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Comparative QSAR studies on PAMPA/modified PAMPA for high throughput profiling of drug absorption potential with respect to Caco-2 cells and human intestinal absorption

Rajeshwar P. Verma · Corwin Hansch · Cynthia D. Selassie

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Abstract Despite the dramatic increase in speed of synthesis and biological evaluation of new chemical entities, the number of compounds that survive the rigorous processes associated with drug development is low. Thus, an increased emphasis on thorough ADMET (absorption, distribution, metabolism, excretion and toxicity) studies based on in vitro and in silico approaches allows for early evaluation of new drugs in the development phase. Artificial membrane permeability measurements afford a high throughput, relatively low cost but labor intensive alternative for in vitro determination of drug absorption potential; parallel artificial membrane permeability assays have been extensively utilized to determine drug absorption potentials. The present study provides comparative QSAR analysis on PAMPA/modified PAMPA for high throughput profiling of drugs with respect to Caco-2 cells and human intestinal absorption.

Keywords Parallel artificial membrane permeability assays (PAMPA) · Bio-mimetic artificial membrane permeation assay (BAMPA) · PAMPA-BBB assay · Caco-2 cells · Human intestinal absorption · Hydrophobicity · Quantitative structure-activity relationship (QSAR)

R. P. Verma · C. Hansch · C. D. Selassie (✉)
Department of Chemistry, Pomona College, 645 North College Avenue, Claremont 91711 CA, USA
e-mail: cselassie@pomona.edu

Introduction

One of the most important challenges for pharmaceutical scientists is to develop a cost effective, high throughput and highly predictive model for the drug absorption that is able to guide design during the process of new drug optimization. Although the computational (in silico) approach for the prediction of oral absorption potential is very attractive, its accuracy is still not satisfactory [1, 2]. Thus, high throughput experimental methods for the determination of drug absorption potential continue to garner interest and wield importance in drug development. Two of the most popular in vitro absorption/permeability models used today involve Caco-2 cells and parallel artificial membrane permeability assays (PAMPA). Caco-2 cells are derived from a human colon carcinoma whereas PAMPA is a non-cell based lipid membrane filter. Caco-2 membrane permeability is widely used for in vitro drug absorption potential, but its use is limited due to the long membrane growth cycle as well as the high cost.

As a less expensive alternative to Caco-2 methods, Kansy et al. [3] developed PAMPA as a rapid, 96-well plate technology-based in vitro system for the evaluation of passive transcellular permeability. It involves the use of a filter based lipid membrane, which is completely artificial, lacking pores and active transporter systems [4]. Phosphatidylcholine (PC) has been used as a membrane constituent in the Kansy's PAMPA system. The determination of artificial membrane permeability values from this system requires two pH conditions (pH 7.4 and 5.5) to predict oral absorption. PAMPA studies are more facile, faster and much less expensive than those involving Caco-2 cells. The whole

PAMPA process is easily automated and commercially available [5]; it and variations of it are widely used as high throughput tools for the determination of in vitro drug absorption potential [6–11].

Recently, a bio-mimetic artificial membrane permeation assay (BAMPA) was introduced as an improved version of PAMPA; it utilized a similar lipid composition to intestinal brush border membrane and significantly increased the predictability of oral absorption [11, 12]. Bio-mimetic lipid (BML) membranes have been prepared by the utilization of phosphatidylcholine (PC, 0.8%)/phosphatidylethanolamine (PE, 0.8%)/phosphatidylserine (PS, 0.2%)/phosphatidylinositol (PI, 0.2%)/cholesterol (CHO, 1.0%)/1,7-octadiene (97.0%). BAMPA has been also used in connection with the paracellular pathway model based on the Renkin function (BAMPA-PP-RF model) resulting in the prediction of drugs absorption via both the passive transcellular as well as paracellular pathway [13]. A recent PAMPA (PAMPA-BBB) study using porcine brain lipids has been successfully utilized to improve the prediction of blood-brain barrier (BBB) penetration. The PAMPA-BBB assay is high throughput, accurate, low cost, reproducible and consumes a minimal amount of sample (< 0.5 mg) [14].

In the current study, we have derived various QSAR (quantitative structure-activity relationship) of drug absorption via PAMPA/modified PAMPA and compared them with those derived in Caco-2 cells as well as human intestinal systems. In the past 40 years, QSAR has been increasingly helpful in understanding chemical–biological interactions in drug, pesticide and toxicology areas [15] as it can shed light on mechanisms of action of various ligands with enzymes, membranes, organelles and cells [16, 17]. It has been widely utilized for the evaluation of adsorption, distribution, metabolism and excretion (ADME) phenomena in many organisms and whole animal studies. The QSAR approach employs extra-thermodynamically derived and computational-based descriptors to correlate biological activity in isolated receptors, cellular systems and in-vivo. The three standard classifications routinely used in QSAR analysis: electronic, hydrophobic and steric including topological indices are invaluable in helping to delineate a large number of receptor-ligand interactions that are critical to biological processes [16]. QSAR models can stand alone, augment other modeling approaches or be examined in tandem with equations of a similar mechanistic genre to establish authenticity and reliability [18].

Materials and methods

All the data for the absorption of drugs by different systems e.g. PAMPA, modified PAMPA, Caco-2 and human intestine have been collected from the literature (see individual QSAR for respective references). In the present study, drug absorption/permeability is represented by the following terms: F (permeation measured as percent flux by PAMPA), F_{ga} (glycocolic acid-aided permeation measured as percent flux by PAMPA), P_{app} (apparent permeability coefficient measured in cm/s by PAMPA or Caco-2 assay), P_{am} (artificial membrane permeability coefficient determined by BAMPA in cm/s), $P_{pp\text{-}rf}$ (permeability measured in cm/s by PAMPA-PP-RF), P_e (permeability measured in cm/s by PAMPA-BBB), Peff (human intestinal permeability measured in cm/s), and F_a (fraction of dose absorbed in humans). With the aid of the C-QSAR program, all physicochemical descriptors are auto-loaded, and multi regression analyses (MRA) is used to derive the appropriate QSAR [19]. For in-depth knowledge about the utility of the QSAR program in comparative correlation analysis, the interested reader is referred to earlier publications [20–22]. While comparing QSAR based on data from different laboratories and via diverse approaches, it must be borne in mind that variations in quality in testing will have an effect that cannot be estimated.

Parameters used in this paper have already been discussed in detail along with their applications [16]. Clog P is a calculated partition coefficient in 1-octanol/water system and is a measure of the overall hydrophobicity of a molecule [20]. The indicator variable I is assigned the value of 1 or 0 for the presence or absence respectively of certain structural features with unusual effects that cannot be parameterized and has been explained wherever it comes into play. The functional groups with hydrogen bonding capabilities include the –COOH, $–SO_2NH_2$, aromatic –OH and the $–N(CH_3)_2$ moieties.

In the derived models, n is the number of data points, r is the correlation coefficient, r^2 is the goodness of fit, s is the standard deviation, q^2 represents the goodness of prediction, calculated by the method of Cramer et al. [23], and the data within the parentheses represents the 95% confidence intervals. For a list of outliers in each data set, refer to the corresponding tables. All the QSAR reported here are derived by us and were not formulated by the original authors.

Results

QSAR for PAMPA

QSAR 1 and 2 were derived from the published data of Kansy et al. [24] for PAMPA flux of a large set of miscellaneous drugs with or without the addition of glycocolic acid.

Permeation of miscellaneous drugs measured as flux by PAMPA at pH 7.4 (Table 1) [24]

$$\begin{aligned}&\log F = 0.22(\pm 0.05)\mathrm{Clog}P - 0.31(\pm 0.10)(\beta \cdot 10^{\mathrm{Clog}P} + 1)\\&\quad - 0.60(\pm 0.14)\ I_1 - 0.57(\pm 0.18)\ I_2 + 1.36(\pm 0.08)\\&n = 94, \quad r^2 = 0.721, \quad q^2 = 0.682, \quad s = 0.206,\\&\quad \log \beta = -2.234 \quad \mathrm{Clog}\ P_{\mathrm{O}} = 2.61\end{aligned} \tag{1}$$

Permeation of miscellaneous drugs measured as flux by PAMPA at pH 7.4 and with the addition of 0.5% glycocolic acid (Table 1) [24]

$$\begin{aligned}&\log F_{\mathrm{ga}} = 0.20(\pm 0.05)\ \mathrm{Clog}P - 0.47(\pm 0.17)\\&(\beta \cdot 10^{\mathrm{Clog}\,P} + 1) - 0.47(\pm 0.16)\ I_1 - 0.45(\pm 0.23)\\&I_2 - 0.43(\pm 0.22)\ I_3 + 1.36(\pm 0.10)\\&n = 65, \quad r^2 = 0.721, \quad q^2 = 0.661, \quad s = 0.212,\\&\quad \log \beta = -3.053 \quad \mathrm{Clog}P_{\mathrm{O}} = 2.92\end{aligned} \tag{2}$$

According to QSAR 1, hydrophobicity of the drug molecules affects PAMPA flux in a bilinear fashion. Permeation first increases with an increase in hydrophobicity to an optimum hydrophobicity of 2.6 and then gradually decreases linearly. Likewise, with the addition of glycocolic acid (0.5%), PAMPA flux was found to be similarly well correlated with hydrophobicity in a bilinear fashion as represented by QSAR 2. I_1, I_2 and I_3 are indicator variables, which acquire a value of one in the presence of –COOH, $–SO_2NH_2$ and aromatic –OH moieties. The negative coefficients of the indicator variables, I_1, I_2 and I_3 indicate that drugs bearing these groups have diminished drug permeability at pH 7.4 with or without the addition of glycocolic acid. The addition of glycocolic acid helped enhance the solubility of lipophilic drugs but the observed permeabilities were only slightly altered. Thus QSARs 1 and 2 are found to be almost identical. It must be noted that –COOH, $–SO_2NH_2$ and aromatic –OH moieties can exhibit both hydrogen bond donor (HBD) and hydrogen bond acceptor (HBA) characteristics. A direct comparison between observed PAMPA flux values with and without the addition of glycocolic acid for a subset of 73 common drug molecules listed in Table 1, reveals the following significant correlation as seen in QSAR 3 and Fig. 1.

$$\begin{aligned}&\log F = 1.01(\pm 0.06)\ \log F_{\mathrm{ga}} - 0.03(\pm 0.09)\\&n = 73, \quad r^2 = 0.940, \quad q^2 = 0.937, \quad s = 0.104\end{aligned} \tag{3}$$

Using the data of Zhu et al. [25], the following QSAR were developed for the apparent permeability of a set of miscellaneous drugs determined by PAMPA at various pH.

Apparent permeability coefficient of miscellaneous drugs determined by PAMPA at pH 5.5 (Table 2) [25]

$$\begin{aligned}&\log P_{\mathrm{app}} = 0.55(\pm 0.11)\ \mathrm{Clog}\ P - 0.75(\pm 0.14)\\&(\beta \cdot 10^{\mathrm{Clog}\,P} + 1) - 0.77(\pm 0.36) I_2 - 0.12(\pm 0.19)\\&n = 60, \quad r^2 = 0.728, \quad q^2 = 0.690, \quad s = 0.376,\\&\quad \log \beta = -2.654 \quad \mathrm{Clog}P_{\mathrm{O}} = 3.08\end{aligned} \tag{4}$$

Apparent permeability coefficient of miscellaneous drugs determined by PAMPA at pH 7.4 (Table 2) [25]

$$\begin{aligned}&\log P_{\mathrm{app}} = 0.51(\pm 0.11)\ \mathrm{Clog}\ P - 0.66(\pm 0.14)\\&(\beta \cdot 10^{\mathrm{Clog}\,P} + 1) - 1.40(\pm 0.37)\ I_2 - 0.03(\pm 0.19)\\&n = 55, \quad r^2 = 0.743, \quad q^2 = 0.700, \quad s = 0.311,\\&\quad \log \beta = -2.487 \quad \mathrm{Clog}P_{\mathrm{O}} = 3.02\end{aligned} \tag{5}$$

QSAR 4 and 5 also reveal a bilinear dependence on hydrophobicity with an optimal log P value around 3.0. I_2 is an indicator variable that acquires a value of one in the presence of a $–SO_2NH_2$ group. The hydrogen bond related effects are more pronounced at the higher pH; the negative coefficient with this term reveals the detrimental nature of a strongly hydrogen bond-donor entity to drug permeability. The pH plays a significant role; it impacts the magnitude of the coefficients of the indicator variable that increase as the pH increases from 5.5 to 7.4.

QSAR for BAMPA (an improved version of PAMPA)

QSAR 6–8 were derived from the published data of Sugano et al. [26] for the bio-mimetic artificial membrane permeability assay (BAMPA) of a diverse set of

Table 1 Permeation of miscellaneous drugs measured as flux by PAMPA with or without the addition of glycocolic acid at pH 7.4 [24]

No.	Drugs name	log F (Eq. (1))			log F_{ga} (Eq. (2))			Clog P	I_1	I_2	I_3
		obsd	calcd	Δ	obsd	calcd	Δ				
1[a]	Acetaminophen	1.01	1.46	–0.45	1.03	1.02	0.01	0.49	0	0	1
2	Acyclovir	0.94	0.84	0.10	0.99	0.88	0.11	–2.42	0	0	0
3	Alprazolam	1.87	1.76	0.11	1.93	1.80	0.12	2.55	0	0	0
4	Alprenolol	1.81	1.76	0.05	–	–	–	2.65	0	0	0
5	Amitriptyline	1.72	1.60	0.12	–	–	–	4.85	0	0	0
6	Antipyrine	1.42	1.40	0.02	1.61	1.40	0.21	0.20	0	0	0
7	Aspirin	0.75	0.97	–0.22	–	–	–	1.02	1	0	0
8	Atenolol	1.04	1.34	–0.30	–	–	–	–0.11	0	0	0
9	Atropine	1.57	1.62	–0.06	–	–	–	1.30	0	0	0
10	Betamethasone	1.84	1.70	0.13	1.85	1.70	0.15	1.79	0	0	0
11	Caffeine	1.63	1.35	0.28	1.64	1.35	0.29	–0.04	0	0	0
12	Carbamazepine	1.86	1.75	0.10	1.93	1.79	0.14	2.38	0	0	0
13	Cefixime	0.70	0.81	–0.11	0.76	0.93	–0.17	0.25	1	0	0
14[a]	Cefoxitin	1.10	0.58	0.52	–	–	–	–0.81	1	0	0
15	Ceftriaxone	0.74	0.76	–0.02	–	–	–	0.02	1	0	0
16	Chlorambucil	1.29	1.11	0.18	1.54	1.28	0.26	3.63	1	0	0
17	Chloramphenicol	1.83	1.62	0.21	–	–	–	1.28	0	0	0
18	Chloroquine	1.67	1.58	0.08	–	–	–	5.06	0	0	0
19	Chlorpropamide	1.65	1.75	–0.10	–	–	–	2.35	0	0	0
20	Chlorprothixene	1.26	1.55	–0.28	1.07	1.31	–0.24	5.48	0	0	0
21	Chorthalidone	0.79	0.89	–0.09	–	–	–	0.45	0	1	0
22[a]	Cimetidine	0.70	1.44	–0.74	–	–	–	0.38	0	0	0
23	Clonazepam	1.74	1.76	–0.01	1.74	1.79	–0.05	2.38	0	0	0
24	Cocaine	1.93	1.76	0.17	–	–	–	2.57	0	0	0
25	Codeine	1.67	1.56	0.11	–	–	–	0.98	0	0	0
26	Corticosterone	1.97	1.76	0.21	1.96	1.80	0.16	2.51	0	0	0
27	Coumarin	1.95	1.64	0.31	1.96	1.63	0.33	1.41	0	0	0
28	Dapsone	1.73	1.54	0.19	1.74	1.53	0.21	0.89	0	0	0
29	Diazepam	1.68	1.75	–0.07	1.68	1.82	–0.14	2.96	0	0	0
30[a,b]	Diflunisal	1.59	1.04	0.55	1.38	0.68	0.70	4.40	1	0	1
31	Dihydrocodeine	1.58	1.62	–0.03	–	–	–	1.26	0	0	0
32	Diltiazem	1.88	1.71	0.17	1.90	1.75	0.15	3.65	0	0	0
33[a]	Doxorubicin	0.76	1.43	–0.67	0.81	0.99	–0.18	0.32	0	0	1
34	Ethinylestradiol	1.11	1.69	–0.57	–	–	–	3.86	0	0	1
35	Etilefrine	1.13	1.45	–0.32	–	–	–	0.44	0	0	1
36	Etofylline	0.97	1.17	–0.20	1.05	1.19	–0.14	–0.87	0	0	0
37	Famotidine	0.83	0.67	0.16	1.03	0.79	0.24	–0.58	0	1	0
38	Felbamate	1.66	1.46	0.19	–	–	–	0.50	0	0	0
39	Fluocortolone	1.80	1.76	0.04	1.84	1.81	0.03	2.61	0	0	0
40	Furosemide	0.85	0.55	0.30	0.75	0.79	–0.04	1.90	1	1	0
41	Griseofulvin	1.79	1.73	0.06	1.77	1.74	0.03	2.05	0	0	0
42	Hydrochlorothiazide	0.70	0.71	–0.01	0.77	0.83	–0.06	–0.36	0	1	0
43	Hydrocortisone	1.96	1.72	0.24	1.98	1.72	0.26	1.89	0	0	0
44	Hydroflumethiazide	0.72	0.75	–0.03	0.73	0.86	–0.13	–0.21	0	1	0
45	Imipramine	1.82	1.59	0.24	1.73	1.42	–0.31	5.04	0	0	0
46[a,b]	Indomethacin	1.85	1.06	0.79	1.79	1.17	0.62	4.18	1	0	0
47	Isoproterenol	1.08	1.39	–0.32	–	–	–	0.15	0	0	1
48[a]	Ketoprofen	1.58	1.16	0.42	1.50	1.34	0.16	2.76	1	0	0
49	Lidocaine	1.85	1.72	0.12	–	–	–	1.95	0	0	0
50	Lorazepam	1.68	1.75	–0.08	1.65	1.79	–0.14	2.37	0	0	0
51[a]	Mebendazole	0.78	1.75	–0.97	–	–	–	3.08	0	0	0
52	Metergoline	1.48	1.62	–0.14	–	–	–	4.69	0	0	0
53	Methylprednisolone	1.86	1.70	0.16	1.84	1.69	0.15	1.74	0	0	0
54	Methysergide	1.63	1.75	–0.11	1.54	1.77	–0.23	2.22	0	0	0
55	Metoclopramide	1.71	1.75	–0.04	1.66	1.77	–0.11	2.23	0	0	0
56[a]	Metolazone	1.85	1.17	0.68	–	–	–	2.06	0	1	0
57	Metoprolol	1.54	1.66	–0.11	–	–	–	1.49	0	0	0
58	Metronidazole	1.16	1.26	–0.10	–	–	–	–0.46	0	0	0
59	Midazolam	1.60	1.72	–0.12	1.56	1.79	–0.23	3.42	0	0	0
60	Molsidomine	1.15	1.63	–0.48	1.24	1.62	–0.38	1.33	0	0	0

Table 1 Continued

No.	Drugs name	log F (Eq. (1))			log F_{ga} (Eq. (2))			Clog P	I_1	I_2	I_3
		obsd	calcd	Δ	obsd	calcd	Δ				
61	Morphine	1.39	1.48	–0.08	–	–	–	0.57	0	0	1
62	Naloxone	1.74	1.39	0.35	–	–	–	0.16	0	0	1
63[a,b]	Naproxen	1.95	1.15	0.79	1.86	1.35	0.51	2.82	1	0	0
64	Nitrazepam	1.77	1.75	0.02	1.84	1.78	0.06	2.32	0	0	0
65[b]	Nitrendipine	1.01	1.70	–0.69	1.08	1.74	–0.66	3.73	0	0	0
66	Nitrofurantoin	1.45	1.26	0.19	1.62	1.26	0.36	–0.47	0	0	0
67	Nordazepam	1.54	1.75	–0.21	1.55	1.82	–0.27	3.02	0	0	0
68	Norfloxacin	0.91	0.59	0.32	0.75	0.73	0.02	–0.78	1	0	0
69	Omeprazole	1.66	1.76	–0.10	–	–	–	2.57	0	0	0
70	Oxazepam	1.67	1.75	–0.08	1.72	1.78	–0.06	2.31	0	0	0
71	Oxprenolol-HCl	1.65	1.74	–0.09	–	–	–	2.09	0	0	0
72	Papaverine	1.79	1.70	0.09	1.49	1.73	–0.24	3.78	0	0	0
73[a,b]	Pentamidine	0.70	1.75	–1.05	0.70	1.78	–1.08	2.31	0	0	0
74	Pentoxifylline	1.44	1.38	0.06	1.47	1.38	0.09	0.12	0	0	0
75	Phenobarbital	1.88	1.64	0.24	–	–	–	1.37	0	0	0
76	Phenylbutazone	1.73	1.73	0.00	1.90	1.79	0.11	3.39	0	0	0
77	Phenytoin	1.61	1.74	–0.12	–	–	–	2.09	0	0	0
78	Pindolol	1.66	1.69	–0.03	1.76	1.68	0.08	1.67	0	0	0
79[a]	Practolol	0.98	1.52	–0.53	1.02	1.50	–0.48	0.75	0	0	0
80	Prednisolone	1.90	1.65	0.26	1.92	1.63	0.29	1.42	0	0	0
81	Prednisone	1.88	1.69	0.19	1.89	1.68	0.21	1.66	0	0	0
82	Primidone	1.51	1.54	–0.03	–	–	–	0.88	0	0	0
83	Probenecid	1.16	1.13	0.03	1.35	1.32	0.03	3.37	1	0	0
84	Procainamide	1.45	1.65	–0.19	1.46	1.63	–0.17	1.42	0	0	0
85	Promethazine	1.40	1.64	–0.24	1.31	1.59	–0.28	4.40	0	0	0
86	Propoxyphene	1.90	1.57	0.33	–	–	–	5.21	0	0	0
87	Propranolol	1.82	1.76	0.06	–	–	–	2.75	0	0	0
88	Propylthiouracil	1.37	1.56	–0.19	1.44	1.55	–0.11	0.97	0	0	0
89	Proquazone	1.78	1.71	0.07	1.76	1.75	0.01	3.65	0	0	0
90	Proxyphylline	1.04	1.24	–0.20	1.05	1.25	–0.20	–0.56	0	0	0
91	Pyrimethamine	1.79	1.75	0.04	1.84	1.82	0.02	3.00	0	0	0
92	Quinidine	1.94	1.76	0.18	–	–	–	2.79	0	0	0
93[a]	Ranitidine	0.86	1.50	–0.64	0.95	1.49	–0.54	0.67	0	0	0
94	Rifampin	1.74	1.70	0.04	1.49	1.31	0.18	3.71	0	0	1
95[a]	Saccharin	0.70	1.51	–0.81	–	–	–	0.72	0	0	0
96	Salicylic acid	0.70	1.14	–0.44	–	–	–	2.19	1	0	1
97	Scopolamine	1.42	1.42	0.00	–	–	–	0.29	0	0	0
98[a,b]	Sulfadiazine	0.75	1.38	–0.63	0.99	1.38	–0.39	0.10	0	0	0
99[a,b]	Sulfinpyrazone	0.70	1.68	–0.99	0.70	1.68	–0.98	1.66	0	0	0
100[a,b]	Sulfisoxazole	0.70	1.41	–0.71	0.70	1.40	–0.70	0.22	0	0	0
101	Sulindac	1.15	1.14	0.01	1.18	1.34	–0.16	3.16	1	0	0
102	Sulpiride	0.70	1.02	–0.32	–	–	–	1.11	0	1	0
103	Suprofen	1.12	1.16	–0.04	1.24	1.33	–0.09	2.54	1	0	0
104	Tacrine	1.73	1.73	0.00	1.61	1.80	–0.19	3.27	0	0	0
105[a]	Terbutaline	0.94	1.46	–0.52	1.00	1.02	–0.02	0.48	0	0	1
106	Testosterone	1.90	1.72	0.17	1.90	1.79	0.11	3.41	0	0	0
107[b]	Tetracycline	1.26	1.17	0.09	1.24	0.75	0.49	–0.90	0	0	1
108	Theophyllin	1.09	1.35	–0.26	1.09	1.35	–0.26	–0.03	0	0	0
109	Trimethoprim	1.64	1.56	0.07	1.75	1.55	0.20	0.98	0	0	0
110	Verapamil	1.83	1.64	0.19	1.78	1.57	0.21	4.47	0	0	0
111	Warfarin	1.85	1.75	0.09	1.85	1.82	0.03	2.90	0	0	0
112	Zolpidem	1.83	1.75	0.08	1.79	1.82	–0.03	3.03	0	0	0

[a] Data points not included in deriving Eq. (1)

[b] Data points not included in deriving Eq. (2)

miscellaneous drugs at different pH conditions such as pH 5.5, 6.5 and 7.4, respectively. In the BAMPA method, Kansy's membrane was replaced by a bio-mimetic lipid (BML) membrane. In the case of BML membrane, the typical intestine pH condition is pH 6.5 for the predictability of oral absorption [11].

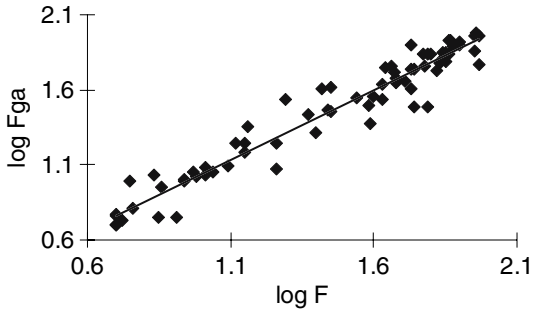

Fig. 1 Comparison of observed PAMPA flux with and without the addition of glycocolic acid (0.5%) at pH 7.4

Artificial membrane permeability of miscellaneous drugs byBAMPA at pH 5.5 (Table 3) [26]

$$\log P_{am} = 0.48(\pm 0.13)\ \mathrm{Clog}P - 0.44(\pm 0.41)\ I_1 + 1.54(\pm 0.59)\ I_4 - 0.14(\pm 0.26) \quad (6)$$
$$n = 24, \quad r^2 = 0.764, \quad q^2 = 0.672, \quad s = 0.400$$

Artificial membrane permeability of miscellaneous drugs by BAMPA at pH 6.5 (Table 3) [26]

$$\log P_{am} = 0.50(\pm 0.13)\ \mathrm{Clog}P - 0.78(\pm 0.37)\ I_1 + 1.47(\pm 0.55)\ I_4 - 0.03(\pm 0.24) \quad (7)$$
$$n = 25, \quad r^2 = 0.802, \quad q^2 = 0.711, \quad s = 0.375$$

Artificial membrane permeability of miscellaneous drugs by BAMPA at pH 7.4 (Table 3) [26]

$$\log P_{am} = 0.52(\pm 0.11)\ \mathrm{Clog}P - 1.28(\pm 0.32) I_1 + 1.12(\pm 0.48) I_4 + 0.04(\pm 0.22) \quad (8)$$
$$n = 24, \quad r^2 = 0.890, \quad q^2 = 0.832, \quad s = 0.314$$

QSAR 6–8, establish the critical importance of hydrophobicity in artificial membrane permeability of a diverse set of miscellaneous drugs. QSAR 6–8 reveal a strong linear dependence on hydrophobicity. The lack of bilinearity in these QSAR may reflect the

Table 2 Apparent permeability coefficient of miscellaneous drugs determined by PAMPA at pH 5.5 and pH 7.4 [25]

No.	Drugs name	log P_{app} (pH 5.5) (eq. (4))			log P_{app} (pH 7.4) (eq. (5))			Clog P	I_2
		obsd	calcd	Δ	obsd	calcd	Δ		
1[a]	Acebutolol	–0.70	0.78	–1.48	0.52	0.79	–0.27	1.71	0
2	Acetaminophen	0.36	0.15	0.21	0.54	0.22	0.32	0.49	0
3	Acetylsalicylic acid	0.51	0.43	0.08	0.58	0.48	0.10	1.02	0
4[a]	Alprenolol	0.15	1.11	–0.96	1.18	1.06	0.12	2.65	0
5[a,b]	Antipyrine	1.30	–0.01	1.29	1.12	0.08	1.04	0.20	0
6	Atenolol	–1.00	–0.18	–0.82	–	–	–	–0.11	0
7	Bromocriptine	0.11	0.53	–0.42	–	–	–	6.58	0
8	Bumetanide	0.66	0.36	0.30	–0.52	–0.34	–0.18	3.37	1
9	Bupropion	1.68	1.14	0.54	1.15	1.08	0.07	3.21	0
10[a,b]	Caffeine	1.31	–0.14	1.46	1.03	–0.05	1.08	–0.04	0
11[b]	Captopril	0.64	0.36	0.28	1.28	0.42	0.86	0.89	0
12	Carbamazepine	1.08	1.05	0.03	1.05	1.01	0.04	2.38	0
13	Ceftriaxone	–1.00	–0.11	–0.89	–	–	–	0.02	0
14	Chloramphenicol	0.83	0.57	0.26	0.23	0.61	–0.38	1.28	0
15[b]	Chlorothiazide	–0.70	–1.05	0.35	0.11	–1.58	–1.69	–0.29	1
16	Chlorpromazine	1.07	0.79	0.28	0.60	0.81	–0.21	5.30	0
17[a,b]	Clofibrate	–0.40	1.10	–1.50	–0.52	1.04	–1.56	3.68	0
18	Clonidine	1.30	0.64	0.66	1.15	0.67	0.48	1.43	0
19	Clozapine	1.35	1.09	0.26	1.45	1.03	0.42	3.71	0
20	Corticosterone	1.59	1.08	0.51	1.34	1.04	0.30	2.51	0
21	Coumarin	1.36	0.64	0.72	1.34	0.67	0.67	1.41	0
22	Cyclosporine	–1.00	–1.05	0.05	–0.52	–0.54	0.02	14.36	0
23	Desipramine	0.97	0.96	0.01	1.16	0.93	0.23	4.47	0
24	Dexamethasone	0.83	0.82	0.01	0.91	0.83	0.08	1.79	0
25	Diclofenac	1.03	0.91	0.12	1.10	0.90	0.20	4.73	0
26	Diltiazem	1.03	1.10	–0.07	1.27	1.04	0.23	3.65	0
27	Doxorubicin	–0.52	0.05	–0.57	–0.30	0.13	–0.43	0.32	0
28[b]	Enalapril	0.53	0.24	0.29	–1.00	0.31	–1.31	0.67	0
29[a,b]	Erythromycin	–1.00	0.74	–1.74	–1.00	0.75	–1.75	1.61	0
30	Etoposide	–0.15	–0.10	–0.05	–0.40	–0.01	–0.39	0.03	0
31	Flumazenil	0.68	0.57	0.11	0.78	0.61	0.17	1.29	0
32	Fluoxetine	0.87	0.94	–0.07	1.15	0.92	0.23	4.57	0
33	Furosemide	–0.22	0.10	–0.32	–0.22	–0.53	0.31	1.90	1
34	Gabapentin	0.08	–0.48	0.56	0.08	–0.36	0.44	–0.66	0

Table 2 Continued

No.	Drugs name	log P_{app} (pH 5.5) (eq. (4))			log P_{app} (pH 7.4) (eq. (5))			Clog P	I_2
		obsd	calcd	Δ	obsd	calcd	Δ		
35	Griseofulvin	0.89	0.93	–0.04	0.72	0.92	–0.20	2.05	0
36[a]	Guanabenz	0.20	1.14	–0.94	1.24	1.08	0.16	2.98	0
37	Hydrochlorothiazide	–1.00	–1.09	0.09	–	–	–	–0.36	1
38	Hydrocortisone	0.49	0.86	–0.37	0.53	0.86	–0.33	1.89	0
39	Ibuprofen	1.03	1.10	–0.07	0.83	1.04	–0.21	3.68	0
40	Imipramine	1.11	0.85	0.26	0.92	0.85	0.07	5.04	0
41	Indomethacin	0.80	1.01	–0.21	0.38	0.97	–0.59	4.18	0
42[b]	Ketoconazole	0.52	1.10	–0.58	0.08	1.04	–0.96	3.64	0
43	Ketoprofen	1.28	1.12	0.16	1.22	1.07	0.15	2.76	0
44	Ketorolac	0.71	0.74	–0.03	0.15	0.76	–0.61	1.62	0
45[a]	Labetalol	–1.00	1.08	–2.08	0.65	1.04	–0.39	2.50	0
46[a,b]	Melphalan	1.01	–0.23	1.24	0.76	–0.13	0.89	–0.21	0
47	Methotrexate	–0.70	–0.41	–0.29	–1.00	–0.29	–0.71	–0.53	0
48	Methylprednisolone	0.41	0.80	–0.39	0.77	0.81	–0.04	1.74	0
49	Metoprolol	0.08	0.67	–0.59	0.54	0.70	–0.16	1.49	0
50[a]	Miconazole	–0.15	0.69	–0.84	–	–	–	5.81	0
51	Naproxen	1.36	1.13	0.23	1.03	1.07	–0.04	2.82	0
52[a,b]	Nicotine	1.17	0.36	0.81	1.33	0.41	0.92	0.88	0
53	Norfloxacine	–0.30	–0.55	0.25	–0.05	–0.42	0.37	–0.78	0
54[a,b]	Penicillin-V	0.20	0.89	–0.69	–1.00	0.89	–1.89	1.94	0
55	Phenytoin	0.88	0.94	–0.06	0.71	0.93	–0.22	2.09	0
56	Pindolol	1.12	0.76	0.36	0.69	0.78	–0.09	1.67	0
57	Piroxicam	0.92	0.86	0.06	0.91	0.87	0.04	1.89	0
58	Prazosin	0.40	0.92	–0.52	1.13	0.91	0.22	2.03	0
59	Prednisolon	0.34	0.64	–0.30	0.76	0.67	0.09	1.42	0
60[b]	Probenecid	0.60	1.13	–0.53	0.38	1.07	–0.69	3.37	0
61[a]	Progesteron	–0.10	1.05	–1.15	0.60	1.00	–0.40	3.97	0
62	Propranolol	1.23	1.12	0.11	1.37	1.07	0.30	2.75	0
63	Quinidine	0.78	1.13	–0.35	1.04	1.07	–0.03	2.79	0
64	Saccharin	0.85	0.27	0.58	–	–	–	0.72	0
65	Salicylic acid	1.33	0.98	0.35	0.52	0.96	–0.44	2.19	0
66	Sotalol	0.46	0.00	0.46	0.04	0.09	–0.05	0.23	0
67[a,b]	Sulfasalazine	–0.52	1.07	–1.59	–1.00	1.01	–2.01	3.88	0
68	Sulpiride	–0.70	–0.29	–0.41	–1.00	–0.88	–0.12	1.11	1
69[a]	Terazosine	0.23	0.98	–0.75	0.94	0.96	–0.02	2.18	0
70	Timolol	0.23	0.53	–0.30	0.71	0.57	0.14	1.21	0
71	Trimethoprim	0.43	0.41	0.02	0.70	0.46	0.24	0.98	0
72	Verapamil	0.99	0.96	0.03	0.87	0.93	–0.06	4.47	0
73	Warfarin	1.02	1.14	–0.12	1.09	1.08	0.01	2.90	0
74[b]	Zidovudine (AZT)	–0.22	–0.10	–0.12	0.69	–0.01	0.70	0.04	0
75	Zopiclone	0.51	0.55	–0.04	0.95	0.59	0.36	1.25	0

[a] Data points not included in deriving Eq. (4)

[b] Data points not included in deriving Eq. (5)

limited range in hydrophobicity of these test sets with only two drugs with hydrophobicities > 3.0. The indicator variable I_4 takes a value of one for the presence of $-N(CH_3)_2$ group, which appears to be conducive to permeability, particularly at the more acidic pH(5.5). I_1 is the –COOH indicator variable whose negative coefficient in QSAR 6–8, indicate that the –COOH moiety is detrimental to permeability. These QSAR at different pH milieus highlight the conflicting roles played by the acidic and basic groups in terms of their negative and positive contributions, to permeability, respectively. As the acidity increases, the $-N(CH_3)_2$ group with its HBA characteristics markedly enhances permeability and the –COOH group's negative influence is also not as marked as it is at higher pH. The BAMPA results are highly sensitive to pH changes as can be seen by remarkable differences in the observed permeabilities. The intercepts of QSAR 6–8 increase as the pH increases. In a direct comparison between the observed artificial membrane permeability coefficient measured by BAMPA at pH 6.5 and pH 7.4 for a subset of 26 common drug molecules listed in Table 3, the following results are obtained. See QSAR 9, and Fig. 2.

Table 3 Artificial membrane permeability coefficient of miscellaneous drugs determined by BAMPA at pH 5.5, pH 6.5 and pH 7.4 [26]

No.	Drugs name	log P_{am} (pH 5.5) (eq. (6))			log P_{am} (pH 6.5) (eq. (7))			log P_{am} (pH 7.4) (eq. (8))]			Clog P	I_1	I_4
		obsd	calcd	Δ	obsd	calcd	Δ	obsd	calcd	Δ			
1[a]	Acebutolol	–0.07	0.67	–0.74	0.57	0.81	–0.24	0.59	0.93	–0.34	1.71	0	0
2	Acyclovir	–1.40	–1.29	–0.11	–1.05	–1.23	0.18	–1.30	–1.22	–0.08	–2.42	0	0
3	Amiloride	–0.08	–0.09	0.01	–0.17	0.02	–0.19	–0.38	0.10	–0.48	0.11	0	0
4	Ceftriaxone	–1.10	–0.57	–0.53	–0.77	–0.80	0.03	–1.52	–1.23	–0.29	0.02	1	0
5	Cefuroxime	–0.70	–0.47	–0.23	–1.40	–0.70	–0.70	–1.30	–1.12	–0.18	0.23	1	0
6	Chlorothiazide	–0.66	–0.28	–0.38	–0.68	–0.18	–0.50	–	–	–	–0.29	0	0
7	Doxycycline	1.45	1.16	0.29	1.32	1.19	0.13	1.30	0.90	0.40	–0.50	0	1
8	Enalapril	0.13	–0.26	–0.39	0.14	–0.48	0.62	–0.20	–0.89	0.69	0.67	1	0
9	Furosemide	0.90	0.32	0.58	–0.14	0.13	–0.27	–0.12	–0.25	0.13	1.90	1	0
10	Guanabenz	0.88	1.27	–0.39	1.10	1.44	–0.34	1.20	1.59	–0.39	2.98	0	0
11	Hydrochloro-thiazide	0.22	–0.32	0.54	0.30	–0.21	0.51	0.01	–0.15	0.16	–0.36	0	0
12	Hydrocortisone	1.33	0.75	0.58	1.36	0.90	0.46	1.29	1.02	0.27	1.89	0	0
13[a]	Ketoprofen	1.78	0.73	1.05	1.27	0.55	0.72	0.37	0.20	0.17	2.76	1	0
14	Metoprolol	0.80	0.56	0.24	0.84	0.70	0.14	0.89	0.81	0.08	1.49	0	0
15	Nadolol	–0.08	0.04	–0.12	0.06	0.15	–0.09	0.42	0.24	0.18	0.38	0	0
16[a]	Olsalazine	–0.24	1.87	–2.11	–	–	–	–	–	–	5.17	1	0
17	Oxytetracycline	0.77	0.80	–0.03	0.79	0.81	–0.02	0.43	0.50	–0.07	–1.27	0	1
18	Pindolol	0.98	0.65	0.33	0.83	0.79	0.04	0.78	0.91	–0.13	1.67	0	0
19	Practolol	–0.29	0.21	–0.50	0.19	0.34	–0.15	0.60	0.43	0.16	0.75	0	0
20	Pravastatin	0.53	0.39	0.14	–0.21	0.20	–0.41	–0.68	–0.17	–0.51	2.05	1	0
21	Procainamide	0.41	0.53	–0.12	0.86	0.67	0.19	0.88	0.78	0.10	1.42	0	0
22	Propranolol	0.94	1.16	–0.22	1.45	1.33	0.12	1.56	1.48	0.08	2.75	0	0
23	Quinidine	0.99	1.18	–0.19	1.06	1.35	–0.29	1.50	1.49	0.01	2.79	0	0
24[a,b,c]	Ranitidine	0.30	1.72	–1.42	0.34	1.77	–1.43	0.62	1.51	–0.89	0.67	0	1
25[b,c]	Sulfasalazine	0.90	1.26	–0.36	–0.17	1.11	–1.28	–1.05	0.78	–1.83	3.88	1	0
26	Sulpiride	0.00	0.38	–0.38	0.35	0.52	–0.17	0.70	0.62	0.08	1.11	0	0
27[b]	Tetracycline	0.72	0.97	–0.25	0.88	0.99	–0.11	0.35	0.69	–0.34	–0.90	0	1
28	Timolol	1.12	0.43	0.69	0.88	0.57	0.31	0.96	0.67	0.29	1.21	0	0

[a] Data points not included in deriving Eq. (6)

[b] Data points not included in deriving Eq. (7)

[c] Data points not included in deriving Eq. (8)

$$\log P_{am}[\text{pH } 6.5] = 0.77(\pm 0.15)\ \log P_{am}[\text{pH } 7.4] + 0.18(\pm 0.14) \quad (9)$$
$$n = 26, \quad r^2 = 0.827, \quad q^2 = 0.796, \quad s = 0.318$$

A similar analysis was conducted on the results obtained by Sugano et al. [13] for the BAMPA of a large set of structurally diverse drug molecules at pH 6.0.

Artificial membrane permeability of miscellaneous drugs by BAMPA at pH 6.0 (Table 4) [13]

$$\log P_{am} = 0.56(\pm 0.11)\ \text{Clog } P - 1.08(\pm 0.27)\ (\beta \cdot 10^{\text{Clog } P} + 1) - 0.94(\pm 0.29)\ I_1 + 1.28(\pm 0.44) I_4 + 0.04(\pm 0.17) \quad (10)$$
$$n = 51, \quad r^2 = 0.763, \quad q^2 = 0.710, \quad s = 0.394, \quad \log \beta = -3.063 \quad \text{Clog} P_O = 3.10$$

The substantial range in hydrophobicity (–2.54 to 6.58) of the drugs in the dataset bodes well for the development of a QSAR with bilinear properties. Permeability first increases with the increase in hydrophobicity to an optimum Clog P of 3.1 and then decreases linearly. I_1 and I_4 denote indicator variables, which acquire a value of one for the presence of –COOH and $-N(CH_3)_2$ groups, respectively. The negative coefficient of the indicator variable I_1 indicates that the presence of –COOH group decreases permeability, whereas the positive coefficient of I_4 suggests that dimethylamino groups enhance permeability. A direct comparison between the observed artificial membrane permeability coefficient measured by BAMPA at pH 5.5 (QSAR 6) and pH 6.0 (QSAR 10) for a subset of 23 common drug molecules listed in Tables 3 and 4, reveals the following relationship (See also Fig. 3.)

$$\log P_{am}[\text{pH } 5.5] = 0.89(\pm 0.14)\ \log P_{am}[\text{pH } 6.0] + 0.08(\pm 0.12) \quad (11)$$
$$n = 23, \quad r^2 = 0.896, \quad q^2 = 0.876, \quad s = 0.244$$

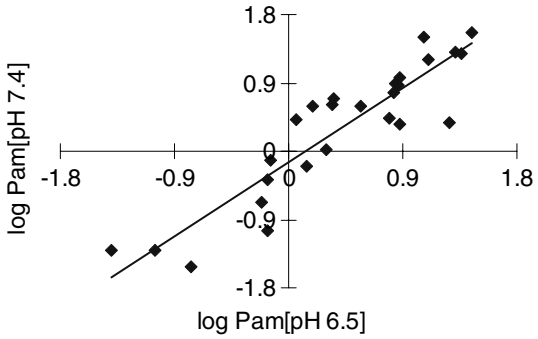

Fig. 2 Comparison of observed BAMPA permeability coefficient at pH 6.5 and 7.4

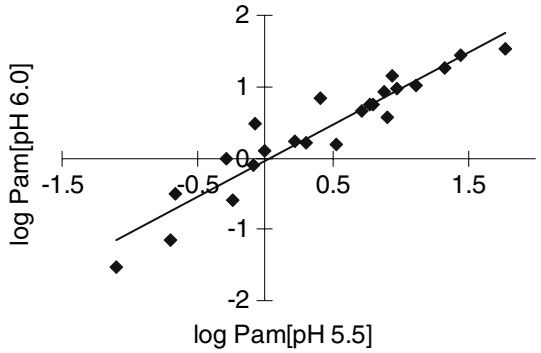

Fig. 3 Comparison of observed BAMPA permeability coefficient at pH 5.5 and 6.0

A good correlation was also found by the comparison of observed artificial membrane permeability measured by BAMPA at pH 6.5 and pH 6.0 for a subset of 22 common drug molecules listed in Tables 3 and 4, shown by Eq. (12) as well as Fig. 4.

$$\begin{aligned} &\log P_{\mathrm{am}}[\mathrm{pH}\,6.5] = 0.90(\pm 0.17)\ \log P_{\mathrm{am}}[\mathrm{pH}\,6.0] \\ &\quad + 0.07(\pm 0.15) \\ &n = 22, \quad r^2 = 0.863, \quad q^2 = 0.805, \quad s = 0.285 \end{aligned} \tag{12}$$

QSAR for bio-mimetic PAMPA-PP-RF

Bio-mimetic artificial membrane permeability of miscellaneous drugs by the paracellular pathway model based on the Renkin function (PAMPA-PP-RF) (Table 5) [27]

$$\begin{aligned} &\log P_{\mathrm{pp-rf}} = 0.48(\pm 0.22)\ \mathrm{Clog}\ P - 0.70(\pm 0.55) \\ &\quad log(\beta \cdot 10^{\mathrm{Clog}\ P} + 1) + 0.48(\pm 0.31) \\ &n = 13, \quad r^2 = 0.773, \quad q^2 = 0.606, \quad s = 0.296, \\ &\quad \log\beta = -2.641 \quad \mathrm{Clog}\ P_{\mathrm{O}} = 3.00 \end{aligned} \tag{13}$$

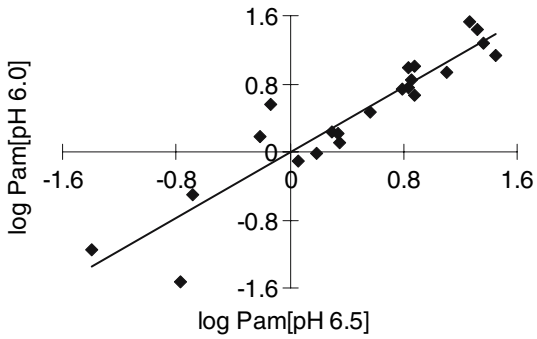

Fig. 4 Comparison of observed BAMPA permeability coefficient at pH 6.5 and 6.0

QSAR 13 was developed from the published data of Sugano et al. [27] Once again our results indicate a bilinear correlation with hydrophobicity; that is, permeability first increases with an increase in the hydrophobicity of the drug molecules up to an optimum value of Clog $P = 3.00$ and then decreases.

QSAR for PAMPA-BBB

QSAR 14 was derived from the data of Di et al. [14] It shows a strong correlation between BBB permeability and hydrophobicity.

Artificial membrane permeability of miscellaneous drugs by PAMPA-BBB (Table 6) [14]

$$\begin{aligned} &\log P_{\mathrm{e}} = 0.43(\pm 0.10)\ \mathrm{Clog}\ P - 0.54(\pm 0.25) \\ &\log(\beta \cdot 10^{\mathrm{Clog}\ P} + 1) - 0.09(\pm 0.19) \\ &n = 23, \quad r^2 = 0.842, \quad q^2 = 0.780, \quad s = 0.249, \\ &\quad \log\beta = -3.087 \quad \mathrm{Clog}\ P_{\mathrm{O}} = 3.67 \end{aligned} \tag{14}$$

QSAR in Caco-2 systems

Caco-2 cells are of human colon adenocarcinoma origin and can form confluent monolayers once they are cultured onto filter membranes. These monolayers differentiate and exhibit a morphology that is reminiscent of the small intestinal epithelium. Utilization of the data of Zhu et al. [25] for permeability coefficients of a large set of miscellaneous drugs in Caco-2 cells, resulted in the formulation of QSAR 15.

Table 4 Artificial membrane permeability coefficient of miscellaneous drugs determined by BAMPA at pH 6.0 [13]

No.	Drugs name	log P_{am} (pH 6.0) (eq. (10))			Clog P	$_1$	I_4
		obsd	calcd	Δ			
1	Acebutol	0.47	0.98	–0.51	1.71	0	0
2	Acetaminophen	0.38	0.32	0.06	0.49	0	0
3	Allopurinol	–0.29	–0.43	0.14	–0.83	0	0
4	Alprenolol	1.05	1.38	–0.33	2.65	0	0
5[a]	Antipyrine	0.96	–0.06	1.02	0.20	0	0
6	Atenolol	–0.25	–0.02	–0.23	–0.11	0	0
7	Bromocriptine	0.00	–0.05	0.05	6.58	0	0
8	Bupropion	1.08	1.44	–0.36	3.21	0	0
9	Ceftriaxone	–1.52	–0.89	–0.63	0.02	1	0
10	Cefuroxime	–1.15	–0.77	–0.38	0.23	1	0
11	Chloramphenicol	1.34	0.76	0.58	1.28	0	0
12	Chlorothiazide	–0.51	–0.12	–0.39	–0.29	0	0
13	Cimetidine	–0.07	0.26	–0.33	0.38	0	0
14[a]	Ciprofloxacin	0.87	–2.09	2.96	–0.73	1	0
15	Cloxacillin	0.28	0.40	–0.12	2.52	1	0
16	Cymarin	0.62	0.41	0.21	0.66	0	0
17	Dexamethasone	1.53	1.02	0.51	1.79	0	0
18[a]	Diclofenac	1.73	–3.34	5.07	4.73	1	0
19	Dicloxacillin	0.62	0.49	0.13	2.98	1	0
20	Doxycycline	1.44	1.04	0.40	–0.50	0	1
21	Ethambutol	–0.28	0.11	–0.39	0.12	0	0
22	Ethionamide	1.10	0.99	0.11	1.73	0	0
23	Flecainide	0.97	1.35	–0.38	3.66	0	0
24	Furosemide	0.56	0.14	0.42	1.90	1	0
25	Ganciclovir	–1.00	–1.39	0.39	–2.54	0	0
26	Guanabenz	0.93	1.44	–0.51	2.98	0	0
27[a]	Hbed	–0.57	–3.12	2.55	–1.35	1	0
28	Hydrochlorothiazide	0.23	–0.16	0.39	–0.36	0	0
29	Hydrocortisone	1.27	1.07	0.20	1.89	0	0
30	Imipramine	1.62	2.02	–0.40	5.04	0	1
31[a]	Ketoprofen	1.53	–2.33	3.86	2.76	1	0
32[a]	Lansoprazole	0.53	–1.30	1.83	2.60	0	0
33	Lincomycin	0.37	0.76	–0.39	1.28	0	0
34	Methylprednisolone	1.49	1.00	0.49	1.74	0	0
35	Metoprolol	0.75	0.87	–0.12	1.49	0	0
36	Nadolol	–0.10	0.26	–0.36	0.38	0	0
37[a]	Naltrexone	1.05	–0.14	1.19	0.36	0	0
38[a]	Naproxen	1.69	–2.36	4.05	2.82	1	0
39[a]	Norfloxacin	0.73	–2.18	2.91	–0.78	1	0
40	Olsalazine	–0.60	–0.27	–0.33	5.17	1	0
41	Oxacillin	0.25	0.21	0.04	2.05	1	0
42	Oxytetracycline	0.74	0.61	0.13	–1.27	0	1
43	Pindolol	0.98	0.96	0.02	1.67	0	0
44	Practolol	–0.02	0.47	–0.49	0.75	0	0
45	Pravastatin	0.18	0.21	–0.03	2.05	1	0
46	Prazosin	1.26	1.14	0.12	2.03	0	0
47	Procainamide	0.84	0.83	0.01	1.42	0	0
48	Propranolol	1.14	1.40	–0.26	2.75	0	0
49	Propylthiouracil	1.02	0.59	0.43	0.97	0	0
50[a]	Ranitidine	0.21	0.97	–0.76	0.67	0	1
51	Sotalol	–0.52	0.17	–0.69	0.23	0	0
52	Sulindac	0.96	0.50	0.46	3.16	1	0
53	Sulpiride	0.11	0.66	–0.55	1.11	0	0
54[a]	Sumatriptan	0.37	0.94	–0.57	0.74	0	1
55	Tetracycline	0.67	0.82	–0.15	–0.90	0	1
56	Theophylline	0.44	0.02	0.42	–0.03	0	0
57	Timolol	1.01	0.72	0.29	1.21	0	0

Table 4 Continued

No.	Drugs name	log P_{am} (pH 6.0) (eq. (10))			Clog P	$_1$	I_4
		obsd	calcd	Δ			
58	Tolbutamide	1.71	1.33	0.38	2.50	0	0
59[a]	Tranexamic acid	–0.66	–3.86	3.20	–1.80	1	0
60	Valsartan	0.32	–0.11	0.43	4.86	1	0
61	Verapamil	1.58	1.02	0.56	4.47	0	0
62	Warfarin	1.86	1.43	0.43	2.90	0	0
63	Zidovudine	0.56	0.07	0.49	0.04	0	0

[a] Data points not included in deriving Eq. (10)

Permeability of miscellaneous drugs determined by Caco-2 assay (Table 7) [25]

$$\begin{aligned}&\log P_{app} = 0.57(\pm 0.10)\ \text{Clog}\ P - 0.72(\pm 0.14)\\&\quad \log(\beta \cdot 10^{\text{Clog}\,P} + 1) - 0.89(\pm 0.45)\, I_3 + 0.25(\pm 0.18)\\&n = 42, \quad r^2 = 0.800, \quad q^2 = 0.745, \quad s = 0.373,\\&\quad \log \beta = -2.533 \quad \text{Clog}P_O = 3.11 \qquad (15)\end{aligned}$$

This QSAR is governed by hydrophobic terms and a bilinear relationship. The optimum hydrophobicity is once again, around 3.0. I_3, which denotes the presence of aromatic –OH group suggests that strong hydrogen bond donor groups are detrimental to intestinal absorption. This has been previously observed in the QSAR for the –COOH groups.

QSAR for human intestinal absorption

Using the data derived by Sugano et al. [13] the following QSAR 16 was developed for human intestinal absorption of a diverse set of drugs.

Percentage of drug absorbed by the human intestine (Table 8) [13]

$$\begin{aligned}&\log F_a = 0.28(\pm 0.05)\ \text{Clog}\ P - 0.69(\pm 0.22) I_1\\&\quad + 0.72(\pm 0.36) I_4 + 1.47(\pm 0.11)\\&n = 57, \quad r^2 = 0.711, \quad q^2 = 0.654, \quad s = 0.328 \qquad (16)\end{aligned}$$

It is of interest to note that in this study (n = 68), there were only three drugs with Clog P values > 3.5 so it is difficult to have a convergence of the bilinear model with such data. Once again we see a positive dependence on the presence of the dimethylamino function (I_4) and a negative correlation with the carboxyl group.

Human intestinal permeability of miscellaneous drugs (Table 9) [27]

$$\begin{aligned}&\log P_{eff} = 0.31(\pm 0.15)\ \text{Clog}\ P - 1.12(\pm 0.62)\\&\quad I_2 - 0.47(\pm 0.41) \qquad (17)\\&n = 11, \quad r^2 = 0.883, \quad q^2 = 0.732, \quad s = 0.325\end{aligned}$$

QSAR 17 was also developed from the published data of Sugano et al. [27]; our results indicate a strong correlation with increased hydrophobicity. The presence of a strong hydrogen bond donor group (SO_2NH_2) leads to decreased permeability.

PAMPA versus BAMPA

There is limited correlation between the experimental values obtained from these two assays, PAMPA and BAMPA, since the pH values utilized in these two assays are different. See Eqs. (18) and (19). In a direct comparison between the observed PAMPA flux at pH 7.4 (log F) and BAMPA membrane permeability coefficient at pH 6.0 (log P_{am}) for a subset of 28 common drug molecules listed in Tables 1 and 4, the following correlation is obtained.

$$\begin{aligned}&\log F[\text{pH}\,7.4] = 0.51(\pm 0.14)\ \log P_{am}[\text{pH}\,6.0]\\&\quad + 0.96(\pm 0.14) \qquad (18)\\&n = 28, \quad r^2 = 0.691, \quad q^2 = 0.562, \quad s = 0.252\end{aligned}$$

A similar comparison is carried out between the observed PAMPA flux at pH 7.4 (with the addition of glycocolic acid) and BAMPA membrane permeability coefficient at pH 6.0 for a subset of 20 common drug molecules listed in Tables 1 and 4. This results in the formulation of QSAR 19.

Table 5 Bio-mimetic artificial membrane permeability of miscellaneous drugs determined by PAMPA-PP-RF [27]

No.	Drugs name	log $P_{pp\text{-}rf}$ (eq. (13))			Clog P
		obsd	calcd	Δ	
1	Amiloride	0.76	0.53	0.23	0.11
2[a]	Antipyrine	1.03	0.58	0.45	0.20
3	Atenolol	0.39	0.43	–0.04	–0.11
4	Carbamazepine	1.73	1.49	0.24	2.38
5	Cimetidine	0.51	0.66	–0.15	0.38
6	Desipramine	1.14	1.36	–0.22	4.47
7[a]	Furosemide	0.57	1.34	–0.77	1.90
8	hydrochlorothiazide	0.45	0.30	0.15	–0.36
9	ketoprofen	1.53	1.56	–0.03	2.76
10	metoprolol	0.87	1.17	–0.30	1.49
11	naproxen	1.70	1.56	0.14	2.82
12	piroxicam	1.77	1.34	0.43	1.89
13	propranolol	1.19	1.56	–0.37	2.75
14	ranitidine	0.48	0.80	–0.32	0.67
15	verapamil	1.58	1.36	0.22	4.47

[a] Data points not included in deriving Eq. (13)

Table 6 Artificial membrane permeability of miscellaneous drugs for the prediction of blood-brain barrier (PAMPA-BBB) [14]

No.	Drugs name	log P_e (eq. (14))			Clog P
		obsd	calcd	Δ	
1	alprazolam	0.73	0.94	–0.21	2.55
2	caffeine	0.11	–0.11	0.22	–0.04
3	chloropromazine	0.81	0.98	–0.17	5.30
4	clobazam	1.23	0.90	0.33	2.44
5	clonidine	0.72	0.51	0.21	1.43
6	desipramine	1.08	1.07	0.01	4.47
7	diazepam	1.20	1.05	0.15	2.96
8	β –estradiol	1.08	1.11	–0.03	3.78
9	imipramine	1.11	1.01	0.10	5.04
10	oxazepam	1.00	0.86	0.14	2.31
11	progesterone	0.97	1.10	–0.13	3.97
12	promazine	0.94	1.07	–0.13	4.40
13	testosterone	1.23	1.10	0.13	3.41
14	thiopental	1.26	1.05	0.21	2.98
15	aldosterone	0.08	–0.04	0.12	0.13
16	astemizole	1.04	0.92	0.12	5.84
17	atenolol	–0.10	–0.14	0.04	–0.11
18	hydrocortisone	0.28	0.70	–0.42	1.89
19[a]	dopamine	–0.70	–0.11	–0.59	0.17
20[a]	enoxacin	–0.05	–1.64	1.59	–1.60
21[a]	isoxicam	–0.52	–0.28	–0.24	1.61
22	lomefloxacin	0.04	–0.14	0.18	–0.11
23	corticosterone	0.71	0.93	–0.22	2.51
24	norfloxacin	–1.00	–0.43	–0.57	–0.78
25	ofloxacin	–0.10	–0.31	0.21	–0.51
26	piroxicam	0.40	0.70	–0.30	1.89
27[a]	tenoxicam	–1.00	–0.28	–0.72	1.61

[a] Data points not included in deriving Eq. (14)

$$\log F_{ga}[\text{pH}\,7.4] = 0.63(\pm 0.20)\ \log P_{am}[\text{pH}\,6.0] + 0.78(\pm 0.22) \quad (19)$$
$$n = 20, \quad r^2 = 0.702, \quad q^2 = 0.647, \quad s = 0.233$$

PAMPA versus bio-mimetic PAMPA-PP-RF

In a direct comparison between the observed PAMPA flux at pH 7.4 and bio-mimetic artificial membrane permeability corrected for paracellular permeability based on the Renkin function (PAMPA-PP-RF) for a small subset of 12 common drug molecules listed in Tables 1 and 5, the following correlation was obtained (Fig. 5).

$$\log F[\text{pH}\,7.4] = 0.85(\pm 0.26)\ \log P_{pp-rf} + 0.50(\pm 0.29)$$
$$n = 12, \quad r^2 = 0.845, \quad q^2 = 0.789, \quad s = 0.201 \quad (20)$$

BAMPA versus bio-mimetic PAMPA-PP-RF

A similar analysis between the observed BAMPA membrane permeability coefficients at pH 6.0 and bio-mimetic artificial membrane permeabilities corrected for paracellular permeability based on the Renkin function (PAMPA-PP-RF) for a subset of 11 common drug molecules listed in Tables 4 and 5, led to the following derivation (Fig. 6).

$$\log P_{am}[\text{pH}\,6.0] = 1.33(\pm 0.25)\ \log P_{pp-rf} - 0.48(\pm 0.26)$$
$$n = 11, \quad r^2 = 0.941, \quad q^2 = 0.914, \quad s = 0.174 \quad (21)$$

PAMPA versus Caco-2 assays

Excellent correlations were obtained between experimental values obtained from PAMPA and Caco-2 cell assays. In a direct comparison between the observed PAMPA flux at pH 7.4 and Caco-2 membrane permeability coefficients for a subset of 26 common drug molecules listed in Tables 1 and 7, the following QSAR was obtained. See Fig. 7.

$$\log F[\text{pH}\,7.4] = 0.46(\pm 0.08)\ \log P_{app} + 1.12(\pm 0.10)$$
$$n = 25, \quad r^2 = 0.849, \quad q^2 = 0.823, \quad s = 0.186 \quad (22)$$

Similarly, a comparison between the observed PAMPA permeability at pH 5.5 (log P_{app} [pH 5.5]) and Caco-2 membrane permeability coefficient (log P_{app}) for a subset of 43 common drug molecules listed in Tables 2 and 7, yielded an adequate correlation as delineated by QSAR 23 and Fig. 8. However, the number of outliers was extensive. Reasons for their aberrant behavior are not clear.

$$\log P_{app}[\text{pH}\,5.5] = 0.87(\pm 0.15)\ \log P_{app} - 0.26(\pm 0.18)$$
$$n = 36, \quad r^2 = 0.811, \quad q^2 = 0.785, \quad s = 0.352 \quad (23)$$

Table 7 Permeability coefficient of miscellaneous drugs determined by Caco-2 membrane [25]

No.	Drugs name	log P_{app} (eq. (15))			Clog P	I_3
		obsd	calcd	Δ		
1[a]	acebutolol	–0.29	1.18	–1.47	1.71	0
2	acetylsalicylic acid	0.38	0.82	–0.44	1.02	0
3	acyclovir	–0.60	–1.12	0.52	–2.42	0
4	alprenolol	1.40	1.49	–0.09	2.65	0
5[a]	amoxicillin	–0.10	–1.70	1.60	–1.87	1
6[a]	antipyrine	1.45	0.37	1.08	0.20	0
7	atenolol	–0.70	0.19	–0.89	–0.11	0
8[a]	caffeine	1.49	0.23	1.26	–0.04	0
9	cephalexin	–0.30	–0.79	0.49	–1.84	0
10	chloramphenicol	1.31	0.96	0.35	1.28	0
11	chlorothiazide	–0.82	0.09	–0.91	–0.29	0
12	chlorpromazine	1.30	1.27	0.03	5.30	0
13	cimetidine	–0.13	0.47	–0.60	0.38	0
14	clonidine	1.48	1.04	0.44	1.43	0
15	corticosterone	1.74	1.47	0.27	2.51	0
16	cyclosporin	–0.05	–0.09	0.04	14.36	0
17	desipramine	1.33	1.39	–0.06	4.47	0
18	dexamethasone	1.09	1.21	–0.12	1.79	0
19	diltiazem	1.47	1.50	–0.03	3.65	0
20	doxorubicin	–0.80	–0.46	–0.34	0.32	1
21	enalapril	0.36	0.63	–0.27	0.67	0
22	erythromycin	0.57	1.13	–0.56	1.61	0
23[a]	furosemide	–0.92	1.26	–2.18	1.90	0
24	griseofulvin	1.56	1.33	0.23	2.05	0
25	guanabenz	1.32	1.53	–0.21	2.98	0
26	hydrochlorothiazide	–0.29	0.05	–0.34	–0.36	0
27	hydrocortisone	1.15	1.26	–0.11	1.89	0
28	ibuprofen	1.72	1.49	0.23	3.68	0
29	imipramine	1.15	1.31	–0.16	5.04	0
30	indomethacin	1.31	1.43	–0.12	4.18	0
31	labetalol	0.97	0.58	0.39	2.50	1
32	methotrexate	0.08	–0.05	0.13	–0.53	0
33	metoprolol	1.37	1.07	0.30	1.49	0
34	nadolol	0.59	0.47	0.12	0.38	0
35	naproxen	1.60	1.51	0.09	2.82	0
36	nicotine	1.29	0.75	0.54	0.88	0
37	phenytoin	1.43	1.34	0.09	2.09	0
38	pindolol	1.22	1.16	0.06	1.67	0
39	piroxicam	1.55	1.26	0.29	1.89	0
40	prazosin	1.64	1.32	0.32	2.03	0
41	progesterone	1.37	1.46	–0.09	3.97	0
42	propranolol	1.62	1.51	0.11	2.75	0
43	quinidine	1.31	1.51	–0.20	2.79	0
44[a]	ranitidine	–0.31	0.63	–0.94	0.67	0
45[a]	salicylic acid	1.62	0.49	1.13	2.19	1
46[a]	salfasalazine	–0.89	0.58	–1.47	3.88	1
47	terbutaline	–0.42	–0.37	–0.05	0.48	1
48	timolol	1.11	0.93	0.18	1.21	0
49	warfarin	1.32	1.52	–0.20	2.90	0
50	zidovudin(AZT)	0.84	0.28	0.56	0.04	0

[a] Data points not included in deriving Eq. (15)

BAMPA versus Caco-2 assays

A good correlation was obtained between the observed BAMPA membrane permeability coefficient at pH 6.5 and Caco-2 membrane permeability coefficients for a subset of 16 common drug molecules listed in Tables 3 and 7, see QSAR 24 and Fig. 9.

$$\log P_{am}[\text{pH } 6.5] = 0.61(\pm 0.19)\ \log P_{app} + 0.23(\pm 0.20)$$
$$n = 14, \quad r^2 = 0.800, \quad q^2 = 0.733, \quad s = 0.298 \tag{24}$$

In a similar vein, the correlation between the observed BAMPA membrane permeability coefficients at pH 7.4 and Caco-2 membrane permeability coefficients for a subset of 15 common drug molecules listed in Tables 3 and 7, is represented by QSAR 25 and Fig. 10.

$$\log P_{am}[\text{pH } 7.4] = 0.91(\pm 0.28)\ \log P_{app} - 0.06(\pm 0.30)$$
$$n = 13, \quad r^2 = 0.822, \quad q^2 = 0.715, \quad s = 0.411 \tag{25}$$

Bio-mimetic PAMPA-PP-RF versus Caco-2 assays

In a direct comparison between the observed bio-mimetic artificial membrane permeabilities corrected for paracellular permeability (PAMPA-PP-RF) and Caco-2 membrane permeability coefficients for a subset of 11 common drug molecules listed in Tables 5 and 7, the following QSAR was developed. See also Fig. 11.

$$\log P_{pp-rf} = 0.43(\pm 0.18)\ \log P_{app} + 0.70(\pm 0.21)$$
$$n = 10, \quad r^2 = 0.790, \quad q^2 = 0.659, \quad s = 0.254 \tag{26}$$

PAMPA versus human intestinal permeability

Excellent correlation is obtained between 11 experimental values of permeabilities obtained from PAMPA, pH 7.4 and human intestine. See QSAR 27 and also Fig. 12.

$$\log F[\text{pH } 7.4] = 0.48(\pm 0.15)\ \log P_{eff} + 1.38(\pm 0.13)$$
$$n = 11, \quad r^2 = 0.854, \quad q^2 = 0.796, \quad s = 0.187 \tag{27}$$

The correlation between the observed PAMPA permeability at pH 5.5 and human intestine permeability for a subset of 12 common drug molecules listed in Tables 2 and 9, is described by QSAR 28 and Fig. 13.

$$\log P_{app}[\text{pH } 5.5] = 0.93(\pm 0.30)\ \log P_{eff} + 0.38(\pm 0.26)$$
$$n = 12, \quad r^2 = 0.827, \quad q^2 = 0.712, \quad s = 0.386 \tag{28}$$

Similarly, the comparison between the observed PAMPA permeability at pH 7.4 and human intestine permeability for a subset of 10 common drug molecules

Table 8 Percentage of miscellaneous drugs absorbed by the human intestine [13]

No.	Drugs name	log F_a (eq. (16))			Clog P	I_1	I_4
		obsd	calcd	Δ			
1	acebutolol	1.95	1.94	0.01	1.71	0	0
2	acetaminophen	1.90	1.61	0.29	0.49	0	0
3	acyclovir	1.30	0.80	0.50	–2.42	0	0
4[a]	allopurinol	1.95	1.24	0.71	–0.83	0	0
5	alprenolol	1.97	2.21	–0.24	2.65	0	0
6	antipyrine	1.99	1.53	0.46	0.20	0	0
7	atenolol	1.70	1.44	0.26	–0.11	0	0
8	aztreonam	0.00	0.88	–0.88	0.34	1	0
9[a]	bromocriptine	1.45	3.29	–1.84	6.58	0	0
10	bupropion	1.94	2.36	–0.42	3.21	0	0
11	ceftriaxone	0.00	0.79	–0.79	0.02	1	0
12	cefuroxime	0.70	0.85	–0.15	0.23	1	0
13	chloramphenicol	1.95	1.83	0.12	1.28	0	0
14	chlorothiazide	1.11	1.39	–0.28	–0.29	0	0
15	cimetidine	1.81	1.58	0.23	0.38	0	0
16[a]	ciprofloxacin	1.84	0.58	1.26	–0.73	1	0
17	cloxacillin	1.69	1.48	0.21	2.52	1	0
18[a]	creatinine	1.90	0.98	0.92	–1.77	0	0
19	cymarin	1.67	1.65	0.02	0.66	0	0
20	dexamethasone	1.90	1.96	–0.06	1.79	0	0
21	dicloxacillin	1.74	1.61	0.13	2.98	1	0
22	doxycycline	1.98	2.05	–0.07	–0.50	0	1
23[a]	erythritol	1.95	1.00	0.95	–1.71	0	0
24	ethambutol	1.90	1.50	0.40	0.12	0	0
25	ethionamide	1.90	1.95	–0.05	1.73	0	0
26	famotidine	1.58	1.31	0.27	–0.58	0	0
27	fenoterol	1.78	1.74	0.04	0.98	0	0
28[a]	flecainide	1.91	2.48	–0.57	3.66	0	0
29	furosemide	1.79	1.31	0.48	1.90	1	0
30	ganciclovir	0.48	0.77	–0.29	–2.54	0	0
31	guanabenz	1.88	2.30	–0.42	2.98	0	0
32	hbed	0.70	0.41	0.29	–1.35	1	0
33	hydrochlorothiazide	1.83	1.37	0.46	–0.36	0	0
34	hydrocortisone	1.96	1.99	–0.03	1.89	0	0
35	lactulose	–0.22	0.48	–0.70	–3.59	0	0
36	lansoprazole	1.93	2.19	–0.26	2.60	0	0
37	lincomycin	1.45	1.82	–0.37	1.28	0	0
38	mannitol	1.20	0.90	0.30	–2.05	0	0
39	metaproterenol	1.64	1.49	0.15	0.08	0	0
40	metformin	1.93	1.74	0.19	–1.63	0	1
41	methylprednisolone	1.91	1.95	–0.04	1.74	0	0
42	metoprolol	1.98	1.88	0.10	1.49	0	0
43	nadolol	1.54	1.58	–0.04	0.38	0	0
44	naltrexone	1.98	1.57	0.41	0.36	0	0
45	naproxen	2.0	1.56	0.44	2.82	1	0
46[a]	norfloxacin	1.85	0.57	1.28	–0.78	1	0
47[a]	olsalazine	0.30	2.21	–1.91	5.17	1	0
48	oxacillin	1.52	1.35	0.17	2.05	1	0
49	oxytetracycline	1.78	1.84	–0.06	–1.27	0	1
50	pindolol	1.95	1.93	0.02	1.67	0	0
51	pravastatin	1.53	1.35	0.18	2.05	1	0
52	prazosin	1.93	2.03	–0.10	2.03	0	0
53	procainamide	1.93	1.86	0.07	1.42	0	0
54	propranolol	1.95	2.23	–0.28	2.75	0	0
55	propylthiouracil	1.88	1.74	0.14	0.97	0	0
56	raffinose	–0.52	0.00	–0.52	–5.32	0	0
57[a]	ranitidine	1.70	2.38	–0.68	0.67	0	1
58	sotalol	1.78	1.53	0.25	0.23	0	0
59	sulindac	1.95	1.66	0.29	3.16	1	0
60	sulpiride	1.54	1.78	–0.24	1.11	0	0

Table 8 Continued

No.	Drugs name	log F_a (eq. (16))			Clog P	I_1	I_4
		obsd	calcd	Δ			
61[a]	sumatriptan	1.76	2.40	–0.64	0.74	0	1
62	terbutaline	1.79	1.60	0.19	0.48	0	0
63	tetracycline	1.89	1.94	–0.05	–0.90	0	1
64	timolol	1.95	1.81	0.14	1.21	0	0
65	tolbutamide	1.93	2.16	–0.23	2.50	0	0
66[a]	tranexamic acid	1.74	0.28	1.46	–1.80	1	0
67	valsartan	1.74	2.13	–0.39	4.86	1	0
68	warfarin	1.99	2.27	–0.28	2.90	0	0

[a] Data points not included in deriving Eq. (16)

listed in Tables 2 and 9, results in the following relationship (Fig. 14).

$$\log P_{app}[\text{pH } 7.4] = 0.60(\pm 0.19)\ \log P_{eff} + 0.57(\pm 0.16)$$
$$n = 9, \quad r^2 = 0.889, \quad q^2 = 0.843, \quad s = 0.160 \qquad (29)$$

BAMPA versus human intestinal permeability

Direct comparisons between the observed BAMPA permeability at pH 6.0 and 6.5 and the percentage dose absorption in human intestine for subsets of common drugs listed in Tables 3, 4 and 8, resulted in the following correlations and Figs. 15 and 16.

$$\log P_{am}[\text{pH } 6.0] = 1.33(\pm 0.20)\ \log F_a - 1.66(\pm 0.35)$$
$$n = 47, \quad r^2 = 0.797, \quad q^2 = 0.773, \quad s = 0.323 \qquad (30)$$

$$\log P_{am}[\text{pH } 6.5] = 2.12(\pm 0.47)\ \log F_a - 3.22(\pm 0.82)$$
$$n = 22, \quad r^2 = 0.816, \quad q^2 = 0.759, \quad s = 0.337 \qquad (31)$$

Bio-mimetic PAMPA-PP-RF versus human intestinal permeability

A similar comparison between the observed bio-mimetic artificial membrane permeability corrected for paracellular permeability and human intestinal permeability for a subset of 15 common drug molecules listed in Tables 5 and 9, led to the following equation and Fig. 17.

$$\log P_{pp-rf} = 0.58(\pm 0.18)\ \log P_{eff} + 1.00(\pm 0.15)$$
$$n = 14, \quad r^2 = 0.797, \quad q^2 = 0.710, \quad s = 0.251 \qquad (32)$$

Discussion

There are two main routes by which compounds cross the epithelial barrier. Hydrophilic compounds utilize the paracellular pathway while hydrophobic compounds rely on the transcellular route for permeation [28]. Compounds subject to efflux and absorptive transporter effects can distort this pattern and add to the complexity of the drug absorptive picture. Nevertheless, it is clear that for transcellular diffusion, desolvation of the hydration shell of a compound is critical to a drug's ability to traverse the cell membrane. Thus hydrogen bonding capability and lipophilicity will enter into play as well as size, albeit on a reduced scale [29].

In all 13 QSAR (1, 2, 4–8, 10, 13–17) pertaining to drug absorption, hydrophobicity as represented by Clog P, plays a significant and omnipresent role. The coefficients with the linear hydrophobic terms range from 0.22 to 0.57, with an average of 0.43. This suggests that the partitioning of these drugs between the aqueous and lipophilic phases is partially similar to their partitioning between octanol and water. Of the 13 QSAR, eight (QSAR 1, 2, 4, 5, 10, 13–15) are bilinear in nature with optimum Clog P values around 3.06 (range extends from 2.6 to 3.67). In these data sets, a bilinear relationship could be formulated due to the large range in hydrophobicities and the reasonable number of compounds with Clog P values > 3.0. At least 14% of the compounds in these datasets met the latter criteria. In QSAR where bilinear relationships were not obtainable (QSAR 6–8), only 4% of the drugs had Clog P values > 3 and hence linear relationships were predominant.

Indicator variables, which pertain to the occurrence of various functional groups such as –COOH (I_1), –SO_2NH_2(I_2), aromatic –OH (I_3), and the –$N(CH_3)_2$(I_4), in the compounds, were present in all but two of the QSAR, i.e. QSAR 13 and 14. QSAR 13 precludes the addition of any more variables because of its limited size. In the case of the acidic moieties, the signs of the coefficients are all negative. The only one with a positive contribution to permeability is the basic –$N(CH_3)_2$ entity, whose positive contribution is enhanced under more acidic pH conditions. The I_4

Table 9 Human intestinal permeability of miscellaneous drugs [27]

No.	Drugs name	log P_{eff} (eq. (17))			Clog P	I_2
		obsd	calcd	Δ		
1[a]	amiloride	0.21	–0.44	0.65	0.11	0
2[a]	antipyrine	0.65	–0.41	1.06	0.20	0
3	atenolol	–0.70	–0.50	–0.20	–0.11	0
4	carbamazepine	0.63	0.27	0.36	2.38	0
5	cimetidine	–0.52	–0.35	–0.17	0.38	0
6	desipramine	0.64	0.93	–0.29	4.47	0
7	furosemide	–1.30	–1.00	–0.30	1.90	1
8	hydrochlorothiazide	–1.40	–1.70	0.30	–0.36	1
9[a]	ketoprofen	0.92	0.39	0.53	2.76	0
10	metoprolol	0.11	–0.01	0.12	1.49	0
11	naproxen	0.92	0.41	0.51	2.82	0
12[a]	piroxicam	0.89	0.12	0.77	1.89	0
13	propranolol	0.46	0.39	0.07	2.75	0
14	ranitidine	–0.57	–0.26	–0.31	0.67	0
15	verapamil	0.83	0.93	–0.10	4.47	0

[a] Data points not included in deriving Eq. (17)

descriptor appears in QSAR 6–8, 10 and 16. Its' coefficients are large and positive (+0.72 to +1.54) and more pronounced at acidic pH.

On the other hand the other prominent indicator variable, I_1, with a significant coefficient is the –COOH group. However, its sign is negative and as the pH increases from 5.5 to 7.4 so does its magnitude. Thus its effect on drug absorption is negative and leads to a decrease. It can be surmised that removal of these acidic hydrogen bond donor groups would improve the absorption profiles of these drugs. It has been shown that removal of hydrogen bonding functionality in a series of X-phenylalanine peptidomimetics led to an improvement in permeability [30]. A similar effect is seen with the $–SO_2NH_2$ group and its indicator variable I_2; its coefficients in QSAR 1, 2, 4, 5 and 17 range from –0.45 to –1.40. Clearly, its presence in a drug is detrimental to absorption in pH ranges from 5.5 to 7.4. The phenolic –OH when present also contributes in a similar, negative fashion.

The importance of hydrogen bonding capability in predicting absorption related activities is underscored by the extensive use of polar surface area in various applications [31, 32]. Oprea and Gottfries' [33] analysis of permeability has revealed that the two predominant molecular attributes are hydrogen bonding capacity and hydrophobic transferability. They have also shown that hydrogen bond donors (HBD) wield a stronger effect than acceptors in drug permeability since lipids that constitute most membranes have ester head groups that are water solvated and can easily form hydrogen bonds with HBDs and not acceptors (HBA). Thus HBD and HBA drugs would experience different microdomains within the absorbing epithelium. HBD solutes would face a significant barrier in movement across the apolar regions of the membrane in which interfacial hydrogen bonds would have to be broken [34]. On the other hand the positive coefficients with I_4 indicate that permeability is enhanced in the case of the HBA. Oprea has suggested that HBA could be

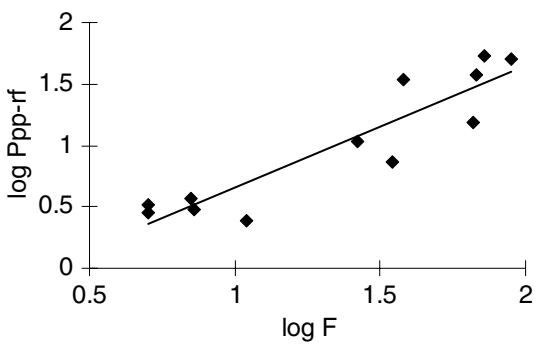

Fig. 5 Comparison of observed PAMPA flux at pH 7.4 and bio-mimetic PAMPA-PP-RF

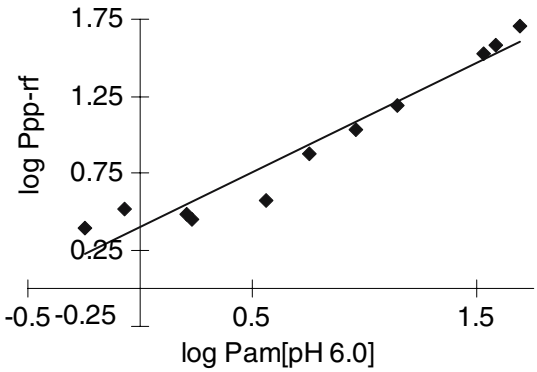

Fig. 6 Comparison of observed BAMPA permeability coefficient at pH 6.0 and bio-mimetic PAMPA-PP-RF

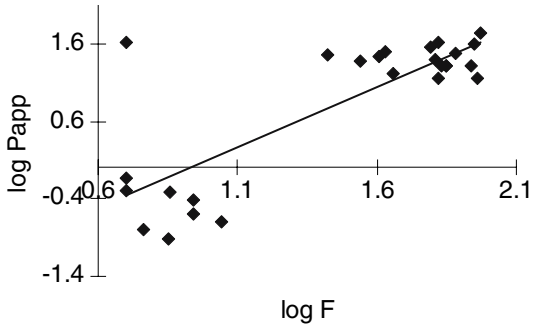

Fig. 7 Comparison of observed PAMPA flux at pH 7.4 and Caco-2 membrane permeability

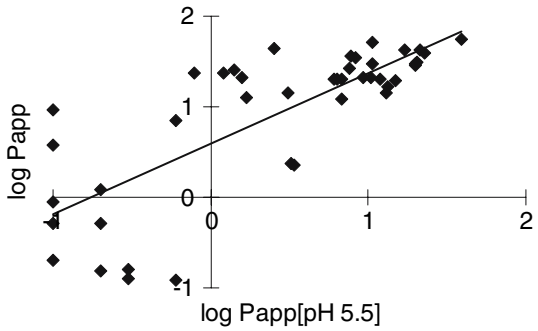

Fig. 8 Comparison of observed permeability coefficient measured by PAMPA at pH 5.5 and Caco-2 membrane

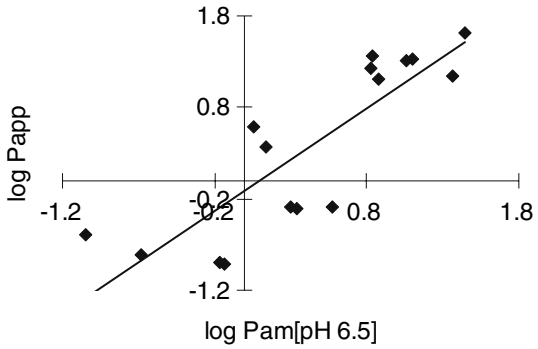

Fig. 9 Comparison of observed permeability coefficient measured by BAMPA at pH 6.5 and Caco-2 membrane

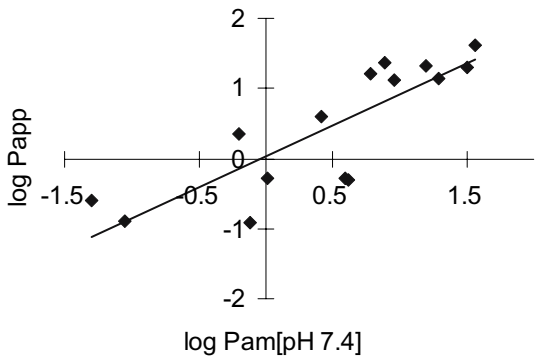

Fig. 10 Comparison of observed permeability coefficient measured by BAMPA at pH 7.4 and Caco-2 membrane

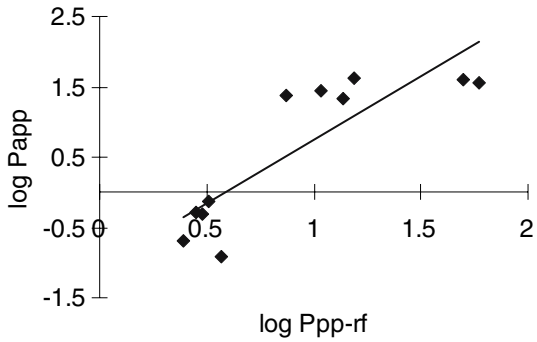

Fig. 11 Comparison of observed permeability measured by biomimetic PAMPA-PP-RF and Caco-2 membrane

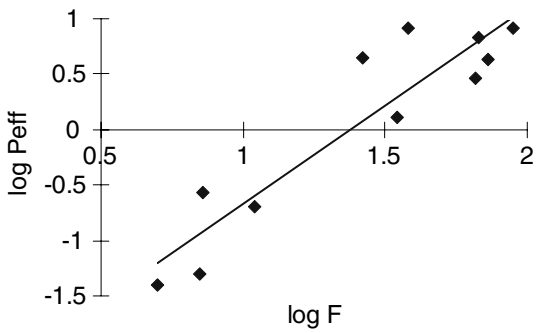

Fig. 12 Comparison of observed PAMPA flux at pH 7.4 and human intestine permeability

water solvated in an analogous fashion to the esteratic lipid head groups and thus be conducive to transmembrane diffusion [35]. The results presented here with the inclusion of indicator variables for the polar entities also lend support to the importance of hydrogen bond capability in membrane permeability.

The importance of hydrophobicity in enhancing membrane permeability has been delineated by the Lien group and Norinder and Osterberg whose models have also emphasized the negative contributions of hydrogen bonding in various transport phenomena [36, 37].

The percentage of outliers in all these datasets range from 7 to 20%. QSAR 15–17 represent in vitro assays (Caco-2 and human intestinal cells) where a number of the outliers may be subject to efflux and/or absorptive transporter effects. e.g. furosemide, acebutolol, pravastatin and ranitidine. QSAR 1–12 all pertain to artificial membrane permeability assays which have some drawbacks that limit their effectiveness [38]. They include the absence of efficient stirring conditions and the lack of constant concentration gradients that

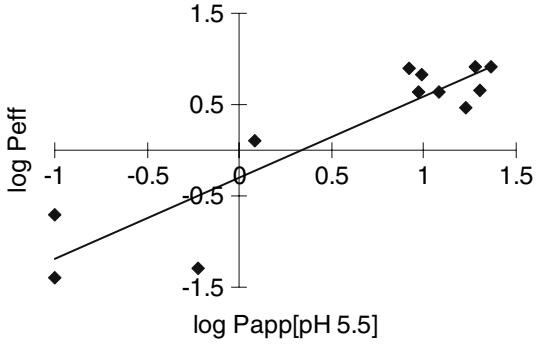

Fig. 13 Comparison of observed permeability measured by PAMPA at pH 5.5 and human intestine

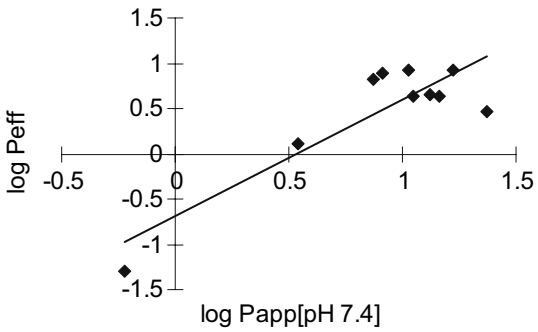

Fig. 14 Comparison of observed permeability measured by PAMPA at pH 7.4 and human intestine

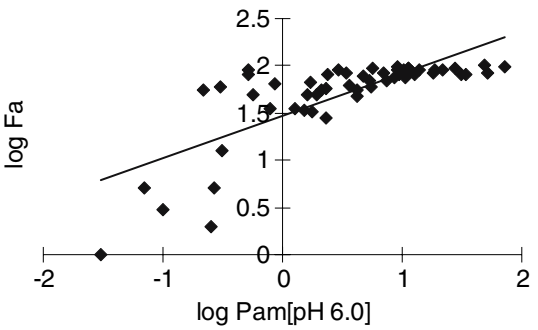

Fig. 15 Comparison of observed BAMPA permeability at pH 6.0 and the percentage dose absorption in human intestine

can result in back-diffusion of the drugs and subsequent erroneous low permeability values.

It is of interest to compare QSAR 1 and 5 for PAMPA flux and PAMPA permeability, respectively with a similar QSAR obtained by Fujikawa et al. (QSAR 33) for PAMPA permeability [39]. They evaluated the absorption of a diverse set of drugs across a membrane via the transcellular route by utilizing PAMPA.

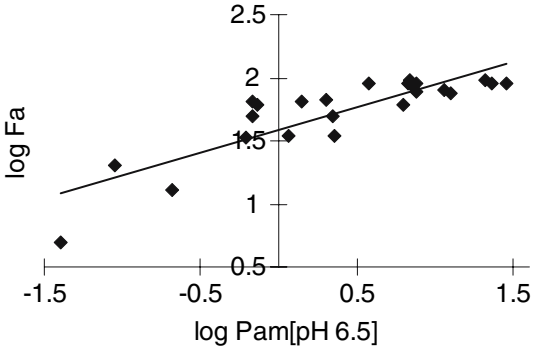

Fig. 16 Comparison of observed BAMPA permeability at pH 6.5 and the percentage drug absorption in human intestine

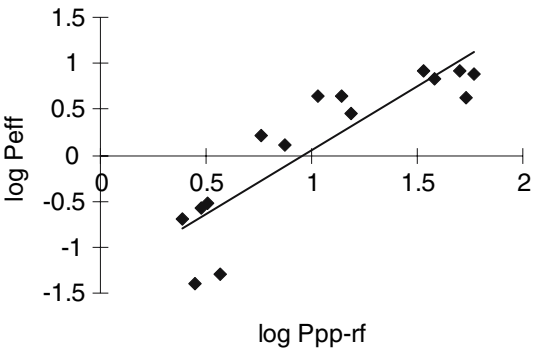

Fig. 17 Comparison of observed permeability measured by biomimetic PAMPA-PP-RF and the human intestine

Permeation of diverse drugs as measured by PAMPA at pH 7.3 [39]

$$\log P_{\text{app}} = 0.36(\pm 0.09)\ \text{Clog}\ P - 0.24(\pm 0.09)\ (\text{calcd pK}_a - \text{pH}) - 1.24(\pm 0.53)\text{SA}_{\text{HA}} - 0.77(\pm 0.47)\ \text{SA}_{\text{HD}} - 4.86(\pm 0.33)$$
$$n = 57, \quad r^2 = 0.720, \quad q^2 = 0.670, \quad s = 0.360 \qquad (33)$$

All three QSAR (1, 5 and 33) show a positive dependence on hydrophicity with Clog *P* coefficients of 0.22, 0.51 and 0.36, respectively. However, QSAR 1 and 5 show a bilinear dependence on hydrophobicity while QSAR 33 does not. An examination of the data used to derive QSAR 33 reveals that it only contains 8 compounds with Clog $P > 3$ while the data sets associated with QSAR 1 and 5 have 26 and 20 such compounds, respectively. Thus an adequate span of hydrophobic space that is also well populated allows for a determination of the optimum hydrophobicity and comparative QSAR analysis of various data sets vouches for the validity of the parameter. QSAR 1, 5 and 33 all stress the deleterious effects of hydrogen bonding on PAMPA permeability.

Fujikawa et al. [39] also established a strong relationship between Caco-2 cell and PAMPA permeabilities, suggesting that PAMPA permeability is an excellent predictor of Caco-2 cell permeability via the transcellular route. Their QSAR is listed as follows:

$$\log P_{\text{app-Caco-2}} = 1.03(\pm 0.21)\ \log P_{\text{app-pampa}} + 0.39(\pm 1.10) \quad (34)$$
$$n = 27, \quad r^2 = 0.81, \quad q^2 = 0.78, \quad s = 0.31$$

A similar comparison between the observed PAMPA permeability at pH 7.4 (log P_{app} [pH 7.4]) and Caco-2 membrane permeability coefficient (log $P_{\text{app-Caco-2}}$) for a subset of 30 common drug molecules in Tables 2 and 7 yielded the following QSAR 35.

$$\log P_{\text{app-Caco-2}} = 1.16(\pm 0.21)\ \log P_{\text{app}}[\text{pH}\,7.4] + 0.16(\pm 0.20) \quad (35)$$
$$n = 30, \quad r^2 = 0.818, \quad q^2 = 0.778, \quad s = 0.323$$

Despite differences in the drugs used to formulate QSAR 34 and 35, the equations are highly similar in terms of their coefficients with PAMPA permeabilities. These results suggest that PAMPA permeabilities are reliable and accurate predictors of Caco-2 cell permeabilities of a wide array of drugs.

The comparative QSAR studies on PAMPA/modified PAMPA with respect to that of Caco-2 cell as well as human intestinal absorption of drug molecules, gave nineteen QSAR (Eqs. (3), (9), (11), (12) and (18–32)). A strong correlation is shown by Eq. (3) and Fig. 1 suggesting that PAMPA flux will not be affected by the addition of glycocolic acid. BAMPA results are highly dependent on pH conditions as revealed by QSAR (Eqs. (6)–(8) and 10). This is the reason that good correlations are only demonstrated between the BAMPA results under similar pH conditions (Eqs. (9), (11) and (12) and Figs. 2, 3 and 4). There is an adequate correlation between PAMPA and BAMPA (Eqs. (18) and (19)); this may be attributed to differences in the pH of the test system. For a small set, bio-mimetic PAMPA-PP-RF has a good correlation with PAMPA (Eq. (20) and Fig. 5) as well as BAMPA (Eq. (21) and Fig. 6).

Caco-2 cell membrane permeability data was well correlated with PAMPA data (Eqs. (22) and (23) and Figs. 7 and 8), BAMPA (Eqs. (24) and (25) and Figs. 9 and 10) and bio-mimetic PAMPA-PP-RF (Eq. (26) and Fig. (11)) indicating that PAMPA/BAMPA/bio-mimetic PAMPA-PP-RF assays constitute ideal high throughput alternatives to Caco-2 cell absorption/permeability models. Similarly, human intestinal absorption permeability has a good correlation with PAMPA (Eqs. (27)–(29) and Figs. 12, 13 and 14), BAMPA (Eqs. (30) and (31) and Figs. 15 and 16) and bio-mimetic PAMPA-PP-RF (Eq. (32) and Fig. 17) indicates that PAMPA/BAMPA/bio-mimetic PAMPA-PP-RF methods provide high throughput alternatives to computational (in-silico) approaches.

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