RBA* value was found to be 3.4% [22]. For both series **4a,4b** and **5a,5b**

TABLE 1. Relative binding affinities towards estradiol receptors of the ferrocenic optically enriched analogs

	RBA[a]			
	(p*S*,3*S*,4*S*)(−)**4a** (p*R*,3*S*,4*S*)(+)**5a**	(p*R*,3*R*,4*R*)(+)**4a** (p*S*,3*R*,4*R*)(−)**5a**	(p*S*,3*S*,4*R*)(−)**4b** (p*R*,3*S*,4*R*)(−)**5b**	(p*R*,3*R*,4*S*)(+)**4b** (p*S*,3*R*,4*S*)(+)**5b**
Series **4**	0.20	0.16	0.24	0.34
Series **5**	0.31	0.33	0.93	1.07

[a]Referenced to estradiol as 100%.

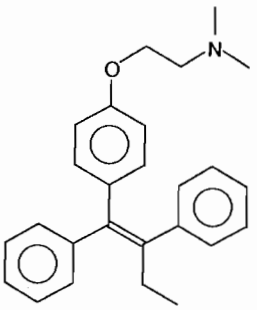
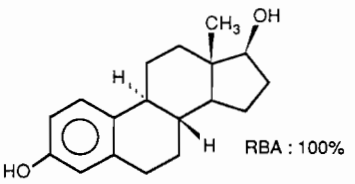
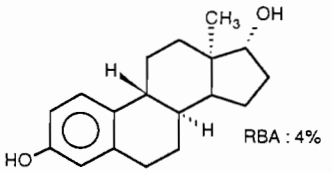


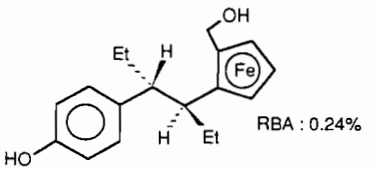
Fig. 12. Tamoxifen.



17β-Estradiol (natural estradiol)



Enantiomeric image of natural estradiol



(pS, 3S, 4R) : (-) **4b**

Fig. 13.

the values of *RBA* are higher for the isomers **4b** and **5b**, which present a *trans* disposition of the ethyl groups like in estradiol or *meso*-hexestrol.

Experimental

General considerations

NMR solvent was $CDCl_3$ or C_6D_6 (only for proton). ^1H NMR data are presented as follows: chemical shift on the δ scale, relative to TMS (multiplicity, number of protons coupling constants in Hertz). ^{13}C NMR data are presented as follows: chemical shift on the δ scale, relative to solvent as 77.0 ppm. NMR spectra were recorded on a Bruker AM 250 spectrometer.

Mass spectra were obtained on a Nermag R 10-G spectrometer.

Rotations were measured on a Perkin-Elmer model 241 MC polarimeter.

Elemental analyses were performed by the 'Service Régional de Micro Analyse' Université P. et M. Curie, Paris.

The adsorbent used for column chromatography was silica gel Merck 60 GF_{254}. Tetrahydrofuran and ether were distilled from sodium/benzophenone under argon and dichloromethane over calcium hydride before use.

Syntheses

3-(2-((*N*,*N*-Dimethylamino)methyl)ferrocenyl)-4-(4-methoxyphenyl)hexane-3-ol (**1a** and **1b**) were synthesized as reported previously [12] and reduced to 3-(2-((*N*,*N*-dimethylamino)methyl)ferrocenyl)-4-(4-methoxyphenyl)hexane (**2a** and **2b**) [17].

Enantiomeric excesses were related to those of the corresponding cyclopalladated compounds: *ee* = 60% for **1a**(+), **1b**(+), *ee* = 68% for **1a**(−), **1b**(−) and consequently for related compounds.

3-(2-((N,N-Dimethylamino)methyl)ferrocenyl)-4-(4-hydroxyphenyl)hexane (3a and 3b)

General procedure. In a cold bath at −40 °C, 81 mg (0.2 mmol) of **2** were dissolved in 5 ml of chloroform, and 0.5 ml (5.3 mmol) of BBr_3 was added under rapid stirring. The mixture was allowed to reach room temperature and after 5 min the solution turned dark brown. The mixture was cooled again before adding 30 ml of aqueous ammonia at 10%, extracted with ether (3×15 ml) and neutralized ($NaHCO_3$). The crude oil obtained after evaporation was flash-chromato-

graphed on silica gel (7730 G F_{254} Merck) (eluent Et_3N/petroleum ether: 1:4). 67 mg (80% yield) of 3-(2-((*N*,*N*-dimethylamino)methyl(ferrocenyl)-4-(4-hydroxyphenyl) hexane (**3**) were recovered.

(pS,3S*,4S*)-**3a**.* ^{1}H NMR ($CDCl_3$) δ: 0.64 (t, 3, $J=7.2$), 0.86 (t, 3, $J=7.4$), 1.67 (m, 3), 1.88 (m, 1), 2.25 (s, 6), 2.53 (m, 1), 3.04 (m, 1), 3.11 (d, 1, $J=12.8$), 3.60 (d, 1, $J=12.8$), 3.77 (dd, 1, $J=1.4$, $J=2.4$), 3.99 (t, 1, $J=2.4$), 4.01 (s, 5), 4.21 (dd, 1, $J=1.4$, $J=2.4$), 5.25 (broad, 1), 6.70 (d, 2, $J=8.5$), 6.98 (d, 2, $J=8.5$).

(pS,3S*,4R*)-**3b**.* ^{1}H NMR ($CDCl_3$) δ: 0.66 (t, 3, $J=7.1$), 1.13 (t, 3, $J=7.2$), 1.48 (m, 2), 1.78 (m, 2), 2.25 (s, 6), 2.42 (m, 1), 2.71 (m, 1), 2.73 (d, 1, $J=13.8$), 3.04 (d, 1, 13.8), 3.79 (dd, 1, $J=1.4$, $J=2.4$), 4.00 (t, 1, $J=2.4$), 4.01 (s, 5), 4.22 (dd, 1, $J=1.4$, $J=2.4$), 5.30 (broad, 1), 6.63 (d, 2, $J=8.5$), 6.73 (d, 2, $J=8.5$).

*Preparation of the protected ferrocenic alcohols: **4a**-acet, **4b**-acet*

In a degassed sealed glass tube, a mixture of 0.063 g (0.15 mmol) of **3** and 5 ml of acetic anhydride was heated at 100 °C for 20 h. Benzene (10 ml) was added and the benzene solution was washed with aqueous sodium hydroxide (10%) then water, dried ($MgSO_4$) and evaporated. The residue was chromatographed on silica gel (eluent pentane:ether 1:1). 0.042 g of **4**-acet were recovered (yield 62%).

(pS,3S*,4S*)-**4a**-acet.* ^{1}H NMR (C_6D_6) δ: 0.70 (t, 3, $J=7.2$), 1.04 (t, 3, $J=7.2$), 1.55 (m, 3), 1.72 (s, 3), 1.90 (m, 1), 2.44 (q, 1), 2.73 (h, 1), 3.45 (dd, 1, $J=1.4$, $J=2.4$), 3.80 (t, 1, $J=2.3$), 3.89 (s, 5), 4.07 (dd, 1, $J=1.4$, $J=2.4$), 4.22 (q, 2, $J=8$), 6.82 (d, 2, $J=9$), 6.99 (d, 2, $J=9$).

(pS,3S*,4R*)-**4b**-acet.* ^{1}H NMR (C_6D_6) δ: 0.67 (t, 3, $J=7.4$), 1.03 (t, 3, $J=7.4$), 1.60 (m, 1), 1.43 (m, 2), 1.71 (s, 3), 1.90 (m, 1), 2.46 (q, 1), 2.68 (q, 1), 3.60 (d, 1, $J=12$), 3.75 (dd, 1, $J=1.4$, $J=2.4$), 3.85 (d, 1, $J=12$), 3.90 (t, 1, $J=2.4$), 3.95 (s, 5), 4.11 (dd, 1, $J=1.4$, $J=2.4$), 6.77 (d, 2, $J=8$), 6.95 (d, 2, $J=8$).

(p*R*,3*R*,4*R*)-**4a**-acet: $[\alpha]_D = +14.6°$ ($C=0.22$, methanol)

(p*S*,3*S*,4*R*)-**4b**-acet: $[\alpha]_D = -37.0°$ ($C=0.17$, methanol)

(p*R*,3*R*,4*S*)-**4b**-acet: $[\alpha]_D = +48.1°$ ($C=0.08$, methanol)

The corresponding diols **4a** and **4b** were recovered quantitatively from the acetates after hydrolysis (6% KOH in methanol), refluxing for 1 h and subsequent workup. All the isomers were chromatographed on silica gel (eluent pentane/ether 1:1).

(pS,3S*,4S*)-**4a**.* ^{1}H NMR (C_6D_6) δ: 0.74 (t, 3, $J=7.2$), 1.07 (t, 3, $J=7.02$), 1.57 (m, 2), 1.93 (m, 1), 2.41 (q, 1), 2.65 (q, 1), 3.53 (dd, 1, $J=1.4$, 2.4), 3.82 (t, 1, $J=2.5$), 3.93 (s, 5), 4.06 (dd, $J=1.4$, 2.4), 4.19 (q, 2, $J=7.2$), 6.52 (d, 2, $J=8.6$), 6.72 (d, 2, $J=8.6$).

Mass spectrum (chemical ionization, NH_3 as reactant gaz): $[M]^+ = 392$, $[M+1]^+ - 18 = 375$.

(pS,3S*,4R*)-**4b**.* ^{1}H NMR (C_6D_6) δ: 0.72 (t, 3, $J=7.2$), 1.06 (t, 3, $J=7.2$), 1.44 (m, 2), 1.72 (m, 1), 1.99 (m, 1), 2.34 (q, 1), 2.67 (m, 1), 3.70 (d, 1, $J=12.4$), 3.83 (m, 1), 3.85 (d, 1, $J=12.4$), 3.88 (s, 5), 3.93 (t, 1, $J=2.5$), 4.11 (dd, 1, $J=1.4$, 2.5), 6.72 (q, 4, $J=8.8$).

Mass spectrum (chemical ionization, NH_3 as reactant gaz): $[M]^+ = 392$, $[M+1]^+ - 18 = 375$.

(p*S*,3*S*,4*S*)-**4a**: $[\alpha]_D = -7.6$ ($C=0.40$, methanol)

(p*R*,3*R*,4*R*)-**4a**: $[\alpha]_D = +6.9$ ($C=0.72$, methanol)

(p*S*,3*S*,4*R*)-**4b**: $[\alpha]_D = -30.3$ ($C=0.48$, methanol)

(p*R*,3*R*,4*S*)-**4b**: $[\alpha]_D = +29.5$ ($C=0.31$, methanol)

Anal. Calc. for $C_{23}H_{28}O_2Fe$: C, 70.42; H, 7.19. Found: for **4a**: C, 70.21; H, 7.30. Found for **4b**: C, 69.43; H, 7.50%.

*3-(2-Methylferrocenyl)-4-(4-hydroxyphenyl)hexane (**5a**,**5b**)*

0.039 g (0.1 mmol) of **4** was dissolved in 5 ml of CH_2Cl_2, and placed in a Schlenk apparatus under a flow of argon. 0.02 g of $NaBH_4$ was added at 0 °C followed by 0.15 ml of trifluoro acetic acid (TFA). After 15 min another 0.02 g of $NaBH_4$ and 0.15 ml of TFA were added and the mixture was stirred for 15 min. The excess of $NaBH_4$ was eliminated by pouring the mixture into an ice-water bath. The organic layer was separated using (3×10 ml) ether, and neutralized. After removing the solvent, the residue was chromatographed on silica gel (eluent ether/pentane 1:1), giving 0.030 g of **5** (yield 80%).

(pR,3S*,4S*)-**5a**.* ^{1}H NMR (C_6D_6) δ: 0.74 (t, 3 $J=7.4$), 1.22 (t, 3, $J=7.4$), 1.55 (q, 2, $J=7.2$), 1.88 (s, 3), 2.03 (m, 2), 2.42 (m, 1), 2.69 (m, 1), 3.42 (dd, 1, $J=1.4$, 2.4), 3.81 (t, 1, $J=2.4$), 3.92 (s, 5), 3.94 (dd, 1), 6.46 (d, 2, $J=8.6$), 6.68 (d, 2, $J=8.6$).

(pR,3S*,4R*)-**5b**.* ^{1}H NMR (C_6D_6) δ: 0.53 (t, 3, $J=7.0$), 0.99 (t, 3, $J=7.0$), 1.29 (s, 3), 1.31 (m, 2), 1.67 (m, 1), 1.91 (m, 1), 2.29 (q, 1), 2.49 (q, 1), 3.73 (m, 1), 3.87 (m, 2), 3.90 (s, 5), 6.61 (q, 4, $J=8.6$).

(p*R*,3*S*,4*S*)-**5a**: $[\alpha]_D = +37.5$ ($C=0.16$, methanol)

(p*S*,3*R*,4*R*)-**5a**: $[\alpha]_D = -29.4$ ($C=0.17$, methanol)

(p*R*,3*S*,4*R*)-**5a**: $[\alpha]_D = -18.3$ ($C=0.93$, methanol)

(p*S*,3*R*,4*S*)-**5b**: $[\alpha]_D = +22.9$ ($C=0.70$, methanol)

Anal. Calc. for $C_{23}H_{28}OFe$: C, 73.42; H, 7.50. Found for **5a**: C, 72.95; H, 7.45. Found for **5b**: C, 72.65; H, 7.41%.

*Trans-3-(2-((N,N-dimethylamino)methyl)ferrocenyl)-4-(4-methoxyphenyl)hexa-3-ene (trans-**6**) and (cis+trans)-3-(2-((N,N-dimethylamino)methyl)-ferrocenyl)-4-(4-methoxyphenyl)hexa-2-ene (cis-**7** and trans-**7**)*

0.45 g of **1a** (1 mmol) in 5 ml of CH_2Cl_2 was stirred with 0.5 ml of TFA under argon for 1 h at room temperature. After neutralization (NH_4Cl, then Na_2CO_3) the organic layer (3×10 ml ether) was dried under ($MgSO_4$) and evaporated. The residue was chromatographed on silica gel (eluent Et_3N/hexane 1:10). Three products were recovered. The most polar product isolated (0.04 g) was found to be *trans*-**6**, and the less polar products of 0.250 and 0.110 g, respectively, were *cis*- and *trans*-(p*S**,4*S**)-**7** (total yield for olefines 92.3%).

*Trans-**6**.* ^{1}H NMR ($CDCl_3$) δ: 0.64 (t, 3, J=7.5), 1.05 (t, 3, J=7.5), 1.87 (m, 1), 1.97 (m, 1), 2.22 (s, 6), 2.36 (m, 2), 3.06 (d, 2, J=13.5), 3.47 (d, 2, J=13.5), 3.84 (s, 3), 4.01 (dd, 1, J=1.4, 2.4), 4.13 (s, 5), 4.17 (t, 1, J=2.4), 4.41 (dd, 1, J=1.4, 2.4), 6.89 (dd, 2, J=7.9, 1.2), 7.03 (dd, 2, J=7.9, 1.2).

^{13}C NMR ($CDCl_3$) δ: 157.8, 143.5, 135.0, 131.6, 129.6, 113.2, 93.0, 83.0, 70.0, 69.9, 68.4, 65.5, 58.0, 55.1, 45.5, 30.9, 29.1, 15.2, 13.0.

Anal. Calc. for $C_{26}H_{33}ONFe$: C, 72.39; H, 7.71; N, 3.25. Found: C, 72.15; H, 7.54; N, 3.20%.

Trans- or cis-(pS,4S*)-**7**.* ^{1}H NMR ($CDCl_3$) δ: 1.02 (t, 3, J=7.5), 1.60 (d, 3, J=6.8), 1.76 (m, 1), 1.92 (m, 1), 2.12 (s, 6), 3.00 (d, 1, J=12.5), 3.42 (d, 1, J=12.5), 3.73 (dd, 1, J=1.4, 2.4), 3.77 (m, 1), 3.81 (s, 3), 3.90 (s, 5), 4.07 (t, 1, J=2.4), 4.33 (dd, 1, J=1.4, 2.4), 5.71 (q, 1, J=6.8), 6.83 (d, 2, J=8.4), 7.23 (d, 2, J=8.4).

^{13}C NMR ($CDCl_3$) δ: 157.7, 137.5, 136.5, 129, 125.5, 113.5, 91.5, 83.5, 69.5, 69.0, 68.3, 65.7, 58.0, 55.5, 55.0, 45.5, 30.0, 15.7, 13.3.

Anal. Calc. for $C_{26}H_{33}ONFe$: C, 72.39; H, 7.71; N, 3.25. Found: C, 72.31; H, 7.70; N, 3.20%.

Cis- or trans-(pS,4S*)-**7**.* ^{1}H NMR ($CDCl_3$) δ: 0.83 (t, 3, J=7.5), 1.54 (m, 1), 1.82 (m, 1), 1.90 (d, 3, J=7), 2.19 (s, 6), 3.27 (d, 1, J=13.4), 3.37 (m, 1), 3.39 (d, 1, J=13.4), 3.83 (s, 3), 3.87 (t, 1, J=2.0), 3.92 (m, 1), 3.94 (s, 5), 4.24 (dd, 1, J=1.4, 2.3), 6.26 (q, 1, J=7.0), 6.87 (d, 2, J=8.8), 7.22 (d, 2, J=8.8).

^{13}C NMR ($CDCl_3$) δ: 157.0, 139.0, 136.0, 129.0, 127.0, 113.0, 89.0, 85.0, 70.0, 68.5, 67.5, 65.5, 58.0, 55.0, 46.5, 45.5, 24.6, 14.5, 12.8.

Anal. Calc. for $C_{26}H_{33}ONFe$; C, 72.39; H, 7.71; N, 3.25. Found: C, 72.53; H, 7.88; N, 3.25%.

*Cis-3-(2-(N,N-dimethylamino)methyl)ferrocenyl)-4-(4-methoxyphenyl)hexa-3-ene (cis-**6**) and (cis+trans)-3-(2-((N,N-dimethylamino)methyl)ferrocenyl)-4-(4-methoxyphenyl)hexa-2-ene (cis-**8** and trans-**8**)*

0.45 g of **1b** (1 mmol) in 5 ml of CH_2Cl_2 was stirred with 0.5 ml of TFA under argon for 1 h at room temperature. After neutralization (NH_4Cl, then Na_2CO_3) the organic layer (3×10 ml ether) was dried under ($MgSO_4$) and evaporated. The residue was chromatographed on silica gel (eluent Et_3N/hexane 1:10). Four products were separated. The less polar, 0.20 g was the unreacted starting material followed by 0.02 g of *cis*-**6** and 0.18 g of *cis and trans* **8**, respectively (total yield for olefines 46%).

*Cis-**6**.* ^{1}H NMR ($CDCl_3$) δ: 0.84 (t, 3, J=7.4), 1.34 (t, 3, J=7.4), 2.18 (s, 6), 2.47 (m, 1), 2.75 (m, 2), 3.18 (q, 1), 2.29 (q, 2, J=10.0), 3.49 (dd, 1, J=1.4, J=2.4), 3.70 (s, 3), 3.78 (t, 1, J=2.4), 4.05 (s, 5), 4.13 (dd, 1, J=1.4, J=2.4), 6.60 (dd, 2, J=8.8, 1.5), 6.81 (dd, 2, J=8.8, 1.5).

^{13}C NMR ($CDCl_3$) δ: 157.3, 142.1, 135.6, 131.8, 130.3, 112.5, 92.5, 83, 71.7, 69.1, 69.0, 64.9, 58.2, 55.0, 45.6, 29.7, 27.4, 15.0, 13.0.

Anal. Calc. for $C_{26}H_{33}ONFe$: C, 72.39; H, 7.71; N, 3.25. Found: C, 72.21; H, 7.80; N, 3.19%.

Cis- or trans-(pS,4R*)-**8**.* ^{1}H NMR ($CDCl_3$) δ: 0.92 (t, 3, J=7.4), 1.52 (d, 3, J=7.4), 1.73 (m, 1), 1.98 (s, 6), 2.08 (m, 1), 2.87 (d, 1, J=13.5), 3.23 (d, 1, J=13.5), 3.77 (m, 1), 3.82 (s, 3), 3.89 (dd, 1, J=2.4, 1.4), 4.13 (t, 1, J=2.4), 4.13 (s, 5), 4.32 (dd, 1, J=2.4, 1.4), 5.57 (q, 1, J=6.8), 6.88 (d, 2, J=7.9), 7.25 (d, 2, J=7.9).

^{13}C NMR ($CDCl_3$) δ: 157.8, 138.7, 136.5, 129.5, 125.5, 113.5, 92.0, 83.0, 69.2, 68.8, 66.0, 57.5, 56.0, 55.0, 45.0, 30.0, 16.0, 13.0.

Anal. Calc. for $C_{26}H_{33}ONFe$: Calc. C, 72.39; H, 7.71; N, 3.25. Found: C, 72.42; H, 7.81; N, 3.13%.

Trans- or cis-(pS,4R*)-**8**.* ^{1}H NMR ($CDCl_3$) δ: 1.00 (t, 3, J=7.3), 1.86 (d, 3, J=7.0), 1.88 (m, 1), 1.92 (s, 6), 2.64 (d, 1, J=13.5), 3.15 (d, 1, J=13.5), 3.72 (s, 3), 3.83 (t, 1, J=7.5), 3.98 (s, 5), 4.08 (t, 1, J=2.5), 4.19 (m, 2), 6.23 (q, 2, J=7.05), 6.68 (d, 2, J=8.4), 6.97 (d, 2, J=8.4).

^{13}C NMR ($CDCl_3$) δ: 157.4, 137.5, 136.0, 127.0, 113.2, 92.0, 69.5, 68.0, 67.5, 65.5, 57.7, 55.0, 48.2, 45.4, 35.0, 27.2, 14.8, 12.8.

Anal. Calc. for $C_{26}H_{33}ONFe$: C, 72.39; H, 7.71; N, 3.25. Found: C, 72.34; H, 7.63; N, 3.15%.

Relative binding affinities

Relative binding affinities (*RBA*) were determined as follows. Lamb uterine cytosol (0.2 ml) fractions containing 4 mg of protein/ml were incubated at 0 °C

for 3 h with 2 nM [^{3}H]-17β-estradiol (Amersham, UK, specific activity 52 Ci/mmol) and increasing amounts of the competiting product (10–1000–fold excess; nine concentrations in duplicate). Bound fractions were measured by protamine sulfate precipitation [25]. The *RBA* of the competition is taken as the ratio [unlabeled estradiol]/[competitor] required to inhibit half of the specific [^{3}H]-17β-estradiol binding, with the affinity of estradiol set at 100%.

Acknowledgements

This work was supported by CNRS-France and the Russian Academy of Sciences according to agreement No. 213, and by Grant 93-03-5827 from Russian Fundamental Science Foundation. We thank A. Cordaville for the measurement of *RBA* values.

References

1 (a) A.D. Ryabov, *Angew. Chem., Int. Ed. Engl., 30* (1991) 931; (b) G. Jaouen, A. Vessieres and I.S. Butler, *Acc. Chem. Res., 26* (1993) 361.
2 (a) G. Jaouen and A. Vessieres, *Pure Appl. Chem., 61* (1989) 565; (b) G. Jaouen, A. Vessieres, S. Top, A.A. Ismail and I.S. Butler, *J. Am. Chem. Soc., 107* (1985) 4778; (c) S. Tondu, G. Jaouen, M.F. D'Agostino, K.L. Malisza and M.J. McGlinchey, *Can. J. Chem., 70* (1992) 1743.
3 (a) M. Gruselle, P. Deprez, A. Vessieres, S. Greenfield, G. Jaouen, J.P. Larue and D. Thouvenot, *J. Organomet. Chem., 359* (1989) C53–56; (b) M. Salmain, A. Vessieres, P. Brossier, I.S. Butler and G. Jaouen, *J. Immunol. Methods, 148* (1992) 65.
4 V.I. Sokolov, L.L. Troitskaya and N.S. Khrushchova, *J. Organomet. Chem., 250* (1983) 439.
5 V.I. Sokolov, *Chirality and Optical Activity in Organometallic Chemistry,* Gordon and Breach, London, 1991.
6 K.E. Dombrowski, W. Baldwin and J.E. Sheats, *J. Organomet. Chem., 302* (1986) 281.
7 (a) P. Köpf-Maier, H. Köpf and E.W. Neuse, *Angew. Chem., 302* (1986) 281; (b) E.W. Neuse and F. Kansawa, *Appl. Organomet. Chem., 4* (1990) 19.
8 (a) A. Vessieres, C. Vaillant, M. Gruselle, D. Vichard and G. Jaouen, *J. Chem. Soc., Chem. Commun.,* (1990) 837; (b) D. Vichard, M. Gruselle, G. Jaouen, M.N. Nefedova, I.A. Mamedyarova, V.I. Sokolov and J. Vaissermann, *J. Organomet. Chem.,* (1994) in press.
9 V.I. Sokolov, L.L. Troitskaya and O.A. Reutov, *J. Organomet. Chem., 182* (1979) 537.
10 M. Gruselle, B. Malezieux, G. Jaouen, L.L. Troitskaya and V.I. Sokolov, *Appl. Organomet. Chem., 4* (1990) 73.
11 P.W. Clark, H.J. Dyke, S.F. Dyke and G. Perry, *J. Organomet. Chem., 253* (1983) 399.
12 M. Gruselle, B. Malezieux, L.L. Troitskaya, V.I. Sokolov, L.M. Epstein, Y.S. Shubina and J. Vaissermann, *Organometallics, 13* (1994) 200.
13 (a) M. Cherest, H. Felkin and N. Prudent, *Tetrahedron Lett.,* (1968) 2199; (b) 2205.
14 D.N. Kursanov, Z.N. Parnes and N.M. Loim, *Synthesis,* (1974) 633.
15 G.W. Gribble, R.M. Leese and B.E. Evans, *Synthesis,* (1977) 172.
16 K.M. Nicholas and J.J. Siegel, *J. Am. Chem. Soc., 107* (1985) 4999.
17 M. Gruselle, B. Malezieux, V.I. Sokolov and L.L. Troitskaya, *Organometallics,* (1994) in press.
18 M. Gerecke, R. Borer and A. Brossi, *Helv. Chim. Acta, 59* (1976) 2551.
19 G. Eberle and I. Ugi, *Angew. Chem., Int. Ed. Engl., 15* (1976) 492.
20 S.G. Davies and T.J. Donohoe, *Synlett., 5* (1993) 323.
21 T. Hayashi, T. Mise, M. Fukushima, M. Kagotani, N. Nagashima, Y. Hamada, A. Matsumoto, S. Kawakami, M. Konishi, K. Yamamoto and M. Kumada, *Bull. Soc. Chem. Jpn., 53* (1980) 1138.
22 J. Raus, H. Martens and G. Leclerq, *Cytotoxic Estrogens in Hormone Receptive Tumors,* Vol. 2, Academic Press, New York, 1980, pp. 40–50.
23 S. Stoessel and G. Leclercq, *J. Steroid Biochem., 25* (1986) 677.
24 G. Jaouen, A. Vessieres and S. Top, *J. Am. Chem. Soc., 107* (1985) 4778.
25 A. Vessieres, S. Top, A.A. Ismail, I.S. Butler, M. Louer and G. Jaouen, *Biochemistry, 27* (1988) 6659.