Pesticide Dynamic MRM Compound Database for Screening and Identification Using the Agilent Triple Quadrupole LC/MS Systems

Technical Note

Introduction

Over the last century more than 1000 pesticides have been in common use for crop protection. Beyond approved and recommended usage, there always exists the possibility that any of these chemicals can be found in the environment and make their way into the food supply. To protect both the environment and human health, detection and identification in survey type monitoring is very important. Liquid chromatography/tandem mass spectrometry with a triple quadrupole LC/MS meets this need by providing the most sensitive and highly selective detection in complex samples. The system must be run in multiple reaction monitoring (MRM) mode, in order to obtain the maximum sensitivity and selectivity from this technology.

Although the triple quadrupole LC/MS is the most sensitive method for multi-residue analysis, the technique can only detect the pesticides that have been included in the methodology. Each pesticide must contain its predetermined precursor ion and an indicative product ion. This single precursor/product ion pair or transition is required for screening in the MRM mode. For confirmation of a compound at least two transitions must be included in the method so that their presence and correct ratio in a sample can be determined along with the correct chromatographic retention time. Because every compound is different there are specific instrument conditions that will provide an appropriate response for each transition. On the Agilent systems this includes both fragmentor voltage, optimizing transmission of the precursor ion into the mass spectrometer, and the collision energy, optimizing the maximum intensity for a specific product ion. Excellent results are obtained using all other mass spectrometer settings provided by the system's Autotune program.

A powerful tool called MassHunter Optimizer has been added to the Agilent 6400 Series triple quadrupole LC/MS systems. This software allows automated optimization of compound specific parameters, and it is within this tool that the Pesticide Database operates. Any compound that the user optimizes can be saved to a Project or to a Database. Agilent has created a pesticide database containing the operating parameters for over 700 pesticides. The Pesticide Database is read-only and can be saved to any name for customization by re-optimization of compounds in

the database or addition/deletion of those present. Presently, modifications can be done only in MassHunter Optimizer or by saving method conditions in MassHunter Acquisition, not in the database browser. The power of the database is due to the fact that the conditions included will provide good results without the user needing to optimize each and every compound. In addition, methods are included that now contain retention times for Dynamic MRM [1,2] acquisition. For analysts needing customized methods with hundreds of pesticides per method, the database will allow fast startup and provide good results without the need to optimize the compounds in the database. This does not negate the need to run standards and validate results with good QC/QA procedures. Compounds that the user needs to analyze but are not in the database will need to be optimized. This is readily facilitated by MassHunter Optimizer, and the compounds can then be added to the user's customized database.

Description

The Pesticide Dynamic MRM Database requires MassHunter Acquisition and MassHunter Optimizer 3.01 or later. The link to the database is from the MassHunter Acquisition software or MassHunter Optimizer. It is here that the user can import selected compounds to rapidly develop a customized method, which meets the analytical needs of a specific analysis. This can include compounds in the supplied Pesticide MRM Database and those added by the user, importing to a custom designed chromatographic method for a specific matrix, or a host of other needs for customization. Figure 1 is a screen capture of the MS QQQ Acquisition setup tab for controlling any of the Agilent 6400 Series triple quadrupole LC/MS systems. Right-clicking on the white or grey area of the "Scan Segments" section of the screen displays the pull-down menu where "Import from optimizer" is accessed. When this is selected, the database browser is opened with the default database. The Agilent pesticide database or a customized version of it can be made the default (please note again that the supplied database is read-only). The user can then select the compounds and product ions to import into the acquisition method of his or her choice. The user can also save the compounds in the method to the database with the retention times used in Dynamic MRM.

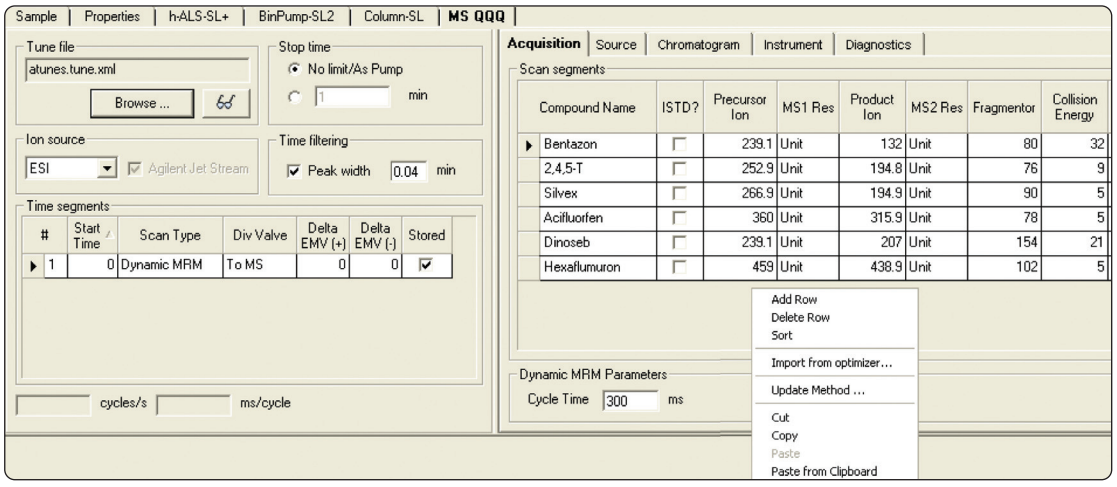


Figure 1. MS QQQ Acquisition tab of MassHunter showing the "Import from optimizer" function.

Once in the Database Browser the Pesticide Database can be opened. Figure 2 shows the Database Browser with the Pesticide Database loaded. The pull-down menu at the top left of the screen in Figure 2 or the Database Save icon allows the user to save the database to a new name. This is a necessary step for changes to be made because the Pesticide Dynamic MRM Database is read-only. Compounds can be deleted from the database in the Database Browser, but they cannot be added. That must be done from MassHunter Optimizer (see below). The copied database can now be customized and set to the default. The database contains the compound name, its formula, the nominal monoisotopic mass (nominal mass plus one decimal place where the second is not significant) of the compound, and the method(s) that were used for analyses. The parameters for analysis include the precursor that gave the optimal signal and its associated fragmentor voltage, at least two product ions (if the compound did produce two significant product ions), and the optimized collision energy for each product ion. In addition, the abundance and response factor of each ion is shown so that the user can distinguish between the quantitation ion and the qualifier by their response. In addition, acquisition methods and retention times with retention time windows are given for Dynamic MRM. The user may select a method and import compounds and their associated retention times with that method. The LC portion of this method must be used or the retention times become invalid. If not all the compounds desired are found in a specific method, the user must find the retention times for those compounds using the desired method and then import the other compounds from the database with the same method. The method must also use the same LC configuration used in the database method. Therefore, if a user has a 1200 SL pump, a method using the Agilent Infinity 1290 LC should not be used without expecting to modify the retention times. If only some of the compounds in a method or compounds from various methods are desired, they can be added to the "Import List" as shown in Figure 3. This shopping cart of compounds allows mixing sources of the compounds found by the various search filters provided (Figure 2).

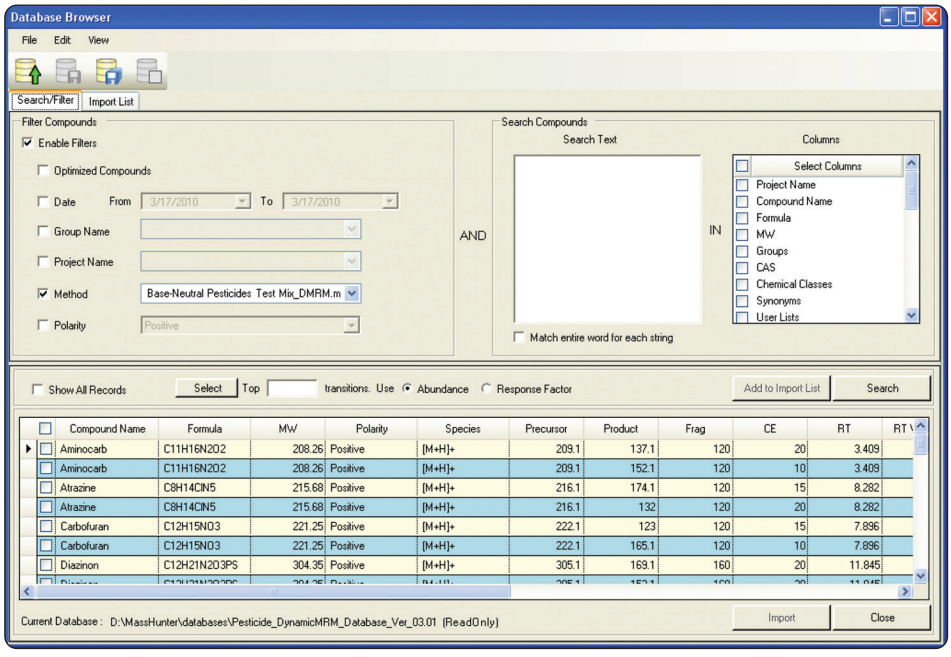
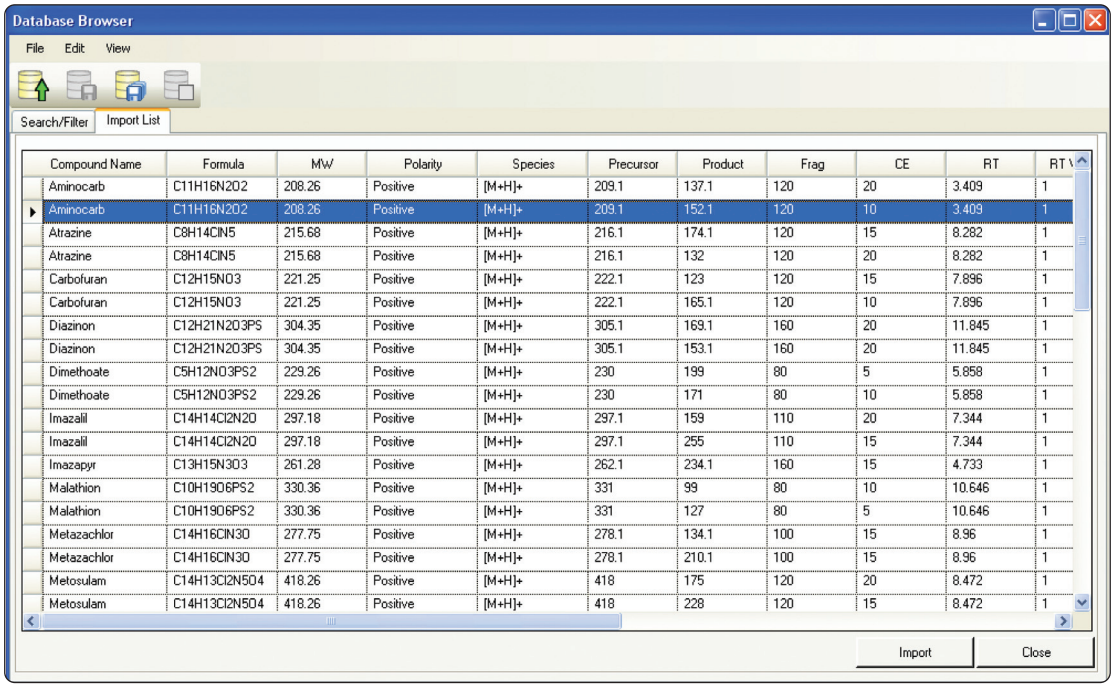


Figure 2. Database Browser view of Pesticide Database.

A good way to search for compounds is to make a list in Excel and copy that list to the search text window (Figure 3). However, the search filter desired MUST be checked. For example, if the user has a list of compound names, then the compound name box must be checked. Likewise if the list is CAS numbers then the "CAS" box must be checked. Selected compounds can be added to the Import list and removed from that list if necessary. However the database itself cannot be edited from the database browser, even in a user-created database.



Compound Name	Formula	MW	Polarity	Species	Precursor	Product	Frag	CE	RT	RT
Aminocarb	C11H16N2O2	208.26	Positive	[M+H]+	209.1	137.1	120	20	3.409	1
Aminocarb	C11H16N2O2	208.26	Positive	[M+H]+	209.1	152.1	120	10	3.409	1
Atrazine	C8H14ClN5	215.68	Positive	[M+H]+	216.1	174.1	120	15	8.282	1
Atrazine	C8H14ClN5	215.68	Positive	[M+H]+	216.1	132	120	20	8.282	1
Carbofuran	C12H15NO3	221.25	Positive	[M+H]+	222.1	123	120	15	7.896	1
Carbofuran	C12H15NO3	221.25	Positive	[M+H]+	222.1	165.1	120	10	7.896	1
Diazinon	C12H21N2O3PS	304.35	Positive	[M+H]+	305.1	169.1	160	20	11.845	1
Diazinon	C12H21N2O3PS	304.35	Positive	[M+H]+	305.1	153.1	160	20	11.845	1
Dimethoate	C5H12NO3PS2	229.26	Positive	[M+H]+	230	199	80	5	5.858	1
Dimethoate	C5H12NO3PS2	229.26	Positive	[M+H]+	230	171	80	10	5.858	1
Imazalil	C14H14Cl2N2O	297.18	Positive	[M+H]+	297.1	159	110	20	7.344	1
Imazalil	C14H14Cl2N2O	297.18	Positive	[M+H]+	297.1	255	110	15	7.344	1
Imazapyr	C13H15N3O3	261.28	Positive	[M+H]+	262.1	234.1	160	15	4.733	1
Malathion	C10H19O6PS2	330.36	Positive	[M+H]+	331	99	80	10	10.646	1
Malathion	C10H19O6PS2	330.36	Positive	[M+H]+	331	127	80	5	10.646	1
Metazachlor	C14H16ClN3O	277.75	Positive	[M+H]+	278.1	134.1	100	15	8.96	1
Metazachlor	C14H16ClN3O	277.75	Positive	[M+H]+	278.1	210.1	100	15	8.96	1
Metosulam	C14H13Cl2N5O4	418.26	Positive	[M+H]+	418	175	120	20	8.472	1
Metosulam	C14H13Cl2N5O4	418.26	Positive	[M+H]+	418	228	120	15	8.472	1

Figure 3. Import list where compounds selected from different search filters can be added to a Dynamic MRM method.

Finally, the other access point for the database is from MassHunter Optimizer program. It is from this access that the user can add compounds to their customized database. Figure 4 shows the initial screen for the MassHunter Optimizer. The icons across the top allow import and export to and from Excel, naming and saving projects and compounds, starting an optimization or breakdown profile, and access to the databases. The circled icon in Figure 4 allows the import from an acquisition method. Figure 5 with "show results" from an import demonstrates that not only the operating parameters, but the retention times and the retention time windows are imported from the method. By selecting the icon circled in Figure 5 the user can then export this to the default database (their custom database). Again, the Pesticide Dynamic MRM database is read-only but once accessed it can be saved as a customized database and then set as the default. As the default, compounds and projects can be saved from MassHunter Optimizer to the database. Compounds can be deleted directly from the Database Browser but can only be added or changed using MassHunter Optimizer. It will be useful for the user to save compounds not in the provided database and at times to re-optimize certain compounds for specific user conditions.

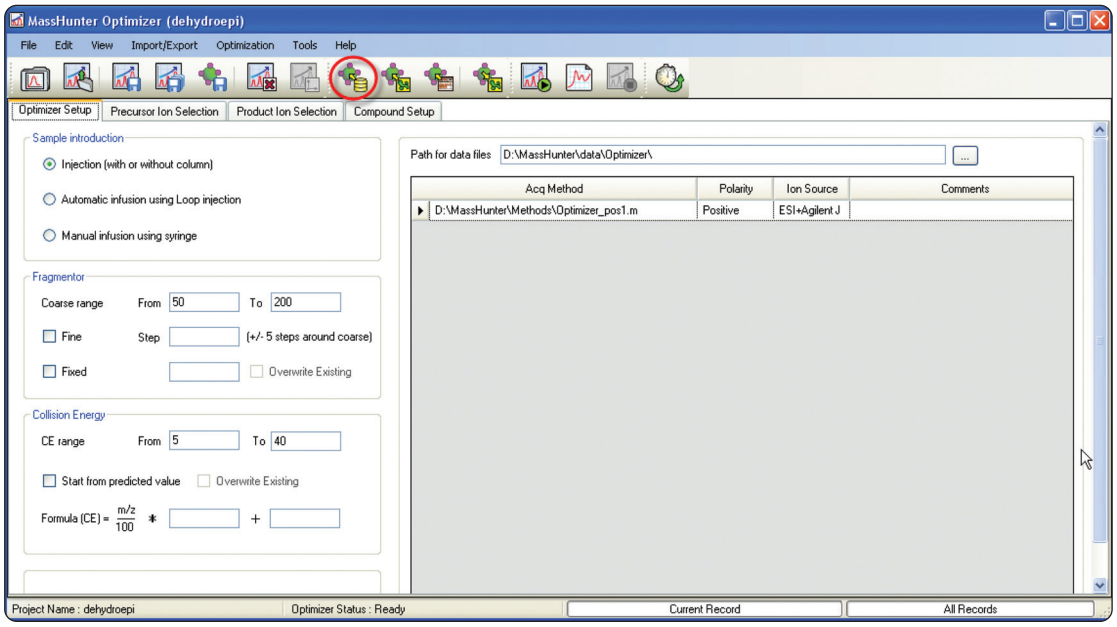


Figure 4. First tab of MassHunter Optimizer where parameter range and method settings are made. Note icon "Import from Database" provides access to Database Browser.

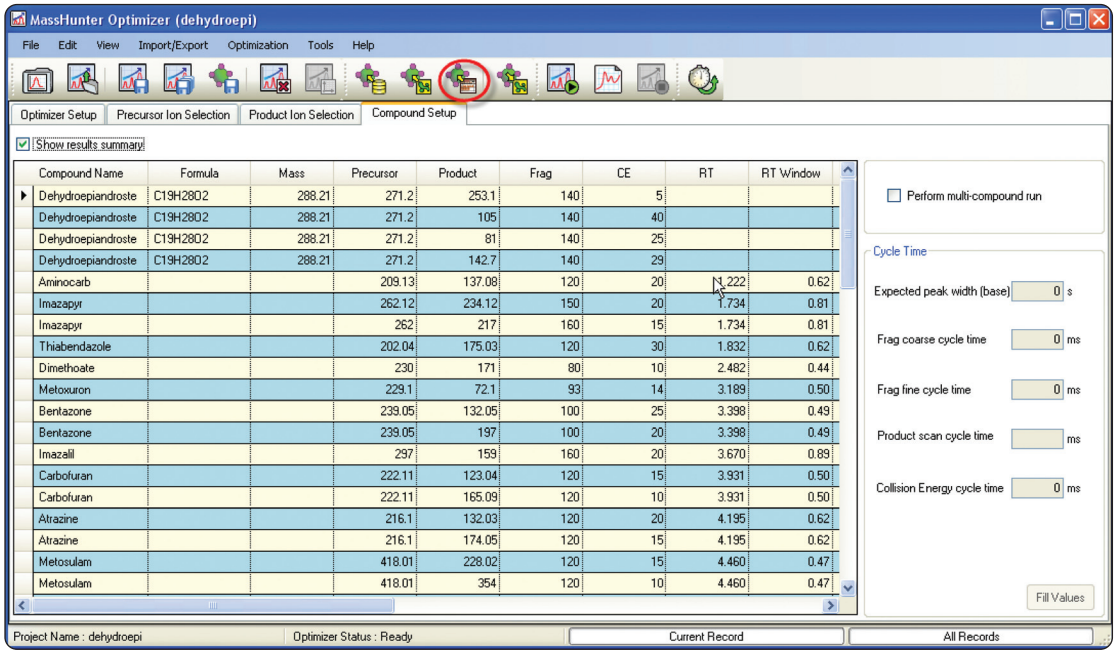


Figure 5. Import from an Acquisition method with "Show Results" displayed. The circled icon allows export of all compound information to the default database.

Summary

The Dynamic MRM Pesticide Database provides over 700 compounds with specific parameters for all Agilent 6400 Series triple quadrupole LC/MS systems. It is designed to meet the needs of laboratories analyzing hundreds of pesticides in one analysis. It allows re-optimization of compounds through the MassHunter Optimizer program and incorporation of the compounds into data acquisition methods for multi-residue analysis where Dynamic MRM is most useful. Its benefits to the analyst are:

- Fast method development with compound-specific parameters for hundreds of compounds
- Storage and retrieval of compounds added to the supplied database
- Customization to meet specific needs of laboratories and their analyses

The database and its functionality will continue to evolve to provide greater search and retrieval capabilities and faster development for Dynamic MRM methods.

For More Information

The Dynamic MRM Pesticide Database is included in only the Pesticide Application kit (Agilent part number, G1733AA). Details of how to use the database are given in the Quick Start Guide that is included in the application kit.

For more information on our products and services, visit our Web site at www.agilent.com/chem.

www.agilent.com/chem

Printed in the USA
April 28, 2010
5990-4255EN

Agilent Technologies