# Building and Editing RTL Screener/Quant Databases and Libraries

**Technical Overview** 



## Author

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# Introduction

The techniques of Retention Time Locking (RTL) [1], Target Compound Screening [2], and Deconvolution Reporting [3] all use retention times (RTs) for an additional level of compound confirmation. It is important that for maximum productivity and quality, RTs are constant/reproducible and embedded in mass spectral libraries [4]. However, not all compounds are in a commercially available mass spectral library, and most mass spectral libraries do not have RTs.

This technical overview describes building and editing (for example, adding compounds) the following:

RTL Screener databases, RTL Quant databases, and Mass Spectral Libraries. These tasks are accomplished using the GC/MSD ChemStation software and Microsoft Excel. RTL Quant databases and Mass Spectral Libraries can also be used with Agilent's Deconvolution Reporting Software [3].

# Software Requirements

- GC/MSD revision D.01.01 (D.01.00 sp1) or higher
- Microsoft Excel

# File Locations (File types: \*.d, \*.l, \*.scd, and \*.tab)

- Data files (\*.d) in C:\MSDchem\1\data\
- Library files (\*.l) in C:\Database\
- Screener database files (\*.scd) in C:\Database
- TAB-delimited files (\*.tab) in C:\Database



# **Building an MS Spectral Library**

1. Build a (Screener) Library from datafiles (\*.d) and a .tab file. See Table 1. The \*.d files consist of the peaks/spectra of the library entries.

Table 1. Explanation of Columns in .tab File

Column	Header
А	open (column A is used in GC for retention time (RT), not used in MSD)
В	name (compound name, for example dimethyl)
C	cas (CAS number without dashes, for example, 12345, not 00012-34-5)
D	molecular formula (for example, C6H14N2O, no commas, dashes, quotes etc)
E	molecular weight (for example, 32.04)
F	RT (GC/MSD, in minutes, for example, 12.34)
G	open (not used)
Н	company ID (a unique identifier, for example, cmpd01 - more explanation on this later)
I	file name (complete file path for .d used for library creation such as C:\MSDchem\1\data\oxymix.d)
J	target ion (scd target ion)
К	q1 (qualifier 1)
L	q1ratio (qualifier 1 ratio)
Μ	q1 unc (qualifier 1 uncertainty)
Ν	q2 (qualifier 2)
0	q2ratio (qualifier 2 ratio)
Р	q2 unc (qualifier 2 uncertainty)
Q	q3 (qualifier 3)
R	q3ratio (qualifier 2 ratio)
S	q3 unc (qualifier 3 uncertainty)
Т	unc tp: 0=rel, 1=abs (uncertainty type)
U	Ref Lib Name (library name)*
V	Ref Lib entry num (entry number in the library, such as 15)*

\* Not used for original library creation.

The columns B, C, D, E, F, and I are required. All other columns are left blank. Use one compound per row. See the example below: Note the first two lines (and only two lines) are for comment and header information.

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1	This is a DEMO	TAB file												
2	open name	cas	mol form	mol wt	R.T.	open	company ID	file name	target ion	q1	q1 ratio	q1 unc	q2	q2ratio
3	Methanol	67561	CH4O	32.04	3.33		cmpd01	c:\msdchem\1\data\oxymix_1.d						
4	Ethanol	64175	C2H6O	46.07	3.85		cmpd02	c:\msdchem\1\data\oxymix_1.d						
5	tert-Butand	75650	C4H10O	74.12	4.85		cmpd03	c:\msdchem\1\data\oxymix_1.d						
6	n-Propanol	71238	C3H8O	60.1	5.54		cmpd04	c:\msdchem\1\data\oxymix_1.d						
7	MTBE	1634044	C5H12O	88.2	6.08		cmpd05	c:\msdchem\1\data\oxymix 1.d						
8	DIPE	108203	C6H14O	102.2	7.26		cmpd06	c:\msdchem\1\data\oxymix 1.d						
9	Isobutanol	78831	C4H10O	74.1	7.9		cmpd07	c:\msdchem\1\data\oxymix 1.d						
10	ETBE	637923	C6H14O	102.2	8.06		cmpd08	c:\msdchem\1\data\oxymix 1.d						
11	tert-Pentar	75850	C5H12O	88.1	8.52		cmpd09	c:\msdchem\1\data\oxymix_1.d						
12	Methylcyc	96377	C6H12	84.16	8.64		cmpd10	c:\msdchem\1\data\oxymix 1.d						
13	Benzene	71432	C6H6	78	9.95		cmpd11	c:\msdchem\1\data\oxymix 1.d						
14	TAME	994058	C6H14O	102.2	11.17		cmpd12	c:\msdchem\1\data\oxymix_1.d						
15	( ) ) original /						10							· · · ·
Rea	ady											NUM		

2. Make sure you save the spreadsheet file in .tab format in the Database folder. Go to **File|Save as|** and choose Text (Tab delimited)(\*.txt). Put the file name in quotes with a .tab extension such as "original.tab". This will save in a tabdelimited format with a .tab extension.

As									
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b Folders	Save as type:	Text (Tab de	alimited) (*.txt)				*	Car	h
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- 3. Click **Yes** to keep the format.
- 4. CLOSE the Excel file, otherwise you will get an error message (Could not open file: C:\ database\original.tab) from MSD ChemStation.

Microsoft	Excel			×
	Do you want to sa	ve the change	es you made to 'origir	nal.tab'?
	Yes	No	Cancel	

5. Click No.

6. Go to Enhanced Data Analysis. On the command line type **rtl\_import** and then click **Execute**.

📇 Enhanced Data A	nalysis - DEFAULT.	M 7 EVALDEMO.	D (MS Data: No	t Quantitated)
<u>File Method</u> Chroma	togram <u>S</u> pectrum Ca	ili <u>b</u> rate <u>Q</u> uantitate	<u>I</u> ools ⊻iew <u>H</u> elp	)
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7. Select Library only, click OK.

Import w	ill Create:	
⊙ Lib ⊂ Lib	ary Only ary and SCD	
<u> </u>	) only	
OK	Cancel	

(The screener database can also be generated at the same time by selecting the "Library and SCD" option as seen in a later section.)

Select TAB i	nformation file				? ×
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Files of type:	Custom (*.TAB)		•		Cancel
	Dpen as read-only			_	

8. You will be prompted for a file to select from the Database folder. Select the .tab you want, click **Open**.

Library Name	
OXYMIX.L	
SAV folder	
C:\DATABASE\0XYMIXSAV	
Chromatographic Approval	
OFF	

9. Give the library a name (maximum eight characters), such as oxymix.L and click **OK**.

Enhanced DA will call each of the files in Column I of your .tab file. It will then go to the RT you specified in Column F and create a Library entry using that spectrum and the information from Columns B, D, and E. You should see chromatograms flashing before your eyes. If you fill in column H of the \*.tab file, you will get a .sav directory when the library is built.

The following screen shows the new library, Oxymix.L, the oxymix.tab file, and the .sav directory.

Lontents of LINDATABASE	Siza	Тире	Modified	Attributes
Ambs 11	5126	File Folder	3/6/01 3:23 PM	Aunduces
backup files		File Folder	10/2/02 2:17 PM	
		File Folder	2/7/02 3:28 PM	
Molstruc		File Folder	4/10/00 9:53 AM	
🛄 Nist98.1		File Folder	4/10/00 9:52 AM	
🛅 Oxymix.l 🛛 👞 🔤		File Folder	10/2/02 2:10 PM	
		File Folder	10/2/02 2:10 PM	
RtI-PCB.I		File Folder	2/9/00 5:06 PM	
RTLpest.I		File Folder	1/3/01 3:46 PM	
RTLPest2.I		File Folder	9/19/01 1:13 PM	
🛄 Wiley275.1		File Folder	9/28/99 5:18 PM	
📓 Amhs_1.scd	67KB	Microsoft Schedule+ Application	3/6/01 12:26 PM	A
📓 Cngr-srt.scd	211KB	Microsoft Schedule+ Application	2/17/00 4:32 PM	A
📓 oxygas.scd	18KB	Microsoft Schedule+ Application	6/7/02 9:49 AM	A
🔳 oxymix.tab 🛛 🛶 🔤	1KB	TAB File	10/2/02 1:59 PM	A
📓 Rtl-pcb.scd	211KB	Microsoft Schedule+ Application	2/17/00 3:53 PM	A
📓 Rtipest.scd	569KB	Microsoft Schedule+ Application	4/12/02 5:14 PM	A
📓 rtlpest2.scd	569KB	Microsoft Schedule+ Application	10/19/01 11:13	A

What is the .sav directory? The .sav is a folder created in Database folder when you create a Library from a .tab. The .sav folder contains one .sav file for each entry in the Library you just created.

Name	Size Type	Modified	Attributes
🛋 cmpd01.sav	5KB SAV File	10/2/02 2:10 PM	A
🛋 cmpd02.sav	6KB SAV File	10/2/02 2:10 PM	A
🛋 cmpd03.sav	6KB SAV File	10/2/02 2:10 PM	A
🛋 cmpd04.sav	6KB SAV File	10/2/02 2:10 PM	A
🛋 cmpd05.sav	6KB SAV File	10/2/02 2:10 PM	A
🛋 cmpd06.sav	6KB SAV File	10/2/02 2:10 PM	A
🛋 cmpd07.sav	6KB SAV File	10/2/02 2:10 PM	A
🛋 cmpd08.sav	7KB SAV File	10/2/02 2:10 PM	A
🛋 cmpd09.sav	7KB SAV File	10/2/02 2:10 PM	A
🛋 cmpd10.sav	6KB SAV File	10/2/02 2:10 PM	A
🛋 cmpd11.sav	7KB SAV File	10/2/02 2:10 PM	А
🛋 cmpd12.sav	6KB SAV File	10/2/02 2:10 PM	A

Each .sav file contains the spectrum of the Library entry. The .sav file name is taken from Column H. If Column H is left blank you will get no usable .sav files. You can rebuild the Library using the .sav files if you lost the original data files. In the dialogue box where you entered the Library name when you first created it, there is a "more" button. Clicking "more" gives you a field to specify whether you're using original data or .sav files.

#### List the Content of a Library (\*.I)

1. After the library is built, type **listlib** on the command line and then click **Execute**.



2. Choose the library name you just created then click **OK**.

D D	atabase		
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	backup files		
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	Molstruc		
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3. Click No to the detailed graphics question.

	×
٩	Do you want detailed graphics? (This process may generate a LOT of paper!!)
	Yes <u>N</u> o Cancel

If all went well you will see a list of your library entries, compound names, MW, CAS, etc.

Entry	Name	MW	CAS	MF	
1)	Methanol	32.0	000067-56-1	CH4O	
2)	Ethanol	46.1	000064-17-5	C2H6O	
3)	tert-Butanol	74.1	000075-65-0	C4H100	
4)	n-Propanol	60.1	000071-23-8	C3H8O	
5)	MTBE	88.2	001634-04-4	C5H12O	
6)	DIPE	102.2	000108-20-3	C6H14O	
7)	Isobutanol	74.1	000078-83-1	C4H10O	
8)	ETBE	102.2	000637-92-3	C6H14O	
.9)	tert-Pentanol	88.1	000075-85-0	C5H12O	
10)	Methylcyclopentane	84.2	000096-37-7	C6H12	
11)	Benzene	78.0	000071-43-2	C6H6	
12)	TAME	102.2	000994-05-8	C6H14O	
Library:	C: DATABASE OXYMIX. L	Wed Oct	02 14:29:57	2002	

If you see only a few lines of text at the top, the library was not built properly. Go back and follow the build-a-library steps again carefully, especially entering the correct datafile name and the data path in column I of the .tab file.

#### Create an SCD and a Library From a .tab File

The best way to create an .scd is to create it when you create the Library. Do ALMOST exactly what you did to create a Library in the previous section.

1. After you execute the **rtl\_import** command, select "Library and SCD".

Import will Cro Library Or Library an SCD only OK Car	eate: Ily d SCD Icel		
Select TAB info	ormation file		? ×
Look in: 🧲	🗟 Database	- 1	
Amhs_1.1 backup files Demo.L Nist98.1 RtI-PCB.1	RTLpest.I RTLPest2.I Wiley275.I		
File <u>n</u> ame: C Files of <u>type</u> : C	original.tab Custom (*.TAB) Open as <u>r</u> ead-only		<u>O</u> pen Cancel



2. Click **Yes**. An .scd will be created with the same name you give the library.

The largest ion will be the target ion, and then the next three largest ions will be the qualifiers. Uncertainty percent will be globally set to 20. Uncertainty type will be globally set to 0=relative. 3. From the Tools menu item, select "List Screen Database...".





4. You will see a listing of your .scd. Select the .scd you just created.

Screen Database : C:\DATABASE Total SCD Conds : 12	VOXANIX	(.scd				
Cpd# Compound Name	TIon	Exp_RT	Q1	Q2	Q3	
1 Methanol 2 Ethanol 3 tert-Butanol 4 n-Propanol 5 MTBE 6 DIFE 7 Isobutanol 8 ETBE 9 tert-Pentanol 10 Methylcyclopentane 11 Benzene 12 TAME	31 359 31 73 45 43 59 56 78 73	3 33 3 85 4 85 5 54 6 08 7 26 7 90 8 06 8 52 8 64 9 95 11 17	29 45 31 59 43 43 41 87 73 69 77 87	30 29 43 42 57 87 42 57 57 55 41 51 43	33 46 41 27 41 41 31 29 31 55 50 55	
Cal À = Àverage L = Linear LO = #Qual = number of qualifiers A/H = Àrea or Height ID R = R.T. B = R.T. & O O =	Linear Ovalue	r w⁄origin e L = Larg	Q = est	Quad QO = A = All	Quad w⁄origin	C.

5. Close the window.

The macro that does the .scd listing also creates a .tab file (in the c:\database folder) with the same name as the .scd. In this case, you will have oxymix.scd and oxymix.tab.

 Go into Excel and open the .tab file (oxymix.tab) of the same name as the .scd you just listed. Click Finish.

Now you have a tab file with the proper headers in row 2. This is a .tab of your .scd NOT of your library.

	Microso	ft Excel - o	oxymix.tab																	L.	
國	] <u>F</u> ile <u>E</u>	dit <u>V</u> iew Ir	nsert F <u>o</u> rm	at <u>T</u> ools	Data W	indow <u>H</u>	<u>l</u> elp														18 ×
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1	A1	-	= 9	SCD tabb	ed list fo	r oxym	ix.scd; list cr	eated Tue	Oct	08 16:50	:42 20	002									
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2	open	name	cas	mol form	mol wt	R.T.	opcompany	file target	q1	q1ratio q1	unc	q2	q2ratio	q2 unc	qЗ	q3ratio	q3 unc	unc tp	Ref Lib Na	Ref Lib	entr
3	3.33	Methanol	67561	CH4O	32.04	3.33	cmpd01	31	29	81.6	20	30	9.2	20	33	2.6	20	0	oxymix.L	1	
4	3.85	Ethanol	64175	C2H6O	46.07	3.85	cmpd02	31	45	71.3	20	29	31.5	20	46	27.2	20	0	oxymix.L	2	
5	4.85	tert-Butar	75650	C4H10O	74.12	4.85	cmpd03	59	31	18.1	20	43	16	20	41	14.8	20	0	oxymix.L	3	
6	5.54	n-Propano	71238	C3H8O	60.1	5.54	cmpd04	31	59	28.8	20	42	19.5	20	27	17.8	20	0	oxymix.L	4	
7	6.08	MTBE	1634044	C5H12O	88.2	6.08	cmpd05	73	43	18.8	20	57	18.7	20	41	15.3	20	0	oxymix.L	5	
8	7.28	DIPE	108203	C6H14O	102.2	7.26	cmpd06	45	43	50.5	20	87	47.6	20	41	18.9	20	0	oxymix.L	6	
9	7.9	Isobutano	78831	C4H10O	74.1	7.9	cmpd07	43	41	79.4	20	42	61.2	20	31	44.2	20	0	oxymix.L	7	
10	8.08	ETBE	637923	C6H14O	102.2	8.06	cmpd08	59	87	57.3	20	57	29.7	20	29	8.3	20	0	oxymix.L	8	
11	8.52	tert-Penta	75850	C5H12O	88.1	8.52	cmpd09	59	73	74.9	20	55	40.7	20	31	13.8	20	0	oxymix.L	9	
12	8.64	Methylcy	96377	C6H12	84.16	8.64	cmpd10	56	69	54.8	20	41	47.1	20	55	26.4	20	0	oxymix.L	10	
13	9.95	Benzene	71432	C6H6	78	9.95	cmpd11	78	77	25.2	20	51	15.4	20	50	14.5	20	0	oxymix.L	11	
14	11.2	TAME	994058	C6H14O	102.2	11.17	cmpd12	73	87	31.3	20	43	27.6	20	55	21.1	20	0	oxymix.L	12	
15	0																				-
		oxymix/										1									
Re	ady												-				-	1	NUM		

This also is NOT the original .tab from which you created the Library and .scd. You will notice the datafile paths (column I) are gone. Columns J-V are filled in. The software has associated each scd entry with a library name (column U) and library entry number (column V). Information depicted in Columns U and V is used for the XCR (cross correlation) on the screener report [2].

#### Add Additional Entries to an Existing Library

When you need to add spectra to an existing library and you have not made changes to the corresponding .scd file, you can rebuild your library and .scd using the procedures described above. Just add the new entries to the original.tab file, sort the RT, and save the file. Then use **rtl\_import** command to build the library.

If you have made changes to your .scd file, for example, picked different target/qualifier ions or changed ion ratios etc., you should follow these steps.

 Display the spectrum (preferably averaged over the width at the half-height of the peak) to be added to the library. Go to **Spectrum | Edit Library**|, and select the library to add the compound into.



#### Edit PBM Library

- Select Library
   Add New Entry
- C ENERGY LINKY
- Edit Existing Entry
   Create Library



Browse for Folder	? ×
Select Library	
Database	
Amhs_1.I	
😟 🛄 backup files	
Demo.L	
Molstruc	
Nist98.1	
🚽 🛶 🛶 📥 🔄 Oxymix I	
OXYMIX.sav	
Rti-PCB.I	
RTLpest.I	
RTLPest2.1	
ОК	Cancel
	- and and -

2. After the library is chosen, select the "Add New Entry" option.

C <u>S</u> e	elect Library
• Ac	ld New Entry
C <u>E</u> c	fit Existing Entry
C <u>C</u> r	eate Library
CDe	elete Library

3. Enter the information associated with each entry. The Retention Index should be filled in with the RT converted to seconds. In this case, it is  $4.36 \times 60 = 261.6$ 

	- (Database (Oxyii		
Description of Mass Sp	ectrum Scan 10	95 (4.365 min): Oxymix_1.d	
Name: Isoproponal			×.
Mol. Eormula: C3H80	$\rightarrow$	Mol. <u>W</u> eight:	60.057
Miscellaneous			*
			<u>*</u>
CAS number: 67	63 0 Com	pany I <u>D</u> : cmpd13 <u>R</u> et. Ind	ex: 261.6
Melting Point (C):		Boiling Point (C):	
✓ Include in search			
		10 IV IV IV	



- 4. Repeat the steps to add all the entries. In this case, we have added a total of four new entries.
- 5. Go to Enhanced DA and execute a **listlib** command on the command line to list all the library entries.

ry	Name	M₩	CAS	MF
1)	Methanol	32.0	000067-56-1	CH40
2)	Ethanol	46.1	000075 45 0	C2H60
4)	n-Propanol	60 1	000075-65-0	C3H8O
5)	MTBE	88.2	001634-04-4	C5H12O
6)	DIPE	102.2	000108-20-3	C6H14O
7)	Isobutanol	74.1	000078-83-1	C4H100
8)	ETBE	102.2	000637-92-3	C6H14O
9)	tert-Pentanol	88.1	000075-85-0	C5H12O
10)	Methylcyclopentane	84.2	000096-37-7	C6H12
12)	Benzene	102.2	000071-43-2	CEHIAO
13)	Isopropanol	60 1	000067-63-0	C3H80
14)	sec-Butanol	74.1	000078-98-2	C4H100
15)	1,2-Dimethoxyethane	90.1	000110-71-4	C4H10O2
16)	n-Butanol	74.1	000071-36-3	C4H100

The entry numbers in the above picture (13, 14, 15, and 16) are needed in the .tab file for building the .scd file.

### **Screener Database**

#### Create Only the SCD From Your .tab

This is used with an existing Library you edited (for example, entries added).

- 1. In Excel, open your exisiting oxymix.tab (not original.tab) file for editing.
- 2. Fill in columns B, C, D, E, F, U, and V (column H is optional). You will need to know the existing Library name (enter in column U) and entry number (enter in column V) for each compound.

2	Cue Edi	t wew Insert	rum	at 1	ools	Dat		NOON	k ⊡atb								-										4	-10
		664	V.	¥	<b>B</b>	ß		<b>K</b> )	• C.M. v	6	$\Sigma f_{*}$	2+ ×	一個小	100	)%	• 6	2] +											
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	3.85	Ethanol		64	175	C2H	60		46.07	•	3.85		cmpd02		31	45	71	20	29	32	20	46	27	20	(	) oxymix.L	1	2
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	8.52	tert-Pentanol		75	850	C5H	120		88.1	Ľ.	8.52		cmpd09		59	73	75	20	55	41	20	31	14	20	(	) oxymix.L	5	9
1	8.64	Methylcyclop		96	377	C6H	12		84.18	5	8.64		cmpd10		56	69	65	20	41	47	20	55	26	20	(	) oxymix.L	10	)
3	9.95	Benzene		71	432	C6H	6		78	3	9.95		cmpd11		78	77	25	20	51	15	20	50	15	20	(	) oxymix.L	11	1
-	11.17	TAME	3	99.40	168	CEH	140	_	102.2	5	11 17	_	cmpd12	-	73	87	31	20	43	28	20	55	21	20	. (	DUTINUL	-	1
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### 3. Sort the rows using the RT (column F).

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3	3.33	Methanol	675	51 CH4O		32.04	3.33		cmpdD1		31	29	82	20	30	9.2	20	33	2.6	20	0	oxymix.L		1
4	3.85	Ethanol	641	75 C2H6	С	46.07	3.85		cmpdD2		31	45	71	20	29	32	20	46	27	20	0	oxymix.L	-	2
5		Isopropanol	676	30 C3H8	C	60.1	4.36		cmpd13													oxymix.L		13
6	4.85	tert-Butanol	756	50 C4H10	00	74.12	4.85		cmpdD3		59	31	18	20	43	16	20	41	15	20	0	oxymix.L		3
7	5.54	n-Propanol	712	38 C3HB	О.	60.1	5.54		cmpd04		31	59	29	20	42	20	20	27	18	20	0	oxymix.L		4
8	6.08	MTBE	16340	44 C5H12	20	88.2	6.08		cmpd05		73	43	19	20	57	19	20	41	15	20	0	oxymix.L		5
9		sec-Butanol	789	22 C4H10	00	74.1	6.81		cmpd14													oxymint		14
10	7.26	DIPE	1082	03 C6H14	10	102.2	7.26		cmpd06		45	43	51	20	87	48	20	41	19	20	0	oxymix.L	5	6
11	7.9	Isobutanol	788	31 C4H10	00	74.1	7.9		cmpd07		43	41	79	20	42	61	20	31	44	20	0	oxymix.L		7
12	8.06	ETBE	6379	23 C6H14	40	102.2	8.06		cmpd08		59	87	57	20	57	30	20	29	8.3	20	0	oxymix.L		8
13	8.52	tert-Pentanol	758	50 C5H12	20	88.1	8.52		cmpd09		59	73	75	20	55	41	20	31	14	20	0	oxymix.L		9
14	8.64	Methylcyclop	963	77 C6H12	2	84.16	8.64		cmpd10		56	69	55	20	41	47	20	55	26	20	0	oxymix.L	1.00	10
15		1,2-Dimethox	1107	14 C4H10	002	90.1	9.13		cmpd15													oxymix.L		15
16		n-Butanol	713	53 C4H10	00	74,1	9.74		cmpd16													oxymineL		16
17	9.95	Benzene	714	32 C6H6		78	9.95		cmpd11		78	77	25	20	51	15	20	50	15	20	0	oxymix.L		11
18	11.17	TAME	9940	58 C6H14	10	102.2	11.17		cmpd12		73	87	31	20	43	28	20	55	21	20	0	oxymix.L		12
19																						102	L	
4 4	> >	oxymix /										1												•
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- 4. Save the **.tab file** and close Excel.
- 5. Go to **Enhanced DA** and execute an **rtl\_import** command on the command line.

Import w	ill Create:	
C Libr	ary Only	
🔿 Lįbr	ary and SCD	
• <u>s</u> ci	) only	
ОК	Cancel	

6. Select "**SCD only**" and click **OK**. You will be prompted to select a .tab file from the Database folder.

7. Select the .tab you want and then click **Open**.



8. Give the scd a name (maximum eight characters), such as oxymix.scd, and click **OK**.

tame or 5	CD to create	
Screen Dal	abase Name ( scd)	
oxymix SCI	D	
(a)		
OK		
UN.	Lancel	

An .scd will be created using the spectra from the library. The largest ion will be the target ion, and then the next three largest ions will be the qualifiers. Uncertainty percent will be globally set to 20. Uncertainty type will be globally set to 0=relative.

#### Make an ASCII File of the Screener Database

The information in a .scd can be saved in ASCII format (text) for sharing or for future reference.

- From MSD software, enhanced DA, go to Tools
   I List Screen Database...]. A dialogue box comes up.
- 2. Choose an existing Screen Database (oxymix.scd) in the Database folder and click **Open**. You will get a text listing in a MultiVu window.

The following graphic shows that the four compounds that were added to the library are successfully added to the screener database in the proper RT order.

SCD Compoun	nd List R	eport				
Screen Database : C:\DÀTÀBÀS Fotal SCD Cpnds : 16	SE/OXYMIX	. scd				
od# Compound Name	TIon	Exp_RT	Q1	Q2	Q3	
Methanol Ethanol Isopropanol A tert-Butanol MTBE 7 sec-Butanol B DIPE J Isobutanol ETBE L tert-Pentanol Methylcyclopentane 1,2-Dimethoxyethane A n-Butanol Benzene TAME	31 31 45 31 73 45 45 45 59 56 56 78 73	$\begin{array}{c} 3.33\\ 3.85\\ 4.36\\ 4.85\\ 5.54\\ 6.08\\ 6.81\\ 7.26\\ 7.90\\ 8.52\\ 8.64\\ 9.13\\ 9.74\\ 9.95\\ 11.17\end{array}$	29 45 43 59 43 59 43 41 87 69 60 41 77 87	30 29 41 42 57 31 87 42 55 41 29 31 51 43	33 46 29 41 27 41 27 41 31 31 35 55 90 43 55	
al À = Average L = Linear LO Qual = number of qualifiers À/H = Area or Height ) R = R T R = R T & O O	= Linear = Qvalue	w∕origin L = Larg	Q=( resti	Quad QO = A = All	Quad w/or:	igin

Save the MultiVu window to a file (**File**|**Save as...**|), for example, oxymix.txt, to be kept as a record.

#### Edit a \*.scd Using a \*.tab

If you want to change individual ions, ion ratios, and/or uncertainties etc.

- 1. Use the "**List Screener Database...**" to generate a .tab file for the .scd to be edited.
- 2. Open the .tab in Excel. Edit existing entries (select a different target ion, change the qualifier ion ratios etc.). See the example below.

2	dicroso	ft Excel - d	oxymix.tab	-					-		_										IX
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2	open	name	cas	mol for	m mol w	R.T.	op compan	y file	target	q1	q1 ratio	q1 une	-2	q2ratio	q2	q3	q3ratio d	3 unc	t Ref Lib Na	a Ref Lib entry nur	m
3	3.33	Methanol	67561	CH4O	32.04	1 3.33	cmpd01		31	29	81.6	30	30	9.2	20	33	2.6 2	0	0 oxymix L	0	1
4	3.85	Ethanol	64175	C2H6C	46.07	3.85	cmpd02		31	45	71.3	30	29	31.5	20	46	27.2 2	0	0 oxymix.L		2
5	4.85	tert-Butan	75650	C4H10	0 74.12	4.85	cmpd03		59	31	18.1	30	43	16	20	41	14.8 2	0	0 oxymix.L		3
6	5.54	n-Propano	71238	C3H8C	60.1	5.54	cmpd04		31	59	28.8	30	42	19.5	20	27	17.8 2	0	0 oxymix.L		4
7	6.08	MTBE	1634044	C5H12	0 88.2	2 6.08	cmpd05	2.	73	43	18.8	30	57	18.7	20	41	15.3 2	0	0 oxymix.L		5
8	7.26	DIPE	108203	C6H14	0 102.2	2 7.26	cmpd06		45	43	50.5	30	87	47.6	20	41	18.9 2	0	0 oxymix.L		6
9	7.9	Isobutano	78831	C4H10	0 74.1	7.9	cmpd07		43	41	79.4	20	42	61.2	20	31	44.2 2	0	0 oxymix.L	1	7
10	8.06	ETBE	637923	C6H14	0 102.2	8.06	cmpd08		99	87	57.3	20	57	29.7	20	29	8.3 2	0	0 oxymix.L	1	8
11	8.52	tert-Penta	75850	C5H12	0 88.1	8.52	cmpd09		99	73	74.0	20	55	40.7	20	31	13.8 2	0	0 oxymix.L		9
12	8.64	Methylcy	96377	C6H12	84.16	8.64	cmpd10		56	69	70	20	41	47.1	20	55	26.4 2	0	0 oxymix.L	11	0
13	9.95	Benzene	71432	C6H6	78	9.95	cmpd11	3	78	77	70	20	51	15.4	20	50	14.5 2	0	0 oxymix.L	1	1
14	11.2	TAME	994058	C6H14	0 102.2	2 11.17	cmpd12		73	87	70	20	43	27.6	20	55	21.1 2	0	0 oxymix.L	1:	2
15		oxymix /									Barran		4								Ìŕ
Re	ady																		N	M	ā 🦷

Be very careful in columns U and V. Do not change anything in these two columns.

- 3. Save the **.tab file**, then close Excel.
- 4. Go to **Enhanced DA**, execute an **rtl\_import** command on the command line.
- 5. Select **SCD only**. The software preserves the edits you have made in the .tab for the .scd.
- 6. Go to **Tools | List Screen Databases...|** to confirm the changes.

### FAQ

#### When to Use "Set Other (via command)..." Function?

Global changes to the \*.scd can be made using the menu item

1. Tools | Change Screen Database Parameters.... The following will appear:



The **Set Other** (via command) is used to make global changes not included in the list.

For instance, if you want to use a specific Chem-Station Integrator Parameter file for target ion integration:

2. Choose Set Other (via command) and click OK.

Input		
Enter the c	ommand	
tgtevents\$	=''myfile.e'	
OK	Cancel	

3. Input the ChemStation Integrator Parameter file you saved, in this case myfile.e, enclosed in quotation marks.

4. Click **OK** and you will see your currently specified method screen database file updated by watching the gray message line at the bottom.

To update qualifier ion intergration parameter files, repeat the above procedure using q1events\$, q2events\$ and q3events\$ for qualifier ion q1, q2 and q3 respectively.

**Caution**: Global changes made using either one of the preset choices or Set Other will AFFECT ALL COMPOUNDS in the \*.scd.

# How to Use the Integration Parameters with the Screener?

The screener is always using the "AutoIntegrate Function" when the ChemStation integrator is selected. Because the "AutoIntegrate Function" does not use any of the events files, none of the integration events will be used in the screening process unless an event file was specified for a compound. The screener does not use the autoint.e parameter file. This file is NOT related to the "AutoIntegrate Function".

The screener is integrating extracted ion chromatograms (EICs), not total ion chromatograms (TIC). Therefore, the screener integration parameters should be determined by integrating the EICs. When the optimal parameters are found, save them in an event file (for example, myfile.e).

To associate an event file to all the compounds, refer to the discussion above.

#### **Does RTE Integrator Use any Parameter File?**

Yes, the RTE integrator uses rteint.p from your method directory.

- After selecting the RTE integrator, go to Chromatogram |MS Signal Integration Parameter...| to change the integrator parameters.
- 2. Click **OK**; the parameters are automatically saved to rteint.p.

The screener will always use rteint.p for integration unless you use Set Other (via command) to select another .p file.

Different from the RTE integrator, the ChemStation Integrator always uses the "AutoIntegrate Function" for integration. You have to associate an .e file to the compounds to specify the integration parameters. How Can I Get the Screener Report to Show Just the Hits and Probable Hits?

- 1. Go to |Quantitate|Report Options...|
- 2. Check the box next to "Omit Target Compounds that Are Missed".

How Can I Get the Screener Report to Show Just the Hits (x)?

- 1. Go to |Quantitate|Report Options...|
- 2. Check the box next to "Omit Target Compounds that Are Missed" and check the box next to "Have qualifiers Out of Range".

#### What Does Tools | Exclude Zero Qualifiers Do?

You get probable hits (designated by a ?) when one or more of the qualifiers do not meet criteria. Many times a qualifier will not meet criteria because its ion abundance is zero. If the ion abundance is zero, the compound probably is not present. Exclude Zero Qualifiers, when checked, eliminates probable hits from the report if any of the qualifiers is zero.

**Caution**: If a qualifier is small, it could be absent even when the compound is present, so do not use small abundance qualifier ions.

#### Can I Append Entries to the Library Using Excel?

If you choose to create a Library only or a Library and SCD after executing the rtl\_import command, your Library or Library and SCD will be created from scratch, with all counters set to zero. In other words, Excel .tab files can NOT be used to append Library entries to an existing library nor to edit existing Library entries.

# How Do I Get Screener Report of a GC Detector on the MS System?

There is no GC mode screener report in the MSD DA, but, here is the work-around. After you load the MS and ECD signal into the ChemStation:

- 1. Go to |File|Select Signals...| and uncheck the MS signal.
- 2. Now, select |Tools|Create Screen Results for Current File|.

This will create a screener report for the GC data and save the report to RTLPEST(2).RES. (Depending on the pesticide library revision, you may or may not get the 2 in the file name.) 3. To generate a "pseudo" GC screener report, go back to |File|Select Signals...| and check the MS signal. Use |Tools|Generate Screener Report for Current File| to get a report.

**Note**: The GC and MS screeners use the same RTLPEST.RES file. They overwrite each other when you screen. If you do not uncheck the MS signal, you will screen and then report the MS signal.

#### Screen 0,1,"F"

This command, when executed on the command line in Data Analysis View, produces a multipage Screener report for a data file that has already been screened. The first page of the report is a summary report identical to the report from Tools | Generate Screen Report for Current File. Each hit is then reported on a separate page, complete with all the graphics you normally see in Results Screener View. This multipage report is sent to the default printer and is not viewable on the monitor.

# What is "Subtraction Method" and Which Method Should I Use ?

The subtraction method that is used is printed near the top of the Screener Report, on the right side. Under Tools | Change Screen Database parameters, one of the choices is Set Subtraction Method. There are four methods from which to choose:

**Use Relative Areas** - qualifier ion ratios are based on area comparisons to the target ion area. It is NOT recommended to use this method. Small areas may not be integrated or matrix may add significantly to an area. There is NO subtraction done when using this method.

**No Subtraction** - qualifier ion ratios are based on ion abundances using the spectrum at the apex of the chromatographic peak. This method can be used if there are no interfering ion abundances from matrix. There is NO subtraction done using this method.

Lower of First and Last - qualifier ion ratios are based on ion abundances using the spectrum at the apex of the chromatographic peak MINUS the ion abundances just before the start of the peak or just after the end of the peak. The ion abundances that are subtracted are those that are the lower of either the start or end of the peak. This subtraction method can compensate for ions abundances due to matrix. Average of First and Last - qualifier ion ratios are based on ion abundances using the spectrum at the apex of the chromatographic peak MINUS the average of the ion abundances just before the start of the peak and just after the end of the peak. This subtraction method can compensate for ion abundances due to matrix, and is the recommended subtraction method to use.

#### How Do I Build a Quant Database from a Screener Database?

A Quant database is used for quantitating identified compounds and is calibrated. A Screener database is used for identifying compounds, without quantitating them. A Quant database (qdb.mth in the mymethod.m folder) has the same format as a Screener database (such as oxymix.scd in the Database directory). To convert a Screener database to a Quant database, do the following:

- 1. In the method folder, say mymethod.m, locate the file **qdb.mth.**
- 2. Rename qdb.mth to qdb.mth.ori (for original).
- 3. Copy your Screener database, say **oxymix.scd** from C:\database, and paste it into your method folder, **mymethod.m**.
- 4. Rename **oxymix.scd** (in the mymethod.m folder) to **qdb.mth**.
- 5. Reload your method, **mymethod.m**, in Enhanced Data Anaylsis.
- 6. Select **Calibrate>Edit Compounds** to view the Quant database.
- 7. Calibrate the Quant database as you normally would.

# What Else Can I Do with my Quant Database and Mass Spectral Library?

Agilent introduced Deconvolution Reporting Software (DRS), product # G1716AA. Application Note 5989-1157EN describes the product, its use and performance in detail. DRS uses the results from Quant databases as part of its report. DRS also uses Mass Spectral Libraries in producing its reports. The DRS Help files provide detailed procedures for converting Agilent format Mass Spectral Libraries for use with DRS. So the Quant databases and Libraries you have built using this technical overview have wider application. DRS is a powerful software tool to aid the analyst in finding trace compounds in complex matrices for different industries, for example, forensic toxicology, food safety, flavor and fragrance (that is, allergens), environmental, and homeland security.

### **Troubleshooting Questions**

# "When I Execute a Listlib Command, I Only See the Header of the Report, No Entries Listed".

Make sure the file name and the data path is entered correctly

#### "Could Not Open File: C:\database\oxymix.tab"

Close the file in Excel

#### "Array Index 6 Out of Bounds"

The TAB file is in Excel (not text tab delimited) format. Re-save the file in tab format.

#### "Index Out of Range"

There is a mismatch between a library entry number in the \*.scd and the library.

The library entry with that number does not exist. To find which \*.scd entry has the wrong number, screen a sample with the \*scd. Go to **View | Results Screener**. Under the menu item Spectrum, select **| Display Reference Spectra**. The lower left panel should now display the unknown spectrum above the library reference spectrum for that compound. Double click through the compound list until no library reference spectrum is displayed. This compound(s) has an incorrect or missing spectrum in the library.

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