Structural Study of Flavonoids and Their Protonated Forms

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Z. Naturforsch. **51 c,** 784–790 (1996); received June 19/August 8, 1996

Flavonoids, Structure, Proton Affinity, PM3, AM1

The highly successful semiempirical quantum chemical methods AM1 (Austin Model 1) and PM3 (a reparametrization of AM1) were applied to an investigation of the conformational properties of flavone, 3-hydroxyflavone, isoflavone and 2-hydroxyisoflavone. The most stable structures correspond to the non-planar forms with an angle of phenyl ring rotation out of the chromone moiety from a relatively narrow interval (28° – 38°). The mono- and diprotonation of these compounds was also investigated. The prominent site of protonation is the oxygen of the carbonyl group with a protonation enthalpy from the interval of about 900–920 kJ.mol^{-1}. The protonation enthalpy for protonation of the ether oxygen was computed to be about 200 kJ.mol^{-1} lower. Adding a second proton to monoprotonated species studied resulted in much lower protonation enthalpies compared to monoprotonation. The geometry of the studied compounds upon protonation changed considerably.

Introduction

Flavonoids, a widely distributed family of natural compounds, derivatives of 2-phenylchromone (flavone) or 3-phenylchromone (isoflavone), have been of great interest for their diverse effects on human, animal and microorganisms in the last years (Middleton and Kandaswami, 1993). They also are intensively coloured chromogenes responsible for much of the colour found in vascular plants (Harborne *et al.*, 1975). Their chromophoric properties were studied both experimentally (Mabry *et al.*, 1970) and theoretically (Remko and Polcin, 1980; Shevchenko, 1994). The biochemical activity of many flavonoids and their metabolites depends primarily upon the structure and relative orientation of the various moieties in the molecule as shown by structure-activity data (Cody, 1988). The molecular structure of some flavonols was determined using X-ray crystallography (Cody, 1988). Some conformational studies of basic flavone using quantum chemical methods have also been reported (Vrielynck *et al.*, 1993; Cornard *et al.*, 1995). However, no experimental and/or theoretical work was published until now on a systematic comparison of four simplest structures (flavone, 3-hydroxyflavone, isoflavone and 2-hydroxyisoflavone) which represent basic molecules of important groups of flavonoids.

The purpose of the present investigation was to determine theoretically the geometric structure of flavone (**I**), 3-hydroxyflavone (**II**), isoflavone (**III**) and 2-hydroxyisoflavone (**IV**) by means of quantum chemical calculations. Further, we studied the protonation of basic centres of these compounds and estimated how the stable confor-

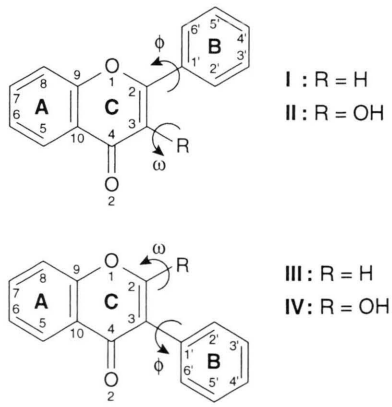

Fig. 1. Flavonoid geometry and numbering.

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0939–5075/96/1100–0784 $ 06.00

mations of the parent compounds could be changed by mono- and diprotonation.

The atomic numbering of the four flavonoids **I** - **IV** used in this work is shown in Fig. 1. It corresponds to the IUPAC numbering generally used for this family of compounds.

Computational Methods

Semiempirical quantum mechanics chemical calculations use parameters derived from experiments to simplify the calculation process. AM1 (Austin Model 1) (Dewar *et al.*, 1985) is a semiempirical method useful for organic molecules. It calculates electronic properties, optimized geometries, total energy and heat of formation. PM3 (Stewart, 1989a; 1989b) is a reparametrization of AM1 and differs from it only in the values of the parameters. *Ab initio* methods are characterized by the introduction of an arbitrary basis set for expanding the molecular orbitals and then the explicit calculation of all required integrals involving this basis set. Expressions like 3–21G and 6–21G are used for conventional basis sets which mathematically describe the orbitals within a system.

Molecular modeling of all molecules studied was carried out by the means of the MOLGEN 4.0 (Baricic and Mackov, 1995) and HyperChem 4.5 (Hypercube, 1995) programs. The geometry of each structure was then fully energy-optimized using the AM1 and PM3 Hamiltonians and parameters set implemented in the MOPAC (version 6.00) program package (Stewart, 1990; 1991). In the first step a potential energy profile was constructed by computing the dependence of the relative heat of formation versus the dihedral angle ϕ (C2'-C1'-C2-C3, molecules **I**, **II**), (C2'-C1'-C3-C2, molecules **III**, **IV**) between the phenyl ring and the chromone part of each molecule by fixing the dihedral angle ϕ by stepwise 10° in the interval from 0° to 180°. Then, the conformers corresponding to each local minimum of the curve were used for the final refinment of the geometry by relaxing all the geometrical parameters for optimisation, including the dihedral angle ϕ. The MOPAC program keyword 'Precise', that imposes a more stringent convergence criterion, was used and the keyword 'Gnorm', allowing an output when the gradient norm drops bellow the set limit, was set to be 0.01 (in kcal.mol^{-1}.Å^{-1}).

The AM1 method allows the calculation of the standard (T = 298 K) enthalpies of formation $\Delta H^0_{f,298}$. The proton affinity of base PA(B) can be computed by the Eq. (1) (Remko *et al.*, 1994):

$$PA(B) = \Delta H^0_{f,T}(H^+,g) + \Delta H^0_{f,T}(B,g) - \\ - \Delta H^0_{f,T}(BH^+,g) \quad (1)$$

$\Delta H^0_{f,T}$ represents the heat of formation of the species stated between parentheses. For $\Delta H^0_{f,298}(H^+,g)$ the experimental value 1537.1 kJ.mol^{-1} is taken (Stull and Prophet, 1971).

Results and Discussion

The results of the AM1 and the PM3 methods

Calculating the potential energy profiles of molecules **I** - **IV**, we found considerable differences between the results obtained using the two Hamiltonians AM1 and PM3 (Figs. 2a and 2b). Therefore we tested the suitability of these two methods for conformational studies of phenolic

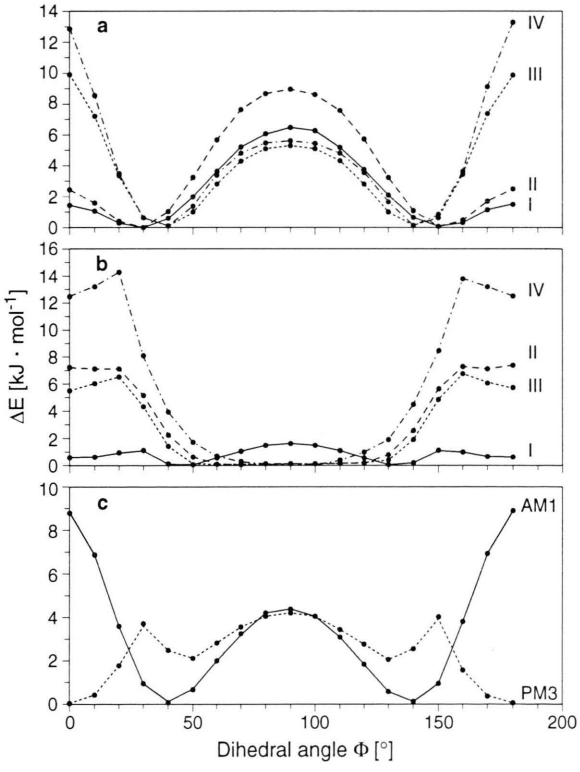

Fig. 2. a) Potential energy profiles of molecules **I** – **IV** calculated using the AM1 and b) the PM3 methods. c) Potential energy profiles of biphenyl calculated using the AM1 and the PM3 methods.

compounds, by applying it to the simple molecule of biphenyl. It has been proven before, that this molecule in the gas phase is favourably twisted to a dihedral angle of 44.4° (Almenningen *et al.*, 1985) between the two phenyl rings. *Ab initio* calculations are also in a close agreement with this value; the optimum torsional angle was found to be 45.5° (6–31G calculations) (Häfelinger and Regelmann, 1987) and 50.8° (3–21G calculations) (Kendrick, 1990), respectively. As shown in the Fig. 2c this is in good agreement with our results obtained using the AM1 method; the minimum of energy corresponds to the torsional angle 40.6°. The PM3 method seems to be less effective; although there is a local minimum on the potential energy profile at 47.5°, the global minimum was found at 0.0° and is about 2.0 kJ mol^{-1} lower. With respect to the better performance of the AM1 method by predicting the optimum structure of extended aromatic conjugated systems, we used in the further work the AM1 method only.

Conformational analysis

Generally, the problem of solving the structure of flavonoid compounds reduces to determination of the value of the dihedral angle (ϕ) between the ring B (the phenyl ring) and the rest of the molecule (the chromone part). The value of this angle depends on the environment (e.g. 3-hydroxyflavone was found nearly planar in the crystal and twisted (ϕ = 28.3°) in vapour state (Cornard *et al.*, 1995)). However, both phenyl and chromone rings stay always in an approximately planar configuration in the ground state (Vrielynck *et al.*, 1993; Cornard *et al.*, 1995). Naturally, this planarity is changed by protonation especially on oxygen O1, which is part of the ring C.

The eight states of protonation considered in this paper are shown in Fig. 3. It is a set of combinations of protonation of the oxygens O1 and O2. The protons were signed H_x and H_y. Each proton can interact with electrons of two lone-pairs of oxygen, what results in the two conformers **a** and **b** for H_x and **c** and **d** for H_y, respectively. The conformers **e** - **h** are further combinations of the conformers **a** - **d** and represent diprotonation. As all ZDO methods can not reproduce lone pairs and they produce a region of negative electrostatic potential only (Kallies and Mitzner, 1995), the approximate values for dihedral angles θ (H_x-O1-C9-C8) and μ (H_y-O2-C4-C10) were predestinated in the input files (Fig. 3).

Flavone

Table I presents in its first part the results of the AM1 computations of molecules **I** and **Ia** - **Ih**. Two minima on the potential energy profile in the range of the dihedral angle ϕ from 0° to 180° are presented and signed m1 and m2. For neutral flavone both minima are energetically equivalent. The protonation of this compound at the etheric

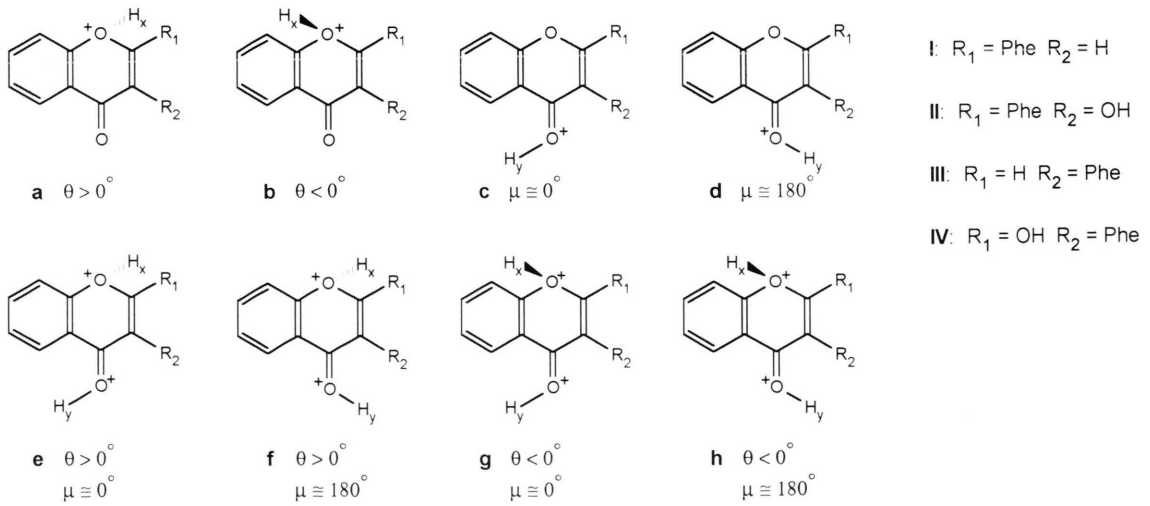

Fig. 3. Schematic view of the geometry of the protonated forms.

Table I. Relative heats of formation and selected AM1 optimized structural parameters of flavone (**I**), 3-hydroxyflavone (**II**), isoflavone (**III**), 2-hydroxyisoflavone (**IV**) and their protonated forms (**a–h**).

	m1[a]					m2[a]				
Species	$\Delta H^{o}_{f,298}$ [kJ/mol]	φ^{b} [°]	ω^{c} [°]	θ^{d} [°]	μ^{e} [°]	$\Delta H^{o}_{f,298}$ [kJ/mol]	φ^{b} [°]	ω^{c} [°]	θ^{d} [°]	μ^{e} [°]
I	28.21	28.8				28.21	150.8			
Ia						852.96	100.9		35.6	
Ib	852.96	77.0		–35.6						
Ic	651.90	13.8			–0.1	651.90	166.3			0.1
Id	647.15	16.3			–179.8	647.14	164.0			179.8
Ie	1879.69	51.0		28.9	1.5	1880.67	142.8		38.4	0.9
If	1873.29	48.1		30.1	–179.1	1873.31	145.2		40.4	179.7
Ig	1880.67	35.1		–38.4	–0.9	1879.69	127.9		–28.9	–1.5
Ih	1873.31	32.9		–40.0	–179.7	1873.29	131.1		–30.1	179.0
II	–146.00	28.3	173.1			–146.00	151.7	–173.1		
IIa						678.50	130.4	–170.8	35.5	
IIb	678.50	48.0	170.8	–35.5						
IIc	498.23	23.2	154.9		–1.1	498.23	157.2	–154.9		1.1
IId	483.31	31.5	52.2		179.7	483.31	149.7	–52.2		–179.7
IIe	1730.84	37.7	143.3	28.7	2.6	1730.77	148.5	–135.3	39.0	–0.2
IIf						1707.30	92.6	–3.0	34.9	–179.1
IIg	1730.77	29.5	135.3	–39.0	0.2	1730.84	141.1	–143.3	–28.7	–2.6
IIh	1707.27	87.3	2.9	–34.9	179.1					
III	23.13	38.5				23.13	141.0			
IIIa	869.78	30.7		81.8		868.12	141.7		84.1	
IIIb	868.12	37.9		–84.1		869.78	149.0		–81.8	
IIIc	670.40	43.8			–2.6	670.40	135.4			2.6
IIId	651.69	60.5			–178.9	651.69	118.6			178.9
IIIe	1928.12	40.7		42.6	0.1	1928.14	139.0		47.1	1.7
IIIf	1906.31	57.8		47.6	–179.0	1905.58	123.1		46.4	179.2
IIIg	1928.14	39.9		–47.1	–1.7	1928.12	137.7		–42.6	–0.1
IIIh	1905.58	55.2		–46.4	–179.2	1906.31	120.3		–47.6	179.0
IV	–173.66	37.4	174.7			–173.66	142.6	–174.7		
IVa	660.61	36.9	82.7	32.3		667.97	150.8	–59.8	27.5	
IVb	667.97	29.7	59.8	–27.5		660.61	143.2	–82.7	–32.3	
IVc	463.02	43.6	178.1		–3.7	463.02	136.4	–178.1		3.7
IVd	441.53	61.0	178.8		–178.9	441.53	118.8	–178.8		178.9
IVe	1711.53	42.9	66.2	31.2	–0.2	1714.07	126.6	10.6	30.3	2.7
IVf	1688.23	77.7	24.6	31.5	–178.9					
IVg	1714.07	53.9	–10.6	–30.3	–2.7	1711.53	136.7	–66.1	–31.2	0.2
IVh						1688.23	103.1	–24.9	–31.5	178.9

[a] Two minima (if present) on the potential energy profile in the range of φ = (0°, 180°); [b] dihedral angle C2′–C1′–C2–C3 (molecules **I–Ih, II–IIh**), C2′–C1′–C3–C2 (molecules **III–IIIh, IV–IVh**); [c] dihedral angle H–O–C3–C2 (molecules **II–IIh**), H–O–C2–C3 (molecules **IV–IVh**); [d] dihedral angle H_x–O1–C9–C8; [e] dihedral angle H_y–O2–C4–C10.

oxygen O1 dramatically changes the potential energy profile of the molecule and the protonated form **Ia** possesses one very broad energy minimum only (Table I). This minimum corresponds to a approximately rectangular structure making the dihedral angle ϕ between the phenyl and chromone parts equal to 100.9°, which is apparently a result of steric repulsion between the ortho hydrogen of phenyl ring B and the H_x proton. The protonation on carbonyl oxygen O2 (species **Ic** and **Id**) makes the benzo-γ-pyrone to become positively charged and increases its capacity to accept negative charge from the phenyl ring. The electron donating ability of ring B leads to the better conjugation and more planar protonated structures **Ic** and **Id** (Table I).

The dihedral angle θ (H_x-O1-C9-C8) is a measure of the deviation of the hydrogen H_x out of

the plane of rings C and A. The value of this angle is from a relatively narrow interval for the whole series of protonated forms studied. The dihedral angle μ (H_y-O2-C4-C10) of the structures **Ic** - **Ih** represents the two possibilities of binding of hydrogen H_y, mentioned above. In all of the studied molecules the value of this dihedral angle is very close to the predefined value of 0° and 180°, respectively, i. e. the hydrogen H_y lies in the plane of the rings C and A. Furthermore it is evident, that the structures **a/b**, **e/g**, **f/h** are enantiomers (non superimposable mirror images), or more exactly that, e. g. the conformer m2 of structure **Ia** is an enantiomer of the conformer m1 of structure **Ib**. (Table I)

3-Hydroxyflavone

The results of the AM1 computations presented in the second part of Table I show the heats of formation and selected torsional angles of 3-hydroxyflavone **II** and its protonated forms **IIa** - **IIh**. It has been observed for the crystalline form (Etter *et al.*, 1986), that **II** exhibits a five-membered intramolecular ring formed by a hydrogen bond between the hydroxyl and the carbonyl group. Therefore we performed theoretical calculations with this structure only.

The calculations of the potential energy curves for the rotation about the C2-C1' bond of the species **IIa** and **IIb** show one very broad minimum. The optimal conformation is the result of a compromise between the stabilization effect of the interaction of the C3-hydroxyl oxygen and the ortho hydrogen atom of the phenyl ring and the repulsive forces between H_x and the other ortho hydrogen. Although the C3-hydroxyl is bent out of plane of the ring C to a dihedral angle of ±9.2°, the distance O2...H_{OH} is still less important than the sum of van der Waals radii. A margin case of hydrogen bonding can be observed for the molecule **IIc**, that is even protonized on the carbonyl oxygen O2; the distance O2...H_{OH} is 2.30Å. The situation for molecule **IId** is, of course, changed due to the fact that here the dihedral angle μ is about 180°. The C3-hydroxyl group is also turned to the opposite direction and, as a consequence of the close interaction between H_{OH} and the ortho hydrogen, bent out to a dihedral angle ω (H-O-C3-C2) of ±52.2°. All these influences culminate in structures **IIf** and **IIh**. By these species only one broad minimum area on the potential energy profile is again observed and the phenyl ring is positioned in a quasi right angle to the chromone part in the global minimum.

Isoflavone

From the third part of Table I it can be concluded that isoflavone **III** and its protonated forms **IIIa** - **IIIh** do not possess structural features that would cause steric interactions of the respective substituents of the chromone part. The C3 phenyl substitution represents a sufficient separation to avoid close contacts between H_x and phenyl. The protonation on the carbonyl oxygen yields a quasi hydroxyl group -O2-H_y and its interaction with the ortho hydrogens of the phenyl group is mastered with a slight increase of the dihedral angle φ (structures **IIId**, **IIIf** and **IIIh**, Table I). One interesting observation is the extremely high value of the dihedral angle θ for molecules **IIIa** and **IIIb** caused also by the fact, that even the whole ring C is twisted more than in any other molecule; dihedral angle (C2-O1-C9-C10) = 18.0°.

2-hydroxyisoflavone

Although **IV** has never been isolated from natural sources it has even the lowest (most negative) calculated heat of formation among the molecules **I** - **IV**.

The results of computation for molecules **IV** and **IVa** - **IVh** are presented in the fourth part of Table I. Likewise molecule **II**, **IV** also exhibits an intramolecular ring formed by a hydrogen bond between the hydroxyl hydrogen and the heterocyclic oxygen O1. Since this is a four-membered ring and the distance H_{OH}...O1 is 2.12Å only, the bond angle O1-C2-C3 is expanded to the value of 127.3°. This intramolecular stabilisation persists also in structures **IVc** and **IVd**, but is evidently broken by the O1 protonation in molecules **IVa**, **IVb** and **IVe** - **IVh**. The C2-hydroxyl group is positioned with the hydrogen oriented in the direction of the phenyl group at C3 in the latter mentioned molecules (see dihedral angle ω (H-O-C2-C3), Table I). This fact, along with the predicted position of hydrogen H_y in structures **IVf** and **IVh** causes that the phenyl ring is forced to a quasi right angle to the chromone part and that there can be only one

Table II. AM1 calculated proton affinities (in kJ·mol^{-1}) of the single steps of protonation of flavonoids **I–IV.**

	I		**II**		**III**		**IV**	
	m1	m2	m1	m2	m1	m2	m1	m2
base → **a**		712.35		712.60	690.45	692.11	702.84	695.47
base → **b**	712.35		712.60		692.11	690.45	695.47	702.84
base → **c**	913.42	913.42	892.87	892.87	889.83	889.83	900.42	900.42
base → **d**	918.16	918.18	907.79	907.79	908.54	908.54	921.91	921.91
a → **e**		509.39		484.83	478.76	477.08	486.17	491.00
a → **f**		516.75		508.30	500.57	499.64	509.48	
b → **g**	509.39		484.83		477.08	478.76	491.00	486.18
b → **h**	516.75		508.33		499.64	500.57		509.48
c → **e**	309.31	308.83	304.49	304.56	279.38	279.36	288.59	286.05
c → **g**	308.33	309.31	304.56	304.49	279.36	279.38	286.05	288.59
d → **f**	310.96	310.93		313.11	282.48	283.21	290.40	
d → **h**	310.94	310.95	313.15		283.21	282.48		290.40

broad minimum area observed on the potential energy profiles of these molecules.

Proton affinity

Flavones are moderatery strong oxygen bases (Davis and Geissman, 1954). The protonation site can be both carbonyl and etheric oxygen. Table II summarizes the AM1 calculated proton affinities (PA) of compounds studied. For evaluation of PA's the fully geometry-optimized structures of base and the corresponding cation were used. Only the PA's of the single steps of protonation, i. e. of the first step to monoprotonated forms and of the further possibilities of diprotonation are presented. Generally, it can be summarized for molecules **I – IV**, that of the two principal protonation sites the carbonyl oxygen O2 exhibits a substantially higher (by about 200 kJ.mol^{-1}) proton affinity, especially when adding proton H_y to position **d**. The transition base → **c** has a value of PA about 5–20 kJ.mol^{-1} lower. This observation is in agreement with earlier works (Dávid *et al.*, 1976) on the reactivity and basicity of flavonoid compounds. The conformers **a** and **b** possess the lowest value of PA among the monoprotonated forms. The subsequent addition of the second proton to protonated species (Table II) is a substantially less exothermic process, especially when adding H_x as a second proton to structures with present H_y proton.

Acknowledgements

Support of this work from the Slovak Ministry of Education (Grant No. 1/1224/96) and from ECC Program COPERNICUS (Grant No. ERB3512PL941009) is gratefully acknowledged.

Almenningen A., Bastiansen O., Fernholt L., Cyvin B. N., Cyvin S. J. and Samdal S. (1985), Structure and barrier of internal rotation of biphenyl derivatives in the gaseous state. Part 1. The molecular structure and normal coordinate analysis of normal biphenyl and perdeuterated biphenyl. J. Mol. Struct. **128,** 59–76.

Baricic P. and Mackov M. (1995), MOLGEN: personal computer-based modeling system. J. Mol. Graphics **13,** 184–189.

Cody V. (1988), Crystal and molecular structures of flavonoids. In: Plant Flavonoids in Biology and Medicine II: Biochemical, Cellular and Medicinal Properties (Cody V., Middleton E. Jr., Harborne J. B. and Beretz A., Eds.), Alan R. Liss, New York, pp. 29–44.

Cornard J. P., Vrielynck L., Merlin J. C. and Wallet J. C. (1995), Structural and vibrational study of 3-hydroxyflavone and 3-methoxyflavone. Spectrochim. Acta **51A,** 913–923.

Dávid É. R., Janzsó G., Bálint J. and Bognár R. (1976), The reactivity of the carbonyl group in flavonoid compounds. II. Basicity of the analogues of flavone and flavanone. Acta Chim. (Budapest) **88,** 309–318.

Davis C. T. and Geissman T. A. (1954), Basic dissociation constants of some substituted flavones. J. Am. Chem. Soc. **76,** 3507–3511.

Dewar M. J. S., Zoebisch E. G., Healy E. F. and Stewart J. J. P. (1985), AM1: a new general purpose quantum mechanical molecular model. J. Am. Chem. Soc. **107,** 3902–3909.

Etter M. C., Urbanczyk-Lipkowska Z., Baer S. and Barbara P. F. (1986), The crystal structures and hydrogen-bond properties of three 3-hydroxy-flavone derivatives. J. Mol. Struct. **144,** 155–167.

Häfelinger G. and Regelmann C. (1987), Refined ab initio 6–31G split-valence basis set optimization of the molecular structures of biphenyl in twisted, planar and perpendicular conformations. J. Comput. Chem. **8,** 1057–1065.

Harborne J. B., Mabry J. J. and Mabry H. (1975), The Flavonoids. Chapman & Hall, London.

Hypercube, Inc. (1995), Hyperchem 4.5. 419 Philip Street, Waterloo, Ontario N2L 3X2 Canada.

Kallies B. and Mitzner R. (1995), The ability of the semiempirical PM3 method to model proton transfer reactions in symmetric hydrogen bonded systems. J. Mol. Model. **1,** 68–78.

Kendrick J. (1990), Calculated energetics of torsional motion in six diphenyl molecules: benzophenone, diphenyl ether, diphenyl sulphide, diphenyl sulphone, diphenylmethane and biphenyl. J. Chem. Soc. Faraday Trans. **86,** 3995–4000.

Mabry T. J., Markham K. R. and Thomas M. B. (1970), The Systematic Identification of Flavonoids. Springer-Verlag, New York.

Middleton E. Jr. and Kandaswami C. (1993), The impact of flavonoids on mammalian biology: implications for immunity, inflamation and cancer. In: The Flavonoids: Advances in Research since 1986. (J. B. Harborne, Ed.),Chapman & Hall, London, pp. 619–652.

Remko M. and Polcin J. (1980), Experimental and calculated (PPP) electronic spectra of flavonoid, stilbene and coumarone structures. Collection Czechoslov. Chem. Commun. **45,** 201–209.

Remko M., Scheiner S. and Rode B. M. (1994), Molecular modelling of the antiarrhytmic-receptor interaction. J. Mol. Struct. (Theochem) **307,** 35–46.

Shevchenko S. M. (1994), Theoretical approaches to lignin chemistry. Croat. Chem. Acta **67,** 95–124.

Stewart J. J. P. (1989a), Optimization of parameters for semiempirical methods. I. Method. J. Comp. Chem. **10,** 209–220.

Stewart J. J. P. (1989b), Optimization of parameters for semiempirical methods. II. Applications. J. Comp. Chem. **10,** 221–264.

Stewart J. J. P. (1990), MOPAC: a semiempirical molecular orbital program. J. Computer-Aided Molecular Design **4,** 1–105.

Stewart J. J. P. (1991), MOPAC version 6.00. Frank J. Seiler Res. Lab., U. S. Air Force Academy, Colorado Springs, CO 80840.

Stull D. R. and Prophet H. (Eds.) (1971), JANAF Thermochemical Tables. Natl. Stand. Ref. Data Ser., Natl. Bur. Stand., NSRDS-NBS **37,** U. S. Govt. Print. Office, Washington, DC.

Vrielynck L., Cornard J. P., Merlin J. C. and Bopp P. (1993), Conformational analysis of flavone: vibrational and quantum mechanical studies. J. Mol. Struct. **297,** 227–234.