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*Research Article*

# Lipophilicity Descriptors Correlate Uniquely with Pharmacokinetic and Blood-Brain Barrier Penetration Parameters for Selected Antipsychotic Drugs

**Adeyemo, M.A., Balogun, F.J. and *Idowu, S.O.**

*Department of Pharmaceutical Chemistry, Faculty of Pharmacy, University of Ibadan, Ibadan, Nigeria.*

**ABSTRACT**

Lipophilicity is an important physicochemical parameter of biological relevance; although its in- vivo predictive capability is dependent on accuracy and reliability of platforms used for its determination. This work examines biomimetic attribute of isocratic chromatographic hydrophobicity index (ICHI), experimental logarithm of octanol – water partition coefficient (LogP) and some computed lipophilicity indices for eight (8) selected antipsychotic agents and their predictive capability in drug discovery. The retention behavior of 5 first-generation and 3 second-generation antipsychotics was determined on reversed-phase chromatographic platform using methanol-phosphate buffer (pH 6.8) mobile phase. The retardation factor obtained was transformed to Rm, and plotted against volume fraction of organic modifier in the mobile phase to generate linear graph whose x- intercept is ICHI. Experimental LogP values were curled from literature while computed LogP were obtained using respective software. The experimentally determined $LogP_{octanol/water}$ and ICHI were first correlated with index of brain permeability (BBB); before all lipophilicity indices were comparatively evaluated and correlated with in-vivo-normalized pharmacokinetic parameters curled from literature. ICHI gave better correlation with BBB index (r = 0.976) compared to Log Poctanol/water (r = 0.557). Comparative lipophilicity evaluation shows clustered pattern for second generation antipsychotics compared to first generation. In vivo correlation was poorer for the 8 drugs (r < 0.7), better with subset of phenothiazine homologues (r = 0.51 to 0.97). The ALogP, $LogP_{octanol/water}$, cLogP and ICHI gave highest correlation with the pharmacokinetic parameters. The biomimetic attributes of ICHI is better than for $LogP_{octanol/water}$ in predicting brain permeability, but lower for in-vivo pharmacokinetic prediction.

**Keywords:** *lipophilicity, hydrophobicity, biomimetic, pharmacokinetics, permeability, antipsychotics*

*Author for correspondence: Email*: olakunleid@yahoo.com;* Tel: *234-805-842-7072*

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## INTRODUCTION

Antipsychotic drugs are a major class of psychotherapeutic agents for ameliorating the symptoms of schizophrenia; which has been classified as one of the top leading 15 diseases responsible for global disease burden and disability (Global Disease Burden, 2017). A variety of typical antipsychotics i.e. first generation antipsychotics such as chlorpromazine, thioridazine etc. and atypical antipsychotics i.e. second generation antipsychotics like clozapine has been used to lessen the positive and negative symptoms of schizophrenia respectively through blockade of dopaminergic neurotransmission in the brain (Bhosale *et al*., 2014). In order to maximize their therapeutic impact, it is critical to control the secondary pharmacologic effect of these compounds and thus their Absorption, Distribution, Metabolism and Excretion (ADME) properties by optimizing lipophilicity. Lipophilicity is the key critical physico-chemical property that regulates ability of antipsychotics to cross the blood brain barrier via p-glycoprotein (Loscher, 2005); regulate their gastro-intestinal resorption and distribution via the albumin (Varshney *et al*., 2010) and also facilitates their diffusion and hence potency (Mikitsh and Chacko, 2014). This makes lipophilicity a vital metric in evaluating drug potential during preclinical trials (Peruskovic *et al*., 2014); and could guide optimization of potential lead molecules with antipsychotic activity (Nielsen and Nielsen, 2009; Hopkins *et al.*, 2014; Meanwell, 2016).

Lipophilicity is among the three critical physicochemical-biomimetic parameters, others being kinetic solubility and artificial membrane binding; for modeling in vivo drug disposition and quality of drug molecules (Gleeson *et al*., 2015; Valko, Teague and Pidgeon, 2017). Several lipophilicity models such as log octanol-water partition coefficient (log P); calculated log P and chromatographic hydrophobicity index are useful in biomimetic profiling, however with relative limitations. For instance log P and log D have been reported unreliable for lipophilicity profiling of poorly aqueous soluble compound (Young and Hill, 2010);

whereas chromatographic index e.g. from reversed phase high pressure liquid chromatographic platform gives reliable estimate (Valko *et al*., 1997), irrespective of the solubility status of the compound (Young *et al*., 2011). Likewise, calculated log P has been reported as "often inaccurate"; and can limit the potential of some promising compounds in drug discovery (Tsopelas *et al*., 2017; Giaginis *et al*., 2018).

In vitro-in vivo correlation (IVIVC) is a way of finding a good correlation between in vivo results and in vitro data for the purpose of optimizing human trials; predictive characterization of in vivo pharmacokinetics towards reducing need for elaborate bioequivalence studies (Chavda *et al*., 2016; Gomeni *et al*., 2019). Since lipophilicity has been reported as vital to determining in vivo pharmacokinetic properties (Constantinescu *et al*., 2019), a reliable model of lipophilicity measurement would guarantee a good in vitro-in vivo correlation (IVIVC); and is required for harnessing the biomimetic attributes of lipophilicity in future drug discovery research with minimal failure rate. The architecture of the blood-brain barrier (BBB), which regulates partitioning of drug into the brain is somewhat different from architecture of bio-membrane barrier bordering other drug compartments of the human body.

Morphologically, brain capillaries are like those found in other tissues, yet brain vessels are functionally bound to the other cells of the brain parenchyma. BBB consists of blood vessels built up by specialized endothelial cells (ECs), astrocytes, pericytes, and neuronal terminations. Astrocytes lay their end-feet over the continuous basal lamina and form a very restrictive barrier (Martas *et al*., 2014).

It is therefore reasonable to surmise that what constitute biomimetic attributes of lipophilicity estimating platforms will vary depending on the intended site of action of a given drug, since bio-membrane architecture in different compartments of the body is not uniform. Thus, we hypothesize that various lipophilicity descriptors will correlate differently with index of BBB penetration and general pharmacokinetic parameters.

This study therefore aims to first, comparatively evaluate lipophilicity characterization from these seven descriptors obtained from three different platforms i.e. octanol/water partitioning, chromatographic and computational algorithms; second, assess correlation of the two experimentally determined lipophilicity i.e. LogPoctanol/water and ICHI with computed blood brain barrier penetration; and third estimate the biomimetic attributes of all the lipophilicity indices in assessing in vivo pharmacokinetics of antipsychotic agents.

## MATERIALS AND METHODS

**Materials:** Methanol (Merck), Liquid paraffin, n-hexane (analar, BDH), distilled water, Phosphate buffer pH 6.8, Potassium dihydrogen phosphate (BDH, UK), Sodium hydroxide (Lobachemie, India), conical flasks, filter paper, pipette, measuring cylinder, volumetric flask, TLC tanks, precoated aluminum TLC plates GF254 (Merck, Germany), Model compounds: first generation antipsychotics: chlorpromazine (1), haloperidol (2), trifluoperazine (3), thioridazine (4), prochlorperazine (5); second generation antipsychotics: clozapine (6), olanzapine (7), risperidone (8) which are shown in Fig. 1.

**Equipment:** Ultraviolet lamp (254/365, Gallenkamp, U.K.), Drying oven (Astell Hearson PBS 040, England), Hot plate, Vacuum pump (Oerlikon Leybold, Germany), Analytical weighing balance (Mettler H80, UK), pH meter (PBS 040, England),

**Chromatographic evaluation:** Lipophilicity profiling of the model compounds was carried out by reversed phase thin layer chromatography on silica gel plates, 5 x 10 cm (Merck, Darmstadt Germany) coated with 5% liquid paraffin as stationary phase; and phosphate buffer (pH 6.8) as mobile phase. The solutions of the model compounds in methanol was spotted onto the plates, air dried and developed by ascending order. After development, the plates were air dried before the retardation factor (Rf) was determined. All measurements were conducted in duplicate and at room temperature ($33 \pm 2^{O}C$). The Rf was transformed to Rm, and plotted against the volume fraction of the organic modifier for the binary mobile phase to generate a linear relationship with the equation below:

$$Rm = Rmw + S\varphi$$

where φ is the volume of organic modifier fraction, S is the slope and indicates the rate of solute partitioning into the aqueous phase while Rmw is the intercept and value of Rm extrapolated to pure water (0 % methanol) as the mobile phase. The lipophilicity index used for this study is $\varphi_0$ known as isocratic chromatographic hydrophobicity index (ICHI); a derived parameter obtained when Rm equals zero i.e. x-intercept of the linear regression; and obtained from the equation:

$$\varphi 0 = -Rmw/S$$

**Calculations:** Apart from the logarithm of octanol-water partition coefficient ($logP_{octanol/water}$) curled from pubchem database (www.pubchem.ncbi.nlm.nih.gov), five other lipophilicity indices were calculated with two online-available software algorithms ALOPS 2.1 (www.vcclabs.org), SwissADME SwissADME (www.swissadme.ch) and a commercially available software Bio-Loom (www.biobyte.com). The index for blood-brain barrier (BBB) penetration was calculated with molsoft drug-likeness and molecular property prediction software (www.molsoft.com) while the Statistical evaluation of the chromatographic linear regression; correlation analysis of the two experimentally based lipophilicity index i.e. log Po/w and ICHI with BBB index; and of all the lipophilicity descriptors with the pharmacokinetic parameters were performed by GraphPad Prism Version 7 (SanDiego, CA).

**Pharmacokinetic Parameters:** The bioavailability of the drug depicted by Area under Curve (AUC∞), maximum plasma concentration (Cmax) and time for maximum concentration (Tmax) for single dose oral administration of the selected drugs were curled from literature. The AUC∞ and Cmax were dose-normalized to remove bias in the correlation analysis

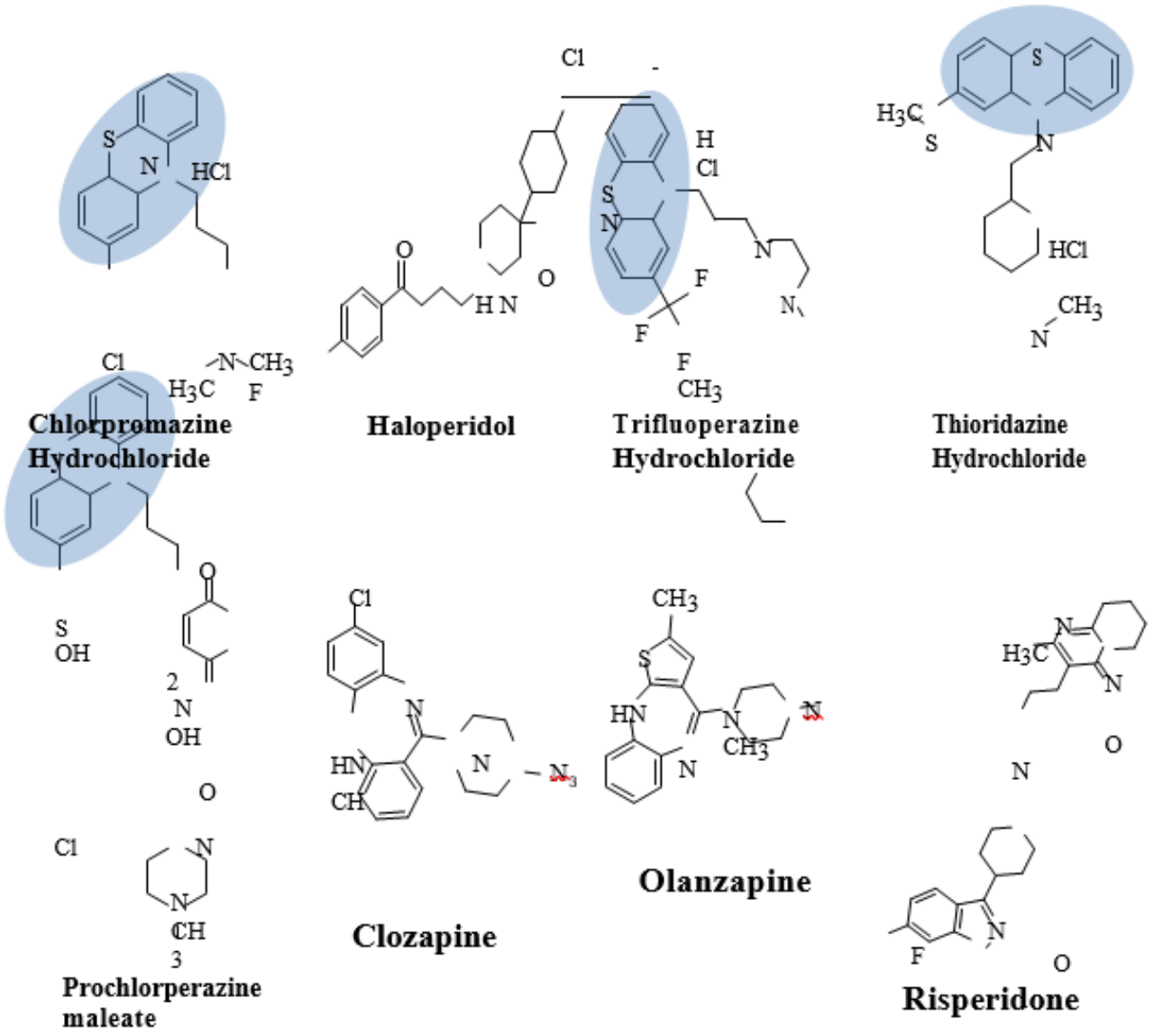


**Figure. 1:**
Chemical structures of antipsychotic compounds studied. The blue oval highlights the phenothiazine heterocycle in compounds 1, 3, 4 & 5.

## RESULTS AND DISCUSSION

**Chromatographic lipophilicity determination:** The linear regression of the Rm against the organic modifier fraction of the mobile phase (ϕ) for the antipsychotic agents is represented in Fig. 2. The regression parameters for this chromatographic evaluation of the compound's lipophilicity are also summarized in Table 1.

The Rm values decreased linearly with increasing concentration of the organic modifier in the mobile phase ("r" ranges from 0.967 to 0.997). The extrapolation of the values to the x-axis i.e. 50 % methanol fraction or Rm = zero; gives the φ0 which is known as the isocratic chromatographic hydrophobicity index (ICHI). ICHI reflects the relative partitioning between the hydrophobic stationary and hydrophilic mobile phase based on the equation:
Rm = Rmw + Sφ; where φ = organic modifier fraction,
at Rm = 0, φ0 = - Rmw/S

Correlation of the experimentally determined lipophilicity indices with computed Blood Brain

**Barrier penetration index:** The association between the experimental lipophilicity indices and the index of brain permeability is described in Fig. 3 below. Chromatographically generated ICHI gave higher positive correlation with BBB index (r = 0.976) while log $P_{octanol/water}$ gave a poorer positive correlation (0.557) reflecting that ICHI could predict therapeutic availability of these drugs in the central nervous system better (Morak-Mlodawska, Pluta and Jelen, 2020).

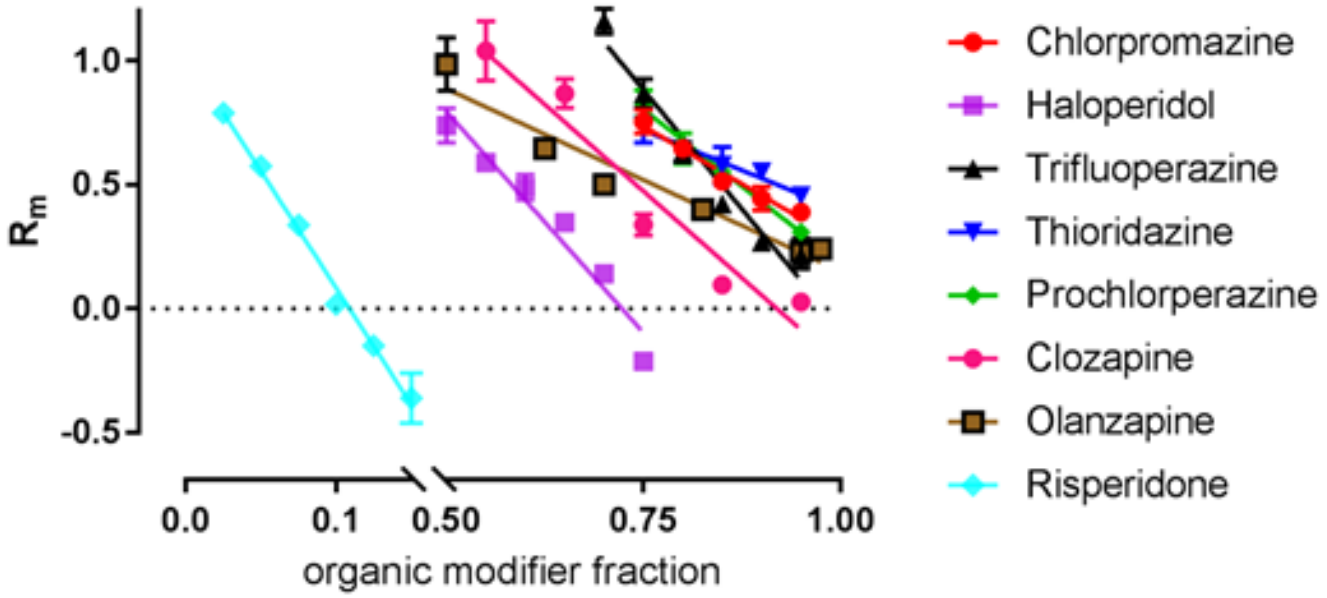


**Figure 2**
Linear regression of Rm versus volume fraction of the organic modifier (methanol)

**Table 1:**
Linear regression parameters for the chromatographic estimation of the antipsychotics' lipophilicity showing ICHI, r, $S_{y.x}$ residual and the equation Rm = Rmw + Sφ

| Compound name | ϕ0 (ICHI) | r | $S_{y.x}$ | Equation |
|---|---|---|---|---|
| Chlorpromazine | 1.140 | 0.986 | 0.0288 | Rm = -1.86ϕ + 2.13 |
| Haloperidol | 0.723 | 0.973 | 0.0881 | Rm = -3.57ϕ + 2.58 |
| Trifluoperazine | 0.979 | 0.979 | 0.0838 | Rm = -3.85ϕ + 3.77 |
| Thioridazine | 1.310 | 0.980 | 0.0242 | Rm = -1.3ϕ + 1.69 |
| Prochlorperazine | 1.070 | 0.994 | 0.0248 | Rm = -2.48ϕ + 2.66 |
| Clozapine | 0.919 | 0.967 | 0.1330 | Rm = -2.8ϕ + 2.57 |
| Olanzepine | 1.100 | 0.970 | 0.0778 | Rm = -1.47ϕ + 1.62 |
| Risperidone | 0.109 | 0.997 | 0.0385 | Rm = -9.41ϕ + 1.02 |

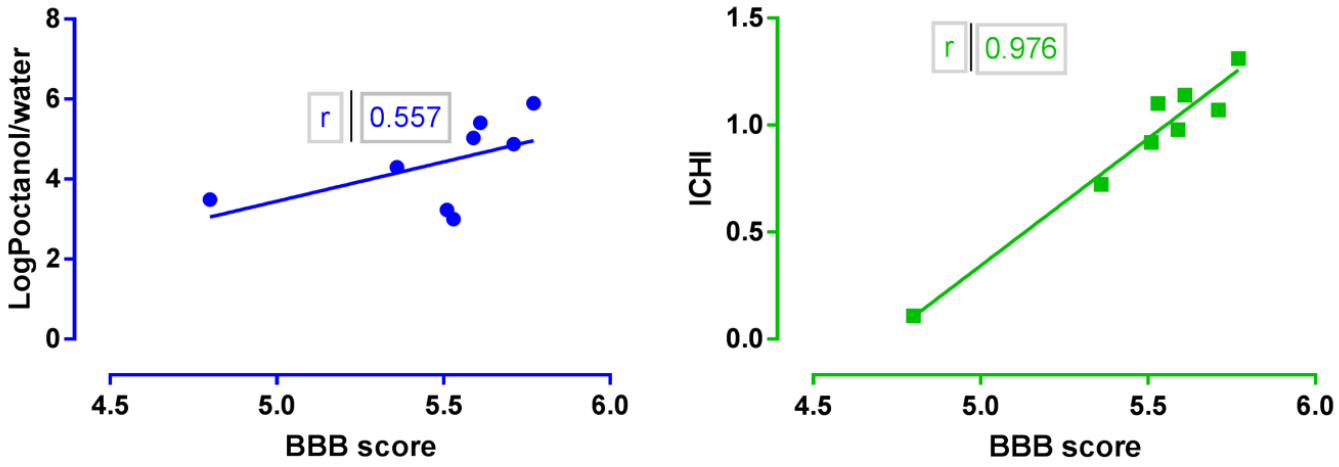


**Figure 3:**
Correlation of experimentally determined lipophilicity descriptors – (a) Log $P_{octanol\ water}$ and (b) ICHI on liquid paraffin film – with computed brain permeability index.

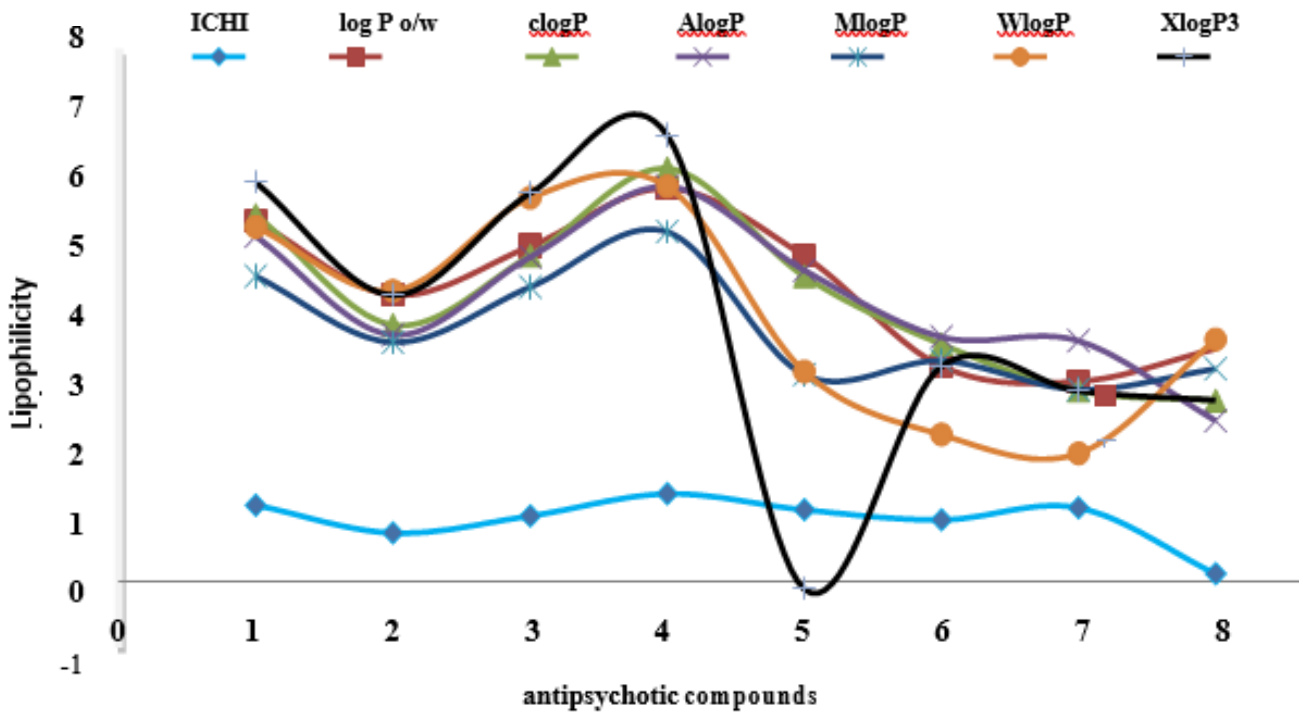


**Figure 4:**
Graphical visualization of the various lipophilicity indices for the selected antipsychotic compounds

**Table 3:**
Correlation of Pharmacokinetic Parameters with lipophilicity descriptors for the selected 8 compounds

| | ICHI | LogP | cLogP | ALogP | MLogP | WLogP | XLogP3 |
|---|---|---|---|---|---|---|---|
| AUC | -0.286 | -0.704 | -0.65 | -0.558 | -0.444 | -0.58 | -0.191 |
| Cmax | -0.564 | -0.24 | -0.282 | -0.4 | -0.0123 | -0.052 | -0.0277 |
| Tmax | 0.27 | -0.0171 | -0.061 | 0.0502 | -0.293 | -0.164 | -0.26 |

**Table 4:**
Correlation of Pharmacokinetic Parameters with lipophilicity descriptors for the compound 1 to 5

| | ICHI | LogP | cLogP | ALogP | MLogP | WLogP | XLogP3 |
|---|---|---|---|---|---|---|---|
| AUC | 0.633 | 0.726 | 0.739 | 0.694 | 0.752 | 0.563 | 0.543 |
| Cmax | 0.708 | 0.782 | 0.789 | 0.763 | 0.756 | 0.556 | 0.503 |
| Tmax | -0.889 | -0.964 | -0.964 | -0.971 | -0.873 | -0.745 | -0.598 |

**Evaluation of the different lipophilicity descriptors and their association with important Predicted ADME properties relevant for antipsychotic activity:** The seven different lipophilicity descriptors comprised of experimentally determined ICHI, LogP octanol-water and computer-generated logP values i.e. cLogP, ALogP, MLogP, WLogP and XLogP3 are summarized in Table 2 below.

The graphical visualization of the various lipophilicity descriptors is represented in Fig. 4. All the computed logP values for the compounds except compound 5 are closely clustered with the log $P_{octanol/water}$ unlike with ICHI (which is a non-logarithmic metric) except for compound 5 which could be due to presence of significant contribution of the oxygen hetero atom in the salt on the overall atomic effect captured by the XLogP3algorithm. However, all the lipophilicity indices were closely clustered from compound 6 to 8; which could be due to similarity in evaluation of the hydrogen bond acceptance capacity of these molecules by the different models (Šegan *et al*., 2017).

Correlation between the different lipophilicity indices and the experimentally determined pharmacokinetic data gave different pattern that showcases the variability in the model algorithms. All the lipophilicity descriptors gave poor negative correlation (i.e. $r < 0.9$) with the pharmacokinetic parameters except poor positive correlations between ICHI, ALogP with Tmax, and XLogP3 with Cmax (Table 3). However, by restricting the correlation analysis to compounds 1 to 5; which are the first generation antipsychotic agents led to significant increase in the correlation coefficient in which all the lipophilicity descriptors had a negative association with the Tmax (Table 4).

**Table 5:**
Correlation of Pharmacokinetic Parameters with lipophilicity descriptors for the compounds with phenothiazine ring (i.e. compounds 1, 3, 4, and 5)

| | ICHI | LogP | cLogP | ALogP | MLogP | WLogP | XLogP3 |
|---|---|---|---|---|---|---|---|
| AUC | 0.908 | 0.932 | 0.912 | 0.97 | 0.76 | 0.547 | 0.543 |
| Cmax | 0.895 | 0.909 | 0.888 | 0.954 | 0.733 | 0.521 | 0.51 |
| Tmax | -0.689 | -0.915 | -0.927 | -0.923 | -0.986 | -0.897 | -0.9 |

Finally, correlation within the phenothiazine compounds i.e. chlorpromazine, trifluoperazine, thioridazine and prochlorperazine gave the highest correlation coefficients in which ALogP had the best correlation with AUC and Cmax (r = 0.97 and 0.954) respectively while MLogP had best but negative correlation (r = -0.986) with the Tmax (Table 5). This underscores that determination of associations within homologous series give higher correlation coefficient since there is less variability in the topological and molecular features of the compounds (Hau *et al*., 1999; Xuefeng *et al*., 2006). Thus, establishing a functional relationship for predicting in vivo pharmacokinetics based on lipophilicity as the physicochemical parameter is weakened by the large chemical diversity of the selected library of compound s (Dambolena *et al*., 2016); which is reflected in the pattern of correlation seen from Table 3 to Table 5. The AUC and Cmax were positively correlated with the lipophilicity descriptors unlike the Tmax, since higher lipophilicity leads to faster absorption and peak level, hence lower Tmax (Ballas and Dinges, 2009; Paul, 2019). Overall, of the five computational algorithms, cLogP and ALogP gave relatively high correlation coefficient, comparable to the experimentally determined logP indices i.e. ICHI and LogP; and underscores why these two are among the most widely used algorithms for log P predictions (Souza *et al*., 2011).

In conclusion, lipophilicity plays an important role in the pharmacodynamics and pharmacokinetics of antipsychotic agents; and must be carefully determined with an appropriate model during drug discovery phase to optimize therapeutic value. The ICHI is a better predictor of blood brain permeability, which is a critical parameter in antipsychotic efficacy, compared to the LogPoctanol/water. In terms of in-vivo pharmacokinetic prediction, this study shows that prediction accuracy is improved within the homologous congeners; and the best predictive accuracy was observed with the following sequence: ALogP > LogP > cLogP > ICHI. Further study using larger sample size would prove the relative merits of these four parameters in predicting pharmacokinetic outcomes of antipsychotic drugs.

## REFERENCES


**Ballas C. and Dinges D. F. (2009):** Stimulant and Wake-Promoting Substances In: L. R. Squire (Eds.) Encyclopedia of Neuroscience pp. 419 – 424 United State: Academic Press

**Bhosale S.H., Kanhed A.M., Dash R.C., Suryawanshi M.R. and Mahadik K.R. (2014)**: Design, Synthesis, Pharma-cological Evaluation and Computational Studies of 1-(Biphenyl-4-yl)-2-[4- (substituted phenyl)-piperazin-1-yl] Etha- nones as Potential Antipsychotics *European Journal of Medicinal Chemistry*, 74, 358 - 365

**Chavda V. P., Shah D., Tandel H. and Soniwala M. (2016):** In Vitro-In Vivo correlation (IVIVC): A strategic tool in drug product development *Research and Reviews: A Journal of Drug Formulation, Development and Production* 3(3), 31 – 54

**Constantinescu T., Lungu C. N. and Lung I. (2019):** Lipophilicity as a central component of Drug-like properties of Chalchones and Flavonoid derivatives *Molecules* 24(1505), 1 – 11

**Dambolena J. S., Zunino M. P., Herrera J. M., Pizzolitto R. P., Areco V. A. and Zygadlo J. A. (2016)** Terpenes: Natural Products for Controlling Insects of Importance to Human Health – A Structure-Activity Relationship Study *Psyche* 2016, 1 – 17

**Giaginis C., Tsopelas F., Tsantili-Kakoulidou A. (2018)**: The impact of lipophilicity in drug discovery: Rapid measurements by means of Reversed-Phase HPLC *Methods in Molecular Biology* 1824, 217 – 228.

**Gleeson M. P., Leeson P. D., van der Waterbeemd H. (2015)**: Physicochemical properties and compound quality In: The handbook of Medicinal Chemistry. pp 1 – 31 x The Royal Society of Chemistry Publications: London.

**Global Disease Burden (2017)**: Global, regional, and national incidence, prevalence, and years lived with disability for 328 diseases and injuries for 195 countries, 1990-2016: a systematic analysis for the Global Burden of Disease Study 2016 *Global Health Metrics* 390(10100), P1211– 1259

**Gomeni R., Fang L. L., Bressolie-Gomeni F., Spencer T. J., Faraone S. V. and Babiskin A. (2019)**: A general framework for assessing in vitro/In vivo correlation as a tool for maximizing the benefit-risk ratio of a treatment using a convolution-based modeling approach. *CPT Pharmacometrics and System Pharmacology* 8, 97 – 106

**Hau K. M., Connell D. W. and Richardson B. J. (1999):** Quantitative Structure Activity relationships for Nasal pungency thresholds of volatile organic compounds *Toxicological Sciences* 47, 93 - 98

**Hill A. P. and Young R. J. (2010):** Getting physical in drug discovery: a contemporary perspective on solubility and hydrophobicity *Drug Discov. Today* 15, 648 – 655

**Hopkins, A. L., Keseru, G. M., Leeson, P. D., Rees, D. C. and Reynolds, C. H. (2014):** The role of Ligand Efficiency Measures in Drug Discovery *Nat. Rev. Drug Discov.*13(2), 105 – 121

**Leeson P.D. and Young, R. J. (2015)**: Molecular Property Design: Does Everyone Get It? *ACS Med. Chem. Lett.* 6 (2015), 722-725.

**Lobo S. (2020)** Is there enough focus on lipophilicity in drug discovery? *Expert Opin. Drug Discov.* 15(3), 261 – 263

**Loscher W. (2005)** Blood-brain barrier active efflux transporters: ATP-binding cassette gene family *NeuroRX* 2, 86 – 98

**Marta T., Eva R., Xian W., Monica B., Biuse G., Abel E., *et al*. (2014)** The blood-brain barrier: Structure, function and therapeutic approaches to cross it *Molecular Membrane Biology* 31(5): 152 – 167

**Meanwell N. A. (2011):** Improving drug candidates by design: a focus on physicochemical properties as a means of improving compound disposition and safety *Chem. Res. Toxicol*. 24, 1420 – 1456.

**Meanwell N. A. (2016):** Improving drug design: An update on Recent Applications of Efficiency metrics, strategies for replacing problematic elements, and compounds in nontraditional drug space Chem. Res. Toxicol. 29(4), 564 – 616 Available online at: https://doi.org/10.1016/S1567-7192(00)80015-7

**Mikitsh J. L. and Chacko A. (2014)**: Pathways for small molecule delivery to the Central Nervous
System across the blood-brain barrier Perspectives in Medicinal Chemistry 6, 11–24 Available online at: https://doi.org/10.4137/PMC.S13384

**Morak-Mlodawska B., Pluta K and Jelen M. (2020)**: Evaluation of the lipophilicity of new anticancer 1,2,3-Triazole-dipyridothiazine hybrids using RPTLC and different computational methods Processes 8(858), 1 – 11 Available online at: https://doi.org/10.3390/pr8070858

**Nielsen R. E. and Nielsen J. (2009):** Antipsychotic Drug Treatment for Patients with Schizophrenia: Theoretical Background, Clinical Considerations and Patient Preferences Clinical Medicine. Therapeutics, Volume: 1: 1053-1068

**Paul A. (2019)**: Drug Absorption and Bioavailability. In: Raj G., Raveendran R. (eds) Introduction to Basics of Pharmacology and Toxicology. Springer Publishers: Singapore.

**Peruskovic D. S., Stevanovic N. R., Lolic A. D., Nikolic M. R. and Baosic R. M. (2014):** Quantitative Structure-Activity Relationship study of some antipsychotics by Multiple Linear Regression Am. J. Analyt Chem. 5, 335 – 342 Available online at: http://dx.doi.org/10.4236/ajac.2014.55041

**Šegan S., Božinovi´c N., Opsenica I., Andri´c F. (2017):** Consensus-based comparison of chromatographic and computationally estimated lipophilicity of benzothiepino[3,2-c]pyridine derivatives as potential antifungal drugs Journal of Separation Science 40, 2089–2096.

**Souza E. S., Zaramello L., Kuhnen C. A. and Junkes B. D. (2011)** Coefficient for aliphatic organic compounds using semi-empirical electrotopological index Int. J. Mol. Sci. 12(10), 7250– 7264 Available online at: https://doi.org/10.3390/ijms12107250

**Tsopelas F., Giaginis C., Tsantili-Kakoulidou A. (2017)** Lipophilicity and biomimetic properties to support drug discovery Expert Opin. Discov. 12(9), 885 – 896.

**Valko K., Bevan C. and Reynolds D. (1997)** Chromatographic Hydrophobicity Index by Fast- gradient RP-HPLC: A high-throughput alternative to log P/log D Analytical Chemistry 69, 2022– 2029

**Valko K., Teague S. and Pidgeon C. (2017)**: In vitro membrane binding and protein binding (IAM MB/PB technology) to estimate in vivo distribution: applications in early drug discovery ADMET 5(1), 14 - 38

**Xuefeng L., Gaoyong Z., Jinfeng D., Xiaohai Z., Xiaoci Y. and Mingdao L. (2006)** Correlation of critical micelle concentration of sodium alkyl benzenesulfonates with molecular descriptors. Wuhan University Journal of Natural Science 11, 409 – 414

**Young R. J., Green D. V., Luscombe C. N. and Hill A. P. (2011)** Getting physical in drug discovery II: the impact of chromatographic hydrophobicity measurements and aromaticity Drug Discov. Today 16, 822 – 830

**Varshney A., Sen P., Ahmad E., Rehan M., Subbarao N. and Khan R. H. (2010)** Ligand Binding Strategies of Human Serum Albumin: How can the cargo be utilized? Chirality 22, 77 – 87