CHARACTERIZATION OF OCHRATOXIGENIC FUNGI IN DRY COFFEE CHERRY, EFFECT OF POST-HARVEST HANDLING PRACTICES AND FACTORS AFFECTING ITS PREVALENCE IN THARAKA NITHI COUNTY, KENYA

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DECLARATION AND RECOMMENDATION

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Declaration

This thesis is my original work and has not been presented for an award of a diploma or degree in any other University.

Signature..... Date 22/10/2024

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Recommendation

This thesis has been examined, passed and submitted with our approval as the University supervisors.

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DEDICATION

I dedicate this thesis to the Almighty God, who has been my source of strength, grace and wisdom throughout my period of study. Through his grace and favor, I have been able to complete my course and scale through the hurdles of my academic pursuit. I also dedicate it to my lovely husband, Patrick Gitonga, and all my children and parents for their prayers, love, encouragement and endless support throughout this journey.

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ABSTRACT

Coffee is an economically significant cash crop valued for its unique flavour, aroma, and potential health benefits. In Kenya, most research efforts have focused on wet-processed coffee beans, with little attention directed towards the dry-processed cherry, known as *dry coffee cherry*, which is roasted and consumed locally. There remains a glaring gap in our understanding of ochratoxin A (OTA) occurrence in dry coffee cherry, particularly in major coffee-producing areas like Tharaka Nithi County. The pathogenic fungal species responsible for OTA production in coffee have not been adequately characterized. The

Objectiveof this study was to investigate the prevalence of Ochratoxin A in dry coffee cherry in Tharaka Nithi County and its relationship with socio-economic factors and post-harvest handling practices for improved control and management. A cross-sectional design was used and the target population of coffee farmers drawn from three sub-counties. A cluster random sampling was applied to collect dry coffee cherry samples and information from farmers through a semi-structured questionnaire. A total of 143 farmers were sampled from 44 villages. Pathogens were isolated on Potato Dextrose Agar (PDA) media and OTA production assessed according to the International Commission on Microbiological Safety for Foods. Morphological characterization of fungal isolates was done using cultural characteristics while molecular identification was done using Sanger sequencing. Data was analysed using the SPSS version 28 and SAS version 9.4. Cluster analysis was carried out using Eucleadian distances. The fungal isolates obtained were aligned and phylogenetic tree was constructed using neighbor-joining clustering as implemented in MEGA X version 11. The results of the study revealed a high prevalence of Ochratoxigenic fungi in coffee samples across the different AEZs, with *Aspergillus niger* being the predominant fungal species. Ochratoxin A was detected by Enzyme-Linked Immunosorbent Assay (ELISA). The analysis of colony-forming units (CFU) across agroecological zones showed significant differences, with the highest CFU recorded in UM1 (941.14), followed by UM2 (852.06), and the lowest in UM3 (716.56). Genetic variability was measured using Principal Component Analysis (PCA) and Cluster Analysis**.** The Eigenvalu**es** and proportion explained variance from the PCA provide insights into the genetic variability, with the first three components accounting for 77.82% of the total variation. Coefficient of Variation (CV**)** was used, with a CV of 27.59% indicating moderate variability in colony-forming units across samples**.** R-squared values were also used to indicate the extent of variability explained by agroecological zones and other factors. The levels of OTA contamination were influenced by several factors which includes; inadequate drying practices and improper storage conditions were identified as major contributors to the growth of Ochratoxin A fungi, Agro-ecological zones with higher humidity and warmer temperatures, socioeconomic factors such as farmers' level of education, awareness about mycotoxins, and access to proper post-harvest facilities played a crucial role in determining the levels of contamination. Coffee beans stored for longer periods in humid, poorly ventilated conditions showed higher contamination levels. The results highlight the urgent need for interventions aiming at improving post-harvest handling practices to reduce OTA contamination. The study emphasizes the importance of educating farmers about the risks of mycotoxins. The findings also provide a basis for developing policies and interventions that can mitigate the risk of mycotoxin contamination and improve the overall quality and safety of Kenyan coffee product.