

Agilent Mass Hunter Software

Quick Start Guide

Use this guide to get started with the Mass Hunter software.

What is Mass Hunter Software?

Mass Hunter is an integral part of Agilent TOF software (version A.02.00). Mass Hunter operates on chromatographic and electrophoretic mass spectral data to extract information to reduce data complexity, eliminate potential interferences, and generate a list of molecular features.

Working with the extracted information

A feature is a discrete molecular entity defined by the combination of retention time and mass. You can use the feature information from Mass Hunter to perform many different tasks. Here are a few examples:

- Compare the original (raw data) total ion chromatogram (TIC) with the processed TIC
- Compare the original and processed mass spectra at a specific retention time (RT), or averaged over an RT range, on the TIC
- Show extracted ion chromatograms based on a mass spectral range.
- View the species clusters (isotopic, dimers, adducts) for each feature

For a complete list of tasks, see the Mass Hunter online help.



Getting started with the Mass Hunter software

Start the Mass Hunter software

This section tells you how to start the Mass Hunter software.

- Either do this:
 - **a** Double-click the Agilent TOF Software folder icon on the desktop, or

Select **Start > Programs > Agilent > TOF Software** from the desktop.

- **b** In the Agilent TOF Software folder, double-click the **Mass Hunter** icon [Q].
- Or do this:

Select **Start > Programs > Agilent > TOF Software > Mass Hunter** from the desktop.

The system displays the Mass Hunter main window.

Mass Hunter				_ 5 ×
File Batch Settings Proces	a Help			
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Learn how to access Mass Hunter functions

You can use the toolbar or the menus to perform many of the Mass Hunter tasks:



HINT To access other functions, double-click or **Ctrl**-click a plot or table row number. You may use the middle mouse button click for **Ctrl**-click in the Mass Hunter software

Learn how to use Mass Hunter

Try these exercises to familiarize yourself with the Mass Hunter application. Try the **Steps** on the left in the exercises on the next pages without the **Detailed Instructions**. If you need more help, follow the detailed instructions.

If you want to do this:	Refer to this section or exercise:
Extract feature information	"Exercises: Extracting feature information" on page 4
Extract feature information for a single file	"Exercise 1—Extract feature information for a single file" on page 4
Extract feature information for a batch	"Exercise 2—Extract feature information for a batch" on page 9
Reprocess a file with different parameters	"Exercise 3—Reprocess files with different parameters" on page 11
Display specific feature information or export and save information	"Exercises: Reviewing and saving feature information" on page 14
Show species clusters, mass spectra and extracted ion chromatograms (EIC) for features	"Exercise 4—Show species clusters, EIC and mass spectra for features" on page 14.
Show possible feature compositions	"Exercise 5—Show possible feature compositions" on page 17
Export and save feature information	"Exercise 6—Export and save feature information" on page 19
Work with chromatograms, mass spectra and contour plots	"Exercises: Working with plots" on page 22
Show mass spectra from chromatograms and show and hide other plots	"Exercise 7—Working with processed chromatograms (TICs or EICs)" on page 22
Show EICs from mass spectra	"Exercise 8—Working with mass spectra" on page 24
Learn to use contour plots	"Exercise 9—Working with contour plots" on page 25

Exercises: Extracting feature information

Exercise 1—Extract feature information for a single file

This exercise guides you through the process to extract feature information from a TOF .wiff file.

CAUTION

To process Agilent TOF data files (.wiff files) with the Mass Hunter software, the .wiff files must be local to the software. That is, the files must reside on the computer where the Mass Hunter software is running. Also, the files must not be read-only. Use Windows Explorer to check and change the file attributes.

Steps	Detailed Instructions	Comments		
 Open UrineNeg 1027_9_1A.wiff. Copy the example Mass Hunter files that come with the TOF software to a folder that only you will use. 	 a Select File > Open File. b Go to your folder that contains the Mass Hunter example TOF data files. c Select UrineNeg 1027_9_1A.wiff. d Click Open. The data file appears in the Raw Data Window. (See Figure 1.) 	 This example file contains a single sample time segment and scan data. When you open a data file that contains multiple data sets, the Select Data Set dialog box (Figure 2) appears. You must then select one combination of sample, time segment and scan segment to extract before the Raw Data Window appears. 		



Figure 1 Raw Data Window



Steps	Detailed Instruction	s	Comments
2 View the mass spectra and p information of the raw data a retention time of 8.9 min.	eak Double-click the T at Two mass spectra below the TIC, an information for th appears in the Ray on the left.	FIC at 8.9 minutes. a for that RT appear d the peak e mass spectrum w Data Peak Viewer	 One mass spectrum shows the baseline, and the other shows the threshold level, which can be changed by the user (see "Exercise 3—Reprocess files with different parameters" on page 11).
	QC:\MassHunter_Profiler\Example_Data\RatUnineMFE-	4\Samples 9\UnneNeg1027_9_1A.mff	_[6]×
	Fin Batch View Settings Process Help		
	Exact 487 Peaks	HO 14 1	
	ID m/z heicht	TIC 12	• • • •
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	20 7 113.9500 614.00 24 25 120.0506 644.00 24 25 120.0506 422.00 24 24 120.0506 120.0506 25 120.0506 1260.00 126.00 26 27 77 120.040 76.00 27 77 120.040 61.00 127.00 28 28 120.040 127.00 138.98 130.0913 127.00 29 40 19.992 200.00 127.00 129.020 127.00 129.020 127.00 129.020 127.00 129.020 127.00 129.020 127.00 129.020 127.00 129.020 127.00 129.020 127.00 129.020 127.00 129.020 127.00 129.020 127.00 129.020 127.00 129.020 127.00 129.020 127.00 129.020 129.020 129.020 127.00 129.020 129.020 129.020 129.020 129.020 129.020 <td>w 1000 10</td> <td><u>∗1</u></td>	w 1000 10	<u>∗1</u>

S	teps	Detailed Instructions Comments	
3	View the threshold level for the mass spectrum between 400 and 600 m/z.	 a In the lower spectra viewer, hold the mouse button down and draw a rectangle at the baseline between 400 and 600 m/z. b Release the mouse button. c Repeat the zoom until you see the blue line. 	
		m/z 40 40 60 60 60 60 60 10 10 10 10 10 10 10 1	
4	Save the TIC image to a file.	a Right-click the TIC.	
		b Select Image > Save to File > Bitmap.	
		c Go to the folder into which you want to	
		d Click Save.	
		TIC 12	

Detailed Instructions

Comments

•

5 Process the file.

Steps

 An .mhd file is created upon processing. This file contains he extraction result. Although you can open this file directly, you should open the original .wiff file so that you access more information.

Note: if you attempt to process the *.wiff file with the same set of extraction parameters (see "Exercise 3—Reprocess files with different parameters" on page 11) as the ones stored in the *.mhd file, the program simply loads the result from the *.mhd file.

- Select Process > Run, or click the Process button on the toolbar. The processed total ion chromatogram appears under the original TIC in the Processed Data Window (Figure 3), and the Feature Summary Table appears in the Features Viewer.
- An Excel file is automatically created containing attributes for each feature. The Excel file takes the name of the **.wiff** file processed. In this case, the file is named UrineNeg1027_9_1A.xls. (Figure 4).





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6	1	124.1	192.027	180.91		0.03	4	-1		1	9.05					
7	2	640.3	246.0202	123.09		0.02	4	-1	0	1	5.89					
8	3	927.1	381.9845	122.44		-0.02	3	-1			14.63					
9	4	533.9	166.0629	74.15		0.06	/	-1	U		3.1					
10	5	927.2	445.9809	64.07		-0.02	5	-1	U		13.43					
11	6	229.7	97.9673	63.86		-0.03	2	-1	L L		18.2					
12		71.9	97.977	63.52		+0.02	3	-	U C		4.0					
1.4	0	44	E0E 0719	46.50		0.01	2	- 1	0		4.19					
14	10	422.2	170.0570	39.32		-0.03	4	-1	0		4.96					
16	11	432.2	406.0364	37.24		0.00		-1	0	1	4.30					
17	17	47.4	169 0436	36.9		0.04	4	-1	0		4.22					
18	12	653.8	244.0045	35.11		0.04	2	-1		1	6.97					
19	14	679.5	213 0103	34.84		0.01	2	-1	0		8.52					
20	15	50	196.058	32.12		0.06	1	-1	0	1	5.4					
21	16	123.8	112.016	30.61		0.02	2	-1	C C	1	86					
22	17	827.4	395,983	28.83		-0.02	3	-1	i a	1	15.53					
23	18	972.7	1034.995	26.55		0	2	-1	C C	1	16.07					
24	19	945.5	521.9175	26.07		-0.08	2	-1	C	1	15.65					
25	20	95.3	168.0285	25.78		0.03	4	-1	0	1	4.41					
26	21	622.3	189.9936	20.04		-0.01	2	-1	0	1	8.04					
27	22	646.9	173.9992	19.25		0	3	-1	0	1	6.73					
28	23	953.2	231.994	16.4		-0.01	2	-1	0	1	16.06					
20	74	401 0	102 0742	10 02		0.07	E	4		4	207					

Figure 4 Excel spreadsheet with feature information automatically saved after processing

Exercise 2—Extract feature information for a batch

This exercise shows you how to automatically extract feature information for a batch of TOF data files for downstream use or later review.

CAUTION

To process Agilent TOF data files (.wiff files) with the Mass Hunter software, the .wiff files must be local to the software. That is, the files must reside on the computer where the Mass Hunter software is running. Also, the files must not be read-only. Use Windows Explorer to check and change the file attributes.

Steps	Detailed Instructions	Comments				
 Create a batch of files to extract. Use the following files: All three sample 9 .wiff files: UrineNeg1027_0_3A.wiff, UrineNeg1027_9_2A.wiff, and UrineNeg1027_9_1A.wiff. The first file in the sample 10 folder: UrineNeg1027_10_2A.wiff 	 a Select Batch > Create. Or, click the Create Batch button in the toolbar. b Hold the Shift key as you select all three files in the sample 9 folder, and click Open. The Batch Process Window (Figure 5) appears with the .wiff files from sample 9 listed. c Click the Create Batch button in the toolbar again. d Select the first file in the sample 10 folder. e Click Open. The Batch Process Window now contains all four files. 	 If any of the .wiff files contains more than one data set, the Batch Data Set Options dialog box appears. You then select if you want to use all the data sets in all the files or one spectra set per file. If you click One spectra set per file, the Select Data Set dialog box appears. For more details about using these dialog boxes, see the Mass Hunter online help. 				
	Files	Sample Time seg. Scan seg. Status				
	C:\Mass Hunter_Profiler\Example_Data\RatUrineMFE4\Samples 9\Uri	neNeg1027_9_3A.wiff 1 1 1 Pending				

Figure 5 Batch Process Window

C:\Mass Hunter_Profiler\Example_Data\RatUrineMFE4\Samples 9\UrineNeg1027_9_1A.wiff 1

💍 C:\Mass Hunter_Profiler\Example_Data\RatUrineMFE4\Samples 10\UrineNeg1027_10_2A.wiff 1

Pending

Pending

Steps	Detailed Instructions	Comments		
2 Process the files.	 Select Process > Execute Batch, or click the Process button in the toolbar. 	 Notice the Progress bar on the left. When the runs are complete for all data files, you see a message saying that the batch run is complete. To see the features, groups and mass spectra for each of the processed .mhd files in the batch, you must open each individually and work with it in the Processed Data Window. 		

Exercise 3—Reprocess files with different parameters

You can reprocess original .wiff files or already processed .mhd files with new extraction parameters.

Steps	Detailed Instructions	
1 Open the file UrineNeg 1027_9_1A.wiff.	 a Select File > Open File. b Select UrineNeg 1027_9 click Open. 	_ 1A.wiff , and
 Change the extraction para RT range of 6 to 12 minu m/z range of 300 to 800 250 x 1000 mass spectra 	aSelect Settings > ExtracutesParameters,or click the Extraction Paralal peaksbutton in the toolbar.	tion Irameters
	■ Extraction Parameters □ Data Ranges □ Use all the available data Min Max RT 0.00 20000. M/Z 0 10000 Spectral Peak Detection 5 S/N threshold 5 Feature Detection 500 ■ Peptidic isotope distribution 5 Single charge only Adducts Positive ions Ne ■ Salt dominated	X 00 min 00 Da 00 K1000 gative ins 0 Add 0 Delete

Steps	Detailed Instructions	Comments
	 b To set the RT range, enter 6 as the Min RT and 12 as the Max RT. c To set the m/z range, enter 300 as the Min M/Z and 800 as the Max M/Z. d For the Max spectral peaks to use, enter 250. e Click OK. 	
 3 Reprocess the file. Because the open .wiff file was previously processed and produced an .mhd file, the system displays the Reprocessing Options dialog box. Make sure to reprocess the file and not load the old results. 	 a Select Process > Run, or click the Process button in the toolbar. b Click Reprocess in the Reprocessing Options dialog box. 	• This dialog box appears only when you try to process a newly opened *.wiff file which has been processed previously. If you try to reprocess a file whose feature information is displayed now, the software assumes that is your intention and will not show the dialog.
	Reprocessing Options	X
	Data has been processed using different parameters	8
	Make your choice	
	Load old result Reprocess Canc	el

Steps	Detailed Instructions	Comments		
4 Review the extraction parameters used for the current result.	 a Select View > Parameter Table. b Close the table after review. 	 You may want to occasionally review the parameters used to display the currently displayed results. 		

	Parameter	Value	
1	Data Ranges		
2	Min BT	6.00 min	
3	Max RT	12.00 min	
4	Min M/Z	300.00 D a	
5	Max M/Z	800.00 D a	
6			
7	Spectral Peak Detection		
8	S/N	5.0	
9			
10	Feature Detection		
11	Max spectral peaks used	250 x1000	
12	Peptidic isotope distribution	no	
13	Single charge only	no	
14			
15	Adducts		
16	Salt Dominated	no	
17	Positive Adducts	K	
18		Na	
19	Negative Adducts		

Exercises: Reviewing and saving feature information

Exercise 4—Show species clusters, EIC and mass spectra for features

The Feature Summary Table presents a list of features and their attributes. When coelution grouping is enabled (default), the list presents group features that coelute at almost equivalent retention times. This exercise shows you how to bring up the Feature Details Table, which lists the features and their species clusters for the coeluting group. When coelution grouping is disabled, the two tables list only individual features.

St	eps	Detailed Instructions	Comments
1	Display the species clusters for the features that coelute together in a group. • Display group #2 in the .wiff file.	a Double-click row 25 of the Feature Summary Table. Double-clicking the row header of any feature of the group brings up the Feature Details Table for the group.	 The Feature Details Table contains the species clusters for the features in the group. The Plot Viewer displays the mass spectra for the group of features ,
	File Batch View Set	vidWeil/MiczehegativeIonAcids/UnineNeg1027_9_LAwdfi ttings Process Help	XOL
	Expot featur	res:1298/groups:501	
	State 1 0 0.07 3 1 0.07 3 1 0.07 3 1 0.07 3 1 0.07 3 1 0.07 3 1 0.07 4 1 0.07 1 0.07 1 0.07 7 1 0.07 1 0.07 1 0.07 9 1 0.07 1 0.07 1 0.07 9 1 0.07 1 0.07 1 0.07 9 1 0.07 1 0.07 1 0.07 10 1 0.07 1 0.07 1 0.07 11 1 0.07 1 0.07 1 0.07 12 1 0.07 1 0.07 1 0.07 13 1 0.07 1 0.07 1 0.07 1 0.07 14	Instruct Model Op/ Model Model <t< td=""><td>EXCRIT-2 DOI - 10 Features</td></t<>	EXCRIT-2 DOI - 10 Features
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Steps	Detailed Instructions	Comments
3 Show ion mass spectra and EICs for the M+H ion of the second feature in the group.	 Double-click the row #9 in the Feature Details table. Image: State of the state of	 The Plot Viewer displays the mass spectrum and EIC for the original data, and a mass spectrum and EIC for the processed ion specified in row 9.
4 Show feature mass spectra for a feature when coelution grouping i disabled.	 a Select Settings > Disable Coelution Grouping. b Click the Process icon to bring up the Feature Summary Table with a listing of features and no groups. c Double-click the row #2. You now see the Feature Details Table with species cluster information for only that feature. 	• The mass spectra for the selected feature appear in the Plot Viewer, and the species clusters for that feature only appear in the Feature Details Table.
5 Re-enable Coelution Grouping.	 a Select Settings > Enable Coelution Grouping. b Click the Process icon. 	

Exercise 5—Show possible feature compositions

The Mass Hunter software calculates the possible compositions for any feature you select either from the Feature Summary Table or the Feature Details Table. This exercise shows you how to display these compositions and set up the "rules" for calculating the composition.

eps	Detailed Instructions	Comments
Display the possible compositions for feature at row 15.	a In the Feature Summary table, right-click row #15 at the row header.	 When you click Composition, the Possible Compositions dialog box appears with the possible compositions listed. If the MW is greater than 800 amu
	4 1 0.897 97.3971 0.21 97.2979 0.01 2 1 1 0.071 5 1 0.898 952.1087 0.11 95.114 0.114 0.11 0.114 0.11 0.014 0.11 0.014 0.11 0.01 1 0.02 1 0.02 1 0.02 1 0.01 1 0.02 1 0.02 1 0.02 1 0.02 1 0.01 0.050 0 1 0.050 0 1 0.050 0 1 0.051 1 0.050 0 1 0.051 1 0.050 0 1 0.051 0 1 0.050 0 1 0.051 1 0.051 1 0.051 1 0.051 1 0.051 1 0.051 1 0.051 1 0.051 1 0.051 1 0.051 1 0.051 1 0.051 1 0.051 1 0.051 <td>a warning message appears to let you know that the calculation may take surbils and to ack you if you</td>	a warning message appears to let you know that the calculation may take surbils and to ack you if you
	10 1 8.89 950.1006 0.10 980.1533 0.16 2 4 1 1 0.056 11 1 8.89 97.6002 0.23 370.033 0.09 2.4 -1 1 0.056 12 1 8.899 97.6002 0.23 370.033 0.09 2.4 -1 1 0.056 12 1 8.899 97.6002 42 370.133 0.07 31 0.04 composition 9 95.1105 472 95.1174 0.18 2.4 -1 1 0.057 14 1 1.556 452.122 1.018 97.1748 0.18 3.1 -1 1 0.057 14 1 2.859 95.1749 1.018 3.1 -1 1 0.047 14 1 0.859 2.47 1 0.108 3.1 -1 1 0.042 15 0.495 2.47 1	want to continue.
	16 1 0.89 0.01 701.299 1 0 1 0.051 17 1 0.859 0.06 470.059 1 0 0 1 0.051 18 1 0.859 472.1358 0.06 470.055 1 0 0 1 0.051 19 1 0.859 472.1358 0.06 470.013 1 1 0.051 15 1 0.859 0.07 475.013 1 0 0 1 0.054 20 1 0.859 0.07 475.013 1 0 0 0.042	
	21 1 8.869 7.81806 0.04 7.81433 0.15 2 -1 1 0.07 22 1 8.900 0.03 681527 1 0 0.1 0.04 23 1 6.900 0.10 437.067 1 0 0 1 0.079 24 2 7.159 0.02 560.097 1 0 0 1 0.079 26 2 7.159 0.02 560.097 1 0 0 1 0.079 26 2 7.159 0.02 560.097 1 0 0 1 0.079	-1

Exp	chemistry	Comp	osition	S	
	chemical formula	dm(Da)	dm(ppm)	DBE	score
1	C18H2006	-0.0002	-0.7	9.0	84
2	C11H20N604S	0.0005	1.4	5.0	62
3	C10H24N208S	-0.0009	-2.7	0.0	54
4	C19H16N4O2	0.0011	3.3	14.0	68
5	C12H16N10S	0.0018	5.4	10.0	63
6	C7H20N609	0.0030	8.9	1.0	71
7	C15H2406S	0.0031	9.4	4.0	85

Steps I		Detailed Instructions	Comments
<u>S1</u> 2	Add isotope N15 to the list of elements used in the calculation.	Detailed Instructions a In the Possible Compositions dialog box, click Chemistry.	Comments • After you follow these instructions, Mass Hunter adds the N15 isotope to the element list. The isotope appears at the bottom of the Periodic Table with a marked checkbox.
		Height uncertainty 10 %	

b In the Composition Info dialog box, click **Add/Remove**.

E Selec	t Elemen	lts															2
H																	He
Li	Be											B	C N	N	0	F	Ne
∟ Na	Mg											AI	Si	P	S	CI	Ar
Г К	Ca	Sc	Ti	v	Cr	∏ Mn	Fe	Co	Ni	Cu	⊂ Zn	Ga	Ge	As	⊂ Se	Br	Г Kr
□ Rb	□ Sr	Y	Zr	Nb	Г Мо	Г Тс	Ru	Rh	Pd	⊢ Ag	⊂ Cd	n In	⊂ Sn	⊂ Sb	Г Те	I	⊂ Xe
Cs	Ba	La - Lu	Hf	Г	w	□ Re	C Os	۲ Ir	□ Pt	∟ Au	□ Hg	П	Pb	E Bi	Г Ро	□ At	□ Rn
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3 Set up the calculation so that the
software uses no fewer than zero
and no more than 10 N15.a Enter the Min number of atoms for
N15 of 0 and Max number of 10.
b Click OK.• When you click OK, the software
automatically recalculates the
possible compositions.

Exercise 6—Export and save feature information

You can export the information in each Mass Hunter table to an Excel file:

- Feature Summary Table
- Feature Details Table
- Possible Compositions Table
- Parameter View

You can also save molecular ion peak information for all the features.

St	eps	Detailed Instructions	Comments		
1	 Export the Feature Composition table to a Microsoft Excel file. If you are already in the Possible Compositions dialog box after Exercise 5, skip to step b. View the Excel file. 	 a Right-click any feature in the Feature Summary Table, and click Composition. b Click Export. c Specify a destination folder. d Specify a file name. e Click OK. f Close the Possible Compositions dialog box. g Go to the folder containing the Excel file, and open the file. 	 If you are in the Mass Hunter main window, open the file UrineNeg1027_9_1A_1_1_1.mhd. Then follow the instructions in this step. 		
2	Export the Feature Summary table to a Microsoft Excel file.View the Excel file.	 a At the top of the Feature Summary Table, click Export. b Specify a destination folder. c Specify a file name. d Click OK. e Go to the folder containing the Excel file, and open the file. 	• See Figure 6, "Feature Summary Table in Excel," on page 20.		
3	Export the Feature Details table.View the Excel file.	 f At the top of the Feature Details Table, click Export. g Specify a destination folder. h Specify a file name. i Click OK. j Go to the folder containing the Excel file, and open the file. 	 See Figure 7, "Feature Details Table in Excel," on page 21. If the Feature Details Table is not present, double-click the row number of the group or feature of interest in the Feature Summary Table. 		

St	eps	D	etailed Instructions	Comments		
4	Save and view the molecular ion information.	a b c d	Select File > Save Feature Info. Specify a destination folder. Click OK. Go to the folder containing the Excel	•	See Figure 8, "Feature Info Table in Excel," on page 21.	

file, and	open the file.	

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Figure 6 Feature Summary Table in Excel

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8	M-H	7.202	379.0915	380.0988	3.78	0.05
9	M-H+1	7.203	380.0955		0.65	0.05
10	M-H+2	7.197	381.0955		0.1	0.0
11	M-H+3	7.204	382.0894		0.16	0.06
12	M-H+4	7.194	383.096		0.03	0.08
13						
14	M	7.199		361.1162	0.29	0.07
15	M-H	7.203	360.109	361.1162	0.24	0.07
16	M-H+1	7.196	361.1095		0.05	0.08
17						
18	M	7.201		516.0737	0.25	0.06
19	M-H	7.206	515.0665	516.0737	0.2	0.06
20	M-H+1	7.195	516.071		0.04	0.06

Figure 7 Feature Details Table in Excel

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7	6	1	-1	M-H	8.899	331.1189	1.97	
8	7	1	-1	M-H	8.839	308.1175	1.73	
9	8	1	-1	M-H	11.112	344.98	1.81	
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11	9	1	-1	M-H	9.502	417.1195	1.42	
12	10	1	-1	M-H	8.031	407.1221	1.26	
13	11	1	-1	M-H	11.428	315.2536	1.18	
14	12	1	-1	M-H	9.061	325.0935	1.17	
15	13	1	-1	M-H	11.435	309.2079	1.06	
16	14	1	-1	M-H	6.985	326.0668	0.92	
17	15	1	-1	M-H	9.322	445.0779	0.79	
18	16	1	-1	M-H	6.931	307.0443	0.83	
19	16	2	-1	2M-H	6.931	615.0997	0.02	
20	17	1	-1	M-H	6.817	369.0831	0.78	
21	18	1	-1	M-H	8.034	385.1407	0.79	-
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Figure 8 Feature Info Table in Excel

Exercises: Working with plots

Exercise 7—Working with processed chromatograms (TICs or EICs)

You can perform the chromatogram operations on total ion chromatograms (TICs) and extracted ion chromatograms (EICs) and on the original and processed chromatograms, either together or separately.

Steps		Detailed Instructions		Comments	
1	 Do the following: Zoom in on the chromatograms between 8 to 10 minutes to include the entire peak around 9 minutes. Zoom out the chromatogram. 	a b c	Hold the mouse button down as you draw a rectangle around the specified time window. Release the mouse button. Click the Full Zoom Out button.	•	When the zoom function is locked (default), the zoom works on both chromatograms simultaneously. If you click the Lock button to unlock the zoom function, you can zoom in or out of each chromatogram separately.
2	View the mass spectra (original and processed) at around RT 9 minutes.	•	Double-click the RT at around 9 minues in the chromatogram.	•	The system displays the mass spectrum at RT 9 min. for both the original and processed chromatograms.
3	View the average mass spectra over the range of RT 8 to 10 minutes.	a b c	CTRL-click the left mouse button (or click the middle mouse button) to place a line on the plot to set the low end of the range over which the average is calculated. CTRL-click the left mouse button again to place a line on the plot to set the high end of the range. Right-click the image, and select Ave MS – Range.	•	See Figure 9, "Chromatographic range context menu," on page 23. The average mass spectra calculated over the selected range appears below the chromatogram plots for the original chromatogram and the processed chromatogram.
4	Hide the TIC.	a	Right-click the image, and select Hide EIC from the context menu (see Figure 9).	•	When an EIC is superimposed on a TIC, you can hide the TIC.
5	Re-show the TIC.	a	Right-click the image, and select Show EIC from the context menu.		



Figure 9 Chromatographic range context menu

Exercise 8—Working with mass spectra

Steps		Detailed Instructions		Comments	
1	Show an extracted ion chromatogram at about 500 m/z (highest peak)	1	Double-click the highest peak in the mass spectrum.	•	The Plot Viewer displays the EIC for this RT in both the original and processed chromatogram windows.
2	Show an extracted ion chromatogram for the m/z range of 500 to 550 m/z (approximate).	a b	CTRL-click the left mouse button (or click the middle mouse button) to place a line on the mass spectrum to set the low end of the range over which the chromatogram is extracted. CTRL-click the left mouse button again to place a line on the mass spectrum to set the high end of the range. Bight-click the image and select Show	•	The EICs appear superimposed on the TICs.
		C	EIC from the context menu.		



Exercise 9—Working with contour plots

The contour plot is a two-dimensional representation of 3D data, with m/z as the x-axis, RT as the y-axis and intensity as the z-axis (the darkness or lightness of the image on the plot). Two plots are shown, one for the original data and the other, for the processed data.

Steps		Detailed Instructions		Comments	
1	Show the contour plots.	a	Right-click any one of the chromatograms, and select Show Contour Plot .	•	See Figure 9, "Chromatographic range context menu," on page 23. The contour plots appear for the original and processed chromatograms.
2	Zoom into and out of the contour plot.	a b c	Hold down the mouse button and draw a rectangle around the area of interest. Release the mouse button. To zoom out, click the Zoom Out button next to the coordinates text field.		
3	Find the position of m/z= 500 and RT=9.	a b c	CTRL-click the left mouse button (or click the middle mouse button). Two perpendicular blue lines (cross-hair cursor) appear in the plot. With the mouse button held down, move the cross-hair cursor to the point specified in step 3. Double-click the plot to remove the cross-hair cursor.	•	You can see the coordinates of the position in the box to the left and below the left contour plot. (Figure 10)
4	Hide the contour plot.	a	Right-click the plot, and select Hide Counter Plot .		



Figure 10 Contour Plots with cross-hair cursor

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In this guide

This Quick Start Guide includes an overview of the Mass Hunter software, quick reference information to get started using the software, and a set of tutorials to learn how to use the software.

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