In Part I of this study [10] we showed that, with both ion-exchange and ion-pair reversed-phase micro-LC, 1-10 µl samples of acidic organophosphorus analytes could be injected without an appreciable increase in peak broadening, which strongly suggested the occurence of solute focusing. In addition, when we tried to improve the low analyte recovery observed when extracting MPA from soil samples with water, by adding *ca.* 4% hydrochloric acid to the water, we observed that due to the presence of hydrochloric acid in the 10 µl sample only, the retention time of MPA shifted from 10 min to *ca.* 4 min. Simultaneously the peak width in the system (PRP-X100 ion-exchange / 0.5% formic acid in water) became much smaller.

Ion-exchange: influence of acids. For a further study of the above effect, experiments were carried out on the PRP-X100 column using MPA as test solute and adding hydrochloric or nitric acid to the sample solution. As an example, **Figure 2A** illustrates the dramatic influence of increasing amounts of hydrochloric acid in the sample solution on the retention time and peak shape of MPA. The effect of the addition of acid is shown in more detail in **Figure 2B**. Whereas the decrease in retention is directly proportional with the concentration of the acid, efficient peak compression obviously requires the presence of, at least, about 1.5 mg/ml of acid in the sample solution (15 µg injected on the micro-LC column).

When the sample contains several analytes of interest, their resolution is expected to be readily lost due to the great retention shifts. However, a 2-µl injection of a sample containing 70 mg/ml of hydrochloric acid (140 µg injected on the micro-LC column) with MPA and MMP as test solutes still yields sufficient resolution of these analytes, despite the large decrease in retention (**Figure 3**). For DMP, which has a high capacity factor of $k' = 20$ (t_R = 40 min) in the absence of hydrochloric acid, retention time and plate number remained unchanged upon the addition of acid.

The influence of the (retention) characteristics of the acid anion on peak compression was demonstrated by adding nitric acid (4 mg/ml) instead of