

Clinton W. Amoss and Bruce Coryell
CHIRAL TECHNOLOGIES INC,

A variety of sample diluents can be used with CHIRALPAK® IA columns. The long term effects of dimethylsulfoxide injections have been studied, and effective regeneration sequences have been demonstrated.

Sample solubility may be a key consideration in chiral HPLC separations. With the recent introduction of an immobilized chiral polysaccharide-based stationary phase, the CHIRALPAK IA column, a wider variety of sample solvents can now be used with a greater likelihood that sufficient sample solubility can be realized.

Studies have confirmed that chloroform, tetrahydrofuran, ethyl acetate, dichloromethane, methyl-t-butyl ether, and acetone can be safely and effectively used as mobile phases and sample diluents with the CHIRALPAK IA column. Due to its ability to solvate proteins and other biologically derived samples, dimethylsulfoxide (DMSO) is a particularly desirable solvent for some samples. Experiments were performed to determine the long term effects of repeated sample injections of DMSO on a CHIRALPAK IA column. Two hundred 5 ul injections (1 mg/ml in DMSO) of Wieland-Miescher ketone (WMK) were made on a CHIRALPAK IA column. Figure 1 shows the initial injection of WMK and Figure 2 shows the 200th injection of the same compound. It can be clearly seen that selectivity is unchanged for the two injections, but column efficiency has declined slightly. This loss in efficiency is believed to be caused by a build-up of adsorbed DMSO.

The CHIRALPAK IA column is readily regenerated by flushing with dimethylformamide (DMF). Table I illustrates restoration of column efficiency following a 2 hour DMF flush of the column.

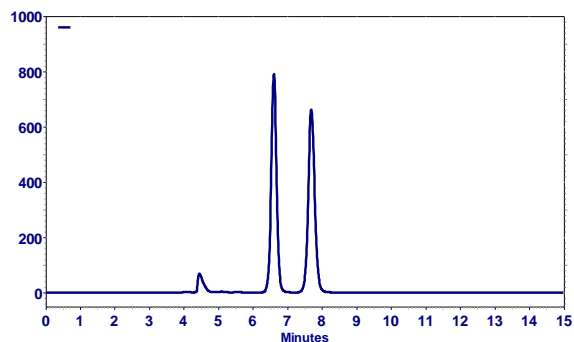


Figure 1. Separation of WMK enantiomers using CHIRALPAK IA (25 cm x 4.6 mm) with 50:50 methanol/ethanol at 0.75 ml/min.

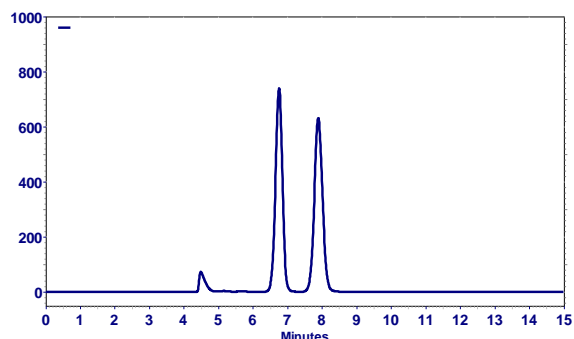


Figure 2. Separation of WMK enantiomers after 200 injections; same conditions as Figure 1.

Table I
Efficiency for WMK Separation
On CHIRALPAK® IA Column

Injection	Peak 1 N	Peak 2 N
Initial	13897	12452
200	7852	7895
Post DMF Wash	13554	12354