pounds were redocked more exhaustively and then subjected to a modified similarity Tanimoto screen based upon the six TPIs. From the resulting ranked similarity and interaction energy scores, 535 ligands were selected for fully flexible ligand docking. The 535 docked solutions were then scrutinized for appealing interactions between the ligand and the active site residues, particularly hydrogen bonds, and packing efficiency in the active site.

Results: The top 40 ranked ligands were then ordered from the NCI repository. 13 were available and are currently undergoing biological evaluation.

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Structural consequences of trans-membrane association and mechanism of molecular recognition at the active site of human estrone sulfatase, a potential target for hormonal breast cancer therapy

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Human estrone (E1)/DHEA sulfatase (ES), along with cytochrome P450 aromatase and 17beta-hydroxysteroid dehydrogenase1 (17HSD1), is responsible for maintaining high levels of the active estrogen, 17beta-estradiol (E2), in tumor cells. ES catalyzes the hydrolysis of E1-sulfate, which is subsequently reduced to E2 by 17HSD1. The presence of ES in breast carcinomas and ES-dependent proliferation of breast cancer cells have been demonstrated. Selective estrogen enzyme modulators that inhibit these enzymes have shown promise as anti-proliferative agents. Rational design of specific ligands requires detailed understanding of molecular structure of the active site. Although the precise sub-cellular localization of the functional ES is not clear, this membrane-bound enzyme is distributed in the rough endoplasmic reticulum (ER). The full-length enzyme has been purified from the microsomal fraction of human placenta in the active form and crystallized. The three-dimensional structure of the enzyme has been determined by X-ray crystallography at 2.6 angstrom resolution. The structure shows a trans-membrane domain consisting of two anti-parallel alpha-helices that protrude from the roughly spherical molecule, thereby giving it a "mushroom"-like shape. These highly hydrophobic helices, each roughly 40 angstrom long and situated between residues 179 and 235, are capable of traversing the membrane, thus presumably anchoring the functional domain to the membrane surface facing the ER lumen. The location of the trans-membrane domain is such that the opening to the active site, buried deep in a cavity in the "gill" of the "mushroom", rests near the membrane surface. Furthermore, a spatially proximal polypeptide segment between residues 468 and 500, consisting of several hydrophobic sidechains and displaying high thermal motion, also presumably associate with the lipid bilayer. The residues from the membrane-associating regions line the entry path leading to the active site. The catalytic amino acid hydroxyl formylglycine 75 is found to be covalently linked to a sulfate moiety. While D35, D36, D342 and Q343 are involved in coordination of the Mg2+ ion, H290, H136, K134 and K368 play important roles in catalysis. Residues V101, F178, V177, L74 and F488 could participate in substrate recognition. Details of steroid-protein and lipid-protein interactions will be presented. This work is partially supported by the grants GM59450 and GM62794 from the NIH (to DG).

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Synthesis, structure and anticancer activity of a novel platinum(II) coordination compound of 4-aminosalicylic acid

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We have previously synthesized binuclear complexes of Pt(II)involving the naphthoquinone bridging ligand,attempting to explore the possible synergistic effect of anthracyclins and cis-platin. The results were encouraging and therefore we extended our research to Pt(II) complexes involving various hydroxyquinonic ligands. Considering that hydroxyquinones are slightly soluble, the selection of smaller ligands exhibiting coordination ability towards Pt(II) metal centers, is a good choice. Along this line, we present preliminary results on a new mixed-ligand Pt(II) complex of 4-amino-salicylic acid with imidazole. The complex was prepared by reacting of sodium 4-aminosalicylate with K2PtCl4 in aqueous medium in a 1:2 molar ratio. To the mixture, excess of saturated imidazole was added. The final mixture was stirred for 24h at RT. The dark yellow precipitate (MT: C10H8O3N3PtCl, MW: 448.5) was filtrated,washed with water and methanol and air-dried. It is soluble in DMSO.

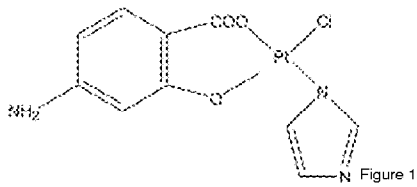

Figure 1

4ASA is coordinated to the Pt(II) central atom in a bidentating way, via the carboxylic and phenolic oxygen donor atoms forming a 6-atom ring. The imidazole and chloride ligands complete the square planar coordination environment of Pt(II). The IR spectrum confirms the above as the band at 3139 cm^{-1} is characteristic for non-coordinated –NH2 groups.Moreover,the ν(C=O) stretching vibration at 1627 cm^{-1} is shifted to lower frequencies compared to the uncoordinated ASA(C=O) group (1638 cm^{-1}).Accordingly,the ν(C-O) vibration is shifted to higher frequencies (1307 cm^{-1}) relative to that of 4ASA (1302 cm^{-1}). Finally,the ν(Pt-O) stretching vibration was found at 334 cm^{-1}. Human cancer cell lines of lung (A549), colon HCT-15), melanoma (SK-MEL-2) and ovaries (A2780) were used for the cytotoxicity test *in vitro* with the SRB assay. The cytotoxic activity was evaluated by measuring the concentration of the complex required to inhibit the protein synthesis by 50% (IC_{50}) compared to cis-platin. Each value is the result of triplicate experiments.

Table 1

Cell	cis-platin	complex
A549	0,275	0,075
HTC-15	1,490	0,650
SK-MEL-2	0,155	0,140
A-2780	0,240	0,148

The present results illustrate that the complex exhibits cytotoxicity against all cell lines tested,with higher rate of activity than that of cis-platin. Nevertheless,the solubility effects are of key importance to the improvement of the complex's toxicity. Therefore, research on the synthesis and study of new, mixed-ligand Pt(II) compounds of 4ASA and a variety of other N-donor ligands is in progress aiming to improve the solubility and bioactivity of the compounds.

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A molecular overlap tool for investigating potential binding mode similarity in sets of compounds

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The Developmental Therapeutics Program (DTP) currently offers web accessible tools such as COMPARE that use biological similarity comparison algorithms to mine the compound databases provided by DTP. By applying a method that focuses on structural aspects of these compounds, we provide an independent measurement that identifies structural commonalities that may help distinguish particular sets of compounds that are more likely to have the same underlying biochemical mechanisms of action. We present a new tool where sets of compounds can be aligned based on maximizing their molecular volume when hydrogen bond donors and acceptors are superimposed. The algorithm employs an evolutionary programming method that overlays structures based on a substitution matrix of atom types. Examples of the utility of this new application will be presented along with details about its accessibility through our web pages and integration with COMPARE.

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Development of an Mdm2/p53 fluorescence polarization high throughput inhibitor screening assay

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Mdm2 regulates p53 tumor suppressor function by three mechanisms: binding to the transactivation domain of p53, exporting p53 out of the nucleus, and ubiquitinating p53 for degradation. Mdm2 hyperactivity, due to amplification/overexpression of Mdm2 or mutational inactivation of ARF locus, is undesirable because it inhibits the function of wild type p53 and can lead