**Description**

This application attempts to find all cross-linked peptides in the given Ion Mobility MS dataset. First, the FASTA file is read in and possible peptides are generated by digesting the protein(s) and assuming 1 max missed cleavage. The possible peptides are searched for in the given features by matching up the monoisotopic mass of the feature to the mass of the peptides. The mass ppm tolerance is an input parameter of the application. All possible cross-links of the unmodified peptides are then calculated and searched for in the Isotopic Peak data. A detailed csv results file is created at the end.

This software was written by Kevin Crowell, but is now maintained by Matthew Monroe. Alternative contact info is proteomics@pnnl.gov

**Program Usage**

CrossLinkingIMSConsole.exe

-f [Features File]

-p [Peaks File]

-fasta [FASTA file]

-ppm Value

[optional arguments]

\*\*\*\*\*\*\*\*\*REQUIRED ARGUMENTS \*\*\*\*\*\*\*\*\*\*\*

-f Features File. LC-IMS-MS Feature Finder Output

-p Peaks File. DeconTools Output

-fasta FastA File. Contains all protein sequences to search.

-ppm [value] Mass tolerance in ppm

\*\*\*\*\*\*\*\*\*OPTIONAL ARGUMENTS \*\*\*\*\*\*\*\*\*\*\*

-c The maximum number of missed cleavages to consider. Defaults to 1.

-t Set to 'full' for fully tryptic only.

Set to 'partial' to consider partially and fully tryptic.

Set to 'none' to consider non, partially, and fully tryptic.

Defaults to 'full'.

-o The desired location and name for the output file. Defaults to workingDirectory/crossLinkResults.csv

Crosslinked peptide pairs are found using a delta mass value that is

typically computed based on the number of carbons and number of nitrogen atoms,

treating carbons in the crosslinked peptide as C13 and nitrogens as N15.

Alter this behavior using switches -C13off, -N15off, and -Static

-C13off Peptides are not labeled with C13

-N15off Peptides are not labeled with N15

-Static [Value] Static (fixed) mass shift to use for all paired peptides

For example, to disable C13 and N15 shifts and only use a

fixed shift of 25.5 Da, your command line would look like this:

CrossLinkingIMSConsole.exe

-f Dataset\_LCMSFeatures.txt -p Dataset\_Filtered\_isos.csv -fasta Database.fasta

-ppm 10 -C13Off -N15off –Static 25.5

**Features File**

This is a tab-delimited file normally generated by the LC-IMS-MS Feature Finder, which is in-house software developed at Pacific Northwest National Lab. If you need a copy of the software, see http://omics.pnl.gov/software/lc-ims-ms-feature-finder or contact proteomics@pnnl.gov

A normal Features file is created by clustering together isotopic profiles that are believed to be from the same isotopic signature eluting over a period of time. Each feature in the file has representative mass, elution time, drift time and charge state values that define the feature.

To fake a Features file, you can create a tab-delimited text file with the following columns required by the CrossLinkingIMS software:

* Feature\_Index: Integer identifier for the feature. Any numbers can be used, but it is suggested to use sequential number.
* Monoisotopic\_Mass: The monoisotopic mass of the LC-IMS-MS Feature.
* Scan\_Start: The first LC Scan where the Feature was observed.
* Scan\_End: The last LC Scan where the Feature was observed.
* Scan: The LC Scan where the Feature was most abundant.
* IMS\_Scan: IMS Scan of the most abundant LC Scan where the Feature was most abundant.
* Drift\_Time: The detected drift time of the Feature.
* Abundance: The total intensity of the Feature.

**Peaks File**

This is a tab-delimited file that can be exported when running DeconTools. If you need a copy of that software,   
see http://omics.pnl.gov/software/decontools-decon2ls or contact proteomics@pnnl.gov

A peaks file can easily be faked since it is simply a dump of all Isotopic Peaks in the data.

To fake a peaks file, you can create a tab-delimited text file with the following columns required by the CrossLinkingIMS software:

* frame\_num: The LC Scan of the peak.
* scan\_num: The IMS Scan of the peak.
* mz: The m/z value of the peak.
* intensity: The intensity of the peak.