Designing new nucleoside-based butyrylcholinesterase inhibitors

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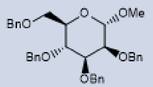
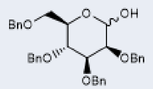
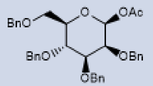
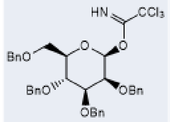
Optimization of N-glycosylation conditions

- ✓ Since the most active compound was one of the minor compounds, an attempt of optimize the *N*-glycosylation reaction conditions was made.
- ✓ The methyl group present at the anomeric position is a poor leaving group: Several glycosyl donors were tried, namely with the anomeric free hydroxy group, or protect with an acetyl group or a glycosyl trichloroacetimidate.
- ✓ Another Lewis acid ($BF_3{\cdot}Et_2O$) was also tried to access the outcome of the *N*-glycosylation reaction.
- ✓ These changes did not give a positive change in regard of the α-N^7 isomer yield.
- ✓ Using the methylated compound and $BF_3{\cdot}Et_2O$ as Lewis acid, there was a change in the ratio N^7/N^9 in favor of the N^9 isomers. With the free hydroxy group no reaction was observed in any condition. The acetyl protecting group gave the same results as the methyl group. Trichloroacetimidate favored the N^9 isomers with both activators.
- ✓ Since none of the changes produced a positive effect in the α-N^7 isomer yield, other improvements were attempted.
- ✓ Runing the reaction with TMSOTf at different temperatures did produce some changes.
- ✓ If the temperature was increased (to 90ºC), reaction time decreased but it gave almost a 1:1 ratio on all four isomers.
- ✓ Decreasing the reaction temperature to 40ºC, reaction time increases to 8 hours and there is a positive change in N^7/N^9 ratio.
- ✓ When the reaction was run at room temperature, the N^7/N^9 ratio was increased in favor of the N^7 isomers with almost 1:1 for the α/β ratio.

Table 1 – Yields of the *N*-glycosylation reaction, with a different leaving group and activator

	Yield (TMSOTf)					Yield ($BF_3{\cdot}Et_2O$)				
	αN⁹	βN⁹	αN⁷	βN⁷	Total	αN⁹	βN⁹	αN⁷	βN⁷	Total
	24%	3%	8%	28%	63%	19%	23%	3%	18%	63%
	N⁹ 27%		N⁷ 36%			N⁹ 42%		N⁷ 21%		
	No reaction					No reaction				
	10%	15%	4%	33%	62%	29%	10%	9%	11%	59%
	N⁹ 25%		N⁷ 37%			N⁹ 39%		N⁷ 20%		
	19%	18%	4%	23%	64%	21%	17%	2%	20%	60%
	N⁹ 37%		N⁷ 27%			N⁹ 38%		N⁷ 22%		

Introduction of methyl groups

- ✓ A series of analogues was synthesized with the objective to decrease the number of benzyl groups, decreasing overall molecular weight.
- ✓ Interestingly, there was a pattern in the results obtained. The introduction of the methyl groups at positions 4, 6 or 4 and 6 produced only a slight decrease in the formation of β-N^7 isomer in favor of the β-N^9. On the other hand, when there were methyl groups present in positions 2 and 3, there was a significant decrease in the formation of the β-N^7 isomer in favor of the β-N^9.

Deoxygenation of key positions

- ✓ Several deoxygenations were made at different positions of the sugar moiety, to access the impact on biologcal activity.
- ✓ Deoxygenation at position 6**,** position 4 and positions 3 and 4 were made after a few regioselective protection steps, with a good overall yield.
- ✓ All deoxygenated compounds were submitted to the optimized *N*-glycosylation conditions to afford deoxynucleosides, under biological evaluation.

Table 2 – Yields of the glycosylation reaction at different temperatures

T (ºC)	Yield (TMSOTf)				
	αN⁹	βN⁹	αN⁷	βN⁷	Total
65°C	24%	3%	8%	28%	63%
	N⁹ 27%		N⁷ 36%		
25°C	17%	3%	25%	27%	72%
	N⁹ 20%		N⁷ 52%		
40°C	16%	4%	22%	23%	65%
	N⁹ 20%		N⁷ 45%		
90°C	15%	10%	11%	14%	50%
	N⁹ 25%		N⁷ 25%		

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