Food Chemistry **44** (1992) 337–342

Determination of apparent molar volumes (AMV) and capacity factors (k_w) for sweet and non-sweet sulphamates, correlation between AMV values, and Corey–Pauling–Koltun volumes and relative sweetness correlations

William J Spillane*, Gabriella Morini† & Gordon G. Birch

Department of Food Science and Technology, University of Reading, Whiteknights, Reading, UK, RG6 2AP

(Received 26 June 1991; revised version received and accepted 26 September 1991)

Apparent molar volume (AMV) and apparent specific volume (ASV) have been determined for 16 sulphamates, 14 of which are sweet. Capacity factors (k_w) have been determined by reversed-phase HPLC for seventeen sulphamates. A good linear correlation ($r = 0{\cdot}917$, $p < 0.001$) has been obtained between previously measured Corey–Pauling–Koltun (CPK) volumes and the AMV values of 16 sulphamates. Log k_w values show a moderately high correlation ($r = 0{\cdot}74$, $p < 0.01$) with the logs of the relative sweetness of 12 sulphamates. Relative sweetness also shows a reasonable correlation ($r = 0{\cdot}770$, $p < 0.01$) with molecular connectivity of the third order, $^3\chi_m$, for the same 12 compounds. Calculated partition coefficients (log P) for the sulphamates correlate well with log k_w ($r = 0{\cdot}756$, $p < 0.001$).

INTRODUCTION

There has been enormous interest in the development of quantitative structure – taste relationships for all classes of sweeteners (Crosby *et al.*, 1979; Daniel, 1989) since the first *QSAR* was reported for nitroaniline sweeteners (Deutsch and Hansch, 1966). We developed some semi-quantitative structure–taste relationships for carbosulphamates (Spillane & McGlinchey, 1981; Spillane & Sheahan, 1989) and aromatic sulphamates (Spillane *et al.*, 1989). We successfully classified sweet and non-sweet cyclic and open-chain heterosulphamates using linear discriminant analysis (Spillane *et al.*, 1983; Spillane & Sheahan, 1989). This work was mainly based on the use of Corey–Pauling–Koltun (CPK) space-filling molecular models for measuring the dimensions (x, y, z and $V = x.y.z$) of the R group in $RNHSO_3^-$.

In the present work in pursuit of QSAR values for the sulphamates we measured apparent molar volume (AMV) and capacity factors (k_w) and calculated partition coefficients (log P and third-order molecular connectivity ($^3\chi_m$) for a series of sulphamates including one non-sweet compound (4-bromophenyl and two having a sweet aftertaste (phenyl- and 3,5-di-fluorophenyl). Some interesting results and correlations have emerged from this work.

MATERIALS AND METHODS

Sodium sulphamates

The synthesis of the sulphamates, their purification and characterization has been described previously (see footnote *b*, Table 2). The synthesis of 3, 5-difluorophenylsulphamate (see footnote *e*, Table 2) and of exo-norbornylsulphamate have not yet been reported by us. Both gave a satisfactory elemental analysis (C, H and N), the usual IR bands and a positive and clean (i.e. free of sulphate ion and chloride ion) sulphamate test.

* Present address: Chemistry Department, University College, Galway, Ireland.

† Present address: Dipartimento di Scienze Moleculari Agroalimentari (DISMA), Facolta' di Agraria, Via Celoria 2, 20122 Milano, Italy.

Food Chemistry 0308-8146/92/$05.00

Determination of apparent molar volumes

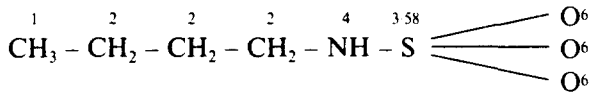
Apparent molar volumes were determined with an Anton–Parr precision density meter (DMA 60) and density measuring cell (DMA 602) from Stanton Redcroft (London) equipped with an automatic sampler (SP2) and Anadex printer. Temperature control was achieved with a Hetofrig bath (Heto Birkerod, Denmark), coupled to the density measuring cell. The density meter was calibrated with air and water and the method was as described by Birch and Catsoulis (1985). All measurements were carried out at 20±0·1°C with 5% (w/w) solution of the sodium sulphamate in water.

Determination of capacity factors (k_w)

The k_w values were determined by reversed-phase HPLC using a Lichrosorb RP-18 column; flow rate, 0·5 ml/min; eluant, water. Potassium bromide was used as a standard in each run and k_w was calculated from the equation: $k_w = (t_{RNHSO_3Na} - t_{KBr})/t_{KBr}$ where the t values are the retention times of the sulphamate and KBr. An LDC Constametric Pump 1 and an Applied Chemistry Systems mass detector, model 750/14, were used in the determinations.

Calculation of the third-order molecular connectivity ($^3\chi_m$)

Values of $^3\chi_m$ were calculated as described by Kier and Hall (1976). The calculation for *n*-butyl sulphamate is illustrated.



$(1 \times 2 \times 2 \times 2)^{-1/2} = 0.354 \quad (2 \times 2 \times 4 \times 3.58)^{-1/2} = 0.132$

$(2 \times 2 \times 2 \times 4)^{-1/2} = 0.177 \quad 3(2 \times 4 \times 3.58 \times 6)^{1/2} = 0.228$

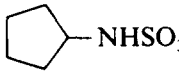
$^3\chi_m = 0.891$

Calculation of partition coefficients (*P*)

Partition coefficients were calculated by the fragmentation method (Hansch & Leo, 1979). Some examples follow:

log *P* for isobutylsulphamate: $(CH_3)_2CHCH_2NHSO_3^-$

$$2f_{CH_3} + f_{CH} + f_{CH_2} + f_{cBr} = 2(0.89) + 0.43 + 0.66 - 0.13 = 2.71$$

log *P* for cyclopentylsulphamate: 

$$4f_{CH_2} + f_{CH} + (5-1)F_b = 4(0.66) + 0.43 + 4(-0.09) = 2.71$$

log *P* for 3-bromophenylsulphamate: $3\text{-}BrC_6H_4NHSO_3^-$

$$f_{C_6H_5} - f_H + f^{\phi}_{Br} = 1.90 - 0.22 + 1.09 = 2.77$$

Measurement of Corey–Pauling–Koltun volumes

These measurements were available in previous work (Spillane & McGlinchey, 1981; Sheahan, 1988, Spillane & Sheahan, 1989). Replicate measurements were, at least, within 5% (usually better).

Statistics

Correlation analysis was performed for the various linear equations ($y = mx + c$) examined, and the data are given later in Table 3. The correlation coefficient (r) given is a Pearson product moment one, s is the standard deviation of the y distribution and p is the probability or the significance, calculated either from the t-test ($t = r(n\text{-}2)^{1/2}/1\text{–}r^2)^{1/2}$ or from tables, knowing r and reading at the n–2 level (Fisher & Yates, 1974).

RESULTS AND DISCUSSION

AMV/ASV correlations with volume

The results of the apparent molar volume measurements are given in Table 1 for 16 sulphamates. (ASV) values have been calculated from these and are given in the third column of the table. Fourteen of these compounds are sweet (see Table 2, incl. footnotes) and most of the ASV volumes fall in the expected range of 0·52 – 0.71 for sweetness (Birch & Kemp, 1989; Shamil & Birch, 1990). The phenyl compound, having a sweet aftertaste, also falls in this range. 4-Bromophenylsulphamate is as expected, in the sour range (0·11 – 0·51). Surprisingly two well-characterised sweeteners, 3-fluorosulphamate and 3-bromophenylsulphamate fall just short of the sweet range and there is no obvious explanation for this.

A plot of CPK volumes, converted to cm³/mol from the more usual $Å^3$, against apparent molar volumes (AMV) in cm³/mol is shown in Fig. 1 for the data in Table 1. Table 3 gives the equation of the best straight line with standard deviation, etc. This relationship is significant at the 0·001 level (t-test).

CPK molecular models have also been used by others interested in SAR values for sweeteners (Brussel *et al.*, 1975; Pautet & Nofre, 1978 *a*, *b*), but a relationship of this type, where 'calculated'; volumes and experimental volumes from the AMV measurements correlate well, appears to be rare. ASV values for amino acids correlate well with specific molecular volumes (V_m/mol.wt./Å³ $r = 0.894$, $p < 0.001$) and specific van der Waals volumes (V_w/mol.wt./Å³ dal⁻¹ $r = 0.875$, $p < 0.001$) (Bartolo, 1989). The specific molecular volumes and specific van der Waals volumes of seven carbohydrates and aspartame also correlate with the experimentally determined ASV values ($r = 0.913$, $p < 0.01$, $n = 8$ and $r = 0.892$, $p < 0.01$, $n = 8$, respectively). The

Table 1. Apparent specific and molar volumes and CPK measured volumes for sodium sulphamates

Compound R in $RNHSO_3^-Na^+$	Mol.Wt	ASV(ΦV), cm^3/g	AMV(ΦV/mol.wt.),[a] cm^3/mol	CPK, $A^{\circ 3}$	Volume, cm^3/mol
Phenyl	195·2	0·531	103·6	123	74
3-MeC_6H_4–	209·2	0·593	124·1	158	95
3-FC_6H_4–	213·2	0·466	99·3	133	80
3-ClC_6H_4–	229·6	0·525	120·6	152	91
3-BrC_6H_4–	274·1	0·439	120·3[b]	172	103
4-BrC_6H_4–	274·1	0·451	123·6[b]	177	106
exo-Norbornyl	213·2	0·596	127·1	210	127
endo-Norbornyl	213·2	0·578	123·2	210	127
Cyclooctyl	229·3	0·625	143·3	246	148
Cycloheptyl	215·2	0·624	134·3	208	125
Cyclohexyl	201·2	0·610	122·7	181	109
Cyclopentyl	187·2	0·578	108·2	155	93
n-Propyl	161·2	0·544	87·7[c]	90	54
n-Butyl	175·2	0·610	106·9	113	68
Isobutyl	175·2	0·616	107·9	158	95
Isopentyl	189·2	0·630	119·2	180	108

[a] Accurate to ±0·4 unless otherwise stated. Measurements were carried out twice in most salts.
[b] Accurate to ±0·5.
[c] Accurate to ±0·3.

molecular and van der Waal's volumes were computed using the GEPOL/87 program (Pascual-Ahuir *et al.*, 1987).

Since the accuracy of the CPK measurements is only about ± 5% the utility of the relationship in Fig. 1 for predicting AMV values (and hence ASV values) would be unsatisfactory since they can be determined with a much higher degree of precision. However, since

Table 2. Partition coefficient (log *P*, third order molecular connectivities ($^3\chi_m$), capacity factors (log k_w) and relative sweetness for sodium sulphamates

Compound R in $RNHSO_3^-Na^+$	Log *P*	$^3\chi_m$	Log k_w[a]	Relative sweetness[b]	
				RS	Log RS
Phenyl	1·90		–0·75	[c]	
3-MeC_6H_4–	2·57	1·581	–0·25	15·3	1·18
3-FC_6H_4–	2·05	0·996	–0·28	12·4	1·09
3-ClC_6H_4–	2·62	1·679	0·11	26·0	1·42
3-BrC_6H_4–	2·77	2·051	0·06	11·2	1·05
4-BrC_6H_4–	2·77		0·21	[d]	
3,5-diFC_6H_3–	2·20		–0·08	[e]	
exo-Norbornyl	3·30		–0·02	[f]	
endo-Norbornyl	3·30		0·09	[g]	
Cyclooctyl	4·42	2·651	0·58	27·8	1·44
Cycloheptyl	3·85	2·401	0·32	34·1	1·53
Cyclohexyl	3·28	2·151	0·02	41·0	1·61
Cyclopentyl	2·71	1·901	–0·67	10·0	1·00
n-Propyl	1·97	0·610	–0·71	0·63	–0·20
n-Butyl	2·51	0·891	–0·30	3·5	0·54
Isobutyl	2·74	1·152	–0·34	2·9	0·46
Isopentyl	3·40	1·490	–0·22	6·8	0·83

[a] Column, Lichrosorb RP-18; eluent, H_2O, flow rate: 0·5 ml/min.
[b] RS values for the aromatics are in Spillane *et al.* (1989), for isopentyl in Benson (1976) and for the other seven alicyclics/aliphatics in Benson and Spillane (1976).
[c] Described as having 'a sweet aftertaste' (Audrieth & Sveda, 1944).
[d] Sourness (Spillane & Sheahan, unpublished work).
[e] Bitter/acidic with a sweet aftertaste (Spillane *et al.*, unpublished work).
[f] Very sweet (Evangelisti *et al.*, 1980).
[g] Reported as faintly sweet/non-sweet (see Benson, 1976; Spillane & Sheahan, 1989).

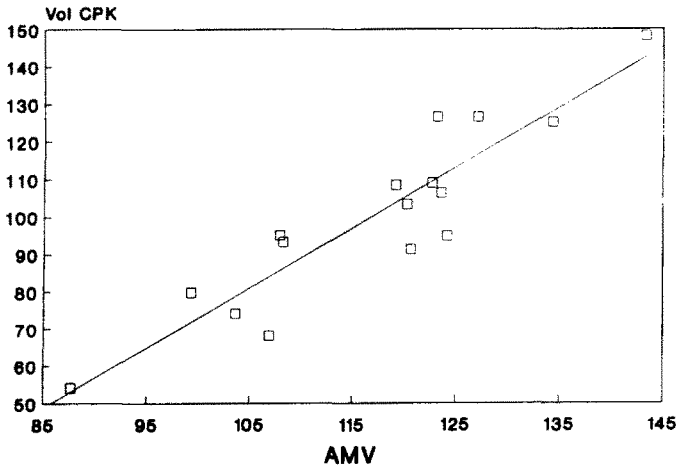


Fig. 1. Plot of Corey–Pauling–Koltun (CPK volumes (cm^3mol^{-1})) versus apparent molar volumes (cm^3mol^{-1}) (AMV).

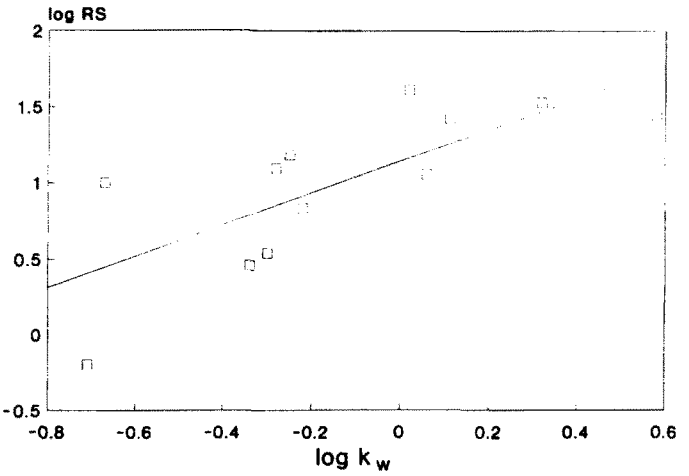


Fig. 2. Plot of log of relative sweetness (log RS) versus log of capacity factor (log k_w).

AMV/ASV values give a direct measure of the displacement and electrostriction of water by solute, they can be related to the events at the chemoreception level (Shamil *et al.*, 1987; Birch & Shamil, 1988) and the above relationships suggest that CPK volumes for the sulphamates and the GEPOL generated volumes for amino acids and carbohydrates provide important numbers that may be relevant to such events.

Log P – log k_w correlations

Numerous examples are known of the correlation of partition coefficients with capacity factors (Braumann, 1986) in log–log plots. We find that the log P of the seventeen sulphamates in Table 2 correlate well (r = 0·756, significant at the 0·001 level) (Table 3, eqn (2)) with log k_w values.)

Relative sweetness (RS) correlations

The log of the relative sweetness (log RS) correlates moderately well with log k_w (Table 2, Fig. 2). This relationship is significant at the 0·01 level (Table 3). Some years ago a similar correlation was made using seven of the 12 sulphamates used in Fig. 2 (Greenberg, 1980). Greenberg used recognition threshold (log $1/c$) plotted against the logs of calculated partition coefficients and obtained a correlation coefficient of 0·86, which is significant at the 0·02 level. We have correlated the same seven sulphamates in a log RS vs log k_w plot (Table 3, eqn (4)) and obtained a slightly poorer correlation (r = 0·767, significant at the 0·05 level).

Daniel has shown (Daniel, 1989) that plots of relative sweetness for some sulphamates and certain carbohydrates against 'HLB' i.e. the ratio of the mol. wt. of the hydrophobic portion of the molecule to that of the mol. wt. of the hydrophilic portion give rise to parabolic plots. For sulphamates, the ratio is mol. wt. of the $NHSO_3^-$ portion divided by the mol. wt. of the R portion of $RNHSO_3^-$. For sulphamates, all the points for the 12 compounds for which relative sweetness data are presently available cannot be accommodated on the same plot. A plot of RS versus $(HLB)^2$. $^3\chi_m$, also suggested by Daniel, showed marked scatter. However, a plot of RS versus $^3\chi_m$ gave a moderate correlation (Table 3, eqn (5)) plotted in Fig. 3 (r = 0·770, significant to the 0·01 level).

Table 3 Correlation analysis of various equations

Equation		Equation [e] ($y = mx + c$)	n[f]	r	s	p
1[a]	CPK =	1·61(±0·188)AMV − 88·21(±22·1)	16	0·917	9·76	0·001
2	log P	1·42(±0·32)log k_w + 3·01(±0·12)	17	0·756	0·46	0·001
3[b]	log RS=	1·03(±0.295)log k_w + 1·15(±0·114)	12	0·742	0.35	0.01
4[c]	log RS=	1·06(±0·398)log k_w + 1·08 (±0·190)	7	0·767	0·43	0·05
5[d]	RS =	16·06(±4.21)$^3\chi_m$ − 10·19(±7.31)	12	0.770	8.41	0.01

[a] Best line is drawn in Fig. 1.
[b] Best line is drawn in Fig. 2.
[c] Using the seven sulphamates previously correlated (Greenberg, 1980).
[d] Best line is drawn in Fig. 3.
[e] The numbers in brackets are standard deviations.
[f] Numbers of sulphamates in plot. See text for definitions of r, s and p.

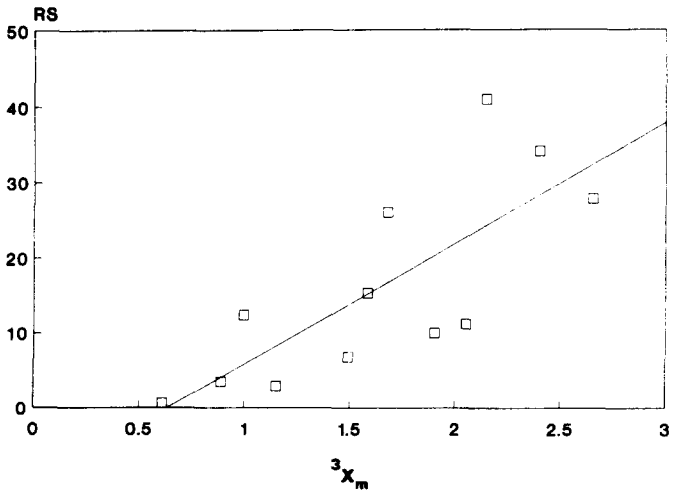


Fig. 3. Plot of relative sweetness (RS) versus third order molecular connectivity ($^3\chi_m$).

Third-order molecular connectivity of $^3\chi_m$ has been discussed at length (Kier & Hall, 1986). This parameter is said to encode information about the conformation of molecules and their flexibility or rigidity. The densities of a series of 9 heptanes gave a good linear relationship with $^3\chi_m$ and for a series of seven heptanes and twelve octanes a steric partition function, Z_g also gave good linear relationships. If $^3\chi_m$ encodes information on the three-dimensional structure and conformation of molecules then its utility in attempting to derive QSAR values for the sulphamate sweeteners is no surprise in the light of our work over some years, which has clearly indicated the importance of volume in deriving SAR values for the sulphamates.

In view of the diversity of structural types encompassed by the twelve compounds for which relative sweetness data are available, it would be surprising if the RS data could be accommodated in an equation containing only one independent variable. Work is continuing in this area and it is hoped to extend the database of compounds for which relative sweetness information is available. A multiparameter equation will probably be needed to correlate the data satisfactorily.

ACKNOWLEDGEMENTS

W. J. S. thanks the Royal Irish Academy and the Royal Society (London) for the award of a Royal Society European Science Exchange Fellowship. We thank Professor L. Merlini and Dr A. Arnoldi for helpful discussions on the k_w measurements. Dr M. G. B. Drew and Mrs C. Usher, Chemistry Department, University of Reading are thanked for their assistance with the graphics and statistical analysis. W. J. S. thanks the Department of Food Science and Technology, University of Reading for facilities and assistance during a period of sabbatical leave. G. G. B. thanks the Royal Society and the Research Board of the University of Reading for travel grants in support of this work.

REFERENCES

Audrieth, L. F., & Sveda, M. (1944). Preparation and properties of some *N*-substituted sulfamic acids. *J. Org. Chem.*, **9**, 89–101.

Bartolo, I. (1989). Taste, solution properties and computer generated molecular dimensions of sapid substances. MSc thesis, University of Reading, UK.

Benson, G. A. (1976). Sulfamate sweeteners: Structure activity and metabolic studies. PhD thesis, National University of Ireland.

Benson, G. A. & Spillane, W. J. (1976). Structure – activity studies on sulfamate sweeteners. *J. Med. Chem.*, **19**, 869–72.

Birch, G. G. & Kemp, S. E. (1989). Apparent specific volumes and tastes of amino acids. *Chem. Senses*, **14**, 249–58.

Birch, G. G. & Shamil, S. (1988). Structure, sweetness and solution properties of small carbohydrate molecules. *J. Chem Soc. Faraday 1*, **84**, 2635–40.

Braumann, T. (1986). Determination of hydrophobic parameters by reversed-phase liquid chromatography: theory, experimental techniques, and application in studies on quantitative structure–activity relationships. *J. Chromatog.*, **373**, 191–225.

Brussel, L. B. P., Peer, H. G. & Van der Heijden, A. (1975). Structure–taste relationships of some sweet-tasting dipeptide esters. *Z. Lebensm. Unters. Forsch.*, **159**, 337–343.

Crosby, G. A., DuBois, G. E. & Wingard, Jr, R. E. (1979). *The design of Synthetic Sweeteners in Drug Design.*, Vol. 8, ed. E. J. Ariens. Academic Press, New York, p.215.

Daniel, J. R. (1989). Sweeteners: Theory and design, In *Frontiers in Carbohydrate Research — 1: Food Applications*, eds. R. P. Millane, J. N. BeMiller and R. Chandrasekaran. Elsevier Applied Science, London, 34–65.

Deutsch, E. W. & Hansch, C. (1966). Dependence of relative sweetness on hydrophobic bonding, *Nature*, **211**, 75.

Evangelisti, F., Bargagna, A. & Schenone, P. (1980). New synthetic sweetener. Preparation of terpenoid sulfamates. *Riv. Soc. Italy. Sci. Aliment.*, **9**, 435–6.

Fisher, R. A. & Yates, F. (1974). *Statistical Tables for Biological, Agricultural and Medical Research.* 6th edn. (revised). Oliver and Boyd, Edinburgh, pp. 46 and 63.

Greenberg, M. J. (1980). The importance of hydrophobic properties of organic compounds and their taste intensities: a quantitative structure – taste intensity study. *J. Agric. Food Chem.*, **28**, 562–6.

Hansch, C. & Leo, A. J. (1979). *Substituent Constants for Correlation Analysis in Chemistry and Biology.* Wiley-Interscience, New York.

Kier, L. B. & Hall, L. H. (1976). Molecular connectivity in Chemistry and Drug Research. Vol. 14 of a series of monographs in medicinal chemistry, ed. G. deStevens, Academic Press, New York.

Kier, L. B. & Hall, L. H. (1986). *Molecular Connectivity in Structure-Activity Analysis.* Academic Press, New York, pp. 53–68.

Pascual-Ahuir, J. L., Silla, E., Tomasi, J. & Bonaccorsi, R. (1987). Area and Volumes of Molecules Quantum Chemistry Program Exchange, QCPE Program No. 554. GEPOL/87.

Pautet, F. & Nofre, C. (1978*a*). Correlation of chemical structure and taste in the cyclamate series and the steric nature of the chemoreceptor site. *Z. Lebensm. Unters. Forsch.*, **166**, 167–70.

Pautet, F. & Nofre, C. (1978*b*). Structure–activity relations in sulfamates with sweetening characteristics and spatial organisation of the receptor site. *Pharm. Acta Helv.*, **53**, 231–5.

Shamil, S & Birch, G. G. (1990). A conceptual model of taste receptors. *Endeavour*, **14**, 191–3.

Shamil, S., Birch, G. G., Mathlouthi, M. & Clifford, M. N. (1987). Apparent molar volumes and tastes of molecules with more than one sapophore. *Chem. Senses*, **12**, 197–409.

Sheahan, M. (1988). Synthetic and structure taste studies of sulphamate sweeteners. PhD thesis, National University of Ireland.

Spillane, W. J. & McGlinchey, G. (1981). Structure–activity studies on sulfamate sweeteners II: Semi-quantitative structure–taste relationship for sulfamate ($RNHSO_3^-$) sweeteners — the role of R. *J. Pharm. Sci.*, **70**, 933–5.

Spillane, W. J. & Sheahan, M. (1989). Semi-quantitative and quantitative structure–taste relationships for carbo- and heterosulphamate ($RNHSO_3^-$) sweeteners. *J. Chem. Soc. Perkin 2*, 741–6.

Spillane, W. J., McGlinchey, G., O. Muircheartaigh, I. & Benson, G. A. (1983). Structure–activity studies on sulfamate sweeteners III: structure–taste relationships for heterosulfamates. *J. Pharm. sci.*, **72**, 852–6.

Spillane, W. J., Sheahan, M., Simmie, J. M., Cunningham, D., McArdle, P & Higgins, T. (1989). The taste of monosubstituted phenylsulphamates. *J. Chem. Soc. Chem. Commun.*, 545–7.