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Mechanism of enantioselective ester cleavage by histidine-containing peptides at a micellar interface. 2. Effect of changing peptide chain length

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Abstract. Chiral p-nitrophenyl esters derived from the amino acid phenylalanine are cleaved by various histidine-containing tripeptides and higher oligopeptides as catalysts at a micellar interface. It is assumed that the oligopeptides adopt an internally hydrogen-bonded C7 conformation when they dissolve into the micellar hydrocarbon phase. Chiral recognition is attributed to the formation of a hydrogen bond in the micellar hydrocarbon phase between one enantiomer of the ester and the peptide. It is important for the activity that the imidazolyl NH moiety of the His residue remains in the aqueous phase. The hydrophilic/hydrophobic balance, which determines the location of each of the amino acid residues of the peptide in the two-phase system, can be controlled by varying the length and stereochemistry of the peptide chain and by attaching hydrophobic groups. The most selective catalyst is a tripeptide with the structure C4H9O-C(0)-L-Phe-L-His-L-Leu. It cleaves the p-nitrophenyl esters of N-protected L- and D-phenylalanine with an enantioselective of \( k_{L}/k_{D} \approx 40 \). Further lengthening of the peptide chain with L-Leu and L-Ala residues and lengthening of the N-protecting group of the catalyst decreases the enantioselectivity down to \( k_{L}/k_{D} \approx 6 \) for the penta-peptide C12H25O-C(0)-L-Phe-L-His-(L-Leu)3. The origin of these effects is discussed.

Introduction

Reactions in aqueous surfactant solutions are the subject of an increasing number of papers. The kinetics of these reactions resemble to some extent the kinetics of enzyme-catalyzed reactions. Recently, high (enantio-)selectivities, characteristic of enzyme-controlled reactions, have been described in a number of papers on micellar-catalyzed reactions. These papers deal with the cleavage of chiral amino acid p-nitrophenyl esters by the imidazolyl moiety of histidine-containing oligopeptides. Until recently, the origin of the observed enantioselectivities was not understood. In our previous paper, we showed that, for a number of histidine-containing dipeptides, the highest enantioselectivity was obtained with L-phenylalanine in position 1. Consequently, in the higher oligopeptides that are presented here, this position 1 is always occupied by this amino acid residue. The peptide chain of the L-Phe-L-His derivative is extended up to a pentapeptide using various L- and D-amino acid residues and the effect of these residues on the esterolytic activity and enantioselectivity was studied. The N-protecting group (R'CO) of the peptide chain is of the alkyloxy carbonyl type, which proved to be the most effective protecting group. The surfactant used is the chiral cationic surfactant (R)-N-hexadecyl-N,N-dimethyl-(x-methylbenzyl)ammonium bromide (R-surf 16).
which are shown in Chart 3, are TV-acyl /?-nitrophenyl esters of the amino acid of the imidazolyl group was not investigated. It will be shown that this assumption is valid only under certain step using the acylation of the imidazolyl moiety. In this step, the enantioselectivity is expressed as the ratio of the rate constants obtained for this acylation step using catalyst and/or substrate. Attention is, therefore, focused on the first step in the catalytic cycle, i.e., the acylation of the imidazolyl moiety. In this step, the enantioselectivity is expressed as the ratio of the rate constants obtained for this acylation step using L and D substrates: \( k_L / k_D \). The deacylation step of the acylated imidazolyl moiety, regenerating the free imidazolyl group was not investigated.

Special attention was paid to the blank (background) reaction. It is generally assumed that the blank reaction in the presence and absence of the catalyst is identical. It will be shown that this assumption is valid only under certain conditions. If these conditions are not met, large errors can occur.

Results

Catalytic activity and enantioselectivity of \( S_{12}-L\)-Phe-L-His-L-(or D-)X

In Table I, the catalytic activities and enantioselectivity of tripeptide catalysts towards \( C_{12}\)-Phe-ONp (L and D) in the presence of the surfactant R-surf 16 are listed. For comparison, the data of the dipeptide catalysts \( S_{12}-L\)-Phe-L-His and \( S_{12}-L\)-Ala-L-His are also included. In general, introducing a third amino acid residue at the 3-position of the catalyst appears to have little effect on the observed selectivity. An exception is \( S_{12}-L\)-Phe-L-His-L-Leu (entry 7) which exhibits an enhanced selectivity of \( k_L / k_D = 23 \) when compared to the corresponding dipeptide catalyst (\( k_L / k_D = 16.6 \)). Increase in hydrophobicity and size of the amino acid residue in the 3-position has no marked effect, up to a certain limit (entries 3–7). However, with the very bulky Phe residue, the
rate and selectivity are considerably suppressed (entry 8) as if a critical point is passed. The catalysts with the very hydrophobic Ile and Phg residues in the 3-position, whose side chains are both branched at C8, also exhibit reduced activity and enantioselectivity (entries 9 and 10). However, with the Trp residue in the 3-position, which is the largest residue tested, the rates and enantioselectivity are not affected to any large extent (entry 11). Neither a proline residue, whose side chain is covalently attached to the amide nitrogen atom of the peptide backbone, nor a hydrophilic threonyl residue has a large effect on the observed selectivity and activity (entries 12 and 13). From the results in Table I, it can be concluded that the reactivity of the imidazolyl group is reduced upon adding a third amino acid residue. The smallest reduction is observed for the S12-L-Phe-L-His-L-Asp catalyst, which has two unprotected COO⁻ moieties in the 3-position. The activity of this catalyst is by far the highest of all tripeptide catalysts examined here, although, its selectivity is somewhat lower than that of the catalyst under these reaction conditions show that the Trp residue in the 3-position from L to D causes a dramatic drop in activity from 23 to 19 (entries 7 and 15). This loss of selectivity is the result of a six-fold reduction in activity towards the L substrate and a two-fold increase in activity towards the D substrate, i.e., changing L-Leu into D-Leu has an opposite effect on the rates towards both enantiomers. Interestingly, the presence of the achiral amino acid residue Gly in the 3-position does not reduce the selectivity (compare entries 3, 7 and 15). The chirality of the surfactant molecules has no significant effect on the rates and enantioselectivity (entries 7 and 16), a fact that was also observed previously for a number of dipeptide catalysts.

This means that no specific interactions (e.g., stacking of aromatic rings) exists between these surfactant molecules and the catalysts or substrate, resulting in chiral recognition. This can also be concluded from the fact that the enantioselective profile of the esterolytic reaction does not change when R-surf 16 is substituted by the surfactant cetyltributylammonium bromide. The only apparent of the surfactant molecules is to form micelles that provide an interface at which the reaction can occur.

Table I Rate constants \(k_1/\text{mol} \cdot \text{m}^{-1} \cdot \text{s}^{-1}\) and enantioselectivity \(k_1/k_D\) in the cleavage of \(C_{12}\)-Phe-ONp (l, and D) by composites of \(12\), and d) are listed in Table II. In this Table, the data for the N-benzylxoyacetyl- (Z-) protected catalyst are also included. Upon increasing the length and thus the hydrophobicity of the N-protecting group from \(n = 2\) to \(12\), the reaction rate strongly increases. This enhanced reactivity reflects the fact that an increasing fraction of the catalyst is bound to the micelle where the substrate resides. The enantioselectivity increases from \(S_2\) to \(S_3\) where it reaches a maximum (\(k_1/k_D = 38.1\)) and decreases upon further chain lengthening.

Fluorescence measurement on the \(S_{12}-L-Phe-L-His-L-Trp\) catalyst

Fluorescence measurements on the \(S_{12}-L-Phe-L-His-L-Trp\) catalyst under these reaction conditions show that the Trp side chain is in a relatively apolar environment. At an excitation wavelength of \(\lambda_{ex} = 286\) nm, the fluorescence emission maximum of the Trp side chain appears at \(\lambda_{em} = 352\) nm. This position is also observed in a 1,4-dioxane/water mixture of 55:45 (v/v), demonstrating that the degree of hydration of the indolyl moiety in this catalyst at the micellar interface is similar. In this case, it is about 40–50% dehydrated. The same conclusion was derived from fluorescence measurements on the dipeptide catalyst \(S_{12}-L-Trp-L-His\), i.e., the indolyl group is partially dehydrated but is still in contact with bulk water.

Higher surfactant concentrations do not cause a further blue shift in the position of the emission maximum indicating that this catalyst is completely micellar-bound at the applied surfactant concentration.

Catalytic activity of \(S_{12}-L-Phe-L-His-L-Leu\) and \(S_{12}-L-Phe-L-His-L-Ala\) catalysts

In this section, the length, \(n\), of the hydrocarbon chain of the \(S_{12}-L-Phe-L-His-L-Leu\) catalyst is varied from \(n = 2\) to \(12\). The results obtained with the substrate \(C_{12}\)-Phe-ONp (l, and D) are listed in Table II. In this Table, the data for the N-benzylxoyacetyl- (Z-) protected catalyst are also included. Upon increasing the length and thus the hydrophobicity of the N-protecting group from \(n = 2\) to \(12\), the reaction rate strongly increases. This enhanced reactivity reflects the fact that an increasing fraction of the catalyst is bound to the micelle where the substrate resides. The enantioselectivity increases from \(S_2\) to \(S_3\) where it reaches a maximum (\(k_1/k_D = 38.1\)) and decreases upon further chain lengthening.

Table II Rate constants \(k_1/\text{mol} \cdot \text{m}^{-1} \cdot \text{s}^{-1}\) and enantioselectivity \(k_1/k_D\) in the cleavage of \(C_{12}\)-Phe-ONp (l, and D) by composites of \(S_{12}-L-Phe-L-His-L-Leu\) catalysts

<table>
<thead>
<tr>
<th>Catalyst</th>
<th>(k_1)</th>
<th>(k_D)</th>
<th>(k_1/k_D)</th>
<th>(\Delta G^\circ) (kcal/mol)</th>
</tr>
</thead>
<tbody>
<tr>
<td>(S_{12}-L-Phe-L-His-L-Leu)</td>
<td>157</td>
<td>15</td>
<td>10.5</td>
<td>1.39</td>
</tr>
<tr>
<td>(S_{12}-L-Phe-L-His-L-Leu)</td>
<td>1257</td>
<td>33</td>
<td>38.1</td>
<td>2.16</td>
</tr>
<tr>
<td>(S_{12}-L-Phe-L-His-L-Leu)</td>
<td>1544</td>
<td>59</td>
<td>26.2</td>
<td>1.93</td>
</tr>
<tr>
<td>(S_{12}-L-Phe-L-His-L-Leu)</td>
<td>2319</td>
<td>100</td>
<td>23.2</td>
<td>1.86</td>
</tr>
<tr>
<td>(S_{12}-L-Phe-L-His-L-Leu)</td>
<td>1284</td>
<td>32</td>
<td>40.1</td>
<td>2.19</td>
</tr>
</tbody>
</table>

- **Conditions as described in Table I.**

The activities and selectivity of \(Z\)- and \(S_{12}\)-protected \(L-Phe-L-His-L-Leu\) catalysts do not differ significantly. Such behaviour was also observed for \(Z\)-Phe-L-His and \(S_{12}\)-Phe-L-His dipeptide catalysts. This indicates that the \(R\) moieties butyl and benzyl are sterically and hydrophobically very similar in the present reaction.

Table III shows that a bell-shaped selectivity dependence on \(n\) is also observed for \(S_{12}-L-Phe-L-His-L-Ala\) (\(n = 2, 4, 6, 12\)). However, the enantioselectivity is approximately 25–30% lower than for the \(S_{12}-L-Phe-L-His-L-Leu\) catalysts. Although not investigated, it is likely that such bell-shaped selectivity curves will also be found for the other tripeptide catalysts listed in Table I. We reported previously a similar but smaller effect in the \(S_{12}-L-Phe-L-His\) series, with a maximum of \(k_1/k_D = 18.3\) at \(n = 3\). This means that the relatively small effect on the selectivity observed when the catalyst is extended with a third amino residue (Table I) is rather fortuitous and probably only occurs in the \(S_{12}-L-Phe-L-His\) series.

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**Notes:**
- Table I, page 461, Recueil des Travaux Chimiques des Pays-Bas, 111/11, November 1992.
- Table II, page 462.
- Table III, page 463.
-protected series. In the $S_4$ series, the enantioselectivity considerably increases from 18 to 38 by adding l-Leu as a third residue.

Table III Rate constants $[k_{obs}/(dm^3 \cdot mol^{-1} \cdot s^{-1})]$ and enantioselectivity $k_l/k_D$ in the cleavage of C$_1$-Phe-ONp (I and II) by cocomplexes of C$_n$:t-Phe-t-His-t-Ala (n = 2, 4, 6, 12) and R-surf 16$^a$.

<table>
<thead>
<tr>
<th>Catalyst</th>
<th>$k_L$</th>
<th>$k_D$</th>
<th>$k_L/k_D$</th>
<th>$\Delta G^*$ (kcal/mol)</th>
</tr>
</thead>
<tbody>
<tr>
<td>S$_{12}$-t-Phe-t-His-t-Ala</td>
<td>128</td>
<td>14</td>
<td>9.1</td>
<td>1.31</td>
</tr>
<tr>
<td>S$_{12}$-t-Phe-t-His-t-Ala</td>
<td>1190</td>
<td>46</td>
<td>25.9</td>
<td>1.93</td>
</tr>
<tr>
<td>S$_{12}$-t-Phe-t-His-t-Ala</td>
<td>1749</td>
<td>89</td>
<td>19.7</td>
<td>1.76</td>
</tr>
<tr>
<td>S$_{12}$-t-Phe-t-His-t-Ala</td>
<td>2420</td>
<td>145</td>
<td>16.7</td>
<td>1.67</td>
</tr>
</tbody>
</table>

$^a$ Conditions as described in Table I.

Catalytic activity of $S_{12}$-t-Phe-t-His-t-Leu towards C$_n$:t-Phe-ONp

Table IV shows the activities and enantioselectivity obtained with the $S_{12}$-t-Phe-t-His-t-Leu catalyst towards the C$_n$:t-Phe-ONp substrates (n = 2, 4, 7, 12, 16). Increasing the length of the acyl chain increases both the activity and selectivity. The enhanced activity will be due to increased binding of the substrate to the micelle. Complete binding is achieved at approximately $n = 12$. The applied catalyst, $S_{12}$-t-Phe-t-His-t-Leu, is also completely bound to the micelle. Upon increasing $n$ in the substrate, the selectivity approaches a limiting value of $k_L/k_D = 23$ at $n = 12$. A different effect on selectivity was observed for the $S_4$ chain in the catalyst, which displays a clear optimum at $S_4$.

Table IV Rate constants $[k_{obs}/(dm^3 \cdot mol^{-1} \cdot s^{-1})]$ and enantioselectivity $k_l/k_D$ in the cleavage of C$_n$:t-Phe-ONp (I and II) by cocomplexes of S$_{12}$-t-Phe-t-His-t-Leu and R-surf 16$^a$.

<table>
<thead>
<tr>
<th>Ester</th>
<th>$k_L$</th>
<th>$k_D$</th>
<th>$k_L/k_D$</th>
<th>$\Delta G^*$ (kcal/mol)</th>
</tr>
</thead>
<tbody>
<tr>
<td>C$_3$:t-Phe-ONp</td>
<td>328</td>
<td>43</td>
<td>7.6</td>
<td>1.20</td>
</tr>
<tr>
<td>C$_5$:t-Phe-ONp</td>
<td>669</td>
<td>75</td>
<td>8.9</td>
<td>1.29</td>
</tr>
<tr>
<td>C$_7$:t-Phe-ONp</td>
<td>1559</td>
<td>90</td>
<td>17.3</td>
<td>1.69</td>
</tr>
<tr>
<td>C$_9$:t-Phe-ONp</td>
<td>2319</td>
<td>106</td>
<td>23.2</td>
<td>1.86</td>
</tr>
<tr>
<td>C$_{11}$:t-Phe-ONp</td>
<td>2230</td>
<td>106</td>
<td>21.9</td>
<td>1.83</td>
</tr>
</tbody>
</table>

$^a$ Conditions as described in Table I.

Activity and enantioselectivity of tetra- and pentapeptide catalysts

Table V shows how the activity and enantioselectivity of the esterolytic reaction is affected when the peptide chain of the catalyst is further extended with t- and/or d-residues. For comparison, the data of the corresponding di- and tripeptide catalysts are also included. Increasing the peptide chain length of the S$_{12}$-t-Phe-t-His catalyst with t-Ala or t-Leu residues causes a dramatic drop in activity. The effect is considerably larger for the more hydrophobic t-Leu residue (entries 1–4) than for the smaller t-Ala residue (entries 1, 5–7). The enantioselectivity remains unaffected up to the tripeptides or even slightly increases (for the S$_{12}$-t-Phe-t-Leu catalyst), as noted above. Upon further chain lengthening, the enantioselectivity also rapidly decreases. The tetrapeptide catalysts with t-Leu-t-Ala and t-Ala-t-Leu in the 3- and 4-positions (entries 8 and 9) display intermediate activities and enantioselectivity in comparison to the catalysts with t-Leu-t-Leu and t-Ala-t-Ala in these positions.

Interestingly, the tetrapeptide catalyst with t-Leu-d-Leu (entry 10) in the 3- and 4-positions exhibits much higher enantioselectivity than the catalyst with the equally hydrophobic t-Leu-t-Leu sequence (entry 3). A similar effect is observed for the tetrapeptide catalysts with t-Ala-t-Ala and t-Ala-d-Ala in the 3- and 4-positions (entries 6 and 11). These differences must be caused by a difference in the shape of the diastereomeric catalysts.

Catalytic activity of $S_{12}$-t-Phe-t-His methylamide

Table VI shows the activities and selectivity obtained with S$_4$- and S$_{12}$-t-Phe-t-His-NMA. In these catalysts, the carboxyl groups are protected by methylamine. The activities and enantioselectivity are strongly reduced compared to the corresponding di- or tripeptide catalysts which have an unprotected carboxyl group (see Tables I, II and III). This almost total collapse of the observed rates and severe suppression of the enantioselectivity cannot be attributed to a steric effect of the NMA-protecting group, as this group is smaller than any of the amino acid residues in the 3-position of the tripeptide catalysts. The catalytic activity of S$_{12}$-t-Phe-t-His-NMA, for instance, should be compared with the S$_{12}$-t-Phe-t-His-Gly catalyst (Table I, entry 3). The structural difference is that, in the latter catalyst, one hydrogen atom of the NMA moiety is replaced by a carboxylate group. These data indicate that the proximity of the carboxylate group increases the activity of the imidazolyl moiety.

Table VI Rate constants $[k_{obs}/(dm^3 \cdot mol^{-1} \cdot s^{-1})]$ and enantioselectivity $k_l/k_D$ in the cleavage of C$_{12}$-t-Phe-ONp (I and II) by cocomplexes of S$_4$/$S_{12}$-t-Phe-t-His-NMA and R-surf 16$^a$.

<table>
<thead>
<tr>
<th>Catalyst</th>
<th>$k_L$</th>
<th>$k_D$</th>
<th>$k_L/k_D$</th>
</tr>
</thead>
<tbody>
<tr>
<td>S$_{12}$-t-Phe-t-His-NMA</td>
<td>62</td>
<td>8</td>
<td>7.8</td>
</tr>
<tr>
<td>S$_{12}$-t-Phe-t-His-NMA</td>
<td>149</td>
<td>26</td>
<td>5.7</td>
</tr>
</tbody>
</table>

$^a$ Conditions as described in Table I.

Solvent isotope effect and apparent $pK_a$ of the imidazolyl moiety

Table VII shows the solvent deuteration kinetic isotope effect for the cleavage of C$_{12}$-t-Phe-ONp by various histidine-containing peptides.

<table>
<thead>
<tr>
<th>Catalyst</th>
<th>$k_L$</th>
<th>$k_D$</th>
</tr>
</thead>
<tbody>
<tr>
<td>S$_{12}$-t-Phe-t-His</td>
<td>6012</td>
<td>363</td>
</tr>
<tr>
<td>S$_{12}$-t-Phe-t-His</td>
<td>2319</td>
<td>100</td>
</tr>
<tr>
<td>S$_{12}$-t-Phe-t-His</td>
<td>156</td>
<td>33</td>
</tr>
<tr>
<td>S$_{12}$-t-Phe-t-His</td>
<td>89</td>
<td>15</td>
</tr>
<tr>
<td>S$_{12}$-t-Phe-t-His</td>
<td>2420</td>
<td>145</td>
</tr>
<tr>
<td>S$_{12}$-t-Phe-t-His</td>
<td>809</td>
<td>77</td>
</tr>
<tr>
<td>S$_{12}$-t-Phe-t-His</td>
<td>345</td>
<td>48</td>
</tr>
<tr>
<td>S$_{12}$-t-Phe-t-His</td>
<td>577</td>
<td>62</td>
</tr>
<tr>
<td>S$_{12}$-t-Phe-t-His</td>
<td>333</td>
<td>26</td>
</tr>
<tr>
<td>S$_{12}$-t-Phe-t-His</td>
<td>1510</td>
<td>69</td>
</tr>
</tbody>
</table>

$^a$ Conditions as described in Table I.
containing catalysts. The kinetic isotope effects do not differ significantly for both enantiomers of the substrate. All values of $k_{11}/k_{12}$ are in the range 1.1–1.5, which indicates that proton transfer does not take place in the rate-determining step, i.e., the reaction is not general base-catalyzed.

Table VII  Solvent kinetic isotope effects for the cleavage of C$_{12}$-Phe-ONp (l. and d) by micelles of His-containing peptides and R-surf 16$^a$.

<table>
<thead>
<tr>
<th>Catalyst</th>
<th>$k_{11}/k_{12}$ (l)</th>
<th>$k_{11}/k_{12}$ (d)</th>
</tr>
</thead>
<tbody>
<tr>
<td>S$_{12}$-t-Phe-1-His</td>
<td>1.1</td>
<td>1.2</td>
</tr>
<tr>
<td>S$_{12}$-t-Phe-1-His-1-Leu</td>
<td>1.5</td>
<td>1.5</td>
</tr>
<tr>
<td>S$_{12}$-t-Phe-1-His-1-Asp</td>
<td>1.4</td>
<td>1.5</td>
</tr>
<tr>
<td>S$_{12}$-t-Phe-1-His-1-Leu-1-Leu</td>
<td>1.1</td>
<td>1.2</td>
</tr>
</tbody>
</table>

$^a$ Conditions as described in Table I.

In Table VIII, the apparent $pK_a$ values are given of two catalysts that are bound to micelles of R-surf. The $pK_a$ values of the S$_2$ compounds were measured in the absence of surfactant. It was impossible to measure the $pK_a$ of the S$_1$-protected catalyst in the absence of surfactant because self-aggregation occurred. Comparison of the $pK_a$ values in the presence and absence of surfactant gives the effect of micellar adsorption on the $pK_a$. The $pK_a$ (ImH$^+$) of 6.9 measured in the absence of surfactant is a normal value for N-protected histidine$^{17}$. The apparent $pK_a$ (ImH$^+$) values of the micellar-bound catalysts are approximately 0.5 $pK_a$ unit lower, which is to be expected for cationic micelles$^{18,19}$.

Due to the positive charge of the micellar surface, protonation of the imidazolyl group is impeded (field effect). The positively charged surface will also attract hydroxide ions from the solution, i.e., the local hydroxide ion concentration (and hence the pH) at the micellar surface is higher than in the bulk aqueous phase. Consequently, the imidazolyl group appears to deprotonate at a lower pH.

Table VIII  Apparent $pK_a$ values of the imidazolyl moieties in various catalysts in the presence (S$_{12}$) and absence (S$_2$) of R-surf 16$^a$.

<table>
<thead>
<tr>
<th>Catalyst</th>
<th>$pK_a$ (ImH$^+$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>S$_{12}$-t-Phe-1-His</td>
<td>6.9</td>
</tr>
<tr>
<td>S$_{12}$-t-Phe-1-His-1-Leu</td>
<td>6.9</td>
</tr>
<tr>
<td>S$_{12}$-t-Phe-1-His</td>
<td>6.4</td>
</tr>
<tr>
<td>S$_{12}$-t-Phe-1-His-1-Leu-1-Leu</td>
<td>6.3</td>
</tr>
</tbody>
</table>

$^a$ Conditions: 0.40 M KCl; $c_{cat}$ $2 \cdot 10^{-4}$ M; $c_{sur}$ $1 \cdot 10^{-2}$ M. $^b$ In the absence of surfactant.

The blank reaction

In order to calculate the catalytic activity of a catalyst ($k_{cat}$) from the observed reaction rate (rate constant $k_{obs}$), the magnitude of the blank or background reaction ($k_{bl}$) must be known ($k_{cat} = k_{obs} - k_{bl}$). The blank reaction rate is determined in the absence of catalyst, and it is assumed that, in the presence of the catalysis, this blank reaction is unaffected. However, it will be demonstrated that this assumption is only valid under certain conditions.

Cationic micelles greatly accelerate hydrolysis of the adsorbed substrates. This effect is due to the attraction of hydroxide ions from the solution, i.e., the local hydroxide ion concentration or local pH at the micellar surface is higher than the pH in the bulk aqueous solution. In spite of the presence of a buffer, the micellar surface is not buffered$^{18,19}$. Most catalysts used are anionic amphiphilic molecules that bind tightly to the micelles. Consequently, the micellar charge density is reduced and fewer hydroxide ions will be bound. This results in a reduction in the rate of spontaneous ester hydrolysis. In order to investigate this effect, we used the catalytically inactive model compound N-(dodecyloxycarbonyl)-L-phenylalanine sodium salt (S$_{12}$-t-Phe-ONa) at a concentration similar to that of our catalysts and determined the reduction of the blank hydrolysis ($\Delta k_{bl}$) of C$_{12}$-D-Phe-ONp at various concentrations of added KCl (pH 7.3, 0.02 mol/dm$^3$ Tris/HCl). The results are shown in Figure 1a. The observed reduction in rate constant is expressed in the same units as the catalytic rate constants in the Tables and their magnitudes can be directly compared. At $c_{KCl} = 0$, the addition of the model compound causes a reduction of $k_{bl}$ from 1112 to 1012 dm$^{-3}$ mol$^{-1}$ s$^{-1}$. In the presence of 0.4 mol·dm$^{-3}$ KCl, the blank reaction is reduced from $k_{bl} = 25$ to $k_{bl} = 22$ dm$^{-3}$ mol$^{-1}$ s$^{-1}$. At low KCl concentrations, many hydroxide ions are bound to the micelles. Consequently, binding of the negatively charged model compound replaces many hydroxide ions and the blank reaction rate is considerably reduced. At high salt concentrations ($c_{KCl} > 0.3$ mol·dm$^{-3}$), the micelles only bind Cl$^-$ ions at their surface which are partially replaced by the model compound. The blank reaction rates now remain almost unaffected. It should be noted that the reduction of the blank reaction rate at low KCl concentration ($\Delta k_{bl} = -100$ dm$^{-3}$ mol$^{-1}$ s$^{-1}$) becomes of similar magnitude as or is even larger than the catalytic reaction rates towards the d substrates ($k_D$ is approximately 100 dm$^{-3}$ mol$^{-1}$ s$^{-1}$). This means that if we calculate $k_D = k_{obs} - k_{bl}$, with the assumption that $k_{bl}$ is unaffected by the presence of the catalyst, we severely underestimate the magnitude of $k_D$ since we use a value for $k_{bl}$ that is too large. The underestimation of $k_D$, which generally has a much larger value than $\Delta k_{bl}$, is of minor importance. The result is that the enantioselectivity of a
catalyst \((k_1/k_D)\) is severely overestimated. This artifact is shown in Figure 1b for the S-2-L-Phe-L-His-L-Leu catalyst as a function of the added amount of KCl. It can be concluded that the assumption \(k_{bl}\) (in the presence of catalyst) = \(k_{bl}\) (in the absence of catalyst) in the present system is only valid at high ionic strength \((c_{cl} > 0.3 \text{ mol/dm}^3)\). High enantioselectivity has been reported due to the above described artifact\(^9\)\(^{11}\)\(^{20}\)\(^{21}\). The experiments described in this paper were carried out in the presence of 0.4 mol/dm\(^3\) KCl, which means that the calculated enantioselectivity values are reliable. Non-ionic molecules that adsorb to the micelles and cause an increase in surfactant headgroup–headgroup distance will also reduce the charge density at the micellar interface. Consequently, fewer OH\(^+\) ions will be bound, resulting in a reduction in the blank reaction rate.

Discussion

A micellar solution is a two-phase system consisting of an aqueous phase and a hydrocarbon-like micellar pseudophase (Figure 2a). When our peptide catalysts dissolve into the micellar hydrocarbon phase, the polar amide groups will be dehydrated, which is very unfavorable. To compensate for this dehydration, the amide groups will try to form internal hydrogen bonds. Since a completely internally hydrogen-bonded conformation is not possible for our peptides, the resulting folded peptide chain has an amphiphilic character. To what extent the peptide chain will dissolve into the micellar hydrocarbon phase will be determined by the hydrophilic/hydrophobic balance of the peptide chain. Peptides shift their preference to the apolar phase if they adopt the internally hydrogen bonded C\(_7\)- or \(\gamma\)-turn conformation\(^22\). A seven-membered ring including the \(\text{H}\) atom is formed (Figure 2b). In the following, we assume that our small peptide catalysts also adopt this C\(_7\) conformation, when they dissolve into the micellar hydrocarbon phase, forming as many intramolecular hydrogen bonds as possible (Figure 2b). As explained in our first paper\(^1\), the NH group of the Phe residue in position 1 of the catalyst forms an intermolecular hydrogen bond with the amide CO moiety of the \(\text{L}\) substrate, when the imidazolyl moiety of the His residue attacks the ester group to form the tetrahedral intermediate (Figure 2c, see also Figure 3a). This hydrogen bond contributes to the stability of the transition state when it is formed in the apolar micellar hydrocarbon phase. Stabilizing hydrogen bond is not formed in the diastereomeric intermediate with the \(\text{D}\) substrate. Consequently, the reaction rate towards the \(\text{L}\) substrate is much higher. When residue 1 in the catalyst is less hydrophobic, the transfer of the NH moiety to the micellar hydrocarbon phase is incomplete and the hydrogen bond with the \(\text{L}\) ester becomes less favorable, resulting in a reduction in the reaction rate (compare entries 1 and 2 in Table I). This feature was discussed in our previous paper\(^1\).

In the tetrahedral intermediate, a positive charge has developed on the imidazolyl moiety which is not present in the ground state (Figures 2c and 3). In order to stabilize this positive charge, the imidazolyl moiety requires a polar environment: the imidazolyl NH group must remain in the aqueous phase. In order to prevent the imidazolyl moiety from dissolving into the micellar hydrocarbon phase, the imidazolyl NH moiety must be structurally connected to the very hydrophilic carboxylate moiety. In liquid water at room temperature, such a connection is possible if the distance between the two groups (\(d\)) does not exceed the space required for two hydrogen-bond water molecules, or approximately 7–8 Å (Figure 2c)\(^23\)\(^{24}\). If this distance is larger, the hydration of the imidazolyl and that of the carboxylate moiety become independent. The imidazolyl moiety can dissolve in the apolar micellar hydrocarbon phase and become a less effective catalyst. This carboxylate-imidazolyl interaction in combination with the above proposed hydrogen bonds affords a satisfying explanation for our observations.

Figure 3 shows the tetrahedral intermediates of a number of catalysts, which have adopted the C\(_7\) conformation. This Figure further indicates whether the imidazolyl group falls within the hydration mantle of the C-terminal carboxylate group (\(d < 8 \text{ Å}\)) or not (\(d > 8 \text{ Å}\)). The distance \(d\) is determined with the aid of CPK space-filling models. Figure 3a gives the intermediate for the reaction with the S-2-L-Phe-L-His catalyst. For both the \(\text{L}\) and \(\text{D}\) substrates, the imidazolyl NH moiety falls within the hydration mantle of the carboxylate group. Consequently, the activity towards both substrates is relatively high. However, due to the additional hydrogen bond with the \(\text{L}\) substrate, the activity towards this enantiomer is higher (\(k_1/k_D = 16.6\); Table I, entry 2).

Figure 3b shows the intermediate for the \(\text{L}\) substrate with the S-2-L-Phe-L-His-L-Leu catalysts. Transfer of the Leu residue to the micellar phase is facilitated because of dehydration of its NH group is compensated for by hydrogen bonding to the Phe C=O. In the micellar phase, the Leu NH moiety forms a hydrogen bond with the \(\text{L}\) substrate and chiral recognition will improve (up to \(k_1/k_D = 40\); Table II). These two internally hydrogen-bonded groups are located on both sides of the His residue (see also Figure 2b in which
The imidazolyl NH moiety is given by d. Consequently, the reaction rate decreases if compared to the 
substrate and the enantioselectivities are strongly reduced. The effect is much larger for the more hydrophobic amino acid residues (l-X-l-Y = l-Leu-l-Leu; Table V, entry 3) than for the less hydrophobic residues (l-X-l-Y = l-Ala-l-Ala; Table V, entry 6). The effect is intermediate when l-X-l-Y = l-Leu-l-Ala or l-Ala-l-Leu.

Figure 3c gives the intermediate for the l-substrate and the S"-L-Phe-L-His-L-X-L-Y catalysts. The imidazolyl NH moiety can easily dissolve into the micellar hydrocarbon phase. This causes a large decrease in activity and enantioselectivity.

Tables II and III show that, in order to obtain maximum enantioselectivity, S" in the catalysts must have optimum hydrophobicity. This behaviour is typical for an amphiphilic transition state in a two-phase hydrocarbon/water system. A shift in the hydrophobic/hydrophilic balance of a transition state affects its distribution between the two phases (which part dissolves into the micellar hydrocarbon phase and which part will remain in the aqueous phase), as already shown above. Increasing the hydrophobicity from S" to S° promotes transfer of the NH moiety of the Phe residue to the micellar hydrocarbon phase. Consequently, the enantioselectivity increases. If the S" group becomes more hydrophobic, the tendency for the imidazolyl group to dissolve into the micellar hydrocarbon phase decreases and the reaction rates decrease.

The S"-L-Phe-L-His-NMA catalysts (Table VI) lack the very hydrophilic carboxylate group. Consequently, the imidazolyl moiety can easily dissolve into the micellar hydrocarbon phase. This causes a large decrease in activity and enantioselectivity.

General remarks

Optimum hydrophobicity is not observed for the C° group of the ester (Table IV). The hydrophobicity of C° of the ester affects its distribution between the aqueous phase and the micellar hydrocarbon phase. The more hydrophobic C° becomes, the greater the tendency of the ester to dissolve into the micellar hydrocarbon phase. At a certain point, the ester is completely dissolved into the micellar hydrocarbon phase. In the micellar hydrocarbon phase, the affinity of the amide CO group of the l enantiomer of the ester to form a hydrogen bond with the NH group of the Phe residue in the catalyst is at its maximum. Consequently, the enantioselectivity increases up to a limit value.

Experimental

General remarks

Thin-layer chromatography (TLC) was performed on silica gel (Merck DC-Plastikrolle, Kieselgel 60 F254) and detection was...
effected by ultraviolet light or iodine. Column chromatography was performed with silica gel (Merck Kieselgel 60, 230–400 mesh). Solvents and reagents were of analytical grade. The preparation of the surfactant solutions with and without catalyst is described elsewhere. The kinetic measurements were carried out as described in Ref. 1. Elemental analyses were made from the final products and were all in good agreement with the calculated values.

Synthesis

N-Dodecylcarboxyl-[(p-phenylalanyl)-l-histidine (S,\textsubscript{i}L-Phe\textsubscript{i}His\textsubscript{i}OH); N-(dodecylcarboxyl)-l-alanyl-l-histidine (S,\textsubscript{i}L-Ala\textsubscript{i}His\textsubscript{i}OH). The synthesis of these compounds is described in our previous paper.

N-(Dodecylcarboxyl)-[(p-phenylalanyl)-l-histidyl-l- (or d) O-XOME (S,\textsubscript{i}L-Phe\textsubscript{i}His\textsubscript{i}-O-XOME). H-X-O-MeCl (X = amino acid) was coupled to N-(dodecylcarboxyl)-[(p-phenylalanyl)-l-histidine in DMF with the aid of 1,3-dicyclohexylcarbodiimide according to a published method. The method can cause some racemization of the histidyl residue but the diastereomers can be removed by chromatography. The crude product was purified by column chromatography (Silica 60, eluent chloroform/methanol, 15:1 v/v) and recrystallized from acetone. TLC: R\textsubscript{i}f 0.3 (silica chloroform/methanol, 10:1 v/v) for all compounds. Traces of diastereomers that were formed by racemization of the histidyl residue showed a R\textsubscript{i}f value on TLC around 0.25. These impurities were absent in the purified final products.

The \textsuperscript{1}H NMR, CDCl\textsubscript{3}, [\textgreek{a}]d/(c 1.0, methanol), m.p./°C and yield/% are:

**S,\textsubscript{i}L-Phe\textsubscript{i}His\textsubscript{i}-Gly\textsubscript{i}OMe:** \textsuperscript{1}H NMR (CDCl\textsubscript{3}), [\textgreek{a}]d/(c 1.0, methanol), m.p./°C and yield/%.

**S,\textsubscript{i}L-Phe\textsubscript{i}His\textsubscript{i}-Asp\textsubscript{i}OMe:** \textsuperscript{1}H NMR (CDCl\textsubscript{3}, and CD\textsubscript{2}OD, 1:1 (v/v)); 0.9 (t, 3H, CH\textsubscript{3}); 1.3 (m, 23H, (CH\textsubscript{2})\textsubscript{10}); 0.9 (t, 3H, CH\textsubscript{3}); 4.2-4.9 (m, 3H, 3xCH); 7.0 and 8.0 (2 x s, 2 x 1H, 2 CH (Im)); 7.1 (s, 5H, ArH) ppm; +5.6°; 206.1.

**S,\textsubscript{i}L-Phe\textsubscript{i}His\textsubscript{i}-Asp\textsubscript{i}OH:** The \textsuperscript{1}H NMR spectrum of this compound closely resembles the spectrum of S,\textsubscript{i}L-Phe\textsubscript{i}His\textsubscript{i}-Asp\textsubscript{i}OMe with the exception: 0.9 (2 x 1H, 2 CH\textsubscript{2} (Im)) ppm; +4.1; 196.1. IR (KBr): 1685 cm\textsuperscript{-1} (CO amide); 1640 cm\textsuperscript{-1} (CO amide).

**S,\textsubscript{i}L-Phe\textsubscript{i}His\textsubscript{i}-Gly-OH:** 1H NMR (CDCl\textsubscript{3}, and CD\textsubscript{2}OD, 1:1 (v/v)); 0.9 (t, 3H, CH\textsubscript{3}); 1.3 (m, 23H, (CH\textsubscript{2})\textsubscript{10}); 0.9 (t, 3H, CH\textsubscript{3}); 4.2-4.9 (m, 3H, 3xCH); 7.0 and 8.0 (2 x s, 2 x 1H, 2 CH (Im)); 7.1 (s, 5H, ArH) ppm; +5.6°; 206.1.

**S,\textsubscript{i}L-Phe\textsubscript{i}His\textsubscript{i}-Thr-OH:** The \textsuperscript{1}H NMR spectrum of this compound closely resembles the spectrum of S,\textsubscript{i}L-Phe\textsubscript{i}His\textsubscript{i}-Thr\textsubscript{i}OMe compound. -13.3°; 92.2; 60.

**S,\textsubscript{i}L-Phe\textsubscript{i}His\textsubscript{i}-Thr\textsubscript{i}OMe:** \textsuperscript{1}H NMR (CDCl\textsubscript{3}, and CD\textsubscript{2}OD, 1:1 (v/v)); 0.9 (t, 3H, CH\textsubscript{3}); 1.3 (m, 23H, (CH\textsubscript{2})\textsubscript{10}); 0.9 (t, 3H, CH\textsubscript{3}); 4.2-4.9 (m, 3H, 3xCH); 7.0 and 8.0 (2 x s, 2 x 1H, 2 CH (Im)); 7.1 (s, 5H, ArH) ppm; -14°; 205.9.

**S,\textsubscript{i}L-Phe\textsubscript{i}His\textsubscript{i}-Ala-OH:** The \textsuperscript{1}H NMR spectrum of this compound closely resembles the spectrum of S,\textsubscript{i}L-Phe\textsubscript{i}His\textsubscript{i}-Ala\textsubscript{i}OMe: +3.9°; 121.4; 60.

**S,\textsubscript{i}L-Phe\textsubscript{i}His\textsubscript{i}-Ala\textsubscript{i}OMe:** \textsuperscript{1}H NMR (CDCl\textsubscript{3}, and CD\textsubscript{2}OD, 1:1 (v/v)); 0.9 (t, 3H, CH\textsubscript{3}); 1.3 (m, 23H, (CH\textsubscript{2})\textsubscript{10}); 0.9 (t, 3H, CH\textsubscript{3}); 4.2-4.9 (m, 3H, 3xCH); 7.0 and 8.0 (2 x s, 2 x 1H, 2 CH (Im)); 7.1 (s, 5H, ArH) ppm; +5.6°; 206.1.

**S,\textsubscript{i}L-Phe\textsubscript{i}His\textsubscript{i}-Leu-OH:** The \textsuperscript{1}H NMR spectrum of this compound closely resembles the spectrum of S,\textsubscript{i}L-Phe\textsubscript{i}His\textsubscript{i}-Leu-OH with the exception: 0.9 (2 x 1H, 2 CH\textsubscript{2} (Im)) ppm; +4.1; 196.1. IR (KBr): 1685 cm\textsuperscript{-1} (CO amide); 1640 cm\textsuperscript{-1} (CO amide).

**S,\textsubscript{i}L-Phe\textsubscript{i}His\textsubscript{i}-Leu\textsubscript{i}OMe:** \textsuperscript{1}H NMR (CDCl\textsubscript{3}, and CD\textsubscript{2}OD, 1:1 (v/v)); 0.9 (t, 3H, CH\textsubscript{3}); 1.3 (m, 23H, (CH\textsubscript{2})\textsubscript{10}); 0.9 (t, 3H, CH\textsubscript{3}); 4.2-4.9 (m, 3H, 3xCH); 7.0 and 8.0 (2 x s, 2 x 1H, 2 CH (Im)); 7.1 (s, 5H, ArH) ppm; +5.6°; 206.1.

**S,\textsubscript{i}L-Phe\textsubscript{i}His\textsubscript{i}-Thr\textsubscript{i}OMe:** \textsuperscript{1}H NMR (CDCl\textsubscript{3}, and CD\textsubscript{2}OD, 1:1 (v/v)); 0.9 (t, 3H, CH\textsubscript{3}); 1.3 (m, 23H, (CH\textsubscript{2})\textsubscript{10}); 0.9 (t, 3H, CH\textsubscript{3}); 4.2-4.9 (m, 3H, 3xCH); 7.0 and 8.0 (2 x s, 2 x 1H, 2 CH (Im)); 7.1 (s, 5H, ArH) ppm; +5.6°; 206.1.
N-Dodecyl(2-L-phenylalanyl)-L-histidyl-L-leucine methyl ester (S$_{2}$,S$_{-}$-Phe-L-His-L-Leu-OMe); N-Butyryl(2-L-phenylalanyl)-L-histidyl-L-leucine methyl ester (S$_{2}$,S$_{-}$-Phe-L-His-L-Leu-OMe); N-(Dodecyloxycarbonyl)-(L-phenylalanyl)-L-histidyl-L-leucine methyl ester (S$_{2}$,S$_{-}$-Phe-L-His-L-Leu-0Me); N-(2-L-phenylalanyl)-L-histidyl-L-alanine methyl ester (S$_{-}$,S$_{-}$-Phe-L-His-L-Ala-OMe).

The IR (KBr) spectra of these compounds were closely similar to the IR spectrum of S$_{-}$,S$_{-}$-Phe-L-His-NMA (see Ref. 1 for the synthesis of these dipeptides). The final yields after recrystallization from methanol amounted to approximately 55%. TLC: R$_{f}$ 0.35 (Silica, CHCl$_{3}$/CH$_{3}$OH, 10:1 v/v) for both compounds. 'H NMR (CDCl$_{3}$): 5 0.9 (m, 9H, 3 x CH$_{3}$); 1.3 and 1.6 (m, 26H, (CH$_{2}$)$_{1}$); 1.9-2.1 (m, 2H, CH$_{2}$ (Im)); 5.3 (d, 1H, NH (carbamate)); 6.7 and 7.4 (2 x s, 2 x CH, 2 x CH (Im)); 7.2 (s, 5H, ArH); 7.2 and 7.5 (2 x d, 2 x 1H, 2 x CH$_{2}$); 7.9 (br, 1H, NH (Im)) ppm; -33.5°; 79.0; 67; 1745, 1690, 1650.

N-Dodecyl(2-L-phenylalanyl)-L-histidyl-L-alanine (S$_{2}$,S$_{-}$-Phe-L-His-L-Ala-OH).

The 'H NMR and IR (KBr) spectra of this compound closely resemble the spectra of S$_{2}$,S$_{-}$-Phe-L-His-L-Ala-OMe; -32.0°; 138.2.

N-Dodecyl(2-L-phenylalanyl)-L-histidyl-L-t-Leu- and (or d)-Tyr-OMe (X and Y are amino acid residues). The tetrapeptides were prepared from the corresponding tripeptides and the methylester of Y in DMF with the aid of 1,3-dicyclohexylcarbodiimide according to a published method.$^{25,26}$ The crude products were purified as described for the corresponding tripeptide methyl esters. The final yields amounted to 50-60%.

TLC: R$_{f}$ 0.35 (Silica, CHCl$_{3}$/CH$_{3}$OH, 10:1 v/v) for both compounds. 'H NMR (CDCl$_{3}$/CD$_{3}$OD, 1:1 v/v) unless noted otherwise; [a]$_{D}$ (c 1.0, methanol); m.p./°C; yield/%; IR (KBr): CO ester, CO carbamate, CO amide/cm$^{-1}$.

- N-Dodecyl(2-L-phenylalanyl)-L-histidyl-L-leucyl-L-leucyl-L-leucine methyl ester (S$_{2}$,S$_{-}$-Phe-L-His-L-Leu-L-Leu-OMe); N-(Dodecyloxycarbonyl)-(L-phenylalanyl)-L-histidyl-L-leucyl-L-leucyl-L-leucine methyl ester (S$_{2}$,S$_{-}$-Phe-L-His-L-Leu-L-Leu-0Me).

- N-Dodecyl(2-L-phenylalanyl)-L-histidyl-L-leucyl-L-Leu-OH; N-(Dodecyloxycarbonyl)-(L-phenylalanyl)-L-histidyl-L-leucyl-L-Leu-OH.

The 'H NMR and IR (KBr) spectra of these compounds closely resemble the spectra of S$_{2}$,S$_{-}$-Phe-L-His-L-Leu-L-Leu-OMe; -0.9°; 65.8; 49.1745, 1690, 1650.

- N-Dodecyl(2-L-phenylalanyl)-L-histidyl-L-t-Leu-OMe and (or d)-Tyr-OMe (X and Y are amino acid residues). The tetrapeptides were prepared from the corresponding tripeptides and the methylester of Y in DMF with the aid of 1,3-dicyclohexylcarbodiimide according to a published method.$^{25,26}$ The crude products were purified as described for the corresponding tripeptide methyl esters. The final yields amounted to approximately 55%.

TLC: R$_{f}$ 0.35 (Silica, CHCl$_{3}$/CH$_{3}$OH, 10:1 v/v) for both compounds. 'H NMR (CDCl$_{3}$/CD$_{3}$OD, 1:1 v/v); [a]$_{D}$ (c 1.0, methanol); m.p./°C; yield/%; IR (KBr): CO ester, CO carbamate, CO amide/cm$^{-1}$.
talization from acetone amounted to approximately 70%. 

'H NMR (CD2OD); [a]D (c 1.0, methanol); m.p.$^\circ$/C; IR (KBr): CO carbamate, CO amide/cm$^{-1}$: 3.0; 7.1; 3.1 (m, 9H, 3xCH); 2.1 (d, 2xH, 2xCH); 4.1 (t, 3xH, 3xCH); 7.0 and 8.0 (2x2, 2x1H, 2x2CH (Im)); 7.15 (s, 2xH, ArH) ppm; -2.9°; 258.7 (dec.); 1685, 1645.

N-(-Dodecylxycarbonyl)-L-phenylalanine 4-nitrophenyl ester [(C$_{20}$-L- (or D-) Phe-ONp) $\tau$ (2.1; 3H, CH$_3$CO); 3.2 (d, 2xH, CH$_2$); 5.0 (2x2, 2x2H, 2x2CH, ArNO$_2$); 7.1 (m, 4H, ArH) ppm; 1755, 1653, 1490, 1347; $\lambda$: 185.0; 139.7; $\beta$: +18.2°; 138.4.

C$_{20}$-L- (or D-) Phe-ONp: $\delta$ 2.1 (s, 3H, CH$_3$CO); 3.2 (d, 2H, CH$_2$); 5.0 (2x2, 2x2H, 2x2CH, ArNO$_2$); 7.1 (m, 5H, ArH) ppm; 1755, 1653, 1525, 1490, 1347; $\lambda$: 185.0; 139.7; $\beta$: +18.2°; 138.4.

C$_{20}$-L- (or D-) Phe-ONp: $\delta$ 0.95 (t, 3H, CH$_3$); 1.3 (m, 2H, CH$_2$); 2.2 (t, 2H, CH$_2$CO); 3.2 (d, 2H, CH$_2$); 5.0 (2x2, 2x1H, 1H, CH); 5.9 (d, 1H, CH$_2$); 7.0 and 8.1 (2x2, 2x2H, 2x2CH, ArNO$_2$); 7.1 (m, 5H, ArH) ppm; IR (KBr): for the C$_{20}$-L- (or D-) Phe-ONp compounds; $\lambda$: -14.2°; 131.5; $\beta$: +14.0°; 130.1.

C$_{20}$-L- (or D-) Phe-ONp: The 'H NMR and IR spectra of these compounds closely resemble the spectra of C$_{20}$-L- (or D-) Phe-ONp.

The 'H NMR and IR spectra of these compounds closely resemble the spectra of C$_{20}$-L- (or D-) Phe-ONp;

References


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