

## **Processing software: Operation and Parameters**

Raw spectrometric data were firstly translated to netCDF files and then imported to XCMS software. With the XCMS software peaks are detected after applying matched filtration using a second-derivative Gaussian as the model peak shape. Then a peak-matching algorithm that takes into account the two-dimensional, anisotropic nature of LC/MS data is applied. After that the retention times of all samples are corrected simultaneously in a single step and missing peak data can also be filled. MarkerLynx uses ApexTrack peak integration to detect chromatographic peaks, whereas MarkerView utilises the Enhance Algorithm. The ApexTrack algorithm takes the second derivative of a chromatogram and locates inflection points, local minima and peak apex. After apex is found for each peak a retention time is assigned and the data is aligned.

The Enhance algorithm first processes each of the mass spectra of a sample with an order of increasing scan numbers. Masses belonging to the same peak cluster are merged together with a resulting area equal to the sum of all intensities for each mass in the cluster. The ion with the largest intensity is considered the base peak mass of the cluster.

### Applied parameters

1) XCMS: (*version 1.15.0 , 10.2008 release*)

```
findPeaks.matchedFilter(object, fwhm = 30, sigma = fwhm/2.3548, max = 5, snthresh = 10, step = 0.1, steps = 2, mzdifff = 0.8 - step*steps, index = FALSE, sleep = 0)
```

```
group(object, bw = 30, minfrac = 0.5, minsamp = 1, mzwid = 0.25, max = 50, sleep = 0)
```

```
retcor(object, missing = 1, extra = 1, smooth = c("loess", "linear"), span = .2, family = c("symmetric"), plottype = c("mdevden"))
```

```
group(object, bw = 10, minfrac = 0.5, minsamp = 1, mzwid = 0.25, max = 50, sleep = 0)
```

```
fillPeaks(object, method="chrom")
```

2) MarkerView (1.2.0.1)

Peak Intensity Threshold:  $10^4$  counts, chromatographic peak width: 3 - 60 scans minimum spectral peak width: 0.25 amu.

m/z tolerance: 0.25 amu and retention time tolerance : 0.3 min

3) MarkerLynx (v. 4.1)

Peak Width at 5% Height : auto, Peak-to-Peak Baseline Noise: auto,

minimum intensity 100, noise elimination level 6, mass tolerance: 0.25 amu and retention time window 0.3 min.