CHAPTER 4

APPROACHES TOWARDS THE 3″-AMINO ANALOGUE OF FEBRIFUGINE

4.1. Background

After achieving the synthesis of model compound (±)-deoxyfebrifugine 14 using our synthetic methodology, we were ready to test this method for the synthesis of 3″-substituted derivatives of febrifugine 1. We opted to begin with approaches to the 3″-amino analogue of 1 (203) because the alkaloid analogue 203 represents a potentially important antimalarial candidate, which has not been synthesized before. One heteroatomic group (hydroxyl) in the natural product 1 is substituted by another such group (amino) in derivative 203. Furthermore, the hydroxyl and amino groups are isoelectronic and of similar size.

Although the prerequisite thiolactam 204 in our proposed synthesis of 203 had not been synthesized before, we found a potentially easy, two-step preparation of 204 in the literature. In 1978, Pifferi’s group published a new method for the preparation of lactams, including (3S)-aminopiperidin-2-one [(S)-205, R= NH₂, Scheme 41], from their corresponding amino acids. This involves conversion of the appropriate amino acid 206 (R = NH₂) into its trimethylsilyl amino ester 207, followed by exo-trigonal ring closure (i.e. during ring closure, the C=O bond broken is exo to the formed ring, and the carbon of the C=O is trigonal) affording lactams 208 of varying ring size, as shown in Scheme 41. This method was claimed to proceed in a non-racemizing fashion, i.e. the 3″-position in (S)-205 does not undergo racemization. Two different methods were used. When using the free amino acid 206, refluxing xylene was used as the solvent and a few drops of TMSCI were added to catalyze the reaction (Method A, Scheme 41). When starting from the amino acid hydrochloride salt 206•HCl, refluxing CH₃CN was used as solvent and HMDS was the only other additive (Method B, Scheme 41).
Scheme 41: Proposed mechanism of lactamization using HMDS, including structures of important lactams (S)-205, 209 and 210.

For simplicity, Scheme 41 also shows the structures of two other important lactams, which will be discussed in this project, i.e. (3S)-hydroxypyrrolidin-2-one (209) and (3S)-hydroxypiperidin-2-one (210). These lactams will be discussed again in Chapter 5. To our knowledge, 210 has never been synthesized by using the method in Scheme 41, but in 1983, Maurey and Srairi prepared 209 in this way. Our use of this method for preparing 210 is further discussed in Chapter 5.

In 1999, Toshima et al. published the first total synthesis of two novel fungitoxic (3-aminopiperidin-2-one)-containing lipids, cepaciamides A and B, by preparing (R)-205 according to Pifferi’s method. The synthesis of cepacamide A is outlined in Scheme 42. Using the conditions mentioned earlier, they obtained (R)-205 from ornithine hydrochloride [(R)-211] as a brownish oil which could not be crystallized. Pifferi’s group did manage to crystallize (S)-205, but they mentioned that it was hygroscopic and needed to be stored in a desiccator under vacuum. Toshima opted to convert crude (R)-205 directly into its amino N-Boc derivative 212 by using 2.6 eq. Boc₂O in CHCl₃. The yield of (R)-212 over two steps was 82%.
Scheme 42: Toshima’s synthesis of cepaciamide A.

From tetradecanol, Toshima managed to prepare key ester 213, which was condensed with \((R)-205\), after both compounds were deprotected using TFA. Diethyl phosphoryl cyanide (DEPC) was found to be most effective for this amide formation reaction, which proceeded in 96% yield. Deprotection of the O-Ac, using \(\text{K}_2\text{CO}_3\) and MeOH, yielded the desired amide 214. The other prerequisite fragment, carboxylic acid 215, was prepared from \((S)\)-malic acid. After esterification between 214 and 215 using DCC, the obtained compound 216 was deprotected using CAN to afford cepaciamide A.
It is important to note in Toshima’s synthesis of cepacamide A that apparently no racemization occurred at position 3 on the piperidin-2-one, even under the basic conditions employed during deprotection of the intermediate amide to form 214 (Scheme 42). The stereochemistry at position 3 remained (R) in 214, as indicated. However, only the stereochemistry at position (3’S) in 214 was confirmed using a modified Mosher’s method. If there were traces of the other (3S)-diastereomer, one presumes Toshima would have observed this compound, and the single diastereomer yields in going from 212 to 214 would not be as high as were found.

In 1983, Winter et al.\textsuperscript{122} prepared both (S)- and (R)-205, using acid-catalyzed ring-closure of (S)-211. The intermediates were then used in a synthesis of enantiomerically pure (S)- and (R)- supidimide 217 (Scheme 43). Importantly, Winter mentioned that the temperature during the HCl-catalyzed ring-closure step to form 205 should not exceed 40 °C so as to prevent racemization from occurring in 205. Unfortunately, Winter presented no evidence that racemization was indeed observed, nor did he mention any references to such a possibility.

Scheme 43: Winter’s preparation of supidimide.

4.2. Approach without \textit{N}-protection of the piperidine ring

4.2.1. Preparation of tert-butyl (S)-2-thioxopiperidin-3-ylcarbamate (204)

Seeing that the absolute stereochemistry in natural, biologically active (+)-1 is 3”S, we started our synthesis of the 3”-amino analogue from (S)-(+)211. This also happens to be the naturally occurring, less expensive enantiomer of ornithine. Using Pifferi’s procedure\textsuperscript{119}, we managed to obtain the intermediate lactam (S)-205
(Scheme 44) as an orange-coloured oil, which we did not attempt to crystallize. As this compound is hygroscopic, it was completely dried under high vacuum and with gentle heating, before proceeding to the next step.

![Scheme 44: Synthesis of 204.](image)

Dried, crude (S)-205 was converted into (S)-212 by acylation with 1.7 eq. of Boc₂O, i.e. considerably less than the 2.6 eq. used by Toshima. The yield of (S)-212 was 73% over 2 steps, comparable to the 82% yield obtained by Toshima. The ¹H NMR spectral data agreed with those obtained by Toshima. The Boc NH was observed as a broad singlet at δ_H 6.59 and the lactam NH was observed similarly at δ_H 5.51. The Boc C(CH₃)₃ protons were observed as a singlet, which integrated for nine protons, at δ_H 1.45. The optical rotation we observed for (S)-212 in CHCl₃ ([α]_D +54.4) agreed very well with what was expected from Toshima’s measurement for (R)-212 ([α]_D – 57.4 in CHCl₃). Next, thiolactam 204 was obtained from (S)-212 in 82% yield using Lawesson’s reagent, or in 81% yield using the P₂S₅/Na₂CO₃ method. It was obvious from the ¹H and ¹³C NMR data that we did indeed obtain the desired thiolactam 204. The thiocarbonyl carbon was observed at δ_C 203.5 and the Boc carbonyl carbon at δ_C 155.5. Furthermore, the deshielding effect of the sulfur atom was again apparent in the ¹H NMR spectrum. The thiopiperidone NH was highly deshielded and observed as a broad singlet at δ_H 8.65, whereas the Boc NH was less deshielded at δ_H 5.93. The hydrogen alpha to the thiocarbonyl group was also highly deshielded at δ_H 4.16, as expected.
As will be seen in the next section, a single crystal of 204 was found to be centrosymmetric. This was a very disappointing result, as we had hoped that racemization of 204 might not occur at all. The specific rotation observed for the obtained product 204 in CHCl₃ was \([\alpha]_D -2.2\) (for the material obtained by the Lawesson’s thionation method, Method A, Scheme 44). This laevorotation was similar to that observed for precursor (S)-205 ([\alpha]_D -12.4, in CHCl₃) but different to that expected for precursor (S)-212 (+ rotation expected, as \([\alpha]_D +54.4\) was found for (S)-212, in CHCl₃). We did not do HPLC work to determine the ratio of enantiomers obtained. It was sufficient evidence from the crystal data that undesired racemization was occurring, the extent of which was not important at this stage as we were simply testing our synthetic methodology. We also hoped that, should diastereomers arise at a later stage in the synthesis, these might be easily resolved.

How might racemization at position 3 in 204 occur? As mentioned before, Winter et al. thought that racemization of precursor lactam 205 occurred above 40 °C under acidic conditions, without presenting conclusive evidence. We know that Toshima encountered no such problems under basic conditions with precursor lactam 212. The answer might lie in the fact that 204 is a thiolactam, bearing a highly deshielding group (NHBoc) at the position alpha to the thiocarbonyl function. It is well known that the sulfur in thioamides is considerably more nucleophilic than the oxygen in corresponding amides. This is why thioamides are easily S-alkylated, e.g. in 1969, Gompper and Elser prepared isolable ketene S,N-acetals 218, from thioamides 219 using methyl iodide and potassium tert-butoxide, as shown in Scheme 45.

![Scheme 45: Synthesis of ketene-S,N-acetals 218.](image)

Similarly, the sulfur in 219 might be easily protonatable under acidic conditions, and the alpha-hydrogen in 219 might be deprotonated under basic conditions. As deduced
from Scheme 45, such proton exchange processes would ultimately result in racemization at the position alpha to the thiocarbonyl group. These processes are not as favoured in amides as in thioamides, which contain strongly nucleophilic and electron-withdrawing sulfur atoms. The “enol” form can be viewed as being more pronounced in thioamides, compared to in the corresponding amides. Furthermore, if the alpha carbon in 219 is substituted by an electron-withdrawing group, e.g. the Boc group in 204, racemization at the alpha carbon might be accelerated owing to the increased acidity of the alpha-hydrogen.

Acidic or basic conditions might therefore cause racemization during the three-step preparation of 204 in Scheme 44. Lawesson’s reagent is known to be slightly acidic \(^{125}\), and the heating required during this method might promote racemization. The basic conditions used in the P\(_2\)S\(_5\) thionation work-up procedure might cause racemization in that case. Furthermore, the slightly acidic silica gel used for purification could be detrimental to the enantiomeric purity of 204. Finally, it is possible that racemization is a slow process, which occurs upon storage of 3-substituted thiolactams such as 204. We obtained single crystals of 204 by keeping it for a prolonged period in EtOAc-hexane solution. It might be coincidental that a centrosymmetric crystal was selected for X-ray diffraction characterization purposes, and that the product 204 was not completely racemized, as suggested by the optical rotation data.

Future work includes the search for milder conditions, especially during thionation, in the preparation of 204 and related thiolactams. Importantly, studies on the racemization of the alpha-position in thioamides, using HPLC, are required. No such studies were found in the literature. Even though the loss of stereochemical purity in 204 was a discouraging outcome, we decided to continue this route to check whether the subsequent steps might work, before trying to solve the racemization problem.

4.2.2. Crystal structure of 204

The crystal data for 204 are summarized in Table 12. Note that all the detailed crystal data for structures determined during this project are available on the accompanying compact disc.
Crystal system: Orthorhombic

Space group: \( \text{Pbca} \)

Unit cell dimensions:
\[
\begin{align*}
a &= 11.1118(14) \text{ Å} & \alpha &= 90^\circ. \\
b &= 11.0591(14) \text{ Å} & \beta &= 90^\circ. \\
c &= 20.603(3) \text{ Å} & \gamma &= 90^\circ.
\end{align*}
\]

Volume: 2531.8(6) Å\(^3\)

\( Z \): 8

Density (calculated): 1.209 Mg/m\(^3\)

Absorption coefficient: 0.241 mm\(^{-1}\)

\( F(000) \): 992

Crystal size: 0.40 x 0.36 x 0.30 mm\(^3\)

Theta range for data collection: 1.98 to 28.30°.

Index ranges:
\[-14 \leq h \leq 14, -14 \leq k \leq 14, -16 \leq l \leq 27\]

Reflections collected: 16492

Independent reflections: 3140 [\( R(\text{int}) = 0.0324 \)]

Completeness to theta = 28.30°: 99.8 %

Absorption correction: None

Refinement method: Full-matrix least-squares on \( F^2 \)

Data / restraints / parameters: 3140 / 0 / 139

Goodness-of-fit on \( F^2 \): 1.021

Final \( R \) indices \([I>2\sigma(I)]\): \( R1 = 0.0391, wR2 = 0.1065 \)

\( R \) indices (all data): \( R1 = 0.0645, wR2 = 0.1213 \)

Largest diff. peak and hole: 0.267 and -0.218 e.Å\(^{-3}\)

Table 12: Crystal data and structure refinement for \( \text{204} \).

In 1989, Valle et al.\(^{126} \) published the crystal structure of precursor lactam, \((3S)-3\text{-}\text{tert-}\text{butyloxy}\text{carbonylaminopiperidin-2-one}, \,(S)-\text{212}.\) It is interesting to compare the structures of lactam \((S)-\text{212}\) and the corresponding thiolactam \( \text{204}.\) In the case of enantiomerically pure \((S)-\text{212}, \) the crystal system is monoclinic (as opposed to orthorhombic for \( \text{204}.\)) and the space group is \( P2_1 \) (as opposed to the centrosymmetrical space group \( \text{Pbca} \) for racemic \( \text{204}.\)) In the crystal of \((S)-\text{212}, \) Valle
*et al.* found two molecules in the asymmetric unit. The δ-lactam ring of Molecule A was in an approximate half-chair conformation, while that of Molecule B was in the boat conformation. We found that related thiolactam 204 crystallized only in an approximate half-chair conformation, as shown in the Ortep diagram for 204 given in Figure 17. It can be seen that N1, C2, C3, C5 and C6 are approximately planar, whereas C4 is out-of-plane.

![Ortep diagram (50% ellipsoid probability) of 204, showing the atomic numbering scheme.](image)

As observed by Valle for (S)-212, the Boc group in 204 is in the expected extended conformation. How does the sulfur in thiolactam 204 affect the surrounding molecular environment, compared to what was observed by Valle for lactam (S)-212? Selected bond lengths in the two structures are compared in Table 13, whereas selected bond angles and torsion angles are compared in Table 14.

<table>
<thead>
<tr>
<th>Bond</th>
<th>(S)-212 Molecule A (Å)</th>
<th>(S)-212 Molecule B (Å)</th>
<th>204 (Å)</th>
</tr>
</thead>
<tbody>
<tr>
<td>C(2)-N(1)</td>
<td>1.337 (9)</td>
<td>1.327 (10)</td>
<td>1.3154 (19)</td>
</tr>
<tr>
<td>C(2)-C(3)</td>
<td>1.529 (11)</td>
<td>1.520 (10)</td>
<td>1.522 (2)</td>
</tr>
<tr>
<td>C(2)-S, or C(2)-O(3)</td>
<td>1.238 (10)</td>
<td>1.220 (9)</td>
<td>1.6755 (16)</td>
</tr>
<tr>
<td>C(3)-N(2)</td>
<td>1.454 (9)</td>
<td>1.458 (8)</td>
<td>1.4547 (18)</td>
</tr>
<tr>
<td></td>
<td>(S)-212 Molecule A (°)</td>
<td>(S)-212 Molecule B (°)</td>
<td>204 (°)</td>
</tr>
<tr>
<td>----------</td>
<td>------------------------</td>
<td>------------------------</td>
<td>---------</td>
</tr>
<tr>
<td>N(1)-C(2)-C(3)</td>
<td>117.7 (9)</td>
<td>113.4 (8)</td>
<td>117.05 (13)</td>
</tr>
<tr>
<td>N(1)-C(2)-S, or N(1)-C(2)-O(3)</td>
<td>121.9 (8)</td>
<td>122.8 (8)</td>
<td>121.43 (12)</td>
</tr>
<tr>
<td>C(3)-C(2)-S, or C(3)-C(2)-O(3)</td>
<td>120.2 (10)</td>
<td>123.6 (10)</td>
<td>121.50 (11)</td>
</tr>
<tr>
<td>N(2)-C(3)-C(4)</td>
<td>112.4 (7)</td>
<td>109.1 (8)</td>
<td>112.66 (13)</td>
</tr>
<tr>
<td>N(2)-C(3)-C(2)</td>
<td>110.2 (8)</td>
<td>112.3 (8)</td>
<td>109.55 (12)</td>
</tr>
<tr>
<td>C(4)-C(3)-C(2)</td>
<td>114.1 (8)</td>
<td>110.7 (7)</td>
<td>112.66 (13)</td>
</tr>
<tr>
<td>C(3)-C(2)-N(1)-</td>
<td>12.4 (14)</td>
<td>0.2 (13)</td>
<td>-10.7 (2)</td>
</tr>
</tbody>
</table>

Table 13: Comparison of bond lengths between (S)-212 and 204. Note that “S” in 204 becomes “O(3)” in (S)-212.

The bond lengths are very similar in the two compounds (Table 13). However, the presence of strongly electron-withdrawing sulfur in 204 does cause a slight shortening of the N(1)-C(2) (1.32 Å), as well as the C(3)-C(4) (1.52 Å) bonds in the ring, compared to these bonds in (S)-212 [ca. 1.34 and 1.54, respectively]. It can also be seen that bond C(7)-O(2) is shorter (1.34 Å) in 204 than in (S)-212 (1.35 and 1.36 Å), and that bond C(8)-O(2) is longer (1.48 Å) in 204 than in (S)-212 (1.47 and 1.45 Å). This is a consequence of the slightly stronger intermolecular H-bonds involving O(1) in 204 (see later), as opposed to those found in (S)-212. The thiolactam C=S bond, C(2)-S, is of expected magnitude [1.6755(16) Å, compared to the mean value of 1.671 Å for (X)2NC(=S), X = C, N, O, S]127.
From Table 14, it can be seen that most of the bond angles and torsion angles are very similar in (S)-212 and 204. The angles correspond better between Molecule A of (S)-212 and 204. This is because both these molecules consist of δ-lactam rings in the half-chair conformation, whereas Molecule B is in the boat conformation. Importantly, similar to what was found for the amide group in lactam (S)-212, the thioamide group in 204 is non-planar as indicated by the torsion angles C(3)-C(2)-N(1)-C(6) (-10.7°) and S-C(2)-N(1)-C(6) (170.8°). The respective rotations of the ring relative to the tert-butyloxycarbonylamino substituent [torsion angles C(2)-C(3)-N(2)-C(7)] for Molecules A (52.3 °) and B (-86.5 °) in (S)-212, and for 204 (143.1 °), differ greatly in value.

Figure 18 is a representation of the unit cell and its contents in 204. It clearly demonstrates the centrosymmetry (i.e. symmetry from the inversion centre) in structure 204, which indicates that racemization had occurred at carbon 3.
Figure 18: Representation of the unit cell in 204 showing centrosymmetry in the crystal. Four molecules were removed for clarity.

Similar to the results obtained by Valle et al. for (S)-212, no intramolecular H-bonding is observed in the structure of 204. Instead, (thiolactam)N(1)-H···O(1)=C(Boc) intermolecular H-bonds are observed along the B plane, as seen in Figure 19. The N(1)···O(1) separation is 2.823 Å, i.e. shorter than that observed by Valle for (S)-212 (2.990 and 2.880 Å between Molecules A and Molecules B, respectively). The corresponding N(1)-H(1)···O(1) distance is 1.984 Å. This results in a layered structure, each layer formed by intermolecular H-bonds extending in one plane. Valle additionally observed (Boc)N(2)-H···O(3)=C(lactam) intermolecular H-bonds in (S)-212. No such close contacts were found between sulfur and the (Boc)N(2)-H in 204. It is important to note that, seeing that the hydrogen atoms in all the structures determined during this project were first located in a difference map and then placed on the parent atoms, hydrogen bonds are indicated between the heteroatoms which share the bond (as shown in Figure 19), and not directly between the hydrogen atom and the hydrogen bond acceptor.
4.2.3. **Sulfide contraction reaction between 204 and 105, and attempted hydrogenation of the product**

With the desired racemic thiolactam 204 in hand, the Eschenmoser reaction with bromide 105 could be attempted. As this reaction worked quite pleasingly in the model studies (see Chapter 3), we decided to use exactly the same experimental conditions as used for the N-protected model thiolactams. The use of the stronger base, 1-methylpiperidine, was explained in section 3.6. Thus, following this procedure, we obtained a moderate yield (apparently 64%) of what initially appeared to be the desired vinylogous amide 220 (Scheme 46) as a viscous oil, which unfortunately could not be induced to crystallize.

![Scheme 46: Reaction between 105 and 204.](image)

Figure 19: Intermolecular H-bonding in 204, as viewed along the b-axis. Four molecules were removed for clarity.
The $^1$H NMR spectrum of product 220 gave cause for alarm. It should be noted that we could not prove unequivocally that we had indeed obtained structure 220. For convenience, the $^1$H NMR spectrum of 220 is compared to that of an analogue of 220 [the 1-(4-methoxybenzyl) derivative 221, see later in Section 4.3.1.], here. All the quinazolinyl proton signals in 220 were observed at the expected chemical shifts, and in the correct multiplicities. The only deviation from the normal trend was H-2 ($\delta_H$ 8.35, i.e. the hydrogen attached to the imine C in Q, Scheme 46). Usually, we would expect this singlet to appear at a lower chemical shift (e.g. at $\delta_H$ 7.91 in 221). Although the remaining signals integrated correctly for the remaining 21 protons in 220, the chemical shifts were very different from those observed for analogue 221. Furthermore, all the signals were broadened. As expected, two distinct doublets were observed for the NCH$_2$C(O) protons, but at considerably higher chemical shifts ($\delta_H$ 5.78 and 5.66, respectively) than in 221 ($\delta_H$ 4.67 and 4.56, respectively). The H-3" alpha to the tert-butoxycarbonylamino substituent also absorbed at a higher chemical shift ($\delta_H$ 5.30) than its counterpart in 221 ($\delta_H$ 4.98). The next four assignments were more ambiguous and questionable. Two broadened multiplets at $\delta_H$ 4.86 and 4.66, integrating for one proton each, were attributed to the ring NCH$_2$ protons, owing to the multiplicities (two multiplets) of the signals. The corresponding protons in 221, however, absorb much lower at $\delta_H$ 3.63 and 3.22. Then a broad singlet at $\delta_H$ 3.5 and integrating for two protons, was assigned to the two NH protons in 220. However, as mentioned before, all signals were very broadened compared to those observed in other analogous compounds. The fact that this signal appears to be a singlet is not conclusive evidence that it belongs to NH protons. Also, the corresponding Boc NH in 221 absorbs at $\delta_H$ 5.88, i.e. at a considerably more deshielded position. Then, two multiplets ($\delta_H$ 2.47 and 2.33) were attributed to the remaining four ring methylene protons. These chemical shifts are again much higher than expected (e.g. $\delta_H$ 2.26, 2.00, 1.85 and 1.70, for the four protons in 221, respectively). The nine Boc protons were observed as a singlet at the expected chemical shift ($\delta_H$ 1.44, compared to $\delta_H$ 1.40 for 221). Importantly, the only peak that could be assigned to the vinyl proton in 220, was a very highly deshielded singlet at $\delta_H$ 8.77. This singlet is observed at $\delta_H$ 5.06 for 221. This was a major deviation, which puzzled us. This signal could also be assigned to the BocNH, although it is perhaps
too sharp a signal for these two assignments to be exchanged. It seems possible that
the presence of the free NH group in the piperidinyl ring of 220 could H-bond in such
a way that it results in substantial deshielding of the atoms surrounding the enaminone
functional group, as compared to the case where this nitrogen is protected by an
electron-donating alkyl group, such as the PMB group in 221. It is important to note
that the enaminone in product 220 is not necessarily in the Z-configuration as drawn
in Scheme 46. The BocNH group can also H-bond, which might lead to a mixture of
Z/E products. This might also explain the differences in chemical shifts observed for
220 compared to 221, and might also lead to a broadening of signals.

The broadened signals in the $^1$H NMR spectrum (which could suggest a mixture or an
unstable compound), together with the fact that no expected molecular ion was
present in the mass spectrum of 220 (see experimental, Section 8.1.3.), were both
discouraging results. The absence of a molecular ion in the mass spectrum also
supports the suspicion that 220 is unstable. Despite several repetitions of this
experiment, the same product was isolated in each case and, even though it appeared
as a single spot on TLC, product 220 was very polar, difficult to purify, and darkened
upon storage, which further suggested decomposition. We nevertheless decided to
attempt the next reaction in our synthetic sequence, i.e. hydrogenation, on freshly
prepared material of 220, in the hope of isolating a more stable, or crystalline
compound, and with the aim of casting more light on the possible structure of 220.

Unfortunately, attempted chemoselective hydrogenation of the putative enaminone
C=C bond in 220, using conditions similar to those used on the model vinylogous
amides, failed. These reaction conditions seemed to lead to the decomposition of 220,
as evident from the five, uncharacterizable spots isolated by column chromatography
of the crude product (see experimental, Section 8.1.4.). At this stage, the structure of
the product (220) obtained remains a mystery.

4.3. Approach using N-(4-methoxybenzyl) protection

As the route without piperidinyl $N$-protection failed, we decided to use a protecting
group on N1″ instead. It was hoped that the advanced intermediates thus obtained
would be more easily isolable and more stable than those obtained in the previous section. A popular amino protecting group is the PMB (4-methoxybenzyl) group. This group can also be used to protect amides. We chose this group primarily because it represents an electron-donating protecting group (as explained in Section 2.2.), which should be easily removable from the amide nitrogen using e.g. CAN \(^ {128}\) (ceric ammonium nitrate) and from the amine nitrogen using e.g. standard catalytic hydrogenation methods \(^ {129}\), or chloroethyl chloroformate \(^ {130}\). The PMB group is also very stable under basic conditions such as those required during the Eschenmoser reaction to produce our key vinylogous amide intermediate.

4.3.1. Preparation of 3-\((E\)-3-\(3\)-(tert-butoxycarbonylamino)-1-(4-methoxybenzyl)piperidin-2-ylidene\)2-oxopropyl\)quinazolin-4(3\(H\))-one (221)

Scheme 47 summarizes the results obtained in this section. The lactam, 1-(4-methoxybenzyl)piperidin-2-one \(^ {222}\), is a known compound, which has previously been prepared before from piperidin-2-one \(^ {131}\) (see later, Section 5.3.4.). Using similar experimental conditions, we managed to prepare our starting lactam \(^ {223}\) (Scheme 47) as a stable and crystalline compound, in 71% yield from \((S)\)-212 and 4-methoxybenzyl alcohol. Tosylation \textit{in situ} of 4-methoxybenzyl alcohol affords 4-methoxybenzyl tosylate. The tosyl group is easily nucleophilically substituted by the amine nitrogen in deprotonated \((S)\)-212 to afford alkylated lactam \(^ {223}\). Fortunately, the Boc nitrogen did not seem to undergo side-reactions under these highly basic (NaH) reaction conditions. Although \(^ {223}\) was optically active \(\left[\alpha\right]_{D}^{20} 14.4\), it cannot be stated with certainty that no racemization had occurred, considering that the presence of NaH could induce base-catalyzed racemization to some extent as explained in section 4.2.1. The \(^ {13}\)C NMR spectrum of \(^ {223}\) clearly showed the presence of both a Boc carbonyl carbon (\(\delta_{C} 159.1\)) and a lactam carbonyl carbon (\(\delta_{C} 169.6\)).

Thionation of \(^ {223}\) using Curphey’s method (\(\text{P}_{2}\text{S}_{5}, \text{HMDO}\))\(^ {94}\) afforded a disappointing yield (40%) of prerequisite thiolactam \(^ {224}\). The \(^ {13}\)C NMR spectrum clearly indicated
the presence of a thiocarbonyl carbon (δC 201.0). The Boc (CH$_3$)$_3$ protons were observed as a singlet at the expected chemical shift (δH 1.47) in the $^1$H NMR spectrum of 224. Although these thionation conditions are mild, the work-up procedure included use of saturated aq. K$_2$CO$_3$ to separate impurities from the product in the organic phase. This could again induce racemization and perhaps the decomposition of 224 by basic hydrolysis. It is recommended that the crude solution be simply filtered through celite prior to column chromatography, without using base in the work-up. Alternatively, Lawesson’s reagent should be used to effect thionation of base-sensitive 3″-substituted lactams. This might increase the yield of the desired thiolactams, whilst reducing the extent of racemization. We did find that the obtained 224 was stable, crystalline, optically active ([α]$_D$ 20 –14.5) and exhibited a sharp melting point (see experimental, Section 8.2.2.).

Scheme 47: Preparation of 221.

Eschenmoser reaction between 224 and bromide 105 afforded a moderate yield (52%) of the desired vinylogous amide 221 as a colourless, stable solid. This was a very pleasing result, as optically active 221 ([α]$_D$ 20 –5.49) may be viewed as an advanced intermediate in our synthesis of 3″-substituted analogues of febrifugine 1. No 3″-nitrogen-containing substituted analogues of 1 had been synthesized before, which makes 221 an interesting candidate for antimalarial testing. As mentioned in Chapter 1, the biological activity of dehydro derivatives of 1, such as enaminone 221, has not been determined. Our synthesis of relatively complex molecule 221 is simple and inexpensive. Owing to time constraints, the yields were not optimized, nor did we
attempt potentially straight-forward Boc-deprotection of 221 to obtain the 3”-amino-dehydro- derivative of 1. Future work includes all the aforementioned studies.

4.3.2. Attempted hydrogenation of 221; the isolation of an apparently dehydrogenated derivative (225/226)

The next step in the synthesis of amino analogue 203 is the chemoselective hydrogenation of the enaminone C=C bond in 221. Using the standard conditions we established in section 3.8.3. for the reduction of the N-alkylated model compounds (PtO₂, glacial AcOH, H₂), we obtained three isolable spots on TLC after stirring the reaction mixture for 72 h under 1 atm of hydrogen gas. The two most polar products, obtained only in trace amounts, were unidentifiable by NMR spectroscopy. The NMR spectra were complex and possibly indicated decomposition of 221. We did, however, manage to isolate an oil, which was apparently a dehydrogenated derivative of 221, characterizable by NMR and mass spectroscopy. It is thought to possess either structure 225 or 226 as shown in Scheme 48, in which case it was obtained in 25% yield from 221. Presumably, the conversion of 225 to isoxazine 226 by an electrocyclic reaction is reversible. Unfortunately, crystallization of the obtained product could not be achieved.

Scheme 48: Two alternative proposed structures formed during the attempted hydrogenation of 221.

It is well known that dehydrogenation of carbamates such as 221 can occur in the presence of metal oxides and base, e.g. Büchi et al.¹³² prepared bright red imine 227
from a carbobenzyloxy compound using MnO₂ or NiO and NEt₃ (see Scheme 49), in
their synthesis of the biologically important cofactor methoxatin.

Scheme 49: Büchi’s synthesis of methoxatin.

Evidence that we might have prepared 225 or 226 comes from the spectroscopic data. The mass spectrum showed a molecular ion, M⁺ 516, which corresponds to dehydrogenated 221. The IR spectrum contained no broad N-H absorption peak. The ¹H NMR spectrum contained signals integrating for the required number of protons. The quinazolinone protons were observed at the expected chemical shifts and of expected multiplicities. Decreased coupling throughout the remaining part of the structure pointed to the absence of a stereogenic centre at carbon 3″. Thus, two singlets (at δH 5.37 and 3.97) were observed for the CH₂Ar and NCH₂C(O) protons, respectively. For precursor 221, these protons were observed as four doublets at δH 4.67, 4.56, 4.52 and 4.34, respectively. The differences in chemical shifts of these protons between 221 and 225/226 are quite surprising, though. As expected for structure 225/226, we did not observe a highly deshielded multiplet for H-3″ (in 221), which is therefore absent. The vinyl hydrogen singlet in 225/226, NC=CH, was observed at δH 5.94 (similar, but more deshielded than in 221, where δH = 5.06). This ¹H chemical shift agrees with that expected for an oxazine ring proton at the corresponding 4-position of the ring. In 1981, Abramovitch and Dupuy 133 obtained 1,2-oxazines “isoxazines” by the thermolysis of 2-azidopyridine 1-oxides in benzene at 90 °C. The simplest isoxazine 228, which they isolated, is shown here (the numbering scheme is included). In the ¹H
NMR spectrum, H-5 was observed [as a ddd, $J_{5,6} = 5.1$ Hz]) at $\delta_H 6.1$, in good agreement with our tentative assignment of the corresponding proton (at $\delta_H 5.94$) if the structure of our isolated compound is that of 226 (Scheme 49). The remaining signals in the $^1$H NMR spectrum of 225/226 (i.e. methoxy protons, ring methylene protons and Boc protons) were as expected, and comparable to those observed for precursor 221, both in chemical shift and multiplicity.

The $^{13}$C NMR spectrum of 225 was complex, owing to the large number of signals in the carbonyl and aromatic region. However, we could assign a few of these (see experimental, Section 8.2.4.), which were comparable to those observed for 221. As expected, the enaminone C=O signal was not observed in 225. Furthermore, it is reasonable to expect the quaternary carbons C-2” and C-3” to be highly deshielded and to absorb in the aromatic region. These could not be assigned with certainty, but two “additional” quaternary carbon peaks were indeed observed in this region. In the $^{13}$C NMR spectrum of 228 (obtained by Abramovitch and Dupuy$^{133}$) carbons 3, 4 and 5 were observed at $\delta_C 148.2, 116.45$ and 124.2, respectively. Although we could not be sure about whether the signal we observed in the $^{13}$C NMR spectrum of 225/226 at $\delta_C 148.1$ was attributed to the analogous C-3, i.e. C-3” in 226, or to the quinazolinone C-8a, we did observe a quaternary carbon at $\delta_C 115.1$ (i.e. similar to C-4 in 228) which might therefore be assigned to C-2” in 226. Five carbon signals were observed at ca. $\delta_C 126-129$ in the spectrum of 226 and it cannot be speculated which one might be attributed to C-3’ (similar to C-5 in 228) in 226. The other carbons in 226 were all tentatively assigned, all within reason, by comparing the assignments to those in precursor 221. Importantly, no carbonyl carbon peak was found corresponding to the carbonyl carbon in structure 225, unless this carbon is more shielded than expected and absorbs at $\delta_C 135.2$ (a quaternary carbon peak which could not be unequivocally assigned). It is therefore more likely that we isolated the isoxazine 226.

Unfortunately, we could not find reported NMR spectral data for isoxazines, which are more similar to structure 226 than the oxazine 228. A Beilstein database search revealed that isoxazine-containing compounds usually consist of the isoxazine ring fused to a benzene ring. None were found which were fused to a tetrahydropyridine
ring, such as 226, nor to a cyclohexene-based ring. Furthermore, no NMR spectral data were found of a compound containing an imine which is substituted on an enaminone group in a way similar to structure 225.

It should be noted that we cannot claim with total certainty that we did isolate 225 or 226 as proposed. The important point is that, for reasons not easily explained, vinylogous amide 221 was either decomposed or untouched when subjected to the hydrogenation conditions which worked well to reduce the model enaminones chemoselectively. The presence of a 3″-substituent therefore alters the properties and the reactivity of cyclic enaminones such as 221 to a greater extent than initially predicted. The presence of an inductively electron-withdrawing group, e.g. NHBoc, on C-3″ seems to prevent easy hydrogenation of the C=C bond in the neighbouring enaminone group.

4.4. Approach using N-(2-methylnaphthyl) protection

Our interest in the NAP (2-methylnaphthyl) group as an alternative alkyl amino-protecting group to the Bn group, which we were unable to remove during the model study, led us to attempt the synthesis of 203 using the NAP group. We also thought that the use of an alternative protecting group might alter the reactivity and isolability of advanced intermediates, which thus far proved to be problematic in our proposed synthesis of 203.

Prerequisite lactam 229 was prepared in 68% yield from lactam (S)-212 and 2-(bromomethyl)naphthalene (Scheme 50) according to the conditions mentioned before (Section 3.4.1.). In the 1H NMR spectrum of 229, the Boc NH proton was observed as a broad singlet at δH 5.59 The NCH2Ar protons were observed as two distinct doublets (at δH 4.80 and 4.69, respectively, with J = 14.6 Hz for each) owing to the stereogenic centre at position 3 of the lactam ring. The signals of two carbonyl carbons were observed in the 13C NMR spectrum (one signal at δC 169.8 can be assigned to the lactam carbonyl group and another, at δC 156.0, can be assigned to the Boc carbonyl group).
Scheme 50: Attempt towards 203 using NAP as an amino-protecting group.

Thionation of 229, this time using the recommended Lawesson’s method, afforded a high yield (97%) of the corresponding thiolactam 230. The $^{13}\text{C}$ NMR spectrum clearly indicated the presence of a thiocarbonyl carbon, as reflected by the peak at $\delta_C 201.5$. The Boc carbonyl carbon was observed at $\delta_C 155.3$ in this case. Owing to the increased electron-withdrawing effect by the nearby thiocarbonyl sulfur atom, the Boc NH proton signal was observed as a broad singlet at $\delta_H 6.41$, which is significantly more deshielded than the analogous proton in precursor lactam 229 (see above).

Although the Eschenmoser reaction between 230 and 105 seemed to lead to the formation of the desired vinylogous amide 231 in moderate yield ($< 47\%$ if the correct structure was obtained), product 231 (a viscous colourless oil) was difficult to isolate and contained trace impurities even after repeated purification attempts using column chromatography. Furthermore, the NMR spectra were complex, owing to the numerous aromatic signals observed. The $^1\text{H}$ NMR spectrum of 231 did show the presence of two identifiable protons in the quinazolinyl group, i.e. H-5 as a doublet at $\delta_H 8.28$ ($J = 7.9$ Hz), and H-2 as the expected singlet at $\delta_H 7.87$. The remaining nine aromatic signals overlapped to form a multiplet at $\delta_H 7.38-7.84$. The Boc NH proton was observed as a broad singlet at $\delta_H 5.98$, and the vinyl proton in the enaminone
group was observed as a singlet at $\delta_H$ 5.12. Furthermore, H-3 (alpha to the Boc substituent) formed a broad singlet at $\delta_H$ 5.01. The $\text{NCH}_2\text{Ar}$ protons were observed as two distinct doublets ($J = 16.1$ and $16.0$) at the expected chemical shifts ($\delta_H$ 4.65 and 4.47, respectively). Two doublets ($J = 16.6$ for each) were also observed for the $\text{NCH}_2\text{C(O)}$ protons (at $\delta_H$ 4.81 and 4.56). Although all the aliphatic and the vinyl ($\text{NC}=$CH) carbon signals could be assigned in the $^{13}\text{C}$ NMR spectrum of 231, a few additional peaks (probably as a result of a small amount of an aromatic impurity) were observed in the aromatic region. However, both the quinazolinone and the Boc carbonyl carbons could be reliably assigned, as well as the peaks belonging to the NC=CH, C-8a and C-4a quaternary carbons (see experimental, Section 8.3.3).

Again in the hope of possibly isolating a crystalline and easily characterizable product, hydrogenation of 231 was carried out in an attempt to produce key amine 232. Unfortunately, a mixture of unidentifiable products (the NMR data of which pointed to decomposition of 231) was obtained. This experimental route was consequently discontinued.

### 4.5. Conclusion

In this chapter, we attempted the synthesis of an important 3-amino analogue of febrifugine (1) from L-ornithine. It was found that the use of an $N$-alkylated piperidine-2-thione was required if a stable, isolable vinylogous amide intermediate was to be obtained. The interesting 3-acylamino dehydro febrifugine analogue 221, which is an analogue of 1 that might be worthwhile submitting for antimalarial testing, was prepared in this way. Attempted chemoselective $\text{C}=\text{C}$ hydrogenation of the enaminone group in 221 unexpectedly resulted in the isolation of putative isoxazine 226. Future work includes the repetition and/or the optimization of the hydrogenation reaction which produced 226. The use of alternative conditions, e.g. NiO and base, to prepare 226 from 221 might be useful to confirm the structure of 226. The attempted chemoselective reduction of 221 to form the desired azafebrifugine analogue should be repeated by employing an alternative reducing agent, preferably one that does not involve catalytic hydrogenation.