Synthesis of Lipid I and Lipid II monophosphate analogues of Moenomycin for inhibition of transglycosylases.

Moenomycin A is a natural product which interrupts cell wall biosynthesis by binding to the bacterial transglycosylases (TGases), causing an accumulation of cell wall intermediates and leading to lysis and cell death. It is one of the most potent transglycosylase inhibitor known to date, but its absorption upon oral administration is relatively poor due to the C25 isoprenoid chain. Removal of this unit completely abolishes its biological activity. Owing to these limitations, intensive synthetic efforts have focused in designing and developing new moenomycin type lead compounds as novel antibiotics.¹

In the light of this and looking at the structural features of the Lipid-II and Moenomycin, both of which interacts actively with transglycosylase, we planned to synthesize hybrid molecules by taking the structural features from both Lipid II and Moenomycin (compound A, B, and C, see Fig. 1).

The synthetic strategy for C-2-hydroxymethyl analogs (A, B, C) involves,

1) The synthesis of a donor fragment and a C₂-hydroxymethyl analog of muramic acid as acceptor fragment (see Fig. 1). The glycosylation reaction between donor and acceptor will give a disaccharide, that upon phosphorylation will give the lipid II precursor (B). Lipid I analogue (A) can be synthesized from one of the intermediate in the synthesis of the acceptor.

2) Alkylation of the phosphates with different alkyl chains or isoprenoid chains (<C20) and glycerate lipid chains (pyrophosphate mimic)

3) Incorporation of suitable peptides

A similar protocol can be used to prepare Lipid II analogue (C) where the C₂-hydroxymethyl group is coupled with a phosphate to form a stable cyclic phosphate.

Fig 1. Design and development of new Moenomycin type lead compounds as novel antibiotic
The convergent approach for the synthesis of C₂-hydroxymethyl analogs (C) involves the synthesis of the donor fragment (as shown in scheme 1) and the synthesis of a C₂-hydroxymethyl acceptor part (as shown in scheme 2).

**Synthesis of donor** (Scheme 1)

The donor fragment was synthesized from d-glucosamine hydrochloride.² The amine functionality was protected with Δ-Troc to obtain 3. Compound 3 was exhaustively acetylated to obtain tetraacetate 4.

Treatment of compound 4 with hydrazine acetate resulted in regioselective hydrolysis of the anomeric O-acetyl group to obtain compound 5. The anomeric position in compound 5 was activated for nucleophilic attack, by treating 5 with trichloroacetonitrile to obtain the donor part (6).

**Synthesis of C₂ hydroxymethylene analog of acceptor** (Scheme 2)

The synthesis of C₂ hydroxymethylene analog of acceptor was envisioned starting from d-Mannose. The anomeric hydroxyl group of d-mannose was protected with a benzyl group to obtain benzyl glycoside 8. Compound 8 was treated with benzaldehyde dimethyl acetal for the protection of the 1,3-dihydroxy functionality to obtain compound 9. The C-3 hydroxy group in compound 9 was regioselectively protected by reaction with tributyl tin oxide followed by treatment with p-methoxy benzyl chloride to obtain 10 in good yield. Compound 30 was subjected to Swern oxidation giving the unstable ketone 11 which was immediately subjected to Wittig reaction to obtain exocyclic olefin 12. This olefin was subjected to hydroboration to obtain C-2 hydroxymethylene compound 13 as a 1:1 diastereomeric mixture. Since we only needed compound with an equatorial hydroxymethyl group (2R-13), this mixture was subjected to Swern oxidation followed by epimerization of 2S to 2R in presence of triethyl amine to afford 2R-13.
Our initial synthetic efforts for the synthesis of C-2 hydroxymethylene analogue of acceptor, involved multistep transformations of compound 10 to compound (2R-13) with an equatorial hydroxymethyl group\(^3\) (Scheme 2).

However, this strategy was cumbersome, five steps from 11 to 2R-13, thus we envisaged an alternative pathway to achieve the target compound 13 from compound 11, is shown in Scheme 3. One carbon homologation at C-2 position of compound 11 was achieved with lithium chlorocarbenoid\(^4,5\) generated from dichloromethane and LDA (lithium diisopropylamide), via stereoselective introduction of dichloromethyl function to compound 11 to obtain benzyl 4,6-O-benzylidene-2-C-dichloromethyl-\(\alpha\)-D-glucopyranoside (12). Hydride reduction of 12 with NaBH\(_4\) (sodium borohydride) in DMSO at 80 °C gave benxyl 4,6-O-benzylidene-2-C-hydroxymethyl-\(\alpha\)-D-glucopyranoside (13) in 65 % yield over two steps.
The compound 13 was subjected to benzyl protection followed by deprotection of the PMP group in presence of DDQ to afforded 16 in 67 % yield over two steps. O-alkylation of 16 with 1,2-chloropropionic acid afforded compound 17 in 80% yield. Further the carboxylic acid moiety of 17 will be protected with 2-(phenylsulfonyl)ethyl ether subsequent treatment with trifile acid in presence of triethylsilane will afford the C-2 hydroxymethylene acceptor 18.

For the synthesis of phosphoglycerates, we have developed a protocol as show in Scheme 4. This protocol was used to scale up C16 phosphoglycerate chains. The synthesis begins from d-mannitol, that serves as a chiral pool for the glycerate moiety and was used to obtain acetonide protected alcohol 19 on multigram scale. The (S)-acetonide-protected glycerol 19 was subjected to p-methoxybenzyl (PMB) protection as it can be selectively removed under neutral conditions with 2,3-dichloro-5,6-dicyanobenzoquinone (DDQ). Acetonide deprotection and regioselective protection of the primary hydroxyl group with TBDPS gave the key intermediate 20a. This key intermediate 20a was cetylated by treatment with sodium hydride and cetyl bromide to afford 21 in 87 % yield. The PMB group was carefully removed with DDQ to afford required product in 84% yield. This PMB deprotected compound was subsequently oxidized, methylated and TBDPS deprotection afforded the desired glycerate cetyl chain (23) in 47 % yield over four steps. The glycerate dodecyl compound (31) will be prepared in the same way.

Scheme 4

Conclusions:

- Synthesis of C-2 methylene analogue of acceptor is near completion with the shorter synthetic pathway (Scheme 3)
- Synthesized C16 phosphoglycerate chain for the phosphorylation of C-2 methylene analogue of acceptor.
References