Synthesis of novel imidazolidinones from hexose–peptide adducts: model studies of the Maillard reaction with possible significance in protein glycation

Štefica Horvat,*† Lidija Varga-Defterdarović, Maja Roščić and Jaroslav Horvat

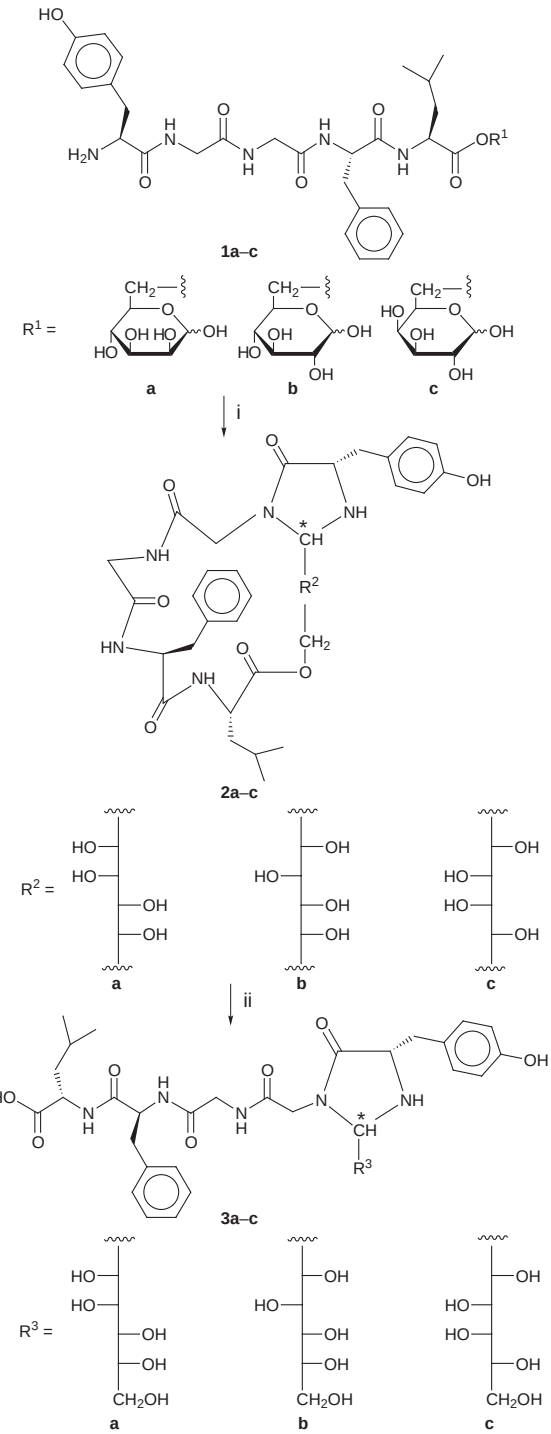
Department of Organic Chemistry and Biochemistry, Ruđer Bošković Institute, POB 1016, 10001 Zagreb, Croatia

Novel hexose-related imidazolidin-4-ones are prepared by intramolecular rearrangements of monosaccharide esters in which D-mannose, D-glucose, or D-galactose are linked through their C-6 hydroxy groups to the endogenous opioid pentapeptide, leucine-enkephalin.

Non-enzymatic glycation of proteins has been increasingly identified as an important factor in the age-dependent chemical modification and cross-linking of tissue proteins,[1] as well as in the pathogenesis of the long term complications of diabetes.[2] The sequence of non-enzymatic glycation involves the reaction of an aldose (or ketose) sugar with free amino groups of proteins (Maillard reaction) leading initially to the formation of labile Schiff bases. With the subsequent Amadori rearrangement, the aminoketoses formed undergo a variety of reactions[3] such as dehydrations, fragmentations, and the formation of highly reactive carbonyl compounds, which continue to react with free amino groups, thus leading to cross-linking of proteins *via* advanced glycation end products (AGEs) which are mainly responsible for the diabetic complications.[2] A few such adducts have been structurally identified by *in vitro* glycation of proteins or by monitoring the breakdown of isolated Amadori compounds.[4]

This study demonstrates for the first time, by *in vitro* experiments, that, in addition to Amadori rearrangement, an alternative pathway for carbohydrate-induced modification of peptides is possible, yielding imidazolidin-4-ones from the initially formed hexose–peptide adducts.

We recently reported[5] on the products of intramolecular Amadori rearrangements formed in pyridine–AcOH as the solvent from monosaccharide esters **1a**–**c**[6] in which D-mannose (**1a**), D-glucose (**1b**), or D-galactose (**1c**) are linked through their C-6 hydroxy groups to the C-terminal carboxy group of the endogenous opioid pentapeptide leucine-enkephalin (H-Tyr-Gly-Gly-Phe-Leu-OH).[7] Herein we provide evidence that synthons **1a**–**c** are also transformed to carbohydrate-related imidazolidinones **3a**–**c** (Scheme 1). Thus, incubation of monosaccharide esters **1a**–**c** in MeOH at 50–60 °C afforded, after purification by semipreparative reverse-phase high-performance liquid chromatography (RP HPLC), the corresponding imidazolidin-4-ones **2a**–**c** in acceptable yields (Table 1). We assume that, in a similar fashion to Amadori product formation from esters **1a**–**c**,[5] the first step of this reaction requires the open-chain form of the carbohydrate moieties in compounds **1a**–**c**, which is attacked by the free amino terminus of the peptide moiety. In the subsequent step, the Schiff base formed, instead of Amadori rearrangement to the corresponding keto-sugar, undergoes nucleophilic attack by the Gly[2] nitrogen to yield imidazolidin-4-ones **2a**–**c** in which C-1 of the sugar moiety forms a bridge between the α-amino group of the *N*-terminal tyrosine residue and the amide nitrogen of the Tyr[1]-Gly[2] peptide bond. As presented in Table 1, the transformation **1a**→**2a** took place completely stereospecifically, while the conversion of esters **1b** and **1c** to the corresponding imidazolidinones exhibited a low degree of stereocontrol. Rearrangements of **1a**–**c** in MeOH yield only 5% of the corresponding Amadori products.



Scheme 1 *Reagents and conditions*: i, MeOH, 50–60 °C; ii, 0.1 M NaOH, 25 °C. Asterisk indicates either (*S*)- or (*R*)-configuration at the new *N*,*N*'-acetal centre.

Table 1 Intramolecular rearrangement of esters **1a**–**c** in MeOH

Starting compound	*t*/h	*T*/°C	Product[a]	Yield (%)[b] Major	Minor
1a	24	50	**2a**	49	—
1b	72	60	**2b**	34	9
1c	48	60	**2c**	12	10

[a] Mixture of diastereomers for **2b** and **2c**. [b] Isolated yields after RP HPLC purification.

Cleavage of the ester bond in both the major and minor isomers of compounds **2a**–**c** was carried out in 0.1 M NaOH at room temperature and led to the corresponding chiral imidazolidin-4-ones of D-mannose (**3a**), D-glucose (**3b**) or D-galactose (**3c**) in 77–95% yield after RP HPLC chromatography.‡

The experimental fact that the monosaccharide esters **1a**–**c**, the behaviour of which closely resembles the reactivity of hexose 6-phosphates, yield either the corresponding Amadori products[5] or imidazolidinones **2a**–**c** points, in our understanding, to the possibility that, depending on the physiological environment, similar imidazolidinon-4-one adducts may be also generated *in vivo*.

In conclusion, the method outlined above, which is based on the intramolecular rearrangement of 6-*O*-peptidyl esters **1a**–**c,** represents an innovative route for the synthesis of hexose-related imidazolidin-4-ones, compounds useful in understanding the details of the mechanism of non-enzymatic glycation *in vivo*.

This work was funded by the Ministry of Science and Technology of Croatia (Grant No. 00980704).

Notes and References

† E-mail: shorvat@rudjer.irb.hr

‡ All new compounds have been fully characterized and have spectroscopic properties compatible with the structures assigned.

1 R. G. Paul and A. J. Bailey, *Int. J. Biochem. Cell Biol.*, 1996, **28**, 1297; R. Sullivan, *Arch. Physiol. Biochem.*, 1996, **104**, 797; D. R. Sell, *Mech. Ageing Dev.*, 1997, **95**, 81.

2 M. Brownlee, *Diabetes*, 1994, **43**, 836; E. Schleicher and A. Nerlich, *Horm. Metab. Res.*, 1996, **28**, 367; H. Vlassara, *Diabetes*, 1997, **46** (Suppl. 2), S19.

3 V.A. Yaylayan and A. Huyghues-Despointes, *Crit. Rev. Food Sci. Nutr.*, 1994, **34**, 321; F. Ledl and E. Schleicher, *Angew. Chem., Int. Ed. Engl.*, 1990, **29**, 565.

4 V. M. Monnier, D. R. Sell, R. H. Nagaraj, S. Miyata, S. Grandhee, P. Odetti and S. A. Ibrahim, *Diabetes*, 1992, **41** (Suppl. 2), 36; P. R. Smith and P. J. Thornalley, *Eur. J. Biochem.*, 1992, **210**, 729; A. Cerami, in *Maillard Reactions in Chemistry, Food and Health*, ed. T. P. Labuza, G. A. Reineccius, V. M. Monnier, J. O'Brien and J. W. Baines, The Royal Society of Chemistry, Cambridge, England, 1994, pp. 1–10.

5 Š. Horvat, M. Roščić, L. Varga-Defterdarović and J. Horvat, *J. Chem. Soc., Perkin Trans. 1*, 1998, 909.

6 Š. Horvat, J. Horvat, D. Kantoci and L. Varga, *Tetrahedron*, 1989, **45**, 4579; Š. Horvat, L. Varga-Defterdarović, J. Horvat, S. Modrić-Žganjar, N. N. Chung and P. W. Schiller, *Lett. Pept. Sci.*, 1995, **2**, 363.

7 G. A. Olson, R. D. Olson and A. J. Kastin, *Peptides*, 1996, **17**, 1421.

Receieved in Glasgow, UK, 24th April 1998; 8/03099E