

Calculations combining  
multiple images

# Select the images that you want to use in the calculations

Image Analysis | C:\Users\shimadzu\Shimadzu Dropbox\yamaguchi shinichi\Yamaguchi\_data\demo\_data\_etc\DemoData\Project\Testicle\_HCA\_SEG.rpf - IMAGEREVEAL

File View Search Settings Option Extensions Help

ROI List

No.	Use	File Name
1	<input checked="" type="checkbox"/>	testicle_9A...

Data Matrix Table

No.	Use	Tag	Label	m/z	PLS Coefficient	All
1	<input checked="" type="checkbox"/>		699.9849-700.9849	700.4849	-1.518e-001	66576.904
2	<input checked="" type="checkbox"/>		700.9849-701.9849	701.4849	-2.800e-001	61319.627
3	<input checked="" type="checkbox"/>		701.9849-702.9849	702.4849	-9.440e-002	40328.424
4	<input checked="" type="checkbox"/>		702.9849-703.9849	703.4849	-1.217e-001	53341.227
5	<input checked="" type="checkbox"/>		703.9849-704.9849	704.4849	-1.238e-001	37124.059
6	<input checked="" type="checkbox"/>		704.9849-705.9849	705.4849	-4.234e-002	60134.142
7	<input checked="" type="checkbox"/>		705.9849-706.9849	706.4849	-4.681e-002	40288.134
8	<input checked="" type="checkbox"/>		706.9849-707.9849	707.4849	-1.887e-001	57736.010
9	<input checked="" type="checkbox"/>		707.9849-708.9849	708.4849	-6.243e-002	41579.730
10	<input checked="" type="checkbox"/>		708.9849-709.9849	709.4849	-1.058e-001	81457.818

Graph

testicle\_9AA\_P1\_SL\_5x\_1\_AREA01.imdx All Average

MS Image

Display

m/z Tolerance: 885.48492±0.5000

Compound Name/Comment: 884.9849-885.9849

File Name: testicle\_9AA\_P1\_SL\_5x\_1\_AREA01.i mdx

Type: Data Matrix

Copy Information

MS Image List

Add MS Image

Sort

Display

m/z Search

Superimposition

Arithmetic Operations

794.9849-795.9... 795.48492

795.9849-796.9... 796.48492

808.9849-809.9... 809.48492

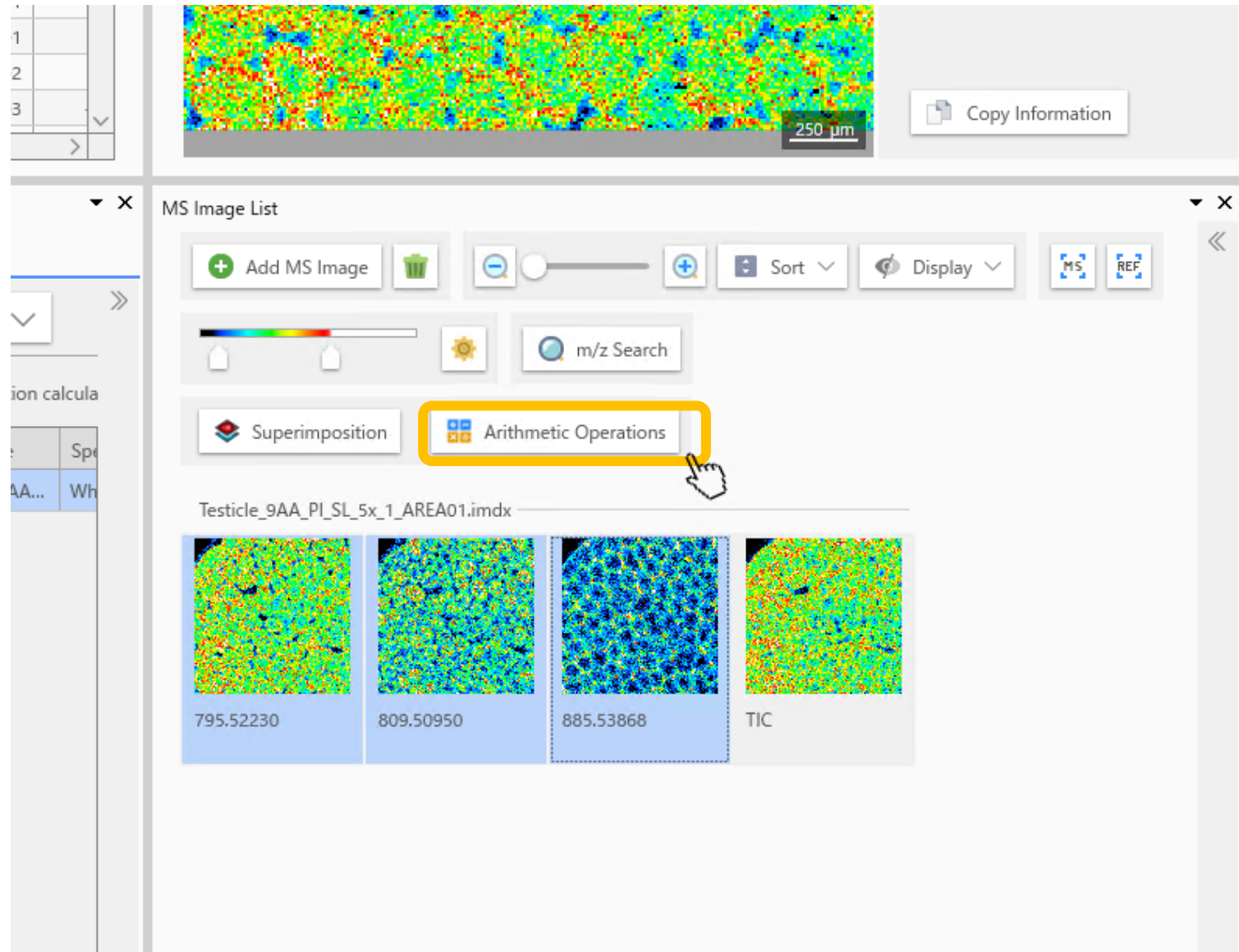
884.9849-885.9... 885.48492

885.9849-886.9... 886.48492

TIC

select

# Click the “Arithmetic Operations” button



# The “Arithmetic Operations” screen opens

Arithmetic Operations

No normalization calculation is applied to the TIC.

Constant

884.9849-885.9849  
885.48492±0.5000

808.9849-809.9849  
809.48492±0.5000

794.9849-795.9849  
795.48492±0.5000

The image you select appears on the right.

Preview Real Time Update Display MS REF

Import Export Add Expression

Expression

(	Coefficient	m/z	Compound Name	Adduct Ion	Alg...	)	Oper...

250 µm

Add to MS Image List

Close

# Double click on the image to add an expression

Arithmetic Operations

No normalization calculation is applied to the TIC.

Constant

884.9849-885.9849  
885.48492±0.5000

808.9849-809.9849  
809.48492±0.5000

794.9849-795.9849  
795.48492±0.5000

Double click the MS image to use in the calculation or click the "Add Expression" button

Import Export Add Expression

Expression

	Coefficient	m/z	Compound Name	Adduction	Arg...		Oper...
None	1	885.48492	884.9849-885.9849		A	None	+

A

Preview Real Time Update Display MS REF

250 µm

Add to MS Image List

Close

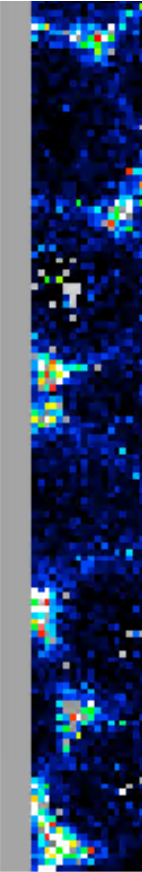
Change the order, operators, brackets, coefficients etc. to create the necessary expressions.

Expression

(	Coefficient		m/z	Compound Name	Adduct Ion	Alg...	)	Oper...
None ▾	1		885.53868	885.53868		<input checked="" type="checkbox"/> A	None ▾	/ ▾
None ▾	1		795.52230	795.52230		<input checked="" type="checkbox"/> B	None ▾	/ ▾
None ▾	1		809.50950	809.50950		<input checked="" type="checkbox"/> C	None ▾	/ ▾







A/B/C




create the necessary expressions



# Example of $(A+B)/C$

Expression

(	Coefficient		m/z	Compound Name	Adduct Ion	Alg...	)	Oper...	
(	1		809.50950	809.50950		<input checked="" type="checkbox"/> A	None	+	
None	1		885.53868	885.53868		<input checked="" type="checkbox"/> B	)	/	
None	1		795.52230	795.52230		<input checked="" type="checkbox"/> C	None	/	

(A+B)/C



# Add to the main window

Arithmetic Operations

⚠ No normalization calculation is applied to the TIC.

Constant

884.9849-885.9849  
885.48492±0.5000

808.9849-809.9849  
809.48492±0.5000

794.9849-795.9849  
795.48492±0.5000

Preview Real Time Update Display MS REF

Import Export Add Expression

Expression

(	Coefficient	m/z	Compound Name	Adduct Ion	Alg...	)	Oper...
(	1	885.48492	884.9849-885.9849		<input checked="" type="checkbox"/> A	None	+
None	1	809.48492	808.9849-809.9849		<input checked="" type="checkbox"/> B	)	/
None	1	795.48492	794.9849-795.9849		<input checked="" type="checkbox"/> C	None	/

(A+B)/C

The image created can be added to the list of MS images on the main window

250 μm

Add to MS Image List

Close



# The image is added to the main screen

The screenshot displays the IMAGEREVEAL software interface with several windows open. The main window shows a large MS Image (Mass Spectrometry Image) with a green border. To the left, there is a sidebar with buttons for File, Image Setting, Image Registration, ROI Setting, Collectively Analyze, Analyze, Analysis Result, PCA Result, and PLS Result. The top menu bar includes File, View, Search, Settings, Option, Extensions, and Help. The top status bar shows 'Differential Analysis | C:\temp\test.rpl\* - IMAGEREVEAL'.

The 'ROI List' window shows a table with columns: No., Use, File Name, ROI Na..., and Attribute. It lists four ROIs, all of which are checked in the 'Use' column.

No.	Use	File Name	ROI Na...	Attribute
1		Testicle_9A...	All	Group A
2	✓	Testicle_9A...	ROI001	Group A
3	✓	Testicle_9A...	ROI002	Group B
4	✓	Testicle_9A...	ROI003	Group C

The 'Data Matrix Table' window shows a table with columns: No., Use, Tag, Label, m/z, PCA-Horizontal Axis, PCA-Vertical Axis, and PLS C. It lists 18 data points, all of which are checked in the 'Use' column.

No.	Use	Tag	Label	m/z	PCA-Horizontal Axis	PCA-Vertical Axis	PLS C
1	✓		PCA-Horizontal Axis	795.5223	5.146e-001	2.805e-001	
2	✓		PCA-Vertical Axis	796.5249	3.226e-001	2.427e-001	
3	✓			797.5244	2.191e-001	1.049e-001	
4	✓			809.5095	2.469e-001	-2.248e-001	
5	✓			767.4923	1.521e-001	1.237e-001	
6	✓			885.5386	-2.091e-001	3.250e-001	
7	✓			810.5122	1.787e-001	-1.261e-001	
8	✓			796.0191	7.455e-002	3.384e-002	
9	✓			798.5249	1.086e-001	1.068e-001	
10	✓			795.7848	8.688e-002	5.959e-002	
11	✓			886.5409	-1.460e-001	2.244e-001	
12	✓			768.4949	9.585e-002	7.644e-002	
13	✓			797.0457	5.680e-002	4.668e-002	
14	✓			796.7837	6.608e-002	4.093e-002	
15	✓			837.5388	1.255e-001	-1.510e-001	
16	✓			857.5078	3.181e-002	1.825e-001	
17	✓			793.5071	9.283e-002	6.814e-002	
18	✓			721.4793	1.093e-001	8.772e-003	

The 'MS Image' window shows a large heatmap of the mass spectrometry image. To the right of the heatmap, there is a 'Compound Name/Comment' section with the following information:

Compound Name/Comment:  
EXP = [(A+B)/C]  
A = <809.50950  
±0.0200/809.50950>  
B = <885.53868  
±0.0200/885.53868>  
C = <795.52230  
±0.0200/795.52230>  
File Name:  
Testicle\_9AA\_PL\_SL\_5x\_1\_AREA01.i  
mdx  
Type:  
Arithmetic

The 'Graph' window shows a 'Spectrum' plot with 'Intensity' on the y-axis and 'm/z' on the x-axis. The plot shows several peaks, with the most prominent one at m/z 795.52084. Other labeled peaks include 721.48186, 767.49182, 796.52374, 796.52363, 837.53900, and 885.53782.

The 'Analysis Parameters' window shows a table with columns: No., Name, and Value. It lists five parameters, all of which are set to 'None'.

No.	Name	Value
1	Normalize	None
2	Data Matrix Analysis Method	Target
3	Compound Template	Peak List
4	Tolerance/Bin Size (Da)	0.0200
5	Threshold Setting	Off

The 'Arithmetic Operations' window shows a table with columns: D..., File Name, and Sp. It lists one operation, 'Testicle\_9AA...', which is checked in the 'D...' column.

D...	File Name	Sp
✓	Testicle_9AA...	Wh

The 'Testicle\_9AA\_PL\_SL\_5x\_1\_AREA01.i.mdx' window shows a series of five small heatmaps, each representing a different m/z value: 795.52230, 809.50950, 885.53868, and TIC. The first heatmap is highlighted with a green border.

The information about the calculations is included in a comment.  
The expression is displayed as a compound name.

# Import and export expressions

Arithmetic Operations

⚠ No normalization calculation is applied to the TIC.

Constant

884.9849-885.9849  
885.48492±0.5000

808.9849-809.9849  
809.48492±0.5000

794.9849-795.9849  
795.48492±0.5000

Preview Real Time Update Display MS REF

The created formula can be exported as a csv file.  
They can also be imported.

Import Export Add Expression

Expression

(	Coefficient	m/z	Compound Name	Adduct Ion	Alg...	)	Oper...
(	1	885.48492	884.9849-885.9849		<input checked="" type="checkbox"/> A	None	+
None	1	809.48492	808.9849-809.9849		<input checked="" type="checkbox"/> B	)	/
None	1	795.48492	794.9849-795.9849		<input checked="" type="checkbox"/> C	None	/

(A+B)/C

250 µm

Add to MS Image List

Close