Complete separation of the quaternary mixture of nadolol stereoisomers using preparative and simulated moving bed chromatography

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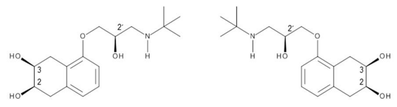
INTRODUCTION

The separation and purification of high added value products by liquid chromatography is a very popular technique. The development of more stable and efficient stationary phases, together with the design of innovative and more flexible separation processes, enhanced the use of chromatographic processes, particularly at preparative and industrial scales through fixed-bed and simulated moving bed (SMB) technologies. Fixed-bed and SMB techniques are more and more used in the separation of a wide range of products for the pharmaceutical, fine chemistry, biotechnology and food industries. In this context, one of the actual main challenges concerns the design and optimization of these chromatographic processes for challenging multicomponent separations. This includes the development of new and innovative chromatographic processes, combining different design strategies and modes of operation, with different types of stationary and mobile phases.

The design and optimization of a chiral separation process for a specific chiral binary or pseudo-binary mixture is based on a careful selection of the proper combination between the chiral stationary phase and the mobile phase composition. When considering multicomponent separations, the complexity deeply increases by the introducing of multi-step separation sequences (or a much more complex multi-region separation process). This can be done by opening the possibility to combine chiral and achiral stationary phases (when in presence of stereoisomers instead of just one pair of enantiomers) and to combine different separation techniques such as the fixed-bed and SMB related processes.

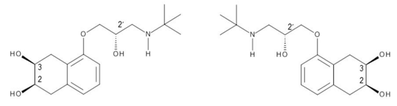
Nadolol is a nonselective beta-adrenergic receptor antagonist (β-blocker) pharmaceutical drug, widely used in the treatment of cardiovascular diseases, such as hypertension, ischemic heart disease (angina pectoris), congestive heart failure, and certain arrhythmias. Its chemical structure has three stereogenic centers which allows for eight possible stereoisomers. However, the two hydroxyl substituents on the cyclohexane ring are fixed in the cis-configuration, which precludes four stereoisomers; in fact, two pairs of enantiomers. Nadolol is presently marketed as an equal mixture of the four stereoisomers, designated as the diastereomers “racemate A” and “racemate B” . Racemate A is a mixture of stereoisomers 2 and 3 and racemate B is a mixture of stereoisomers 1 and 4.

Racemate A



2R3S2´R - Nadolol 2S3R2´S - Nadolol

Racemate B

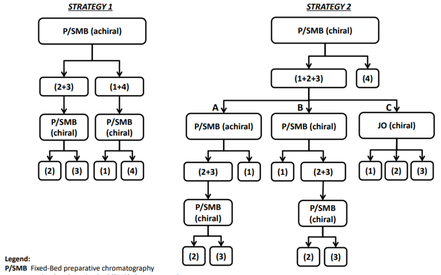


2R3S2´S - Nadolol 2S3R2´R - Nadolol

Since it is composed by four stereoisomers, being two pairs of enantiomers. In this way, it introduces the possibility of alternative strategies, using different kind of preparative separation sequences and techniques, and also the use of different packings (chiral and achiral stationary phases), and the corresponding mobile phase optimization at both normal and reversed-phase modes.

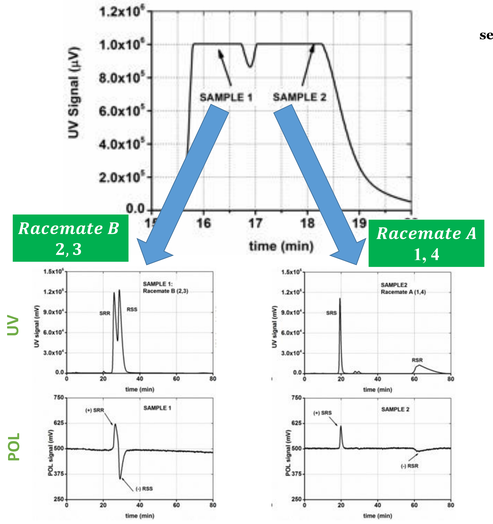
OBJECTIVES

The design of the complete preparative separation of nadolol stereoisomers asks for a global experimental and simulation methodology considering both the characterization and optimization of each separation step and its sequences to achieve the four nadolol components pure. New strategies using combinations of achiral and chiral stationary phases and sequences of different separation techniques will be presented. Extensive experimental and simulation results for the complete separation, using fixed-bed (Azura preparative HPLC) and SMB (LSRE-FlexSMB) pilot units, of all the four nadolol stereoisomers using Chiralpak IA (chiral) and different Waters C18 (achiral) stationary phases will be presented.

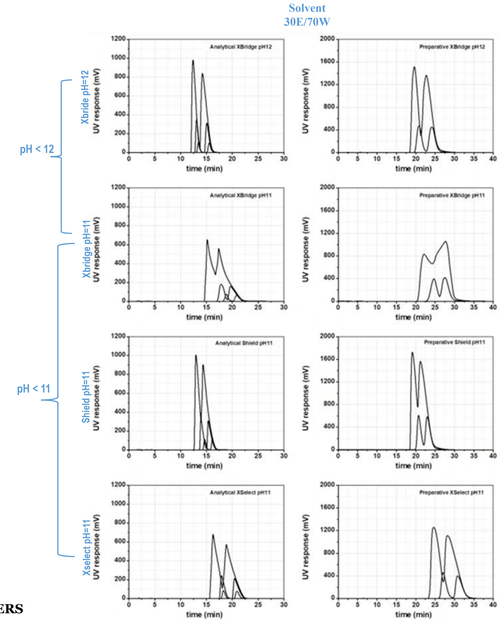


RESULTS AND DISCUSSION

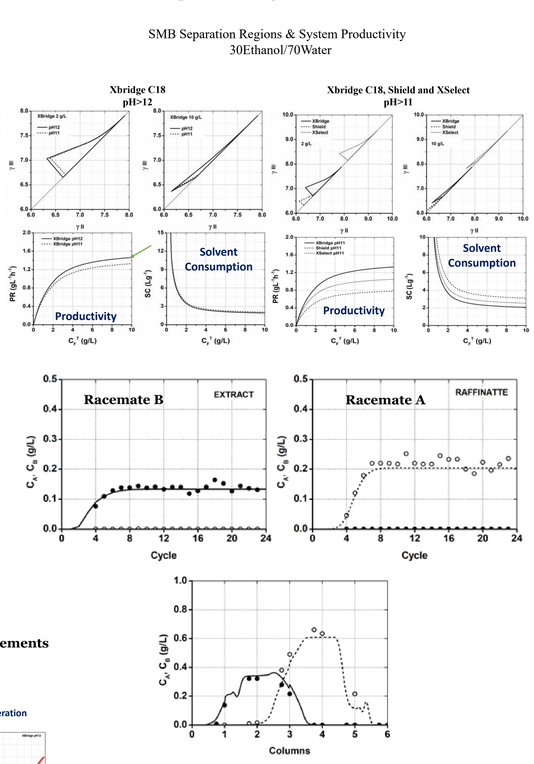
IDENTIFICATION OF ALL THE FOUR NADOLOL STEREOISOMERS



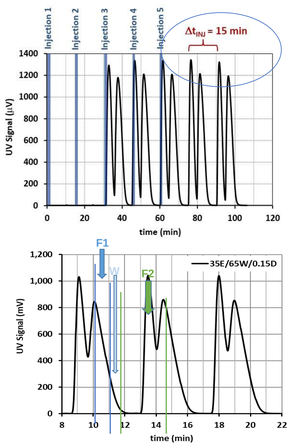
Screening of the achiral adsorbent



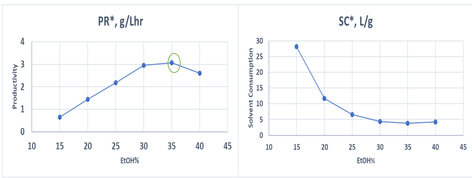
Previsions for SMB operation using the different achiral adsorbents



Sequence of five injections

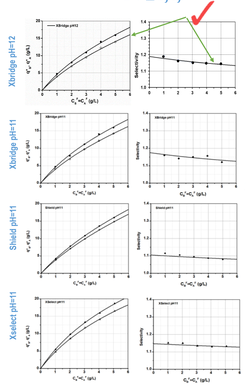


Main Performance Parameters

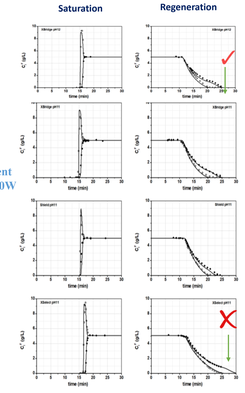


Equilibrium adsorption isotherm and selectivity measurements and model fitting

$$q_i^* = mC_i + \frac{Qb_iC_i}{1+\sum b_jC_j}$$



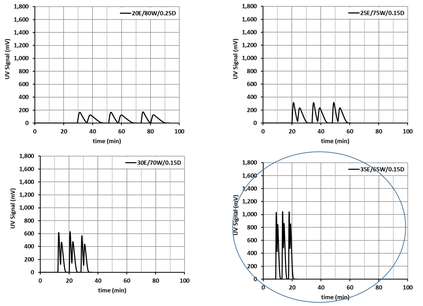
Breakthroughs measurements and simulation



PREPARATIVE HPLC SEPARATION OF NADOLOL

Most Promising Solvents to Perform Enantiomer Separation of Nadolol

pH < 12 ΔP ≈ 150 bar $C_F^T = 10\ g/L$



CONCLUSION

- Strategies for complete separation of nadolol stereoisomers were well defined.
- Reversed phase mode can be successfully applied for the separation of nadolol racemates.
- The selected column to perform enantiomer separation of nadolol was XBridge C18 column using 30%ethanol/70%water/0.1DEA mobile phase composition.
- Real simulated moving bed separation was carried out using 2 g/L of nadolol, Purities for both extract and raffinate outlet streams was 100%.
- Alternatively, 35E/65E/0.15DEA mobile phase composition was selected to perform (2+3/1+4) enantiomer separation of nadolol using fixed bed technic.
- Real enantiomer separation of nadolol was carried out using 10 g/L of nadolol, Purities for both collected fractions were 100%.

Acknowledgements

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