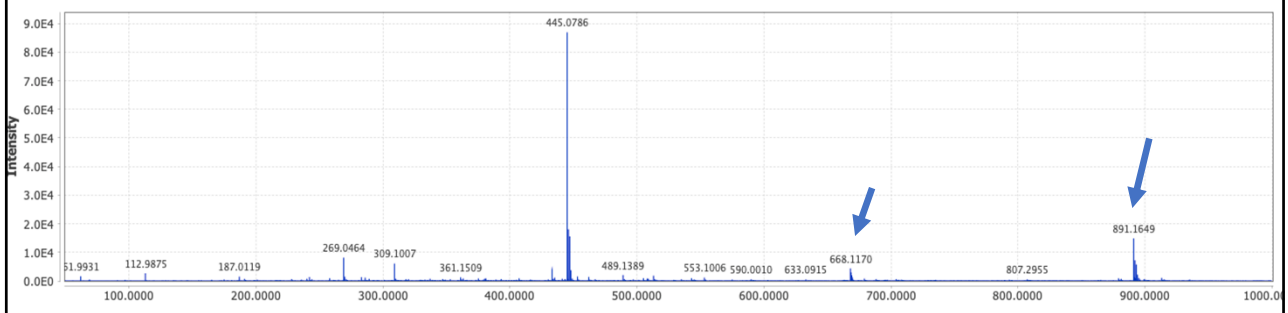
Using the METLIN database

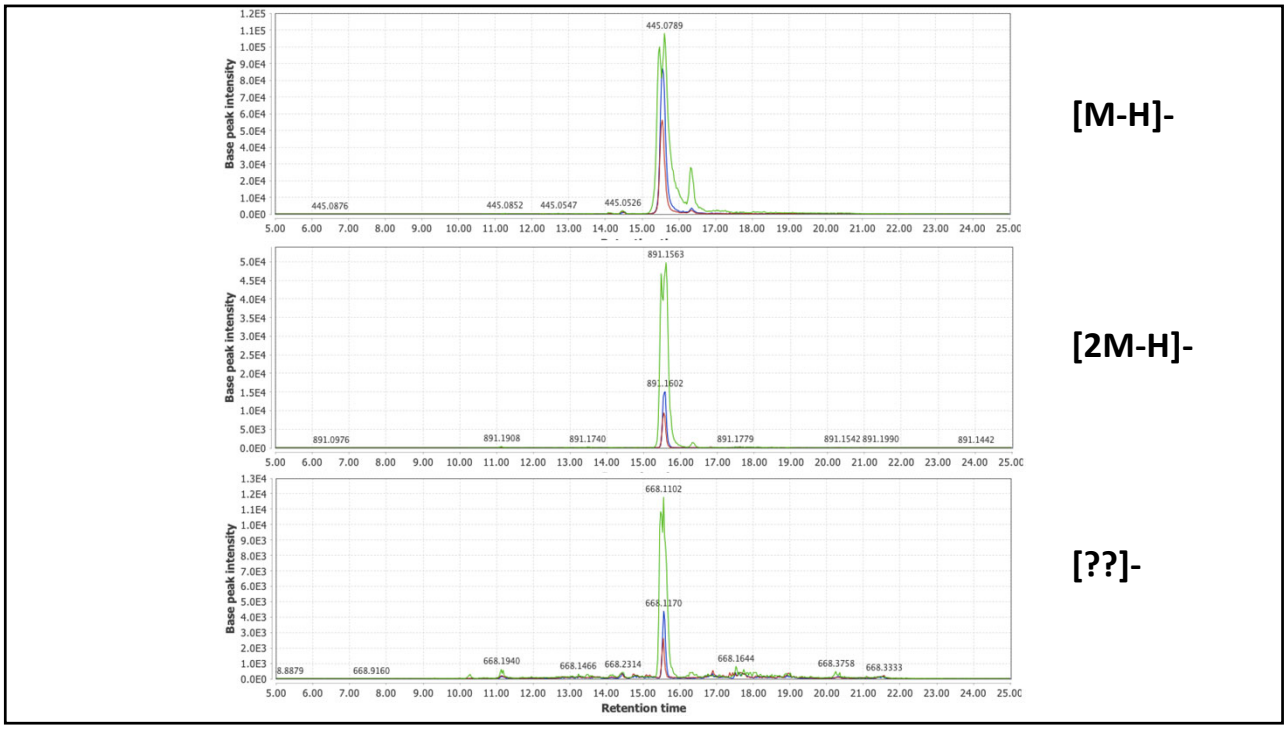
Stephen Barnes, PhD

1

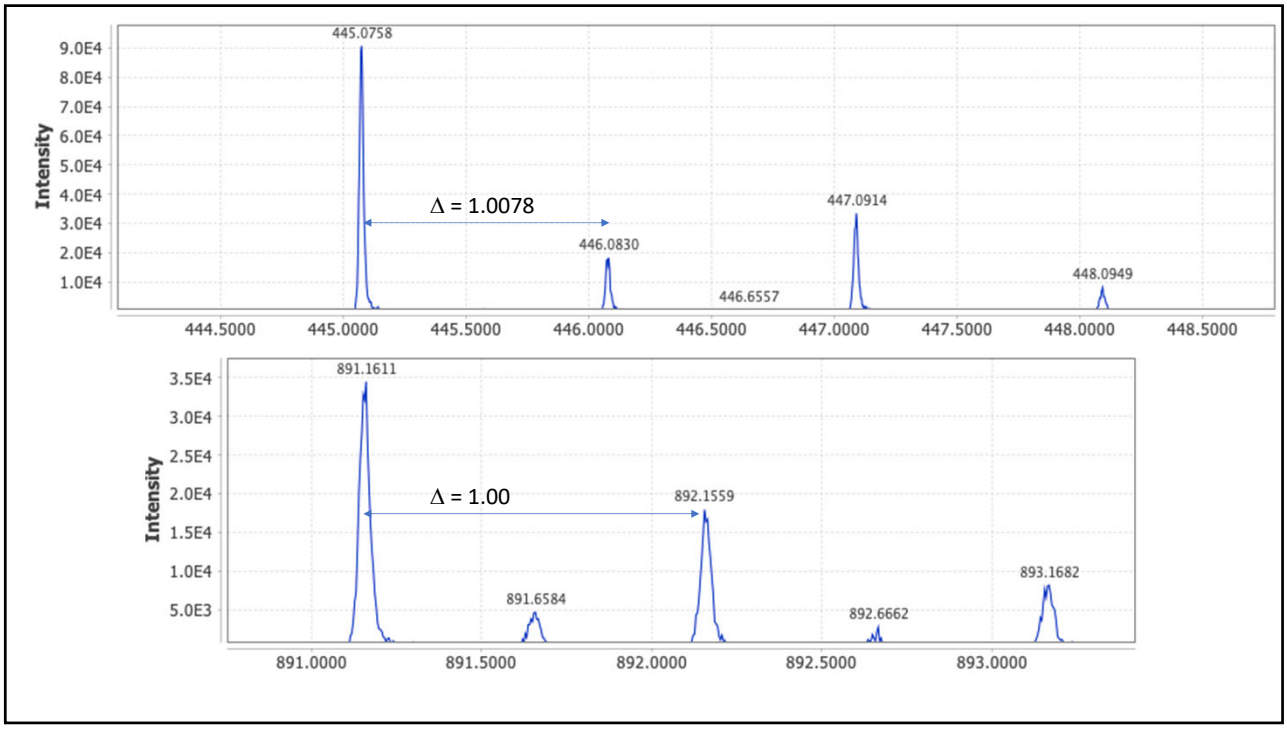
Spectrum at the top of the genistein GlcA peak



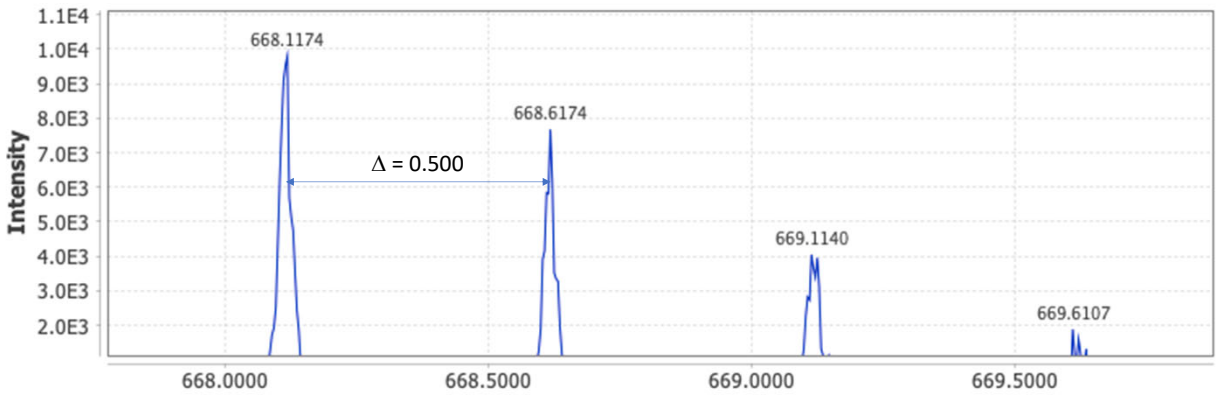
2


Base peak intensity
445.0789
445.0876
445.0852
445.0547
445.0526
[M-H]-
891.1563
891.1602
891.0976
891.1908
891.1740
891.1779
891.1542
891.1990
891.1442
[2M-H]-
668.1102
668.1170
668.1940
668.1466
668.2314
668.1644
668.3758
668.3333
668.9160
[??]-
Retention time

3


Intensity
445.0758
Δ = 1.0078
446.0830
446.6557
447.0914
448.0949
891.1611
Δ = 1.00
892.1559
891.6584
892.6662
893.1682

4



5

Resolving the *m/z* 445, 668 and 891 ions

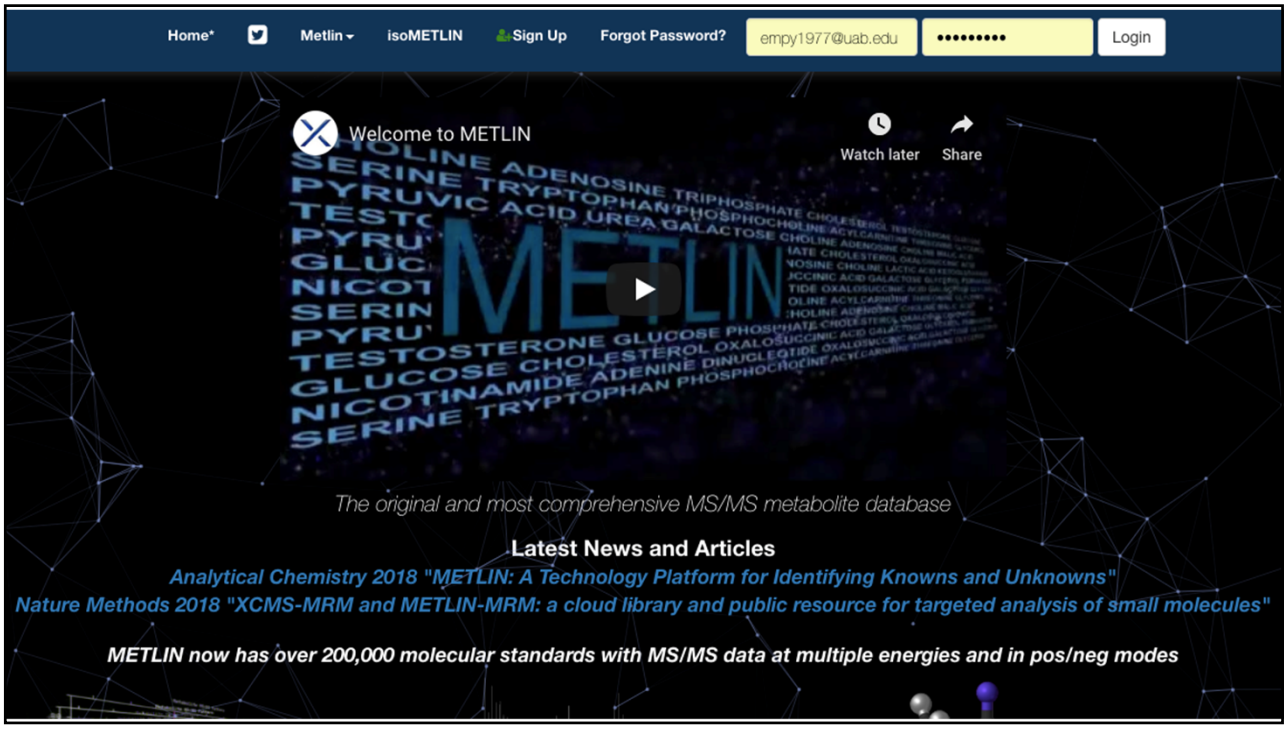
- ***m/z* 445.077 is genistein glucuronide $[M-H]^-$**
  - M is 446.086
- **$[2M-H]^-$ is *m/z* 892.172-1.008 = 891.164 (obs. *m/z* 891.163)**
  - The *m/z* 891-893 series are the 2M-H series with ^{13}C-isotopes
- **The *m/z* 668-669 series are doubly charged**
  - The ^{13}C-isotope spacing is 0.5
  - *m/z* 668.119 is $[M-2H]^{2-}$
  - M is therefore 2 x 668.119 + 2 x 1.0072 = 1336.238 + 2.014 = 1338.252
  - Divide 1338.252 by 446.086 = 3
- ***m/z* 668.119 is therefore $[3M-2H]^{2-}$**
- **All these ions are from genistein glucuronide**

6

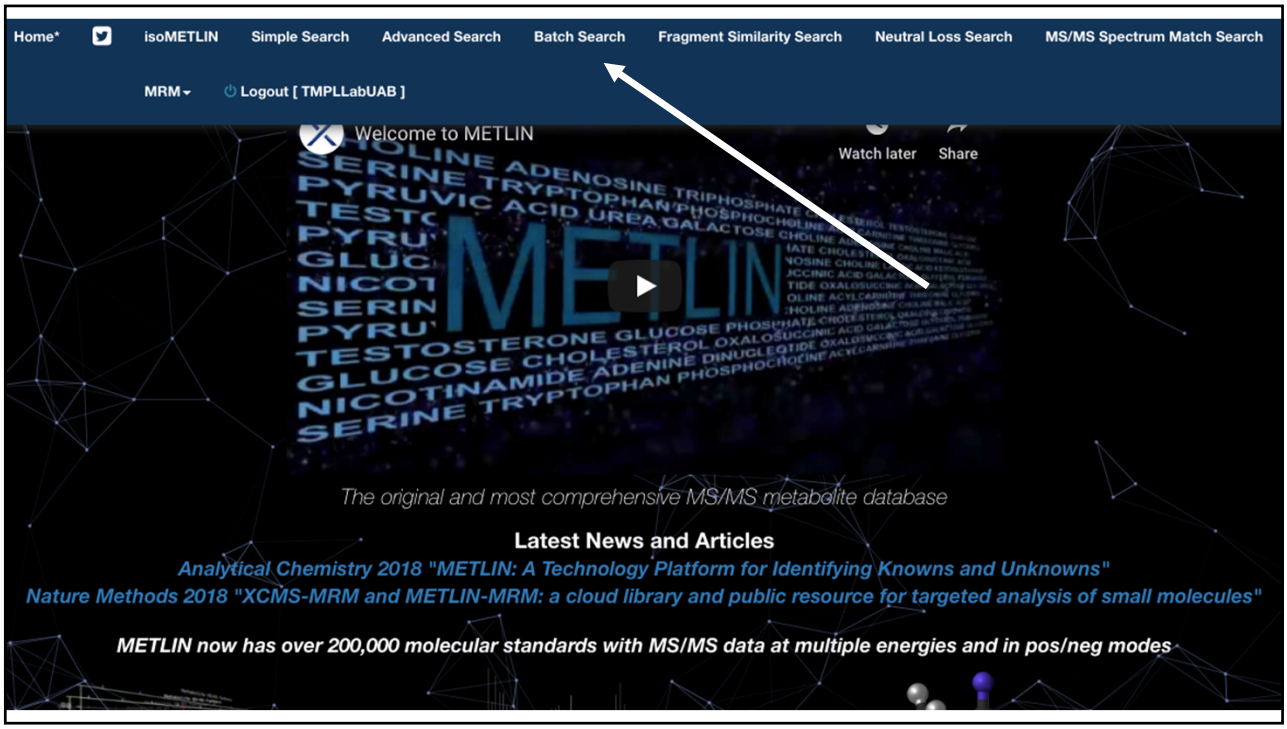
Now let's go to METLIN

Put METLIN in your browser
use your account information

7



8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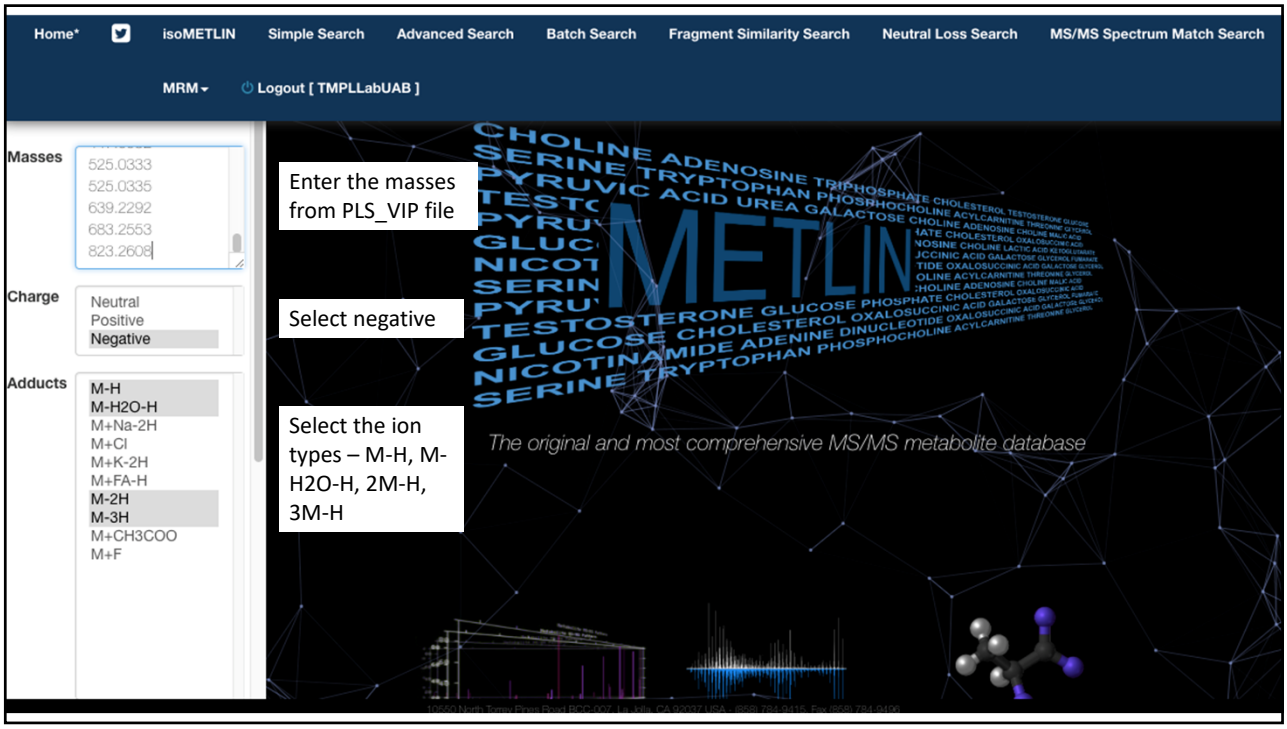
Latest News and Articles
Analytical Chemistry 2018 "METLIN: A Technology Platform for Identifying Knowns and Unknowns"
Nature Methods 2018 "XCMS-MRM and METLIN-MRM: a cloud library and public resource for targeted analysis of small molecules"
METLIN now has over 200,000 molecular standards with MS/MS data at multiple energies and in pos/neg modes

9


Home*
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MS/MS Spectrum Match Search
MRM
Logout [TMPLLabUAB]
Masses
525.0333
525.0335
639.2292
683.2553
823.2608
Charge
Neutral
Positive
Negative
Adducts
M-H
M-H2O-H
M+Na-2H
M+Cl
M+K-2H
M+FA-H
M-2H
M-3H
M+CH3COO
M+F
Enter the masses from PLS_VIP file
Select negative
Select the ion types – M-H, M-H2O-H, 2M-H, 3M-H
METLIN
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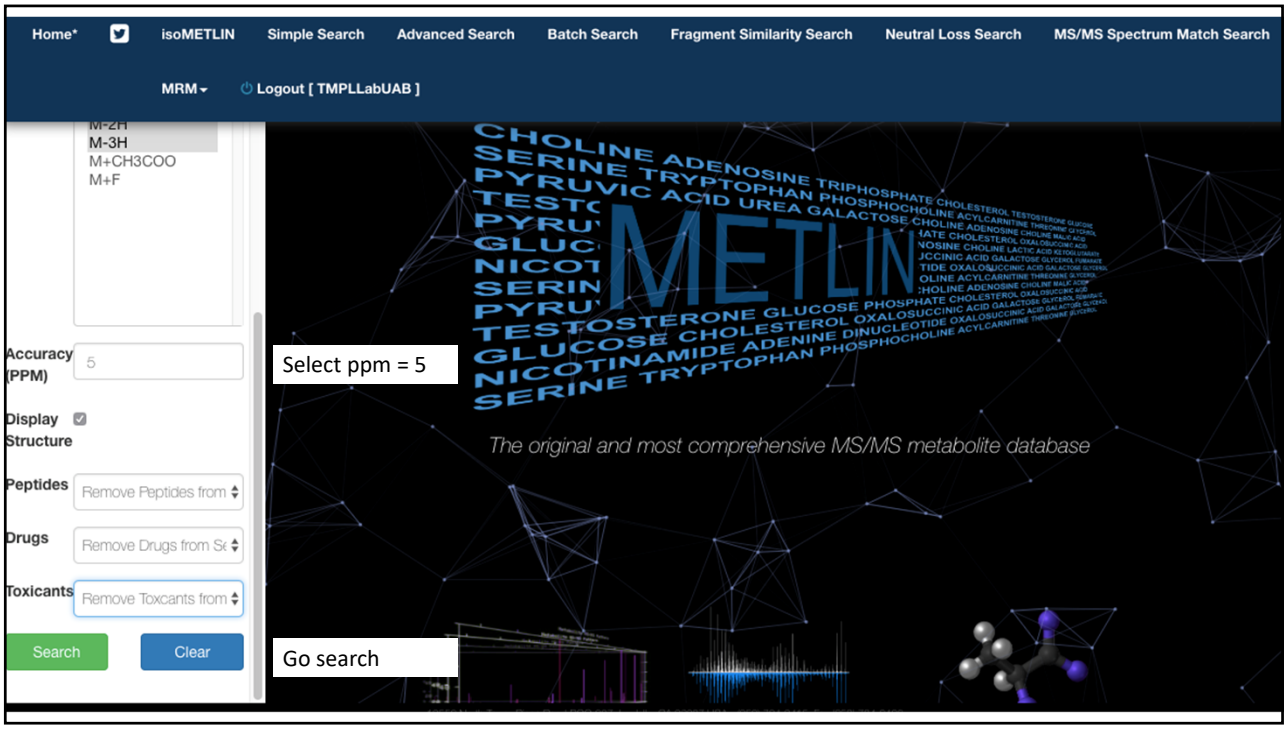
10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Logout [TMPLLabUAB]
M-3H
M+CH3COO
M+F
Accuracy (PPM)
5
Display Structure
Peptides
Remove Peptides from
Drugs
Remove Drugs from Se
Toxicants
Remove Toxcants from
Search
Clear
METLIN
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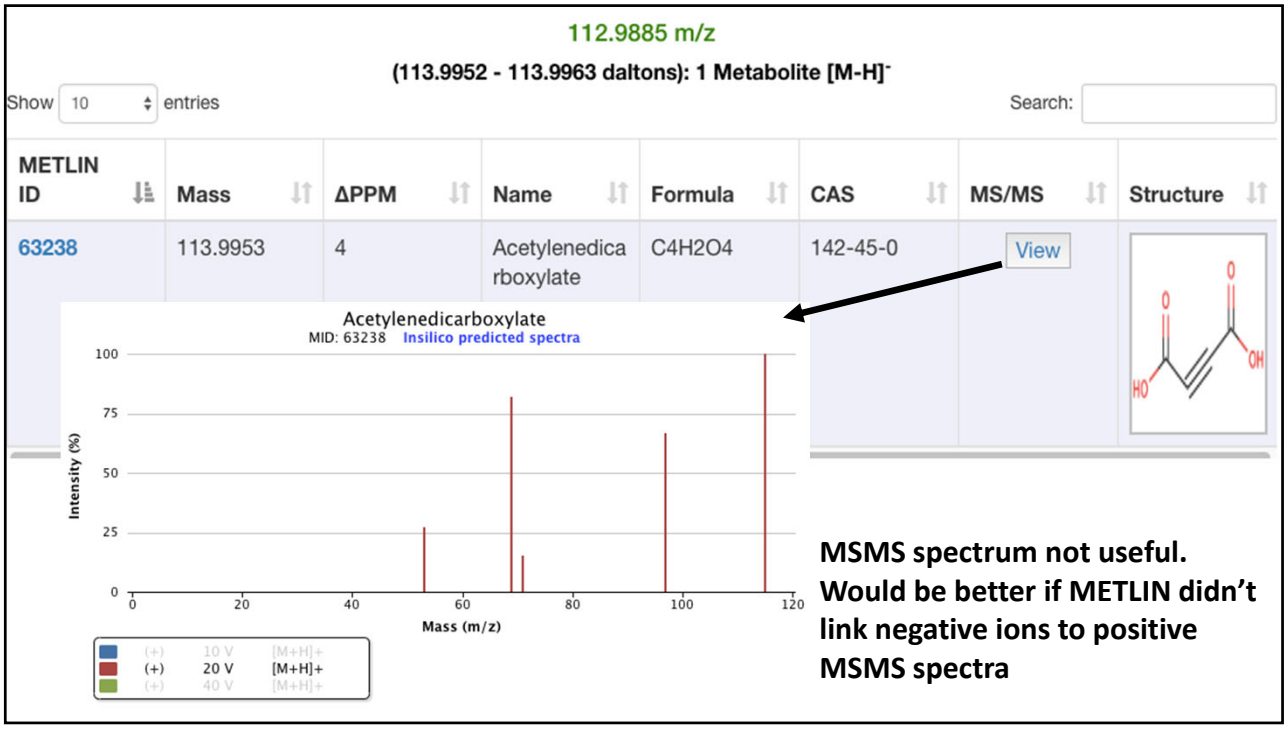
Select ppm = 5
Go search

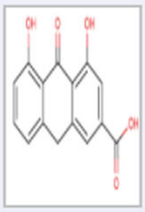
11


112.9885 m/z
(113.9952 - 113.9963 daltons): 1 Metabolite [M-H]-
Show 10 entries
Search:
METLIN ID
Mass
ΔPPM
Name
Formula
CAS
MS/MS
Structure
63238
113.9953
4
Acetylenedicarboxylate
C4H2O4
142-45-0
View
Acetylenedicarboxylate
MID: 63238 Insilico predicted spectra
Intensity (%)
Mass (m/z)
(+) 20 V [M+H]+
MSMS spectrum not useful. Would be better if METLIN didn't link negative ions to positive MSMS spectra

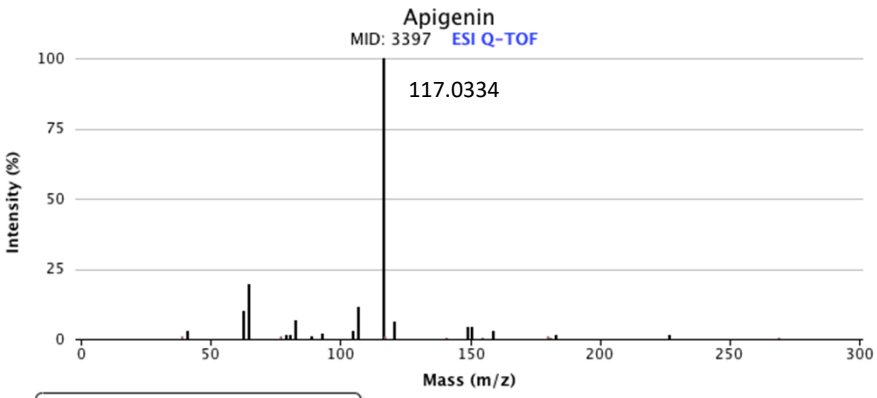
12

(270.0523 - 270.055 daltons): 29 Metabolites [M-H]⁻

Show 10 entries Search:

METLIN ID	Mass	ΔPPM	Name	Formula	CAS	MS/MS	Structure
2412	270.0528	3	Rhein-9-anthrone	C15H10O5	480-09-1	View	
3397	270.0528	3	Apigenin	C15H10O5	520-36-5	View	

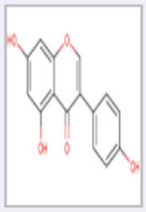
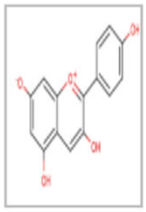
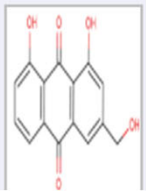
13



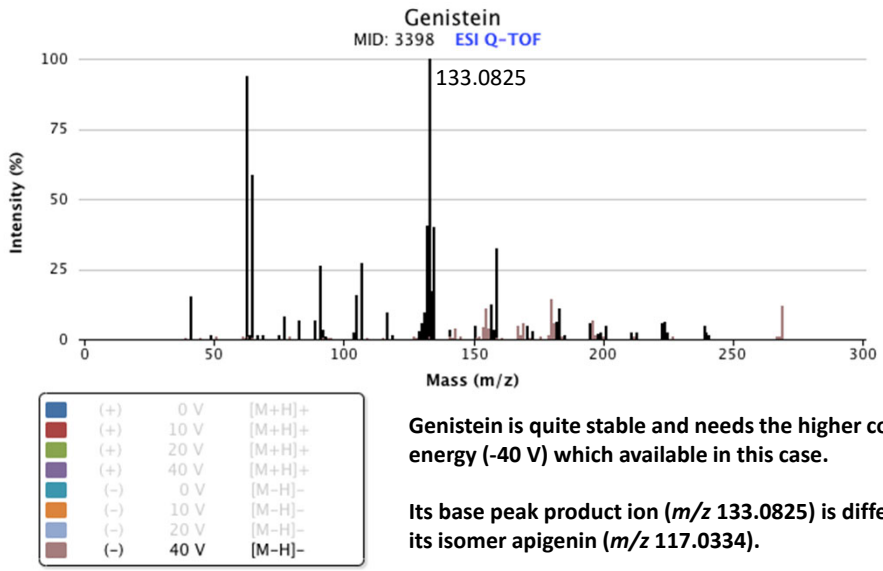
Apigenin is quite stable and needs the higher collision energy (-40 V) which available in this case.

The base peak product ion is *m/z* 117.0334

14

3398	270.0528	3	Genistein	C15H10O5	446-72-0	View	
3399	270.0528	3	Pelargonidin	C15H10O5	7690-51-9	View	
41039	270.0528	3	Aloe-emodin	C15H10O5	NA	View	

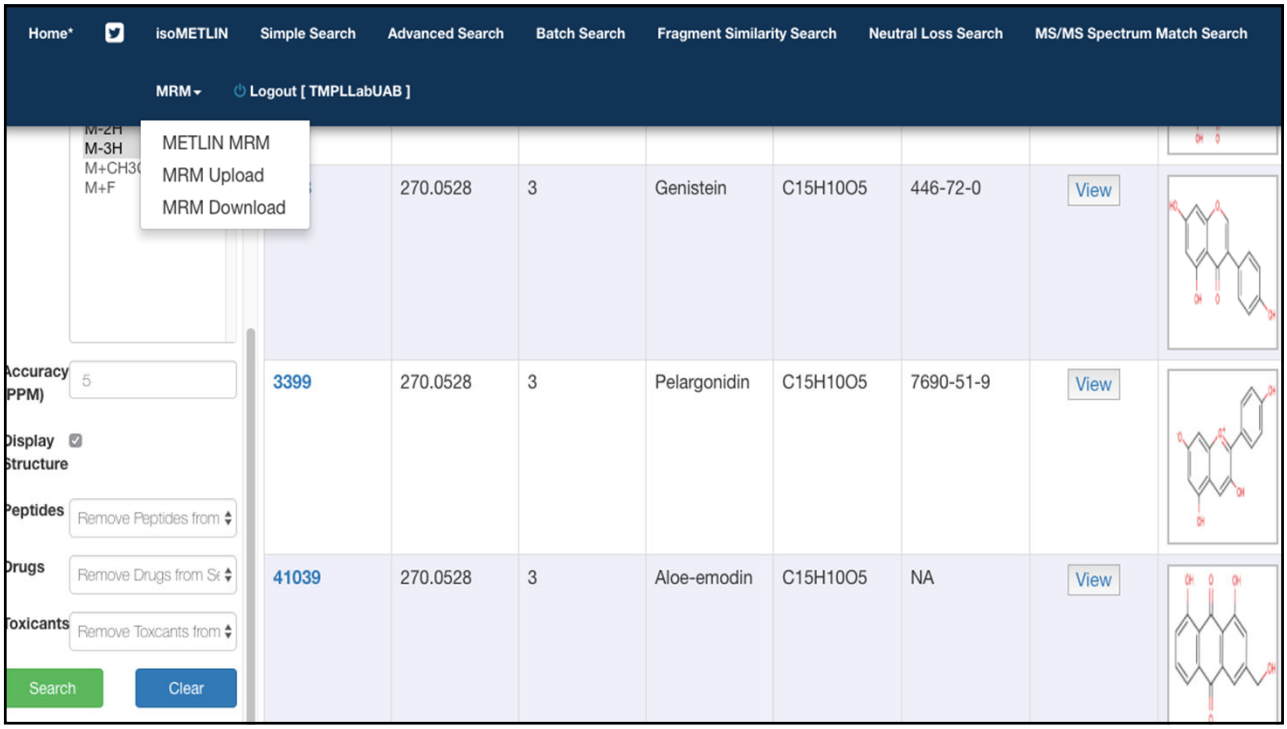
15



Genistein is quite stable and needs the higher collision energy (-40 V) which available in this case.

Its base peak product ion (*m/z* 133.0825) is different from its isomer apigenin (*m/z* 117.0334).

16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Neutral Loss Search
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MRM
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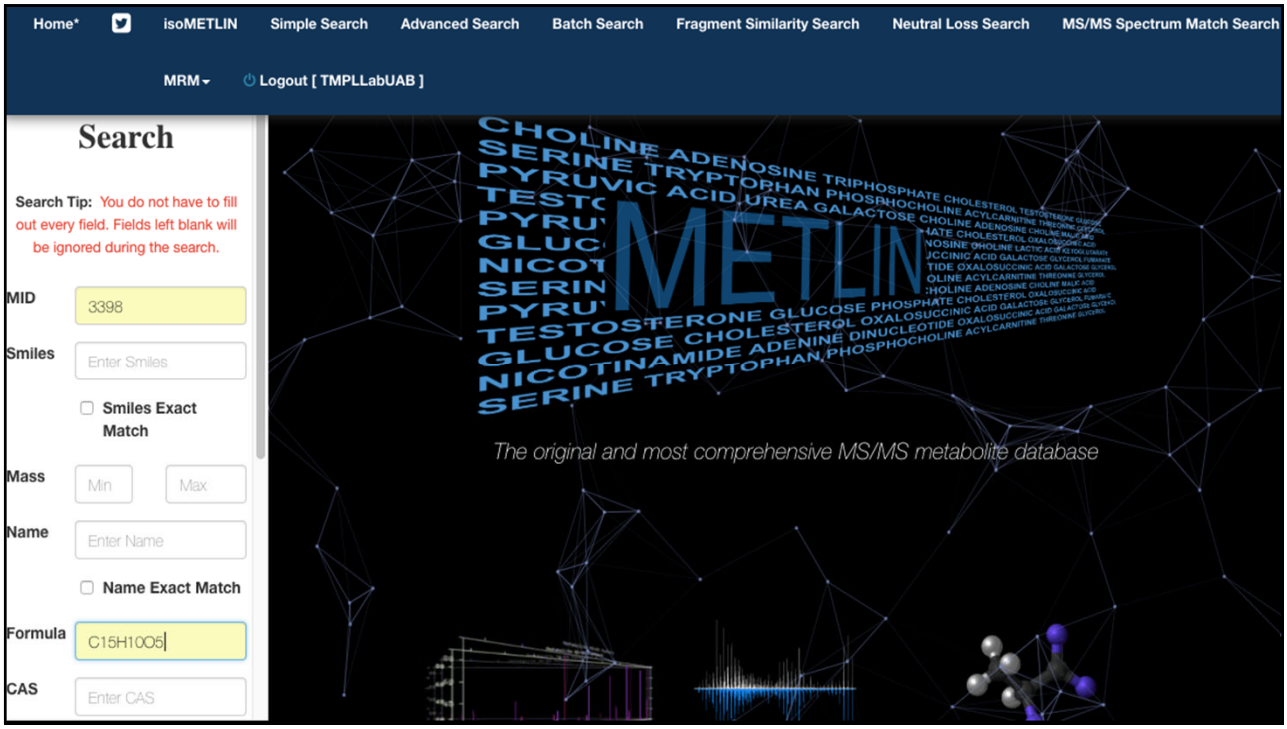
METLIN MRM
MRM Upload
MRM Download
M-3H
M+F
Accuracy (PPM)
5
Display Structure
Peptides
Remove Peptides from
Drugs
Remove Drugs from
Toxicants
Remove Toxcants from
Search
Clear
270.0528 3 Genistein C15H10O5 446-72-0 View
3399 270.0528 3 Pelargonidin C15H10O5 7690-51-9 View
41039 270.0528 3 Aloe-emodin C15H10O5 NA View

17


Home*
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Simple Search
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Batch Search
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Search
Search Tip: You do not have to fill out every field. Fields left blank will be ignored during the search.
MID
3398
Smiles
Enter Smiles
Smiles Exact Match
Mass
Min
Max
Name
Enter Name
Name Exact Match
Formula
C15H10O5
CAS
Enter CAS
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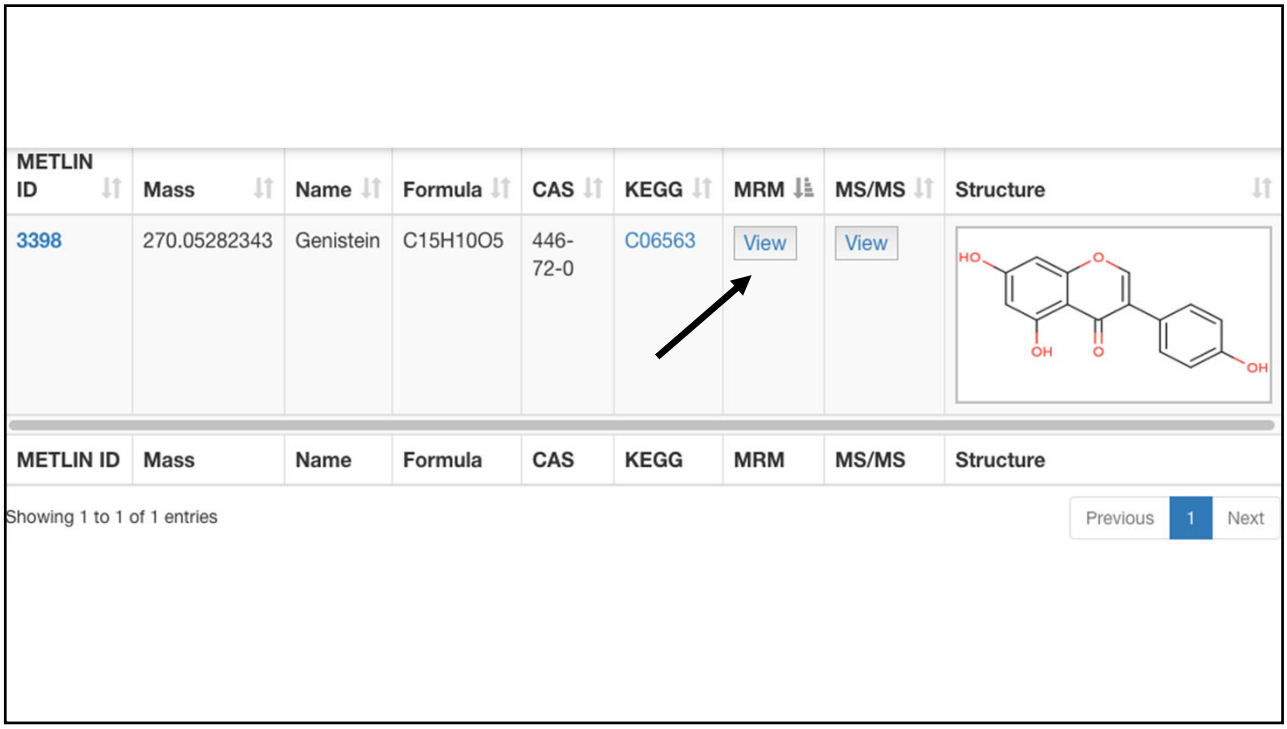
19



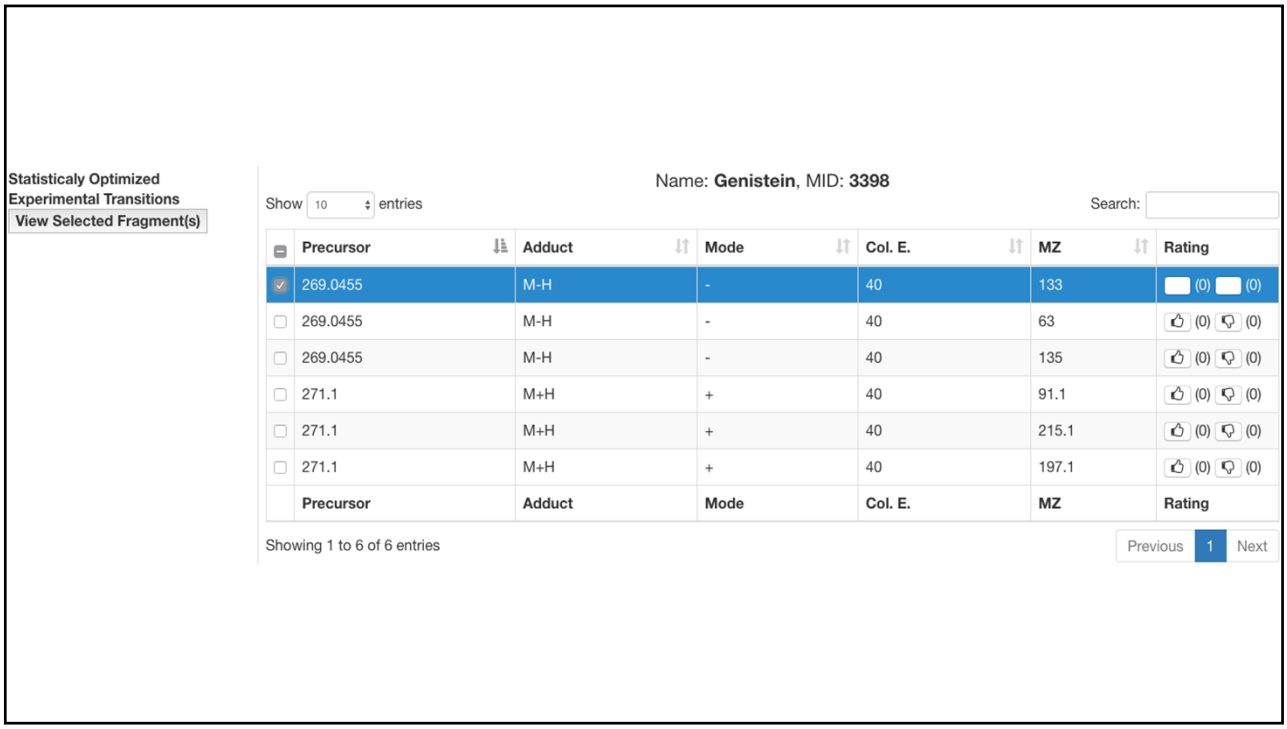
METLIN ID	Mass	Name	Formula	CAS	KEGG	MRM	MS/MS	Structure
3398	270.05282343	Genistein	C15H10O5	446-72-0	C06563	View	View	

METLIN ID	Mass	Name	Formula	CAS	KEGG	MRM	MS/MS	Structure

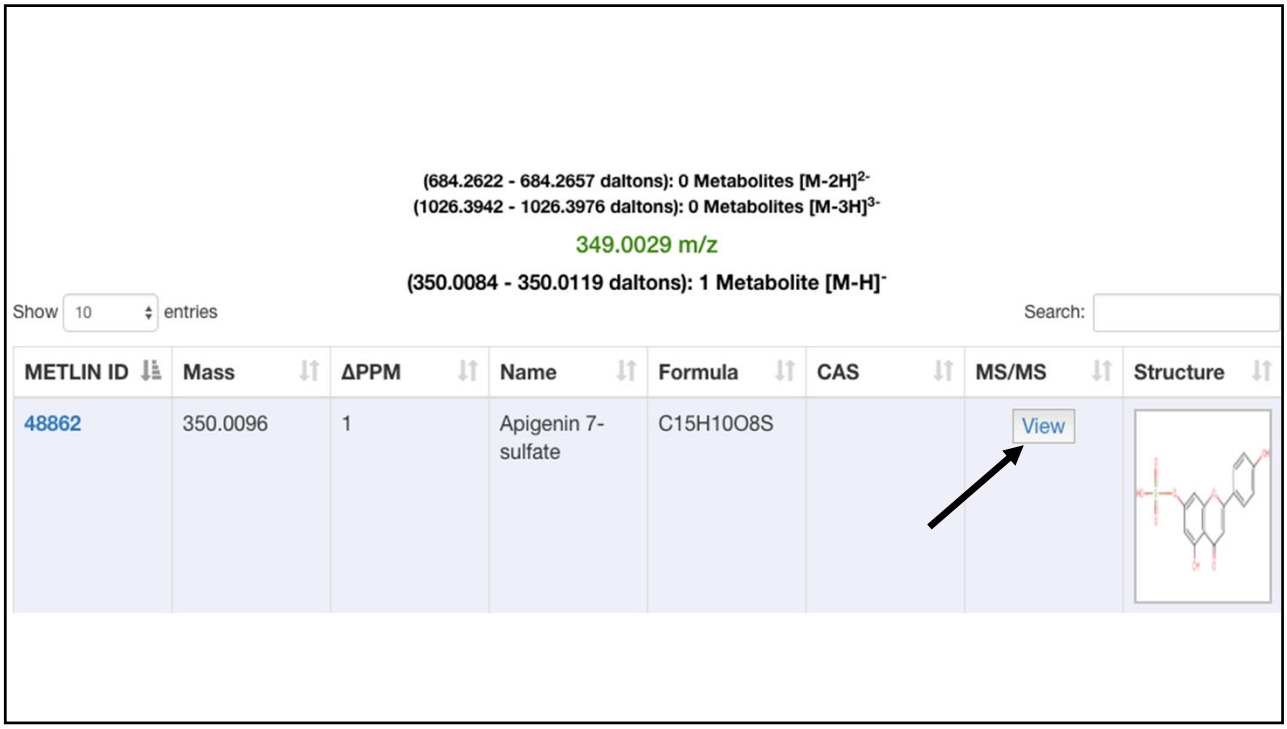
Showing 1 to 1 of 1 entries

Previous 1 Next

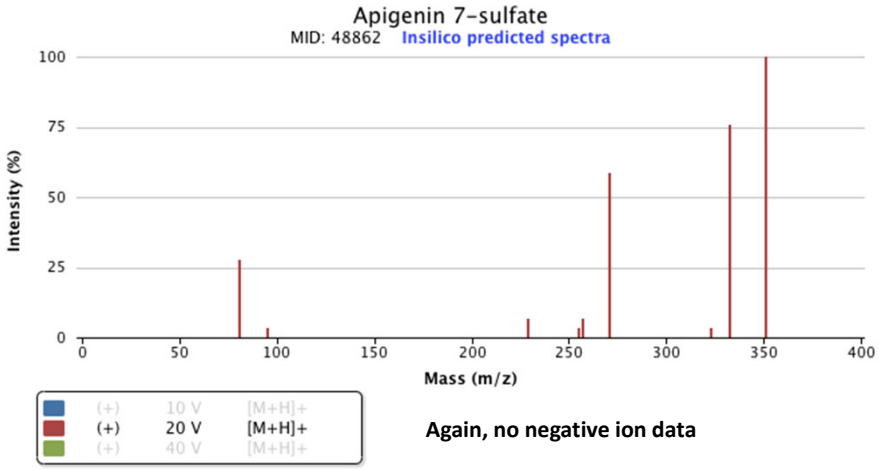
20


Statisticaly Optimized
Experimental Transitions
View Selected Fragment(s)
Name: Genistein, MID: 3398
Show 10 entries
Search:
Precursor | Adduct | Mode | Col. E. | MZ | Rating
269.0455 | M-H | - | 40 | 133 | (0) (0)
269.0455 | M-H | - | 40 | 63 | (0) (0)
269.0455 | M-H | - | 40 | 135 | (0) (0)
271.1 | M+H | + | 40 | 91.1 | (0) (0)
271.1 | M+H | + | 40 | 215.1 | (0) (0)
271.1 | M+H | + | 40 | 197.1 | (0) (0)
Showing 1 to 6 of 6 entries
Previous 1 Next

21


(684.2622 - 684.2657 daltons): 0 Metabolites [M-2H]2-
(1026.3942 - 1026.3976 daltons): 0 Metabolites [M-3H]3-
349.0029 m/z
(350.0084 - 350.0119 daltons): 1 Metabolite [M-H]-
Show 10 entries
Search:
METLIN ID | Mass | ΔPPM | Name | Formula | CAS | MS/MS | Structure
48862 | 350.0096 | 1 | Apigenin 7-sulfate | C15H10O8S | | View

22



Again, no negative ion data

23

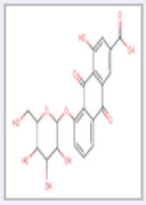
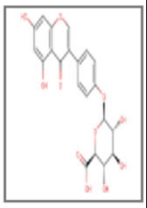
445.0773 m/z

(446.0824 - 446.0868 daltons): 12 Metabolites [M-H]⁻

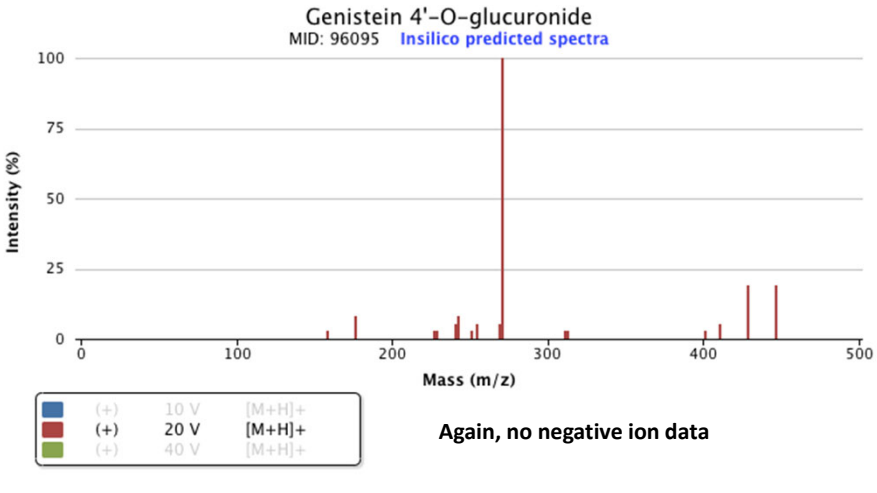
Show 10 entries Search:

METLIN ID	Mass	ΔPPM	Name	Formula	CAS	MS/MS	Structure
48550	446.0849	0	5,7,2'-Trihydroxyflavone 7-glucuronide	C21H18O11		View	
48775	446.0849	0	Apigenin 7-glucuronide	C21H18O11		View	

24

88791	446.0849	0	Glucorhein	C21H18O11	34298-86-7	View	
96095	446.0849	0	Genistein 4'-O-glucuronide	C21H18O11		View	

25



Again, no negative ion data

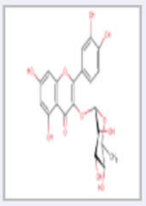
26

This is really the $^{13}C_2$-isotope ion of *m/z* 445.077

447.0932 m/z

(448.0982 - 448.1027 daltons): 74 Metabolites [M-H]⁻

Show 10 entries Search:

METLIN ID	Mass	ΔPPM	Name	Formula	CAS	MS/MS	Structure
43747	448.1006	0	Quercitrin	C21H20O11	522-12-3	View	

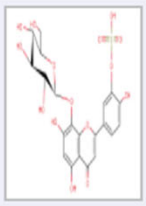
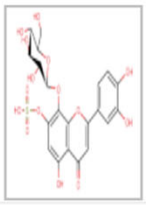
27

525.0333 m/z

(526.038 - 526.0432 daltons): 0 Metabolites [M-H]⁻

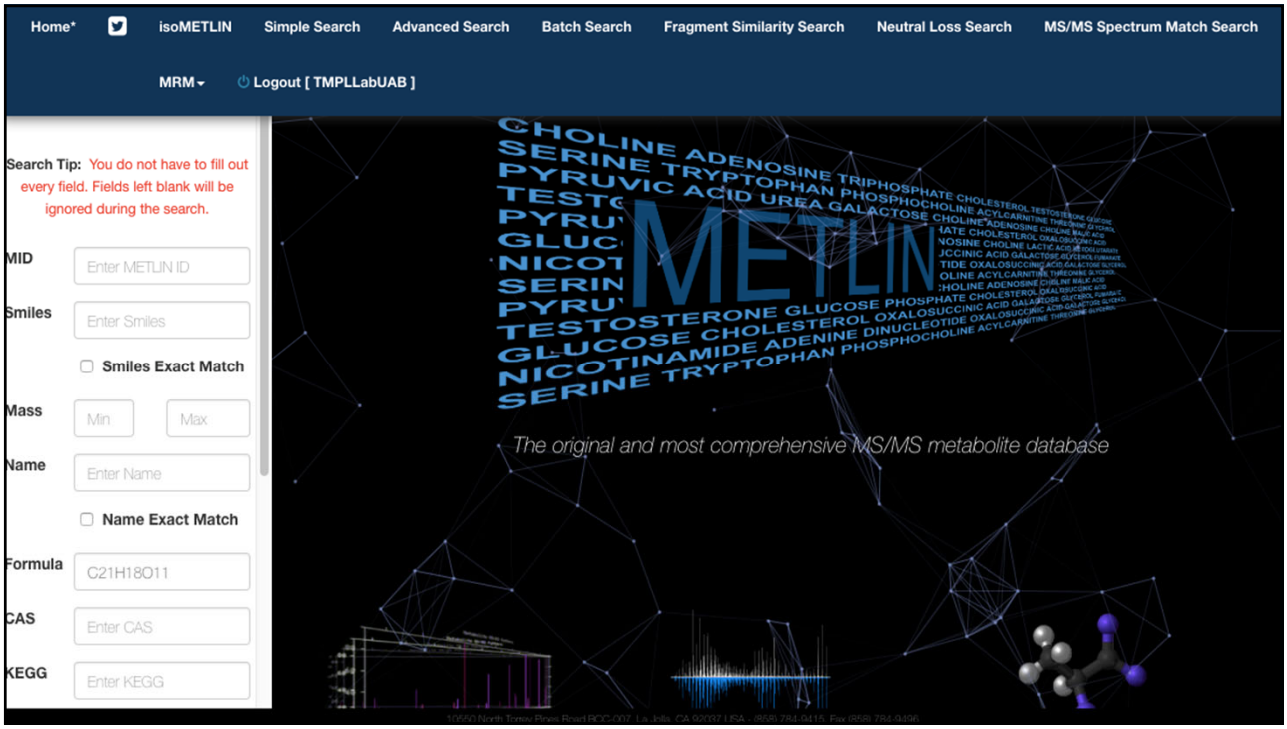
(544.0491 - 544.0543 daltons): 5 Metabolites [M-H2O-H]⁻

Show 10 entries Search:

METLIN ID	Mass	ΔPPM	Name	Formula	CAS	MS/MS	Structure
49817	544.0523	1	8-Hydroxyluteolin 8-glucoside-3'-sulfate	C21H20O15S		View	
49819	544.0523	1	Hypolaetin 7-sulfate-8-glucoside	C21H20O15S		View	

None of the five records are of genistein β-glucuronide sulfate

28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MRM
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MID
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Smiles
Enter Smiles
Smiles Exact Match
Mass
Min
Max
Name
Enter Name
Name Exact Match
Formula
C21H18O11
CAS
Enter CAS
KEGG
Enter KEGG
METLIN
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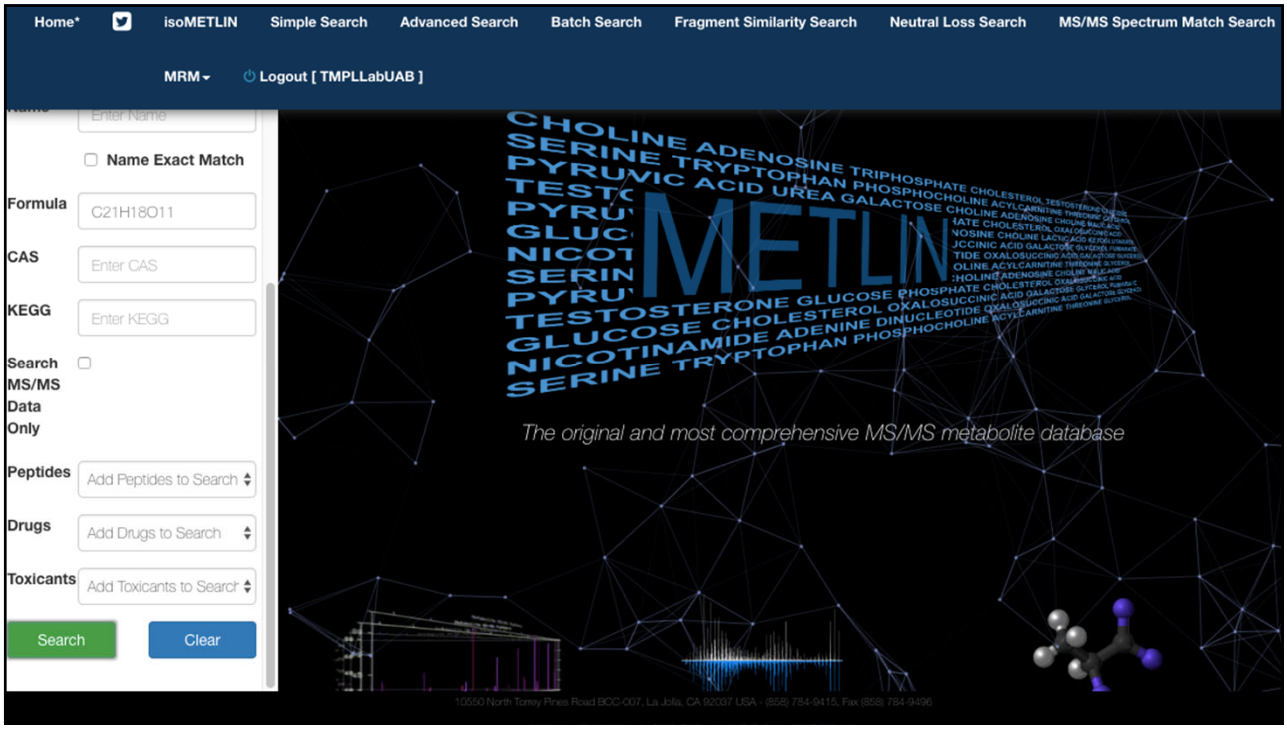
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Neutral Loss Search
MS/MS Spectrum Match Search
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Name Exact Match
Formula
C21H18O11
CAS
Enter CAS
KEGG
Enter KEGG
Search MS/MS Data Only
Peptides
Add Peptides to Search
Drugs
Add Drugs to Search
Toxicants
Add Toxicants to Search
Search
Clear
METLIN
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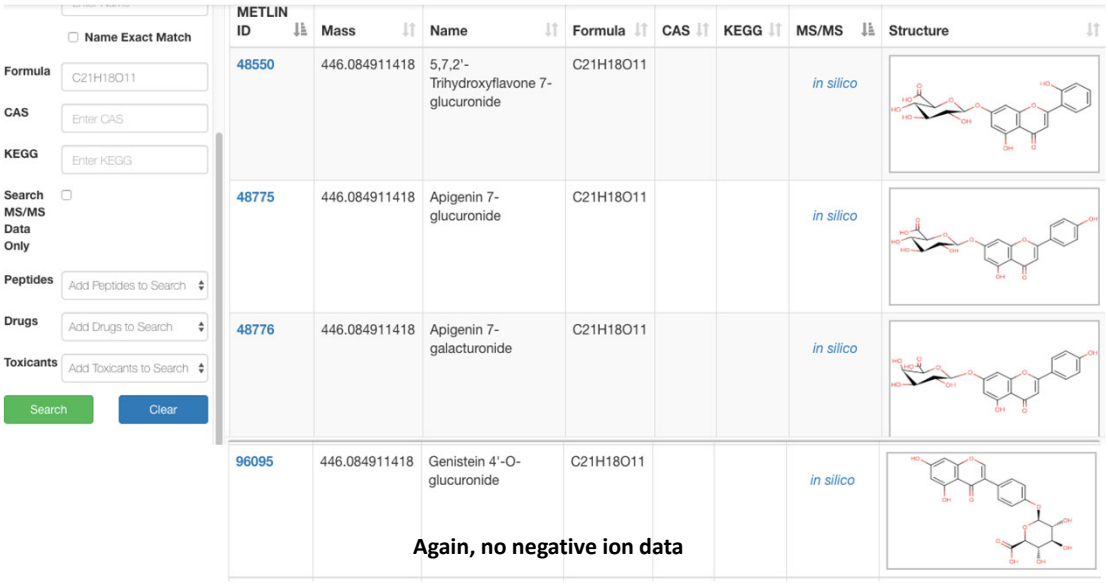
Name Exact Match
Formula
C21H18O11
CAS
Enter CAS
KEGG
Enter KEGG
Search MS/MS Data Only
Peptides
Add Peptides to Search
Drugs
Add Drugs to Search
Toxicants
Add Toxicants to Search
Search
Clear
METLIN ID
Mass
Name
Formula
CAS
KEGG
MS/MS
Structure
48550
446.084911418
5,7,2'-Trihydroxyflavone 7-glucuronide
C21H18O11
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446.084911418
Apigenin 7-glucuronide
C21H18O11
in silico
48776
446.084911418
Apigenin 7-galacturonide
C21H18O11
in silico
96095
446.084911418
Genistein 4'-O-glucuronide
C21H18O11
in silico
Again, no negative ion data

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