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Docking and CoMSIA studies on steroids and non-steroidal chemicals as androgen receptor ligands

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ABSTRACT

While some synthetic chemicals have been demonstrated to disrupt normal endocrine function by binding to the androgen receptor (AR), the mechanism by which ligands bind to the ligand binding domain (LBD) remained unclear. In this study, docking and comparative molecular similarity index analysis (CoMSIA) were performed to study the AR ligand binding mechanism of steroids and non-steroidal chemicals. The obtained docking conformations and predictive CoMSIA models (r^2_{pred} values as 0.842 and 0.554) indicated the primary interaction site and key residues in the binding process. The major factors influence the binding affinity of steroids and non-steroidal chemicals were electrostatic and hydrophobic interactions, respectively. The results indicated that besides amino-acid residues Gln711, Arg752 and Thr877 which have previously been reported to be important in binding ligands, Leu701 and Leu704 are also important. Residues Val746, Met749 and Phe764 are crucial only for steroids, while Met742 and Met787 are important only for non-steroidal chemicals. This knowledge of key interactions and important amino-acid residues governing ligands to the AR allow better prediction of potency of AR agonists so that their potential to disrupt AR-mediated pathways and to design less potent alternatives.

1. Introduction

As a member of the steroid and nuclear receptor superfamily, the androgen receptor (AR) and AR-mediated signal transduction pathways, through transcription of specific genes is important in development and maintenance of male sexual characteristics (AM., 1994; Gao et al., 2005; Lee and Chang, 2003; Mangelsdorf et al., 1995). Some synthetic chemicals that occur in the environment can cause adverse effects on male reproduction by mimicking androgen hormones as agonists or antagonists of the AR (Luccio-Camelo and Prins, 2011; Meeker et al., 2009). Results of several epidemiology studies have suggested disruptors of AR-mediated pathways can reduce quality of semen (Diamanti-Kandarakis et al., 2009; Swan, 2006) and cause testicular cancer (Martin et al., 2008). Considering about these results, identifying AR binding activity of different compounds is important for human health, environmental conservation and economic development.

Attention has been paid to detecting AR activity of compounds through bioassay *in vitro* or *in vivo* studies (McEwan et al., 2010; Orton et al., 2011; Svobodova et al., 2009). However, conducting bioassays on all the chemicals is time and labor consuming. Predicting activity of chemicals and safe commercial substitutes design *in silico* are more effective alternative methods. Understanding key factors and mechanisms involved in binding of ligands to the AR is import for predicting potency of AR agonists and antagonists and development of chemicals that are less likely to behave an a disruptor of AR-mediated pathways.

Mechanisms of binding of chemicals to the AR are not completely elucidated (Gao et al., 2005; Li and Al-Azzawi, 2009; Salvati et al., 2005). Some literatures have reported mechanism of binding between AR and natural steroidal ligands, xeno-steroids (Hur et al., 2004; Matias et al., 2002; Otsuka et al., 2011; Sack et al., 2001) and non-steroidal chemicals (Bohl et al., 2005; Morris et al., 1991; Poujol et al., 2000) in empirical study. However, studies focused primarily natural and synthetic pharmaceuticals but less attention has been given to synthetic, industrial chemicals. Furthermore, previous studies did not distinguish the key factors influencing binding of steroidal and non-steroidal chemicals to the AR.

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The objective of this study was to determine the most important amino-acid residues for binding of chemicals to the AR. A molecular modeling study combining molecular docking and three-dimensional quantitative structure–activity relationship (3D-QSAR) analysis was performed. Docking was used to obtain conformations of chemicals binding to the LBD of the AR and to create the alignment for developing a predictive model (Roncaglioni and Benfenati, 2008). Comparative molecular similarity index analysis (CoMSIA) was used to indicate the foremost interaction and create contour maps to find key amino-acid residues. Docking and CoMSIA were performed for steroidal and non-steroidal chemicals to determine the most likely significant amino acid residues. Predicted conformations and binding amino acids predicted by these two processes were compared and used to develop more accurate predictions of the complicated interactions between natural and synthetic chemicals and the AR LBD.

2. Materials and methods

2.1. Data sets for analysis

In this study, experimental values of the AR binding affinity for 31 steroids and 45 non-steroidal chemicals were collected from the literature (Fang et al., 2003). Fang et al. reported the AR binding affinities of 44 steroids and 102 non-steroidal compounds. Of these two classes of steroids (steroidal androgens and steroidal estrogens) and three classes non-steroidal chemicals, including phytoestrogens, phenols, and diphenylmethanes were selected for use in developing a predictive model of binding affinity to the AR LBD because of their environmental significance (Atkinson et al., 2012; Bellet et al., 2012; Brix et al., 2010; Gago-Ferrero et al., 2011; Guo et al, 2009; Liu et al., 2011, 2010). Thirty-two steroidal androgens and estrogens were included in the training set for the predictive models, but a robust predictive model based on these substances could not be developed. However, after removing 5α-androstan from the set, the model was improved. This phenomenon may due to a conflict between 5α-androstan and general AR binding properties. Potencies were expressed as the logarithm of relative binding affinity (logRBA) to the AR. The RBA was determined in a manner similar to that used in the competitive binding assay to determine relative potencies of ER agonists (Blair et al., 2000). Experimental procedures have been reported previously (Fang et al., 2003). Data for steroids were randomly divided into a training set (25 chemicals) and a test set (six chemicals). Correspondingly, non-steroidal chemicals were divided into two groups, with 38 chemicals in the training set and seven chemicals for testing. Chemicals in the test set were selected based on whether they represented the structural diversity and distribution of biological activities of chemicals in the training set. Most of the investigated chemicals are environmentally persistent and or commercially important, and some have been reported to be androgenic or antiandrogenic (Tamura et al., 2006; Wang et al., 2010). Names and potencies of these chemicals are shown (Tables 1 and 2). Molecular structures of steroids are also shown (Fig. 1) with numbering of carbon atoms. Structures of typical non-steroidal chemicals, such as flavones, *p,p′*-DDT, bisphenol A and nonylphenol are also shown (Fig. 2).

2.2. Molecular modeling, docking and alignment

Three-dimensional structures of all compounds were sketched in SYBYL7.3 molecular modeling software package (Tripos Inc, St. Louis, MO). Geometry of each molecule was optimized by use of the Tripos force field (Clark et al., 1989) with the conjugate gradient method to an energy change convergence criterion of 0.001 kcal/mol Å. Gasteiger–Hückel charges were assigned to each compound. These energy minimized structures were used as initial reasonable conformations for use in the docking study. The crystal structure of human AR complexed with R1881 (PDB code: 1XQ3) was obtained from the Protein Data Bank (http://www.rcsb.org/pdb/) He et al. (2004). The Surflex-Dock program interfaced with SYBYL 7.3 was adopted to dock the compounds to the ligand binding domain (LBD) of human AR. Prior to docking, R1881 was extracted from the crystal structure. Water molecules were removed and polar hydrogen atoms were added in standard geometry using the Biopolymer modulator. Kollman-all atom charges were assigned to protein molecule. During the docking process, automatic-based docking mode was applied with consideration of ring flexibility and other parameters set to default values. Finally, 10 binding conformations for each ligand were obtained.

The greatest TotalScore conformation of each ligand was selected as the bioactive conformation. Bioactive conformations were assigned MMFF94 charges and aligned for CoMSIA studies without further energy minimization (Castilho et al., 2006; Yang et al., 2010).

Table 1
The experimental and predicted logRBA values of steroids.

Group	No.	Name	CAS	logRBA	
				Exp	Pre
Training					
	1	Trenbolone	10161-33-8	2.05	2.199
	2	DHT benzoate	1057-07-4	0.07	0.295
	3	4-androstenediol	1156-92-9	−0.31	−0.161
	4	5α-androstan-17β-ol	1225-43-0	1.45	0.872
	5	5α-androstane-3,11,17-trione	1482-70-8	−1.64	−2.251
	6	3α-androstanediol	1852-53-5	−0.81	−0.716
	7	3-deoxyestradiol	2529-64-8	0.54	0.554
	8	3-methylestriol	3434-79-5	−2.25	−2.311
	9	Mibolerone	3704-9-4	2.27	2.17
	10	Epitestosterone	481-30-1	−1	−0.564
	11	Estradiol	50-28-2	−0.12	−0.345
	12	16β-OH-16α-Me-3-Me-estradiol	5108-94-1	−2.08	−2.153
	13	Androstenediol	521-17-5	−0.66	−0.358
	14	DHT	521-18-6	2.14	2.108
	15	5,6-didehydroisoandrosterone	53-43-0	−1.98	−1.793
	16	17-deoxyestradiol	53-63-4	−2.13	−1.83
	17	11-keto-testosterone	564-35-2	0.54	0.571
	18	3β-androstanediol	571-20-0	0.36	0.17
	19	etiocholan-17β-ol-3-one	571-22-2	−0.1	0.036
	20	ethynylestradiol	57-63-6	−1.42	−1.333
	21	T propionate	57-85-2	−0.79	−0.8
	22	17α-estradiol	57-91-0	−2.4	−2.544
	23	T	58-22-0	1.28	1.051
	24	4-OH-estradiol	5976-61-4	−0.91	−0.95
	25	4-androstenedione	63-5-8	−0.62	−0.437
Test					
	26	5α-androstan-3β-ol	1224-92-6	−0.74	−0.242
	27	2-OH-estradiol	362-05-0	−1.44	−1.12
	28	Estriol	50-27-1	−3.15	−1.615
	29	Androsterone	53-41-8	−2.12	−1.922
	30	Methyltestosterone	58-18-4	1.28	1.955
	31	R1881	965-93-5	2	1.773

2.3. CoMSIA modeling and validation

A predictive model of binding affinity was developed by use of the CoMSIA model by use of SYBYL 7.3. The same operations of modeling were performed for both steroidal and non-steroidal chemicals. To obtain CoMSIA descriptor fields, aligned training set molecules were placed into a 3-D cubic lattice with grid spacing of 2 Å in *x*, *y*, and *z* directions. Properties were calculated according to previously described methods (Klebe et al., 1994). A default value of 0.3 was used for the attenuation factor (α). Partial least-squares (PLS) regression was used for analysis of chemicals in the training set by correlating logRBA with variations in CoMSIA interaction fields. A set of descriptors was reduced to a few principal components that were linear combinations of the original descriptors by use of PLS. To evaluate reliability of the model, the leave-one-out (LOO) cross-validation method was used with a column filtering of 2.0 kcal/mol. In this method, each compound was systematically removed from the training set, after which potency was predicted by a model derived from the rest of the set. Subsequently, the cross-validated correlation coefficient, q^2_{LOO}, which characterized the predictive ability of the model, was calculated. Then, a noncross-validation was performed with the optimal number of components, and a predictive QSAR model with a conventional correlation coefficient (r^2_{ncv}), standard error of estimates (SEE) and *F* value, were generated. To refine the model, region focusing was conducted on a conventional model. Discriminant power and grid spacing were also performed to achieve a more predictive model. In addition to these validations, Monte Carlo cross-validation of groups using 10 groups repeating the procedure 50 times and bootstrap analysis for 100 runs were also carried out. Then the mean Monte Carlo cross-validated correlation coefficient (q^2_{MC}) mean bootstrap correlation (q^2_{BS}) and mean bootstrap standard error of estimates (SEE_{BS}) were generated (Mouchlis et al., 2012; Hao et al., 2011).

To evaluate the predictive capability of the CoMSIA model obtained from the training set, logRBA of the external test set was predicted. The predictive capability of the model was expressed as the predictive correlation coefficient, r^2_{pred}(Eq. (1)),

$$r^2_{pred} = \frac{SD - PRESS}{SD} \quad (1)$$

where SD is the sum of squared deviations between the logRBA of the external test set and mean potency of the training set chemicals, PRESS is the sum of squared deviations between empirical and predicted activities of the test set chemicals.

Table 2
The experimental and predicted logRBA values of nonsteroidal chemicals.

Group	No.	Name	CAS	logRBA	
				Exp	Pre
Training					
	1	igepal CO-210		−1.78	−1.881
	2	4-benzyloxylphenol	103-16-2	−2.89	−2.581
	3	4-dodecylphenol	104-43-8	−1.81	−1.616
	4	3-chlorophenol	108-43-0	−3.17	−3.008
	5	4-hydroxybenzophenone	1137-42-4	−2.78	−2.785
	6	Benzophenone	119-61-9	−2.63	−2.504
	7	4-heptyloxyphenol	13037-86-0	−1.69	−1.951
	8	2,4-dihydroxybenzophenone	131-56-6	−2.53	−2.644
	9	Dihydroxymethoxychlor olefin	14868-03-2	−1.31	−2.008
	10	4-n-octylphenol	1806-26-4	−1.8	−1.973
	11	p,p′-methoxychlor olefin	2132-70-9	−2.2	−2.101
	12	Nonylphenol	25154-52-3	−1.57	−1.701
	13	4′-hydroxychalcone	2657-25-2	−2.27	−2.459
	14	o,p′-DDE	3424-82-6	−1.81	−1.897
	15	Zearalenol	36455-72-8	−1.64	−1.608
	16	β-zearalanol	42422-68-4	−1.72	−1.657
	17	6-hydroxyflavanone	4250-77-5	−1.78	−1.826
	18	Genistein	446-72-0	−2.44	−2.467
	19	Flavanone	487-26-3	−2.25	−2.437
	20	Flavone	525-82-6	−2.4	−2.513
	21	o,p′-DDD	53-19-0	−1.52	−1.585
	22	Zearalanone	5975-78-0	−2.14	−1.862
	23	p-cumyl phenol	599-64-4	−2.11	−2.137
	24	4′-hydroxyflavanone	6515-37-3	−2.48	−2.446
	25	6-hydroxyflavone	6665-83-4	−2.77	−2.532
	26	β-zearalenol	71030-11-0	−2.09	−1.902
	27	p,p′-methoxychlor	72-43-5	−1.94	−2.062
	28	p,p′-DDD	72-54-8	−1.7	−1.656
	29	p,p′-DDE	72-55-9	−1.7	−1.746
	30	Monohydroxymethoxychlor olefin	75938-34-0	−1.84	−1.779
	31	Bisphenol B	77-40-7	−2.09	−2.073
	32	o,p′-DDT	789-02-6	−1.69	−1.329
	33	bisphenol A	80-5-7	−2.39	−2.141
	34	4-tert-amylphenol	80-46-6	−2.39	−2.509
	35	propyl parabene	94-13-3	−3	−2.782
	36	chalcone	94-41-7	−2.32	−2.502
	37	4-tert-butylphenol	98-54-4	−2.67	−2.646
	38	4-sec-butylphenol	99-71-8	−2.44	−2.445
Test					
	39	4-chloro-2-methyl phenol	1570-64-5	−2.59	−2.778
	40	4-hydroxychalcone	20426-12-4	−2.19	−2.734
	41	HPTE	2971-36-0	−1.47	−1.923
	42	p,p′-DDT	50-29-3	−1.76	−1.668
	43	equol	531-95-3	−2.39	−2.261
	44	4,4′-dihydoxybenzophenone	611-99-4	−2.67	−2.578
	45	Isoeugenol	97-54-1	−2.81	−2.429

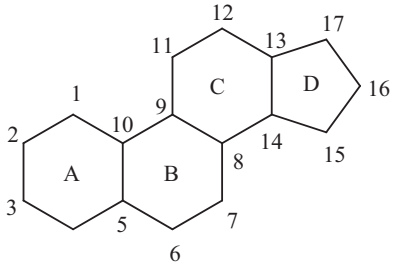

Fig. 1. The framework of steroid.

3. Results and discussion

3.1. Docking and alignment

Considering the structure of studied compounds in complex with AR has not been empirically resolved yet, docking was used to generate the active conformations and to create the alignment

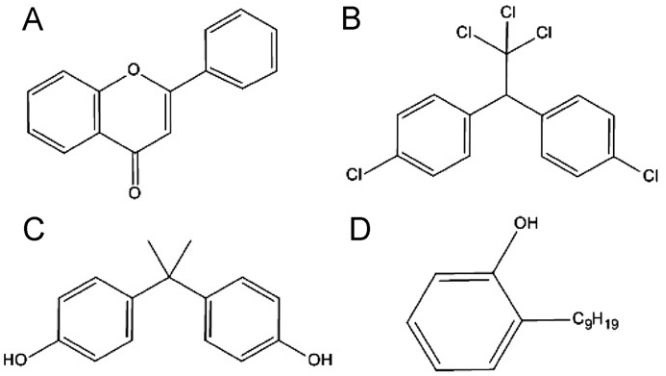

Fig. 2. The structure of nonsteroidal chemicals. (A) flavone; (B) p,p′-DDT; (C) bisphenol A; (D) nonylphenol.

for modeling (Yang et al., 2010). Docking reproduced an X-ray pose of ligand R1881 with a root-mean-square deviation (RMSD) of 0.38 Å. The proximity suggests that the conformations of the ligands analyzed are reasonably well predicted. Alignment determined based on the top-ranked docking pose among all the poses generated for each ligand shows that conformations of steroids share the same plane, and their positions are similar. The 3-keto or 3-OH groups of most steroids can form hydrogen bonds with the side chains of Gln711 and Arg752 in the LBD of the AR. Polar substituents of the C-17 position always form hydrogen bonds with amino acids Asn705 and Thr877 of the AR LBD. Similar to steroidal chemicals, all the non-steroidal chemicals are also located in the hydrophobic pocket and their positions were all proximate. Particularly, the alkylphenols which occupied only one ring are all located near Arg752, which is similar to the A-ring of steroids. Some polar groups of non-steroidal chemicals mimic the A-ring or D-ring polar substituents of steroids to form hydrogen bonds with amino acids Asn705, Gln711, Arg752 and Thr877, which is consistent with the binding of steroids. Some chemicals can also form hydrogen bonds with other residues. For example, bisphenol B can hydrogen bond to Ala765. These phenomena demonstrate that patterns of binding of steroids and non-steroidal chemicals with the AR are similar. However, there are also important differences because of the different base structures of these chemicals. Results of docking conformations of 6 chemicals, R1881, dihydrotestosterone, p,p′-DDT, bisphenol A, flavones and nonylphenol, indicated similarities in binding positions of these chemicals with divergent structures (Fig. 3).

3.2. CoMSIA statistical results and validation

Statistical results obtained from CoMSIA models for chemicals in the training set of steroids, the CoMSIA model yielded $q^2_{LOO}=0.337$ and $r^2_{ncv}=0.976$, which is less satisfactory. Regional focusing was weighted by a discriminant power value of 1 and a grid spacing of 1.0 Å. Use of regional focusing of the model yielded values of $q^2_{LOO}=0.661$, $r^2_{ncv}=0.970$. After performing the Monte Carlo cross validation and bootstrap analysis, values of were: $q^2_{MC}=0.656$, $q^2_{BS}=0.901$ and $SEE_{BS}=0.155$. In addition, the r^2_{pred} was 0.842 for the test set. For the training set of nonsteroidal chemicals, $q^2_{LOO}=0.413$ and $r^2_{ncv}=0.852$, which was not satisfactory. After regional focusing, by use of a discriminant power value of 0.5 and a grid spacing of 1.0 Å, q^2_{LOO} of 0.513, r^2_{ncv} of 0.825, q^2_{MC} 0.561, q^2_{BS} of 0.877, SEE_{BS} of 0.213 and r^2_{pred} of 0.554 were generated. For the CoMSIA model for steroids, the relative field contributions of parameters were 2.2%, 38.8%, 25.1%, 16.5%, and 17.3% for steric, electrostatic, hydrophobic, hydrogen bond donor, and hydrogen bond acceptor field, respectively. Electrostatic field was the most important descriptor in the CoMSIA model, but

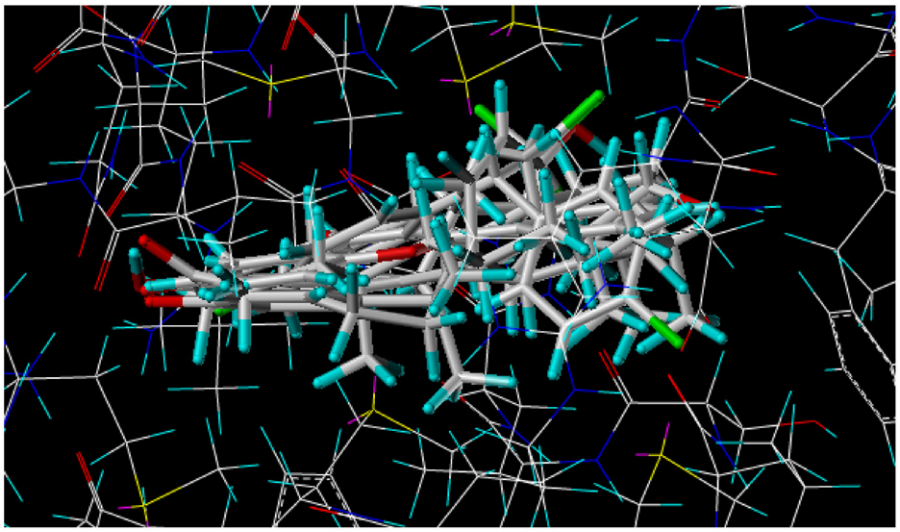

Fig. 3. Docking result of 6 key ligands in AR.

Table 3
Statistics and field contribution of CoMSIA models for the training and test sets.

Parameter	Steriods	Nonsteriodal chemicals
q^2_{LOO}	0.661	0.513
PLS components	6	3
r^2_{ncv}	0.970	0.825
SEE	0.285	0.210
F_{test}	95.362	53.310
Contribution		
Steric	2.2%	20.1%
Electrostatic	38.8%	19.3%
hydrophobic	25.1%	29.7%
H-bond donor	16.5%	12.6%
H-bond acceptor	17.3%	18.3%
r^2_{pred}	0.84	0.55

hydrophobic field also contributed significantly (Table 3). Volumes of these steroids are similar, which is why the contribution of the steric field parameter was small. In the CoMSIA model for non-steriodal chemicals, steric, electrostatic, hydrophobic, hydrogen bond donor, and hydrogen bond acceptor field contributed 20.1%, 19.3%, 29.7%, 12.6% and 18.3%, respectively. Hydrophobic field was the most important descriptor included in Table 3.

The predictive power of the two models were deemed to be acceptable ($q^2_{LOO} > 0.5$, $r^2_{pred} > 0.4$) (Wang et al., 2005; Golbraikh and Tropsha, 2002), thus further analysis based on these models would be appropriate. Comparison of the experimental versus predicted logRBA values for the steroidal and non-steroidal models for both training and test sets are given (Tables 1 and 2).

3.3. CoMSIA contour maps

In the CoMSIA steric field map (Fig. 4A), green represents sterically favorable regions and yellow represents sterically unfavorable regions contours. These two regions represent 80% and 20% level contributions to contour maps, respectively. To aid in the visualization, R1881 is overlaid on the map. Large green contours appearing around Phe876, Thr877 and Val746, Leu873 indicate sterically bulky groups on ligands are favored in these regions. The electrostatic field is the major contributor to the prediction of binding of steroids to the AR LBD. According to the graphic of CoMSIA hydrophobic field (Fig. 4B), represented by the yellow contour, which represents a hydrophobically favorable region near Leu701 and Leu704 indicates that hydrophobic groups are favored to bind to this region. As shown in Fig. 4C, the blue polyhedra indicate that a strong electropositive substituent around amino acids Gln711 and Met745 is important contributor to binding affinity. This might be due to the strong interaction between glycine/methionine and electropositive groups on the ligands. A cyan region represents a hydrogen bond donor favorable contour and a red region represents a hydrogen bond acceptor unfavorable contour in the hydrogen bond donor and receptor contour maps of the CoMSIA model were observed near Leu880, Phe876, and Thr877, respectively (Fig. 4D). This observation suggests that hydrogen bond donors near these amino acids will promote binding of chemicals to the AR LBD and a hydrogen bond acceptor will reduce binding. Chemicals with greater binding affinities form hydrogen a bond with Thr877. This result is consistent with those of previous studies (Sack et al., 2001). The cyan and magenta contours, representing hydrogen bond acceptor favorable regions near Val746, Met749 and Phe764 suggest that chemicals with a hydrogen bond donor or acceptor group at this region would be bound with greater affinity.

Contour maps of non-steroidal chemicals are shown in Fig. 5, and flavone was superimposed on them to be more viewable. In the CoMSIA steric field (Fig. 5A), the large yellow contour near Leu701, Leu704, Asn705 and Thr877 indicates that sterically bulky groups are unfavored in that region. According to the graphic of the CoMSIA hydrophobic field (Fig. 5B), a large white contour, represents a hydrophobically unfavorable region near Leu701, Leu873, Phe876 and Thr877. This indicates that hydrophobic groups on the ligand are unfavored in this region. In the contour map of the electrostatic field contributor to binding of chemicals to the AR LBD (Fig. 5C) the fragmentary blue polyhedron indicates that strong electropositive groups on the ligand would be favorable for binding to amino acid Met742. The importance of Met742 in binding of ligands to the AR LBD has been previously reported (Bohl et al., 2004). The hydrogen bond donor and acceptor contour map of the CoMSIA model is shown (Fig. 5D). One large and two small purple contours representing hydrogen bond donor unfavorable contours around Gln711 and Arg752 indicate that hydrogen bond donor moieties on ligands near these residues contributes negatively to affinity of binding. These residues are important in AR because the 3-keto group of natural ligands can a form hydrogen bond with them directly or

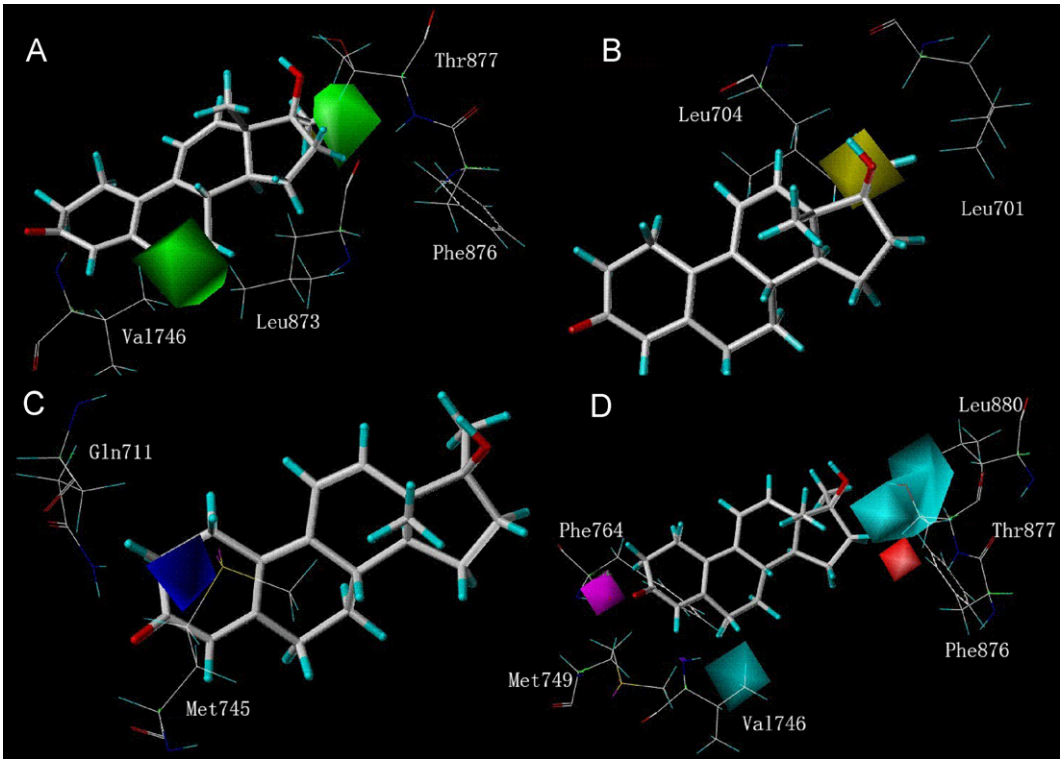

Fig. 4. Comparative molecular similarity indices analysis (CoMSIA) standard deviation/coefficients (stdev.coeff) contour map, the compound R1881 is displayed as a reference. (A)CoMSIA steric contour map; (B) CoMSIA hydrophobic contour map; (C) CoMSIA electrostatic contour maps; (D) CoMSIA hydrogen bond donor and hydrogen bond acceptor contour map.

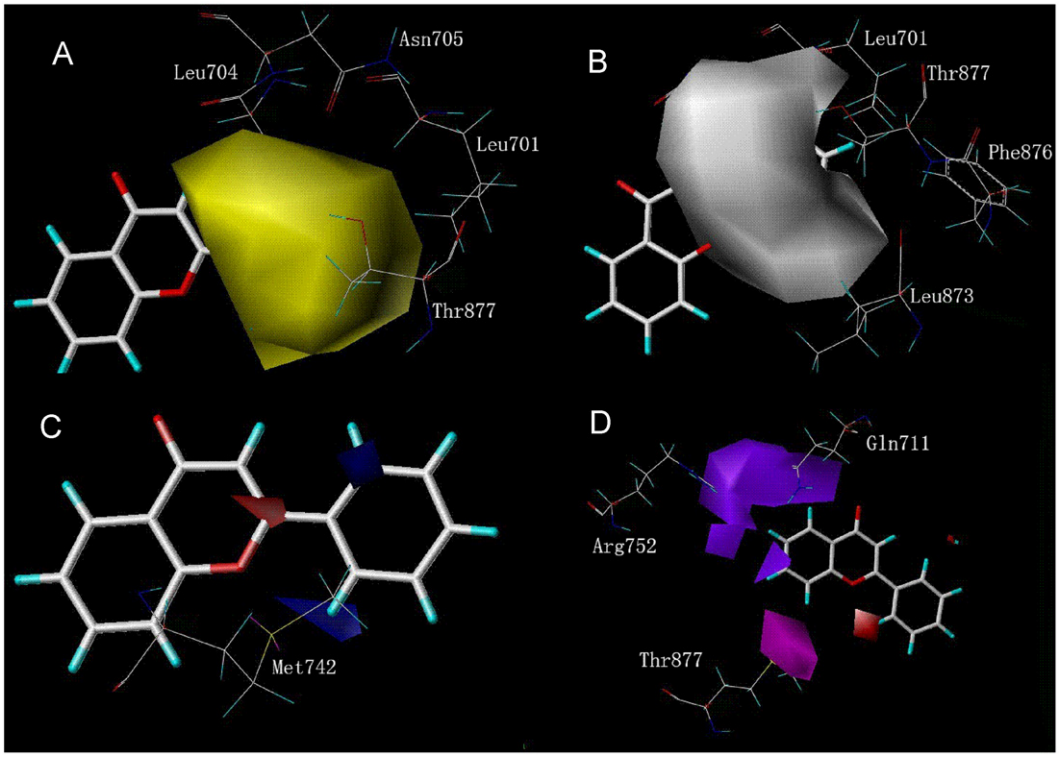

Fig. 5. Comparative molecular similarity indices analysis (CoMSIA) standard deviation/coefficients (stdev.coeff) contour map. The compound flavone is displayed as a reference. (A)CoMSIA steric contour map; (B) CoMSIA hydrophobic contour map; (C) CoMSIA electrostatic contour map; (D) CoMSIA hydrogen bond donor and hydrogen bond acceptor contour map.

indirectly as a hydrogen bond acceptor. The magenta contour, which represents an area favorable for hydrogen bonding is proximate to Met787. The area represented in red is a region favorable for hydrogen bonding, but that region is more distant from Met787 (4 Å versus 2 Å), a distance that can impair the effect for AR binding.

Table 4
Key amino-acid residue in AR for compounds.

interaction type	key residues	
	Steroids	Nonsteroidal chemicals
Steric	Val746, Leu873, Phe876, Thr877	Leu701, Leu704, Asn705, Thr877
Electrostatic	Gln711, Met745	Met742
Hydrophobic	Leu701, Leu704	Leu701, Leu873, Phe876, Thr877
H bond	Val746, Met749, Phe764, Leu880, Phe876, Thr877	Gln711, Arg752, Met787

Based on the results of the docking studies, most steroid hormones could form hydrogen bonds with either Gln711 or Arg752. The hydrogen bond donor near Gln711 and Arg752 are an unfavorable factor for binding of non-steroidal chemicals. Thr877 is significant in the steric field, including for hydrogen bonding of steroids and the hydrophobic field for binding of non-steroidal chemicals. Thr877 is also an important residue in the steric field of non-steroidal chemicals. Leu701 and Leu704 are present in the steric field of non-steroidal chemicals and the hydrophobic field of steroids. Amino acids Leu701, Leu704, Gln711, Arg752 and Thr877 were determined to be important for binding of all ligands to the AR LBD. The three amino acids, Leu701, Leu704 and Gln711 are part of the helix 3 region of the AR LBD. Arg752 and Thr877 belong to helix 5 and helix 11, respectively (Matias et al., 2000). These three helices are important for repositioning helix 12 of the AR LBD and stabilizing ligands of all types (Germain et al., 2006). Some amino acids such as Val746, Met749 and Phe764 are only important for steroids, while Met742 and Met787 were important for non-steroidal chemicals. The similar but slightly different docking orientation of chemicals might contribute to these observed phenomena. All residues involved in binding of steroids and non-steroids to AR are summarized (Table 4).

4. Conclusions

Understanding intermolecular interactions of steroidal chemicals and non-steroidal chemicals with AR was achieved by molecular docking and CoMSIA analysis. The most important interaction for binding of steroids to the AR LBD was electrostatic interaction and that for non-steroidal chemicals was hydrophobic interaction. Leu701, Leu704, Gln711, Arg752 and Thr877 were important for both kinds of chemicals. Val746, Met749, Phe764 were only important for steroids, while Met742, Met787 were important for non-steroidal chemicals. The results of this study will provide useful insights into prediction of agonists and antagonists of the AR and allo for screening of chemicals in silico and allow less potent alternatives to be designed.

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