Estimation of Relationship Between the Structure of 1,2,3,4-Tetrahydroisoquinoline Derivatives Determined by a Semiempirical Molecular-Orbital Method and their Cytotoxicity

MARIKO ISHIHARA[1], HAJIME HATANO[2], MASAMI KAWASE[3] and HIROSHI SAKAGAMI[2]

Divisions of [1]Basic Chemistry and [2]Pharmacology, Meikai University School of Dentistry, Sakado, Saitama 350-0283; [3]Faculty of Pharmaceutical Sciences, Matsuyama University, Matsuyama, Ehime 790-8578, Japan

Abstract. *A semiempirical molecular-orbital method (CAChe 4.9, PM5) was applied to delineate the relationship between the cytotoxicity (evaluated by 50% cytotoxic concentration, CC_{50}) of nineteen 1,2,3,4-tetrahydroisoquinoline derivatives, their molecular weight and the sixteen chemical parameters (descriptors) determined by CONFLEX/PM5 method. There was little or no correlation between the CC_{50} in HL-60 cells and the heat of formation, stability of hydration (ΔH), dipole moment, electron affinity, ionization potential, highest occupied molecular orbital energy (E_{HOMO}), lowest unoccupied molecular orbital energy (E_{LUMO}), absolute hardness (η, softness and hardness of the molecule) or molecular weight (r^2<0.312). On the other hand, there was a good correlation between the CC_{50} and the hydrophobicity (log P) (r^2=0.503), and the descriptors for the molecular size such as surface area (r^2=0.771), volume (r^2=0.805) and width (r^2=0.757). Similar, but not so clear-cut correlation was found in HSC-2, HSC-3 and HSC-4 human oral squamous cell carcinoma cell lines. The present study demonstrates that the cytotoxicity of 1,2,3,4-tetrahydroisoquinoline derivatives depends more on the descriptors for molecular size rather than the physicochemical descriptors.*

Tetrahydroisoquinoline have been reported to display antitumor activity (1), anti-inflammatory activity (2) and to prevent Parkinson's disease in an animal model (3). We investigated here the relationship between the cytotoxicity against human oral squamous cell carcinoma cell lines (HSC-2, HSC-3, HSC-4) (evaluated by 50% cytotoxic concentration, CC_{50}) of nineteen 1,2,3,4-tetrahydroisoquinoline derivatives, their molecular weight and the sixteen chemical parameters (descriptors) determined by CONFLEX/PM5 method.

Correspondence to: Mariko Ishihara, Division of Basic Chemistry, Department of Oral Biology and Tissue Engineering, Meikai University School of Dentistry, Sakado, Saitama 350-0283, Japan. Tel: +81 492855511 ext. 563, Fax: +81 492855171, e-mail: mariko@dent.meikai.ac.jp / sakagami@dent.meikai.ac.jp

Key Words: 1,2,3,4-Tetrahydroisoquinoline, QSAR, cytotoxicity, semiempirical molecular-orbital method, absolute hardness.

Materials and Methods

Materials. The following chemicals and reagents were obtained from the indicated companies: Dulbecco's modified Eagle's medium (DMEM) (Gibco BRL, Grand Island, NY, USA); fetal bovine serum (FBS) (JRH Bioscience, Lenexa, KS, USA); 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) (Sigma Chem. Co., St. Louis, MO, USA). All tetrahydroisoquinolines were provided by Dr. Kawase, Matsuyama University.

Assay for cytotoxicity. Human promyelocytic leukemic cell line (HL-60) and human oral squamous cell carcinoma cell lines (HSC-2, HSC-3, HSC-4) were cultured in RPMI-1640 or DMEM supplemented with 10% heat-inactivated FBS under a humidified 5% CO_2 atmosphere, respectively. These cells were incubated for 48 hours with different concentrations of each compound, and the viable cell number was determined by cell counting after staining with trypan blue (for HL-60 cells) or MTT method (for other cell lines) (4). The 50% cytotoxicity (CC_{50}) against these cell lines was determined from the dose–response curve.

Calculation. The most stable configuration of nineteen 1,2,3,4-tetrahydroisoquinoline was calculated by CONFLEX 5 (Conflex Co. Ltd., Tokyo). The optimization of the structure was achieved using a semiempirical molecular-orbital method (PM5), using a CAChe Worksystem 4.9 (MOPAC, PM5, non-COSMO, COSMO) (Fujitsu Co. Ltd., Tokyo). The following descriptors were used: i) heat of formation (COSMO, non-COSMO; kcal/mole); ii) stability of hydration (=COSMO – nonCOSMO (ΔH); kcal/mole); iii) dipole moment (D); iv) electron affinity (eV); v) ionization potential (eV); vi) hydrophobicity (log P); vii) highest occupied molecular orbital energy (E_{HOMO}; eV); viii) lowest unoccupied molecular orbital energy (E_{LUMO}; eV); ix) absolute hardness [η=(E_{UMO} - E_{HOMO})/2;

Table I. *CC_{50} and chemical descriptors for 1,2,3,4-tetrahydroisoquinoline derivatives.*

Compd.	CC_{50} (μM)				Log CC_{50} (μM)				Heat of formation	ΔH	Dipole moment	Electron affinity
	HL60	HSC-2	HSC-3	HSC-4	HL-60	HSC-2	HSC-3	HSC-4				
1	315	187	317	343	2.498	2.272	2.569	2.535	–76.237	–17.988	2.303	0.412
2	385	400	400	378	2.585	2.602	2.602	2.577	–69.863	–17.806	2.404	0.402
3	288	355	400	400	2.459	2.550	2.602	2.602	–179.061	–43.084	9.209	1.433
4	277	384	400	400	2.442	2.584	2.602	2.602	–182.764	–41.328	8.592	1.460
5	193	244	248	208	2.285	2.387	2.394	2.318	–127.151	–22.010	4.666	0.484
6	276	370	400	400	2.440	2.568	2.602	2.602	–292.552	–24.362	5.030	0.741
7	56	276	298	250	1.748	2.441	2.474	2.398	–54.850	–21.797	0.401	0.382
8	68	255	238	222	1.832	2.407	2.377	2.346	–54.352	–21.485	2.232	0.397
9	10	24	50	44	1.000	1.380	1.699	1.643	–179.881	–37.936	4.377	0.683
10	98	296	390	286	1.991	2.471	2.591	2.456	–212.785	–26.283	12.317	1.801
11	43	175	90	92	1.633	2.243	1.954	1.964	–212.823	–26.321	12.200	1.799
12	38	161	166	148	1.580	2.207	2.220	2.170	–220.166	–36.236	11.316	1.805
13	176	304	353	358	2.246	2.483	2.548	2.554	–209.720	–22.059	7.109	0.737
14	373	329	363	282	2.572	2.517	2.560	2.450	–65.479	–18.924	3.239	0.937
15	43	209	155	153	1.633	2.320	2.190	2.185	–231.555	–23.904	3.782	1.455
16	32	133	94	94	1.505	2.124	1.973	1.973	–247.909	–29.114	2.231	1.466
17	34	152	141	182	1.531	2.182	2.149	2.260	–329.531	–26.791	6.364	1.468
18	104	266	240	248	2.017	2.425	2.380	2.394	–66.147	–23.620	7.573	1.454
19	255	389	400	400	2.407	2.590	2.602	2.062	1.723	–24.254	9.912	0.855

Compd.	Ionization potential (eV)	Log *P*	E_{HOMO} (eV)	E_{LUMO} (eV)	η	χ	ω	Max. Length (Å)	N–R^3 (Å)	R^2-R^3 (Å)	Surface Area (Å^2)	Volume (Å^3)	MW
1	9.313	1.048	–9.313	–0.412	4.451	4.862	2.656	9.183	1.020	2.963	180.755	117.585	192
2	9.248	1.410	–9.248	–0.402	4.423	4.825	2.632	10.239	1.020	2.183	198.826	130.125	207
3	9.404	0.630	–9.404	–1.433	3.985	5.418	3.683	11.371	2.514	4.066	226.950	147.475	271
4	9.449	1.044	–9.449	–1.460	3.994	5.454	3.724	10.268	2.946	5.663	244.347	158.990	285
5	9.385	0.676	–9.385	–0.484	4.451	4.934	2.735	9.786	2.867	3.813	206.613	133.115	222
6	9.474	1.721	–9.474	–0.741	4.366	5.107	2.987	9.981	4.386	3.798	232.085	149.775	290
7	9.220	3.438	–9.220	–0.382	4.419	4.801	2.608	15.706	6.153	5.462	280.912	188.920	206
8	9.228	3.438	–9.228	–0.397	4.415	4.813	2.623	15.625	1.020	4.973	281.011	188.675	296
9	9.421	2.121	–9.421	–0.683	4.369	5.052	2.921	13.704	8.306	10.335	314.496	207.555	357
10	9.814	4.527	–9.814	–1.801	4.006	5.808	4.210	11.547	4.880	6.746	256.778	172.580	371
11	9.814	4.527	–9.814	–1.799	4.007	5.806	4.206	11.547	3.437	9.781	256.782	172.475	273
12	9.817	5.045	–9.817	–1.805	4.006	5.811	4.215	12.229	3.436	6.509	270.708	182.540	333
13	9.495	2.348	–9.495	–0.737	4.379	5.116	2.989	10.788	2.292	3.240	216.989	140.725	435
14	9.424	1.249	–9.424	–0.937	4.243	5.180	3.162	9.317	2.108	3.090	191.105	125.005	205
15	9.662	4.824	–9.662	–1.455	4.104	5.558	3.764	12.410	8.105	8.537	272.541	182.070	368
16	9.628	4.053	–9.628	–1.466	4.081	5.547	3.770	14.009	11.573	5.981	284.226	187.985	363
17	9.218	3.326	–9.218	–1.468	3.875	5.343	3.684	10.066	4.650	7.830	314.039	201.125	373
18	9.238	2.516	–9.238	–1.454	3.892	5.346	3.672	11.021	5.050	5.597	219.019	149.450	251
19	9.090	1.425	–9.090	–0.855	4.117	4.973	3.003	8.698	3.410	2.620	170.457	114.000	198

eV)]; x) absolute electron negativity [$\chi=-(E_{LUMO}+E_{HOMO})/2$; eV]; xi) reactivity index ($\omega=\chi^2/2\eta$; eV); xii) maximum length of the molecule (Å); xiii) distance between N-R3 (Å); xiv) distance between R^2-R^3 (Å); xv) surface area of the molecule (Å^2); xvi) volume of the molecule (Å^3) (5-7). The values of xii – xiv were measured using the 3-dimensional pictures of the most stable structure of each molecule. The quantitative structure–activity relationship (QSAR) was investigated from each descriptor (determined from molecular structure) and CC_{50} value (plotted as logarithmic scale), using a CAChe Worksystem 4.9 project reader.

Results and Discussion

Calculation with CONFLEX soft ware demonstrated that the most stable structure of all nineteen 1,2,3,4-tetrahydroisoquinoline derivatives showed the protrusion of substituents on the planar backbone (Figures 1 and 2).

The QSAR analysis was performed using HL-60 cells. The CC_{50} value, 16 descriptors and molecular weight of each compound are shown in Table I. QSAR between the CC_{50}

Table II. *Correlation coefficients between CC_{50} and each chemical descriptor in four different cell lines.*

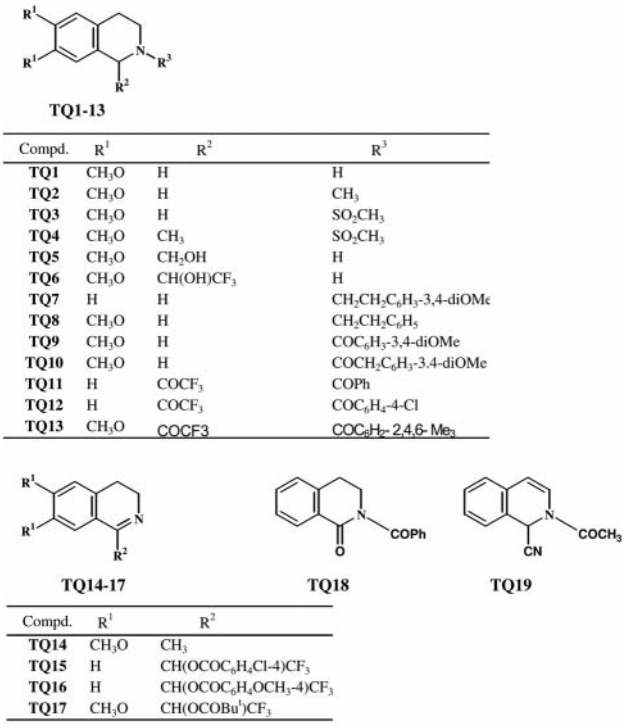
Cell line	Heat of formation (kcal/mol)	ΔH	Dipole moment (D)	Electron affinity (eV)	Ionization potential (eV)	Log *P*	E_{HOMO} (eV)	E_{LUMO} (eV)
HL-60	0.155	0.050	0.000	0.097	0.105	0.503	0.105	0.097
HSC-2	0.075	0.079	0.013	0.002	0.035	0.070	0.035	0.002
HSC-3	0.147	0.058	0.001	0.076	0.108	0.255	0.108	0.076
HSC-4	0.097	0.043	0.002	0.058	0.150	0.264	0.150	0.058

Cell line	η	χ	ω	Max. length (Å)	N-R^3 (Å)	R^2-R^3 (Å)	Surface Area ($Å^2$)	Volume ($Å^3$)	MW
HL-60	0.050	0.115	0.107	0.445	0.471	0.757	0.771	0.805	0.312
HSC-2	0.002	0.009	0.004	0.149	0.285	0.493	0.382	0.375	0.143
HSC-3	0.031	0.098	0.084	0.213	0.412	0.684	0.502	0.509	0.184
HSC-4	0.010	0.093	0.070	0.255	0.413	0.666	0.470	0.489	0.150

value logarithmically plotted and each descriptor of HL-60 cells are shown in Table II and Figure 3. There was a good correlation between the CC_{50} value and hydrophobicity (r^2=0.503, Figure 3f), the maximum length of molecule (r^2=0.445, Figure 1l), distance between N-R3 (r^2=0.471, Figure 3m), distance between R^2-R^3 (r^2=0.757, Figure 3n), surface area of the molecule (r^2=0.771, Figure 3o) and the volume of the molecule (r^2=0.803, Figure 3p). However, there was no correlation between the CC_{50} value and the other descriptors.

The QSAR between the CC_{50} value and the electron state of the molecule can be estimated by E_{HOMO} or E_{LUMO} (Figure 3 g and h, respectively) which are the indicators of electron donating and electron attracting capability, respectively, with an increase in E_{HOMO}, the electron donating capability is enhanced, while a decrease in E_{LUMO}, the electron attracting capability is enhanced. E_{HOMO} also reflects the ionization potential, hence no correlation existed between the CC_{50} and the ionization potential. In a QSAR analysis of endocrine disruptors, positive correlation has been reported between biological activity and chemical hardness (8-10). These papers showed that the biological activity of endocrine disruptors are well fitted to the η value. In contrast, the present results demonstrate the lack of positive correlation between the CC_{50} value of 1,2,3,4-tetrahydroisoquinoline and the η value (Table II, Figure 3).

We then performed the QSAR analysis using HSC-2, HSC-3 and HSC-4 cells. There was a good correlation between the CC_{50} values and the surface area of the molecule and the volume of the molecule (r^2=0.805~0.375) (Table II, Figure 4). These results show that descriptors xiv–xvi can be utilized to estimate the cytotoxicity of 1,2,3,4-tetrahydroisoquinoline related compounds.



Compd.	R^1	R^2	R^3
TQ1	CH_3O	H	H
TQ2	CH_3O	H	CH_3
TQ3	CH_3O	H	SO_2CH_3
TQ4	CH_3O	CH_3	SO_2CH_3
TQ5	CH_3O	CH_2OH	H
TQ6	CH_3O	$CH(OH)CF_3$	H
TQ7	H	H	$CH_2CH_2C_6H_3$-3,4-diOMe
TQ8	CH_3O	H	$CH_2CH_2C_6H_5$
TQ9	CH_3O	H	COC_6H_3-3,4-diOMe
TQ10	CH_3O	H	$COCH_2C_6H_3$-3.4-diOMe
TQ11	H	$COCF_3$	COPh
TQ12	H	$COCF_3$	COC_6H_4-4-Cl
TQ13	CH_3O	COCF3	COC_6H_2-2,4,6-Me_3

Compd.	R^1	R^2
TQ14	CH_3O	CH_3
TQ15	H	$CH(OCOC_6H_4Cl$-4)CF_3
TQ16	H	$CH(OCOC_6H_4OCH_3$-4)CF_3
TQ17	CH_3O	$CH(OCOBu^t)CF_3$

Figure 1. *The structure of 1,2,3,4-tetrahydroisoquinoline derivatives.*

In conclusion, the present QSAR analysis demonstrates that the CC_{50} value of 1,2,3,4-tetrahydroisoquinoline depends on their molecular size (surface area, volume, width), but not on the most of the other electronic factors. The molecular size determined by the CONFLEX/PM5 method is useful to evaluate the biological activity of 1,2,3,4-tetrahydroisoquinoline.

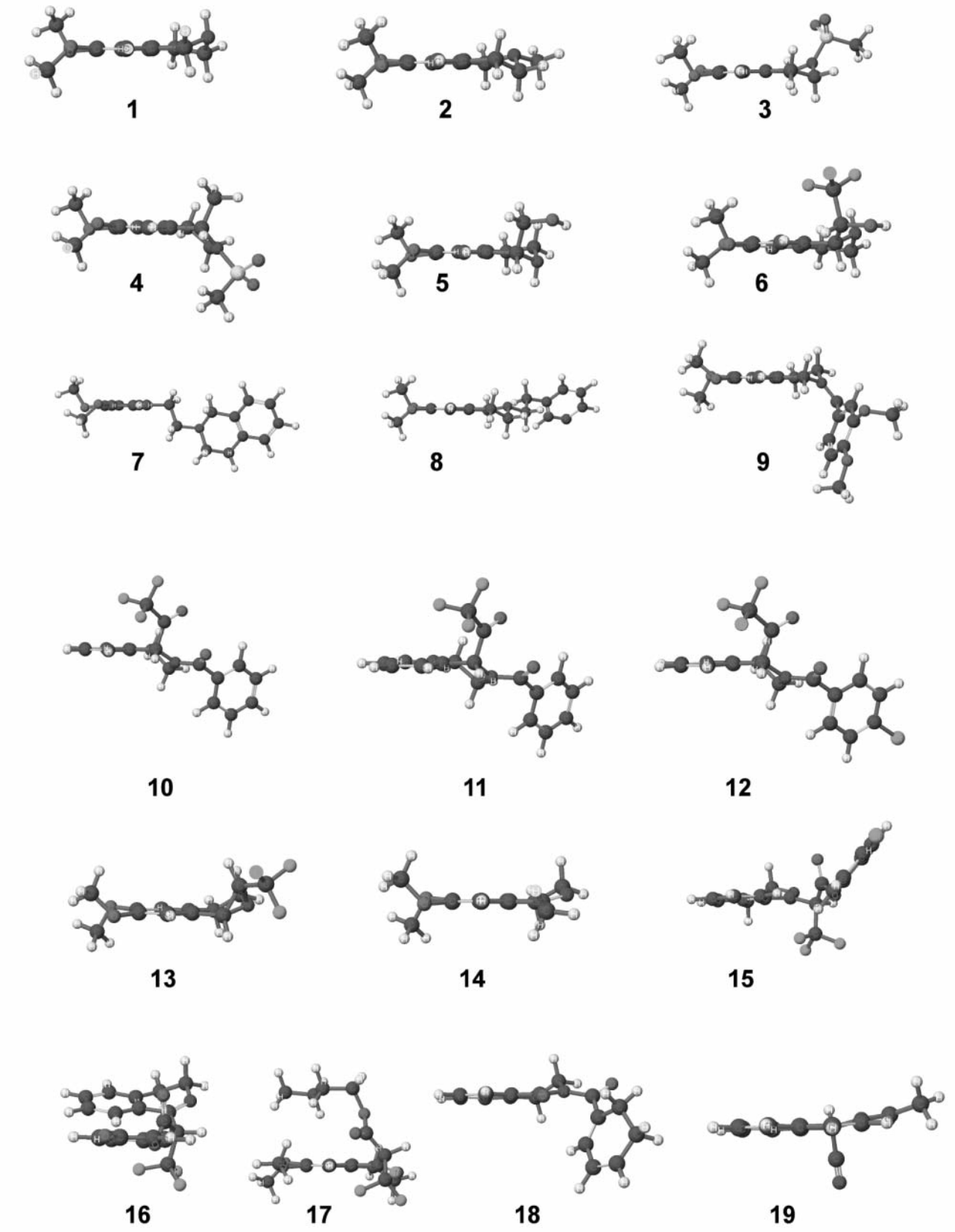


Figure 2. *The most stable conformation of 1,2,3,4-tetrahydroisoquinoline derivatives used.*

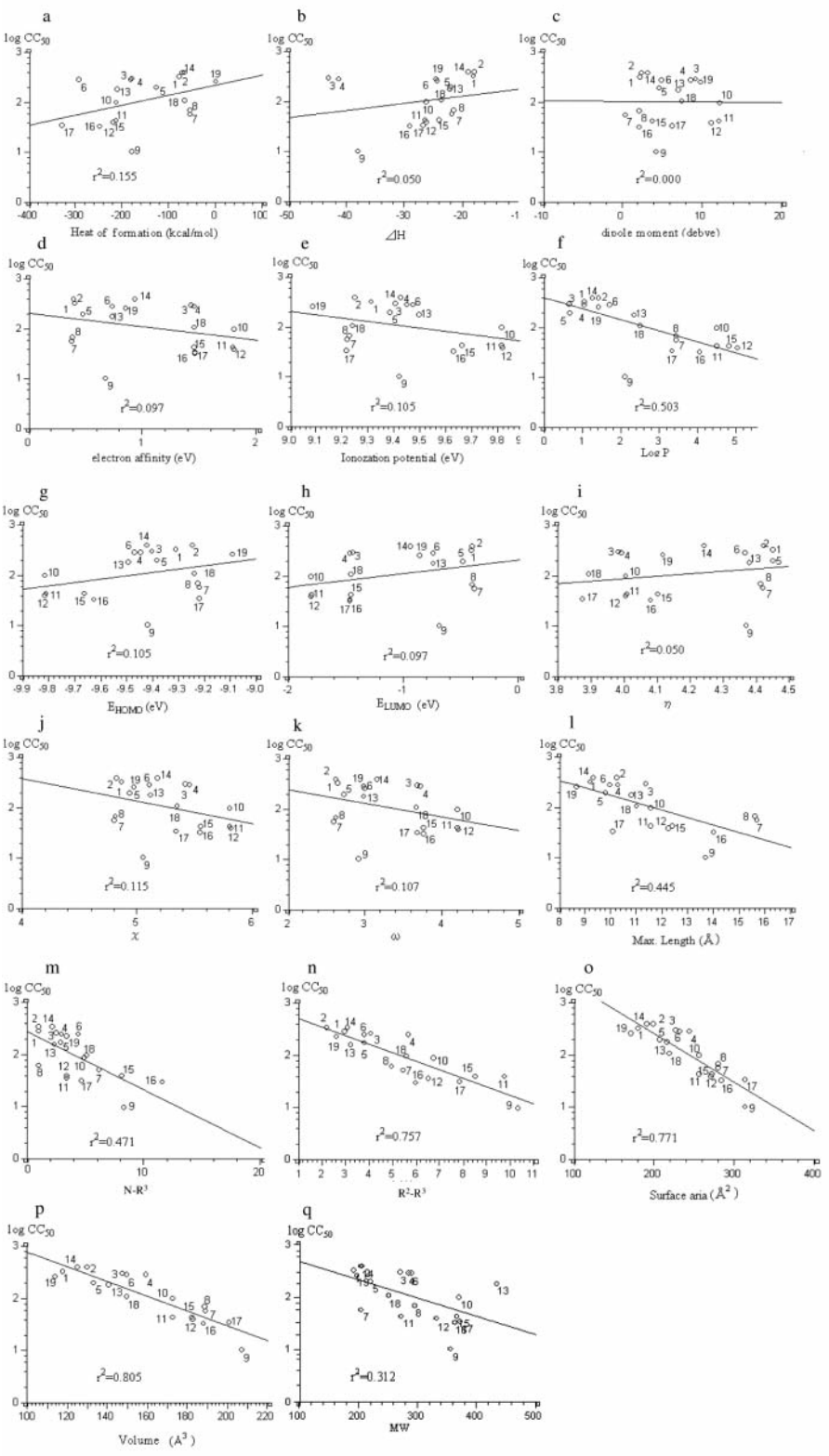


Figure 3. *Correlation between CC_{50} value (log scale) and each descriptor of 1,2,3,4-tetrahydroisoquinoline derivatives against HL-60 cells. The investigated descriptors are a, heat of formation; b, stability of hydration (ΔH); c, dipole moment; d, electron affinity; e, ionization potential; f, hydrophobicity (log P); g, E_{HOMO}; h, E_{LUMO}; i, absolute hardness; j, absolute electron negativity; k, reactivity index (ω); l, maximum length; m, distance between N-R^3; n, distance between R^2-R^3; o, surface area; p, volume of the molecule and q, MW.*

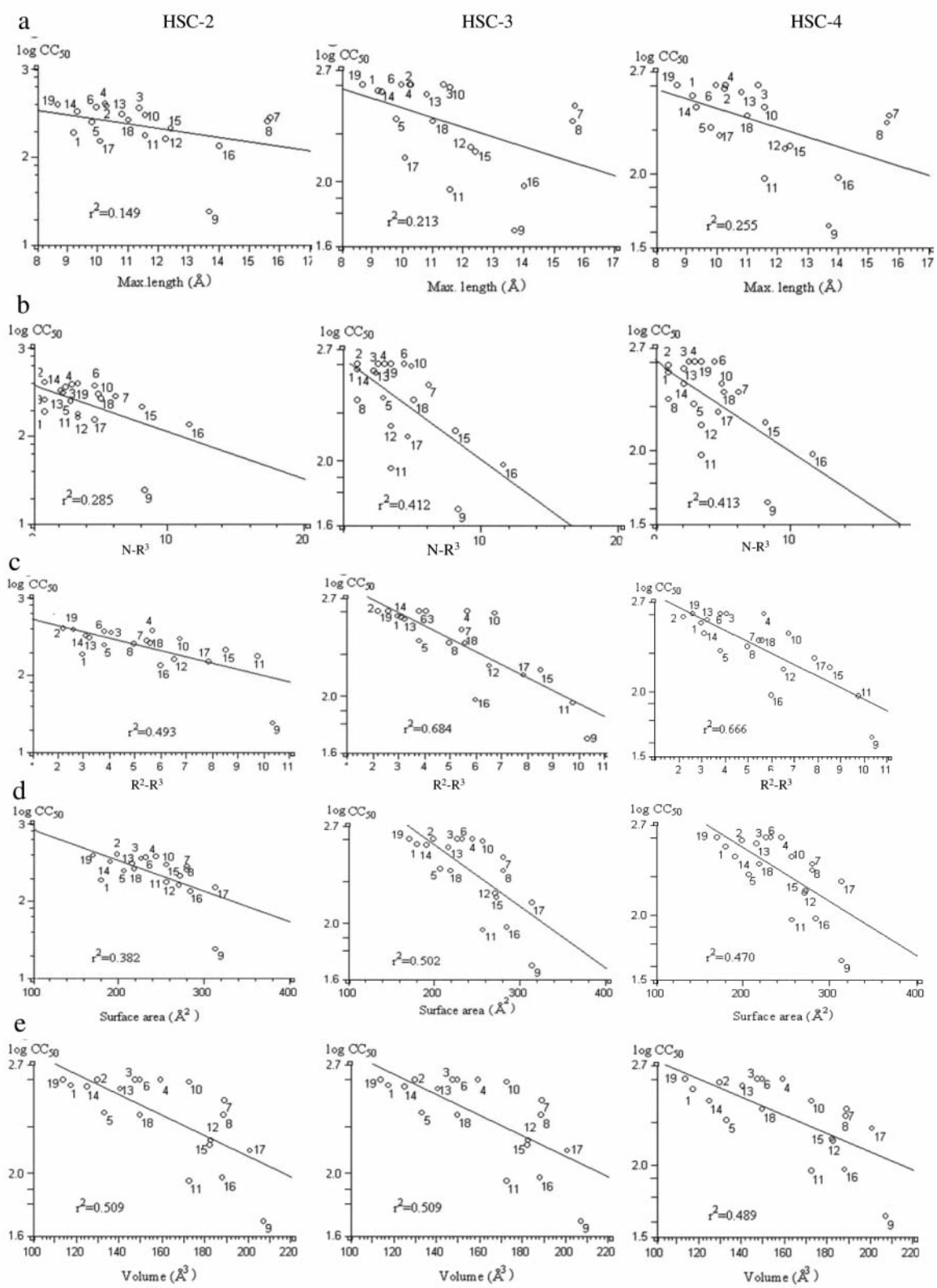


Figure 4. *Correlation between CC_{50} value (log scale) and selective descriptors of 1,2,3,4-tetrahydroisoquinoline derivatives against HSC-2 (left column), HSC-3 (center column) and HSC-4 cells (right column). Only descriptors that were found to show higher correlation coefficients in HL-60 cells (Figure 3) were selected.*

Acknowledgements

This study was supported in part by a Grant-in-Aid from the Ministry of Education, Science, Sports and Culture of Japan (Ishihara, No. 15659444; Sakagami, No. 19592156).

References

1 Lin HR, Safo MK and Abraham DJ: Identification of a series of tetrahydroisoquinoline derivatives as potential therapeutic agents for breast cancer. Bioorg Med Chem Lette *17*: 2581-2589, 2007.
2 Seo JW, Srisook E, Son HJ, Hwang O, Cha YN and Chi DY: Synthesis of tetrahydroisoquinoline derivatives that inhibit NO production in activated BV-2 microglial cells. Eur J Med Chem *43*: 1160-1170, 2008
3 Abe K, Saitoh T, Horiguchi Y, Utsunomiya I and Taguchi K: Synthesis and neurotoxicity of tetrahydroisoquinoline derivatives for studying Parkinson's disease. Biol Pharm Bull *28*: 1355-1362, 2005.
4 Sekine T, Takahashi J, Nishishiro M, Arai A, Wakabayashi H, Kurihara T, Kobayashi M, Hashimoto K, Kikuchi H, Katayama T, Kanda Y, Kunii S, Motohashi N and Sakagami H: Tumor-specificity and type of cell death induced by trihaloacetylazulenes in human tumor cell lines. Anticancer Res *27*: 133-144, 2007.
5 Ishihara M, Yokote Y and Sakagami H: Quantitative structure–cytotoxicity relationship analysis of coumarin and its derivatives by semiempirical molecular-orbital method. Anticancer Res *26*: 2883-2886, 2006.
6 Ishihara M, Kawase M and Sakagami H: Quantitative structure–activity relationship analysis of 4-trifluoromethyl-imidazole derivatives with the concept of absolute hardness. Anticancer Res *27*: 4047-4052, 2007.
7 Ishihara M and Sakagami H: Quantitative structure–cytotoxicity relationship analysis of 3-formylchromone derivatives by semiempirical molecular-orbital method with concept of absolute hardness. Anticancer Res *28*: 277-282, 2008.
8 Kobayashi S, Tanaka A and Sameshima K: Relationship between the electron structure and the expression of toxicity or biological activity of endocrine disruptors. Chemistry and Engineering *52*: 42-44, 1999 (in Japanese).
9 Kobayashi S, Sameshima K, Ishii Y and Tanaka A: Toxicity of Dioxins: Role of absolute hardness-absolute electronegativity diagram (η–χdiagram) as a new measure in risk assessment. Chem Pharm Bull *43*: 1780-1790, 1995.
10 Kobayashi S, Hamashima H, Kurihara M, Miyata N and Tanaka A: Hardness controlled enzymes and electronegativity controlled enzymes: role of an absolute hardness-electronegativity (η–χ) activity diagram as a coordinate for biological activities. Chem Pharm Bull *46*: 1108-1115, 1998.

Received February 24, 2009
Revised April 8, 2009
Accepted April 21, 2009