

## Intermediate Practical HPLC Course- 2 days

### DAY 1

#### 0900-1100

Silica-based column packing materials: Type A, Type B, Base-deactivated and Monolithic  
Comparison between C8 and C18 columns  
When to use 3  $\mu\text{m}$ , 5 $\mu\text{m}$  and 10  $\mu\text{m}$  particle size packing  
Normal phase columns- Si, CN, Diol,  $\text{NH}_2$   
Polymer-based Reverse phase columns  
Scale down to smaller diameter columns and microbore  
Ion exchange columns- SAX, SCX, WAX, WCX

#### 1115-1300

Guard columns and Precolumns  
Scale up to Prep and semi-prep scale  
Understanding Isocratic Elution: effect of changing flow rate and effect of changing %B  
Understanding Gradient Elution: %B range, Run time, gradient profile, effects of changing column length and gradient dwell volume.

#### 1400-1700

##### **Laboratory Session I**

Internal standard calibration

### DAY 2

#### 0900-1100

Ion chromatography and Ion exchange Chromatography  
UV Detection  
Diode Array Detection  
Fluorescence Detection

#### 1115-1300

Data handling  
Peak Integration  
Quantitation and Calibration

#### 1400-1600

##### **Laboratory Session II**

Running a gradient method: determination of gradient dwell volume  
The analysis of mono- and di-valent anions by ion chromatography

#### 1600-1700

Assessment and Discussion