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## Introduction

A key advantage of SFC is minimal solvent waste, which is particularly important in preparative SFC, leading to fast sample recovery. Hence it is important to explore whether SFC, which also promises to be cheaper and more environmentally friendly than conventional HPLC, can be applied more widely as a complimentary method.

The objective is to develop a generic method focussing on speed by using smaller 50mm columns. Compounds with certain common substructures show peak splitting on the 2-ethylpyridine column. An approach to investigate the problem and possible solutions are presented.

## Peak Splitting: artefact or reality ?

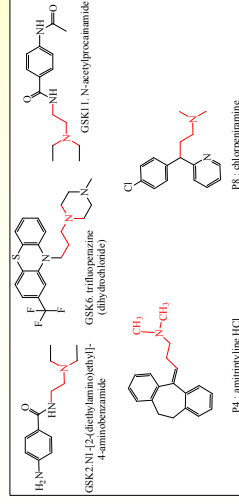


Figure 1: Structures of compounds showing peak splitting

- > Isomers / stereoisomer
  - > In source CID–MS, NMR
- > Attempt to separate peaks (Figure 2)
- > Peaks due to solvation
  - > MeOH / DMSO (Figure 3)
- > Choice of solvents for
  - > sample preparation (Figure 3)
  - > Modifier (Figure 3)



## Difference in mobile phase mixing

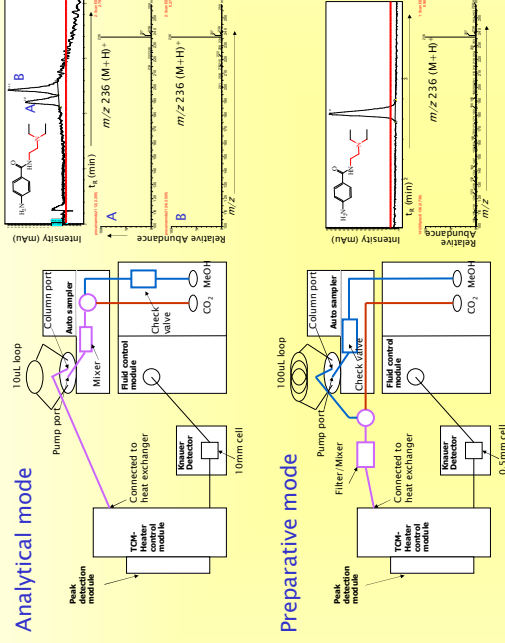
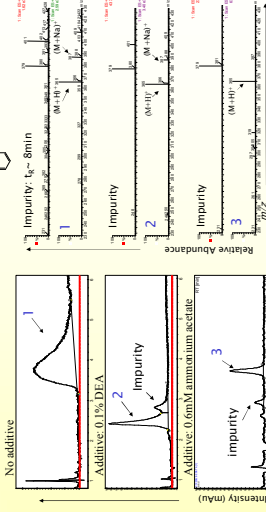


Figure 2: Schematic of instrument in analytical and preparative mode showing difference in the mixing of mobile phase

## Improved peak symmetry and separation using ammonium acetate additive

250mm Cyano column,  
 25% MeOH with/without additive,  
 Flow : 4mL/min



- Figure 4: Structures of compounds showing peak splitting
- > Impurity co-eluting with DEA as additive
  - > Peak symmetry improved using ammonium acetate

## Conclusions

- ✓ Peak splitting ~Biphasic system  
 ~Methanol\DMSO solvated molecule
- ✓ DCM solvent ~Improved peak shape for this class of compounds
- ✓ Ammonium acetate ~improved peak symmetry and separation.

## References

1. Pinkston, J. D.; Stanton, D. T. Wen, D.; *Journal of Separation Science*, 2004, 27, 115–123.
2. Zheng, J.; Taylor, L. T.; Pinkston, J. D.; Mangels, M. L. *Journal of Chromatography A*, 2005, 1082, 220–229.

## Biphasic system or solvated molecule ?

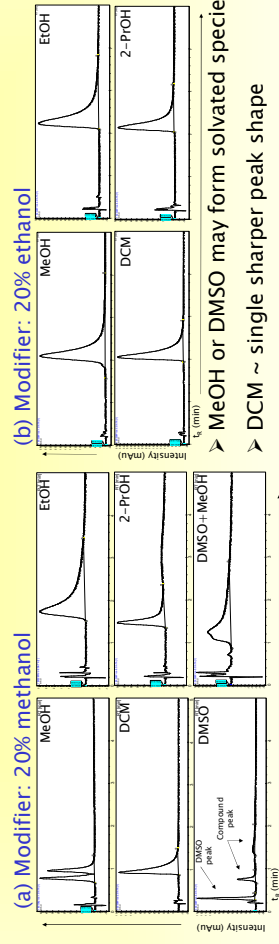


Figure 3: Chromatograms from changing the solvent for preparing samples (a) 20% MeOH modifier, (b) 20% EtOH modifier

## Acknowledgements

