Agilent

Agilent MassHunter Workstation Unknowns Analysis

Familiarization Guide

Notices

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Contents

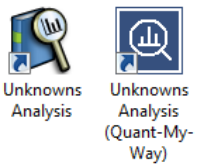
In this Guide...

This guide presents step-by-step exercises to help you learn to use the Unknowns Analysis program. You can do these exercises with the demonstration analysis, method, and library files, shipped with the system installation disk, or with data you acquire.

Choosing Unknowns Analysis Desktop Icons

Quantitative Analysis B.09.00 offers Unknowns Analysis desktop icons for the **Classic** user interface and the **Quant-My-Way** user interface. The **Classic** user interface has a look and feel similar to the user interface offered in Quantitative Analysis B.08.00, with tools and options located in a menu bar. The **Quant-My-Way** user interface has a modern ribbon, with tools and options located on tabs and ribbons instead of in a menu bar. You can select to install the Classic user interface desktop icon, the **Quant-My-Way** user interface desktop icon, or both.

This Familiarization Guide follows the **Classic** user interface. However, where the **Quant-My-Way** user interface navigation differs, those steps are included and highlighted in orange.



Before you begin these exercises

Copy files from the installation media to your hard disk

1 Navigate to the **Data** folder on the installation media.

2 If the folder is in a compressed format, extract the data files from their zip format.

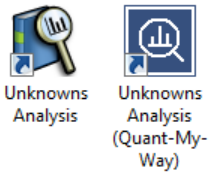
3 Copy the **Data** folder from your installation media in uncompressed format to any location on your hard disk.

This folder contains all of the data, method, and library files needed for these exercises. Do not reuse the example data files on your system unless you know that they are identical to the originals on the media. If the example data files already on the system do not match the original ones of the media exactly, then the results obtained during these exercises will not match those shown in this guide.

Task 1: Identify Compounds with TIC Analysis

Create a new analysis

1 Start **Unknowns Analysis** by double-clicking the desktop icon.
or
Click **Start > Agilent > MassHunter Workstation > Unknowns Analysis.**



When you open the program, the default layout appears.

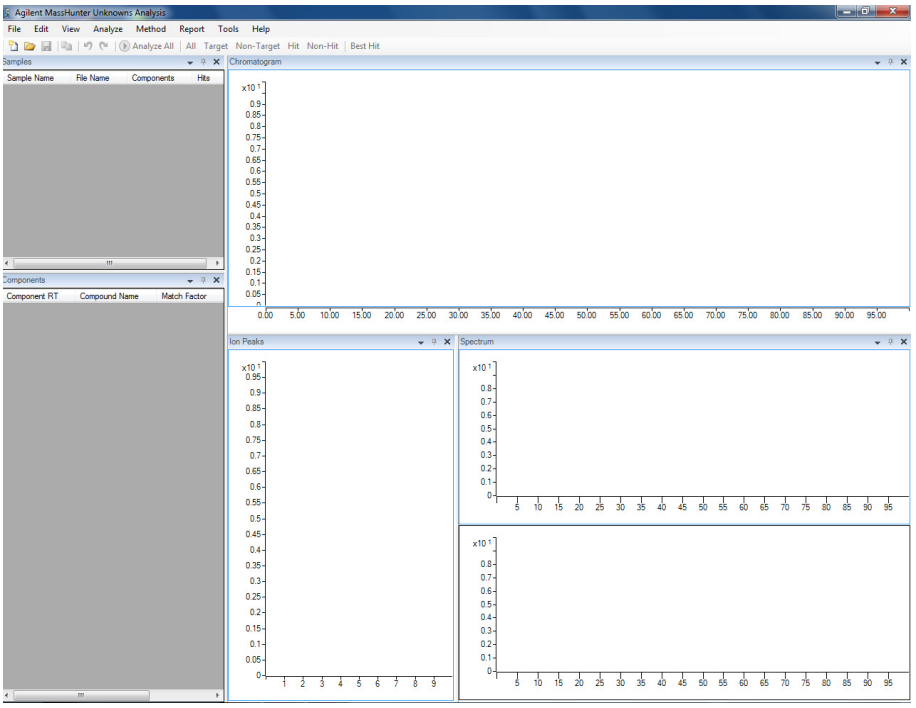


Figure 1.

If the default layout is not present, click **View > Preset Layout > Standard** to restore the default layout before creating a new analysis.

In the **Quant-My-Way** user interface, on the **View** tab, click **Preset Layout > Standard**.

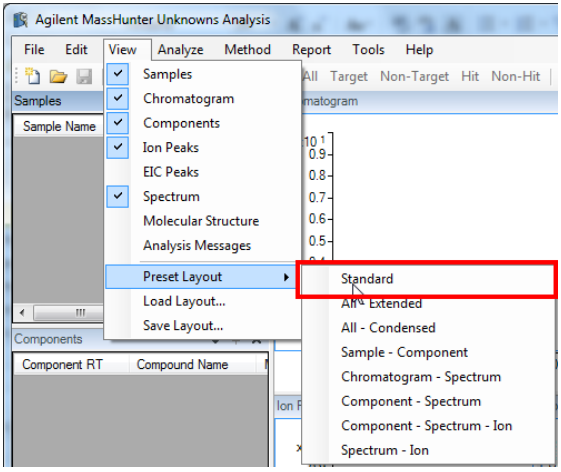


Figure 2.

2 Select **File > New Analysis**.
In the **Quant-My-Way** user interface, on the **Home** tab, click **New Analysis**.

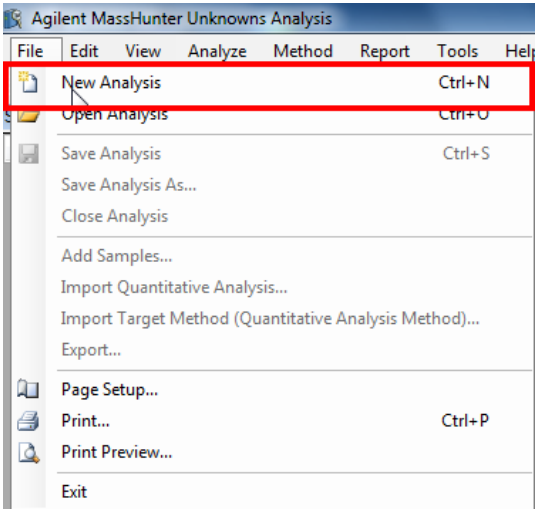


Figure 3.

3 Navigate to **MassHunter\Data\Evaldemo**, or the folder where the data file to be analyzed is stored.

4 Type the analysis name **evaldemo** for the analysis, and click **Create**.

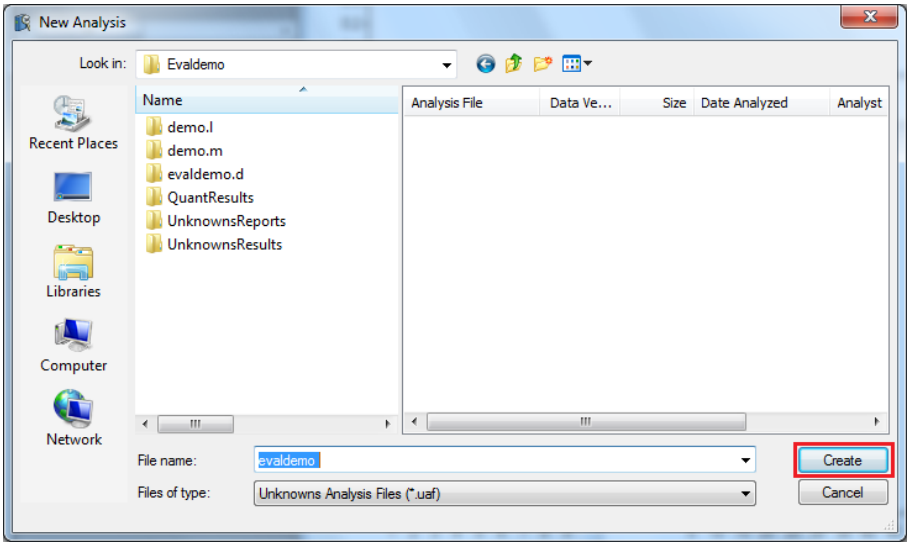


Figure 4.

Add samples to the analysis

1 Select **File > Add Samples**.
In the **Quant-My-Way** user interface, on the **Home** tab, click **Add Samples**.

2 Select the sample file(s) and click **OK** to add the sample to the batch.

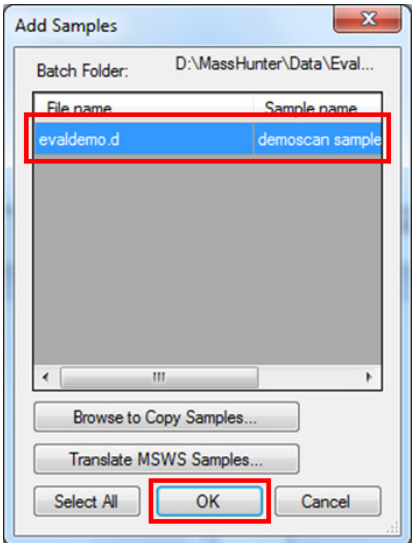


Figure 5.

The Analysis table is no longer empty. It now contains the demo sample.

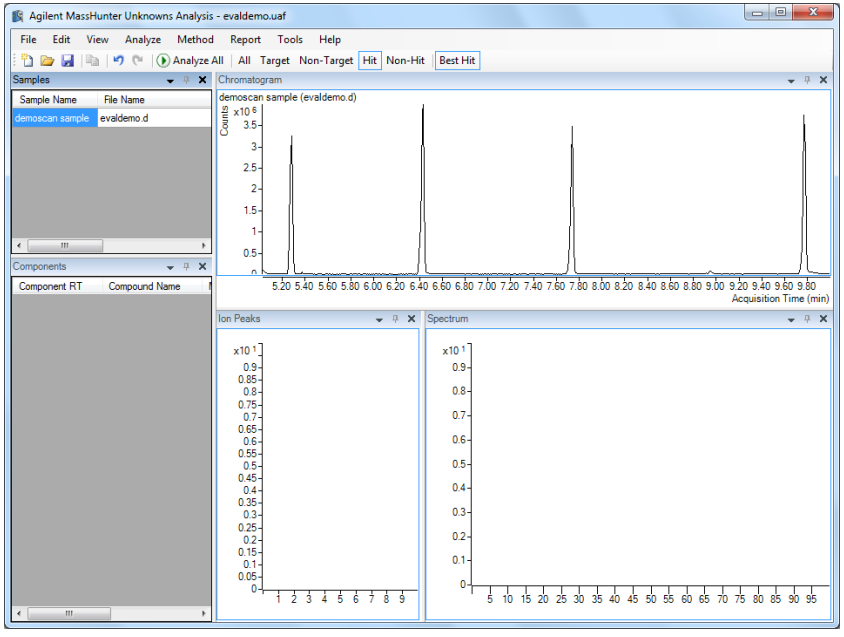


Figure 6.

Set up the method for the analysis

Select **Method > Edit**.

In the **Quant-My-Way** user interface, on the **Home** tab, click **Edit Method**.

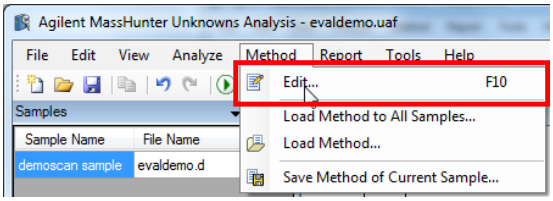


Figure 7.

The Method dialog box standard view appears. For this task, we will use the **Standard** view.

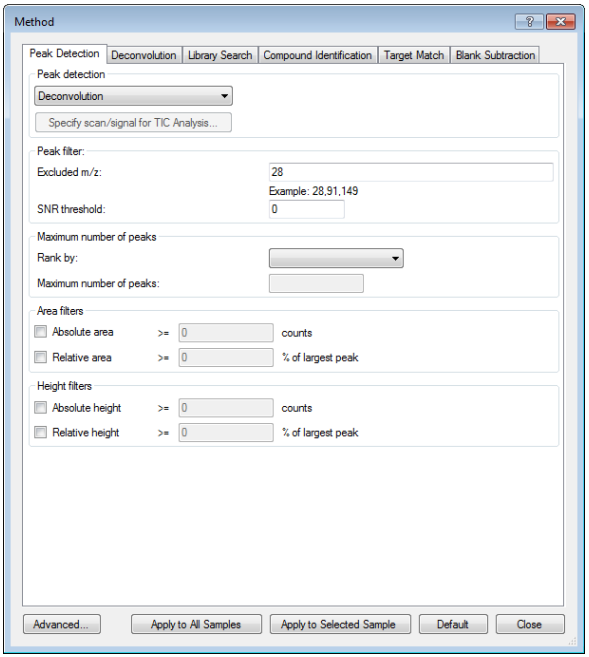


Figure 8.

Note that these are the default parameters for the method. You can click **Default** at the bottom of the Method dialog box to restore default parameters before creating a new method in the next step.

Set Peak Detection options

1 Select **TIC Analysis** from the **Peak detection** drop-down menu.

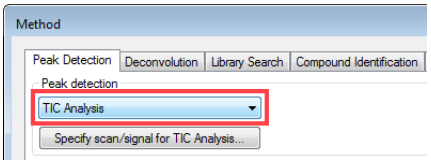


Figure 9.

- **TIC Analysis**: Identifies the chromatographic peaks using integration instead of deconvolution.
- **Deconvolution**: Deconvolutes the components in the chromatogram and extracts the 'clean' spectra from background noise based on both retention time and peak shape.

2 Click **Specify scan/signal for TIC Analysis** to process GC signals.

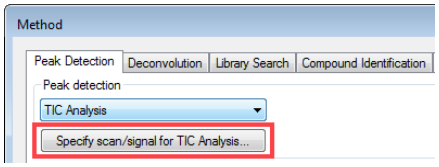


Figure 10.

3 In the **Maximum number of peaks** section, select **Area** from the **Rank by** drop-down menu, and enter **5** for the **Maximum number of peaks**.

Figure 11.

4 In the **Area filters** section, select **Relative area** and enter **1** for the **% of largest peak**.

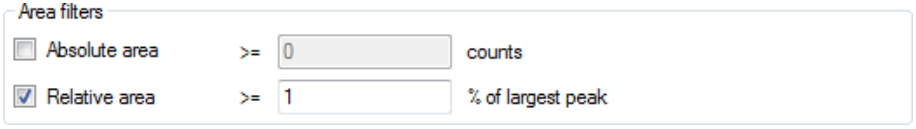


Figure 12.

Set Library Search options

1 Click **Library Search**.

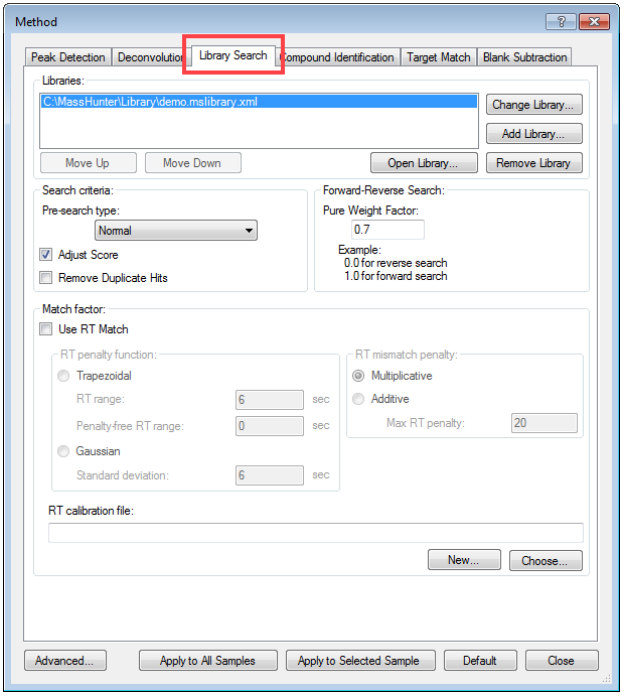


Figure 13.

2 Click **Change Library**.

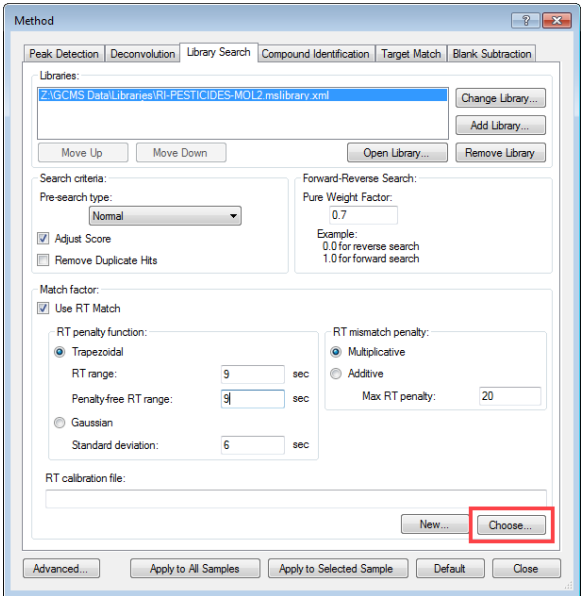


Figure 14.

3 Navigate to **MassHunter\Data\Evaldemo**, or the relevant folder, select **demo.L**, and click **Open**.

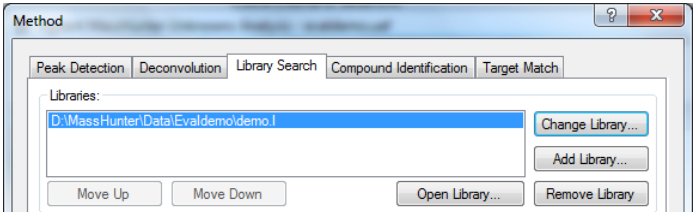


Figure 15.

4 In the **Search criteria** section, select **None** from the **Pre-search type** drop-down menu.

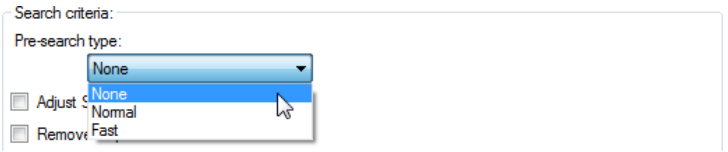


Figure 16.

There are 3 Pre-search types: **None, Normal**, and **Fast**. By default, Unknowns Analysis uses **Normal**.

- **None**: The library search is not subjected to a preliminary screening process.
- **Normal**: The screening algorithm uses the entire library as the list of candidates if the indexing scheme does not produce enough candidates. It is 50-100 times faster than no pre-search, with essentially zero false negatives rate for high-scoring hits (match score > 80).
- **Fast**: The screening algorithm uses whatever list of candidates it gets from the index and avoids the entire library-search even if there are not enough candidates found. The speed is 100-1000 times faster than no pre-search, with ≥1% false negatives rate for high-scoring hits.

Set Compound Identification options

1 1. Click **Compound Identification**.

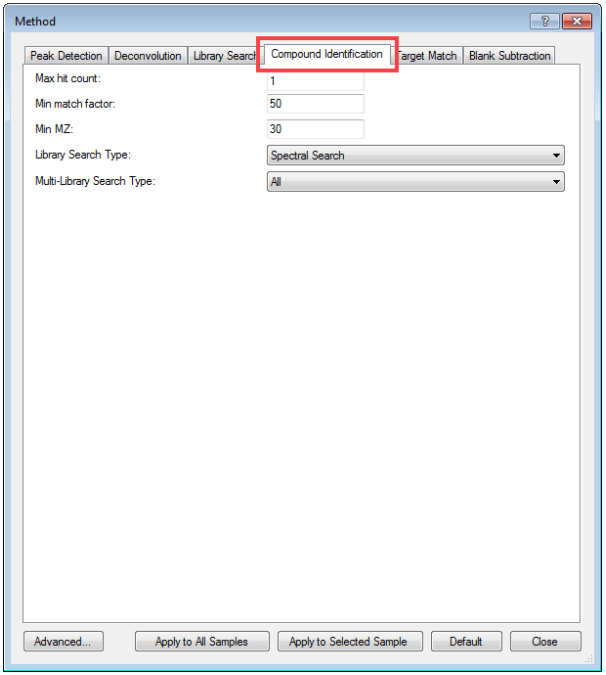


Figure 17.

For this task, we will use the default Compound Identification parameters.

2 Click **Apply to All Samples**, and then click **Close**.

Analyze and review results

1 Click **Analyze All**.

After the analysis is complete, the main view that appears should look like the example below. This is the default layout and contains the default column settings.If you see a different layout than the one in the example below, select **View > Preset Layout > Standard** to reset the standard layout.

In the **Quant-My-Way** user interface, on the **View** tab, select **Preset Layouts > Standard**.

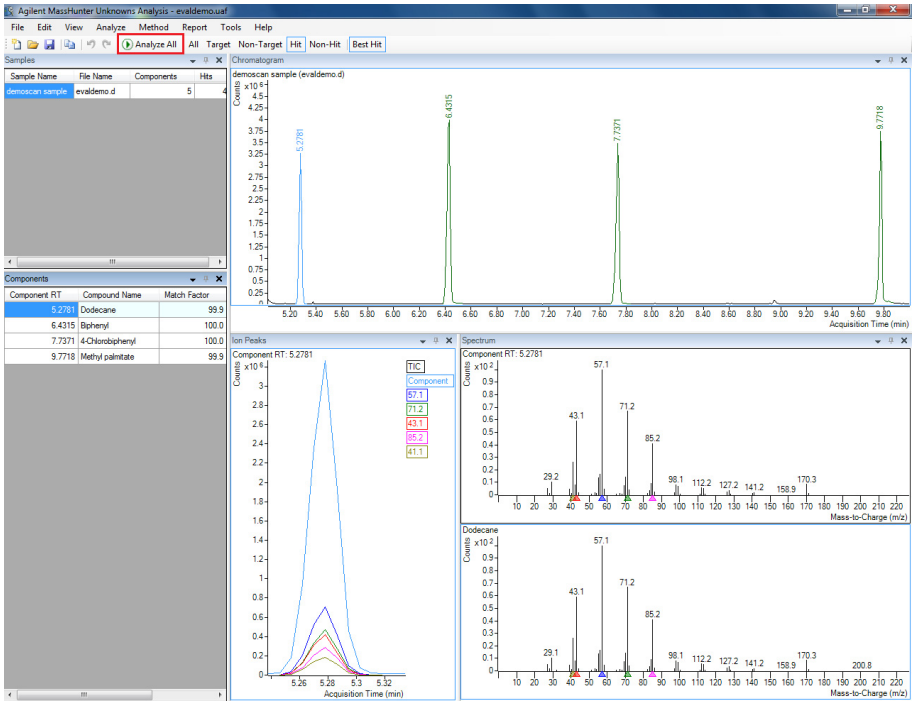


Figure 18.

2 Select **View > Preset Layout > All-condensed**.

The system displays the All-condensed view.

In the **Quant-My-Way user** interface, on the **View** tab, select **Preset Layouts > All-condensed**.

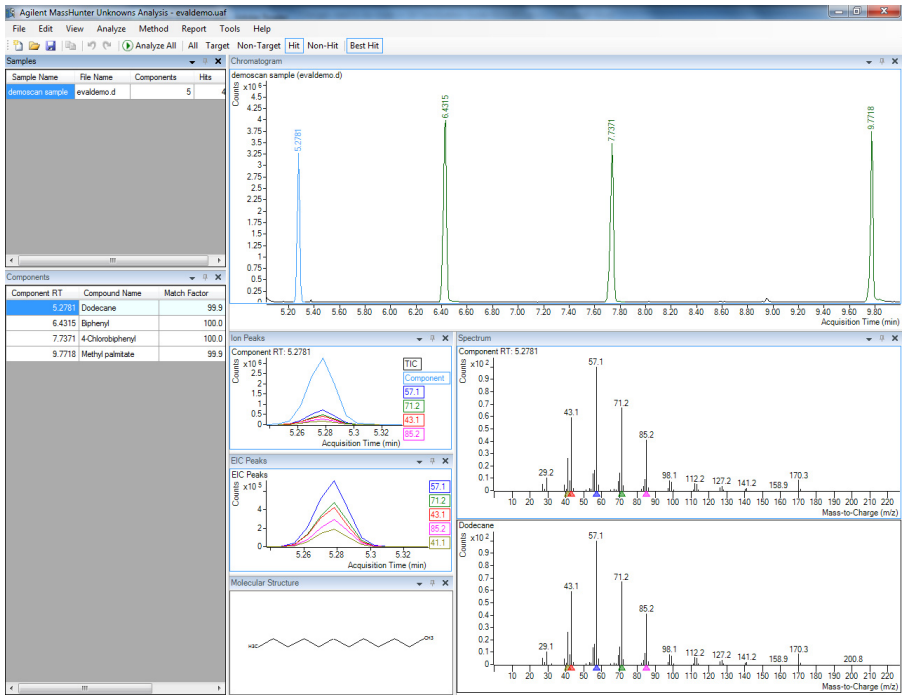


Figure 19.

3 Select the **demoscan sample** from the **Sample** table.

Click one of the following toolbar buttons to view the changes in the **Components** window.

In the **Quant-My-Way** user interface, click one of the following buttons on the **Home** tab.

- **All**: View all the peaks.
- **Hit**: View the peaks that are found in the library search.
- **Non-Hit**: View the peaks that are not found in the library search.

4 Right-click any column header in the **Components** window, and select **Add/Remove Columns**.

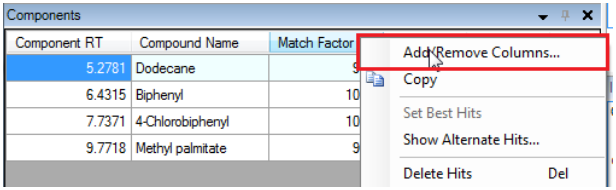


Figure 20.

5 Select **Component** from the **Select columns from** drop-down menu.

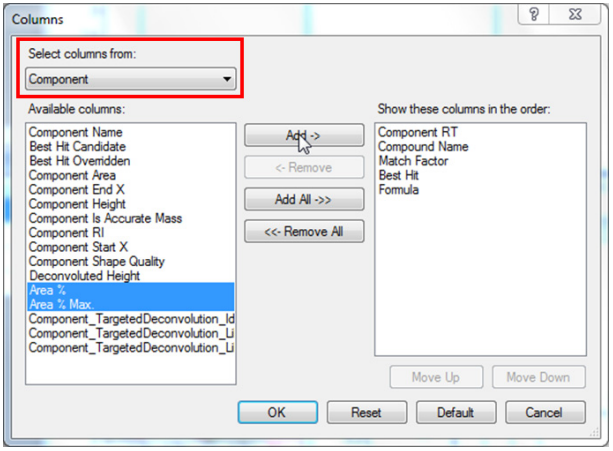


Figure 21.

6 Select **Area %** and **Area % Max** from the **Available columns** list, and click **Add**.

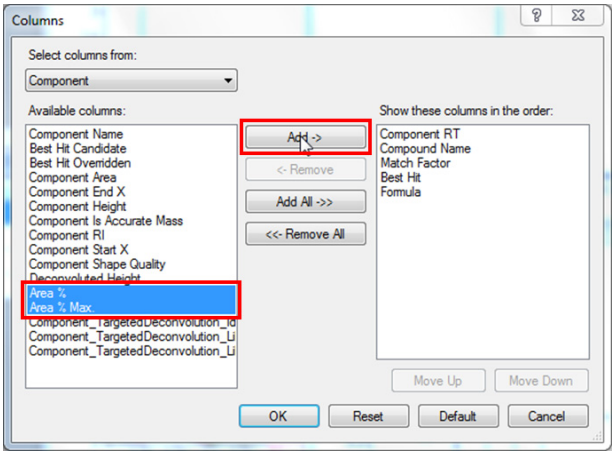


Figure 22.

- **Area %**: Percentage of the peak area sum
- **Area % Max**: Percentage of the largest peak area

7 Verify that the selected columns are moved to the **Show these columns in the order** list, and click **OK**.

8 From the **Components** table, select a component in the **Component RT** column.

Components

Component RT	Compound Name	Match Factor	Best Hit	Formula	Area %	Area % Max.
5.2781	Dodecane	99.9	☑	C12H26	19.158	63.44
6.4315	Biphenyl	100.0	☑	C12H10	30.196	100.00
7.7371	4-Chlorobiphenyl	100.0	☑	C12H9Cl	25.572	84.69
9.7718	Methyl palmitate	99.9	☑	C17H34O2	23.731	78.59

Figure 23.

View the **Chromatogram, Spectrum, Ion Peaks, EIC Peaks**, and **Molecular Structure** for the selected component.

In the **Spectrum** window, the top spectrum is from the component, and the bottom spectrum is from the library. The **Match Factor** in the **Components** table reflects how closely the two spectrum match.

To change to the Header-to-tail view, right-click inside the **Spectrum** window and select **Header-to-tail**.

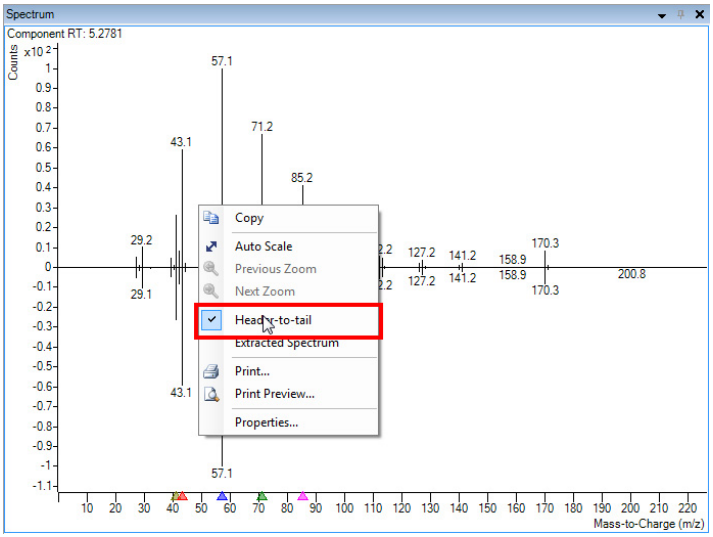


Figure 24.

The **Ion Peaks** and **EIC Peaks** windows show the extracted chromatograms of the selected ions. The EIC traces and their numeric identifiers to the right of the display are color-coded.

To interactively add the ion chromatogram traces in the **Ion Peaks** and **EIC Peaks** window to the display, click in any **Mass Spectral Display** area of the **Spectrum** window. If the selected m/z chromatogram is not already displayed, it will be added to the **Ion Peaks** and **EIC Peaks** window and ▲ symbol of the same color will be at the appropriate m/z position below the x-axis in the **Spectrum** window.

To remove an ion chromatogram trace (and its numeric identifier) from the **Ion Peaks** and **EIC Peaks** window, click on its numeric identifier or on the corresponding m/z value position in the **Spectrum** window.

The **Molecular Structure** is from the library. If the searched library does not contain the structures for the entries, nothing will be displayed in the **Molecular Structure** window.

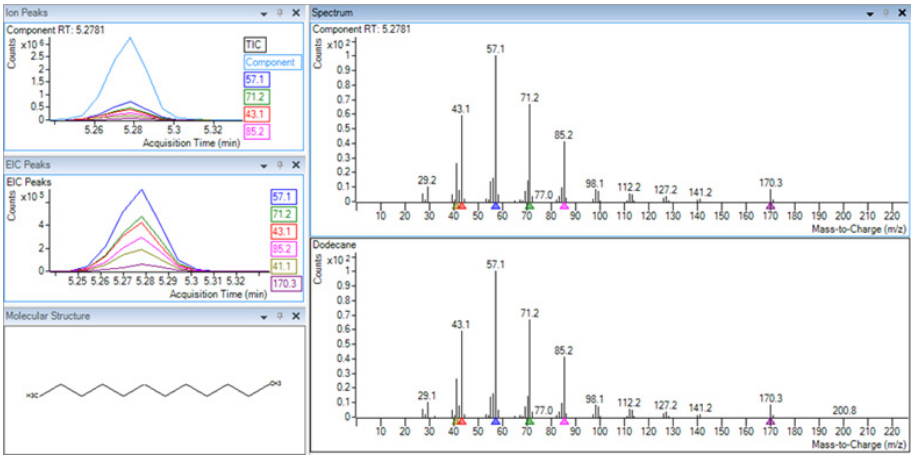


Figure 25.

9 To save the analysis, select **File > Save Analysis**.

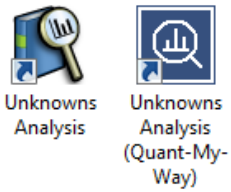
In the **Quant-My-Way** user interface, on the **Home** tab, click **Save Analysis**.

10 . Click **File > Exit**.

Task 2: Identify Compounds with Deconvolution

Create a new analysis

1 Start **Unknowns Analysis** by double-clicking the desktop icon.
or
Click **Start > Agilent > MassHunter Workstation > Unknowns Analysis**.



2 Select **File > New Analysis**.

In the **Quant-My-Way** user interface, on the **Home** tab, click **New Analysis**.

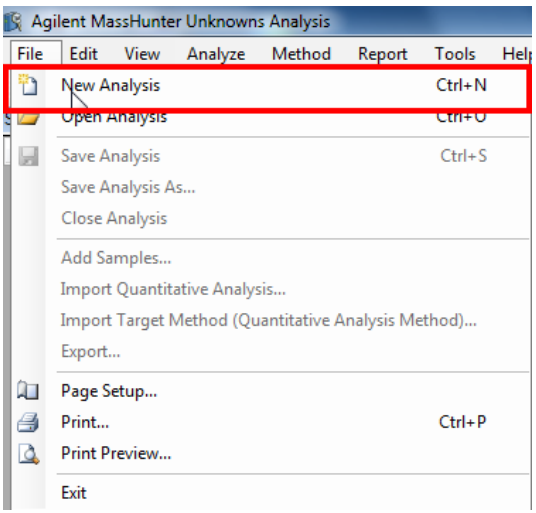


Figure 26.

3 Navigate to **\Your Directory\RI-PEST-MATRIX**.

4 Type the analysis name demo, and click **Create**.

Add samples to the analysis

1 Select **File > Import Quantitative Analysis**.

In the **Quant-My-Way** user interface, on the **Home** tab, select **Import > Import Quantitative Analysis**.

2 Select **TargetDemo.batch.bin**, and click **Open**.

Verify the batch is imported. The **Sample** window now contains one matrix blank and five spiked samples at the different concentration levels. The **Chromatogram** shows the TIC of the sample selected in the **Sample** window.

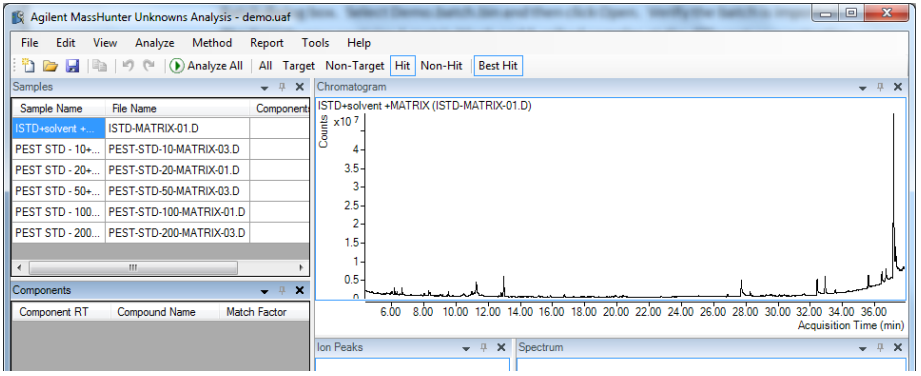


Figure 27.

Set up the method for the analysis

Press **F10** or select **Method > Edit**.

In the **Quant-My-Way** user interface, on the **Home** tab, click **Edit Method**.

Set Peak Detection options

Select **Deconvolution** from the **Peak detection** drop-down menu, and click **Apply to All Samples**.

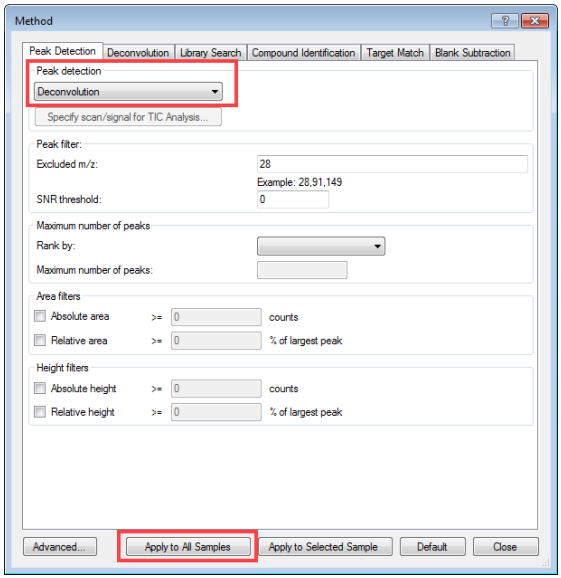


Figure 28.

Set Deconvolution options

1 Click **Deconvolution**.

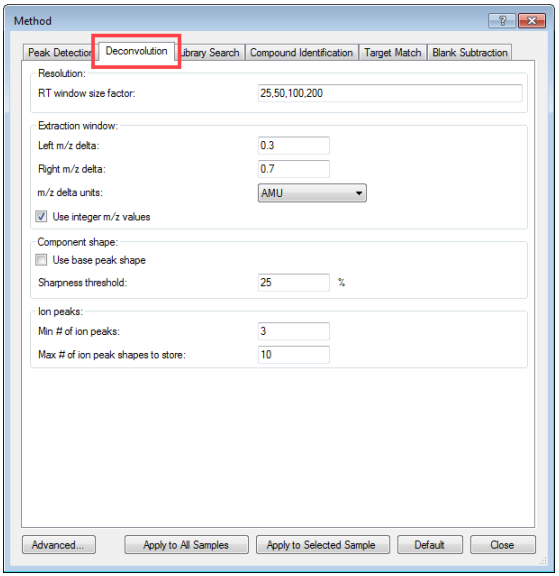


Figure 29.

The default parameters for deconvolution display. By default, there are four values (25, 50, 100, 200) for the **RT window size factor**. Select any set of Window Size Factor (WSF) values in a comma-separated format.

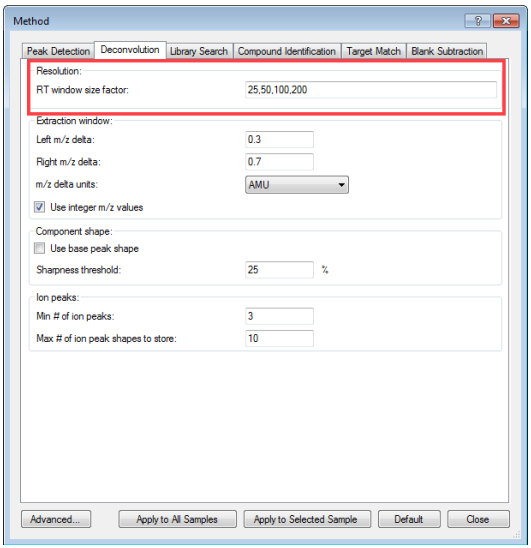


Figure 30.

The WSF represents a dimensionless scale of the correlation window for grouping ion peaks into components, equivalent to Resolution and AMDIS. A smaller value (higher resolution) separates closely spaced peaks, finds more components, and runs longer. A larger value is used for wider peaks. Using multiple values covers all kinds of peaks without manual optimization.

2 In the **Extraction Window** section, select **Use integer m/z values**.

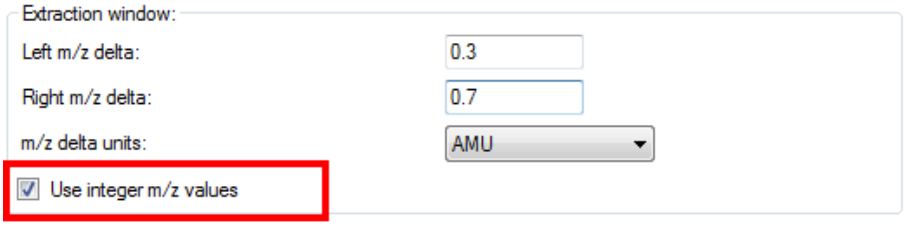


Figure 31.

Use integer m/z values runs the deconvolution with both integer and filtered m/z, and provides the best results.

3 Click **Apply to All Samples**.

Set Library Search options

1 Click **Library Search**.

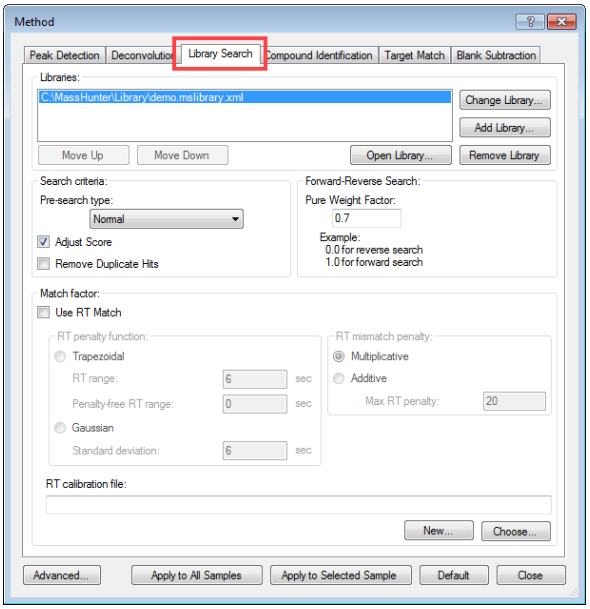


Figure 32.

2 Click **Change Library**.

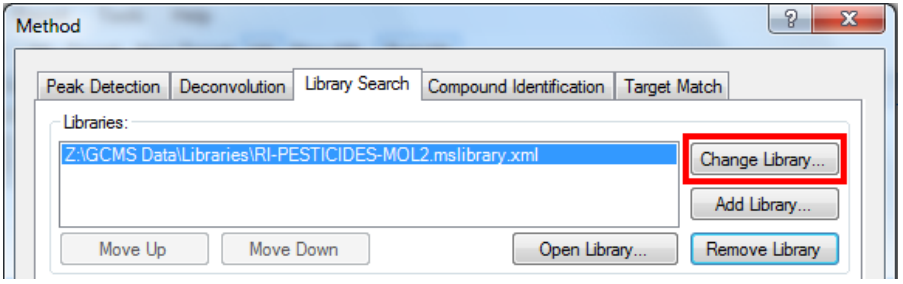


Figure 33.

3 Navigate to the relevant folder, select **RI-PESTICIDESMOL2. mslibrary.xml**, and click **Open**.

4 In the **Search criteria** section, select **Normal** from the **Pre-search type** drop-down menu.

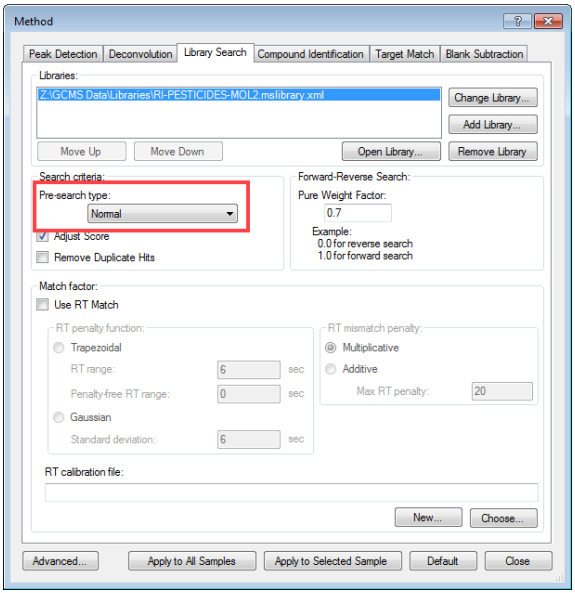


Figure 34.

- Select **Adjust Score** to give the closest library match scores to NIST.
- Select **Remove Duplicate Hits** to remove duplicate hits that appear in the hit list for a given target spectrum. This deals with duplicate and highly similar library entries such as seen in NIST, and only returns the single library entry with the highest fit score.

5 In the **Match factor** section, select **Use RT Match**.

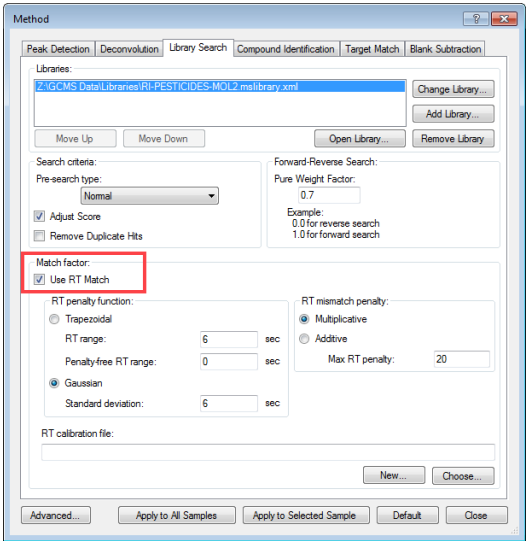


Figure 35.

6 In the **RT penalty function** section, select **Trapezoidal** and enter the following:
RT range: 9
Penalty-free RT range: 9

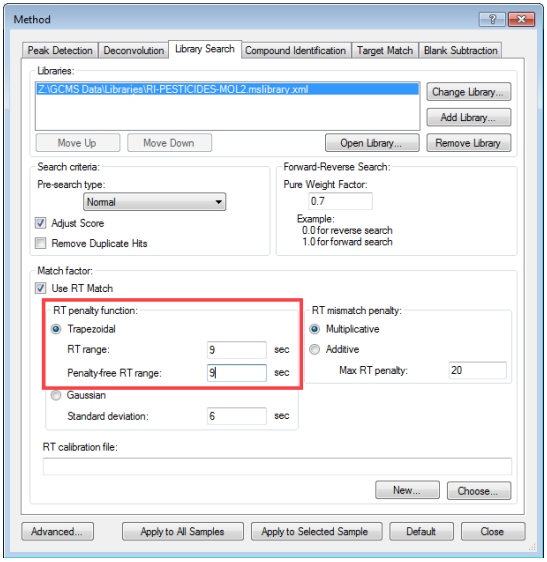


Figure 36.

7 In the RT calibration file section, click **Choose**.

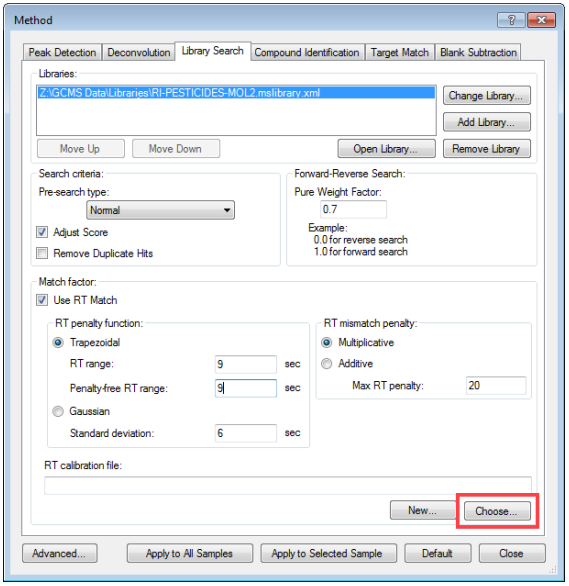


Figure 37.

8 Navigate to the relevant folder, and select **HCs-RTCAL1.rtc**.

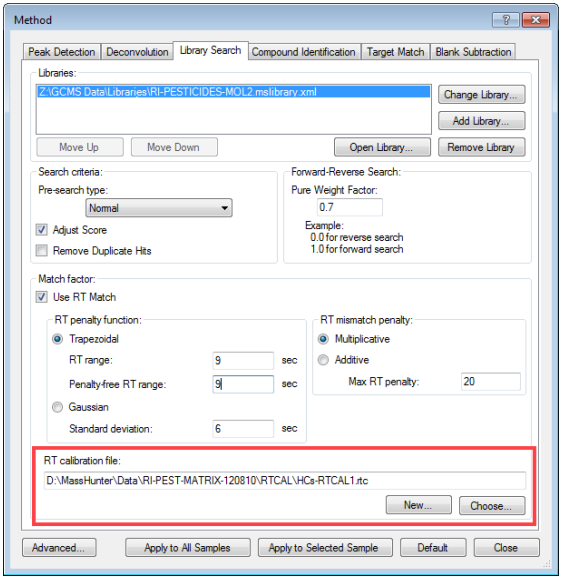


Figure 38.

RT/RI calculation is used with library matching to lower the false positive rate. The window is set to ±9 seconds to qualify the hits from the Library Search.

9 In the **Libraries** section, click **Add Library**.

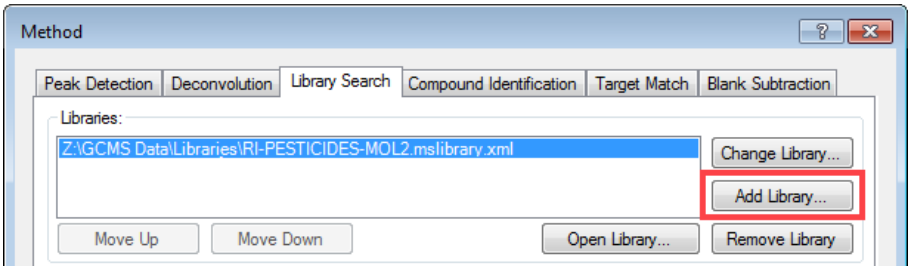


Figure 39.

10 Navigate to the relevant folder, and select **NIST11.L**.

Multiple libraries can be used in Library Search. For this example, the target MS library contains 900+ pesticides with Retention Indexes (RI) information. NIST11.L can be used for the additional confirmation.

11 Select **Fast** from the **Pre-search type** drop-down menu.

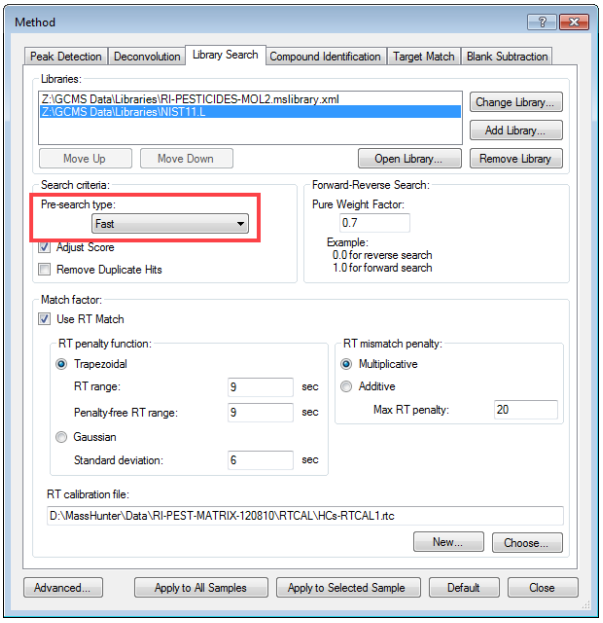


Figure 40.

12 Click **Apply to All Samples**.

You are able to set different Library Search parameters for different libraries.

Set Compound Identification options

Click **Compound Identification**.

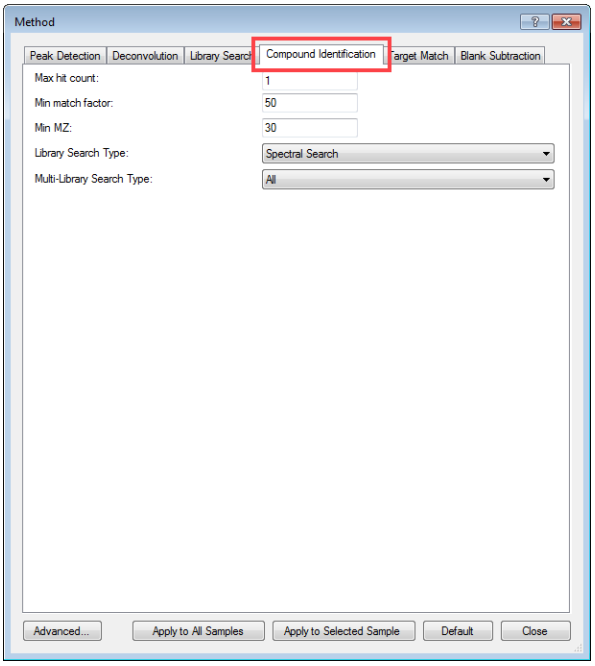


Figure 41.

For this example, the **Min match factor** is set to 50 for the compound identification from the Library Search.

- **Max hit count**: The maximum number of Library Search hits to report per component.
- **Min MZ**: The lower m/z limit for library match score calculation.
- **Library Search Type**: Three search modes are available:
  - Spectral Search: searches Spectral data
  - Retention Time Match: searches GC or LC data
  - Accurate Mass Pattern Match: searches Accurate Mass LCMS data

- **Ratio percent uncertainty**: Only applicable when **Pre-search type** is selected in Library Search. The larger the value, the more Library Search candidates are generated, and the longer the library search process.
- **Multi-Library Search Type**: If multiple libraries were used, two search modes are available:
  - **All**: Search all libraries (default)
  - **StopWhenFound**: Stop searching the library when enough candidates are found

Set Target Match options

1 Click **Target Match**.

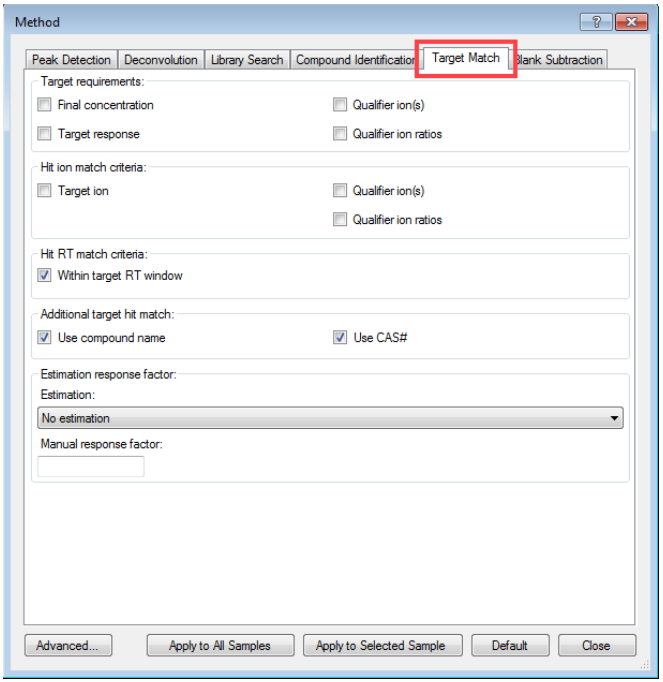


Figure 42.

Target Match identifies quantitation targets using the quantitation method. The goal of identifying non-target compounds is simplified by filtering out the target matches. RT window, compound name, and CAS# can be applied for **Target Match**.

2 In the **Hit RT match criteria** section, select **Within target RT window**.

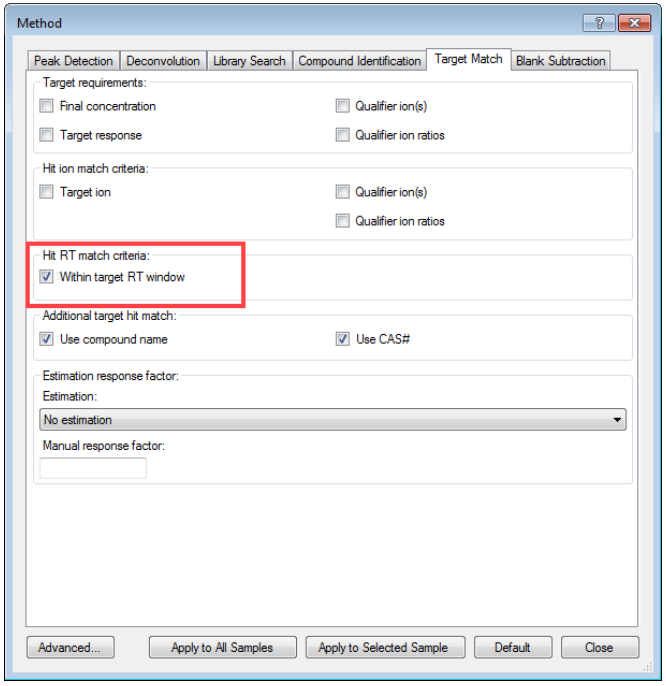


Figure 43.

3 In the **Additional target hit match** section, select **Use compound name** and **Use CAS#**.

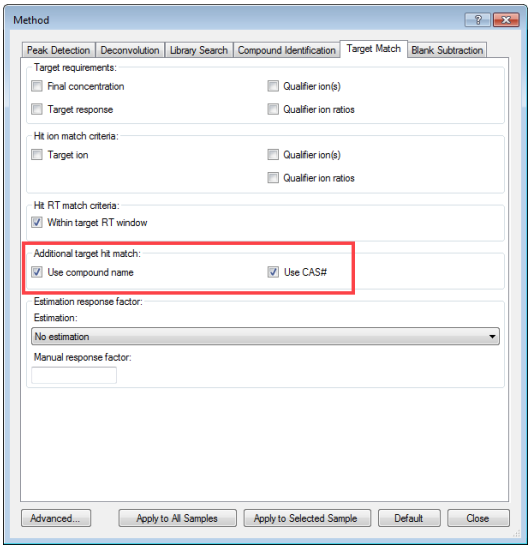


Figure 44.

4 In the **Estimation response factor** section, select **Relative ISTD Estimation** from the **Estimation** drop-down menu.

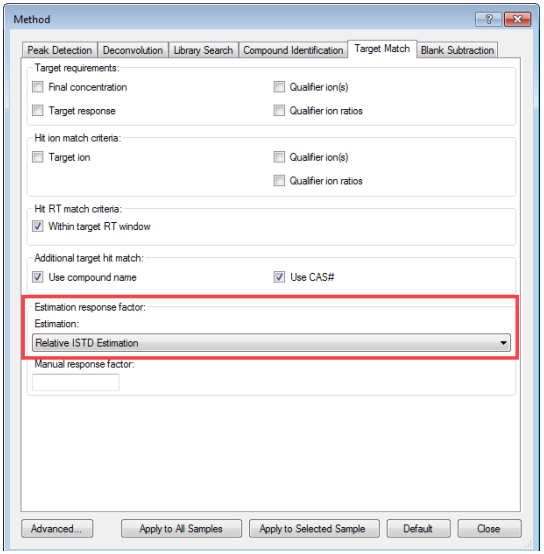


Figure 45.

Concentration estimation leverages the Quant target **Response Factors** (RF), which are applied to Non-Target hits as well. Estimation of response factors is flexible, and can be adjusted to suit the particular analytical requirements.

5 Click **Apply to All Samples**, and then click **Close**.

6 To save the analysis, select **File > Save Analysis**.

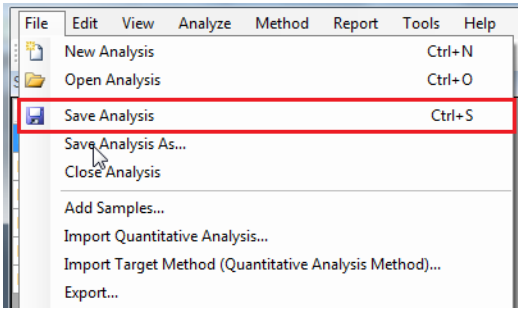


Figure 46.

Set Blank Subtraction options

Click **Blank Subtraction**.

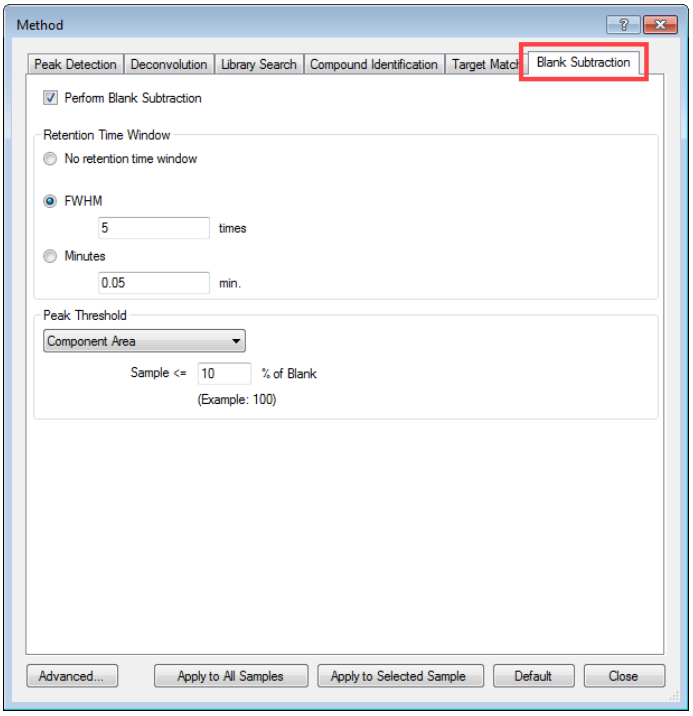


Figure 47.

- **Perform Blank Subtraction**: Select to perform blank subtraction.
- **Retention Time Window**: Select **No retention time window**, **FWHM**, or **Minutes**. If FWHM is selected, specify the number of time. If **Minutes** is selected, specify the number of minutes.
- **Peak Threshold**: Select **None**, **Component Area**, or **Estimated Concentration**. Enter a percentage.

Analyze and review results

1 In the **Sample** window, select the sample **ISTD+solvent+MATRIX**.

2 Select **Analyze > Analyze Sample**.

In the **Quant-My-Way** user interface, on the **Home** tab, select **Selected Samples > Analyze Sample**.

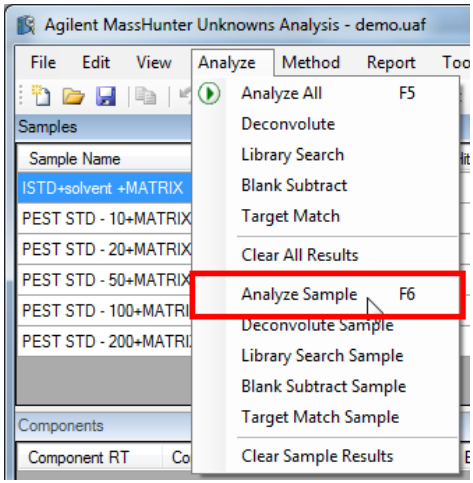


Figure 48.

To analyze the rest of the sample, click **Analyze All**. The analysis starts from where it left off and skips the sample(s) previously analyzed if no parameter in the method has been changed.

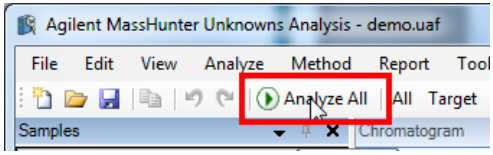
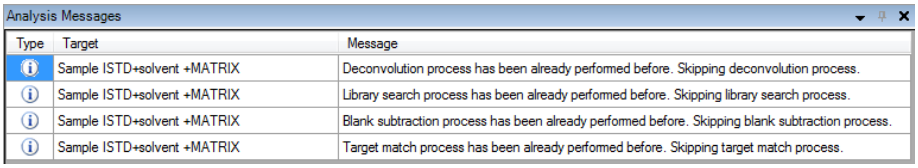


Figure 49.

View validation information in the **Analysis Messages**.



Analysis Messages

Type	Target	Message
ⓘ	Sample ISTD+solvent +MATRIX	Deconvolution process has been already performed before. Skipping deconvolution process.
ⓘ	Sample ISTD+solvent +MATRIX	Library search process has been already performed before. Skipping library search process.
ⓘ	Sample ISTD+solvent +MATRIX	Blank subtraction process has been already performed before. Skipping blank subtraction process.
ⓘ	Sample ISTD+solvent +MATRIX	Target match process has been already performed before. Skipping target match process.

Figure 50.

After the analysis is complete, the main view that appears should look like the example below. This is the default layout and contains the default column settings.

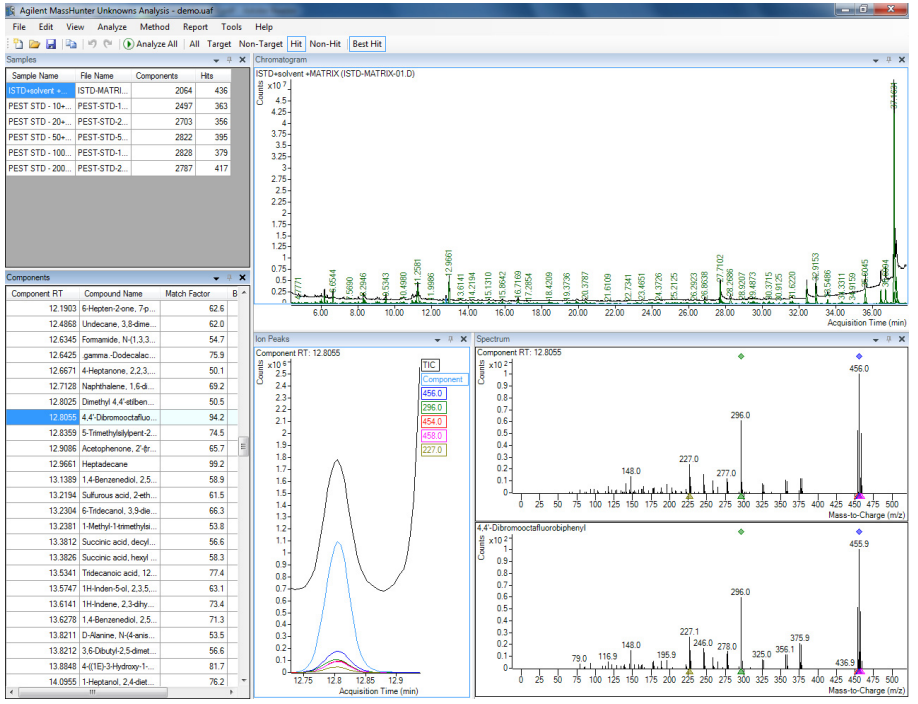


Figure 51.

Review best hit results

1 Right-click any column header in the **Samples** window, and select **Add/Remove Columns**.

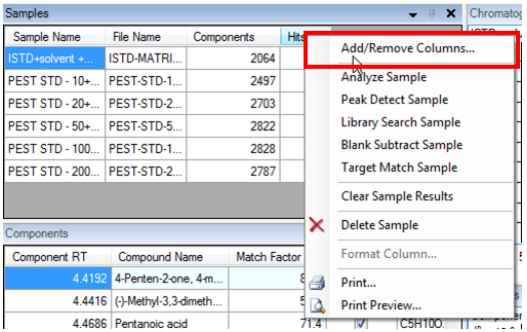


Figure 52.

2 Select **Target Matches** from the **Available columns** list and click **Add**.

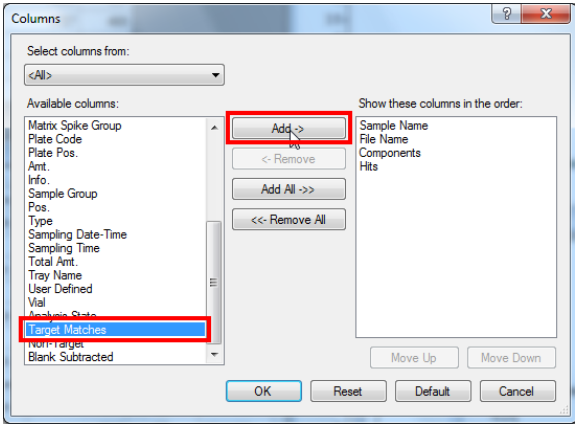


Figure 53.

3 Verify that the selected column is moved to the Show these columns in the order list, and click OK.

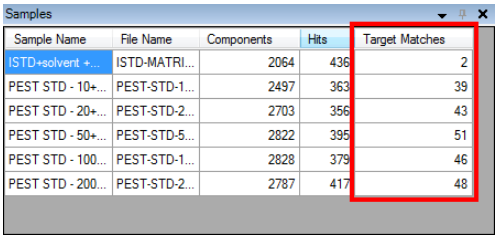


Figure 54.

The Target Matches column is added to the **Samples** window.

4 Select the last sample in the **Samples** window.

Click one of the following toolbar buttons to view the changes in the **Components**, **Chromatogram**, **Ion Peaks**, and **Spectrum** windows.

In the **Quant-My-Way** user interface, click one of the following buttons on the **Home** tab.

- **All**: View all the peaks.
- **Target**: View the peaks that are also in the quantitation method.
- **Non-Target**: View the peaks that are not in the quantitation method.
- **Hit**: View the peaks that are found in the library search.
- **Non-Hit**: View the peaks that are not found in the library search.
- **Best Hit**: View the component with the highest library match score amongthe multiple hits of the same compound from different resolutions.

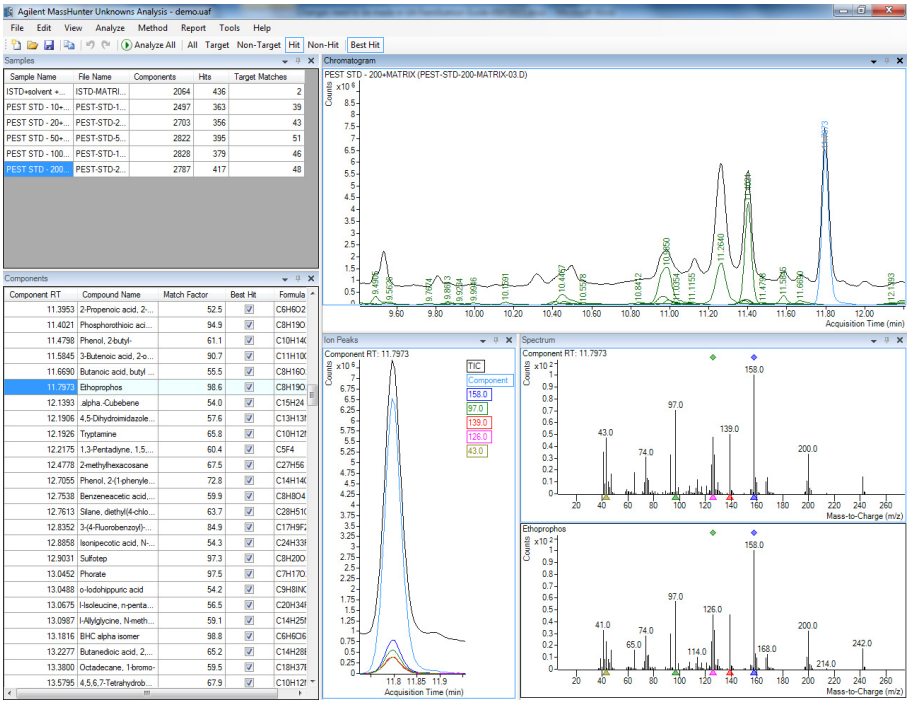

Agilent MassHunter Unknowns Analysis - demo.uaf
File Edit View Analyze Method Report Tools Help
Analyze All All Target Non-Target Hit Non-Hit Best Hit
Samples
Sample Name File Name Components Hits Target Matches
ISTD+solvent +... ISTD-MATRI... 2064 436 2
PEST STD - 10+... PEST-STD-1... 2497 363 39
PEST STD - 20+... PEST-STD-2... 2703 356 43
PEST STD - 50+... PEST-STD-5... 2822 395 51
PEST STD - 100... PEST-STD-1... 2828 379 46
PEST STD - 200... PEST-STD-2... 2787 417 48
Components
Component RT Compound Name Match Factor Best Hit Formula
11.3953 2-Propenoic acid, 2-... 52.5 C6H6O2
11.4021 Phosphorothioic aci... 94.9 C8H19O
11.4798 Phenol, 2-butyl- 61.1 C10H14
11.5845 3-Butenoic acid, 2-o... 90.7 C11H10
11.6690 Butanoic acid, butyl ... 55.5 C8H16O
11.7973 Ethoprophos 98.6 C8H19O
12.1393 alpha.-Cubebene 54.0 C15H24
12.1906 4,5-Dihydroimidazole... 57.6 C13H13
12.1926 Tryptamine 65.8 C10H12
12.2175 1,3-Pentadiyne, 1,5,... 60.4 C5F4
12.4778 2-methylhexacosane 67.5 C27H56
12.7055 Phenol, 2-(1-phenyle... 72.8 C14H14
12.7538 Benzeneacetic acid,... 59.9 C8H8O4
12.7613 Silane, diethyl(4-chlo... 63.7 C28H51
12.8352 3-(4-Fluorobenzoyl)-... 84.9 C17H9F
12.8858 Isonipecotic acid, N-... 54.3 C24H33
12.9031 Sulfotep 97.3 C8H20O
13.0452 Phorate 97.5 C7H17O
13.0488 o-Iodohippuric acid 54.2 C9H8INO
13.0675 l-Isoleucine, n-penta... 56.5 C20H34
13.0987 l-Allylglycine, N-meth... 59.1 C14H25
13.1816 BHC alpha isomer 98.8 C6H6Cl6
13.2277 Butanedioic acid, 2,... 65.2 C14H28
13.3800 Octadecane, 1-bromo- 59.5 C18H37
13.5795 4,5,6,7-Tetrahydrob... 67.9 C10H12
Chromatogram
PEST STD - 200+MATRIX (PEST-STD-200-MATRIX-03.D)
Counts x10 6
Acquisition Time (min)
Ion Peaks
Component RT: 11.7973
TIC
Component
158.0
97.0
139.0
126.0
43.0
Acquisition Time (min)
Spectrum
Component RT: 11.7973
Counts x10 2
158.0
97.0
43.0
139.0
74.0
200.0
Mass-to-Charge (m/z)
Ethoprophos
158.0
97.0
126.0
41.0
74.0
65.0
114.0
200.0
168.0
242.0
214.0
Mass-to-Charge (m/z)

Figure 55.

Review blank hit subtraction results

1 Right-click any column header in the **Samples** window, and select **Add/Remove Columns**.

2 Select **Type** and **Blank Subtracted** from the **Available columns** list, and click **Add**.

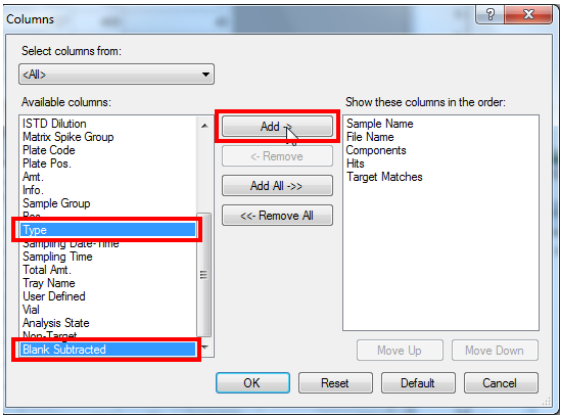


Figure 56.

3 Verify that the selected columns are moved to the **Show these columns in the order** list, and click **OK**.

The **Type** and **Blank Subtracted** columns are added to the **Samples** window.

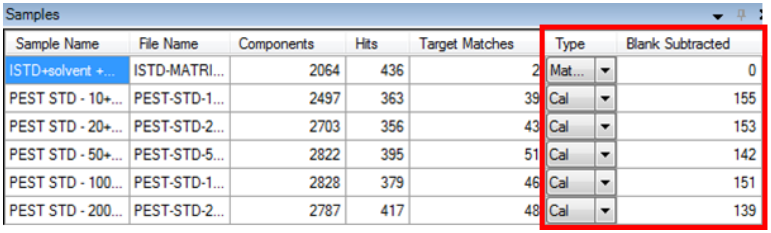


Figure 57.

4 Note the list of available samples in the **Type** drop-down menu.

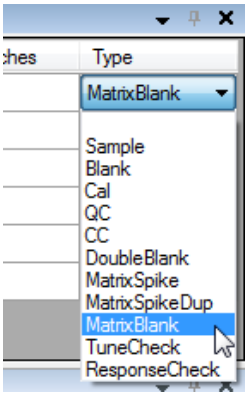


Figure 58.

The values shown in the **Blank Subtracted** column in the **Samples** window represent the number of hits that were blank subtracted from the samples.

Hits in a sample are marked as **Blank Subtracted Hits** when the same hit is found in the blank with RT±5FWHM. FWHM of a typical GC-MS peak is 1-2s. If we use 2s on this estimation, 5FWHM = 10s = 0.17min. You can see the **Blank Subtracted** hits only when you click **All** in the toolbar.

Blank Hit Subtraction is performed against the "blank" sample(s). The hit(s) in any sample(s) with **Sample Type** classified as **Blank**, **DoubleBlank**, or **MatrixBlank** will automatically get subtracted from all the standard samples during the process. You can designate the "blank" sample for blank subtraction purposes by changing the **Sample Type** in the **Sample**. No **Blank Subtraction** happens if there is no "blank" sample(s). Change the sample type to turn off **Blank Subtraction**.

Use Show Alternate Hits to evaluate results

1 Right-click any column header in the **Components** window, and select **Add/Remove Columns**.

2 Select **Library File** from the Available columns list, and click **Add**.

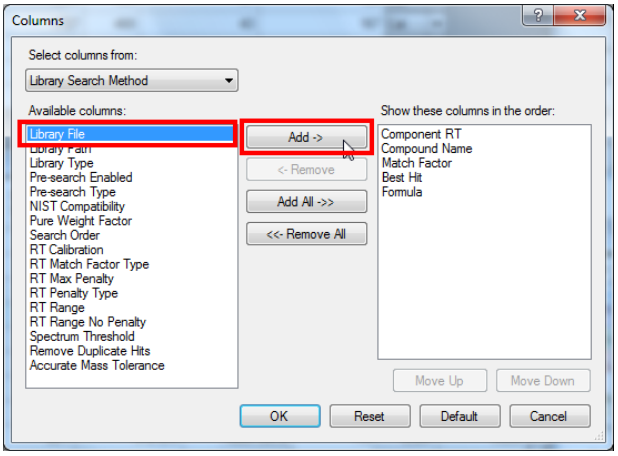


Figure 59.

3 Verify that the selected columns are moved to the **Show these columns in the order** list, and click **OK**.

4 Select the **PEST STD-10+MATRIX** sample in the **Samples** window and click **Hit** in the toolbar to view the changes in the **Components** window.

In the **Quant-My-Way** user interface, on the **Home** tab, click **Hit**.

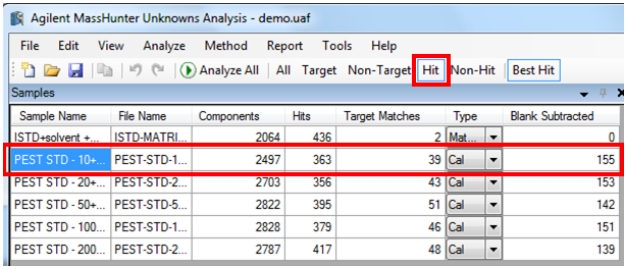


Figure 60.

Verify that the **Best Hits** are from different libraries.

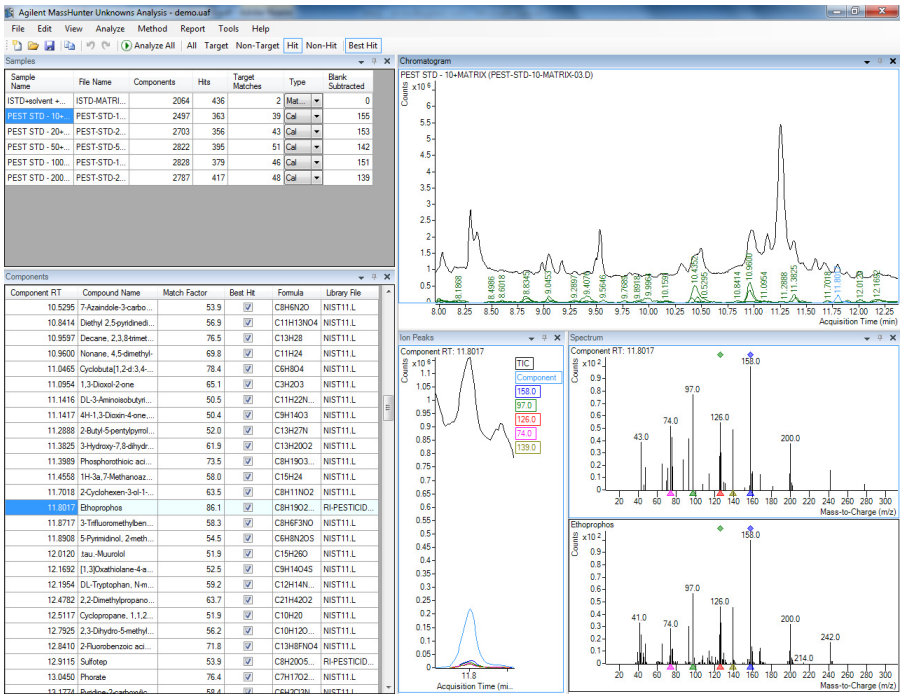


Figure 61.

5 Right-click **Phosphorothioic acid** in the **Compounds** window and select **Show Alternate Hits**.

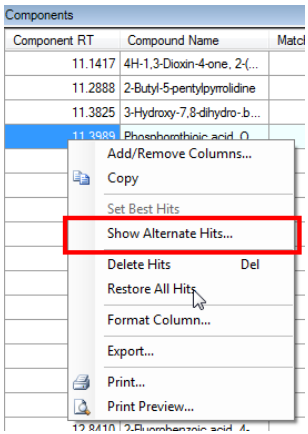


Figure 62.

The **Library Search Results** are displayed.

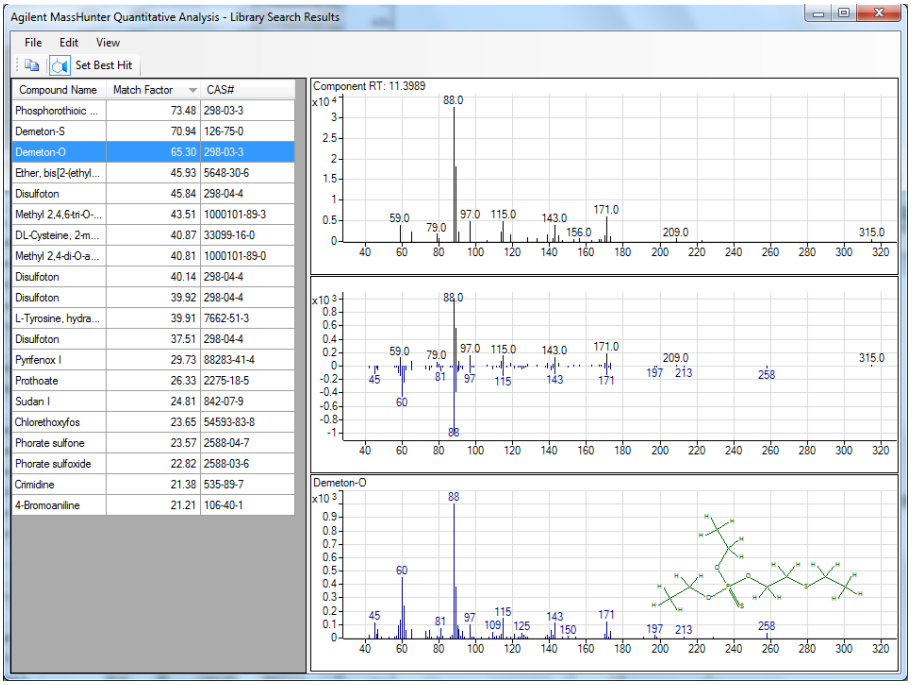


Figure 63.

6 Right-click any column header in the **Library Search Results** window, and select **Add/Remove Columns**.

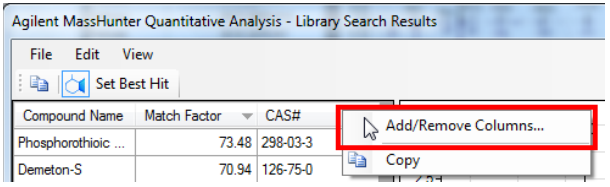


Figure 64.

7 Select **Library** from the **Available Columns** list, and click **Add**.

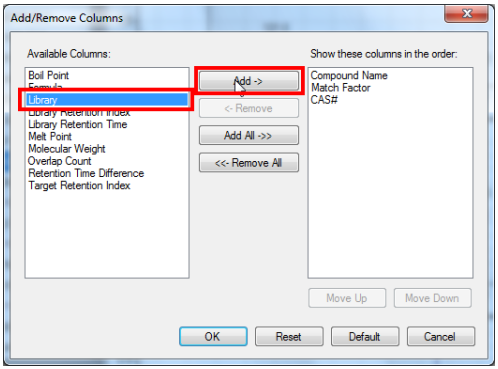


Figure 65.

8 Verify that the selected columns are moved to the **Show these columns in the order** list, and click **OK**.

The **Library** column is added to the table.

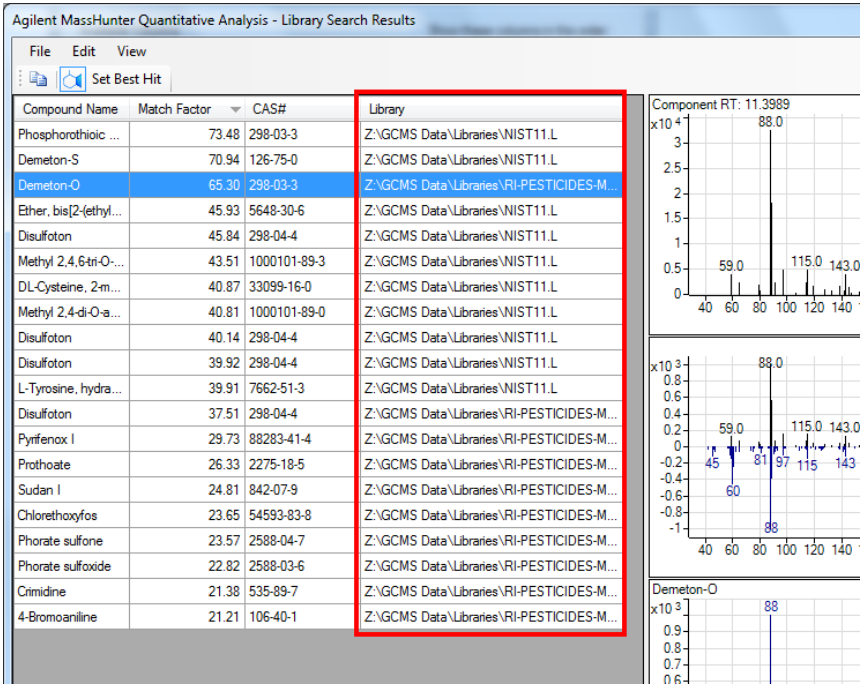


Figure 66.

9 Select **Demeton-0** and click **Set Best Hit**.

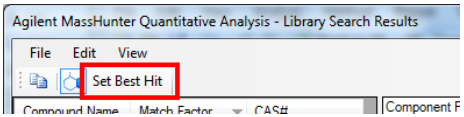


Figure 67.

Verify that the selected compound replaced the previous compound as the current **Best Hit** in the **Component** table.

Components

Component RT	Compound Name	Match Factor	Best Hit	Formula	Library File
10.5295	7-Azaindole-3-carbo...	53.9	☑	C8H6N2O	NIST11.L
10.8414	Diethyl 2,5-pyridinedi...	56.9	☑	C11H13NO4	NIST11.L
10.9597	Decane, 2,3,8-trimet...	76.5	☑	C13H28	NIST11.L
10.9600	Nonane, 4,5-dimethyl-	69.8	☑	C11H24	NIST11.L
11.0465	Cyclobuta[1,2-d:3,4-...	78.4	☑	C6H8O4	NIST11.L
11.0954	1,3-Dioxol-2-one	65.1	☑	C3H2O3	NIST11.L
11.1416	DL-3-Aminoisobutyri...	50.5	☑	C11H22N...	NIST11.L
11.1417	4H-1,3-Dioxin-4-one,...	50.4	☑	C9H14O3	NIST11.L
11.2888	2-Butyl-5-pentylpyrrol...	52.0	☑	C13H27N	NIST11.L
11.3825	3-Hydroxy-7,8-dihydr...	61.9	☑	C13H20O2	NIST11.L
11.3989	Demeton-O	65.3	☑	C8H19O3...	RI-PESTICID...
11.4558	1H-3a,7-Methanoaz...	58.0	☑	C15H24	NIST11.L
11.7018	2-Cyclohexen-3-ol-1-...	63.5	☑	C8H11NO2	NIST11.L
11.8017	Ethoprophos	86.1	☑	C8H19O2...	RI-PESTICID...
11.8717	3-Trifluoromethylben...	58.3	☑	C8H6F3NO	NIST11.L
11.8908	5-Pyrimidinol, 2-meth...	54.5	☑	C6H8N2OS	NIST11.L
12.0120	.tau.-Muurolol	51.9	☑	C15H26O	NIST11.L
12.1692	[1,3]Oxathiolane-4-a...	52.5	☑	C9H14O4S	NIST11.L

Figure 68.

Review concentration estimation results

1 Right-click any column header in the **Components** window, and select **Add/Remove Columns**.

2 Select **Base Peak Deconvoluted Area**, **Response Factor for Estimation**, **Target Multiplier**, **Estimated Conc.**, and **Target Calc. Conc**. from the **Available columns** list, and click **Add**.

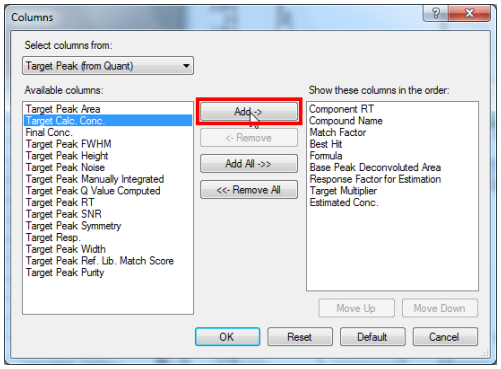


Figure 69.

3 Verify that the selected columns are moved to the **Show these columns in the order** list, and click **OK**.

4 Select the **PEST STD-200+MATRIX** sample in the **Samples** window, and click **Target** in the toolbar to view the changes in the **Components** window.

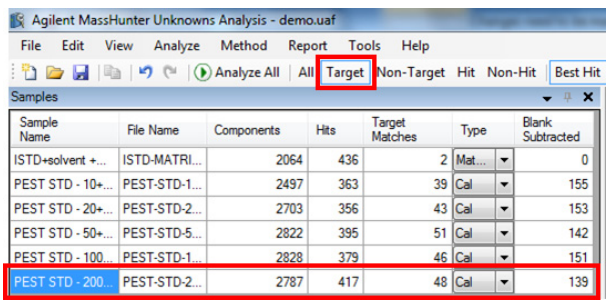


Figure 70.

The estimated concentration results are listed in the **Estimated Conc**. column. For target compounds, you are able to compare with the Quant calculated concentrations.

Estimated Concentration is calculated using the following formula:

$$EstimatedConcentration = \frac{BasePeakDeconvolutedArea}{RFforEstimation} \times Multiplier$$

Components

Component RT	Compound Name	Match Factor	Base Peak Deconvoluted Area	Response Factor for Estimation	Target Multiplier	Estimated Conc.	Target Calc. Conc.
11.4021	Phosphorothioic aci...	94.9	2721304.4	14882.6150	1.0	182.9	144.4
11.7973	Ethoprophos	98.6	2129122.9	23030.7854	1.0	92.45	80.33
12.9031	Sulfotep	97.3	1015154.5	14085.4273	1.0	72.07	60.25
13.0452	Phorate	97.5	3420580.5	36958.8482	1.0	92.55	69.61
13.1816	BHC alpha isomer	98.8	1624103.4	30149.9823	1.0	53.87	50.11
13.7016	Pentachloroanisole	98.8	1731347.2	31680.1152	1.0	49.84	50.17
13.7651	Dimethoate	96.0	2679103.0	27597.5245	1.0	97.08	79.42
14.3218	BHC beta isomer	98.5	1014024.0	16466.2678	1.0	59.17	49.7
14.5841	Lindane	98.0	1123973.2	20441.2941	1.0	54.99	50.04
15.0133	Fonofos	97.4	2705421.8	35661.4284	1.0	75.86	60.31
15.5881	Diazinon	98.2	2028613.6	19208.4327	1.0	98.63	79.78
15.6787	Disulfoton	85.7	1203625.2	14752.9912	1.0	81.59	61.33
15.6842	BHC delta isomer	96.2	1201797.5	22136.8200	1.0	54.29	50.04
17.7267	Methyl parathion	86.9	1539303.6	7893.1866	1.0	108	71.86
17.7336	Chloropyriphos-methyl	91.9	2612062.4	25588.5789	1.0	102.1	81.29
17.9452	Heptachlor	95.7	598014.1	9867.5025	1.0	60.6	50.24

Figure 71.

5 Click **Non-Target** in the toolbar to view the estimated concentrations for Non-Targets.

In the **Quant-My-Way** user interface, on the **Home** tab, click **Non-Target**.

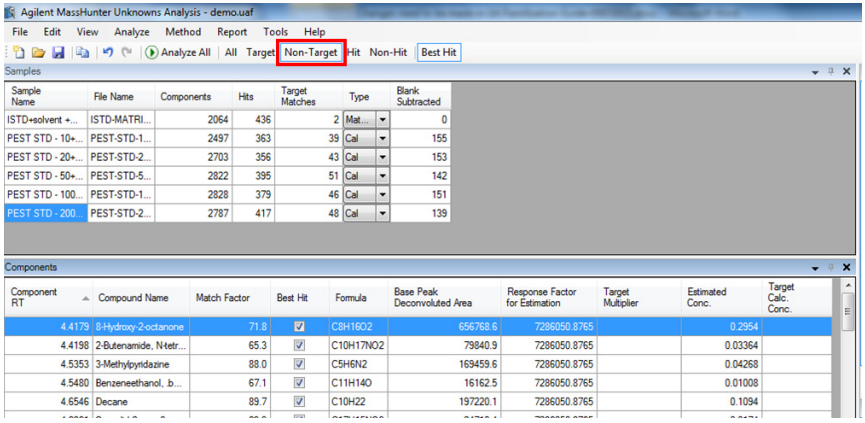


Figure 72.

6 To save the analysis, select **File > Save Analysis**.

In the **Quant-My-Way** user interface, on the **Home** tab, click **Save Analysis**.

Task 3: Generate the Report

1 Select **Report > Generate**.

In the **Quant-My-Way** user interface, on the **Home** tab, click **Generate Report**.

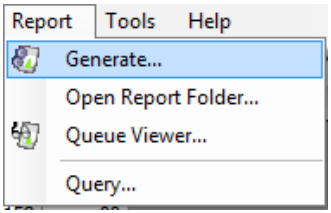


Figure 73.

2 Under **Report method**, click **New**.

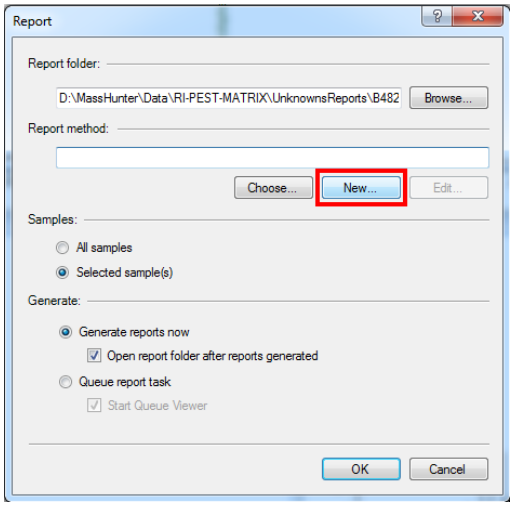


Figure 74.

3 Right-click in the window and select **Add Template**.

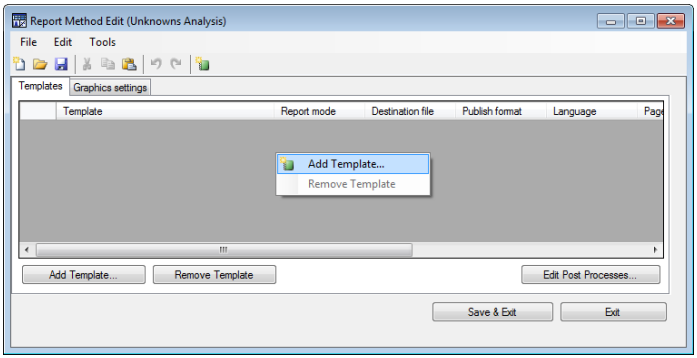


Figure 75.

4 Navigate to **D:\MassHunter\Report Templates\ Quant\PDF-Report-Builder\ Unknowns**, select **LSR_NonTarget_Hits.template.xml**, and click **Open**.

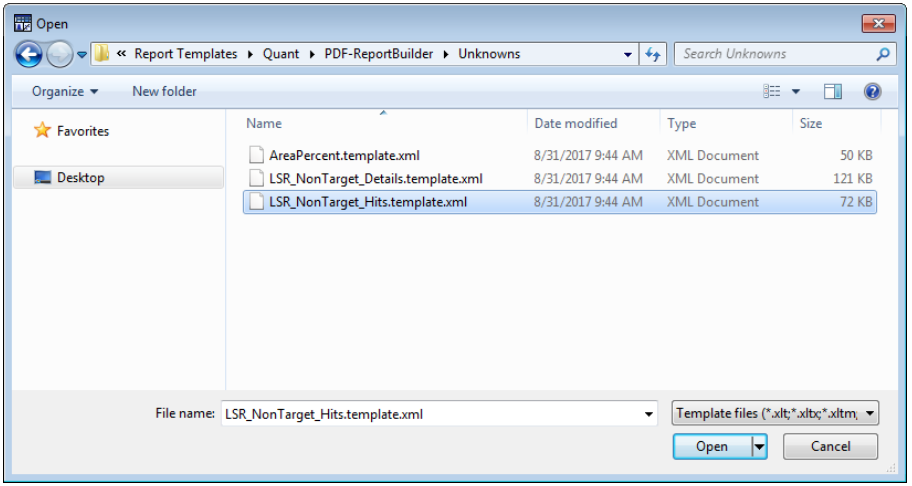


Figure 76.

Once the template(s) is selected, you can configure the **Report Publish Format** with *PDF*, *TEXT*, and *CSV*, **Language** with *English*, *Chinese*, *Japanese*, and *Russian*, **Page Size**, **Printer** with *A4* and *Letter*, and whether or not to **Open published file** after generating the report. The **Post Process** is also available to process the report further after finishing the report task.

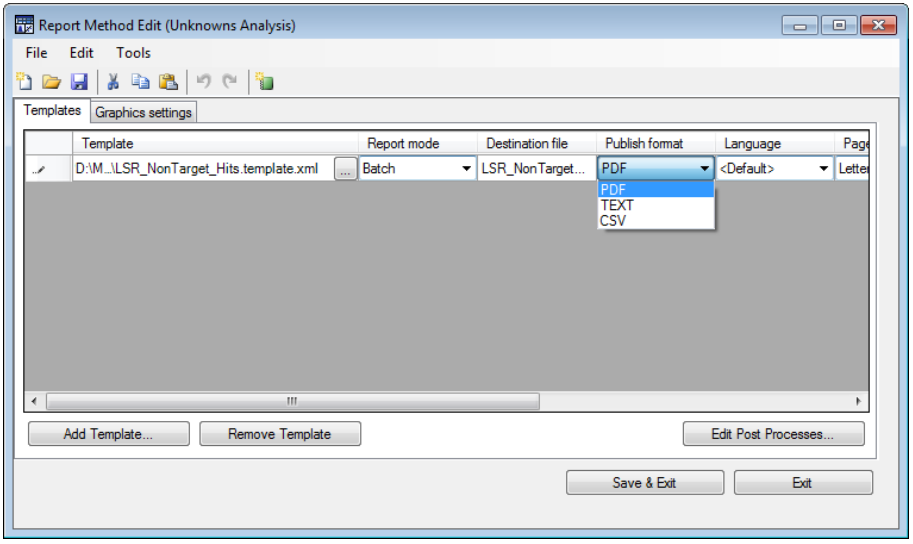


Figure 77.

5 Click **Graphics settings**.

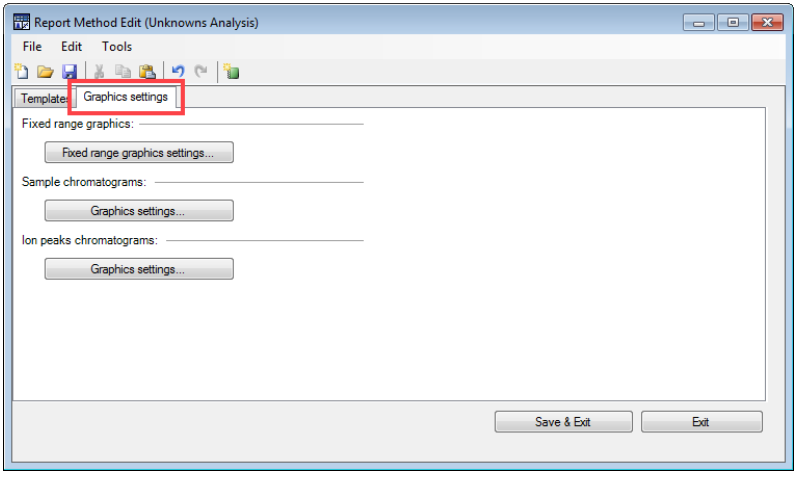


Figure 78.

- Click **Fixed range graphics settings** to manipulate the way to want to present the graphics generated in your report by restricting the scale of your graphs.
- Click **Sample chromatograms: Graphics settings** to adjust the appearance of the sample chromatograms.
- Click **Ion peaks chromatograms: Graphics settings** to adjust the appearance of the ion peaks chromatograms.

6 Click **Save & Exit** to save the Report Method in a desired location.

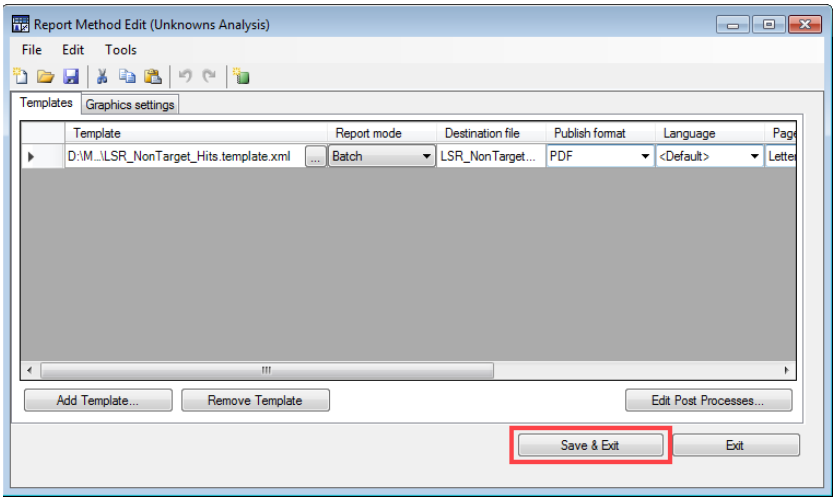


Figure 79.

Report Methods have a **.m** extention.

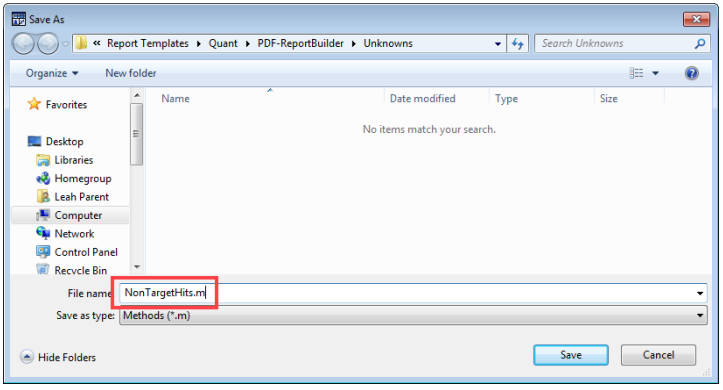


Figure 80.

7 For samples, you can generate a report for **All samples** or the **selected Sample(s)**.

For Report Generating modes, you can select **Generate reports now** or **Queue report task**.

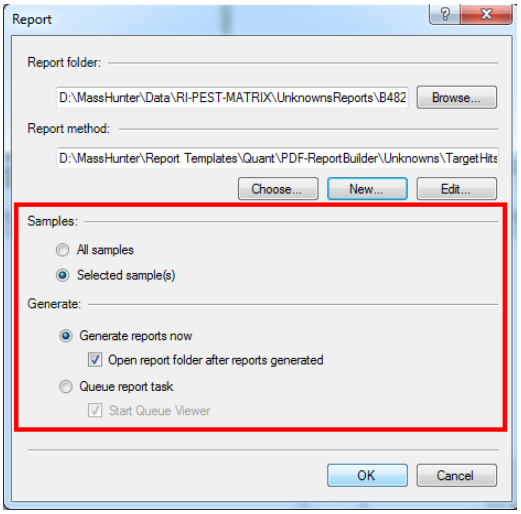
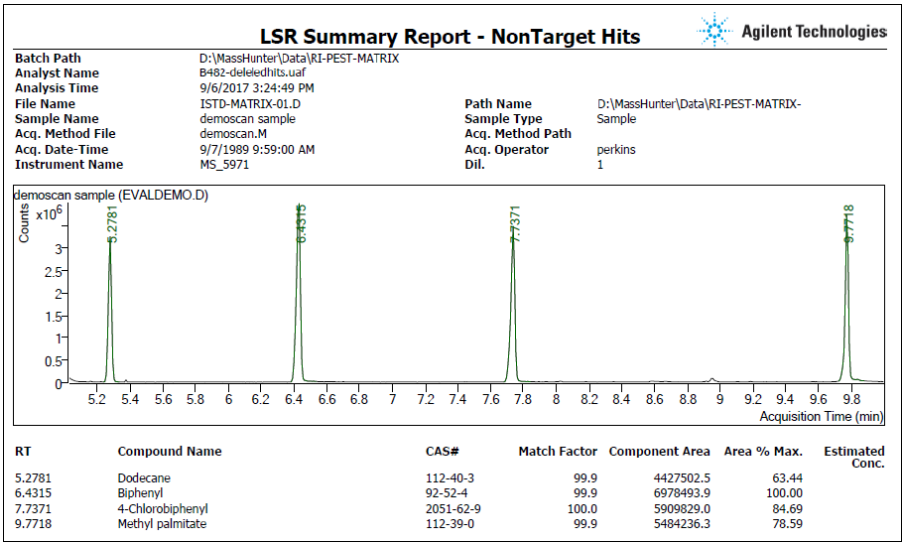


Figure 81.

8 Click **OK** to begin generating reports.

The report folder opens automatically when the report generation is complete.

Alternatively, you can select **Menu > Open Report Folder** to view the newly generated report **LSR_NonTarget_Hits.pdf**. The report opens in Adobe Reader.



LSR Summary Report - NonTarget Hits — Agilent Technologies

Batch Path	D:\MassHunter\Data\RI-PEST-MATRIX		
Analyst Name	B482-deleledhits.uaf		
Analysis Time	9/6/2017 3:24:49 PM		
File Name	ISTD-MATRIX-01.D	**Path Name**	D:\MassHunter\Data\RI-PEST-MATRIX-
Sample Name	demoscan sample	**Sample Type**	Sample
Acq. Method File	demoscan.M	**Acq. Method Path**	
Acq. Date-Time	9/7/1989 9:59:00 AM	**Acq. Operator**	perkins
Instrument Name	MS_5971	**Dil.**	1

RT	Compound Name	CAS#	Match Factor	Component Area	Area % Max.	Estimated Conc.
5.2781	Dodecane	112-40-3	99.9	4427502.5	63.44	
6.4315	Biphenyl	92-52-4	99.9	6978493.9	100.00	
7.7371	4-Chlorobiphenyl	2051-62-9	100.0	5909829.0	84.69	
9.7718	Methyl palmitate	112-39-0	99.9	5484236.3	78.59	

Figure 82.

9 Close the report.

10 To exit the program, select **File > Exit**.

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September 2017
REVISION2 (use for SAP revision if different from REVISION)

G3335-90243 REVISION

Agilent