ASMS 2019 Annual Meeting: Metabolomics Evening Workshop

Moderators: Gary Patti and Jon Sobus
Guest Speakers: Dean Jones, James Cox, Oliver Fiehn, Lloyd Sumner

Jon Sobus (US EPA) and Gary Patti (Washington University) opened the workshop by discussing that while the field of metabolomics has matured over the past decade, lingering issues exist upon which consensus has not been reached. Perhaps the most glaring topic of dissent relates to the acceptance and implementation of reporting levels for metabolite identification. Various reporting strategies have been proffered by experts in the fields of metabolomics and environmental chemistry. Yet, workshop attendees agreed that no single method is universally accepted. Perhaps more importantly, it was acknowledged that no single method is universally implemented. This lack of consistency in adoption and implementation has contributed to an inability of those in the metabolomics field to confidently communicate research findings to scientists, policymakers and the public. To underscore the importance of this issue, four prominent scientists in the field of metabolomics were invited to present their views on metabolite ID practices.

Dean Jones (Emory University) first described strategies used by his laboratory to identify/report on known and putative metabolites. Strategies described by Professor Jones include using computational tools and known metabolic pathways to: 1) predict metabolites of known parent compounds; 2) generate theoretical spectra for predicted metabolites; and 3) match experimental spectra for unknowns against theoretical spectra for predicted metabolites to generate lists of putative identifications. Professor Jones argued that these hits, when rooted in known metabolic/enzymatic pathways, can be annotated with the highest levels of confidence. This notion was met with some resistance by the audience, with several participants arguing that verification with reference standards will always be needed to achieve the highest level of confidence.

James Cox (University of Utah) emphasized in his presentation the need to have targeted validation of any non-targeted discovery. Specifically, Professor Cox clearly outlined the default analytical boxes that must be checked in order to reach the highest level of confidence for metabolite ID; these include reference standard matching based on accurate mass, retention time, and MS/MS pattern. Professor Cox further stressed the value in reference standard matching using orthogonal separation techniques, and orthogonal detection methods (e.g., ELISA vs. MS vs. NMR). Finally, Professor Cox urged the audience to always think critically about the biological system when making a metabolite identification. Simply put, investigators should weigh a plausible biological rationale when determining confidence for an identified metabolite.

Oliver Fiehn (University of California, Davis) conveyed in his presentation that, while refinements to metabolite ID criteria have been made over the past decade, the current paradigm of subjective reporting is not ideal. Professor Fiehn proposed the development of software that could be used to automate metabolite ID and reporting. As described by Professor Fiehn, an effective software tool would benefit from: 1) improved MS/MS predictions for specific compound classes; 2) decoy MS/MS libraries that enable calculation of false-discovery rates; 3) improved MS scoring algorithms that consider mass error, isotope peaks, and retention time error; 4) considerations for spectral matching (library vs. experimental) based on instrument type and energy; 5) high-resolution ion mobility to distinguish positional isomers; and 6) consideration for the biological plausibility of the identified metabolites.

Lloyd Sumner (University of Missouri) provided a succinct history of metabolite ID efforts. Specifically, Professor Sumner first summarized the four levels of metabolite ID as defined in 2007 by the Chemical Analysis Working Group (CAWG) Metabolomics Standards Initiative (MSI) (https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3772505/). He then contrasted these levels with those proposed by Schymanski and colleagues in 2014 (https://pubs.acs.org/doi/abs/10.1021/es5002105). While both of these reporting methods were based on a series of confidence levels, Professor Sumner also described a more refined series of "quantitative and alphanumeric metabolite identification metrics" that were proposed in 2014 (https://link.springer.com/article/10.1007/s11306-014-0739-6). Finally, Professor Sumner described ongoing efforts of a Metabolite ID Task Group who have proposed a refined 7-level scheme for classifying metabolite IDs based on confidence levels.

Professor Sumner led the final discussion of the workshop, which focused on two key questions: 1) Does everyone agree that confidence reporting for metabolite ID is necessary?; and 2) Are metabolomics practitioners willing to abide by the general recommendations going forward? The workshop participants seemed unanimous in the answer to question #1; metabolite ID levels are necessary to communicate results of metabolomics studies. The participants also generally agreed that metabolomics practitioners must work closely with co-investigators, media, journal editors, funding agencies, etc. to ensure that assigned ID levels are interpreted correctly by those who would use these data as a decision support tool. Regarding question #2, most participants seemed willing to abide by general recommendations for reporting, but it was noted that a balance must be struck in which reporting levels are clearly enough defined to minimize ambiguity, but not so refined and complex that they discourage widespread adoption. Dr. Sobus and Professor Patti closed the meeting by reflecting on the workshop presentations and encouraging continued discussion amongst guest presenters and attendees. The evening's spirited discussions surely provided a look into areas of future discussion as refinements to metabolite ID levels are further considered.