Selectivity versus Efficiency in Ultra High Pressure Liquid Chromatography

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Introduction

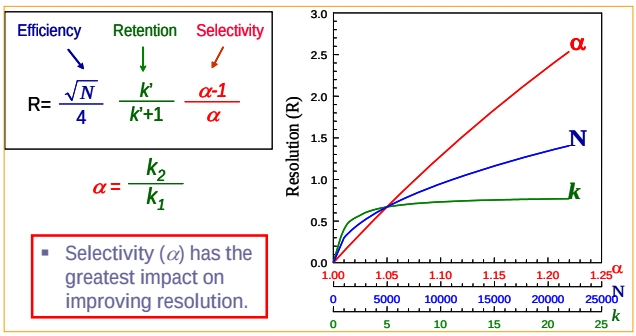
The current trend towards using high pressure in LC is well documented, high efficiencies, good resolution and fast throughput being the goal that has driven the move towards the use of sub 2um particles.

In previous work we have shown that for short fast gradients well packed 3um Fortis columns can provide equivalent, if not more, peak capacity than other commercial sub 2um columns. In this poster we show that for those analysts already working with ultra high pressure LC systems and 2um particle columns that it is important to consider the role of stationary phase selectivity when trying to maximise resolution and not rely on efficiency alone.

Improving Resolution

Approaches to improving resolution involve making changes to one or more of three variables; efficiency, retention and selectivity. The move towards using sub 2um particles has been driven by the theory that the resulting jump in efficiency will lead to significant improvements in resolution. As can be seen in figure 1 efficiency (N) does play a significant part in improving resolution, however by far the greatest factor is column selectivity.

FIGURE 1. Contributions to Resolution

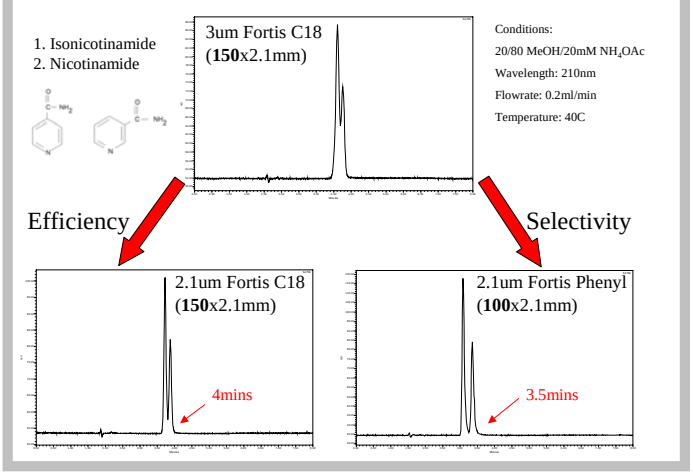


With new small particle columns being released by column manufacturers it is important that a range of phase chemistries are offered to allow the analyst an opportunity to maximise resolution rather than depending on efficiency alone. Also it should not be assumed that all commercial C18 products on the market have the same selectivity, therefore as well as evaluating new phase chemistries it might be wise to test some alternative manufacturers C18's.

Resolution Gain from Alternative Selectivity

Figure 2 shows an example of where increasing efficiency by moving from 3um C18 to 2.1um C18 does not provide the full resolution of two closely related compounds. However by altering selectivity, using Fortis Phenyl™ chemistry, at the same time as decreasing particle size we are able to obtain resolution whilst decreasing column length and as a result analysis time.

FIGURE 2. Positional Isomers - Resolution of Nicotinamides



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Fortis Technologies Ltd recognises the trademarks of all other manufacturers
All columns are original manufacturers packed columns

Speed Increase from Alternative Selectivity

Figures 3a+3b show how the use of an alternative phase chemistry such as Phenyl can significantly reduce analysis time of a set of compounds whilst maintaining resolution of closely eluting peaks. Figure 3c shows how the analysis time can be further reduced by the use of 2.1um particles to increase efficiency.

FIGURE 3. Positional Isomers – Speed of Analysis

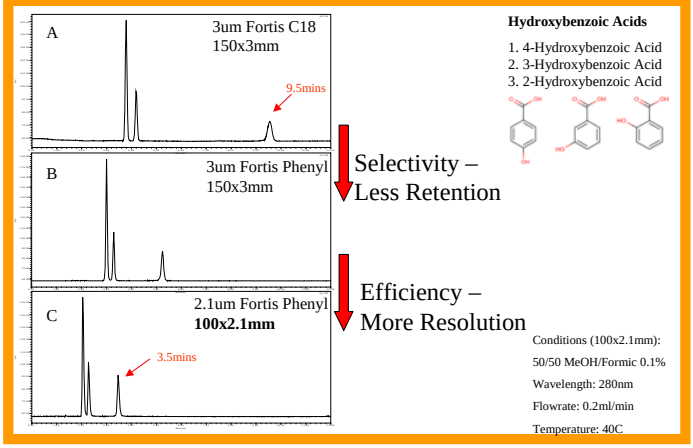
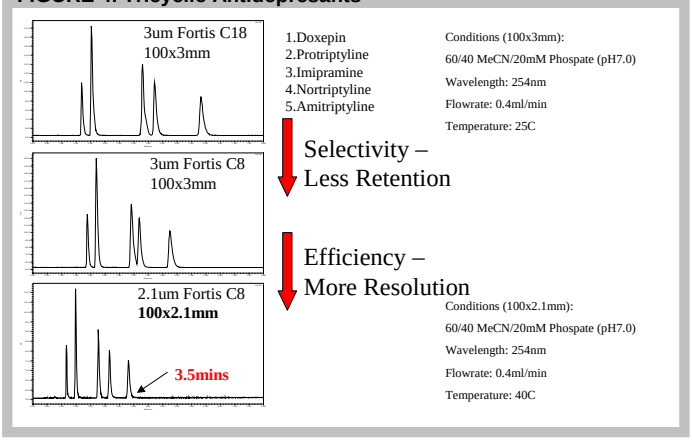
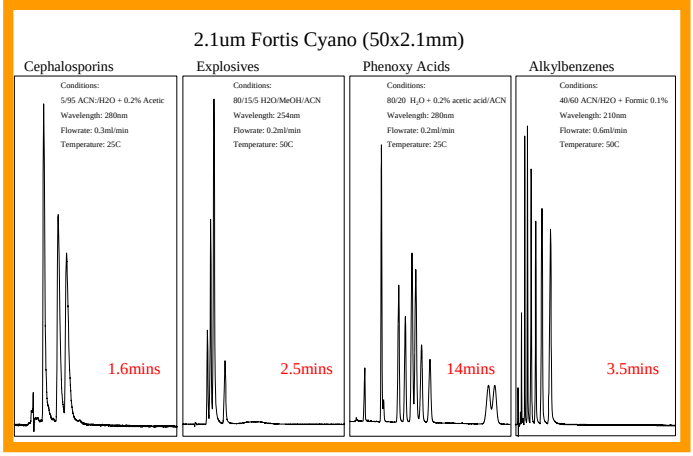


FIGURE 4. Tricyclic Antidepresants



In figure 4 we can see how the application of alternative phase chemistry can speed up analysis times. Figure 5 shows the application of 2.1um Fortis Cyano™ which can be used in both reverse and normal phase.

FIGURE 5. Selectivity for Reverse and Normal Phase



Conclusion

The use of small particles in Ultra High Pressure LC can provide the analyst with increased sensitivity and resolution. We have shown that an important consideration when trying to maximise resolution of closely eluting compounds is the role of selectivity. The application of alternative chemistries based on 2.1um particles can provide greater increases in resolution than the application of small particles alone.