10 Penning, T.D. *et al.* (1997) Synthesis and biological evaluation of the 1,5-diarylpyrazole class of cyclooxygenase-2 inhibitors: identification of 4-[5-(4-methylphenyl)-3-(trifluoromethyl)-1*H*-pyrazol-1-yl]benzene sulfonamide (SC58635, Celecoxib). *J. Med. Chem.* 40, 1347–1365

11 Ehrich, E. *et al.* (1996) Demonstration of selective COX-2 inhibition by MK-966 in humans. *Arthritis Rheum.* 39, S81 (Abstract 328)

12 Palomer, A. *et al.* (2002) Identification of novel cyclooxygenase-2 selective inhibitors using pharmacophore models. *J. Med. Chem.* 45, 1402–1411

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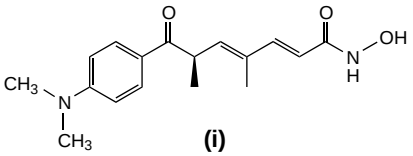
Novel antitumour molecules

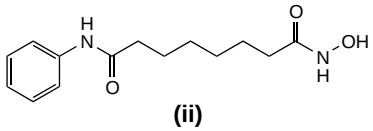
Inhibitors of human histone deacetylase as potential antitumour agents

Reversible acetylation of nuclear histones is a major regulator of gene expression, and cell-specific patterns of gene expression in normal cells result from the balance between histone acetyl transferase (HAT) and histone deacetylase (HDAC) activity. Perturbation of this balance has been linked to cancer and several natural product and synthetic HDAC inhibitors, such as trichostatin A **i** and suberoylanilide hydroxamic acid (SAHA, **ii**) have been reported to have antitumour activity.

Remiszewski and co-workers at the Novartis Institute for Biomedical Research (Summit, NJ, USA) and Westfälische Wilhelms-Universität (Münster, Germany) have reported the synthesis of analogues of SAHA and trichostatin A and their evaluation in a human HDAC enzyme inhibition assay, a $p21^{waf1}$ (p21) promoter assay and in growth inhibitory assays against selected human tumour cell lines *in vitro* [1]. The SAHA analogues prepared displayed comparable activity in the HDAC enzyme inhibition and p21 promoter assays, and among the trichostatin A analogues, enzyme and cellular



(i)



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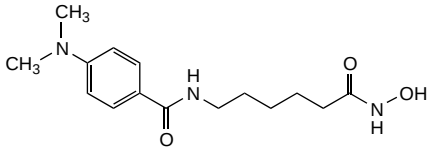

(iii)

potency was found to be related to chain length, with substitution at the 4-position of the benzamide significantly affecting enzyme potency. Compound **iii** was the most potent enzyme inhibitor among the analogues examined (IC_{50} = 0.046 μM). In addition, compound **iii** was found to affect the growth of a panel of eight human tumour cell lines differentially in the low micromolar IC_{50} range.

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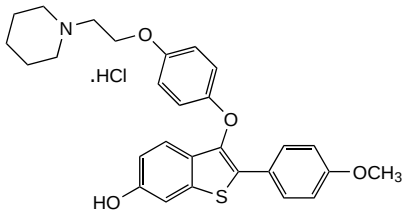
A selective oestrogen receptor modulator for chemoprevention of breast cancer

The development of safe chemopreventative strategies for women at high risk from breast cancer is a desirable alternative to the rather drastic surgical intervention through prophylactic bilateral mastectomy. The use of selective estrogen receptor modulators (SERMs) for this purpose has provided important clinical advances in this area through the development of the triphenylethylene tamoxifen and the benzothiophene raloxifene. However, the widespread clinical data on these two agents show that neither is the ultimate agent for breast cancer chemoprevention, either in terms of total efficacy or freedom from undesirable side effects, such as the increased risk of endometrial cancer with prolonged use of tamoxifen.

Sporn and co-workers at the Dartmouth Medical School (Hanover, NH, USA) and Lilly Research Laboratories (Indianapolis, IN, USA) have now reported the development of a new chemopreventative SERM termed arzoxifene **iv** [2]. Arzoxifene was found to be a selective SERM that is a potent estrogen antagonist in mammary and uterine tissue, and also acts as an estrogen agonist to maintain bone density and to lower serum cholesterol. In addition, arzoxifene is a highly effective agent (more potent than raloxifene) for the prevention of mammary cancer induced in the rat by the carcinogen nitrosomethylurea, and was also found to be devoid of the uterotrophic effects of tamoxifen, suggesting that the use of arzoxifene will not increase the risk of endometrial carcinoma.



(iv)

2 Suh, N. *et al.* (2001) Arzoxifene, a new selective estrogen receptor modulator for chemoprevention of experimental breast cancer. *Cancer Res.* 61, 8412–8415

ADEPT for selective cancer treatment: proof-of-principle

The concept of antibody-directed enzyme prodrug therapy (ADEPT) has been known for about the past 15 years, without yet resulting in widespread clinical application. The technique involves the enzymatic conversion of a non-toxic prodrug into a cytotoxic agent at the surface of tumour cells, by employing an enzyme-immuno conjugate, which docks selectively onto the surface of malignant cells prior to administration of the prodrug. Proposed requirements for the prodrug are that the corresponding

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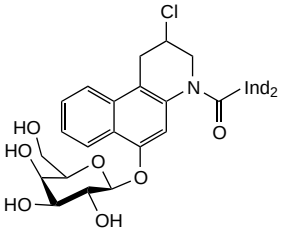
cytotoxic agent should possess an IC_{50} value of <10 nM for the target cells and the quotient of the IC_{50} of the prodrug and prodrug in the presence of the related enzyme should be >1000.

Tietze and co-workers at the George-August-Universität (Göttingen, Germany) have now described a proof-of-principle for an ADEPT system [3]. The novel prodrug developed by this group is a seco-CBI [cyclopropa(c)benz(e)indol-4-one] galactoside **v**, which on enzymatic cleavage by β-D-galactosidase results in the formation of the known cytotoxic agent CBI **vi** (related to the cytotoxic antibiotic CC1065). It should be noted that prodrug **v** contains a secondary rather than primary chloride moiety (as previously described) suggesting that direct alkylation by DNA should be diminished through steric hindrance.



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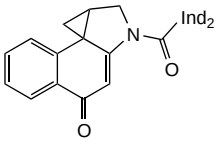

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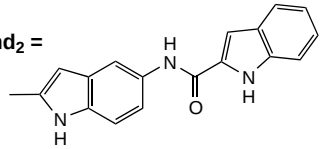

The cytotoxicity of **v** in the presence of β-D-galactosidase was examined in the human bronchial carcinoma cell line A549 and the human pancreatic ductal adenocarcinoma cell line PancTu1 for 24 h, and was found to have IC_{50} values of 0.20 nM and 0.13 nM, respectively. By contrast, the cytotoxicity was diminished in the absence of enzyme by a factor of 1600 and 3140, respectively, fulfilling ADEPT requirements in this respect. Administration of **v** to SCID mice (20 μg kg^{-1}) indicated that animals tolerated the treatment well and showed that **v** is essentially nontoxic to normal organs and blood parameters of SCID mice at therapeutic doses; this indicates that **v** is not converted to cytotoxic **vi** by enzymes expressed by the mice, an important prerequisite for ADEPT application.

Finally, *in vivo* examination of **v** was conducted in mice carrying human A549 and PancTu1 tumour cells. Treatment started on day eight after intravenous administration of the immunoconjugate of galactosidase and a human epithelial monoclonal antibody. Intraperitoneal application of compound **v** every second day (25 μg kg^{-1}) until day 30 showed that primary tumour volume in treated mice was significantly decreased. Histological examination of lung tissue showed that in treated animals the tumour is located only in small areas, compared with invasion of the whole lung in control animals. One further advantage of this prodrug is that it is reasonably water soluble and seems not to penetrate the cell membrane. Further clinical development of this ADEPT system is eagerly anticipated.

3 Tietze, L.F. *et al.* (2002) Proof-of-principle in the selective treatment of cancer by Antibody-Directed Enzyme Prodrug Therapy: the development of a highly potent prodrug. *Angew. Chem., Int. Ed. Eng.* 41, 759–761

An antiangiogenic heparin-binding arginine dendrimer

Anti-angiogenic chemotherapy has become an area of intense research interest. The process of angiogenesis is stimulated by various cytokines, such as vascular endothelial growth factor (VEGF) and basic fibroblast growth factor (FGF2). In addition, the interaction of these cytokines with their endothelial cell-surface receptors depends on the presence of the extracellular macromolecule heparin or heparan sulfate proteoglycan (HSPG).

Angiogenesis can be suppressed by endogenous inhibitors, such as angiostatin or endostatin. X-ray analysis of the mouse endostatin structure revealed a compact globular fold with one exposed positive-charge-rich face composed of 15 arginine residues. It has been proposed that the clustered arginines in endostatin compete for heparin binding sites (which act as co-receptors for several cytokines) thereby inhibiting angiogenesis. The suggestion that heparin and HSPG are potential targets for anti-angiogenic therapy has been validated to some degree by the observation that arginine-rich hexapeptides inhibited the interaction between VEGF and its receptor and suppressed angiogenesis.

Hori and co-workers at the University of Tokushima, Japan have reported the design and synthesis of two arginine-rich dendrimers and a citrulline-rich dendrimer as mimics of the surface structure of endostatin [4]. TX1944, a dendrimer containing 16 arginine residues, possessed the most potent anti-angiogenic activity in the chicken embryo chorioallantoic membrane (CAM) assay with activity comparable to endostatin and angiostatin. In addition, TX1944 was found to be the strongest heparin binder (measured by release from a heparin-agarose column) and the most active against rat lung endothelial (RLE) cells of the dendrimers tested.

4 Kasai, S. *et al.* (2002) Design and synthesis of antiangiogenic–heparin-binding arginine dendrimer mimicking the surface of endostatin. *Bioorg. Med. Chem. Lett.* 12, 951–954

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