Product Specification:

Progenesis™ LC-MS v2.5

Data Import

- Supports the following data formats:
  - .mzXML including zlib compression
  - NetCDF
  - Thermo .RAW
  - Waters .RAW
  - Profile or centroid (excluding NetCDF)
  - Output from low resolution ion trap instruments
  - Output from FTMS (low noise data)
- Import, analyse and visualise single LC-MS runs
- Add LC-MS/MS runs from multiple folders
- Ability to add additional LC-MS/MS runs to an existing analysed experiment
- Automatic check performed to ensure data files are in the correct format for analysis
- Manually exclude any areas within each run from alignment and detection processes
- Intelligent peak-modelling algorithm to remove background noise and reduce data file size. Peaks are identified and peak models created that retain all relevant quantitation and positional information

Run Alignment

- Displays a 2D image view (“run image”) of your LC-MS runs
- Scan level correction of retention time variation using a unique alignment algorithm
- Automatic placement of alignment vectors
- Assisted placement of manual vectors

Image Display

- Preset image enhancement options to access a wide range of image intensity levels
- Automated contrast stretching of a run image

Alignment visualisation

- 4 simple integrated and interactive run image views
  - Zoomed alignment view
  - Alpha blend view with auto zoom
  - Overlaid view of two run images
  - Total Ion Chromatogram (TIC) view
- Select the size of area within a run image to focus on for visualising alignment
- Option to change alignment overlay colours
- Display/remove alignment vectors
- Display/remove aligned grid
- Individually include/exclude run images from further analysis

Nonlinear Dynamics Group
info@nonlinear.com | www.nonlinear.com
Nonlinear Dynamics Ltd
Keel House | Garth Heads | Newcastle upon Tyne | NE1 2JE | UK
tel: +44 (0)191 230 2121 | fax: +44 (0)191 230 2131
Nonlinear USA Inc. | toll free: 1-866 GELS USA
4819 Emperor Blvd | Suite 400 | Durham | NC27703 | tel: 919 313 4556 | fax: 919 313 4505

Alignment controls

- Automatic or manual control of the size and position of an area of focus within the run image
- Show aligned view with options to:
    - Apply alignment changes
    - Always show aligned
    - Display unaligned view to review vector length and orientation
- Options for vector removal
    - Single vector removal by right mouse click
    - Remove automatic vectors in the current area of focus
    - Remove automatic vectors from the whole image
    - Remove all vectors in the current area of focus
    - Remove all vectors

Detection and quantitation of peptide ions

- Creation of “aggregate run” using retention time aligned data from all runs in analysis
- Automatic analysis of aggregate run with the same peptide ion outlines applied across all runs in the experiment
- Peptide ion abundance determined using total isotope peak volumes
- Median based normalisation to compare peptide ion quantitation between runs
- Charge, m/z and retention time assigned to each peptide ion

Filtering

- Filter out features to exclude from the analysis results using the following criteria:
    - m/z
    - Retention time
    - Charge state
    - Number of isotopes

Experimental group set-up

- Group runs and name groups according to experiment structure
- Set-up different groups within the same experiment e.g. control/treated and male/female
- Run name search facility to assist with creating groups in large experiments
- Colour coding to differentiate experimental groups
- Ability to add and remove runs from experimental groups
- Ability to delete experimental groups

Review peptide ions of interest

- Automatic highlighting of interesting peptide ions according to ANOVA p-values
- Peptide ions can be ordered by any characteristics displayed
- Selecting a different experimental grouping automatically updates all views
- Peptide ion identity remains consistent when experiment groupings are changed
- View peptide ion information
    - ANOVA p-value
    - Maximum fold change
    - m/z
    - Charge
    - Mass
    - Retention time
    - Abundance
    - Tags
    - Notes
    - Retention Time window
    - Intensity
    - MS/MS count
    - Protein
    - Peptide score

    - Peptide sequence
    - Modifications
    - Protein description
- Protein accession number, peptide score, peptide sequence, variable modifications and protein description displayed for each identified peptide ion
- Tick/cross buttons to individually include / excluding interesting peptide ions from statistical analysis
- Highlight and select multiple peptide ions to be ticked/crossed
- Select and remove multiple features from the analysis results as required
- Visible count of number of peptide ions ticked/crossed
- Automatically advance through peptide ion list
- Export inclusion list based on selected peptide ions*
- Export peptide ion measurements**

Peptide Ion tags

- Colour coded tags to assist with data exploration
- Right click to tag a highlighted group of peptide ions
- Add name label to peptide ion tag
- Filter peptide ion list displayed based on assigned tags
- Peptide ions can be tagged multiple times and tags can be removed
- Peptide ion tags maintained throughout the workflow

Viewing options

- Colour coding of peptide ion outlines to indicate charge
- Edit peptide ion charge colours
- Display any individual run image or the aggregate run image with corresponding details
- Search by mass and retention time to locate peptide ions of interest
- Whole run image view with option to zoom in
- Zoomed run image view
    - Click and drag to explore run at any resolution
    - View detected peptide ion outlines in detail
    - Click to select peptide ion and zoom
- 1D view
    - Mass spectrum of selected peptide ion
    - Ion chromatogram for selected peptide ion
    - Click on peptide ions in zoomed run image view to change mass spectrum and chromatogram displayed
    - Mass spectrum and chromatogram ranges automatically adjust to match zoomed run image view
- 2D montage view
    - Show current peptide ion or all peptide ions within an area of run image focus
    - Adjustable contrast
    - Adjustable montage view size
- 3D montage view
    - Select runs to display in 3D
    - Show current peptide ion or all peptide ions
    - Click and drag to reposition 3D view
    - Rotate option
    - Adjustable peak scale
    - Contour display option (where supported by graphics card)
- Expression profile view
    - Plot of mean arcsinh transformed normalised volume for each group
    - Error bars showing 3 standard errors within groups

Peptide ion editing tools

- Any edit performed on a single run image is propagated across all the run images
- Editing tools:
    - Edit peptide ion
        - Add isotope
        - Remove isotope

        - Adjust isotope bounds
    - Split peptide ion
    - Merge peptides ion
    - Delete peptide ion
    - Add peptide ion
- Undo / redo peptide ion editing
- Automatic recalculations and update of measurements following editing

Peptide Search

- Integration of MS/MS ion search data via export of peak lists to third party protein identification software (for data files containing MS/MS scans) and import of returned results with support for:
    - Phenyx ® (export .mgf and import Progenesis.tsv files)
    - Mascot™ (export .mgf and import .XML files)
    - SEQUEST® (export .dta and import .out or .pepXML files)
    - Option to add customised file export and import of results from other search engines that are specific to your workflow (nb: this may involve extra cost)
- Automatic matching of MS/MS scans to detected peptide ions
- View of MS/MS precursor m/z and retention time on detected peptide ion outlines
- Graphical view of MS/MS peaks
- Filter MS/MS spectra before you export them for identification based on:
    - Rank
    - Feature ID
    - Charge
    - Scan number
    - Exported before? (yes/no)
    - Isotope
    - ID score
    - Feature intensity
    - Precursor intensity
    - Precursor intensity (%)
    - Run name
    - Peptide sequence
    - Protein accession
    - Protein description
- MS/MS peak processing to reduce the size of the spectra but retain the essential data required by search engines
- Imported protein identification information automatically linked to detected peptide ions
- Links to perform internet searches on proteins and peptides

Peptide Filter

- When you have imported peptide identifications using the Peptide Search features you can filter out identification results based on:
    - Score
    - Hits
    - Mass
    - Charge
    - m/z
    - Retention time
    - Sequence length
    - Sequence
    - Accession
    - Description
    - Modification
- Links to perform internet searches on proteins and peptides

Protein View

- Protein Information reported
    - Accession number

    - Number of peptides
    - Score
    - ANOVA p-value
    - Abundance
    - Description
    - Conflict count
- Peptide ion information reported for each protein
    - Feature number
    - Score
    - Number of hits
    - Mass
    - Retention time
    - Charge
    - Abundance
    - Conflict count
    - Sequence
    - Variable modifications
- Expression profiles and outlines displayed for peptide ions associated with a protein
- Protein resolution view to allow user to select between conflicting protein identifications
- Tags applied to peptide ions are displayed with the proteins they are associated with
- Export of protein measurements***

Reporting and Export of measurements

- HTML report for easy sharing of results
- Whole run view showing locations of selected peptide ions
- Peptide ions selected for reporting using tag filtering
- Report title and creation date
- Customisable reporting options include
    - Overview run image annotated with peptide ion number or notes
    - Data processing methods
    - Experiment design
    - Interactive Protein Report
        - Protein table
        - Peptide table
    - Detected feature table
        - Peptide ion number
        - ANOVA p-value
        - Fold change
        - Tags
        - Notes
        - Average normalised volumes
        - Charge
        - m/z
        - Retention time
    - Detected feature details
        - Tags
        - Expression profile
        - Protein scores
        - 2D Run montage (with option to select runs to include)

*Export inclusion list

- Export of inclusion lists as .txt for:
    - ABI
    - Thermo Finnigan
    - MassLynx
- Option to create customised inclusion list specific to your lab needs (nb: may involve additional charges)

*Export peptide ion measurements

- Export of peptide ion measurements as .csv (commas separated value)

    - ID number
    - m/z
    - Retention time
    - Retention time window
    - Charge
    - Maximum fold-change
    - ANOVA p-value
    - Included in analysis (ticked or crossed)
    - Normalized abundance
    - Raw abundance
    - Intensity
    - Sample retention time
    - Notes
    - Tags selected
    - Best peptide ion match
        - Score
        - Protein
        - Sequence
        - Variable modifications
        - Protein description

**Export protein list

- Export of protein measurements as .csv (comma separated value)
    - Accession
    - Peptide count
    - Confidence score
    - ANOVA p-value
    - Maximum fold change
    - Description
    - Normalized abundance
    - Raw abundance
    - Spectral counts
    - Tags