## Revised Structure of Tolyporphin A**

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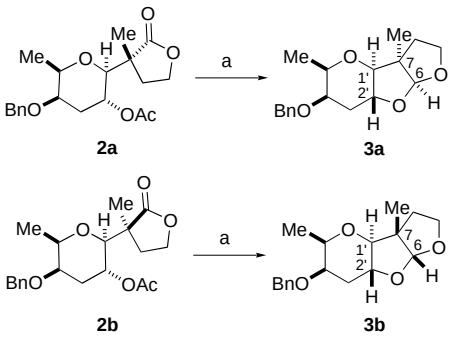
As reported in the preceding communication,[1] we have completed the total synthesis of **1b**, the structure proposed for (+)-tolyporphin A *O*,*O*-diacetate, but have found the synthetic compound to be different from the *O*,*O*-diacetate prepared from natural (+)-tolyporphin A.[2] The structure of the natural product was originally proposed to be **1a** on the basis of $^1H-^{13}C$ HMBC and $^1H-^1H$ ROESY experiments.[3] In particular, the configuration at the quaternary centers C7 and C17 was assigned on the basis of NOEs observed between the C1′ protons on the carbohydrate residues and the C5 and C15 *meso* porphyrin protons and between the C2′ acetyl methyl protons on the carbohydrate residues and the C10 and C20 *meso* porphyrin protons.[4] Here we describe spectroscopic studies on both the synthetic and natural products and propose a revised structure for tolyporphin A, the representative member of the tolyporphin class of natural products.[5]

**1a** (X=H) : "tolyporphin A"
**1b** (X=Ac) : "tolyporphin A *O,O*-diacetate"

Since rings A and C of the synthetic substance were derived from precursor **2a**, the stereochemical assignment of the quaternary center was first addressed at the monocyclic stage. Both precursor **2a**[1] and its diastereomer **2b**[1] were converted into the conformationally rigid tricyclic acetals **3a** and **3b**, respectively (Scheme 1), and subjected to NOE experiments. Irradiation of the C7 quaternary methyl group of **3a** enhanced the signals for both the C6 acetal proton and the C1′ axial anomeric proton of the sugar. On the other hand, irradiation of the C7 quaternary methyl group of **3b** enhanced the signals for the C6 acetal proton and the C2′ axial proton of the sugar.

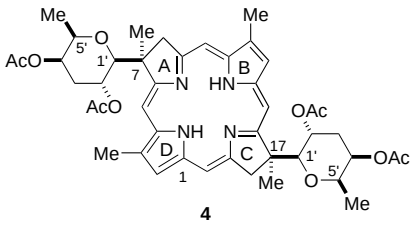



**2a** **3a**
**2b** **3b**

Scheme 1. Synthesis of **3a** and **3b**. a) 1. diisobutyl aluminum hydride (DIBAL-H), toluene, −78 °C; 2. pyridinium-*p*-toluenesulfonate (PPTS), DMF, RT.

Based on this experiment, the configurations at the C7 quaternary centers of **2a** and **2b** were assigned as *R* and *S*, respectively, and the diastereomer **2a** was then used to assemble the tetrahydroporphyrin ring.

Considering the synthetic route and the full characterization of all intermediates, we were confident of the structure of synthetic tetrahydroporphyrin tetraacetate **4**. Nonetheless,



**4**

thorough spectroscopic studies were conducted to confirm this structure. In particular, a $^1H-^1H$ NOESY[6] experiment in $C_6D_6$ allowed us to unambiguously confirm the connectivity of the porphyrin skeleton (Table 1, Figure 1). As we had expected, **4** exhibited a continuous circle of NOEs around the porphyrin ring (Table 1). In addition, strong cross-peaks representing NOEs from the C5 and C15 *meso* protons to

Table 1. Data on NOEs for tetrahydroporphyrin tetraacetate **4**.

| | | |
|---|---|---|
| H2 ↔ $CH_3$(C3) | H8 ↔ H10 | H15 ↔ $CH_3$(C17) |
| $CH_3$(C3) ↔ H5 | H10 ↔ $CH_3$(C12) | $CH_3$(C17) ↔ H18 |
| H5 ↔ $CH_3$(C7) | $CH_3$(C12) ↔ H13 | H18 ↔ H20 |
| $CH_3$(C7) ↔ H8 | H13 ↔ H15 | H20 ↔ H2 |

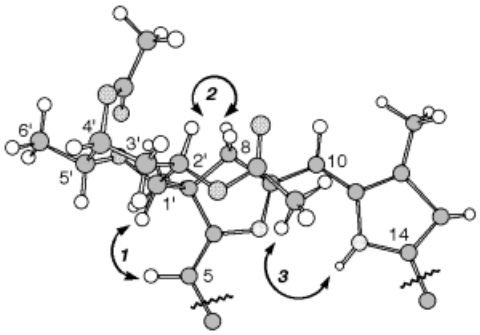


Figure 1. The partial structure of **4**. The NOEs are represented by two-ended arrows: ***1***: H1′ ↔ H5, ***2***: H2′ ↔ H8, ***3***: H2′Ac ↔ HN.

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the carbohydrate C1′ protons, as well as NOEs from the C8 and C18 protons to the carbohydrate C2′ protons, were observed; NOEs were also seen between the protons of the C2′ acetyl methyl groups of the carbohydrate moieties and the porphyrin NH protons (Figure 1). These NOEs confirmed the configuration at the quaternary centers C7 and C17 and also suggested restricted rotation around the C-glycosidic bonds.

The tetrahydroporphyrin tetraacetate **4** was then converted into synthetic *O*,*O*-diacetate **1b**. The structure of **1b** was fully consistent with its spectroscopic data, including the $^1H-^1H$ NOESY data collected in $CD_3COCD_3$ (Table 2). Because the

Table 2. Data on NOEs for synthetic tolyporphin A *O*,*O*-diacetate (**1b**).

| | |
|---|---|
| H20 ↔ H2 | H10 ↔ $CH_3$(C12) |
| H2 ↔ $CH_3$(C3) | $CH_3$(C12) ↔ H13 |
| $CH_3$(C3) ↔ H5 | H13 ↔ H15 |
| H5 ↔ $CH_3$(C7) | H15 ↔ $CH_3$(C17) |

C8 and C18 methylene groups were oxidized to the corresponding ketones, the complete NOE circle observed for **4** was discontinued at C8 and C18 in **1b**, resulting in the two NOE sequences listed in Table 2. The other significant difference in NOEs observed for **1b** and **4** was the NOE pattern detected between the carbohydrate moieties and the porphyrin ring; the C5 and C15 meso protons of **1b** exhibited NOEs to both the carbohydrate C1′ and C2′ protons, whereas the same *meso* protons of **4** exhibited NOEs only to the C1′ protons. These observations suggest that, unlike **4**, **1b** exists as a mixture of rotamers (**1b-A** and **1b-B**, Figure 2) with rotation

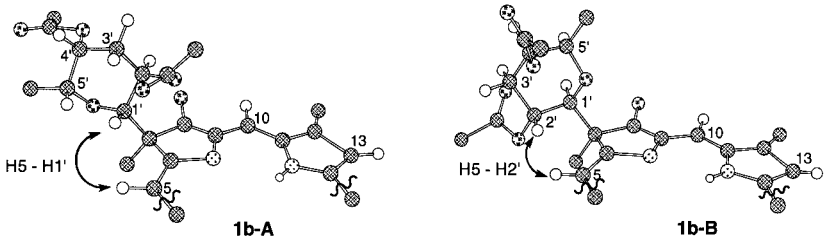


Figure 2. Partial structures of **1b-A** and **1b-B**. The NOEs are represented by two-ended arrows.

around the C-glycosidic bonds due to a lower energy barrier. The difference in the conformational flexibility between **1b** and **4** could be attributed to the difference in the steric effects between the C8 and C18 methylene groups in **4** and the C8 and C18 ketone moieties in **1b**. No NOEs from the carbohydrate C2′ acetate groups to the porphyrin ring were observed.

Based on the evidence presented, there is no doubt concerning the structure of synthetic **1b**. However, the $^1H$ NMR spectrum (Figure 3)[7] of this product does not match that of the *O*,*O*-diacetate prepared from natural (+)-tolyporphin A.[2] We therefore set out to elucidate the structure of (+)-tolyporphin A.

We collected the $^1H-^1H$ NOESY data for natural (+)-tolyporphin A *O*,*O*-diacetate in $CD_3COCD_3$ and compared that with the data for the synthetic compound. The connectivity of the porphyrin skeleton (Table 3) was found to be identical to the connectivity of the synthetic material. As in the synthetic product, natural tolyporphin A *O*,*O*-diacetate

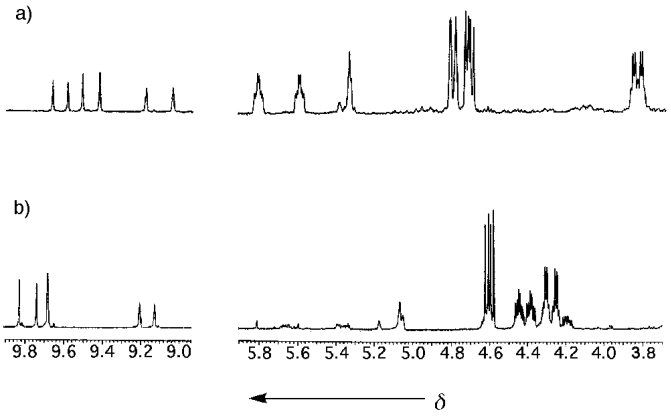


Figure 3. $^1H$ NMR spectra (600 MHz, $CD_3COCD_3$) of a) synthetic and b) natural tolyporphin A *O*,*O*-diacetates. The spectra cover the regions of carbohydrate ($\delta = 3.7-5.9$) and porphyrin ring protons ($\delta = 8.9-9.9$).

Table 3. Data on NOEs for natural tolyporphin A *O*,*O*-diacetate.

| | |
|---|---|
| H20 ↔ H2 | H10 ↔ $CH_3$(C12) |
| H2 ↔ $CH_3$(C3) | $CH_3$(C12) ↔ H13 |
| $CH_3$(C3) ↔ H5 | H13 ↔ H15 |
| H5 ↔ $CH_3$(C7) | H15 ↔ $CH_3$(C17) |

exhibited NOEs between both the C1′ and C2′ protons on the carbohydrate groups and the C5 and C15 *meso* porphyrin protons. Again, this suggests rotational freedom around the C-glycosidic bonds. These results led us to conclude that the synthetic and natural products must differ only in the configuration at their quaternary centers, and thus there are four possibilities for the structure of natural (+)-tolyporphin A (Figure 4). Structure **A** represents our synthetic product, which does not match the natural product. Under the assumption that rings A and C are derived from the same biosynthetic precursor through the same biosynthetic pathway, it is unlikely that structures **B** or **C** represent the natural product, since the absolute configurations of their C7 and C17 quaternary

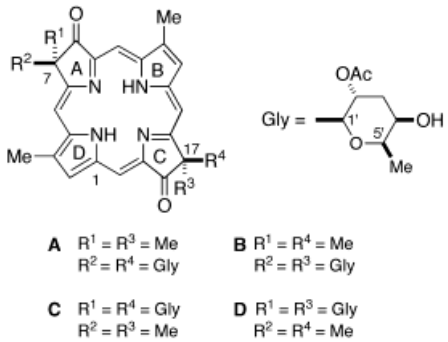


Figure 4. Four possible structures for tolyporphin A, the representative member of the tolyporphin class of natural products.

centers are opposite. Therefore, we propose that the structure of (+)-tolyporphin A is most likely represented by structure **D**. This revised structure is consistent with all the spectroscopic data observed for the natural product and can be

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unambiguously established by the synthesis of **D** starting with the monocyclic precursor **2b**.

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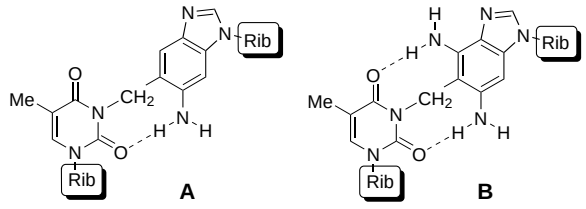
[1] T. G. Minehan, Y. Kishi, *Angew. Chem.* **1999**, *111*, 972; *Angew. Chem. Int. Ed.* **1999**, *38*, 923.
[2] Tolyporphin A *O*,*O*-diacetate was prepared ($Ac_2O$, pyridine, RT) from natural (+)-tolyporphin A.[3]
[3] M. R. Prinsep, F. R. Caplan, R. E. Moore, G. M. L. Patterson, C. D. Smith, *J. Am. Chem. Soc.* **1992**, *114*, 385.
[4] The numbering according to the proposed structure **1a** is adopted here.
[5] Eight additional tolyporphins have been isolated from the cyanophyte microalga: M. R. Prinsep, G. M. L. Patterson, L. K. Larsen, C. D. Smith, *Tetrahedron* **1995**, *51*, 10523.
[6] The $^1H-^1H$ NOESY and $^1H-^1H$ ROESY data were identical for each of the individual compounds studied. We chose to focus on the $^1H-^1H$ NOESY data because it displayed a better signal-to-noise ratio.
[7] The $^1H$ NMR spectra for the synthetic and natural tolyporphin A *O*,*O*-diacetates were shown to be concentration-independent in $C_6D_6$.

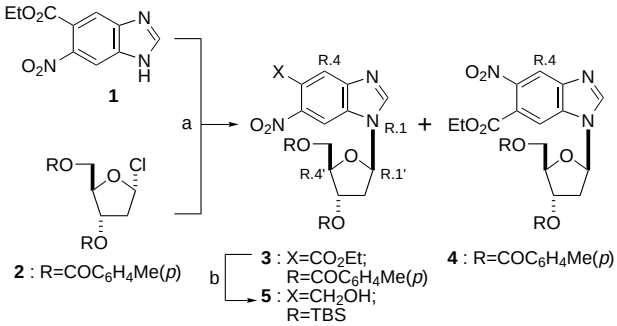
## Covalently Cross-Linked Watson – Crick Base Pair Models**

Xiaoxin Qiao and Yoshito Kishi*

The concept of covalently linked cross sections with molecular architecture similar to Watson – Crick hydrogen-bonded base pairs was introduced by Leonard in the mid-1980s.[1] Since then, several types of covalently linked systems have been developed and used for a variety of purposes.[2] In our view, base pairs linked with a methylene bridge such as **A** and **B** may add some new aspects to the chemistry of covalently linked nucleosides/nucleotides. These include: 1) only Watson – Crick and reversed Watson – Crick base pairings are possible for these models,[15] and 2) conformational flexibility is expected to be retained along the bridging methylene bonds, in addition to their expected increased chemical stability. Evidently, there is concern about such models; introduction of the $CH_2$ bridge would not allow the two bases to be in the same plane, and consequently models such as **A** and **B** might not properly mimic the Watson – Crick base pairs. However, molecular mechanics calculations have suggested that the deformation caused by the introduction of the methylene bridge to a double-stranded oligomer may be insignificant.[3] Herein we report a general synthetic route to the type **A** base pairs and the elucidation of their structures in both solid and solution states, thereby demonstrating that they indeed possess (some of) the structural characteristics of Watson – Crick hydrogen-bonded base pairs.




Ethyl 6-nitro-5-benzimidazole carboxylate (**1**; m.p. 154 – 157 °C (EtOH)) was readily prepared from commercially available 5-benzimidazole carboxylic acid in two steps (1. EtOH/cat. $H_2SO_4$/reflux and 2. fuming $HNO_3/H_2SO_4$/50 °C) in 85 % overall yield. Under phase-transfer conditions,[4] **1** was coupled with 2-deoxy-3,5-di-*O*-*p*-toluoyl-D-*erythro*-pentosyl chloride (**2**)[5] to give a 1:1 mixture of N1- and N3-glycosides **3** and **4**, which were readily separated by silica gel column chromatography ($CH_2Cl_2 \rightarrow CH_2Cl_2$/EtOAc (4/1)) in 95 % combined yield (Scheme 1). A better than 20:1




Scheme 1. Reagents and conditions. a) KOH/[18]crown-6/MeCN/RT; b) 1. $K_2CO_3$/EtOH/RT; 2. TBS-Cl/imidazole/DMF/RT; 3. NaOH/*t*BuOH/RT; 4. $ClCO_2Et/Et_3N$/THF/0 °C; 5. $NaBH_4$/EtOH/0 °C. TBS = *tert*-butyldimethylsilyl.

$\beta$ selectivity was observed for the N1- and N3-glycosides. The structures of **3** and **4** were established by NOESY experiments. In **3**, the R.1′-H exhibited strong crosspeaks with R.4′-H, R.2-H, and the proton *ortho* to the nitro group (R.7-H), respectively. In **4**, the R.1′-H showed strong crosspeaks with R.4′-H, R.2-H, and the proton *ortho* to the carboethoxy group (R.7-H), respectively.[6]

The N1-glycoside **3** was converted to the primary alcohol **5** in five steps in 55 % overall yield (Scheme 1). After mesylation, **5** was coupled with 3′,5′-di-*O*-(*tert*-butyldimethyl)silylthymidine (**6**)[7] to give the $CH_2$-linked product **7** in nearly quantitative yield (Scheme 2). Alternatively, the coupling of **5** with **6** to yield **7** was achieved in one step in 96 % yield under the Mitsunobu conditions.[8] Reduction of the nitro group of **7**,

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