Introduction

Anchoring of homogeneous catalysts to polymers is receiving much attention. The advantages are an easier recovery and sometimes an activity and selectivity higher than of the original species. To some extent the system approaches an enzyme in which the active site is also attached to a polymer. Therefore, the anchored catalyst systems have been used as enzyme models, e.g. imidazole anchored to vinyl and ethylenimine polymers as catalysts for the hydrolysis of activated esters. However, these systems did not show enantioselectivity in the hydrolysis of asymmetric esters. The reason might be that the structural characteristics of the supports so far studied, are too much different from those of enzymes. We are investigating the use of polycaprolactone, (R—C=C(TH)O—C(TH)N—C(TH)OH) as supports for homogeneous catalysts. In earlier papers, we showed that these polymers probably have the configuration of a tightly coiled helix. Helices are chiral molecules. Poly(tert-butyl isocyanide) could even be resolved partly into its enantiomers and its screw sense derived from steric considerations as well as from CD spectra.

Polycaprolactones are obtained from the corresponding isocyanides, which in turn are synthesized from the amines. By starting from naturally occurring amino acids it might be possible to prepare polymers and copolymers of isocyanides which have a highly chiral structure and contain substituents which are catalytically active in enzymes. These polymers could be attractive model systems for the enantiospecific action of enzymes.

We decided to start from histidine; its imidazole group is a possible to prepare polymers and copolymers of isocyanides which have a highly chiral structure and contain substituents which are catalytically active in enzymes. These polymers could be attractive model systems for the enantiospecific action of enzymes.

Results and discussion

Masked carbylhistidine (7a) and carbylhistamine (7b) were synthesized from L-histidine and histamine, respectively. In order to obtain the former isocyanide in optically active form, it was necessary to work at low basicity and to prevent temperatures higher than ambient. Polymerization occurred with 1 mol % nickel chloride and a small amount of trifluoroacetic acid in methanol. The poly(carbylhistidine) showed no optical activity. The apparent pKa of the imidazole function in poly(carbylhistamine) has decreased to 5.2 ± 0.1 compared with free imidazole (7.2), whereas it rises to 9.4 ± 0.2 in poly(carbylhistidine).

Our first results as to the activity of these systems in the hydrolysis of esters are reported elsewhere.

Polyisocyanides 1. Synthesis and polymerization of carbylhistidine and carbylhistamine 2

J. M. van der Eijk, R. J. M. Nolte and W. Drenth

Department of Organic Chemistry of the University, Croesestraat 79, Utrecht, The Netherlands (Received March 2nd, 1977)

Abstract. Polymer anchored imidazole functions were obtained by polymerization of carbylhistidine and carbylhistamine. These isocyanides were synthesized from L-histidine and histamine, respectively. In order to obtain the former isocyanide in optically active form, it was necessary to work at low basicity and to prevent temperatures higher than ambient. Polymerization occurred with 1 mol % nickel chloride and a small amount of trifluoroacetic acid in methanol. The poly(carbylhistidine) showed no optical activity. The apparent pKa of the imidazole function in poly(carbylhistamine) has decreased to 5.2 ± 0.1 compared with free imidazole (7.2), whereas it rises to 9.4 ± 0.2 in poly(carbylhistidine).

Le traitement des données a été fait en lissant la courbe exponentielle observée par la méthode des moindres carrés, à l’aide de l’ordinateur IBM 360–40 de l’U.S.T.L.

Le calcul de la constante cinétique temporelle est fait en divisant la constante expérimentale par la concentration en proton et par la concentration en alcool. Les valeurs indiquées dans le Tableau I représentent la moyenne d’au moins sept déterminations indépendantes.

2 According to IUPAC nomenclature rules the compounds are named 3-[4(5)-imidazolyl]-2-isocyanopropanoic acid and 2-[4(5)-imidazolyl]-1-isocyanopropane, respectively.
addition of a small amount of acid started the polymerization. Under these conditions no polymerization was observed. However, without acid, imidazole residues block free coordination sites on nickel, which are necessary for polymerization. Probably, without acid, imidazole residues block free coordination sites on nickel, which are necessary for polymerization. The highest optical rotation, measured on solutions of clear and colourless crystals, amounted to \( \alpha = -4.8^\circ \). Polymerization of racemic carbylhistidine and of carbylhistamine was attempted by 1 mol \% of nickel chloride in methanol. Under these conditions no polymerization was observed. However, addition of a small amount of acid started the polymerization. Probably, without acid, imidazole residues block free coordination sites on nickel, which are necessary for polymerization. By protonation of imidazole this coordination is prevented.

Optically active carbylhistidine is expected to give on polymerization a polymer with predominantly one screw sense. However, so far no optical activity of the polymer could be detected, because of either a low value of its specific rotation or the low enantiomeric stability of the monomer. The optical activity of poly(carbylhistidine) as well as the synthesis of other optically active imidazole-containing polysiocyanides, like poly(carbylhistidinol) and poly(2-methylcarbylhistamine) are currently under investigation.

Polymers 8 were isolated as creamish brown solids. They were soluble in chlorinated hydrocarbons, acetone, methanol and acidiﬁed water. Their spectroscopic data are in agreement with the structures given. Removal of the protecting groups in 8 gave light-brown solids. After ultraﬁltration and freeze-drying the purified products were analyzed as the monohydrochlorides of polymers 9, containing a varying amount of water of crystallization. The polymers 9 were soluble in the lower alcohols and in water.

In most reactions catalyzed by imidazole, its unprotonated form appeared to be the catalytically active species. Therefore, it was of interest to determine the state of ionization of our polyisocyanide anchored imidazole. The relation between \( pH \) and degree of dissociation of imidazolium residues in 9, \( \alpha \), was measured by ultraviolet and potentiometric titrations. In accordance with the modified Henderson–Hasselbach equation

\[
pH = pK_{im} - n \log \left( \frac{1 - \alpha}{\alpha} \right)
\]

\( pK_{im} \) and \( n \) are constants.

References:

plots of $\log[(1 - x)/x]$ versus pH were linear. In Table I the values calculated for $p\text{K}_\text{im}$ and $n$ are presented as well as the $p\text{K}_\text{im}$'s of model compounds. The $n$ values calculated from ultraviolet titration plots at different wavelengths, showed a wavelength dependency and are omitted from Table I.

<table>
<thead>
<tr>
<th>Compound</th>
<th>$p\text{K}_\text{im}$</th>
<th>$n$</th>
</tr>
</thead>
<tbody>
<tr>
<td>9c</td>
<td>9.4 ± 0.1b</td>
<td>1.5 ± 0.2</td>
</tr>
<tr>
<td>9d</td>
<td>9.4 ± 0.1b</td>
<td>1.45 ± 0.1</td>
</tr>
<tr>
<td>1-Histidine</td>
<td>6.0</td>
<td></td>
</tr>
<tr>
<td>Histamine</td>
<td>6.0</td>
<td></td>
</tr>
<tr>
<td>Imidazole</td>
<td>7.2</td>
<td></td>
</tr>
<tr>
<td>Poly(t-His, t-Asp)</td>
<td>7.0</td>
<td></td>
</tr>
</tbody>
</table>

*a In 29% v/v EtOH/H$_2$O at 25°C; for the polymers the $p\text{K}_\text{im}$ is the apparent $p\text{K}$ of the imidazole in the polymer.
*b Ultraviolet titration.
*c Potentiometric titration.
*d In water; ref. 26.
*Ref. 27.

This wavelength dependency of $n$ is probably caused by interferences with absorption bands of the polymer $\text{C}=$ and backbone.

The potentiometrically determined $n$ values of both polymers 9 are equal within experimental error. Their magnitude suggests that more than one imidazole group participates in the reversible binding of a proton. Apparently, our $n$ value is independent of the presence of a carboxylic group in 9. It is remarked that in polyvinylpyridine the $n$ value of the imidazole residues is affected by the introduction of a carboxylic function 17. Table I shows that the carboxylic function appreciably increases the apparent dissociation constant of an imidazole residue in 9 but not in 1. This behaviour suggests the presence of a strong anionic field in polymer 9c 18.

**Experimental part**

Chemical shifts (δ) in the $^1$H-NMR spectra are given in ppm downfield from internal tetramethylsilane or sodium 2,2,3,3-tetradetero-3-(trimethylsilyl)propionate. Abbreviations used are: s = singlet, d = doublet, t = triplet, br = broad, dist = distorted. Elemental analyses were carried out by the Element Analytical Section of the Institute for Organic Chemistry TNO, Utrecht, The Netherlands, under supervision of W. J. Buys. NMR spectra were recorded under supervision of Dr. M. J. A. de Bie.

**Starting materials**

1-Histidine monohydrochloride, [α]$^22$D + 9.3° (c 5, 5 mol/l HCl), and histamine dihydrochloride were purchased from Fluka and Aldrich, respectively.

N((Im))-Benzy1-L-histidine (2)

This compound was synthesized by the method of Vignaud22 from 1 and benzy1 chloride in liquid ammonia. M.p. 241-242°C, [α]$^22$D + 10.0° (c 2, water, 1 eq. HCl); lit. 22 m.p. 248-249°, [α]$^22$D + 20.5° (c 2, water, 1 eq. HCl); lit. 23 [α]$^22$D + 10.1 ± 0.7° (c 2, water, 1 eq. HCl); the latter is more reliable.

N((Im))-Benzy1-L-histidine methyl ester hydrochloride (3)

This compound was obtained as an oil from 2 through esterification with hydrogen chloride 22 or thionyl chloride 23 in methanol. It was used without further purification for the synthesis of 6a: [α]$^20$D + 9.90° (c 2.5, methanol).

N((Im))-Benzyl-N(a)-formyl-L-histidine methyl ester (6a)

A suspension of 10 g (34 mmol) of 3 in 50 ml chloroform/methanol (9:1 v/v) was treated at 0°C with dry ammonia gas for 30 min. After evaporation of the ammonia at 0°C the precipitated ammonium chloride was removed by filtration and the resulting clear solution concentrated in vacuo at 30°C. The oily residue was dissolved in 100 ml formic acid and treated with 35 ml acetic anhydride while cooling in an ice-salt mixture. After stirring for 1 h at room temperature and removal of the excess of reagents by prolonged evacuation at 50°C/1 mm, compound 6a was obtained in quantitative yield and used without further purification for the synthesis of 7a upon repeated stirring with acetone the syrup partly solidified. Filtration of the solid and recrystallization from acetone afforded white crystals. M.p. 120°C (dec.), [α]$^22$D + 17.4° (c 4.8, methanol); IR (KBr): 3180 (NH), 1730 (COOCH$_3$), 1670 (CHO), 730 and 700 (benzyl) cm$^{-1}$; $^1$H-NMR (CD$_3$OD): 6.80 and 7.35 (2H, 2 × s, imidazole), 8.05 (1H, s, CHO), 7.40 (5H, s, benzyl), 5.35 (2H, s, benzyl), 4.80 (1H, t, CH), 3.70 (3H, s, OCH$_3$), 3.20 (2H, dist, d, CH$_2$).

Benzy1-L-carboxyhistidine methyl ester (7a)

To a stirred solution of 5 g (17.4 mmol) of 6a in 150 ml of N,N-dimethylformamide (DMF) was added, under a nitrogen atmosphere, a solution of 7.5 ml (100 mmol) of thionyl chloride dissolved in 30 ml of DMF, at such a rate that the temperature was kept at about −60°C. After addition, the cooling bath was temporarily removed to allow the temperature to rise to −38°C; then it was replaced and 23 g (210 mmol) of anhydrous sodium carbonate were slowly added, maintaining a temperature of −45°C. The mixture was subsequently stirred at this temperature for 30 min. The cooling bath was then removed and the temperature allowed to rise to 0°C. After addition of 50 ml of DMF, stirring was continued for 20 h while cooling in ice. To the reaction mixture 250 ml of methylene chloride were added followed by 450 ml of ice cold water. The aqueous layer was separated and rapidly extracted with three 100 ml portions of methylene chloride. The combined extracts were washed, dried over MgSO$_4$ and concentrated in vacuo. Addition of ethyl acetate to the resulting yellow-red syrup gave 3.3 g (70%) of light yellow crystals of 7a. Further purification by chromatography on a silica gel column (acetone as eluent) afforded transparant, colourless crystals. M.p. 77.5-78.5°C; [α]$^22$D −4.8° (c 5, acetone); C$_3$H$_5$N$_2$O$_5$ (269.1); calc. C 66.4, H 5.7, N 15.4, O 12.4; found C 66.4, H 5.7, N 15.4, O 12.4; MS: M$^+$ 269, M$^+$-OCH$_3$ 238, M$^+$-COOCH$_3$ 210, M$^+$-CH(NCCOCH$_3$) 171, troplyphonium ion 91; IR (KBr): 2152 + 1 (NC; CO was used for calibration), 1750 (COOCH$_3$) cm$^{-1}$; $^1$H-NMR (CD$_3$OD, CD$_2$OD): δ 7.65 and 7.00 (2H, 2 × s, imidazole), 7.30 (5H, s, benzyl), 5.25 (2H, s, benzyl), 4.85 (1H, t, CH), 3.75 (3H, s, OCH$_3$), 3.15 (2H, dist, d, CH$_2$).

Isocyanide 7a can be recognized on thin-layer plates as red-brown spots by spraying with a solution of nickel chloride in ethanol.

**Poly(benzyl carboxyhistidine methyl ester)** (8a)

A solution of 1.5 g (5.6 mmol) of 7a, 0.015 g (0.06 mmol) of nickel chloride hexahydrate and 5 drops of trifluoroacetic acid in 5 ml of methanol was stirred for 2 days at 25°C. After evaporation of the solvent, 8a was isolated in quantitative yield and used without further purification for the synthesis of 9c. Dropwise addition of a chloroform solution of 8a into a half-volumed excess of ether afforded a

19. For more details see the forthcoming thesis by J. M. van der Eijk.
This compound was prepared from 3 g (10 mmol) of (dec.); IR (KBr): 2148.5 ± 1 (NC, CO was used for calibration), afforded 1.55 g (55%) of pale yellow, crystalline 7b; m.p. 110°. Column chromatography on silica gel (chloroform/acetone 1:1) afforded 4 g (90%); m.p. 118.5-120°. IR (KBr): 3300 (NH), 1670 (C=O) cm⁻¹. Yields of 9c and 9d were obtained as syrups from histamine dihydrochloride and tosylcarbylhistamine chloride as described for the synthesis of 9d.

Poly(carbylhistidine) (9c)

An amount of 1.0 g (3.72 mmol) of finely powder 8a was suspended in 125 ml of dry liquid ammonia. Finely divided metallic sodium was added over a period of 5 h until a blue colour persisted. The excess of sodium was then discharged with ammonium chloride. The mixture was stirred for 2 days at ambient temperature, extracted with ethyl acetate, dried over MgSO₄, and subjected to ultracentrifugation (Diaflo Ultra-Filter, UM 2) and freeze-dried. Yield 0.65 g (3.66 mmol, 52%) of 9d. As indicated by elemental analysis 9d still contained 4% of tosylated imidazole residues and some water of crystallization or N-oxide groups: [C₆H₅-N=HCl]₀.₆₈[H₂O]₀.₃₈[C₆H₅-N=H₂S₇O₄]₀.₃₈; calc. C 46.5, H 5.8, N 26.0, O 8.4, Cl 12.6, S 0.8; [C₆H₅-N=HCl]₀.₉₂[H₂S₇O₄]₀.₀₈; calc. C 46.9, H 4.9, N 26.2, O 8.4, Cl 12.8, S 0.8; found C 46.5, H 4.8, N 26.2, O 8.7, Cl 13.2, S 0.8; IR (KBr): 3600-2200 (NH₂, H₂O), 1617 ± 1 (C=N, polystyrene was used for calibration), 1740 (COOCH₃), 1650 (C=N), br, CH₂), the major fraction of 9c failed to pass through a Diaflo Ultra-Filter UM 10 (retentivity MW > 10,000).

N(α)-Formylhistamine (5)

This compound was obtained as a syrup from histamine dihydrochloride as described for the synthesis of 6a. Yield 3.5 g (96%); ¹H-NMR (D₂O): 8 8.70 and 7.40 (2H, 2 × s, imidazole), 8.20 (1H, s, CHO), 3.60 and 3.00 (4H, 2 × H₂CH₂CHO). IR (KBr): 3600-2200 (NH₂, H₂O, NH), 1675 (COOH), 1617 ± 1 (C=N, polystyrene was used for calibration) cm⁻¹; ¹H-NMR (CDCl₃): 8 4.9 (3H, br, CH₃). Mass: M+ 275, 263, 251, 239, 227, 215, 193, 171, 159, 147, 135, 123, 111, 99, 87, 75, 63, 51, 39, 27, 15, 13.

N(Im)-Tosyl-N(α)-formylhistamine (6b)

To a solution of 7.5 g (54 mmol) of 5 and 15 g of Na₂CO₃ in 100 ml of water was added 15 g (78 mmol) of p-toluene-sulfonyl chloride and 10 ml of ether. The mixture was stirred for 2 days at ambient temperature, extracted with ethyl acetate, dried over MgSO₄, and concentrated in vacuo. A first crop of crystals was collected from ether; after purification of the mother liquid by column chromatography on silica gel (chloroform/acetone 1:1) there remained a residue, which was dried over PO₄₅₀ at 40°/12 mm. Yield 0.6 g (2.43 mmol. 5, 7b and 8b). The pA'₀ value was evaluated from the value of half-neutralization of the imidazolium group. The pH was determined by using its absorbance difference at 340 nm: e 2170 at pH > 11.00 and 1311 at pH < 5.20; for 9d the adsorbance difference at 300 nm was used: e 1838 at pH > 12.00 and 1221 at pH < 2.20.

Ultraviolet titrations

Solutions (3.5 × 10⁻⁴ mol/l) of polymers 9 in 29% v/v ethanol/water, acidified (pH 2) by adding a small amount of 1 mol/l hydrochloric acid and adjusted to an ion strength of 0.02 mol/l by adding KCl, were titrated with 0.1 mol/l of sodium hydroxide. After each addition of hydroxide the pH of the solution was measured and the ultraviolet spectrum recorded at 250°. In the region from 300 to 350 nm both polymers 9 showed a shoulder on the onset of a strong absorption band in the far ultraviolet. This shoulder shifted to longer wavelength on increasing the pH. The pKₐ value of the imidazol unit in 9c was determined by using its absorbance difference at 340 nm: e 2170 at pH > 11.00 and 1311 at pH < 5.20; for 9d the adsorbance difference at 300 nm was used: e 1838 at pH > 12.00 and 1221 at pH < 2.20.

Potentialiometric titrations

Polymers 9 were dissolved in 29% v/v ethanol/water to a concentration of 3.4 × 10⁻³ mol/l. Samples of 20 ml were acidified (pH 2-3) by adding 1 mol/l HCl. An amount of KCl was added, such that at the end point of titration μ = 0.02 mol/l. The solutions were titrated with 0.1 mol/l NaOH in 29% v/v ethanol/water at 25° under a nitrogen atmosphere while stirring. Blank titration curves were obtained by titrating 20 ml aliquots of 29% ethanol/water acidified to the same pH values and adjusted to the same ion strength. Differential titration curves were derived graphically, from which the degrees of dissociation were evaluated. In the titration curve of 9c the inflection point between the dissociation of the imidazolium ion and the carboxyl group was not clearly observed. Therefore, the pKₐ was calculated from the value of half-neutralization of the imidazol group. The n value was evaluated by applying eq. [1] to the pH values for which pH > pKₐ.

Acknowledgement

This investigation was supported by the Netherlands Foundation for Chemical Research (SON) with financial aid from the Netherlands Organization for the Advancement of Pure Research (ZWO). The authors are indebted to Charles F. Gusdorf for experimental assistance.

References: