

Iodination of aromatic residues in peptides by reaction with IPy₂BF₄

José Barluenga,^{*a} Miguel A. García-Martín,^a José M. González,^a Pere Clapés^b and Gregorio Valencia^b

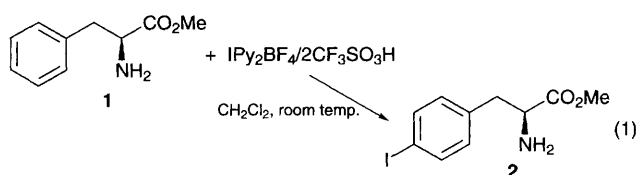
^a Instituto Universitario de Química Organometálica 'Enrique Moles' -Unidad Asociada al CSIC, C/Julián Clavería 8, Universidad de Oviedo, 33071 Oviedo, Spain

^b Unit for Protein Chemistry and Biochemistry, Centro de Investigación y Desarrollo-CSIC, C/Jordi Girona 18-26, 08034 Barcelona, Spain

A simple and straightforward approach to selectively iodinate Tyr residues on peptides using IPy₂BF₄ is described which is compatible with Met residues on the side chain; preliminary studies reveal that the method is also suitable to iodinate Phe derivatives.

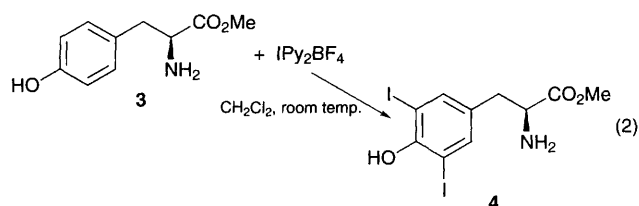
Iodine isotopes are routinely employed in medical nuclear imaging and radioimmunoassays.¹ Single photon emission computed tomography (SPECT) is a useful technique for brain studies² and relies mainly upon Tc-99m labelled agents,³ although I-123 is also well suited for this purpose.⁴ Peptides are ligands of potential interest for this computerized clinical procedure, however their labelling by electrophilic iodination⁵ requires an oxidizing agent that can be harmful to the peptide; thus, indirect⁶ and conjugation based⁷ approaches have become very popular.

We have previously reported that IPy₂BF₄ efficiently iodates arenes under mild conditions, a process that shows wide scope.⁸ Here we report preliminary results concerning the use of this reagent in the iodination of the aryl residues of some amino acids, and the selectivity observed for its reaction with small peptides that could lead to a convenient approach for their direct labelling. First, we established suitable conditions to iodinate amino acid derivatives. The methyl ester of phenylalanine **1** was ring iodinated at room temperature in CH₂Cl₂ by treatment with IPy₂BF₄ and CF₃SO₃H (1 : 1.1 : 2.2 molar ratio, where acid is added to formally liberate I⁺ in solution by precipitation of PyH⁺), furnishing the 4-iodophenylalanine methyl ester **2** in 70% isolated yield [eqn. (1)]. Minor amounts of the correspond-

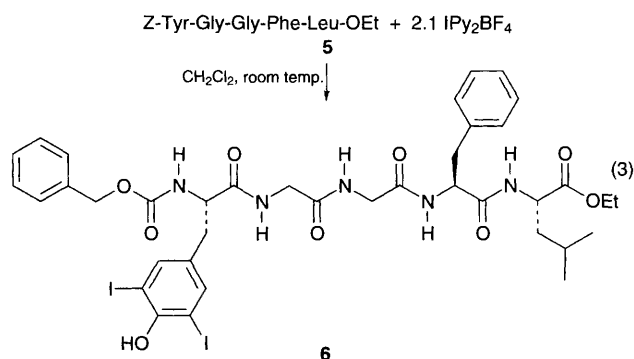


ing *ortho*-isomer are also formed, as evidenced by ¹H and ¹³C NMR analyses of the crude reaction mixture prior to purification. Importantly, this fact proves that iodination of phenylalanine can be directly accomplished without requiring previous activation of the C-4 ring position.⁹ Tyrosine **3** can also be easily iodinated using this approach and, remarkably, without the need for addition of triflic acid to liberate the electrophile. In this regard, the reaction of equimolar amounts of IPy₂BF₄ and **3** furnishes mixtures of the mono- and di-iodinated derivatives; moreover, the diiododerivative **4** is quantitatively obtained after mixing the reagents in CH₂Cl₂ at room temperature, but using 2 : 1 molar ratio of IPy₂BF₄ and **3** respectively, [eqn. (2)].

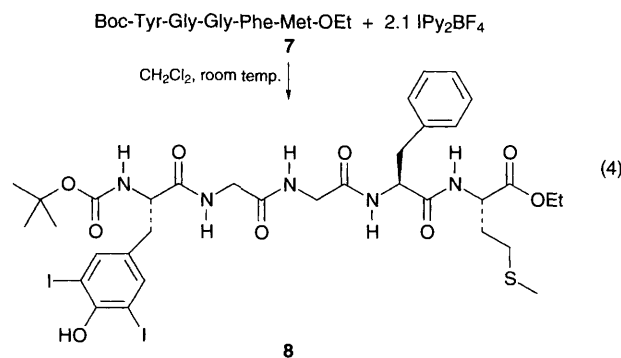
Furthermore, iodination of a Leu-enkephalin derivative **5** (Z-Tyr-Gly-Gly-Phe-Leu-OEt) follows the same pattern as the amino acids, with either CH₂Cl₂ or Me₂SO as solvent. Z-Leu-enkephalin-OEt reacts at room temperature with IPy₂BF₄



(molar ratio 1 : 2.1, respectively) giving instantaneously the diiodo peptide **6**,[†] in quantitative yield [eqn. (3)]. Starting from



a [²H]₆Me₂SO solution of **5** of known concentration, adding gradually weighed amounts of IPy₂BF₄ into the NMR tube, and analysing changes of diagnostic aryl signals allows the progress of the reaction to be monitored by ¹H NMR. With a 1 : 1 molar ratio peptide : iodinating reagent, a mixture containing unreacted starting material (13%) and the mono- and di-iodinated (68 and 19% respectively) compounds was formed. Nevertheless, the starting material essentially disappeared after addition of 1.4 equiv. of IPy₂BF₄, giving rise to the formation of the mono- and di-iodinated compounds. Parallel results were found using HPLC analysis for this clean transformation. Finally, the iodination of the related Boc-Met-enkephalin-OEt derivative **7** (Boc-Tyr-Gly-Gly-Phe-Met-OEt) with IPy₂BF₄ was examined to analyse the effect of the reagent upon a side chain containing a sensitive amino acid [eqn. (4)]. Using a



2.1:1 excess of IPy₂BF₄ with respect to the peptide furnishes **8**,[‡] corresponding again to the diiodination of the aryl residue of tyrosine, in 73% yield (recovered starting material accounted for the remainder of the material). The formation of by-products arising from oxidation of sulfur on the Met residue was ruled out by HPLC analysis of an oxidized sample obtained after treatment of the peptide with H₂O₂.

Footnotes

† Compound **6** was purified by HPLC. MS analysis for **6** (MALDI TOF) shows *m/z* = 992 for [M + Na]⁺, in agreement with the proposed structure.

‡ Compound **8** was purified by HPLC. ESI-MS for **8** shows *m/z* = 954, [M + H]⁺, accounting well for the proposed structure.

References

- 1 R. H. Seevers and R. E. Counsell, *Chem. Rev.*, 1982, **82**, 575.
- 2 M. I. Posner, *Science*, 1993, **262**, 673; G. B. Saha, W. J. MacIntyre and R. T. Go, *Semin. Nucl. Med.*, 1994, **24**, 324.

- 3 S. Meegalla, K. Plössl, M.-P. Kung, D. A. Stevenson, L. M. Liable-Sands, A. L. Rheingold and H. F. Kung, *J. Am. Chem. Soc.*, 1995, **117**, 11 037.
- 4 J. L. Neumeyer, S. Wang, R. A. Milius, R. M. Baldwin, Y. Zea-Ponce, P. B. Hoffer, E. Sybirska, M. Al-Tikriti, D. S. Charney, R. T. Malison, M. Laruelle and R. B. Innis, *J. Med. Chem.*, 1991, **34**, 3144.
- 5 G. S. Bailey, in *Methods in Mol. Biol.*, ed. J. M. Walker, Humana; Towota, NJ, 1994, vol. 32, ch. 45, pp. 441–448.
- 6 (a) S. D. Sharma, G. Toth and V. J. Hruby, *J. Org. Chem.*, 1991, **56**, 4981; (b) M. Ceusters, D. Tourwé, J. Callaerts, J. Mertens and A. Peter, *J. Org. Chem.*, 1995, **60**, 8324.
- 7 A. E. Bolton and W. M. Hunter, *Biochem. J.*, 1973, **133**, 529; Z.-G. Zhao and K. S. Lam, *J. Chem. Soc., Chem. Commun.*, 1995, 1739.
- 8 J. Barluenga, J. M. González, M. A. García-Martín, P. J. Campos and G. Asensio, *J. Org. Chem.*, 1993, **58**, 2058.
- 9 S. R. Wilson and L. A. Jacob, *J. Org. Chem.*, 1986, **51**, 4833.

Received, 11th April 1996; Com. 6/02578A