KnowItAll Mass Spectrometry Training

Vendor Neutral Data Processing Solution for Spectral Analyses

WILEY

KnowItAll™

"Novel Wiley Adaptive MS Search for the Identification of Unknowns"

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Handouts for Videos:
Website: "Little Mass Spec and Sailing"
https://littlemsandsailing.wordpress.com

Note: In-depth training videos/handouts on this new mass spec software on my website!

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KnowItAll Adaptive Search (*Patented*)

- Based on initial research found in NIST hybrid search reference*
- Finds similar compounds where a molecular fragment is *absent* or *present* in the reference spectrum compared to the unknown
- Presence or absence designated by its Δmass, difference of nominal molecular weight of known vs. reference
- Peaks in the unknown spectrum compared to the reference spectrum *shifted* by Δmass
- The "*blended*" hit quality index value reflects *both* the shifted and unshifted peaks

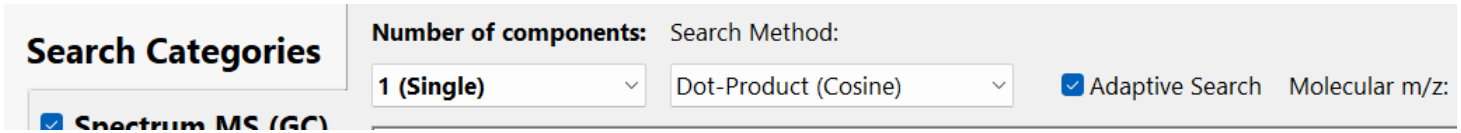
Results: Adaptive search *routinely* yields *useful results* not found with the more traditional identity search if the component is *not present* in the reference libraries

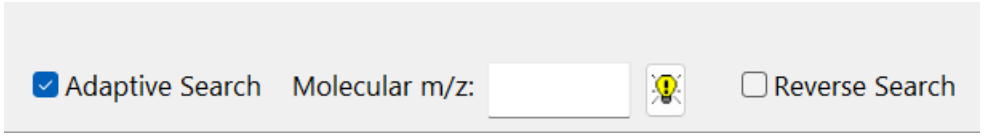
Thus: Greatly extends the *effectiveness* of *all* reference and *even* user libraries

*"Combining Fragment-Ion and Neutral-Loss Matching during Mass Spectral Library Searching: A New General Purpose Algorithm Applicable to Illicit Drug Identification," A. Moorthy, W. Wallace, A. J. Kearsley, D. Tchekhovskoi, and S. Stein, *Analytical Chemistry* **2017** *89* (24), 13261-13268.

Setup for Adaptive Search

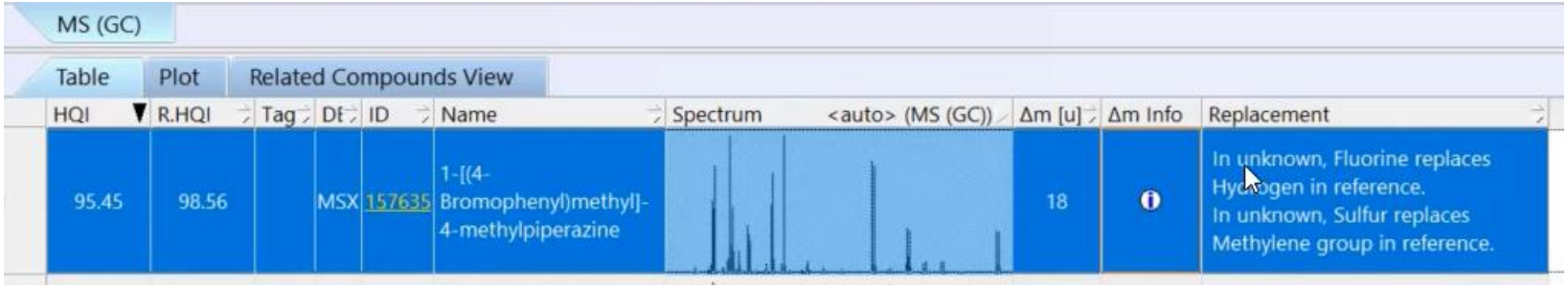
- Setup of libraries searched and advanced parameters same for the standard similarity and adaptive search the same, see previous information for the former in previous handout and video
- Again specify the single dot-product (Cosine) and this time selected Adaptive Search
- Adaptive search works best when the molecular weight is specified, so enter in the value in the molecular m/z box
- If unsure, let the program determine and populate the box selecting the "light bulb" next to the Molecular m/z field
- Send the spectrum to search using the "Search" button at bottom of page





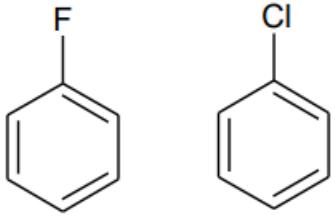
Viewing Results of the Adaptive Search

- Many of the results for the adaptive search and standard similarity search are the same
- The differences are the last three fields to the right
- The first is the Δm [μ\ field which shows the difference in the nominal molecular weight of the known versus the unknown
- The second is the "Δm info" field which will show the user how the reference spectrum was changed to get the observed HQI (hit quality index) for the adaptive search result
- The last column, "Replacement," gives some possible clues for the elements or substructure elements that yield the observe Δm
- *All this better explained in video*



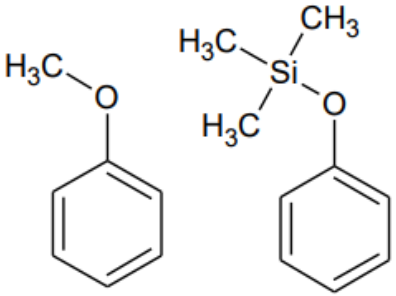
Associating Some *Simple* Structures with DeltaMass Values

- Some simple ***small*** MW compounds to illustrate types of substructural information
- Of course, these substructures can be a part of ***much larger*** molecules
- ***Note:*** Odd values of DeltaMass contain one nitrogen change in structure, thus ***"Nitrogen Rule"***
- Isotope ratios and/or accurate mass helpful with redundancies



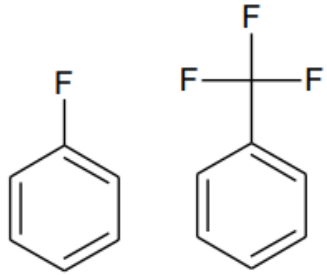
MW 96 MW 112

DeltaMass 16



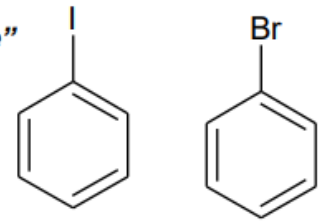
MW 108 MW 166

DeltaMass 58



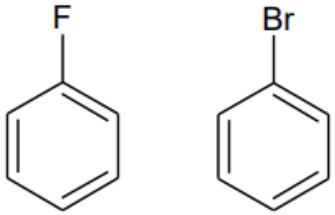
MW 96 MW 146

DeltaMass 50



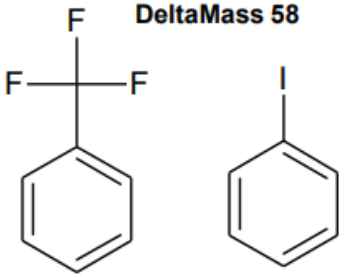
MW 204 MW 156

DeltaMass 48



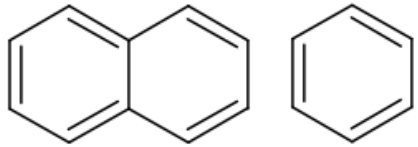
MW 96 MW 156

DeltaMass 60



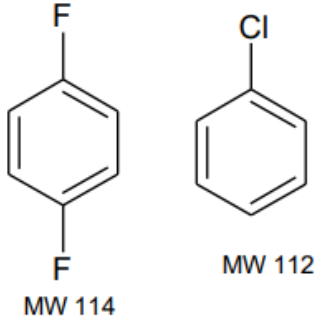
MW 146 MW 204

DeltaMass 58



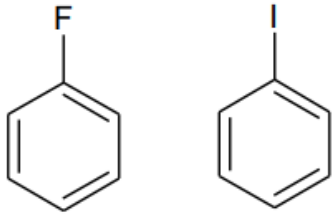
MW 128 MW 78

DeltaMass 50



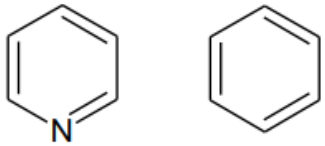
MW 114 MW 112

DeltaMass 2



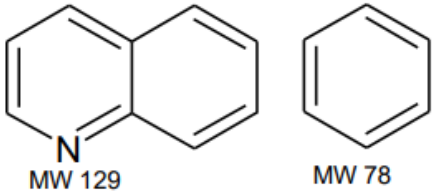
MW 96 MW 204

DeltaMass 108



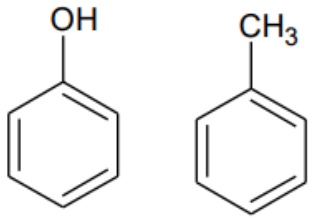
MW 79 MW 78

DeltaMass 1



MW 129 MW 78

DeltaMass 51



MW 94 MW 92

DeltaMass 2

Some Other Resources on the Internet for Wiley Adaptive and NIST Hybrid Searches

- Many of the resources for the NIST hybrid search are useful for Wiley Adaptive Search
- Slightly different results will be noted though when comparing the results
- The adaptive search is easier to visualize using the "Δm info" window than NIST's multicolor display
- The NIST search ***does not*** have a "Replacement" type field to suggest changes in the structure

Click on the Hyperlinks below:

1. Combining Fragment-Ion and Neutral-Loss Matching during Mass Spectral Library Searching, NIST Reference, Stein et. al
2. Real World Wiley Adaptive Example Video for fentanyl related species
3. Real World Wiley Adaptive Example Video for fentanyl related species
4. NIST Hybrid Example of Illicit Drug
5. Slides used for NIST Illicit Drug Video
6. NIST Hybrid Search Training Video
7. NIST Hybrid Search Training Handout
8. Delta Mass Table of Common Values Noted in Excel Format

Conclusions

- Greatly extends the effectiveness of all EI libraries in the characterization of unknowns compared to standard similarity search
- Standard search when unknown present in library *directly* yields proposed structure
- Adaptive research yields *clues* that allow user to propose structure (s)
- Always consider sample history
- Take into consideration chemical ionization, accurate mass, IR, NMR, mechanistic data
- Often more than one useful delta mass noted