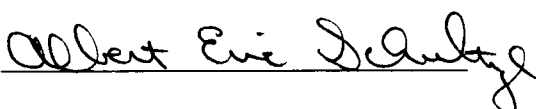


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I am submitting herewith a thesis written by Tonya Louise Ailey Carver entitled "Structure-Toxicity Relationships for Aminoalkanols: A Comparison with Alkanols and Alkanamines". I have examined the final copy of this thesis for form and content and recommend that it be accepted in partial fulfillment of the requirements for the degree of Master of Science, with a major in Life Sciences.


T. Wayne Schultz, Major Professor

We have read this thesis
and recommend its acceptance:



Joyce I. Merryman

Accepted for the Council:


Associate Vice Chancellor
and Dean of The Graduate School

**STRUCTURE-TOXICITY RELATIONSHIPS FOR AMINOALKANOLS:
A COMPARISON WITH ALKANOLS AND ALKANAMINES**

A Thesis

Presented for the

Master of Science Degree

The University of Tennessee, Knoxville

Tonya Louise Ailey Carver

December, 1995

DEDICATION

This thesis is dedicated to my husband, Ethan Allan Carver, who has encouraged and silently endured my pursuit of a higher education.

ACKNOWLEDGMENTS

I would like to thank my major professor, Dr. Terry Schultz, for his assistance in planning my program and in setting the objectives of my thesis research. His guidance and encouragement throughout this investigation were much appreciated. I would also like to thank my committee members, Drs. Joyce Merryman and Eric Schultze, for their advice and review of the thesis. I am especially grateful to Mr. Glendon Sinks for his technical assistance, and patience with me in the laboratory. Gratitude is expressed to Ms. Wanda Aycock, whose word processing expertise facilitated the final draft of this thesis, and Ms. Kim Cline, who aided with the generation of the Figures. I also would like to acknowledge Lockheed Martin Energy Systems for financing my continuing education and Stonewall Jackson Van Hook III for his support of my efforts to continue my education.

ABSTRACT

The relative toxicity ($\log \text{IGC}_{50}^{-1}$) of 35 selected alkanamines and aminoalkanols was evaluated in the static *Tetrahymena pyriformis* population growth assay. Excess toxicity, indicated by potency greater than predicted for alkanols, was associated with both classes of chemicals. Moreover, the aminoalkanols were found to be more toxic than corresponding alkanamines. A high quality 1-octanol/water partition coefficient ($\log K_{\text{OW}}$) dependent quantitative structure-activity relationship (QSAR), $\log \text{IGC}_{50}^{-1} = 0.72 (\log K_{\text{OW}}) - 1.32$; $r^2 = 0.939$, was developed for alkanamines. This QSAR represented the amine narcosis mechanism of toxic action. Several structure-toxicity features were observed for the aminoalkanols. These included 2-amino-1-hydroxy derivatives being more toxic than corresponding derivatives where the amino and hydroxy moieties were separated by methylene groups, and hydrocarbon branching next to the amino moiety resulting in decreased toxicity. Regression analysis of $\log K_{\text{OW}}$ versus the $\log \text{IGC}_{50}^{-1}$ for selected aminoalkanols yielded the model, $\log \text{IGC}_{50}^{-1} = 0.17 (\log K_{\text{OW}}) - 0.70$; $r^2 = 0.833$.

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LIST OF ABBREVIATIONS

DMSO, Dimethyl Sulfoxide

$\log \text{IGC}_{50}^{-1}$, Relative Toxicity

$\log K_{ow}$, 1-octanol/water Partition Coefficient

mg/L, Milligrams Per Liter

mM, Millimoles Per Liter

$\text{Pr} > f$, Level Of Significance

QSAR, Quantitative Structure-Activity Relationships

r^2 , Coefficient Of Determination

s, Root Of The Mean Square For Error

SAS, Statistical Analysis System

T_e , Excess Toxicity

1. INTRODUCTION

The most robust toxicological quantitative structure-activity relationships (QSARs) have been those developed on the basis of a common mechanism of toxic action (Bradbury, 1994). The selection of the correct mechanism of action is no easy task. The interaction of toxicants with living systems depends upon a plethora of physical and/or chemical properties (Nendza and Russom, 1991) and often involves a poorly defined molecular site of action. When the mechanism of action is unknown, the development of structure-toxicity relationships is often begun by analysis of a homologous series of chemicals (e.g., methyl, ethyl, propyl, etc.). This conservative approach places limitations on molecular structure, thereby enhancing the probability of a uniform mechanism of action. It is, in large, part the basis by which the U.S. Environmental Protection Agency estimates toxicity of organic chemicals to aquatic organisms (Clements et al., 1994) and is the essence of the ECOSAR computer program (Clements and Nabholz, 1994). While this approach provides a working basis to explore toxicity, it does not address the question about QSAR selection when a chemical has more than one model-defining substituent. For instance, Schultz et al. (1992) noted that the toxic mechanism of action for a given "phenol derivative" may be a polar narcotic (e.g., phenol), a respiratory uncoupler (e.g., pentachlorophenol), or a precursor to a redox cyclor (e.g., hydroquinone).

Previous studies have used 1-octanol/water partition coefficient ($\log K_{OW}$) dependent QSAR as a means of predicting toxic potency A ($\log IGC_{50}^{-1}$), measured as *Tetrahymena pyriformis* population growth inhibition by neutral organics or nonreactive, nonpolar narcotics (Schultz and Tichy, 1993). However, a variety of aliphatic chemicals have been shown to be more toxic than predicted by the baseline neutral organic model. Such chemicals are deemed bioreactive (Schultz et al., 1995b).

Bioreactive chemicals, especially those with low hydrophobicity, exhibit excess toxicity (T_e). Excess toxicity has been defined as a ratio of predicted-to-observed toxicities (Lipnick et al. 1987) > 2.0 where predicted toxicity is calculated with the baseline or nonpolar narcosis QSAR. Bioreactive chemicals often cause problems in the application of QSAR methods for evaluating the effects and hazard of chemicals.

The toxicity of saturated aliphatic alcohols has been examined in a wide variety of test systems (see Hansch et al., 1989 for a review). In addition, such alcohols were part of a potency comparison study of aquatic test systems by Schultz et al. (1990b). Saturated aliphatic alcohols are considered "classical" neutral organic toxicants (Jaworska and Schultz, 1993). They elicit the baseline nonpolar narcosis mechanism of toxic action (Schultz, 1989). This mechanism of action has recently been reviewed (van Wezel and Opperhuizen, 1995).

The toxicity of aliphatic amines has been examined in a number of aquatic assays (Newsome et al., 1991; 1993a; 1993b; Schultz et al., 1991; 1995a). While the molecular site of action has yet to be identified, amine narcosis (Schultz et al., 1991) is considered a separate mechanism of toxic action. However, the results of the studies of amine toxicity have been clouded by a number of predicaments. In some cases both aliphatic and aromatic amines were included in the same analyses. Other studies evaluated simultaneously primary, secondary and tertiary amines, or, in other cases, hydroxyl-substituted amines were included in the studies.

In an effort to resolve many of these issues, the present investigation utilized a strict chemical class approach. The test system used was the rapid, reliable, and inexpensive *Tetrahymena pyriformis* population growth assay. The aims of this study were to: (1) determine the relative toxicity to a group of aliphatic aminoalkanols and (2) compare

these observed toxicities to those predicted by the alkanol narcosis QSAR and the alkanamine narcosis QSAR.

2. MATERIALS AND METHODS

Tetrahymena Assay

Relative toxicity was determined by utilizing the *Tetrahymena pyriformis* population growth impairment system (Schultz et al., 1990a). The ciliates were grown in axenic cultures and evaluated under static conditions. The endpoint (population density) was evaluated after approximately 44 hrs (i.e., 10 generations of the control cultures) of incubation at $27 \pm 1^\circ\text{C}$ by spectrophotometric measurement @ 540nm. Assays were conducted in 250ml Erlenmeyer flasks containing 50 ml of proteose peptone-based media. After conducting range-finding tests, each chemical was tested three times in duplicate. Each definitive test consisted of a minimum six-step graded concentration series. Flasks were prepared by adding a designated amount of test chemical stock solution to the media. Cultures were inoculated with 150 μl of 2-day (i.e., late log-growth-phase) *Tetrahymena pyriformis* culture. The graded series selected covered a range from no effect to complete inhibition of growth, with at least three partial responses. Only tests with control cultures in late log-phase growth with absorbance units between 0.6 and 0.9 were considered for evaluation.

Chemicals Tested

Twenty-two chemicals consisted of 16 straight-chain and branched aliphatic primary aminoalkanols, 2 straight-chain aliphatic secondary aminoalkanols, 1 aliphatic secondary amine, and 1 aliphatic tertiary amine were assessed. The chemicals were obtained commercially (Aldrich Chemical Co., Milwaukee, Wisconsin, USA; MTM Research Chemicals Lancaster Synthesis Inc., Windham, New Hampshire, USA; or ICN Biomedicals Inc., Irvine, California, USA) at sufficient purity (>95%) that made further

purification unnecessary. Stock solutions were prepared by weighing an appropriate amount of chemical and diluting it with dimethyl sulfoxide (DMSO) to obtain concentrations of 2.5, 5, 10, 25, or 50 parts-per-million. In all cases, the volume of stock solution added did not exceed 0.35 ml. This amount of DMSO does not alter *Tetrahymena* population growth kinetics (Schultz and Cajina-Quezada, 1982).

Analyses

Population growth inhibition was computed by the probit procedure of Statistical Analysis System (SAS). The IGC₅₀ (50% inhibitory growth concentration) and the 95% fiducial interval were determined for each chemical by measuring the percent control-normalized absorbance (Y) and the toxicant concentration in mg/l (X).

Tetrahymena population growth inhibition data for 17 aliphatic primary amines were secured from Schultz et al. (1991).

The log K_{OW} value of each compound was obtained as either a measured value from Hansch et al. (1995) or as estimated values determined by the fragment method (Hansch and Leo, 1979).

Excess toxicity (T_e) evaluations were conducted for each chemical by calculating the ratio of predicted toxicity to observed toxicity (Lipnick et al., 1987). Experimental toxicity was measured as the IGC₅₀ in mM. Predicted toxicity was determined from the relationship,

$$\log \text{IGC}_{50}^{-1} = 0.80 (\log K_{OW}) - 2.04;$$

$$n = 34, r^2 = 0.982, s = 0.171, f = 1744.53;$$

$$\text{Pr} > f = 0.0001 \quad [1].$$

developed by Schultz and Tichy (1993) from data on aliphatic alcohols.

QSARs were developed by modeling the data using linear regression analysis (general linear model procedure) of SAS. Log IGC₅₀⁻¹ in mM was the dependent variable while log K_{OW} was used as the independent variable. Model adequacy was quantified with the r² value (coefficient of determination). The s value (root of the mean square for error) in addition to the f value and the probability greater than the f value (Pr > f) of each model were also noted.

3. RESULTS

Relative Toxicity

The toxicity and hydrophobicity data for the 37 chemicals used in these analyses are listed in Table I. The log K_{OW} values uniformly covered a range of -1.31 to 4.88. Observed toxicity varied over more than three orders of magnitude. Undecylamine demonstrated the least toxicity while ethanolamine was shown to have the highest toxicity. One compound, dodecylamine was not toxic at saturation.

Excess Toxicity

A scatterplot of the predicted toxicity versus the observed toxicity for the alkanamines and the aminoalkanols is presented in Figure 1. Excess toxicity (T_e) is defined as the ratio of predicted-to-observed toxicities. This is basically a diagram showing that excess toxicity is associated in both groups of compounds. The aminoalkanols are observed typically to have higher T_e values with ethanolamineamine being the most toxic. Excess toxicity tends to be inversely related to hydrophobicity, and as hydrophobicity decreases, there is an increase in toxicity.

Structure-Toxicity Relationships

Data for the secondary and tertiary amines tested were combined with toxicity data for the aliphatic primary amines reported by Schultz et al. (1991). A plot of log K_{OW} versus the log IGC_{50}^{-1} for these chemicals (compounds 1-19) is presented in Figure 2. Regression analysis of the data for the primary amines resulted in the equation,

$$\log IGC_{50}^{-1} = 0.72 (\log K_{OW}) - 1.32;$$

Table 1. Hydrophobicity and toxicity data for selected alkanamines and aminoalkanols

	Chemical	CAS Number ^a	Log K _{ow} ^b	log IGC ₅₀ ⁻¹ (mM)	Observed Potency (mM)	Predicted Potency ^c (mM)	Excess Toxicity ^d
1	isopropylamine	75-31-0	0.26 ^e	-0.88	7.58	67.92	8.96
2	(tert)butylamine	75-64-9	0.40 ^e	-0.90	7.89	52.48	6.65
3	propylamine	107-10-8	0.48 ^e	-0.71 ^g	5.10	45.29	8.88
4	(+/-)secbutyl amine	33966-50-6	0.63 ^f	-0.67 ^g	4.69	34.36	7.33
5	butylamine	109-73-9	0.97 ^e	-0.57 ^g	3.75	18.37	4.90
6	(tert)amylamine	594-39-8	1.06 ^f	-0.70	4.99	15.56	3.12
7	isoamylamine	107-85-7	1.49 ^f	-0.58	3.78	7.05	1.87
8	amylamine	110-58-7	1.49 ^e	-0.48 ^g	3.05	7.05	2.31
9	2-methylbutyl- amine	96-15-1	1.49 ^f	-0.48	3.00	7.05	2.35
10	3-phenyl- 1-propylamine	2038-57-5	1.96 ^f	0.28	0.53	2.96	5.59
11	hexylamine	111-26-2	2.06 ^e	-0.22 ^g	1.66	2.47	1.49
12	4-phenyl -1-butylamine	13214-66-9	2.49 ^f	0.62	0.24	1.12	4.70
13	heptylamine	111-68-2	2.57 ^e	0.21 ^g	0.62	0.96	1.57
14	octylamine	111-86-4	2.90 ^e	0.61 ^g	0.24	0.52	2.15
15	decylamine	2016-57-1	4.22 ^f	2.06 ^g	0.01	0.05	5.24
16	undecylamine	7307-55-3	4.88 ^f	2.33 ^g	0.00	0.01	2.91
17	dodecylamine	124-22-1	5.54 ^f	NTAS ^h	----	----	----
18	n-methyl- butylamine	110-69-9	1.33 ^e	-0.68	4.77	9.46	1.98
19	n,n-dimethyl- ethylamine	598-56-1	1.65 ^e	-0.91	8.10	5.25	0.648

Table I. (continued)

	Chemical	CAS Number ^a	Log K _{ow} ^b	log IGC ₅₀ ⁻¹ (mM)	Observed Potency (mM)	Predicted Potency ^c (mM)	Excess Toxicity ^d
20	ethanolamine	141-43-5	-1.31 ^e	-1.01	10.18	1224.62	120.28
21	dl-1-amino- 2-propanol	78-96-6	-0.96 ^e	-0.93	8.47	642.69	75.88
22	2-amino- 1-propanol	6168-72-5	-0.96 ^f	-0.82	6.55	642.69	98.13
23	3-amino- 1-propanol	156-87-6	-1.12 ^e	-0.81	6.41	862.98	134.59
24	(+/-)2-amino -1-butanol	13054-87-0	-0.30 ^f	-0.72	5.22	190.55	36.48
25	2-amino-2-methy- 1-propanol	124-68-5	-0.30 ^f	-0.96	9.09	190.55	20.96
26	4-amino- 1-butanol	13325-10-5	-0.46 ^e	-0.98	9.45	255.86	27.089
27	dl-2-amino-1 -pentanol	4146-04-7	0.36 ^f	-0.67	4.70	56.49	12.03
28	-dl-2-amino-3-methyl- 1-butanol	16369-05-4	0.36 ^f	-0.585	3.85	56.49	14.68
29	3-amino-2,2-dimethyl- 1-propanol	26734-09-8	0.36 ^f	-0.92	8.41	56.49	6.72
30	5-amino- 1-pentanol	2508-29-4	0.20 ^f	-0.99	9.69	31.92	7.83
31	dl-2-amino- 1-hexanol	5665-74-7	1.02 ^f	-0.58	3.84	16.75	4.36
32	(s)-tert- leucinol	112245-13-3	1.02 ^f	-0.72	5.22	16.75	3.21
33	(s)-(+)-leucinol	7533-40-6	1.02 ^f	-0.62	4.16	16.75	4.03
34	(s)-(+)- isoleucinol	24629-25-2	1.02 ^f	-0.66	4.56	16.75	3.67

Table I. (continued)

	Chemical	CAS Number ^a	Log K _{ow} ^b	log IGC ₅₀ ⁻¹ (mM)	Observed Potency (mM)	Predicted Potency ^c (mM)	Excess Toxicity ^d
35	6-amino- 1-hexanol	4048-33-3	1.33 ^f	-0.96	9.08	22.49	2.477
36	2-(methylamino) ethanol	109-83-1	-0.94 ^e	-0.89	7.80	619.44	79.415
37	2-(ethylamino) ethanol	110-73-6	-0.48 ^f	-0.76	5.80	265.46	45.74

Notes:

^a Chemical Abstract Service registry number.

^b 1-octanol/water partition coefficient.

^c Predicted from log K_{ow} values and the QSAR, $\log \text{IGC}_{50}^{-1} = 0.80 (\log K_{ow}) - 2.04$.

^d Excess toxicity parameter defined as predicted potency/observed potency.

^e Measured values.

^f Estimated values.

^g From Schultz et al. (1991).

^h Not Toxic At Saturation.

$$n = 16, r^2 = 0.939, s = 0.253, f = 217;$$

$$\text{Pr} > f = 0.0001 \quad [2].$$

An examination of Figure 2 reveals the secondary amine (compound 18) is modeled adequately by Eq. [2]. This is confirmed in Figure.2 where the 30 amines fall below baseline. However, the tertiary amine (compound 19) is less toxic than predicted by Eq. [2]. A comparison of the observed potency and potency predicted by Eq. [1] (see Table I) shows the tertiary amine models well as a nonpolar narcotic.

A comparison of Eq. [1] and [2] showed while the difference in slopes is minimal, the difference in intercept is better than 0.7 of a log unit with each primary amine being more toxic than its corresponding alcohol.

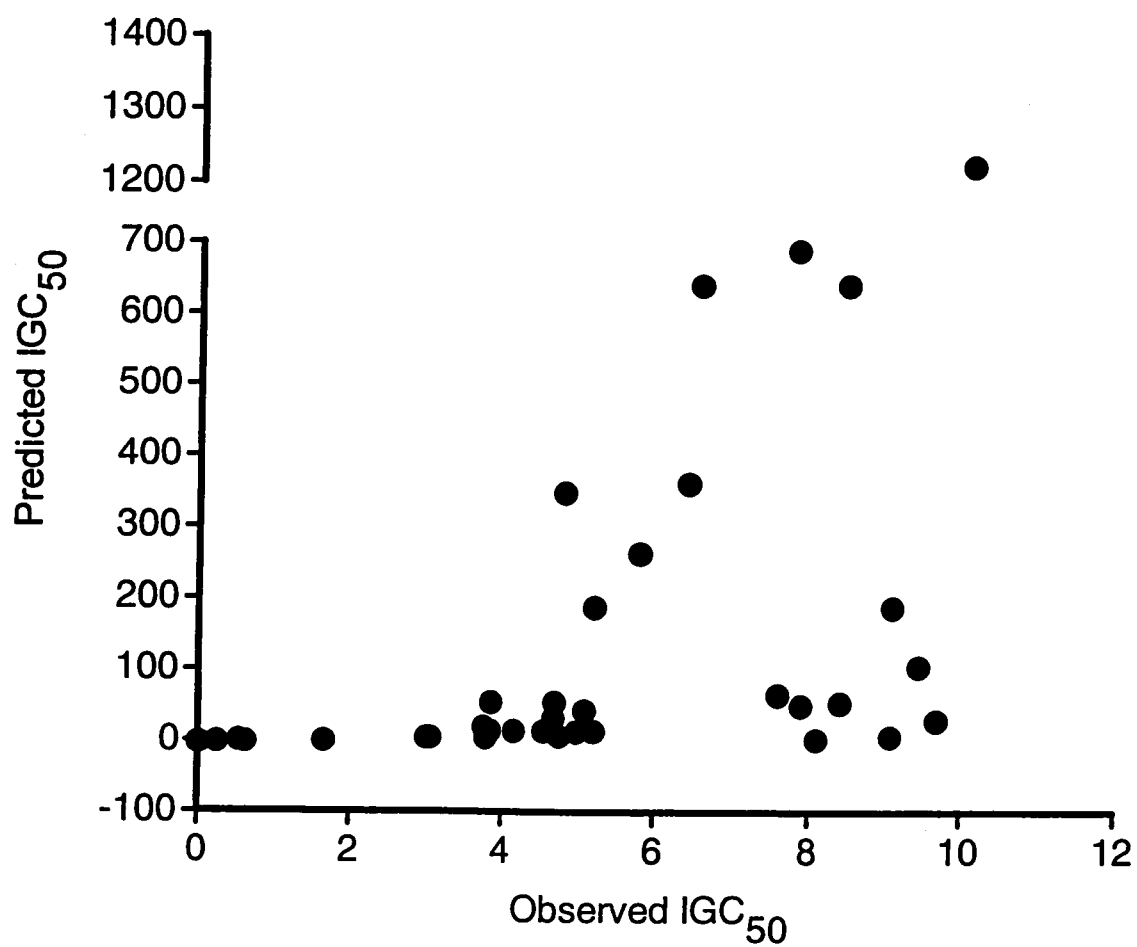


Figure 1. A scatter plot of predicted versus observed toxicities for Aminoalkanols and Alkanamines.

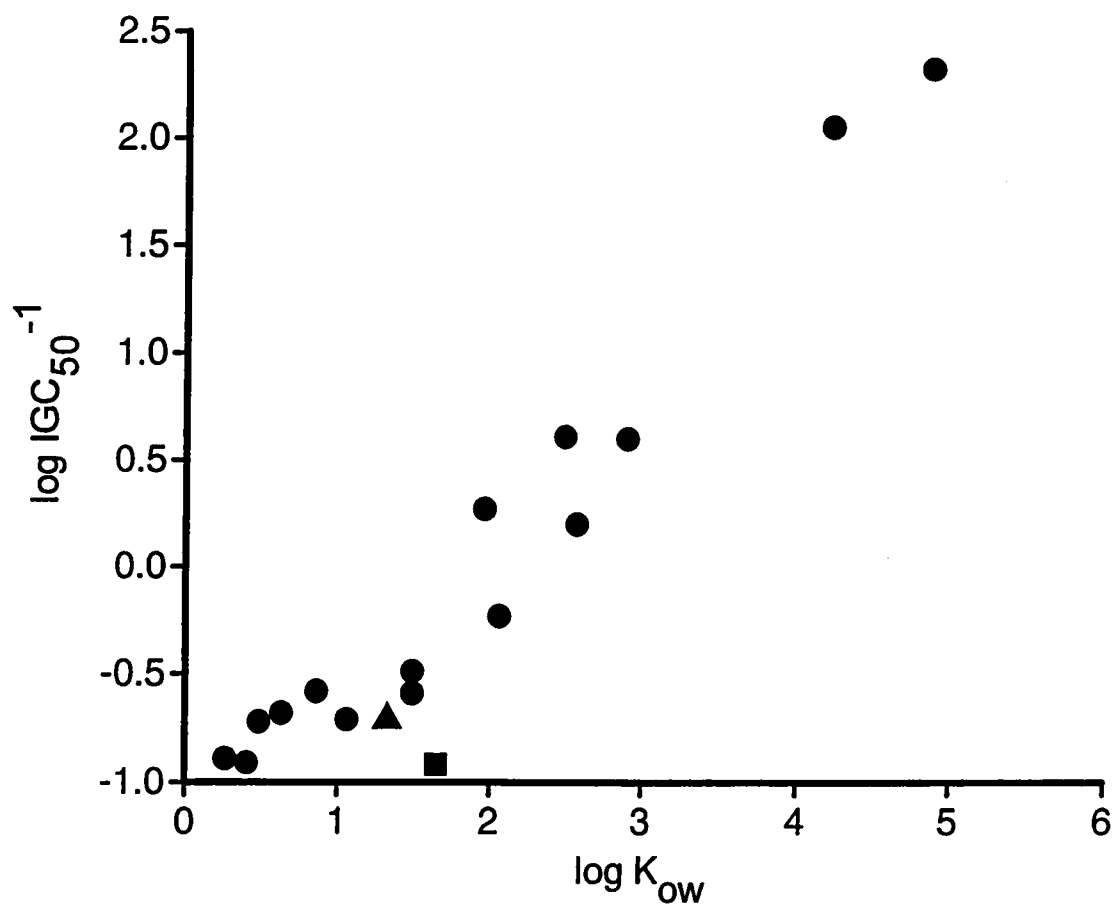


Figure 2. A scatter plot of toxicity (log IGC₅₀⁻¹) versus hydrophobicity (log K_{ow}) for the alkanamines.

A plot of $\log \text{IGC}_{50}^{-1}$ versus the $\log K_{\text{OW}}$ for the aminoalkanols is presented in Figure 3. While Figure 3 demonstrates a general linear relationship between toxic potency and hydrophobicity, several structure-toxicity features were observed. A comparison of toxic potency for the 2-amino-1-hydroxyl derivatives with their corresponding derivative where the amino and hydroxyl moieties were separated by methylene groups (i.e., compounds 24, 26, 27, 30, 31, 35) showed consistently the 2-amino-1-hydroxyl derivatives to be the more toxic. A comparison of potencies between compound 27, 2-amino-1-pentanol, and compounds 28 and 29, as well as compound 31, 2-amino-1-hexanol, and compounds 32-34 demonstrate that hydrocarbon branching near the amino moiety results in a decrease in toxicity. Secondary aminoalkanols (compounds 36 and 37) plotted in the linear aspect of the concentration-response graph. Regression analysis of $\log K_{\text{OW}}$ versus the $\log \text{IGC}_{50}^{-1}$ for the aminoalkanols yielded the model,

$$\log \text{IGC}_{50}^{-1} = 0.17 (\log K_{\text{OW}}) - 0.70;$$

$$n = 10, r^2 = 0.833, s = 0.061, f = 39.8;$$

$$\text{Pr} > f = 0.0002 \quad [3].$$

Attempts to improve the predictability of Eq. [3] by the addition of a molecular orbital energy descriptors was unsuccessful.

The observed toxicity (data not reported) of tertiary aminoalkanols was below that predicted by the baseline QSAR (i.e., Eq. [1]). These chemicals were removed from further consideration.

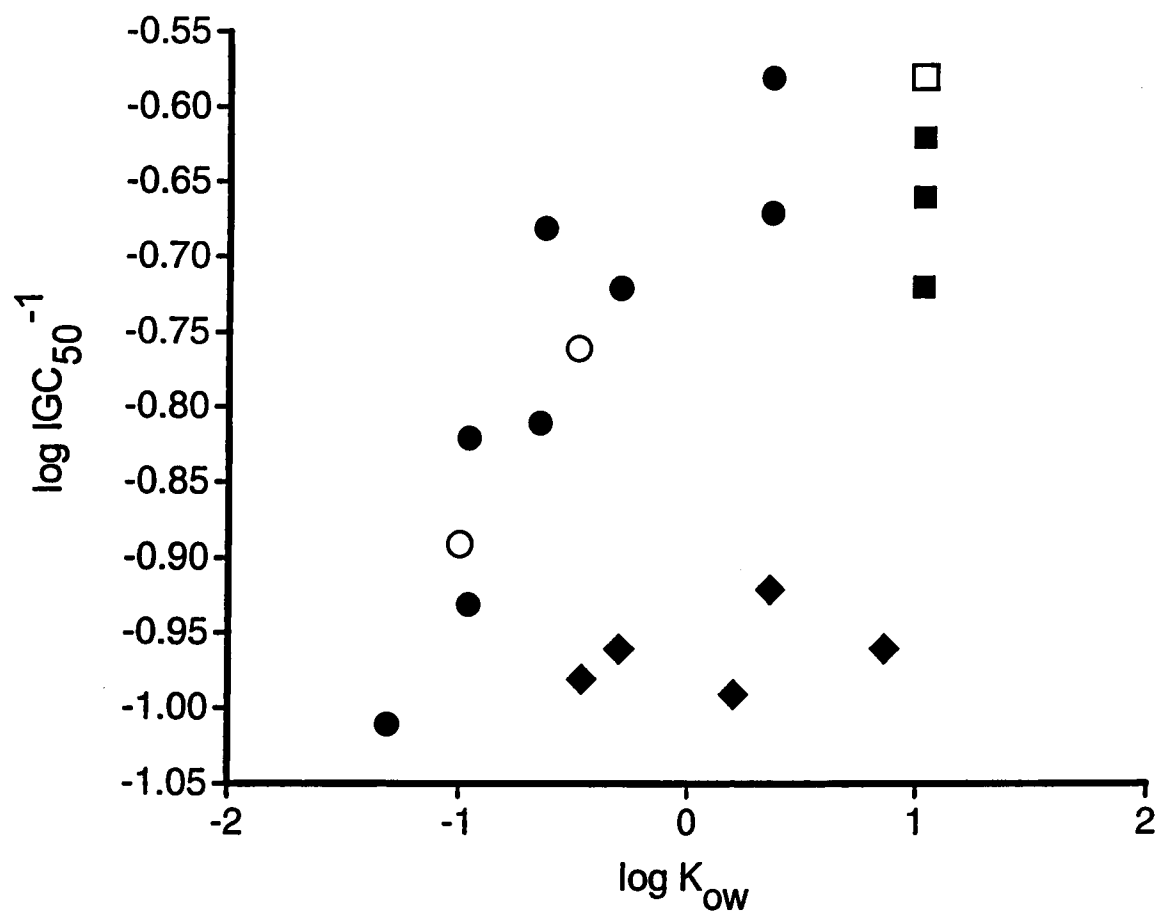


Figure 3. A scatter plot of toxicity ($\log \text{IGC}_{50}^{-1}$) versus hydrophobicity ($\log K_{ow}$) for aminoalkanols.

4. DISCUSSION

The most stalwart structure-toxicity relationships have been those developed for aquatic systems and based on a common mechanism of action. Since activity of chemical toxicants towards living systems depends upon a combination of physical and/or chemical interactions, molecular mechanisms of action are most often ill defined. Despite a general lack of knowledge concerning the molecular basis for toxicity, several methods have been developed to group toxicants by mechanism of action. These include: (A) membership in the same high quality QSAR based on experimental data; (Schultz et al., 1990); (B) strict concentration addition in joint toxic action models (Broderius and Kahl, 1985); (C) similar whole animal behavioral and cardiovascular-biochemical response sets (McKim et al., 1987), and (D) similar volume fraction analyses values (Jaworska and Schultz, 1993).

When the mechanism of action is unknown, the development of structure-toxicity relationships is often initiated via examination of a homologous series of chemical derivatives (e.g., methyl, ethyl, propyl, butyl, etc.). This conservative approach places limitations on molecular structure, and thereby enhances the probability of a uniform mechanism of action. The evaluation of toxicity by chemical class, especially for congeneric series, has allowed for the identification of toxicophores and the elucidation of trends in toxicity (Lipnick, 1991; Jackel and Nendza, 1994).

Quantitative structure-activity relationships based on $\log K_{OW}$ values can be used to model toxicity, with an increase in hydrophobicity resulting in an increase in potency. Congeneric series not having statistically different slopes or intercepts for their respective $\log K_{OW}$ -dependent toxicity regression equations are assumed to have the same mechanism of toxic action. Moreover, their toxicity can be predicted from the same QSAR. This approach has been used successfully to build QSARs for several

mechanisms of toxicity. For example, it has lead to, among others, QSAR models for neutral organics including saturated aliphatic alcohols, ketones, ethers and aromatic hydrocarbons. The aliphatic alcohol model (Eq. [1]) developed by Schultz and Tichy (1993) can be considered a baseline neutral organic model for the *Tetrahymena* population growth endpoint.

The majority of industrial organic chemicals act via the nonpolar narcosis mode of toxic action (Bradbury and Lipnick, 1990). Nonpolar narcosis (Ferguson, 1939) is a nonspecific and reversible mode of action resulting in the arresting of general cytoplasmic activity (Veith and Broderius, 1990). Nonpolar narcotics yield simple log K_{OW} -dependent QSARs which are extremely consistent regardless of the test system (Hansch et al., 1989). Nonpolar narcosis has been the subject of several reviews, including that of van Wezel and Opperhuizen (1995), and it is the least toxic of all acute mechanisms and is important in risk assessment. QSARs for this mechanism serve to estimate "baseline" or minimum toxicity in the absence of any stereoelectronic or bioreactive interactions (Schultz, 1989).

As previously noted, the aquatic toxicity of amines has been examined in a number of test systems, including algae (Newsome et al., 1993b), daphnids (Newsome et al., 1993b), fish (Newsome et al., 1991; 1993a), and protozoa (Schultz et al., 1991). More recently, Jaworska et al. (1995) conducted volume fraction analyses for the amine narcosis mechanism of toxic action. Eq. [2] represents the QSAR for the amine narcosis mechanism of toxic action. The lack of a toxic response for dodecylamine reflects the aquatic solubility cutoff for the amine narcotics in the *Tetrahymena* population growth assay. Equation [2]: demonstrates the regression analysis for the data from the primary aliphatic amines. When compared to the aliphatic alcohol model, the difference in slope is minimal. However, the difference in the intercept is > 0.7 of a log unit. Each primary

amine is more toxic than its corresponding alcohol. Equation [2] represents the QSAR for amine narcosis. While the molecular site has yet to be identified, amine narcosis is considered a separate mechanism of toxic action. Low molecular-weight, aliphatic-primary amines exhibit a modest excess toxicity. The specific nature of this excess toxicity is unknown at this time but metabolism can not be ruled out.

To resolve the differences between Eqs. [1] and [3], McFarland's probabilistic model of toxicity is of great value (McFarland, 1970). McFarland (1970) stated that the manifestation of chemical toxicity is preceded by two circumstances. First, the ingress of the chemical into the cell, and second, the interaction of the chemical with the molecular site of action. Movement into the biophase, a passive diffusion process, requires crossing cell membranes. It is modeled customarily by the hydrophobic term, $\log K_{OW}$. The chemical-molecular site interaction is more complex, and classically, such interactions have been modeled by electronic and/or steric parameters. This two step process can be modeled by the generic QSAR, $\log 1/\text{toxic effect} = A [\log \text{ of the probability of entering the biophase}] + B [\log \text{ of the probability of interacting with the site of action}] + \text{intercept}$.

The slope of the lines specified by Eqs. [1] and [3] represent the A term of McFarland's equation. These slopes can be used to compare the relative importance of partitioning into the biophase in explaining potency between the chemical groups. There is a trend for this term to be indirectly related to bioreactivity. In the case of baseline narcosis, the slope of the entrance term (i.e., $\log K_{OW}$) theoretically equals 1.00. As seen in Eq. [1], the slope may be < 1.00 , especially for systems with hydrophobic components in the test medium (Jaworska and Schultz, 1994). In addition, in the case of the baseline model represented by Eq. [1], it is assumed that interaction of the toxicant with a site of action is null. Thus, the slope of the stereoelectronic term, B, is 0.

For the aminoalkanols modeled by Eq. [3], one limitation of the model is the ability of these compounds to reach the site of action such that $A \neq 0$. However, a comparison of Eqs. [1] and [3] revealed the A value for the aminoalkanols to be less than for the baseline neutral organics. However, because the aminoalkanols are bioreactive they also must have a stereoelectronic interaction at the molecular site of action so $B \neq 0$. Since the interaction terms are constant, it drops out of Eq. [3], and a hydrophobic-dependent linear relationship with a smaller A value is the result.

A comparison of Figures 2 and 3 demonstrates that the quantitative modeling becomes more complicated when more than one reactive group or toxicophore is involved. In the case of the aminoalkanols with more than one reactive center, intramolecular interactions of a nonhydrophobic nature have a strong influence on toxic potency. Toxic potency for the amino-1-hydroxyl-derivatives separated by more than one methylene group (the diamonds on Figure 3) is, for the most part, independent of hydrophobicity. Moreover, the comparative toxicity of 2-amino-1-hexanol, and its branched isomers (the squares on Figure 3) reveal the toxic potency is inversely related to the degree of hydrocarbon branching near the amino moiety. This is an indication of steric hindrance of bioreactivity.

For several other chemical classes, *Tetrahymena* population growth data of sufficient size exist to provide both structural indicators of mechanism and to develop QSARs. Primary propargylic alcohols (Schultz and Tichy, 1993) and aldehydes (Schultz et al., 1994) exhibit structure-toxicity relationships similar to those observed for the alkanamines. On-the-other-hand, alkenoates (Schultz et al., 1995) and isothiocyanates (Schultz and Comeaux, 1996) exhibit structure-toxicity relationships similar to those seen for the alcoholamines.

The observed toxicity of tertiary aminoalkanols was below that predicted by the baseline QSAR. This was thought to be due to their high volatility and low density which effected bioavailability and pipeting accuracy, respectively.

As noted earlier, T_e is defined as the ratio of baseline toxicity to observed toxicity (Lipnick et al., 1987). An examination of the literature revealed that chemical classes having toxicity in excess of baseline are indicated by their smaller slope. For each of these classes, T_e disappears at some $\log K_{OW}$ value with the points of intersection for the baseline model with each of the other classes being between a $\log K_{OW}$ value of 4 and 5. This is also the hydrophobicity at which aqueous solubility becomes the limiting factor in toxicity. Thus, the more hydrophilic-bioreactive toxicants exhibit acute aquatic toxicities in greatest excess of values predicted by the baseline hydrophobic-dependent model. The trend of T_e being inversely related to hydrophobicity is consistent with the observations of Lipnick et al. (1987) for epoxides, Schultz and Tichy (1993) for primary propargylic alcohols, and Schultz and Comeaux (1996) for isothiocyanates. Moreover, these observations are in agreement with the suggestion of Ferguson (1939) that as hydrophobicity increases within a homologous series of electrophiles, there is a transition from "chemical toxicity" (i.e., bioreactive) to "physical toxicity" (i.e., narcosis).

In the case of primary, unbranched amine data set, the most hydrophobic compounds suggest a shift in mechanism for the lowest members of the series. (Newsome). Newsome studies demonstrated that $\log K_{OW}$ is sufficient by itself to model amines.

It has been observed that toxicity will decrease as a homologous series is ascended in a definite quantitative manner. The rapid decrease in toxic concentration does not continue indefinitely. Eventually, a homologue of maximum toxicity is reached and anything beyond that is feebly or nontoxic (Ferguson, 1939). An example of this occurs with the primary unbranched amines. For each additional methyl group, added toxicity

decreased to the point of no effect. However, there is also a correlation between toxicity and physical properties.

After nonpolar narcotics, soft(electro)nucleophils form the second largest group of industrial organic chemicals. Soft interactions between toxicants and biomolecules are influenced by the ability to accept or donate electron density (superdelocalization) through orbital rather than electrostatic charge transfers (Veith and Mekenyan, 1993). However, this mechanism of action was eliminated for the alcoholamines since there was no significant improvement in the predictability of Eq. 3 when the molecular orbital energy parameters were included in the regression analyses. This suggests that the toxicity aminoalkanols is not orbitally controlled (i.e., not soft electrophilic in nature).

In conclusion, potency of bioreactive toxicity is difficult to quantify at this time. It is believed that since the aminoalkanols are neither nonpolar narcotic nor soft(electro)nucleophilic, then they are possibly driven by a mechanism of action which remains undefined at this time. It is feasible that the compound is driven by a metabolic mechanism. We may actually be observing an excess toxicity caused by a metabolite of the compound as opposed to the original compound itself.

In this study, the relative toxicity of aliphatic amines and aminoalkanols was determined and compared to aliphatic alcohols. Aliphatic alcohols are considered to exhibit minimum toxicity. Amines at one time were also considered to be nonpolar narcotics (Newsome et al., 1991). Since then, it has been demonstrated that amines, being slightly more toxic than aliphatic alcohols, appear to have their own mechanism of action: amine narcosis. Amine narcosis, like nonpolar narcosis, is physical and reversible. However, amine narcosis contains a reactive component quite possible via metabolism.

The analyses of the aminoalkanols determined them to be more toxic than the aliphatic alcohols. This conclusion is supplemented by the shallower slope for hydrophobicity. In determining a possible mechanism of action for the aminoalkanols, nonpolar narcosis, which is the most common mechanism of action, neutral organic chemicals could be eliminated since the alcoholamines had shown significant excess toxicity. Therefore, they are considered to have bioreactivity as a mode of action like the amines. Bioreactivity involves a number of competing alkylation, redox and free radical processes. These interactions are dependent upon electronic and/on steric interactions.

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