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Annual Congress on

Mycology and Fungal Infection

November 16-17, 2017 Atlanta, Georgia, USA

Poster Presentations
Day1

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Variation of anti-fungal saponin concentration in *Apostichopus japonicus*

Akira Yano

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Apostichopus japonicus is an edible sea cucumber inhabits the coastal waters of Japan. The sea cucumber has been eaten over thousands of years. In China, dried sea cucumbers are expensive but popular medicinal foods. In Japan, slice of raw sea cucumber is usually eaten with soy sauce and vinegar. Japanese researcher, Dr. Shigetoshi Shimada, found the anti-fungal saponin, holotoxins, from *A. japonicus* in 1969. He developed the drug for athlete's foot by sea cucumber extract. Recently, we tried to use *A. japonicus* as a functional food for oral care of elderly, and reported the *A. japonicus* jelly could reduce the oral Candida of elderly in the care-house¹. *A. japonicus* processed foods will be valuable for prevention of oral candidiasis. However, we found the large individual differences of saponin concentration of sea cucumbers. It is necessary for establishment of the system for quality control of *A. japonicus* functional foods. First of all we surveyed the saponin of *A. japonicus* at the various places. We also analyzed saponins of *A. japonicus* at the same place throughout a year. We revealed that *A. japonicus* from the northern part of Japan tended to be higher concentration of saponins, and saponin contents were markedly decreased in Apr., Aug. and Nov. It has to select the proper districts and season of *A. japonicus* for producing anti-fungal functional foods. It was also described as *Stichopus japonicus*.

Biography

Akira Yano has completed his PhD from University of Tokyo and postdoctoral studies from National Institute of Advanced Industrial Science and Technology. Then he worked at the National Institute of Infectious Diseases and National Institute of Public Health as Government Researcher. In 2006, he moved in Iwate Biotechnology Research Center, and became the Manager of the Department of Bioresource Sciences in 2015. He has studied on local bioresources, especially of functional foods for healthy life.

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Yeast assay for amyloid aggregation in proteopathies

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Amyloid proteins (including transmissible amyloids, prions) cause heritable, sporadic and infectious diseases in humans. Formation of the amyloid fibril is postulated to occur through a two-step process. First, the normal soluble protein is converted into small aggregates or nuclei of the prion isoform of that protein by a process called nucleation. Second, these nuclei seed the conversion of protein molecules containing the same or similar amino acid sequence thereby sequestering them into long fibrils. A similar molecular mechanism is employed by yeast prions, which are not homologous to known mammalian amyloid and prion proteins by sequence, and control heritable traits. We have developed a yeast-based assay that allows us to study the initial nucleation mechanism of any mammalian amyloidogenic protein. Here, we show that chimeric proteins composed of Sup35 fragments, including prion-forming domain and fused to aggregation-prone regions of mammalian prion protein (PrP), human amyloid beta (associated with Alzheimer's disease), human α -synuclein (associated with Parkinson's disease), human amylin (associated with type II diabetes), or the M-region of tumor suppressor protein 53 (associated with many forms of cancer), nucleate new Sup35 prions even in the absence of the Rnq1 prion or any other pre-existing nuclei. Our data indicate that prion/amyloid properties of mammalian amyloidogenic proteins that are detected in yeast and mammalian (or *in vitro*) systems are controlled by the same sequence elements.

Biography

Zachery Deckner has completed his BS in Biology from GCSU and is currently a PhD student at Georgia Tech. He is working on identifying and studying new amyloidogenic proteins implicated in various proteopathies. He has mentored undergraduate students and wants to take up a teaching career after his graduation.

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Physiological regulation of heritable protein aggregation

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Ordered protein aggregates (amyloids) and their transmissible forms (prions) are associated with a variety of neurodegenerative disorders, which can be studied using yeast as a model. In yeast and other fungi, prions control heritable traits. Prion formation and loss are modulated by environmental and physiological conditions, including nutrient limitation and heat stress. Our data show that propagation of yeast prions is controlled by the same cytosolic chaperones that are responsible for the protection of yeast cells against proteotoxic stress. Yeast prions are adjusted to physiological levels of chaperone proteins and hijack the cellular stress defense machinery for their own propagation. Chaperones of the ribosome associated complex, which are involved in proper folding of a nascent polypeptide, antagonize initial prion formation. During stress, the decrease in overall translational activity is accompanied by a relocation of the ribosome associated chaperones into the cytosol, resulting in the impairment of a prion-like propagation of misfolded proteins. Cellular asymmetric segregation apparatus, controlling the asymmetry of mitotic division, influences maintenance and properties of self-perpetuating protein aggregates both during recovery from stress and in the process of replicative aging. Overall, this intimate relationship with the protein quality control machinery of the cell plays a key role in the processes of prion formation and propagation in yeast.

Biography

Rebecca L Howie is pursuing a PhD at Georgia Tech in the School of Biological Sciences, with a focus on protein misfolding. Before becoming a full-time graduate student, she worked at the CDC as part of the National Antimicrobial Resistance Surveillance Team in the Enteric Diseases Laboratory Branch for over six years, studying antimicrobial resistance in foodborne pathogens. Prior to her work at CDC, she worked in anti-bioterrorism as a contractor in the Asymmetric Threat Protection Division at Tyndall AFB, FL.

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Aggregate formation by prionogenic proteins in yeast

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Cross-beta protein polymers (amyloids) cause diseases in mammals and control heritable traits in yeast. Initial amyloid formation is poorly understood. Amyloid (prion) form of the *Saccharomyces cerevisiae* protein Sup35 ($[PSI^+]$) is induced by overproduction of the Sup35 prion domain (PrD) either in the presence of the prion isoform of another protein (for example, Rnq1), or when Sup35 PrD is attached to another amyloidogenic protein, e. g. human Abeta peptide. This is accompanied by generation of various types of protein aggregates, among them filamentous structures representing intermediates of prion formation. We studied if filaments could be formed by Sup35 PrDs from other yeast species, or by chimeric constructs including both Abeta peptide and a fluorophore. Divergent Sup35 PrDs from various yeast species, or a chimeric protein composed of *S. cerevisiae* Sup35 PrD and human Abeta were tagged with fluorophores and expressed in the *S. cerevisiae* cells, either containing ($[PIN^+]$) or lacking ($[pin^-]$) the Rnq1 prion. Sup35 PrDs from various yeast species differed from each other by morphology of aggregates formed in the $[PIN^+]$ cells. Some divergent proteins produced almost no filaments, although this did not necessarily correlate with the evolutionary distance. The Sup35 PrD-Abeta-CFP construct rapidly and efficiently formed dot-like aggregates in the $[pin^-]$ cells. However, this aggregation did not result in $[PSI^+]$ induction, indicating that either prion formation or immobilization of full size sup35 into a prion is inhibited by the attachment of fluorophore to the C-terminus of Abeta. Supported by SPbSU grant 1.50.1038.2014, RFBR 15-04-06650 and RSF 14-50-00069.

Biography

Anastasia V Grizel has received her PhD in Biophysics from Lomonosov Moscow State University in 2012, and performed postdoctoral studies at St. Petersburg State University. She currently is a Research Scientist at St. Petersburg State University (Russia). Her area of research includes genetic, cytological and structural analysis of protein aggregation, primarily in the yeast model. She has published six papers in scientific journals.

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Short-length DNA marker for the determination of Malayan box turtle (*Cuora amboinensis*) materials in food chain and traditional Chinese medicines

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Malayan box turtle (*Cuora amboinensis*) (MBT) is a protected species and prohibited in Muslim foods and medicines. Despite having medicinal values, turtles are reservoirs of heavy metals and potential carriers of health-threatening microbes and allergens. To monitor turtle trafficking, there is a need of a convenient and reliable method for the quantitative tracing of turtle materials in food chain and medicines. Several polymerase chain reaction (PCR) assays have been proposed for the detection of MBT species under various routes but they are based on long-length targets which break down under the state of decomposition, making them unsuitable for the forensic and archaeological detection in food chain, medicines and other potential routes. To overcome this research gap, for the first time, we developed and validated a short length DNA marker for the qualitative and quantitative detection of MBT tissues by Conventional PCR, PCR-RFLP and SYBR green real-time PCR systems. It combined a 120 bp-site of the MBT mitochondrial cytochrome b gene and a 141bp-site of 18S rRNA gene as the universal marker for the eukaryotes. The assay specificity was checked against 20 different species and biomarker stability was tested under various food processing conditions, including boiling, autoclaving and micro oven heating under pure, admixed and commercial food matrices. The limit of detection (LOD) of the conventional PCR and PCR-RFLP assays was 0.0001 ng MBT DNA under pure state and 0.01% (w/w) MBT meat under admixed and commercial matrices. In contrast, the LOD of the SYBR green duplex PCR system was 0.00001 ng DNA and 0.001% (w/w) MBT meat under mixed matrices. PCR amplified target was further validated by sequencing and restriction digestion with *Bfa*I endonuclease and distinctive fingerprints (72, 43 and 5 bp) were obtained. The MBT target was further quantified by a duplex SYBR green real time PCR system consisting of MBT target and internal positive control, wherein the melting curve clearly reflected two distinctive peaks at $74.63 \pm 0.22^\circ\text{C}$ and $81.40 \pm 0.31^\circ\text{C}$ for the MBT and eukaryotic targets, respectively, under pure, admixed and commercial matrices. The quantification limit (ng) was 0.00001 for pure meat, 0.0030 ± 0.00001 for binary mixtures, 0.0021 ± 0.00008 for meatball, 0.0042 ± 0.0037 burger and 0.0013 ± 0.00006 frankfurter products. The analysis of 150 reference meat samples reflected 98.19 to 166.57 % target recovery, 92.23-98.15 % PCR efficiency and 0.001% LOD under various matrices. A total of 183 commercial meat products were screened but no turtle contamination was found. Finally, 153 and 120 TCM samples were surveyed by PCR-RFLP and SYBR Green PCR and 40% and 23% of them were found to be MBT-positive (0.00157 to 0.0612 ng/ μL), respectively. These authentications provided better security, firstly, through short-length biomarker target which offer extraordinary stability and sensitivity and secondly, through molecular fingerprints which authenticated the post-amplified target by restriction digestion. Thus, the novel assay demonstrated sufficient merit for use in any forensic and/or archaeological authentication of MBT, even under a state of decomposition.

Biography

Asing is a graduate student at University of Malaya, Malaysia and he is working under the supervision of Dr Md Eaquab Ali.

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Draft genome sequence of 11 clinical and environmental Colombian isolates of genus *Aspergillus*

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Background: The genus *Aspergillus* is a group of opportunistic fungi that cause infections, with high morbidity in immunosuppressed patients. Approximately 350 species have been described in this genus, classified in 4 subgenus and 20 sections. In Colombia, *Aspergillus fumigatus* is the most frequent species in these infections. However, in the last years an increase in the incidence of other species has been observed. This added to an increase in the quantity and diversity of *Aspergillus* isolates in hospital environments.

Aims: Characterize the phenotype and genotype of clinical and environmental Colombian isolates of *Aspergillus* spp.

Methods: We collected 11 Colombian isolates of *Aspergillus*: 4 from diferents clinical samples, six from hospital environments and 1 from extra-hospital environments. The isolates were identified according to their macro and microscopic characteristics. Genomic DNA for sequencing was prepared from mycelium culture using phenol/chloroform extraction. Library preparation and 150-bp paired-end sequencing was performed using the Illumina HiSeq 2500 platform. The reads was de novo assembled using SPAdes 3.10 pipeline. The draft genome assembly quality was analyzed by QUASt. Augustus v3.0.1 and GlimmerHMM were used for gene prediction. Inference by sequence homology was performed with OrthoFinder v2.0.9. IQtree v1.4.4 software was used for phylogenetic reconstruction by ML.

Results: In this study, four of the eleven isolates classified phenotypically were re-classified correctly after genome analysis. We obtained whole genome sequence of 4 clinical and 7 environmental isolates, including 3 novel genome sequences of the genus *Aspergillus* not reported in the databases: *A. tritici* (Clinical), *A. tamari* (hospital environment) and *A. amoenus* (extra-hospitalenvironment).

Conclusion: The results of this study provide useful data for the genomic comparison between clinical and environmental isolates of *Aspergillus*. Valuable information that can be used to understand the evolutionary relationships, determine the diversity and level of production of enzymes and secondary metabolites, predict the pathogenicity and response of this opportunistic fungus to antifungal agents.

Biography

Gomez OM is currently working as Faculty at Universidad de Antioquia, Colombia.

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The rise and the fall of irradiation treatment for ringworm during the first half of the 20th century: A comprehensive historical perspective

Shifra Shvarts

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Throughout the course of human history, ringworm has always been considered as a difficulty to treat disease that carried a stigma, said to be a disease of the poor and the uneducated. The spread of the industrial revolution which prompted countless people to migrate from the farms to the cities; crowded living conditions, poor hygiene and a high incidence of contagious diseases in the cities led to the spread of ringworm on a massive scale, particularly among children. Ringworm had often led to social ostracism that was very detrimental to the life of the child and his family. Traditional treatment entailed manually plucking out hair from the roots to, a painful process that caused mental trauma to children. The invention of the X-ray machine and the discovery that exposure to low doses of X-rays triggers hair loss transformed irradiation into the preferred treatment strategy for ringworm and it was quickly adopted by the medical community as a humane and most advanced treatment protocol for children with ringworm. Irradiation for ringworm from the outset of the 20th century until 1960 was the standard treatment for the disease in all western countries. This work examines the rise of irradiation treatment for ringworm and profiles the mass ringworm irradiation campaigns (over 200,000 children) conducted in Eastern Europe, Serbia, Morocco, Israel and the United States in the first half of the 20th century. The work focuses on historical, social and health aspects of various groups treated with irradiation for ringworm, how they coped with the social stigmas associated with the disease and its treatment, as well as the impact of treatment over time on the lives of the irradiated children.

Biography

Shifra Shvarts is a Professor for the history of Medicine and Health Sciences, Ben Gurion University and The Gertner Institute of Epidemiology and Health Policy Research, Sheba Medical Center, Israel. Since 2005, she has concentrated her research on the ways various countries in the world, including Israel, has grappled with latent negative health risks and other ramifications of irradiation in childhood as a treatment regime for ringworm, and subsequent social aspects of this treatment that has arisen, reflected in legislation of a compensation for ringworm victims law by Israel and debate elsewhere over how best to inform former patients and monitor them.

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Review of species distribution and susceptibility of invasive isolates of *Candida spp* as evaluated using the previous and recently revised clinical breakpoints and method dependent epidemiological cut of values

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Invasive *Candida* infections often cause high morbidity and mortality especially in the critically ill or immunosuppressive patients. Although *Candida albicans* was the most frequently isolated species as the causative agent of *Candida* infections, geographical differences and changes over time in the species distribution and the susceptibility to antifungals were reported in several surveillance programs. Some variations have been shown to occur among institutions, localities, or countries. It is significant to determine the species distribution and antifungal resistance in large medical centers. We reviewed the species distribution and antifungal susceptibility data of 1371 invasive *Candida* strains isolated in a large university hospital mycology laboratory over 16 years. Susceptibility tests against amphotericin B and azoles were routinely performed using Clinical and Laboratory Standards Institute guidelines from 1998 to 2012 and using Etest from 2012 to 2014. The Sensititre YeastOne colorimetric method was used to test *Candida* echinocandin susceptibility between 2012 and 2014. All test results were routinely reported to clinicians. In this retrospective analysis, resistance or non-wild type phenotypes to systemic antifungals were determined by the previous and recently revised CLSI breakpoints (BPs) and by method dependent species-specific epidemiological cutoff values respectively. The new epidemiological BPs provided by CLSI changed the percentage of resistant *C. albicans*, *C. parapsilosis* and particularly *C. tropicalis* isolates to fluconazole.

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Diversity of *Aspergillus* species associated with groundnut (*Arachis hypogaea* L.) in eastern Ethiopia as revealed by InDels and their potential for aflatoxin production

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Aspergillus species cause aflatoxin contamination, which becomes a health threat in agricultural products and leads to commodity rejection by domestic and international markets. Hence, it is necessary to discriminate diversity and aflatoxin producing species in the eastern Ethiopia. Therefore, the current study was undertaken to elucidate the genetic diversity of *Aspergillus* isolates through InDel (Insertion and Deletions of sequences) markers and evaluate *in vitro* aflatoxin production abilities of the isolates using YES (Yeast Extract Sucrose) medium. A total of 276 isolates were used for genetic diversity fingerprinting of DNA using 23 InDel markers based on aflatoxin biosynthesis gene cluster. Cluster analysis was analyzed by NJ (neighbor joining) and by PCoA (Principal Coordinate Analysis). The *Aspergillus* isolates studied in the current work grouped into three clusters. In addition, 269 isolates were tested for aflatoxin production using UPLC (Ultra Performance Liquid Chromatography). Aflatoxigenic isolates had a maximum of 247 $\mu\text{g mL}^{-1}$ aflatoxin B1 and 139 $\mu\text{g mL}^{-1}$ aflatoxin G₁. This study provides insight into the genetic biodiversity of aflatoxin biosynthesis gene cluster of *Aspergillus* in relation to *in vitro* aflatoxin production of isolates in the country. The prevalence of aflatoxigenic isolates was much higher (93% of the tested isolates) than the non-aflatoxigenic.

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Invasive mucormycosis in chronic granulomatous disease

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Mucormycosis is an uncommon fungal infection caused by members of the order *Mucorales*. Populations at risk for this potentially life-threatening infection include hematopoietic stem-cell transplant (HSCT) recipients, patients with hematological malignancies, diabetes mellitus, ketoacidosis, burns, trauma, premature neonates, and patients receiving iron chelation. *Rhizopus* is the most commonly identified species, followed by *Mucor* spp. Common patterns of mucormycosis are cutaneous, gastrointestinal, rhinocerebral, and pulmonary. Amphotericin B is the antifungal drug of choice for treatment of mucormycosis. Combination polyene-caspofungin treatment was found to be associated with improved survival in patients with rhino-orbital-cerebral mucormycosis, compared to polyene monotherapy. Surgery is an important adjunctive therapy and was shown to decrease mortality in patients with mucormycosis. We described rare presentations of pulmonary mucormycosis caused by *Rhizopus* spp. in 2 patients with CGD; with chest wall and spinal involvement in a child, and cardiovascular involvement in an adult patient. Case 1: A 2-year-old girl presented with pneumonia and pleural effusion that failed to respond to prolonged courses of broad spectrum antibiotics and pleural drainage. Examination revealed a febrile, malnourished child with enlarged liver and spleen. Chest examination showed a firm mass extending from the axial to the posterior aspect of the right chest wall. CT scan showed consolidation involving right lower lobe, middle lobe, and posterior segment of the right upper lobe with pleural effusion. A right chest wall mass with intraspinal extension was also noted. Cultures of tissue obtained from surgical biopsy of the chest wall mass grew *Rhizopus* spp. She was subsequently diagnosed to have CGD based on oxidative burst test result. Treatment with liposomal amphotericin B was initiated at a dose of 5 mg/kg/day then increased to 7 mg/kg/day. Caspofungin and interferon γ (IFN- γ) were added to treatment. She underwent surgical debulking of the chest wall mass and near-total pneumonectomy. She was then referred to a specialized center for HSCT.

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Detection and diagnosis of wood decay fungi in wooden heritage using different image techniques

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This study was based on the current deteriorated status of wood slats from locomotive turntable of Provincial Railway Station La Plata. Wood specie of the slats was determined by conventional methods being *Schinopsis sp.* while fungal species was determined morphological being *Phellinus chaquensis* (white-rot fungus). Determination of the fungus and its *in-vitro* cultural features were based on Iaconis and Wright and Robledo and Urcelay. Fungal degradation wants be measured by non-destructive methods: area occupied by mycelium and basidiomata were observed by x-ray radiography and computer tomography (CT) and quantified by image analysis with Image J software. Greyscales of the images obtain indicated density changes, being black scale the less dense and white scale the densest. To establish the microstructural wood deterioration (cell wall), scanning electron and optical microscopy (SEM and OM) images were analyzed. It was concluded that deterioration analysis by images is a non-destructive alternative methodology, which allows to measure structural condition of material. This is essential in heritage conservation because it allows defining correctly the deteriorated status useful to planning a conservation strategy, avoiding the asset loss.

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Do climate changes affect the fruiting timing of wild mushroom populations in Israel?

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Despite the semi-arid climate of Israel the mushrooms mycobiota is quite varied and has been studied over the years. We found both early and late fruiting in many wild mushroom species in the 2015 mushroom season, that displayed unusual temperatures and rain. Distribution in some cases, several species fruited twice on that year. Similar findings were also reported by researchers from Norway, England, France, Germany and other countries. Is this phenomenon related to global warming changes? Our findings cannot answer this question. We conclude that due to the abnormal weather we could detect a fluctuation in the timing of mushroom appearance, which could be the result of global changes. In order to investigate the causes of these observations, we suggest examining and combining previous herbarium mushroom fruiting records and meteorological information.

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Ergotism and the ergot fungus in Ethiopia

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Ergotism is a current human health problem on the Ethiopian highlands. A few outbreaks of ergotism in the country have been documented in the last few decades. The problem continues to date and a multi-purpose research was undertaken. Field studies showed that the problem is prevalent on highlands between 2,300- 3,000 m above sea level and where barley and wild oats (*Avena abyssinica*) are grown as major crops. It was found that *A. abyssinica* is the only cereal host for the Ethiopian ergot fungus. Based on molecular mycological studies, the fungus is characterized as a pathovar of *Claviceps purpurea*. Both gangrenous and convulsive ergotism are apparent in affected village communities with symptoms typical of ergotism. It was noted that extensive invasion by the fungus and development of the sclerotia on wild oats in farmers' fields and the consequent outbreak of ergotism in Ethiopia is unpredictable in time and space. Farmers, unfortunately, are not aware of the source and cause of the problem. Chemical analysis of ergot sclerotium showed that a cocktail of 16 toxic ergot alkaloids including ergocornine, ergocryptine, ergometrine, ergosine and lysergic acid derivatives are contained in the sclerotium. From studies based on affected communities, it was apparent that ingestion of the sclerotium from ergot infested oats is the cause of the problem. Prevention and control of ergot toxicosis requires a deeper understanding of environmental variables and a systematic ecological study.

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Genetic diversity of *Aspergillus flavus* and occurrence of aflatoxin contamination in stored maize across three agro-ecological zones in Kenya

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Aflatoxin contamination at post-harvest poses a serious challenge in achieving millennium development goals on food security especially in the developing world. In Kenya, major outbreaks of aflatoxicoses have been attributed to poor post-harvest storage practices. In this study, we conducted a cross-sectional survey within three Agro-ecological zones in Kenya, to determine occurrence and distribution of total aflatoxin in stored maize and the aflatoxigenicity potential of *Aspergillus flavus* in stored maize. The counties selected were; Kitui, Nakuru and Kitale (in Trans-Nzoia County). Sampling sites were selected based on previous aflatoxicoses outbreaks (Kitui) and major maize production areas (Nakuru and Kitale) where little information exists on the occurrence of aflatoxin contamination. A total of one hundred and thirty (130) kernel maize samples were random collected during the period between June and August 2012. Moisture content was determined using the standard oven method and *Aspergillus flavus* was isolated by direct plating technique. Genetic diversity of the isolates was determined by PCR and Single Sequence Repeats (SSR) micro satellites analysis. Positive strains were induced to produce B1 aflatoxins on Yeast Extract Sucrose Agar (YESA) and quantified using competitive ELISA technique. The results indicated mean moisture content of maize ranged between 6% and 34%, although this was found not to be significantly different ($p=0.23>0.05$). However, total aflatoxin contamination of postharvest stored maize samples between sites was significantly different ($p=0.000, <0.05$); with the highest contamination in Kitale at a mean of (9.68 $\mu\text{g}/\text{kg}$). *A. flavus* was isolated in 70% (N= 91) of the maize samples collected at postharvest. *A. flavus* isolates with the highest aflatoxigenicity potential were from Nakuru County with mean aflatoxin level at 239.7 $\mu\text{g}/\text{kg}$. Genetic distance based on Neighbor Joining (NJ) clustered the *A. flavus* isolates into five main clusters. Principal coordinate Analysis (PCA) analysis showed five distinct clusters with both axes explaining 60.17% of the variance. This study showed widespread distribution of aflatoxin contamination and a highly toxigenic *A. flavus* in stored maize in three major agro ecological zones in Kenya. These results suggest a potential health risk of aflatoxin outbreaks within these areas, thus call for more investigations.

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Biodiversity investigation and potential of fungal endophytes of peppermint and their extract effect on chickpea rot pathogens

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India is the highest producer of *Cicer arietinum* (Chickpea), however the crop is highly susceptible to plant fungal diseases i.e. *Sclerotinia sclerotiorum*, *Botrytis cinerea*, *Fusarium oxysporum* and *Rhizoctonia solani*. For a sustainable and environment friendly alternative, anti-plant pathogenic efficacy of fungal endophytes were investigated. Endophytic fungal agglomerate of Indian medicinal plant, *Mentha piperita* was investigated for biodiversity, bio control potential towards chickpea rot causing phytopathogens and their metabolite profiling. 63 pure fungal isolates were recovered from medicinal plant sampled in different seasons from distinct regions of India. Endophytic fungi were identified by ITS-rDNA sequence process. PCA divulged seasonal variability with exclusive presence of *Colletotrichum sp.*, *Diaporthe phaseolorum*, *Alternaria sp.*, *Hypocrea sp.* and *Rhizopus oryzae* in second sampling season. Shannon diversity index (H') was found to be highest in leaf (1.253) from Mukteshwar. Menhinick's index discern that stem tissues from Mukteshwar have maximum species richness ($Dmn=1.75$). Best antifungal activity was exhibited by extracts of *Acremonium sp.* (MPM-2.1) with $< 1\text{mg/ml}$ IC₅₀ value towards phytopathogens. GC-MS chromatography of potent biocontrol fungus *Acremonium sp.* (MPHSS-2.1) confirmed presence of antifungal compounds 1-heptacosanol and 1-nonadecane.

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Biofilm induction in mucormycosis-causing fungi and the synergistic antifungal activity of Amphotericin B and thyme oil

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Fungal infections caused by opportunistic pathogens have gained clinical importance in the last decade, with a significant increase in infections due to the *Zygomycetes*, *Mucor*, *Rhizopus* and *Absidia*. These serious and sometimes fatal infections are often associated with biofilm formation. The formation of biofilm often increases resistance to antifungal agents when compared to free living colonies. This study investigates both the biofilm formation and the antifungal susceptibility of two species known to cause mucormycosis infections namely: *Rhizopus oryzae* and *Absidia corymbifera*. Upon successful biofilm formation, the synergistic effects between thyme oil and amphotericin B were tested. Results indicate that both *R. oryzae* and *A. corymbifera* are able to form biofilms under specific conditions and that these biofilms were significantly inhibited by Thyme oil. The MIC₅₀ of thyme oil on *Absidia corymbifera* and *Rhizopus oryzae* was 0.0005 µL/mL and 0.0001 µL/mL respectively. Results also indicate a strong synergistic relationship between Amphotericin B and Thyme oil when used in combination against fungal biofilms.

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Investigating the effect of arbuscular mycorrhizal fungi (*Glomus etonicatum*) and air pollutants on growth parameters of maize (*Zea mays* L.)

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To investigate the effect of *Glomus etonicatum* and air pollutants and acidic rain on growth parameters of maize plants (*Zea mays* L.), a completely randomized experiment with ten replications was conducted from February 2016 to April 2017. Experimental treatments included four categories containing the plants treated by mycorrhiza fungus irrigation by acidic rains and control water with (PH=7), and witnessing plants irrigation by acidic rain and control water. Results from analysis of variance revealed that the effect of mycorrhizal inoculation on chlorophyll, protein and carotenoid content of leaves, plant height, leaf dry wt, leaf fresh wt, root fresh wt, fruit number and leaf number and surface, were significant ($p < 0.05$). Mycorrhizal inoculation enhanced all parameters significantly in comparison to the witnessing plants and the highest value for these traits obtained by the plants inoculated with mycorrhiza irrigation by control water and the least obtained in witnessing plants irrigation by acidic rain water. Furthermore, the research revealed that the amount of these parameters in plants inoculated with mycorrhiza irrigation by acidic rain, is significantly more than witnessing plants irrigation by control water. In general, inoculation by mycorrhizal fungi in addition to enhancing growth parameters can enhance the photosynthesis and production of oxygen in maize even under acidic rain circumstances and air polluted environments, compared to non-mycorrhizal plants in regular circumstances.

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Pulmonary zygomycosis among HIV/AIDS subjects with respiratory symptoms in Calabar, Nigeria

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Introduction: Zygomycosis is a rare infection but the incidence is on the rise as a result of increased use of chemotherapy and steroids among immunosuppressed patients. It is an invasive angiotropic infection with fungi of the *Mucorales* order, which includes *Mucor* species, *Rhizopus* species, *Rhizomucor* species, and multiple others. The second most common form of the infection is pulmonary.

Materials & Methods: HIV-positive subjects with respiratory symptoms were enrolled for the study. Subjects selection was based on HIV screening and the ability to produce sputum. A structured questionnaire was administered to all the subjects after obtaining their informed consent for demographic data. Ethical approval was obtained from the ethical research committee, UCTH, Calabar, Nigeria. Blood samples were obtained for CD4 count determination to ascertain the immune status of the patients. Sputum samples produced early in the morning were obtained twice from the subjects and subjected to macroscopy, microscopy and culture. The immune status of the subjects was assessed by CD4 count levels. Identification to the species complex level was performed by macroscopic and microscopic morphology.

Results: *Rhizopus arrhizus* 50.0% and *Lichtheimia* species 50% were the only *Mucorales* encountered among subjects in this study. 3.0% pulmonary zygomycosis prevalence was recorded in the study. Subjects with *Rhizopus arrhizus* infection presented hemoptysis and cough while those with *Absidia* infection presented with variable symptoms including; cough, chest pain, sinusitis and fever. The mean CD4 counts of subjects with and without zygomycosis were 123.0 ± 136.2 351.3 ± 254.3 respectively. There was a statistically significant effect of zygomycosis on the CD4 counts of subjects ($t=2.18$, $p=0.02$).

Conclusion: This study reveals that pulmonary zygomycosis is a health problem among HIV/AIDS patients in our locality. The immune status may have been influenced by the infection.

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Glomus fasciculatum fungi as a bio-converter and bio-activator of inorganic and organic P in dual symbiosis

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This article describes mechanism of Phosphorus supply from the soil into plants under VAM fungal inoculation. It was already established that VAM fungi symbiosis helps in uptake of nutrients especially P which is not easily accessible under ordinary condition of soil. For this investigation, *Conocarpus erectus L* species and *Glomus fasciculatum* was identified and selected in four experimental set up including i) control (E₁), ii) VAM inoculated (E₂), iii) VAM and excess of CO₂ (E₃) and iv) VAM and drought (E₄). All Plants except E₃ were cultivated under natural condition and watered alternative day whereas E₄ was watered after regular interval of 4 days. Analysis of soil and plant's P were carried out after 12 months. Phosphorus in plants were analysed in roots, stem, and leaves separately and correlated with soil remaining phosphorus. Results showed that soil P was less in four experimental set up when compared with P of standard soil. It was found that P accumulation in plants was varied in all four experimental conditions based on VAM symbiosis. It was found that the roots of E₂ and E₃ plants showed highest P accumulation as compared to E₁ and E₄ which were linked with absence of VAM and drought conditions. P translocation into E₄ plants showed that movement of P based on availability of water condition due to which it was less in leaves too over E₁, E₂ and E₃ plants. Experimental facts and nonstop growth of plants recommended that VAM fungi act as a bio-converter and bio-activator of soil nutrients especially of Phosphorus, and their hypal interaction absorb soil nutrients and convert inorganic P to organic one for plant development. Continuous growth of one year old *conocarpus* plant support the proposed idea that phosphorus cycle exists during VAM inoculations which strengthen the plant and activate photobiological activity that helps in increasing photosynthetic rate and stimulate all biological processes of plant including H⁺ co-transporter couple with inorganic phosphorus and its ultimate supply to plants.

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Primary cutaneous mucormycosis in a patient with burn wounds due to *Lichtheimia ramosa*

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Mucormycosis is usually an invasive mycotic disease caused by fungi in the class mucormycetes. Here, we report a case of cutaneous mucormycosis due to *Lichtheimia ramosa* in a 20-year-old female patient with burn injuries. She was admitted to the hospital with accidental flame burns covering 60% total burn surface area. After 15 days of admission to hospital, the burn wound showed features of fungal infection. Culture showed white cottony growth belonging to the *Mucorales* order. Morphological identification confirmed it as *L. ramosa*. She was managed surgically and medically with the help of amphotericin B. Patient survived due to prompt diagnosis and appropriate medical and surgical treatment. Early diagnosis is critical in prevention of morbidity and mortality associated with the disease. Fungal infection in burn wounds can be difficult to diagnose and manage.

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