Fusarium wilt of banana, is one of the major constraints on global banana production. Recently, its causal agent, *F. oxysporum* f sp. *cubense*, was re-classified and divided over eleven distinct species; including *F. odoratissimum,* also known as Tropical Race 4 (TR4) which is highly aggressive on Cavendish bananas but also poses a significant threat to many other regionally important banana varieties, such as those in Cuba. Since, it is unknown which *Fusarium* species are present in Cuba, we sampled symptomatic banana plants across the country at geographically and environmentally different locations and obtained a collection of 166 *Fusarium* isolates. We used DArT genotyping-by-sequencing technology to explore genetic diversity across this suite of isolates and compared it with the diversity in a global panel of *Fusarium* strains. Our analysis revealed that the Cuban *Fusarium* strains infecting bananas belong to the species: *F. purpurascens*, *F. tardicrescens* and *F. tardichlamydosporum* and to races 1(R1) and 2(R2). The sampling did not reveal any TR4 strain in Cuba. The distribution of the three species throughout the country was associated with the cultivated banana varieties in each region. Oxford Nanopore DNA sequencing of a representative isolate of each species resulted in (near) chromosome-level genome assemblies of 50.6, 51.2 and 52.6Mbp with 14, 16 and 18 contigs, respectively. Whole-genome comparisons revealed that R2 isolates of *F. tardicrescens* and *F. tardichlamydosporum* from Cuba, contain two accessory chromosome that could not be identified in the genome assemblies of *F. purpurascens* (R1) nor in TR4, suggesting that these extra chromosomes are specific for R2 isolates. Thus, we here provide the first report on the genomic diversity and genomic structure of banana infecting *Fusarium* species across Cuba, which will facilitate the identification of effector candidates that are crucial for disease development.

582V ***Fusarium musae*** **diversity from a mitochondrial comparative perspective** *Valeria Tava*[1], Degradi Luca[1], Kunova Andrea[1], Pizzatti Cristina[1], Cortesi Paolo[1], Saracchi Marco[1], Vande Velde Greetje[2], Pasquali Matias[1] 1) University of Milan, Department of Food, Environmental and Nutritional Sciences, Milan, Italy; 2) KU Leuven, Department of Imaging and Pathology, Biomedical MRI unit/ MoSAIC, Leuven, Belgium.

Fusarium musae is a pathogenic species, previously misclassified as *F. verticillioides*, described in 2011 and belonging to the *Fusarium fujikuroi* species complex. It has the ability to infect taxonomically distant hosts: banana fruits mostly in Central and South America as well as human patients in Europe and USA. We studied a worldwide collection of 19 *F. musae* strains isolated from banana and human hosts in central America, northern America and Europe. We thereby verified the ability of the different strains to cause infection on banana fruits and *Galleria mellonella* as "human proxy". All strains were able to cause comparable levels of infection in both hosts. Sequencing and comparative studies of the mitochondrial genomes in *F. musae* led to the identification of a specific endonuclease polymorphic site that allows the distinction from *F. verticillioides* and that differentiates *F. musae* geographic subgroups. By analysing the distribution of the endonuclease polymorphism in *F. musae* we could potentially trace the geographic origin of the clinical infections in Europe and US. We did not find any correlation between the mitochondrial subgroups and their pathogenic activity in the two hosts.

583V Tandem-approach of direct-infusion HRMS and LC-QTOF-MS for the evaluation of food safety and useful secondary metabolites in *Aspergillus oryzae* *Sharon Marie Bahena-Garrido*[1], Ryota Saito[1], Yuko Komatsu[1], Ken Oda[1], and Kazuhiro Iwashita[1] 1) National Research Institute of Brewing, Higashi-Hiroshima, Japan.

Aspergillus oryzae has a plenty number of secondary metabolite gene clusters (SMGCs) of unknown functions and its investigation on genome and secondary metabolite (SM) production particularly on mycotoxins is still limited. There is also a wide array of *A. oryzae* species used in the brewing industry, therefore it is necessary to evaluate the safety of the entire *A. oryzae* which is closely related to *Aspergillus flavus*-notorious for its aflatoxin production, as well as to explore the potential wealth of useful SMs among *A. oryzae* species. In detail, there were 13 *A. oryzae* strains selected based on our previous phylogenetic tree and these strains along with *A. flavus* NRRL3357 were grown in various culture conditions, including rice-*koji a*nd soy sauce-*koji.* The SMs from the extracted fractions were analyzed by adopting a tandem-approach of direct-infusion high-resolution mass spectrometry (DI-HRMS) based metabolomics for efficient, high-throughput screening of metabolites and liquid chromatography quadrupole time-of-flight mass spectrometry LC-QTOF-MS (MS/MS) for further metabolite validation.

In the first approach, DI-HRMS analysis focused on 21 mycotoxins regulated by Joint FAO/WHO Expert Committee on Food Additives (JECFA). Aflatoxin B2 putatively detected in soy sauce-*koji* condition, aflatoxin G2 in corn and citrinin, ergot alkaloids among others detected in different conditions were further validated by LC-QTOF-MS (MS/MS). Results revealed no significant traces of 21 mycotoxins found in all 13 *A. oryzae* strains grown in various conditions. In the second approach, DI-HRMS analysis detected putative SMs which were further subjected to multivariate analysis to determine the SM production pattern resulting from diverse responses among the species. Distinct SM pattern was observed among the strains particularly in *A. oryzae* RIB40, RIB128, RIB915, RIB1172 grown in rice-*koji* and in RIB301, RIB915, RIB1108 grown in soy sauce-*koji* conditions possibly contributed by varying putative production of useful known and nonelucidated SMs. Furthermore, it was observed that *A. flavus* was clearly separated among the *A. oryzae* when grown in corn, YES and CYA suggesting the production of aflatoxins as well as other metabolites likely induced by plant material and nutrient-rich culture media under laboratory conditions.

Taken together, the efficient tandem-approach of metabolomic analysis in various growth conditions provides a plethora of candidate metabolites such as possible novel biomarkers useful for rapid discrimination between *A. oryzae* and the aflatoxigenic *A. flavus* as well as the interesting SM candidates produced by the dependable *A. oryzae* for promising pharmaceutical and other bio-industrial uses.

584V Can the quality of ITS regions in genome assemblies be trusted? *Barbara Robbertse*[1] 1) National Center for Biotechnology Information, National Library of Medicine, National Institutes of Health, Bethesda, MD.

The internal transcribed spacer (ITS) region of the nuclear ribosomal cistron is the primary barcode marker for Fungi and together with any taxon-specific secondary barcodes, serve to help identify species. The ITS marker is widely used in biodiversity studies, which depend on accurate reference databases for identification. The RefSeq ITS database at NCBI curates ITS PCR amplicons from type material specimens, submitted to GenBank. Since sequences from type material provide the most unambiguous link to species names it is generally recommended that taxonomists publicly deposit ITS sequences in addition to sequences from multiple other markers when describing new species. A recent trend in new species descriptions is that only genome assemblies are included. In addition, the