Amino Acids (1999) 16: 381–389

Amino
Acids

Synthesis of perfluoroalkylated β-alanine and some peptide derivatives: an access to original surfactants

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Accepted April 21, 1998

Summary. The reaction of amines or sodium azide with 3-perfluoroalkyl-3-fluoroprop-2-enoate, followed by hydrogenation, affords perfluoroalkylated β-alanine analogues in very good yields. These compounds can be linked via an amide bond to produce peptide analogues such as carnosine or carcinine derivatives, which could have surfactive and complexing properties.

Keywords: Amino acids – Perfluorinated compounds – Surfactants – Complexing properties – Carnosine – β – Amino acids – Blood substitutes

Introduction

Numerous medical applications involving fluorocarbons and highly fluorinated compounds are under investigation and organofluorine chemistry has had a marked impact on the design and synthesis of many kinds of biologically active molecules such as steroids, carbohydrates, amino acids, peptides and other natural products (Ojima et al., 1996; Welch and Eswarakrishnan, 1991). In particular, the development of the chemistry of fluoro-amino acids and related compounds has been one of the major achievements of fluorobioorganic chemistry in the past two decades (Kukhar and Soloshonok, 1995; Resnati and Soloshonok, 1996). Advances in fluorine-related amino acid chemistry have revealed that these artificial amino acids are biologically highly active molecules with a wide range of potential applications (Greiner et al., 1995; Riess, 1995).

Moreover, in recent years, there has been a growing interest in β-amino acid synthesis (Juaristi, 1997; Drey, 1985). β-Amino acids are major constituents in many classes of natural products. Therefore their synthesis should pave the way for the development of new pharmaceutical compounds.

However, most of the research in this field has focused on the chemistry of fluorinated α-amino acids (Kukhar and Soloshonok, 1995). Only a few methods for the synthesis of fluorine-containing β-amino acids are available.

In the present paper, perfluoroalkylated peptides containing β-alanine, in particular analogues of carnosine or carcinine will be investigated. These compounds are potential surfactive, antioxydant and complexing agents. Furthermore, perfluoroalkylated surfactants are generally both more effective and more efficient than their hydrogenated counterparts: compared to the latter, perfluoroalkylated surfactants induce lower surface-tensions at lower concentrations (Fielding, 1978).

Besides, they could be equally useful for the oxygenation of biological media. Thus, fluorocarbons are under close scrutiny for temporary blood replacement together with more specific applications such as medical imaging, cancer therapy, organ preservation, etc. (Riess, 1994, 1995). They may also supply oxygen to immobilized cells (Cho and Wang, 1988; King et al., 1989). However, owing to their great hydrophobicity these compounds have to be emulsified in order to be used in biological aqueous media (Mattiasson and Adlercreutz, 1987; Nivet et al., 1991). Consequently, we have considered the preparation of perfluorinated surfactants.

On the other hand, the presence of a large amount of oxygen in biological systems generates peroxides and toxic free radicals (Imlay and Linn, 1988). To avoid this risk, we have introduced carnosine as an hydrophilic module. The peptidoamine carnosine is a naturally occuring histidine-containing compound (Quinn et al., 1992). It is to be found in various tissues, notably in muscle tissues.

Numerous studies have demonstrated that carnosine displays strong and specific antioxydant properties (Boldyrev, 1993; Brown, 1981). Carnosine and related dipeptides such as carcinine have been shown to prevent model membrane system peroxidation. They may therefore be regarded as water soluble counterparts to lipid-soluble antioxidants such as α-tocopherol which protect cell membranes from oxydative damage (Babizhayev et al., 1994). Carnosine is particularly well known to form antioxidant complexes with copper ions (Babizhayev et al., 1994).

The perfluorinated module has been introduced here in the β-position not to interfere with the complexing properties of the pseudopeptide. Thus, the amine function remains available for complexation.

The approach of the present study involves two stages: i) synthesizing perfluoroalkylated β-alanine analogues with or without substituents at the nitrogen atom; ii) preparing peptides or peptidoamine derivatives.

Result and discussion

The synthesis begins with the preparation of the perfluoroalkylated β-alanine analogues.

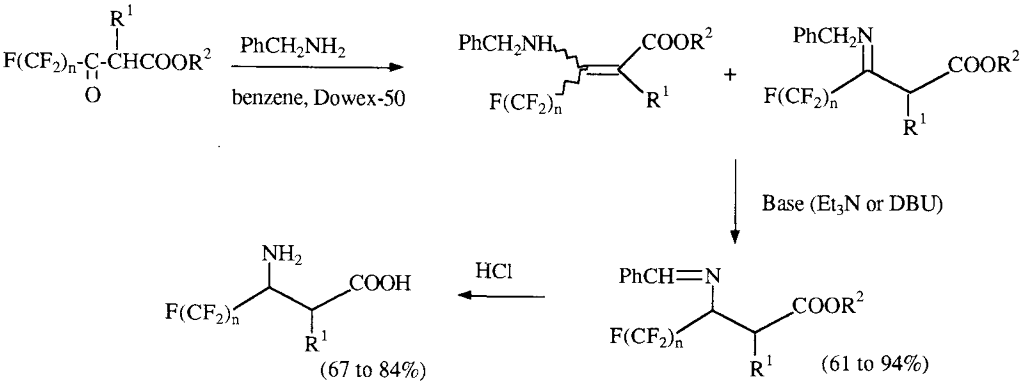
Comparatively few studies cover this subject. Soloshonok developed a β-fluoroalkyl-β-amino acid synthesis from β-perfluoroalkyl-β-ketoesters (Soloshonok et al., 1993). These compounds yielded the corresponding enamines after benzylamine treatment. The Schiff bases obtained after isomerization were hydrolysed by HCl to produce *β*-perfluoroalkyl-*β*-amino acids (Scheme 1).

Likewise, he recently developed (Soloshonok et al., 1997) a methodology for preparing enantiomerically pure *β*-alanines via the enantioselective biomimetic transamination of *β*-ketocarboxylic acid derivatives (Scheme 2).

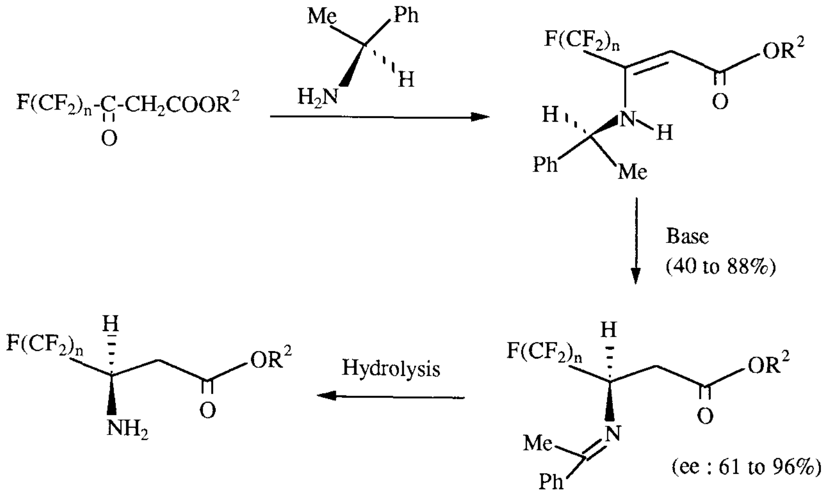
Cen reported on a synthesis of fluorinated enamines by reacting fluoroalkynes with amines or ammonia in alcoholic solution (Cen et al., 1995). The fluorine-containing enamines were hydrogenated to yield the precursor of fluoroalkyl-*β*-alanine (Scheme 3).

However, in both cases, the method has been described for compounds containing relatively short perfluorinated chains.

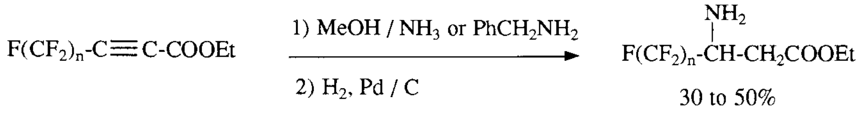
The process suggested in this paper involves the introduction of an amine or an azide moiety into the 3-perfluoro-3-fluoropropenoate **3** prior to



Scheme 1. Synthesis of *β*-polyfluoroalkyl-*β*-aminoacids by transamination of fluorinated ketocarboxylic esters (Soloshonok, 1993)



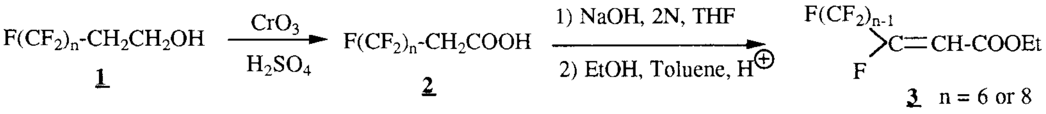
Scheme 2. Enantioselective synthesis of analogues of *β*-alanine (Soloshonok, 1997)



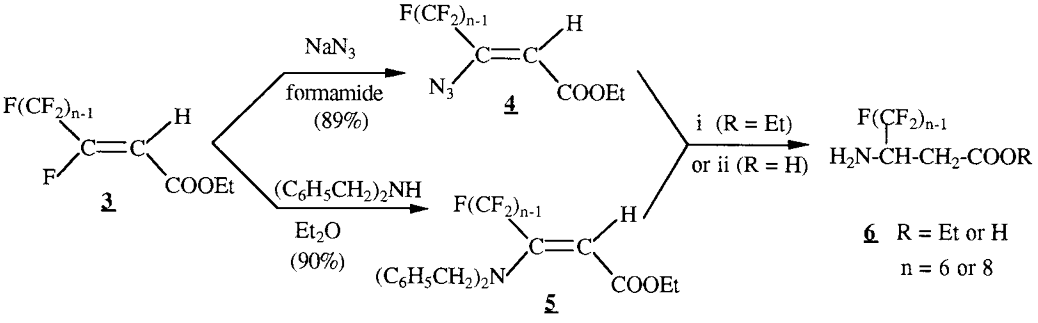
$$F(CF_2)_n\text{-}C{\equiv}C\text{-}COOEt \xrightarrow[\text{2) } H_2\text{, Pd / C}]{\text{1) MeOH / }NH_3\text{ or }PhCH_2NH_2} F(CF_2)_n\text{-}\overset{NH_2}{\overset{|}{C}}H\text{-}CH_2COOEt$$

30 to 50%

Scheme 3. Synthesis of perfluoroalkylated β-aminoacids from fluoroalkynes (Cen, 1995)



Scheme 4. Synthesis of the perfluoroalkenoic ester **3**



i : H_2 / Raney Ni (70 to 80%)

ii : a) NaOH, 2N, dioxane, b)$H_3O^{\oplus}$, c) H_2 / Raney Ni (60 to 65%)

Scheme 5. Synthesis of perfluoroalkylated analogues of β-alanine

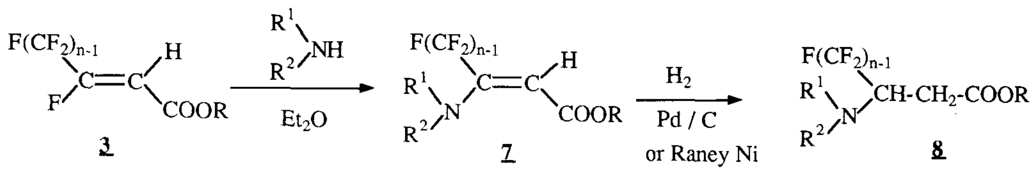
hydrogenation (Özer et al., 1997 and Özer, 1998). This study falls within the scope of a broader research project dealing with the reactivity of perfluoroalkylated ethanoic acids and esters in the presence of nucleophiles such as alcohols or amines (Thiebaut et al., 1995, 1997).

The starting material 3-perfluoroalkyl-3-fluoropropenoate **3** is prepared as described in an earlier paper (Achilefu et al., 1995). The method used is well-known in our laboratory. It begins with the oxidation of 2-perfluoroalkyl ethanols under particular Jones conditions (Scheme 4).

Subsequent sodium azide or dibenzylamine condensation leads to compounds without substituents at the nitrogen atom (Scheme 5).

The sodium azide mechanism comprises certainly two steps: i) Michael addition at the double bond and ii) fluoride elimination (Thiebaut et al., 1997).

The alkaline hydrolysis is straightforward and the hydrogenation of the compounds **4** or **5** produces the perfluorinated β-alanine analogues **6** in good yields, but drastic reaction conditions are required (80 bar of hydrogen pressure, 80°C).



Scheme 6. Synthesis of precursors of perfluoroalkylated N-alkylated β-alanine analogues

Table 1. Synthesis of enamines **7** and hydrogenated compounds **8**

Series	n	R	R^1	R^2	**7** yield %	Z/E	**8** yield %
a	8	CH_3	H	C_4H_9	95	100/0	
b	6	C_2H_5	H	C_6H_{13}	92	100/0	70
c	8	CH_3	H	C_8H_{17}	98	100/0	
d	6	C_2H_5	H	C_2H_4OH	40	100/0	
e	6	C_2H_5	H	$C_2H_4OC_2H_4OH$	97	100/0	
f	6	CH_3	H	$C_6F_{13}C_2H_4$	90	75/25	95
g	8	CH_3	H	$CH(CH_2C_6H_5)COOCH_3$	80	100/0	
h	6	C_2H_5	CH_3	CH_3	40	98/2	
i	6	C_2H_5	C_2H_5	C_2H_5	85	98/2	90
j	6	C_2H_5	CH_3	$C_{12}H_{25}$	80	98/2	95
k	6	C_2H_5	$(CH_2CH_2)_2O$ (a)		92	98/2	98
l	8	C_2H_5	$(CH_2CH_2)_2O$ (a)		90	98/2	
m	8	$CH_3O(C_2H_4O)_2$	$(CH_2CH_2)_2O$ (a)		97	100/0	
n	6	CH_3	$(CH_2CH_2)_2O$ (a)		80	98/2	

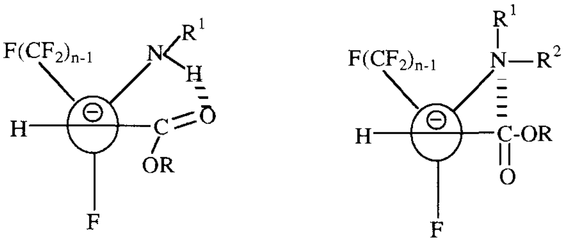
(a) morpholine

The condensation of a mono or dialkyl amine leads to the perfluoroalkylated enamines **7** (Scheme 6).

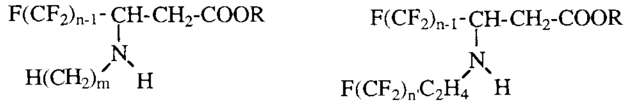
Unlike other procedures (Froissard et al., 1981), fluorine substitution by a primary or secondary amine only yields enamines without imine compounds. This reaction proceeds smoothly in refluxing ether (Et_2O or THF) or methylene chloride.

This mechanism is again a Michaël addition followed by fluoride elimination. The results presented in Table 1 illustrate the effectiveness of the method. The yields are very good for both kinds of amines. This reaction is not limited by the nature of the amine unlike the Wittig reaction proposed by Bégué et al. (1992). The steric hindrance has no influence on the reactivity since we have also successfully introduced an aminoacid. The enamine bearing two perfluoroalkylated chains has also been obtained in good yield (compounds 8f). The preparation of the initial perfluoroalkylated amine is described in a previous paper (Selve et al., 1991).

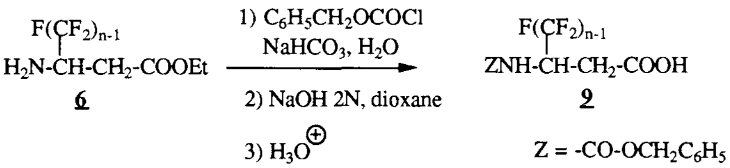
The ^1H NMR, ^{19}F NMR, and IR spectra indicate that in all cases the appearance of the Z-isomer is pratically exclusive. This stereoselectivity could be explained, as discussed in a previous paper (Thiebaut et al., 1997), on the basis of a carbanionic intermediate (Scheme 7).



Scheme 7. Stereoelectronic interactions in the carbanionic intermediates



Scheme 8. Original analogues of β-alanine with perfluorinated or perhydrogenated chain



Scheme 9. Synthesis of the perfluoroalkylated analogues of the Z β-alanine

The steric hindrance of the perfluorinated chain and the electronic effect of the amino group on the ester moiety favours the Z-enamine formation over other isomer. It may be noted that the electronic interactions are stronger with the primary than with the secondary amines.

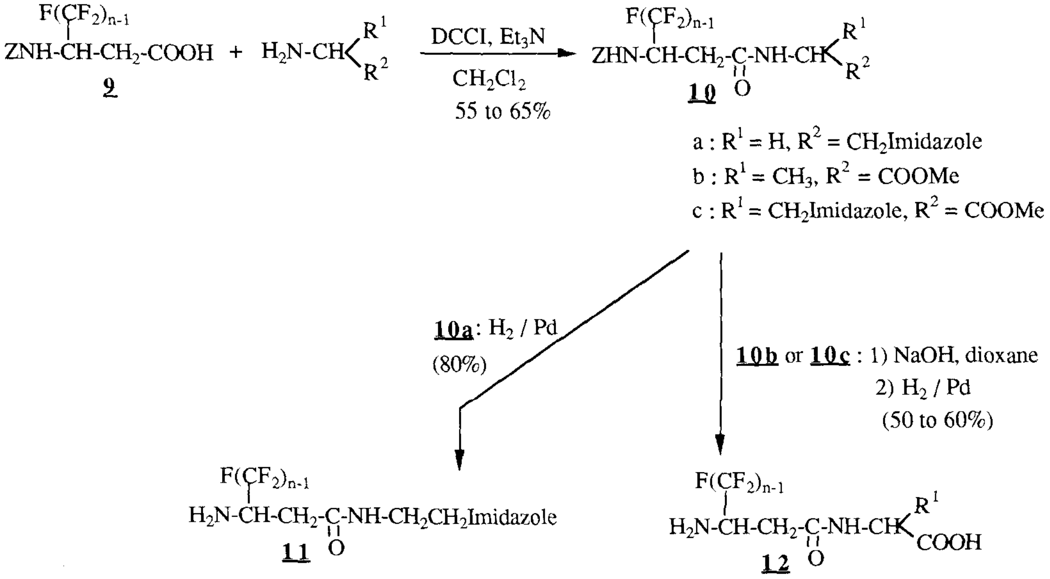
The hydrogenation of these compounds makes it possible to obtain the β-alanine analogues. Strict hydrogenation conditions are required: a pressure of eighty bars and a temperature of 80°C.

The overall yields are very satisfactory and both methods entail a large variety of analogues. Original compounds, namely β-alanine analogues containing either two perfluorinated chains or one perfluorinated substituent and one perhydrogenated chain can thus be obtained (Scheme 8).

The second step involves the protection of the amine function by a benzyloxycarbonyl moiety; saponification of the ester group leads to the perfluoroalkylated Z-β-alanine analogues (Scheme 9) (Özer et al., 1998).

These compounds can easily be bonded to another aminoacid (Özer et al., unpublished result). We choose a classical method using DCCI as a peptide coupling reagent. The dicyclohexyl urea is filtered off and after removing the solvent in vacuo, the product is purified by chromatography on silica gel (Scheme 10).

The method described here has been first tested with non-functionalized aminoacids (alanine) and has permitted the successful coupling with histidine



Scheme 10. Synthesis of peptides and peptidoamines containing perfluoroalkylated *β*-alanine

or histamine. The perfluoroalkylated carnosine has been obtained after hydrolysis in satisfactory yield.

This procedure can readily be extended to N-alkylated *β*-alanine analogues.

Conclusion

In this paper a simple and efficient synthesis of perfluoroalkylated *β*-alanine analogues and of peptidoamine derivatives are described. The method proposed is suitable for large-scale preparation. The complexing and surfactant properties of the compounds synthesized are currently being investigated. The first results of these studies are promising and studies of their physicochemistry will be continued in our laboratory.

Acknowledgments

We are indebted to the Atochem Society for donating the perfluoroalkyl ethanols and we wish to thank Dr. Bernard Henry for his useful suggestions.

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Received February 27, 1998