Agilent’s NEW MassHunter Profinder

The Most Advanced Batch Feature Extraction Software for Metabolomics

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MassHunter Profinder
A Batch Feature Extraction and Review Tool

INTRODUCING THE NEW MASSHUNTER PROFINDER SOFTWARE FOR ACCURATE MASS LC/MS

- Only commercially available batch processing tool
- Fast and robust feature extraction from TOF/Q-TOF raw data
- Optimized manual re-integration for single or batch files
Which Metabolomics Customers can benefit from using MassHunter Profinder?

**Biological Research**
- Basic, clinical Research
- Drug Development

**Food**
- Food process development
- Food quality/ adulterants
- Nutrition

**Ag Chem**
- Crop development
- Pesticide / herbicide development

**Synthetic Biology**
- Biofuels

**Toxicology**
Metabolomics Workflow

**Acquire Data**
- Analyze metabolomics samples
- Use accurate mass LC/MS to analyze samples

**Extract Data**
- **Batch Feature Finding**: MassHunter Profinder

**Analyze Data**
- Differential Analysis in Mass Profiler Professional

**Identify**
- Annotate & Identify metabolites

**Pathway Analysis**
- Map results onto pathways using Pathway Architect
- Export MS/MS inclusion list for next experiment
Agilent’s Comprehensive Metabolomics Solution

- Separate & Detect
- Feature Finding
- Alignment & Statistics
- Identify
- Pathway Analysis

MassHunter Profinder (NEW)

Mass Profiler Professional

- Statistics Visualization
- Annotation & Identification
- Pathway Analysis

**LCMS and GCMS Data can be analyzed together in the same project**
Customer Goal: Find and correctly extract all chromatographic peaks in a sample

Challenges:

• Incomplete peak separation

• Unresolved peaks contribute to an increase in:
  • False peak detection, excessive missing values, incorrect identifications
  • Misdirected and wasted efforts
  • Missed or inaccurate biomarkers
  • Decreased productivity
MassHunter Profinder Workflow solution:  
So what’s new?

- A one-shot process for untargeted and targeted feature extraction
- Designed specifically for the needs of the metabolomics user
- Processes many samples!
- Recursive analysis
- Compound Group Centric: new manual review and editing functionalities
- Major reduction in processing time
- It’s FREE!!
First step: Add/Remove Sample Files and Grouping

- Projects consist of multiple data (.d) files
- One sample group column, but multiple sample groups may be assigned
- This helps with filtering, and plot overlay of the results
Profinder software demo1: Add/Remove Sample Files and Grouping
Profinder’s **Three** Feature Extraction Algorithms

1. **Batch Molecular Feature Extraction**
   - Recursive MFE reduces false positives

2. **Batch Recursive Feature Extraction**
   - Greater “missing feature” recovery
   - Allows manual editing of compounds

3. **Batch Targeted Feature Extraction**
   - Also allows manual editing
   - Data source
What is Batch Molecular Feature Extraction?

- Batch Molecular Feature Extraction begins with MFE on each data file:
  - From the raw data it finds co-eluting ions that are related:
    - isotopes, adducts (such as Na+ / K+), and dimers
  - Filters noise
  - Creates a compound chromatogram for the group of ions
  - Sums all ion signals into one value: one Feature = one compound
What is Batch Molecular Feature Extraction?

- In MassHunter Qualitative Analysis, features are extracted one file at a time.
- However, in MassHunter Profinder Batch MFE aligns features across all sample files to build a consensus spectrum for each compound group, enabling recursive, re-extraction of the batch files.
When should I use Batch MFE?

• Regular MFE doesn’t work across multiple data files
• Batch MFE is based on recursive analysis (rMFE) across multiple data files
• This results in a an averaged consensus spectrum, that is used for re-extracting all the data files
• Use Batch MFE to optimize settings first, prior to using Batch Recursive Feature Extraction
Profinder’s “Batch Recursive Feature Extraction”

1. **Batch Molecular Feature Extraction**
   - **MFE** → **rMFE**
   - Recursive MFE reduces false positives

2. **Batch Recursive Feature Extraction**
   - **MFE** → **rMFE** → **Find by Ion**
   - Find by Ion reduces false negatives
   - Allows manual editing of compounds

3. **Batch Targeted Feature Extraction**
   - **Find by Formula**
   - Also allows manual editing
   - Data source can be .CEF files, a .CSV file
Profinder’s “Batch Recursive Feature Extraction”: Post-alignment filtering after rMFE and Find by Ion

There are two post-alignment filters:

1. After rMFE $\rightarrow$ improves the quality of your target list for Find by Ion

2. After Find by Ion $\rightarrow$ improves the quality of your final compound group list, reducing the amount of manual cleanup
Profinder software demo2: Batch Recursive Feature Extraction Wizard
Four Profinder Windows:
Compound centric visualization and editing of results
Profinder software demo3: Batch Recursive Feature Extraction
Profinder’s “Batch Targeted Feature Extraction”

1. Batch **Molecular** Feature Extraction
   - MFE $\rightarrow$ rMFE
   - Recursive MFE reduces false positives

2. Batch **Recursive** Feature Extraction
   - MFE $\rightarrow$ rMFE $\rightarrow$ Find by Ion
   - Find by Ion reduces false negatives
   - Allows manual editing of compounds

3. Batch **Targeted** Feature Extraction
   - Find by Formula
   - Also allows manual editing
   - Formula source can be .CEF files, a .CSV file or a PCD/L database
Profinder software demo 4: Batch Targeted Feature Extraction
## The Profinder Advantage: 
**A Complete Solution**

<table>
<thead>
<tr>
<th>Features</th>
<th>Agilent Profinder</th>
<th>Customer Benefits</th>
</tr>
</thead>
<tbody>
<tr>
<td>Supported instruments</td>
<td>LC/TOF, and LC/Q-TOF GC/MS planned</td>
<td>Load raw accurate mass LC/MS data without any conversion</td>
</tr>
<tr>
<td>Feature finding</td>
<td>Untargeted and targeted</td>
<td>Maximum flexibility for discovery profiling</td>
</tr>
<tr>
<td>Ion grouping</td>
<td>Compound-centric</td>
<td>Let’s the software determine which ions are related</td>
</tr>
<tr>
<td>Recursive analysis</td>
<td>Yes</td>
<td>Higher quality results</td>
</tr>
<tr>
<td>Feature quality score</td>
<td>Q-score</td>
<td>Helps in ranking which compound groups to review</td>
</tr>
<tr>
<td>Feature visualization and editing</td>
<td>Yes</td>
<td>Quickly re-integrate or remove compounds</td>
</tr>
<tr>
<td>Peak alignment</td>
<td>Yes, by compound</td>
<td>Takes advantage of two separate peak alignment steps</td>
</tr>
</tbody>
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How Can You Obtain a copy of MassHunter Profinder?

• MassHunter Profinder is FREE for MPP customers
• It will be placed on the MPP Supplemental DVD
• It will also be available on Agilent SubscribeNet in the near future to update existing customers
• Please contact your local product specialist for further information