

Submitted to: *J. Am. Chem. Soc.*

Supporting Information

A Solid State ^{13}C NMR, Crystallographic and Quantum Chemical Investigation of Phenylalanine and Tyrosine residues in Dipeptides and Proteins

By

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Full citation of Ref 21:

- (21) Frisch, M. J.; Trucks, G. W.; Schlegel, H. B.; Scuseria, G. E.; Robb, M. A.; Cheeseman, J. R.; Zakrzewski, V. G.; Montgomery, J., J. A.; Stratmann, R. E.; Vreven, T.; Burant, J. C.; Dapprich, S.; Millam, J. M.; Daniels, A. D.; Kudin, K. N.; Strain, M. C.; Farkas, O.; Tomasi, J.; Barone, V.; Cossi, M.; Cammi, R.; Mennucci, B.; Pomelli, C.; Adamo, C.; Clifford, S.; Ochterski, J. W.; Petersson, G. A.; Ayala, P. Y.; Cui, Q.; Morokuma, K.; Salvador, P.; Dannenberg, J. J.; Malick, D. K.; Rabuck, A. D.; Raghavachari, K.; Foresman, J. B.; Cioslowski, J.; Ortiz, J. V.; Baboul, A. G.; Stefanov, B. B.; Liu, G.; Liashenko, A.; Piskorz, P.; Komaromi, I.; Gomperts, R.; Martin, R. L.; Fox, D. J.; Keith, T.; Al-Laham, M. A.; Peng, C. Y.; Nanayakkara, A.; Challacombe, M.; Gill, P. M. W.; Johnson, B.; Chen, W.; Wong, M. W.; Andres, J. L.; Gonzalez, C.; Head-Gordon, M.; Replogle, E. S.; Pople, J. A. *Gaussian 98* Pittsburgh, PA, 1998.

Full citation of Ref 22:

- (22) Frisch, M. J.; Trucks, G. W.; Schlegel, H. B.; Scuseria, G. E.; Robb, M. A.; Cheeseman, J. R.; Montgomery, J., J. A.; Vreven, T.; Kudin, K. N.; Burant, J. C.; Millam, J. M.; Iyengar, S. S.; Tomasi, J.; Barone, V.; Mennucci, B.; Cossi, M.; Scalmani, G.; Rega, N.; Petersson, G. A.; Nakatsuji, H.; Hada, M.; Ehara, M.; Toyota, K.; Fukuda, R.; Hasegawa, J.; Ishida, M.; Nakajima, T.; Honda, Y.; Kitao, O.; Nakai, H.; Klene, M.; Li, X.; Know, J. E.; Hratchian, H. P.; Cross, J. B.; Adamo, C.; Jaramillo, J.; Gomperts, R.; Stratmann, R. E.; Yazyev, O.; Austin, A. J.; Cammi, R.; Pomelli, C.; Ochterski, J. W.; Ayala, P. Y.; Morokuma, K.; Voth, G. A.; Salvador, P.; Dannenberg, J. J.; Zakrzewski, V. G.; Dapprich, S.; Daniels, A. D.; Strain, M. C.; Farkas, O.; Malick, D. K.; Rabuck, A. D.; Raghavachari, K.; Foresman, J. B.; Ortiz, J. V.; Cui, Q.; Baboul, A. G.; Clifford, S.; Cioslowski, J.; Stefanov, B. B.; Liu, G.; Liashenko, A.; Piskorz, P.; Komaromi, I.; Martin, R. L.; Fox, D. J.; Keith, T.; Al-Laham, M. A.; Peng, C. Y.; Nanayakkara, A.; Challacombe, M.; Gill, P. M. W.; Johnson, B.; Chen, W.; Wong, M. W.; Gonzalez, C.; Pople, J. A. *Gaussian 03*; Gaussian, Inc.: Pittsburgh, PA, 2003.

Table S1. Crystallographic Data Summary for Phenylalanine and Tyrosine Dipeptides

	Gly-Phe (6)	Phe-Tyr (11)
formula	C ₁₁ H ₁₄ N ₂ O ₃	C ₁₈ H ₂₀ N ₂ O ₄
mol wt	222.24	328.36
cryst syst	monoclinic	orthorhombic
space group	P2 ₁	P2 ₁ 2 ₁ 2 ₁
<i>a</i> (Å)	6.433(2)	5.4527(9)
<i>b</i> (Å)	5.4926(17)	12.754(2)
<i>c</i> (Å)	16.535(5)	23.021(3)
<i>α</i> (deg)	90.00	90.00
<i>β</i> (deg)	97.711(5)	90.00
<i>γ</i> (deg)	90.00	90.00
<i>V</i> (Å ³)	579.0(3)	1600.9(4)
<i>Z</i>	2	4
ρ_{calcd} (g cm ⁻³)	1.275	1.362
<i>T</i> (K)	193(2)	193(2)
μ (mm ⁻¹)	0.094	0.097
R1 (all data)	0.0399	0.0526
wR2 (all data)	0.0954	0.1317
crystallization	H ₂ O	H ₂ O (5% DMSO)

Table S2. Experimental Chemical Shifts for the Nonprotonated C^δ in Tyrosine Dipeptides^a

Compound	Dipeptide ^a	Chemical Shift (ppm)
17	Tyr-Val	156.0
7	Tyr-Glu	156.7
8	Tyr-Leu	156.5
18	Tyr-His	156.4
19	Tyr-Ile	157.0
9	Tyr-Phe	156.4
20	Tyr-Trp	156.4
21	Tyr-Gln	155.9
22	Tyr-Gly	158.0
23	Gly-Tyr	155.0
24	Ala-Tyr	155.1
10	Leu-Tyr	154.2
11	Phe-Tyr	153.9
25	His-Tyr	155.4
26	Ile-Tyr	153.0

^aBold indicates amino acid of interest.

Table S3. Distribuiton of the C γ Chemical Shifts Within the N δ^1 -H and N ϵ^2 -H Tautomers of Histidine Dipeptides.

Tautomer	Dipeptide	δ_{expt} (ppm)
N δ^1 -H	His-Leu	124.7
	His-Met	124.9
	Gly-His	127.8
	Leu-His	129.6
N ϵ^2 -H	His-Ala	131.7
	His-Glu	132.6
	Ala-His	137.4

Table S4. Experimental Chemical Shifts and Computed Chemical Shieldings for Phe-C^γ from the Charged NH₃⁺-phenylalanine-Amide and N-formyl-phenylalanine-CO₂⁻ Models Using the Hartree-Fock Method ^a

Compound ^a	δ_{expt} (ppm)	σ_{calc} (ppm)
1 Phe-Pro	134.1	57.6
2 Phe-Phe	135.3	59.3
3 Phe-Ala	135.6	56.7
11 Phe-Tyr	136.4	55.1
4 Phe-Val	136.9	59.8
9 Tyr-Phe	138.9	31.4
5 Ile-Phe	139.2	30.4
2 Phe-Phe	139.8	31.5
6 Gly-Phe	140.3	37.8

^aBold residue denotes the residue of interest.

Table S5. Experimental Chemical Shifts and Computed Chemical Shieldings for Phe-C γ from the Monomer Calculations Using Exact Crystal Structures Using the Hartree-Fock Method ^a

Compound ^a	δ_{expt} (ppm)	σ_{calc} (ppm)
1 Phe-Pro	134.1	58.9
2 Phe-Phe	135.3	58.3
3 Phe-Ala	135.6	53.1
11 Phe-Tyr	136.4	56.6
4 Phe-Val	136.9	62.4
9 Tyr-Phe	138.9	42.5
5 Ile-Phe	139.2	36.0
2 Phe-Phe	139.8	37.6
6 Gly-Phe	140.3	46.6

^aBold residue denotes the residue of interest.

Table S6. Experimental Chemical Shifts and Computed Chemical Shieldings for Phe-C γ from the Hydrogen-bond Partner “Supermolecule” Cluster Calculations Using the Hartree-Fock Method^a

Compound ^a	δ_{expt} (ppm)	σ_{calc} (ppm)
1 Phe-Pro	134.1	53.5
2 Phe-Phe	135.3	52.4
3 Phe-Ala	135.6	48.0
11 Phe-Tyr	136.4	52.9
4 Phe-Val	136.9	38.4
9 Tyr-Phe	138.9	40.8
5 Ile-Phe	139.2	37.1
2 Phe-Phe	139.8	40.3
6 Gly-Phe	140.3	41.1

^aBold residue denotes the residue of interest.

Table S7. Experimental Chemical Shifts and Computed Chemical Shieldings for Phe-C γ from the Hydrogen-bond Partner “Supermolecule” Cluster Calculations Using Density Functional Theory^a

Compound ^a	δ_{expt} (ppm)	σ_{calc} (ppm)
1 Phe-Pro	134.1	42.3
2 Phe-Phe	135.3	41.6
3 Phe-Ala	135.6	37.3
4 Phe-Val	136.9	26.9
9 Tyr-Phe	138.9	29.1
5 Ile-Phe	139.2	27.2
2 Phe-Phe	139.8	31.0
6 Gly-Phe	140.3	30.8

^aBold residue denotes the residue of interest.

Table S8. Calculated Chemical Shielding Values (in ppm) for the SCRF Calculations with Hydrogen-bond Partner “Supermolecule” Cluster at Different Dielectric Constants (ϵ).

ϵ	Phe-Pro (1)	Gly-Phe (6)
	ppm	ppm
2.0	53.1	43.1
4.0	52.9	44.3
6.0	52.8	44.8
8.0	52.7	45.0
10.0	52.7	45.2
12.0	52.6	45.3
14.0	52.6	45.4

Table S9. Experimental Chemical Shifts and Computed Chemical Shieldings for Tyr-C γ from the Hydrogen-bond Partner “Supermolecule” Cluster Calculations Using the Hartree-Fock method^a

Compound ^a	δ_{expt} (ppm)	σ_{calc} (ppm)
7 Tyr-Glu	123.2	57.1
8 Tyr-Leu	123.7	64.0
9 Tyr-Phe	124.7	63.6
10 Leu-Tyr	130.4	51.7
11 Phe-Tyr	130.8	48.9

^aBold residue denotes the residue of interest.

Table S10. Statistical Results for HF/DFT Chemical Shift/Shielding Calculations

	Method	Slope	RMSD (ppm)	R
C ^γ (Phe)	CH ^a	-5.32	6.7	0.88
	EX ^b	-3.58	6.2	0.81
	HB-HF ^c	-2.44	4.1	0.81
	HB-DFT ^d	-2.11	4.1	0.79
	HF-SCRF ^e	-1.85	2.4	0.88
	B3LYP-SCRF ^f	-1.66	2.1	0.88
C ^γ (Tyr)	HB-HF ^c	-1.58	4.2	0.84
	HF-SCRF ^e	-1.28	3.1	0.86
	B3LYP-SCRF ^f	-1.24	3.6	0.83
Dipeptides	HF-SCRF ^g	0.99	1.6	0.97
	B3LYP-SCRF ^g	1.00	1.7	0.96
Proteins	HF-SCRF ^g	0.98	1.1	0.97
Overall	HF-SCRF ^g	1.00	1.5	0.96

^a N/C-terminus charged models. ^b Exact crystal structures. ^c Exact crystal structures with surrounding hydrogen bond partner molecules included (HF method). ^d Same as footnote c but using DFT (B3LYP). ^e Same as footnote c but using the SCRF-PCM model and HF method. ^f Same as footnote e but using DFT (B3LYP). ^g using regression lines from equations (1), (2), (3), and (4).

Table S11. Experimental Chemical Shifts (in ppm) and Computed Chemical Shieldings (in ppm) for Phe-C γ and Tyr-C γ in Proteins.

PDB # (amino acid)	BMRB #	Residue #	δ_{expt} (ppm)	HF-SCRF	
				σ_{calc} (ppm)	δ_{pred} (ppm)
1CZP (Phe)	447	3	136.3	44.1	139.0
1CZP (Phe)	447	65	137.2	45.5	138.2
3LZT (Phe)	568	34	137.7	44.1	139.0
3LZT (Phe)	791	38	139.2	43.4	139.4
3LZT (Phe)	791	3	139.5	43.4	139.4
1MO1 (Phe)	5757	34	137.0	44.3	138.9
1MO1 (Phe)	5757	22	137.2	46.3	137.8
1EY0 (Phe)	1704	61	136.2	45.6	138.2
1CZP (Phe)	447	39	136.3	45.7	138.1
1EY0 (Phe)	1704	76	138.4	44.4	138.8
1EY0 (Phe)	1704	34	138.6	42.3	140.0
1CZP (Tyr)	447	25	128.0	55.7	129.9
1CZP (Tyr)	447	82	128.4	55.2	130.3
3LZT (Tyr)	568	20	128.9	59.0	127.3
1CZP (Tyr)	447	35	129.3	54.2	131.1
1EY0 (Tyr)	1704	85	129.9	52.0	132.8
1EY0 (Tyr)	1704	113	130.3	54.5	130.9
3LZT (Tyr)	568	23	130.7	54.4	130.9

Table S12. Experimental Chemical Shifts and Computed Chemical Shieldings for His C^γ , C^{δ^2} and C^{ϵ^1} from the SCRF Calculations with Hydrogen-bond Partner “Supermolecule” Cluster Using the Hartree-Fock Method.

Compound	C^γ		C^{δ^2}		C^{ϵ^1}	
	δ_{expt}	σ_{calc}	δ_{expt}	σ_{calc}	δ_{expt}	σ_{calc}
	(ppm)	(ppm)	(ppm)	(ppm)	(ppm)	(ppm)
His-Leu	124.7	63.1	128.3	58.7	135.5	42.8
His-Met	124.9	61.2	124.9	65.3	135.0	41.6
Gly-His	127.8	55.4	124.0	65.0	133.6	45.1
Leu-His	129.6	55.8	120.1	67.9	133.1	48.4
His-Asp	128.9	56.4	120.6	69.2	132.8	44.8
His-Ala	131.7	52.0	117.4	72.6	136.4	44.3
His-Glu	132.6	50.7	117.6	72.7	140.5	37.7
Ala-His	137.4	47.3	114.5	79.9	135.5	45.3

Table S13. Computed ^{13}C Chemical Shielding Tensor Element Magnitudes for Phenylalanine and Tyrosine Residues in Peptides and Proteins.

Residue	System	σ_{iso} (ppm)	σ_{11} (ppm)	σ_{22} (ppm)	σ_{33} (ppm)	
Phe	1	52.7	-57.0	19.4	195.6	
	2	52.7	-68.8	34.4	192.5	
	3	49.6	-64.7	22.0	191.5	
	11	52.3	-63.1	27.7	192.1	
	4	46.5	-68.3	14.4	193.3	
	9	44.6	-66.4	5.4	194.9	
	5	40.7	-75.2	6.1	191.3	
	2	41.9	-75.0	8.2	192.5	
	6	45.2	-69.0	11.7	192.9	
	1CZP_Phe3	44.1	-66.7	12.9	186.1	
	1CZP_Phe65	45.5	-67.9	18.2	186.1	
	3LZT_Phe34	44.1	-65.3	10.7	186.8	
	3LZT_Phe38	43.4	-67.4	9.9	187.6	
	3LZT_Phe3	43.4	-65.3	8.0	187.4	
	1EY0_Phe61	45.6	-65.0	14.8	186.9	
	1CZP_Phe39	45.7	-66.7	16.2	187.5	
	1EY0_Phe76	44.4	-65.9	14.0	185.0	
	1EY0_Phe34	42.3	-66.6	6.2	187.3	
	Tyr	7	60.8	-49.1	39.9	191.5
		8	64.1	-51.2	50.0	193.5
9		66.0	-45.0	46.8	196.3	
10		56.1	-51.7	28.1	191.8	
11		53.6	-54.5	21.3	193.9	
1CZP_Tyr25		55.7	-50.0	33.3	183.7	
1CZP_Tyr82		55.2	-50.2	29.4	186.5	
3LZT_Tyr20		59.0	-45.7	34.5	188.1	
1CZP_Tyr35		54.2	-51.1	24.8	188.9	
1EY0_Tyr85		52.0	-51.5	20.8	186.7	
1EY0_Tyr113		54.5	-52.2	30.0	185.6	
3LZT_Tyr23		54.4	-48.7	24.0	188.0	
3LZT_Tyr53		44.6	-57.4	11.5	179.7	

Table S14. Experimental Shift Values and NBO Charges of C γ for Phenylalanine and Tyrosine Residues in Peptides and Proteins.

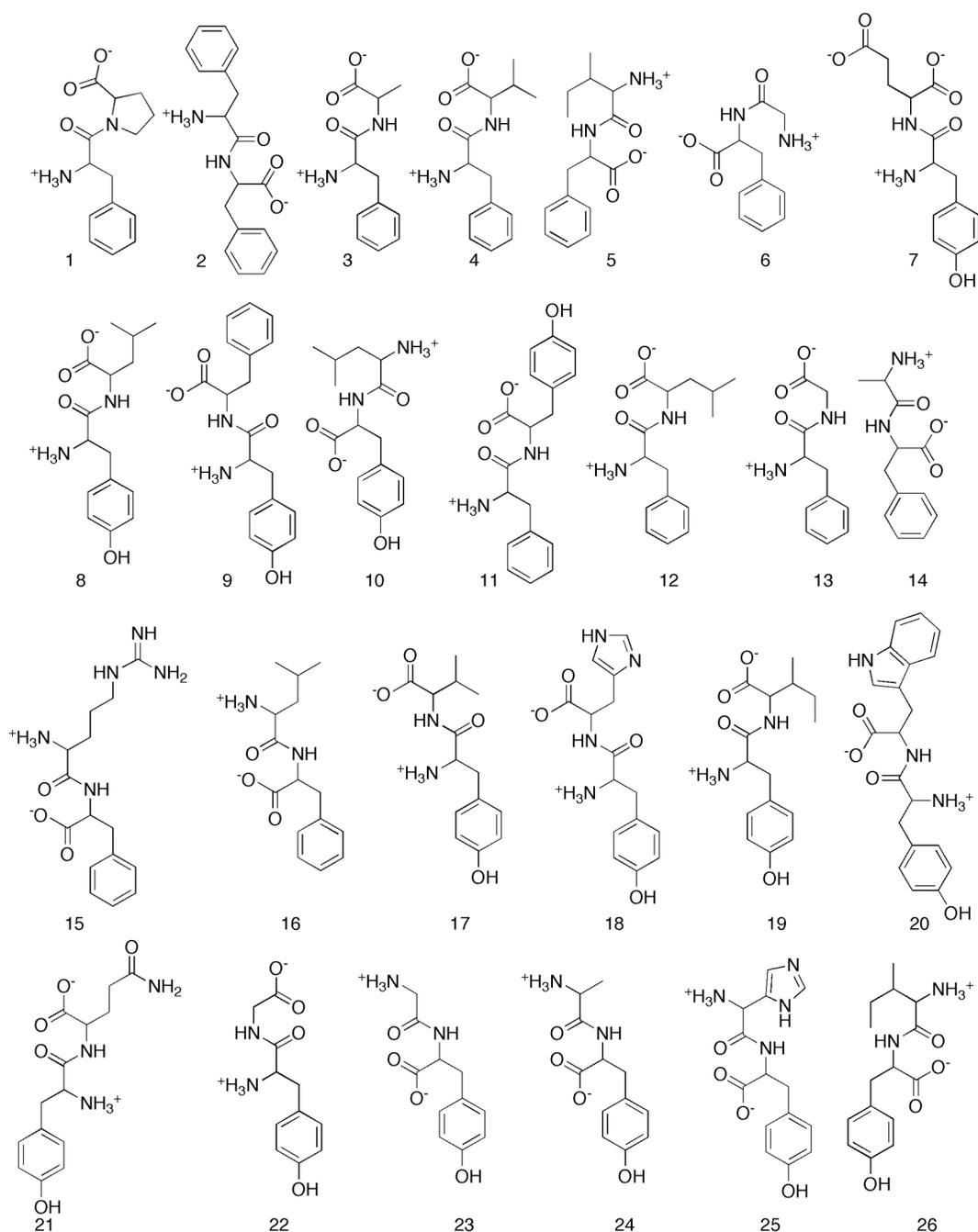
Residue	System	δ_{expt} (ppm)	Q_{NBO} (e)
Type			
Phe	1	134.1	-0.0313
	2	135.3	-0.0422
	3	135.6	-0.0361
	11	136.4	-0.0306
	4	136.9	-0.0094
	9	138.9	-0.0073
	5	139.2	-0.0046
	2	139.8	0.0140
	6	140.3	0.0056
	1CZP_Phe3	136.3	-0.0278
	1CZP_Phe65	137.2	-0.0367
	3LZT_Phe34	137.7	-0.0343
	3LZT_Phe38	139.2	-0.0142
	3LZT_Phe3	139.5	-0.0284
	1MO1_Phe34	137	-0.0237
	1MO1_Phe22	137.2	-0.0332
	1EY0_Phe61	136.2	-0.0304
	1CZP_Phe39	136.3	-0.0361
	1EY0_Phe76	138.4	-0.0305
	1EY0_Phe34	138.6	-0.028
Tyr	7	123.2	-0.0866
	8	123.7	-0.1022
	9	124.7	-0.1096
	10	130.4	-0.0616
	11	130.8	-0.0455
	1CZP_Tyr25	128.0	-0.0799
	1CZP_Tyr82	128.4	-0.0777
	3LZT_Tyr20	128.9	-0.099
	1CZP_Tyr35	129.3	-0.0774
	1EY0_Tyr85	129.9	-0.0722
	1EY0_Tyr113	130.3	-0.0763
	3LZT_Tyr23	130.7	-0.0807

Table S15. Calculated ^{13}C Shielding Values and NBO Charges for Aromatic Ring Systems.

Compound	σ_{calc} (ppm)	Q_{NBO} (e)
Cyclobutadiene dication	-61.9	0.1798
Cyclopropenium	23.9	0.0408
Tropylium	31.9	-0.0931
Benzene	58.2	-0.2040
Cyclononatetraenyl	80.5	-0.2558
Cyclopentadienide	88.3	-0.3731
Cyclooctatetraenyl dianion	101.8	-0.3941

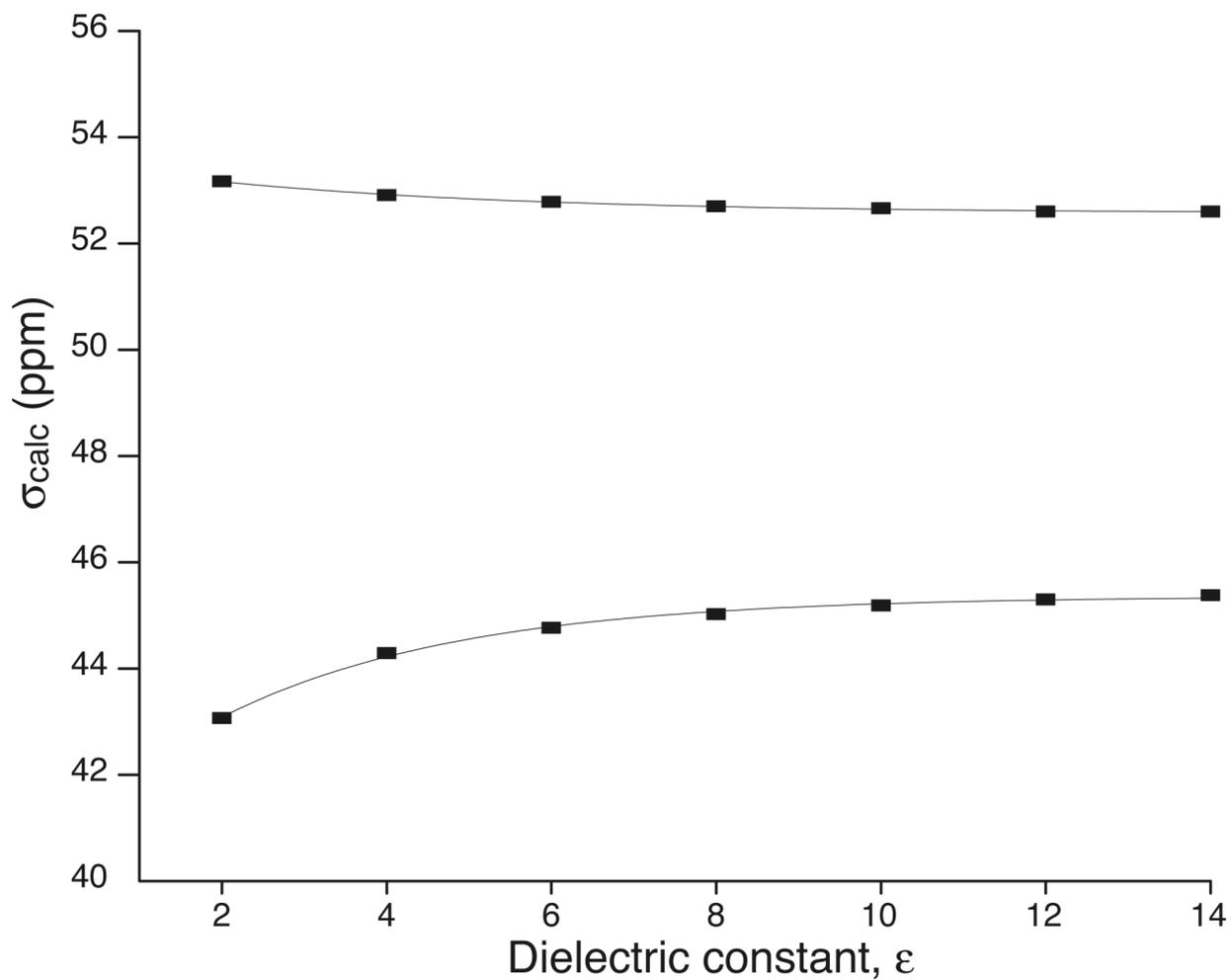
Table S16. Calculated ^{13}C Shielding Values and NBO Charges for C1 in Aromatic Ring Systems.

Compound	σ_{calc} (ppm)	Q_{NBO} (e)
Methylcyclobutadiene dication	-28.2	0.3726
Methylcyclopropenium	13.1	0.2238
Methyltropylium	12.4	0.1040
Toluene	46.1	-0.0275
Methylcyclononatetraenyl	72.1	-0.1026
Methylcyclopentadienide	73.6	-0.1774
Methylcyclooctatetraenyl dianion	97.2	-0.2188



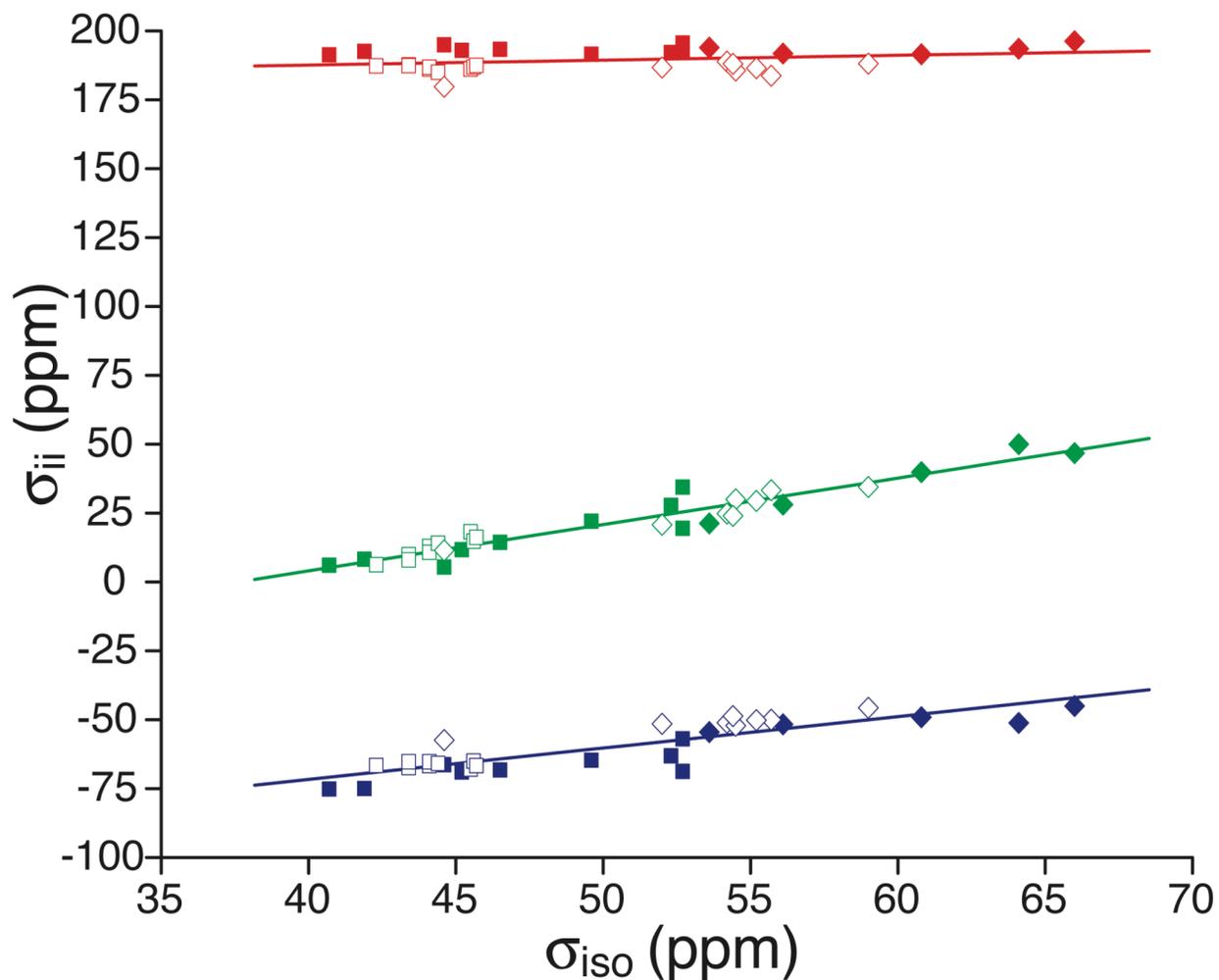
Mukkamala et al., Supplementary Figure S1.

Structures of the phenylalanine and tyrosine-containing dipeptides investigated. 1, Phe-Pro; 2, Phe-Phe; 3, Phe-Ala; 4, Phe-Val; 5, Ile-Phe; 6, Gly-Phe; 7, Tyr-Glu; 8, Tyr-Leu; 9, Tyr-Phe; 10, Leu-Tyr; 11, Phe-Tyr; 12, Phe-Leu; 13, Phe-Gly; 14, Ala-Phe; 15, Arg-Phe; 16, Leu-Phe; 17, Tyr-Val; 18, Tyr-His; 19, Tyr-Ile; 20, Tyr-Trp; 21, Tyr-Gln; 22, Tyr-Gly; 23, Gly-Tyr; 24, Ala-Tyr; 25, His-Tyr; 26, Ile-Tyr.



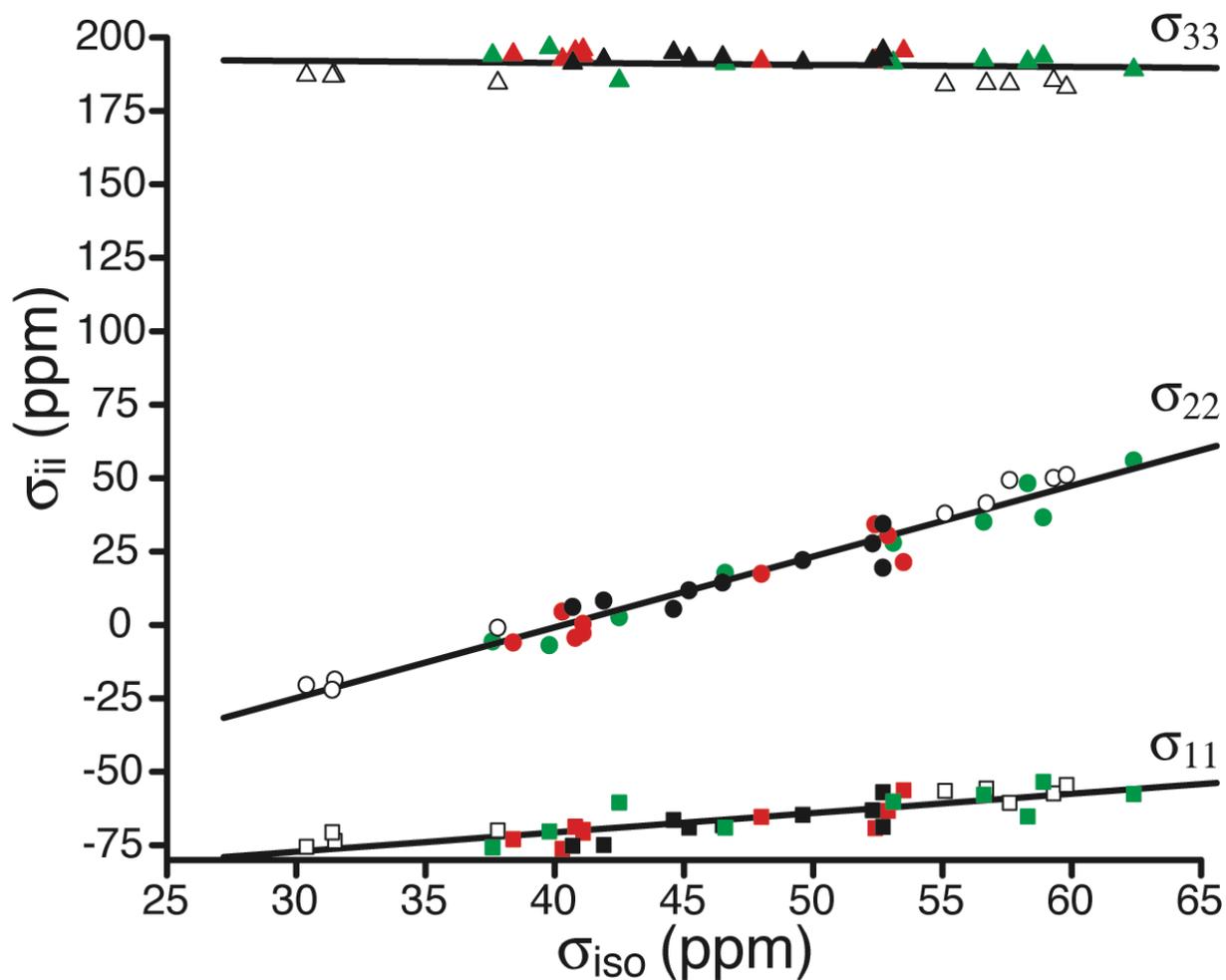
Mukkamala et al., Supplementary Figure S2.

Plot of the calculated chemical shielding of **Phe-Pro (1)** and **Gly-Phe (6)** vs. the dielectric constant used in the HF-SCRF calculation.



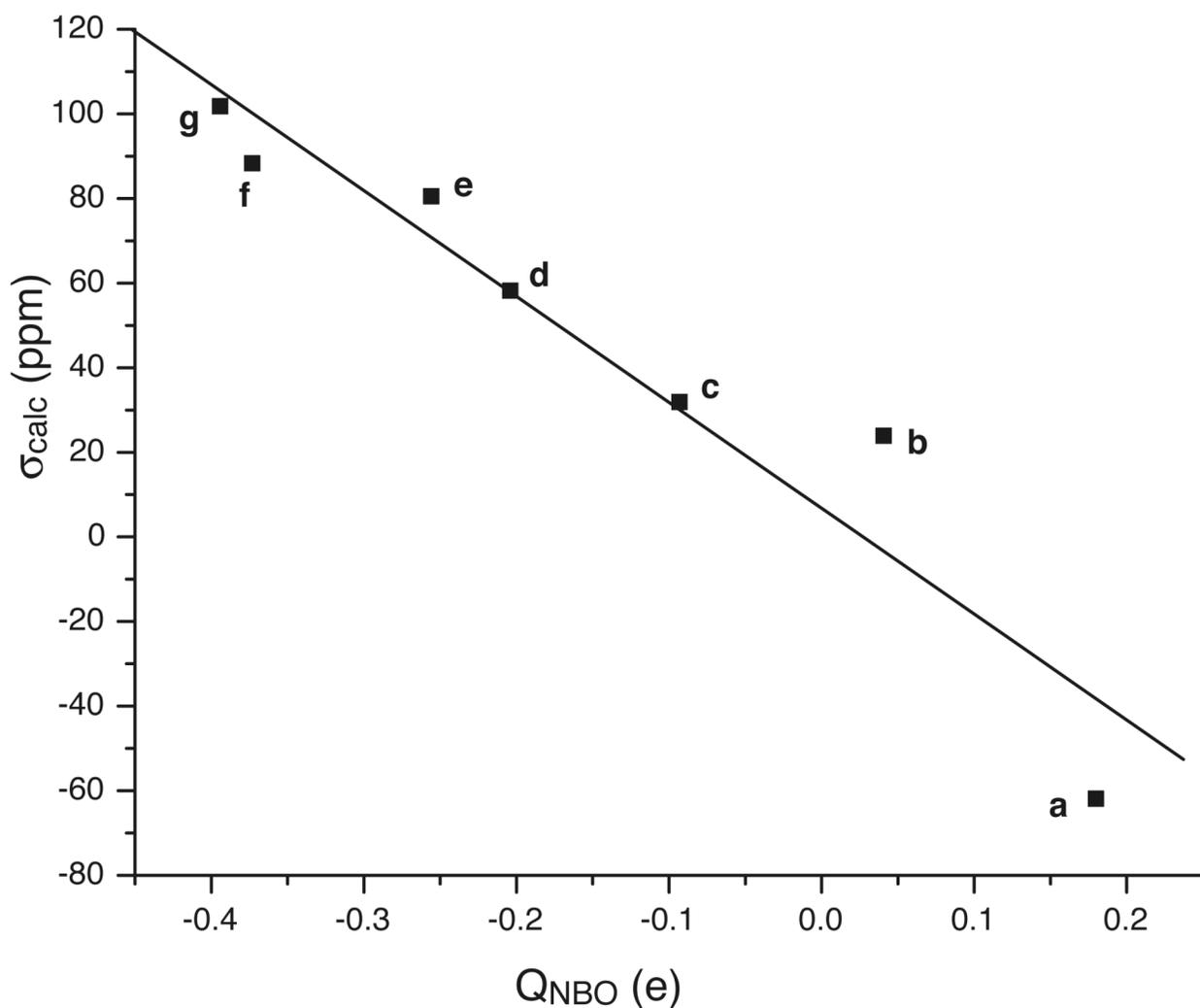
Mukkamala et al., Supplementary Figure S3.

Computed shielding tensor element magnitudes as a function of the calculated C' isotropic shielding: filled squares - σ_{11} , σ_{22} and σ_{33} of Phe C' in dipeptides; open squares - σ_{11} , σ_{22} and σ_{33} of Phe C' in proteins; filled diamonds - σ_{11} , σ_{22} and σ_{33} of Tyr C' in dipeptides; open diamonds - σ_{11} , σ_{22} and σ_{33} of Tyr C' in proteins.



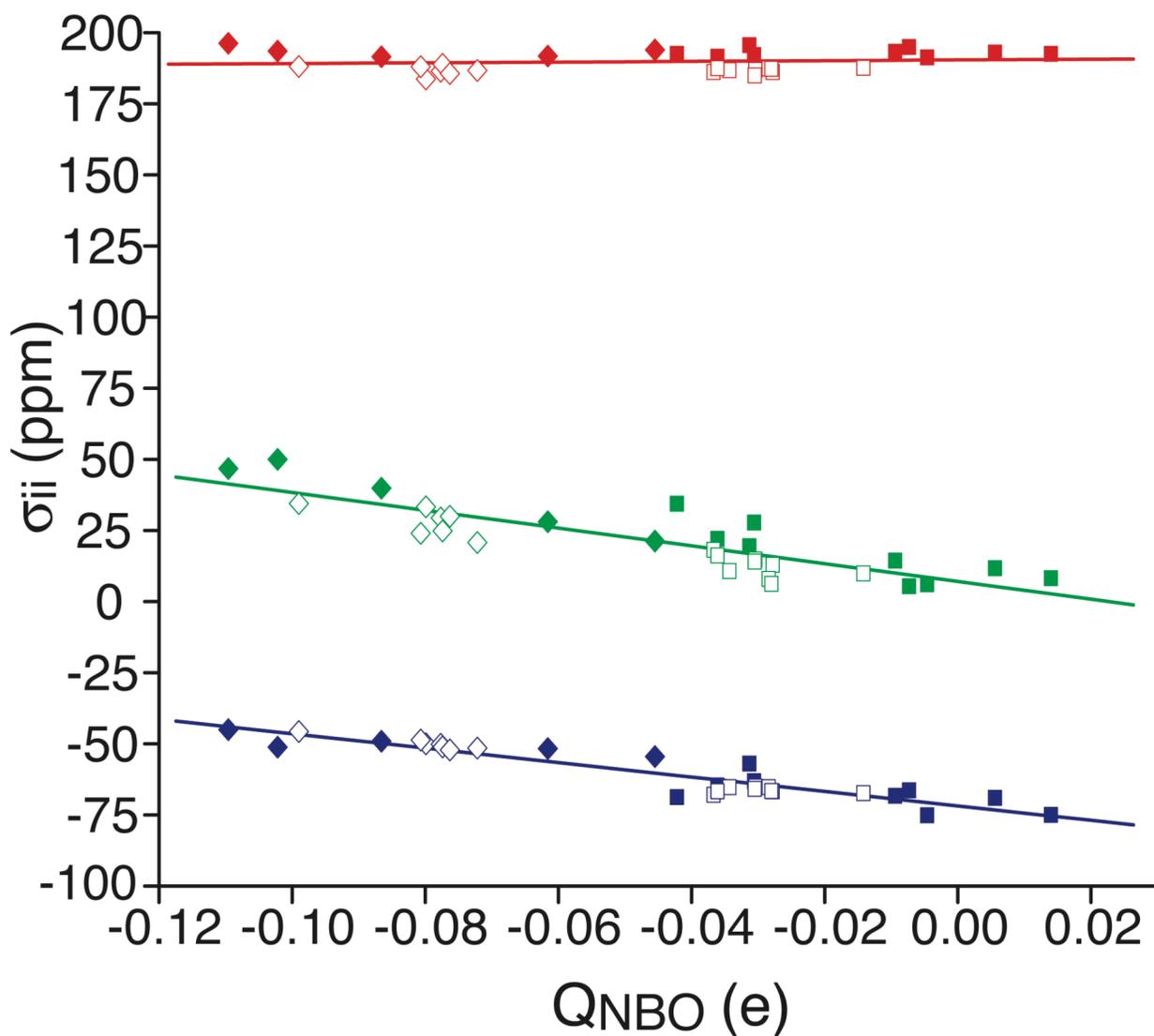
Mukkamala et al., Supplementary Figure S4.

C^{13} shielding tensor magnitudes as a function of the isotropic shielding for different computational models: open symbols - charged models; green symbols - crystal structures without hydrogen-bond partners; red symbols- hydrogen bond partner supermolecule clusters without a reaction field; black symbols - HF-SCRF.



Mukkamala et al., Supplementary Figure S5.

Plot of the calculated C^γ chemical shieldings versus carbon NBO charges of the aryl carbons of various aromatic ring systems (a, Cyclobutadiene dication; b, Cyclopropenium; c, Tropylium; d, Toluene; e, Cyclononatetraenyl; f, Cyclopentadienyl; g, Cyclooctatetraenyl dianion).



Mukkamala et al., Supplementary Figure S6.

Computed shielding tensor element magnitudes plotted as a function of C' NBO charge: filled squares - σ_{11} , σ_{22} and σ_{33} of Phe C' in dipeptides; open squares - σ_{11} , σ_{22} and σ_{33} of Phe C' in proteins; filled diamonds - σ_{11} , σ_{22} and σ_{33} of Tyr C' in dipeptides; open diamonds - σ_{11} , σ_{22} and σ_{33} of Tyr C' in proteins.