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Mycotoxin and molecular characterization of *Alternaria* species contaminated wheat grains in Riyadh City, KSA

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The mycological profile of the wheat grains selling in different markets in Riyadh, Saudi Arabia was studied with a focus on the different species of *Alternaria*. Fifty fungal species and one species variety belonged to 18 genus namely: *Alternaria, Acremonium, Aspergillus, Cladosporium, Drechslera, Mycovellosiella, Mycelia sterilia, Fusarium, Embellisia, Penicillium, Phoma, Scytalidium, Stachybotrys, Stemphylium, Staphylotricum, Torula, Ulocladium* and *Xylohypha*. Twenty-nine strains of *Alternaria alternata* were screened for their ability to produce altenuene, alternariol and alternariol monomethyl ether. Toxicity bioassay for the extracts of fungal isolates was conducted using *Bacillus subtilus*. Fungal extracts differed in their ability to inhibit the growth of bacteria before and after boiling. Random Amplified Polymorphic DNA (RAPD) technique using 5 different random primers (OPA03, OPA04, OPA10, V6 and M13) was used. All tested primers gave positive results with all tested isolates. The constructed dendrogram based on the results of 5 primers collectively showed that there is no correlation between DNA banding patterns and geographical distribution for the isolates.

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Serotype-specificity and immunogenicity of domain-swapped Virus Like Particles (VLPs) from dengue virus serotypes 1 and 2

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The dengue virus (DENV) remains the most important and rapidly emerging mosquito-borne pathogen worldwide. While humans develop long-lived, strongly neutralizing antibodies (NtAbs) after initial or primary dengue infection, there is conflicting information on the specific targets of NtAbs on the envelope (E) glycoprotein, which confounds the development of more effective vaccines. To determine the specific target/s of NtAbs against DENV 1 and 2, we used dengue virus-like particles (VLP) consisting of E domains I/II and III from DENV type 1 (D1) and type 2 (D2) to produce two domain-swapped VLPs (D2-D1 I/II and D2-D1 III) and two parental VLPs (D1 and D2). DNA plasmid vaccine expressing the E proteins of parental and domain-swapped VLPs were intramuscularly administered to 25 female BALB/c mice, divided into five groups at three 4-week intervals. The dengue VLPs and DNA vaccines were characterized through immunofluorescence assay (IFA), antigen-capture (Ag-capture) ELISA and SDS-PAGE. IFA detected that the parental and domain-swapped VLPs were intracellularly produced in COS-1 cells. SDS-PAGE confirmed the expected VLP structure and showed that the VLPs have comparable maturation states. Ag-capture ELISA showed that mice vaccinated with parental and domain-swapped (D2-D1 I/II) VLPs produced cross-reactive antibodies targeting different E domains (interdomain) and broad and high neutralization titers against DENV1 Hawaii. On the other hand, mice immunized with D2 and domain swapped VLPs generated type-specific and high neutralizing titers against DENV2 16681. This implies that the protective ability of immune mice sera against DENVs is serotype-dependent. Interestingly, there is loss of neutralization against DENV1 Hawaii in D2-D1 III immunized sera, suggesting that domain I/II determines the serotypespecificity of NtAbs. ELISA and focus reduction microneutralization test (FRμNT) showed that the domain-swapped VLP antigens can be used to determine the dengue serotype in primary infection. Knowledge on the character and immunogenicity of the VLPs will lead to the development of next-generation, antigen-specific dengue vaccines.

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