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Development of QRAR/QSAR Method by Biopartioning Micellar Chromatography and Application in Prediction Toxicity of Bioactive Ingredients of Traditional Chinese Medicines

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The knowledge of drug potential toxicity is great need for risk assessment and screening candidates of drugs in the drugs development. The use of biopartitioning micellar chromatography has proven to be valid in predicting several biological activities of different kinds of drugs in development new drugs. The use of quantitative retention-activity relationship/quantitative structure-activity relationship basing on biopartitioning micellar chromatography to estimate acute toxicity is an attractive alternative to experimental measurements. In this paper, a data set of 58 chemical drugs from various structure classes with median lethal dose (LD_{50}) data available expressed as pLD_{50} in this paper was studied to construct acute toxicity model. The pLD_{50} was reciprocally correlated to the negative value of the capacity factor (-1/k). The correlation was better with the addition of molecular descriptors ($R^2 = 0.823$). The method of quantitative retention-activity relationship/quantitative structure-activity relationship developed by construction model of chemical drugs was applied to predict toxicity of bioactive ingredients of traditional chinese medicines. The results showed that application was predictable and practical in bioactive ingredients of traditional Chinese medicines.

Key Words: Biopartioning micellar chromatography, Quantitative structure-activity relationship, Quantitative retention-activity relationship, Toxicity, Traditional Chinese medicines.

INTRODUCTION

The primary goal of the drug discovery and development process is to find a molecule with both good pharmacodynamic and good pharmacokinetic properties. Ideally, a new drug should be efficacious and selective, target-tissue(s)-specific and orally-absorbed, cause minimal or no adverse effects due to metabolite activity or toxicity and be distributed / excreted in such a fashion as to permit dosage once a day. However, sub-optimal absorption, distribution, metabolism and excretion pharmacokinetic properties are the major reason for the high attrition rates of compounds in development, where more than 90 % of all candidates fail[1]. Similarly, toxicity play a considerable part role in candidates' failure.This problem has persisted due to difficulties in obtaining data on pharmacokinetic properties (ADME/T) early in drug discovery. Therefore, the ideal situation for the medicinal chemist is that the pharmacokinetic properties of a compound can be predicted on its physicochemical properties. Traditional methods *in vivo* and *in vitro* have been developed in order to predict pharmacokinetic properties. A drawback for most of these methods is that they are time consuming and have a limited throughput.

Most drugs are qualified but toxicity is barrier to go on to the market. Drugs *in vivo* must be transported across the biomembranes and then arrive at the site of action where it must accumulate certain concentration to produce biological response. It is known to all, when drugs are administered beyond effective dose, high bioconcertation of the drug in the action site can attribute to the acute toxicity to the tissues or organs because of easy transmembrane diffusion which usually lead to drug accumulate in the action site. Therefore, toxicity production processes of drug action are also considered to have much in common with the processes on which chromatographic separations are based. The molecular features (hydrophobicity, electrical charge and steric effects, degree of ionization, molecular shape, size, *etc.*) affect not only transport processes and drug-biological target interactions, but also the drug retention in a chromatographic system under specific experimental conditions.

Chromatographic models are universally used in this aspect due to experimental simplicity, low cost, accuracy and high throughput, among which the immobilized artificial membrane, immobilized-liposome chromatography, biopartitioning

micellar chromatography system are well recognized models. Biopartitioning micellar chromatography is a chromatographic modality optimized in order to describe the biological behaviour of drugs, which usually is comprised of a C_{18} reversed stationary phase and polyoxyethylene (23) lauryl ether (Brij35) mobile phase. It has been testified to be useful to predict HOA[2,3], rapid toxicity prediction of organic chemicals to *Chlorella vulgaris*, chemical toxicity[4], ecotoxicity[5] bioconcentration of pesticides in fish[6] and skin permeability[7,8].

The success of QRAR models based on biopartitioning micellar chromatography could be attributed to the similarities among biopartitioning micellar chromatography systems, biological barriers and extracellular fluids. This methodology has been applied for describing and predicting the biological activity of different pharmacological kinds of drugs, namely QSAR model when structure features are introduced. QSAR models describe a mathematical relationship between the structural features of a set of chemicals and the particular activity associated with them[9]. QRAR and QSAR models developed separately by biopartitioning micellar chromatography were studied more in screening bioactivity chemical drugs in drug development. QRAR was used to study on oral drug absorption and biological activity[10], HMG-CoA reductase inhibitors[11], quinolones[12], cephalosporins[13], angiotens inconverting enzyme inhibitors[14], angiotensin enzyme inhibitors[15], alkaloids by mixed micellar liquid[16], dihydropyridine selective calcium channel antagonist toxicity[17], quantitative retention-structure and retention-activity relationships of barbiturates[18] and local anesthetics[19]; QSAR studied on *para*-substituted aromatic sulfonamides as carbonic anhydrase II inhibitors using topological information indices[20], acute chemical toxicity for aquatic environment[21] *etc*. Moreover, the bioactive ingredients of traditional Chinese medicines have much common in retention behaviour on biopartitioning micellar chromatography column for their physical nature and toxicity mechanisms are analogous with chemical drugs. So prediction toxicity of traditional Chinese medicines can be studied by constructing QRAR and QSAR models using biopartitioning micellar chromatography.

But when biopartitioning micellar chromatography is used to predict toxicity of bioactive ingredients of traditional Chinese medicines, the retention may not be obtained accurately because of interference of other ineffective and unknown components. Therefore, in this paper, parts of monomers of bioactive ingredients of traditional Chinese medicines with available literatures about their LD_{50} were just preliminarily studied. Firstly, we constructed toxicity prediction QRAR/QSAR model of chemical drugs which was introduced to capacity factor (k) and molecular structure descriptors (describe molecular structure mathematically) of 58 selected chemical drugs basing on previously studied QRAR and QSAR models. Finally, application toxicity prediction model of chemical drugs on prediction toxicity monomers of bioactive ingredients of traditional Chinese medicines and then comparison predicted value with literatures reported value of their LD_{50} were conducted.

EXPERIMENTAL

The mobile phase consisted of 0.04 mol/L polyoxyethylene (23) lauryl ether (Brij35, Acros, NJ, USA)) with 0.01 mol/L sodium dihydrogen phosphate (analytical-reagent grade, Kelong, Chengdu, China) and was adjusted to pH 7.4 which is the plasmatic pH value by sodium hydroxide. In order to reproduce the osmotic pressure of biological fluids, sodium chloride (9.20 g/L, analytical-reagent grade, Kelong, Chengdu, China) was added to the micellar mobile phase. Sodium chloride concentration was close to physiological concentration of biological fluids. Water was from a Millipore (Billerica, MA, USA) synergy™ 185 system and was degassed before HPLC. The mobile phases injected into the chromatograph were filtered through 0.45 μm micro porous membrane.

Among the 58 chemical drugs, piroxicam, isoniazid, caffeine, meloxicam, furosemide, pindolol were used as control articles provided by analysis test center of West China School of Pharmacy and the others were crude drug or formulations which would not affect their retention behaviour in this study because of the dilution of the mobile phase, donated by the pharmaceutical and pharmaceutical chemistry laboratories of West China School of Pharmacy, Sichuan University (Chengdu, China). Reference substances of traditional Chinese medicines including chlorogenic acid, caffeic acid, cinnamic acid, rhein, aconitine, hypaconitine and sinomenine which were purchased from national institute for the control of pharmaceutical and biological products.

Water-soluble drugs were dissolved in mobile phase solution. Lipophilic drugs were first dissolved in methanol (analytical-reagent grade, Kelong, Chengdu, China) and then were diluted with water to get appropriate concentration. Stock standard solutions of reference substance and crude drug of analytes were prepared by dissolving 10 mg of the compound in 10 mL volumetric flask. Working solutions were prepared by dilution of the stock standard ones using the Brij35 solution. For those pharmaceutical preparations of analytes, working solutions were prepared by dissolving 10 mg of the tablet or capsule powders of the drugs in 10 mL volumetric flask, then centrifuged at 1000 × g for 5 min. The working solutions injected into the chromatograph were filtered through 0.45 μm microporous membranes (Xinya, Shanghai, China), respectively. All the solutions were stored under refrigeration at 4 ºC before analysis.

Instrumental and measurement: The retention of drugs was measured using an LC-6A chromatograph with an LC-6A pump, an SPD-6AV UV-visible detector and a CTO-6A column thermostat (Shimadzu, Japan). Data were collected and processed on a Compaq computer installed with HP-Chemstation software (A0402, 1996). The solutions were injected into the chromatograph through a Rheodyne valve (Cotati, CA, USA), with a 20 μL loop. The HPLC column was a Kromasil C_{18} column (5 μm, 150 × 4.6 mm i.d.) with a phenomenex security Guard ™C_{18} guard cartridge. The mobile phase flow rate was1.0 mL/min. The UV detection of chemical drugs was monitored at 220, 254, 270 and 300 nm and the detection wavelength of bioactive ingredients of traditional Chinese medicines was set at 270, 280 and 240.

All the assays were carried out at 37 ºC for simulating human body temperature. The retention data in biopartitioning micellar chromatography were calculated as capacity factors, $k = (t_r - t_0)/t_0$, where t_r is the retention time of the test compound and t_0 is the column dead time, which is the first fluctuation of baseline, determined by injecting water. The k values used in

this study were the average value of triplicate. The retention data were highly reproducible.

Statistical analysis: The data set was analyzed using Microsoft® Excel 2003 (Microsoft Corporation). MLR was used to carry out on the date set using SPSS 12.0 software (the SPSS for windows version 12.0, SPSS Inc. Chicago, USA). Stepwise regression analysis which is one of the MLR methods was used to determine the most significant descriptors. Molecular descriptors were calculated by Discovery Studio 2.5 (Accelrys Software Inc., San Diego, CA, USA). ChemDraw® Ultra 8.0.3 (Cambridgesoft corporation, USA)was used. For each regression, the following requirements of significant regression analysis were observed: n, R^2, SE, F and p, in which n is the number of points used in the regression, R^2 is the square of the overall correlation coefficient, SE is the standard deviation and F is Fischer's F-statistic, which are used to control fit ability and statistics significance of regression mode. T test is used to prove that partial regression coefficient before each variable is meaningful or not in MLR equation. VIF is determined the multicollinearities among the descriptors. VIF was calculated for each descriptor in the model as $1/(1- r^2)$, where r is a multiple correlation coefficient. Multicollinearities were considered to exist when the VIF was greater than 10 and the model was considered to reconstruct. In addition, root mean squared error of calibration (RMSEC) was estimated predictive ability of the QRAR/QSAR model.

$$RMSEC = \sqrt{\frac{\sum_{i=1}^{n}\left(\overline{Yi} - Yi\right)^2}{n}}$$

where, $\overline{Yi}$ is the predicted activity when all the n molecules are included in the model construction.

RESULTS AND DISCUSSION

Development of QRAR/QSAR method by biopartitioning micellar chromatography: As mentioned above, the molecular structure features of drugs determine not only their toxicity but also the biopartitioning micellar chromatography retention. Therefore, retention-toxicity relationship and structure-toxicity relationship could be expected. Accordingly, in this paper, the first step in current study was to calculate bulk properties and molecular descriptors (Table-1) of the selected chemical drugs which were passively absorbed in intestinal wall and to measure the retention (capacity factor k) of each drug on the biopartitioning micellar chromatography column as an indicator for the drug partitioning into cell membrane. The next step was to correlate calculated descriptors and experimentally measured capacity factor against pLD_{50}. Since an equation containing an excessive number of independent variables can be too cumbersome to use and is likely to be overparameterized, we utilized stepwise regression to refine the model and to select most important descriptors to generate regression equation. The sample set including 58 different chemical drugs with LD_{50} date (expresses as pLD_{50}) available was random divided into training set including 46 chemical drugs which was used to build model and test set 12 chemical drugs which was used to evaluate predictability of model. Table-1 shows the specific compounds, molecular descriptors, experimental pLD_{50} and predicted pLD_{50} calculated by eqn. (1).

Through the MLR method preformed for the training set, the better linear equation with four parameters is listed as follows:

$$pLD_{50} = -3.022\ (\pm 0.419) - 0.22\ (\pm 0.052)1/k_{BMC} - 0.012\ (\pm 0.002)\ \text{Molecular polar surface area} + 0.005\ (\pm 0.002)\ \text{Absolute energy} + 0.748\ (\pm 0.316)\text{IAC\_Mean}$$
$$N = 48;\ R^2 = 0.823;\ R^2\,adj = 0.806;\ SE = 0.29;\ F = 49.83;\ P < 0.001 \quad (1)$$

In the eqn. (1), pLD_{50} was reciprocally correlated the negative value of the biopartitioning micellar chromatography capacity factor (-1/k). Lipophilicity is one of the vital parameters commonly used to predict membrane permeability[22,23] and is approximately correlated to passive transport across cell membranes and the ability of a compound to partition a membrane[24], indicating by retention value measured by biopartitioning micellar chromatography. The large value of k reveal, the strong permeability, which suggest, high concentration of drug in action site accumulates easily and accompany with toxicity effect. The lipophilicity by itself is inadequate for the estimation of the solute's ability to penetrate a membrane barrier. Therefore, both hydrophobic effects and hydrogen bonding forces must be considered rather than just lipophilicity. PSA is a surface descriptor, defined as the part of the surface area of a molecule contributed by nitrogen, oxygen and connected hydrogen atoms. This descriptor can be loosely related to hydrogen-bonding capability[25]. Formation of hydrogen bonds could be linked with the toxic effect in some cases, as it facilitates the creation of the intermolecular interactions between the compound and the biological structure, but the negative contribution in eqn. (1) suggests that more likely here is that it characterizes the ability of a drug to penetrate through the biomembranes to/within the living organism. A higher value of this descriptor indicates lower penetration to cell membranes and small toxicity of a compound. Absolute energy is quantum chemical descriptors, which has large value suggesting that drug toxicity is relative high because molecule is easy to gain and lose electrons and consequently leading to high bioactivities. IAC_Mean which is one of topological structure descriptors is an average information index of atoms composed of molecule. It represents information of molecular shape and molecular size. In the regression equation, IAC_Mean is proportional to pLD_{50} but is against to toxicity. That is large value IAC_Mean indicting low toxicity.

As can be observed, the *p*-value obtained for toxicity model was less than five, which indicated that the relationships between these parameters and the pLD_{50} were statistically significant at the 95 % confidence level. The coefficients obtained for this model was also significant at the same confidence level. The standard error for the toxicity model can be used to construct prediction limits for new observations. The QRAR/QSAR model obtained by biopartitioning micellar chromatography was adequate to predict the toxicity of chemical drugs ($R^2 = 0.823$).

When multiple linear regression was used to construct model, each variable should perform t test. From the Table-3 listed, t value of each variable was large than the standard value of t ($t_{a/2} = 2.018$) for this model at 95 % confidence level. VIF calculated for each variable in this model was less than 5,

TABLE-1
MOLECULAR DESCRIPTORS, EXPERIMENTAL AND PREDICTED pLD_{50} AND RESIDUAL VALUES (EXPERIMENTAL pLD_{50} -PREDICTED pLD_{50}) OF CHEMICAL DRUGS

NO	Compound	$1/k_{BMC}$	MPSA[a]	AE[b]	IAcM[c]	pLD_{50} [d,e] Exp	pLD_{50} [f] Pre	RV[g]
Training set								
1	Amoxapine	0.03	36.85	82.67	1.61	-2.02	-1.85	-0.17
2	Amoxicillin	3.7	158.25	25.74	1.8	-4.4	-4.27	-0.13
3	Amrinone	0.24	68.01	25.45	1.63	-2.46	-2.55	0.09
4	AmLodipine	0.05	19.88	75.59	1.65	-1.57	-1.66	0.09
5	Orphenadrine	0.02	12.47	33.52	1.26	-2	-2.07	0.07
6	Alprenolol	0.05	41.49	26.28	1.34	-2.26	-2.4	0.14
7	Atenolol	1.96	84.57	28.16	1.5	-3.3	-3.21	-0.09
8	Atropine	0.24	49.77	32.93	1.41	-1.88	-2.45	0.57
9	Aciclovir	2.7	114.76	29.85	1.85	-4	-3.47	-0.53
10	Phenylbutazone	0.1	40.62	51.6	1.45	-2.38	-2.19	-0.19
11	Phenobarbital	0.08	75.26	28.97	1.66	-2.3	-2.56	0.26
12	Phenytoin sodium	0.05	58.19	48.36	1.55	-2.22	-2.33	0.11
13	Allopurinol	0.85	74.68	73.4	1.84	-2.25	-2.36	0.11
14	Piroxicam	0.12	107.98	59.17	1.85	-2.4	-2.66	0.26
15	Imipramine	0.02	60.48	54.31	1.21	-2.27	-2.58	0.31
16	Diazepam	0.03	32.67	54.93	1.59	-1.68	-1.96	0.28
17	Paracetamol	0.26	49.32	24.24	1.6	-2.53	-2.35	-0.18
18	Felodipine	0.06	64.62	41.62	1.69	-2.4	-2.34	-0.06
19	Haloperidol	0.02	40.53	36.78	1.57	-1.85	-2.15	0.3
20	Furosemide	0.14	131	32.62	2.04	-3.2	-2.94	-0.26
21	Mannitol	4.76	121.38	14.9	1.46	-4.34	-4.37	0.03
22	Ganciclovir	2.56	134.99	34.26	1.85	-3.3	-3.66	0.36
23	Trimethoprim	0.17	105.51	59.19	1.67	-2.8	-2.78	-0.02
24	Caffeine	0.47	58.44	27.68	1.78	-2.1	-2.36	0.26
25	Lamotrigine	0.08	90.7	52.27	1.84	-2.39	-2.49	0.1
26	Ranitidine	1.41	127.6	47.94	1.73	-2.94	-3.33	0.39
27	Ribavirin	3.57	143.71	29.53	1.87	-3.6	-4	0.4
28	Lidocaine	0.08	32.34	33.35	1.35	-2.34	-2.25	-0.09
29	Chlordiazepoxide	0.03	36.75	54.42	1.5	-2.3	-2.08	-0.22
30	Minoxidil	0.92	81.06	28.57	1.45	-3	-2.97	-0.03
31	Primidone	0.14	58.19	26.35	1.56	-2.45	-2.45	0
32	Tramadol	0.14	32.7	40.94	1.32	-2.43	-2.25	-0.18
33	Diclofenac	0.07	49.32	46.76	1.73	-1.98	-2.1	0.12
34	Diclofenac	0.02	75.26	20.05	1.56	-2.23	-2.66	0.43
35	Bromperidol	0.02	40.53	36.8	1.57	-2.24	-2.15	-0.09
36	Pindolol	0.29	57.28	94.15	1.47	-2.37	-2.2	-0.17
37	Indometacin	0.07	28.53	122.58	1.63	-1.07	-1.55	0.48
38	Terbutaline	1.53	72.71	24.97	1.46	-2.31	-3.02	0.71
39	Clobazam	0.04	60.01	64.04	1.67	-2.76	-2.18	-0.58
40	Oxprenolol	0.13	50.71	34.84	1.41	-2.72	-2.43	-0.29
41	Hexobarbital	0.05	69.96	25.86	1.6	-2.67	-2.55	-0.12
42	Urapidil	0.2	68.35	49.41	1.56	-2.71	-2.47	-0.24
43	Warfarin sodium	0.11	63.6	37.95	1.37	-2.57	-2.6	0.03
44	Aspirin	2.78	63.6	26.01	1.51	-3.4	-3.15	-0.25
45	Soniazid	1.52	68	20.45	1.74	-2.72	-2.77	0.05
46	Fluphenazine	1.79	55.25	60.81	1.46	-2.34	-2.69	0.35
47	Nadolol	1.69	81.95	39.3	1.41	-3.58	-3.13	-0.45
48	Benazepril	0.12	78.87	49.97	1.45	-2.8	-2.66	-0.14
Test set								
1	Aminophenazone	0.31	39.78	48.95	1.489	-2.54	-2.21	-0.33
2	Metformin	2.78	75.99	46.05	1.439	-3.16	-3.25	0.09
3	Digoxin	0.13	53.82	100.33	1.993	-1.25	-1.7	0.45
4	Theophylline	0.63	69.29	27.7	1.837	-2.37	-2.48	0.11
5	Ketoprofen	0.17	54.37	39.21	1.346	-2.56	-2.51	-0.05
6	Propranolol	0.23	41.49	33.04	1.366	-2.51	-2.38	-0.13
7	Naproxen	0.14	46.53	34.69	1.362	-2.56	-2.42	-0.14
8	Procaine	0.26	58.35	25.2	1.43	-2.72	-2.58	-0.14
9	Tetracycline	0.11	81.61	64.2	1.626	-2.83	-2.49	-0.34
10	Cimetidine	0.33	114.18	61.07	1.628	-3.41	-2.94	-0.47

[a]MPSA: Molecular polar surface area; [b]AE: Absolute energy; [c]IAcM: IAC_Mean; [d]$pLD_{50\ Exp}$: Experimental pLD_{50} (- $logLD_{50}$); [e]$pLD_{50\ Pre}$: Predicted pLD_{50} (- $logLD_{50}$); [f]http://www.drugfuture.com; [g]RV: Residual value (experimental pLD_{50}-predicted pLD_{50}).

TABLE-2
VIF VALUE, T VALUE OF THE DESCRIPTORS IN THE MODEL

Variables	$1/k_{BMC}$	Molecular polar surface area	Absolute energy	IAC_mean
VIF	1.995	3.042	1.228	1.790
t	4.301	5.305	-2.432	-2.365

suggesting that the model was robust because multicollinearities did not exist among variables.

In order to evaluate the predictive ability of this model in terms of RMSEC and residuals value (Table-1) were obtained. Applying equation 1 to predict pLD_{50} of the training set and test set, respectively. Figs. 1and 2 show the activities of predicted values *vs.* experimental values of training set and test set, respectively. As can be observed, the better correlation was obtained (training set, $R^2 = 0.8176$; test set, $R^2 = 0.8046$). Fig. 3 shows the corresponding residual plots. There was a random distribution of the residuals and practically they all were statistically small except several drugs due to other factors such as steric force and electrostatic effect may affect the accuracy prediction, which suggests, from a qualitative point of view, the adequacy of the model to data. At the same time, fit error of the model for the chemical drugs (training set, RMSEC = 7.76; test set, RMSEC = 5.06) were relative low which suggested the better predictive ability was obtained and application in predicting toxicity of traditional Chinese medicine was practical.

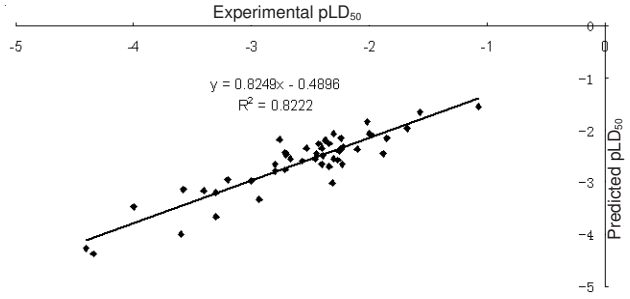

Fig. 1. Predicted value calculated by equation (1) *vs.* experimental value of training set of chemical drugs

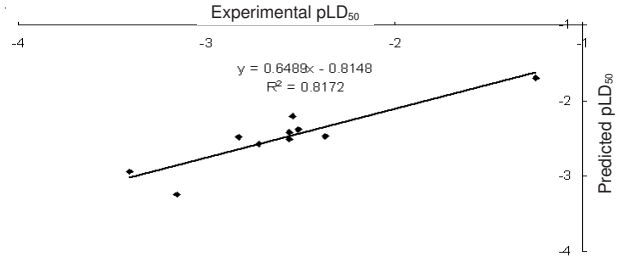

Fig. 2. Predicted value calculated by equation (1) *vs.* experimental value of test set of chemical drugs

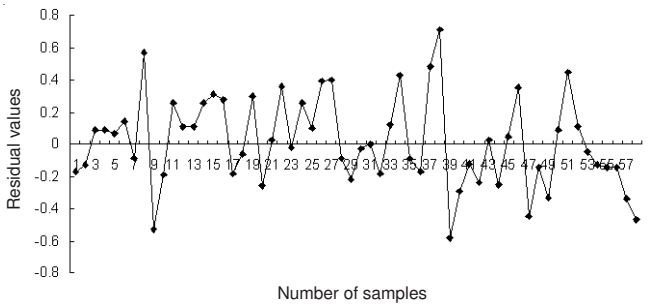

Fig. 3. The residual values [experimental OA (%)-predicted (%)] of all chemical drugs

Application QRAR/QSAR methods developed to predict toxicity of bioactive ingredients of traditional Chinese medicines by biopartitioning micellar chromatography: Applied toxicity prediction model of chemical drugs to predict toxicity of bioactive ingredients of traditional Chinese medicines. Table-3 shows the molecular descriptors of bioactive ingredients of traditional Chinese medicines, experimental pLD_{50} value and predicted pLD_{50} value.

Accurate LD_{50} values about bioactive ingredients of traditional Chinese medicines were not obtained, because researches about LD_{50} of bioactive ingredients of traditional Chinese medicines have not been studied. Table-3 showed the predicted values of pLD_{50} about bioactive ingredients of traditional Chinese medicines were relatively close to literature reported value except aconitine and hypaconitine. Many uncertain reasons contribute to the differences between predicted value and experimental value of aconitine and hypaconitine. One reason is the experimental conditions such as drug dose, time of administration, individual variance of subject, *etc*. Another possible reason is that predicted value was predicted by model of constructed chemical drugs. We hypothesized that the molecular descriptors of monomer of traditional Chinese medicines were same with chemical drug's in description toxic property. But structure descriptors of chemical drugs in toxicity aspect may not impact significantly on traditional Chinese medicines. As a result, existence of prediction error is inevitable.

Conclusion

In this study, prediction toxicity of diverse structural drugs was investigated by biopartitioning micellar chromatography technique. When the molecular structure descriptors were introduced, better correlation with toxicity (expressed as pLD_{50}) was obtained. Meanwhile, constructed model processes certain fit ability and statistics significance after statistics analysis. It is practical to apply this toxicity prediction model

TABLE-3
MOLECULAR DESCRIPTORS VALUES, THE EXPERIMENTAL pLD_{50}, PREDICTED pLD_{50} OF MONOMERS OF ACTIVE INGREDIENTS OF TCMs

Compound	$1/k_{BMC}$	MPSA[a]	AE[b]	IACM[c]	pLD_{50} [d] $_{Exp}$	pLD_{50} [e] $_{Pre}$
Ferulaic acid	0.21	106.76	23.48	1.483	-3.51[26]	-3.12
Hypaconitine	0.1	105.21	56.95	3.055	-0.76[27]	-1.74
Rhein	0.07	111.9	129.61	3.475	-0.70	-1.13
Tuduranine	0.65	109	94.16	1.453	-2.76[28]	-2.92
Cnnamic acid	0.16	137.29	31.59	1.378	-3.70[29]	-3.52
Aaconitine	0.08	109.51	157.19	3.469	-0.00[27]	-0.97

[a]MPSA: Molecular polar surface area; [b]AE: Absolute energy; [c]IAcM: IAC_mean; [d]$pLD_{50\ Exp}$: Experimental pLD_{50} (- $logLD_{50}$); [e]$pLD_{50\ Pre}$: Predicted pLD_{50} (- $logLD_{50}$)

of chemical drugs to predict toxicity of monomers of bioactive ingredients of traditional Chinese medicines because of robustness and reliability of this model.

It is known that the traditional Chinese medicines are complex mixtures containing many kinds of ingredients, effects on body are usually produced by these ingredients interaction with each other, few of which are responsible for their pharmaceutical and/or toxic effects. However, the screening and analysis of active/or safety ingredients in traditional Chinese medicines is very important not only for the quality control of crude drugs but also for elucidating the therapeutic principle of traditional Chinese medicines. In traditional, screening of toxicity ingredients is carried out on animals' models, which are time-consuming, arduous and inappropriate for directly discriminate toxic ingredients from traditional Chinese medicines. The method of construction of QSAR and QRAR models can be applied to discriminate toxicity drugs and predict toxicity degree according to retention on chromatography and calculate molecular descriptors for description molecular structure feature in toxicity through the mathematical statistics methods, such as multiple linear regression (MLR), principal component regression (PCA) and artificial neural network(ANN) *etc*.

Up to now, with the growth of computational chemistry, molecular structure and active relationships can be quantitative studied. And toxicity of active ingredients for certain species of traditional Chinese medicines can be tried to directly predict by construction QSAR model. In addition, retention of all kinds of monomers of active ingredients of traditional Chinese medicines on chromatography and correletionships with physicochemical properties such as partition coefficients (P) in the biphasic octanol-water solvent will be further studied in order to predict toxicity of traditional Chinese medicine compound preparation, hypothesizing that certain links between them can be find by construction mathematical models. Refinement and improvement of the predictive model is possible with the evaluation of different aspects of molecular descriptors and construction large date set as well as possible basing on available date of toxicity for traditional Chinese medicines.

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