

# Ubiquitous contamination with multiple mycotoxins and other fungal metabolites in dietary rations of dairy cattle in Punjab, Pakistan

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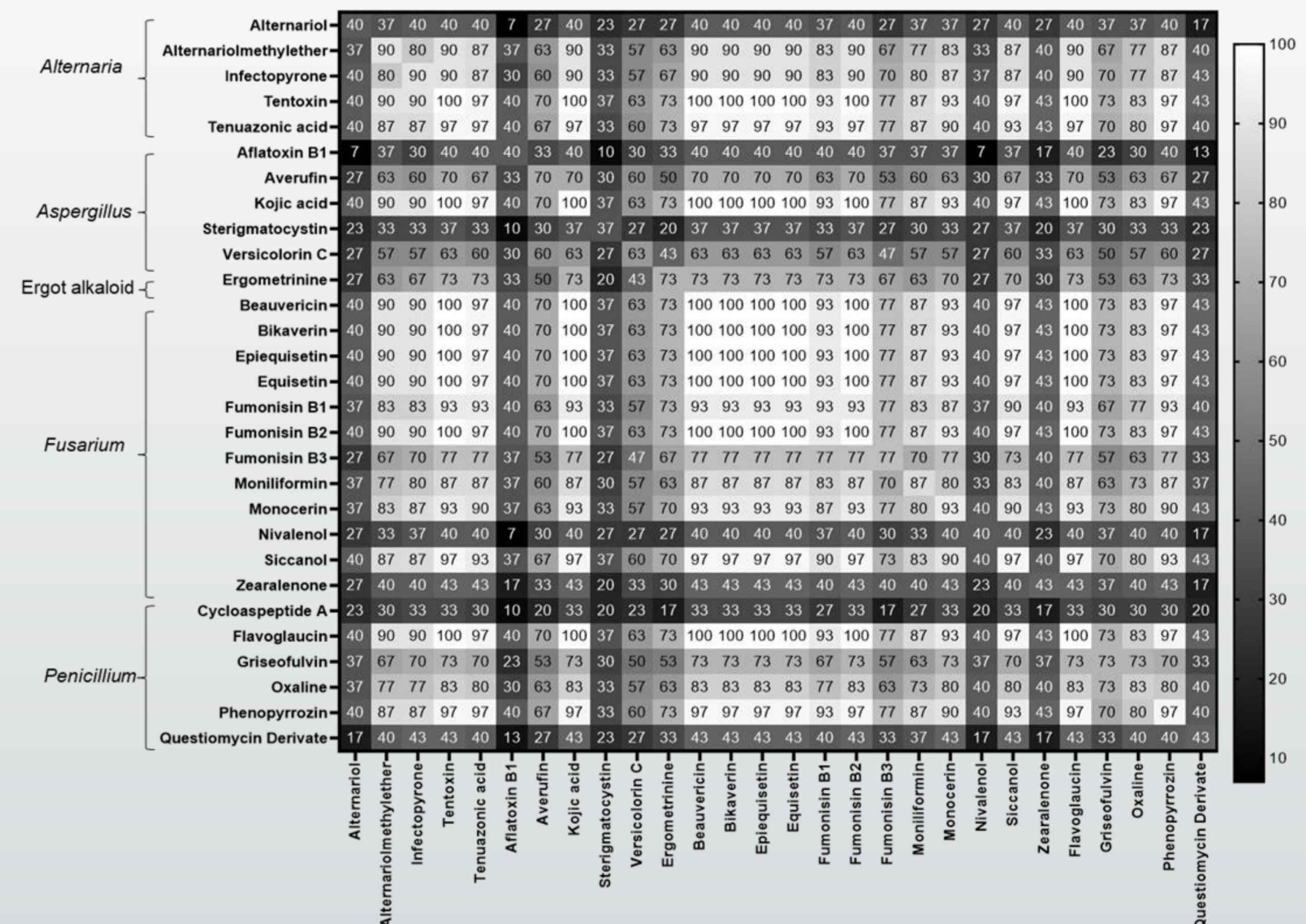
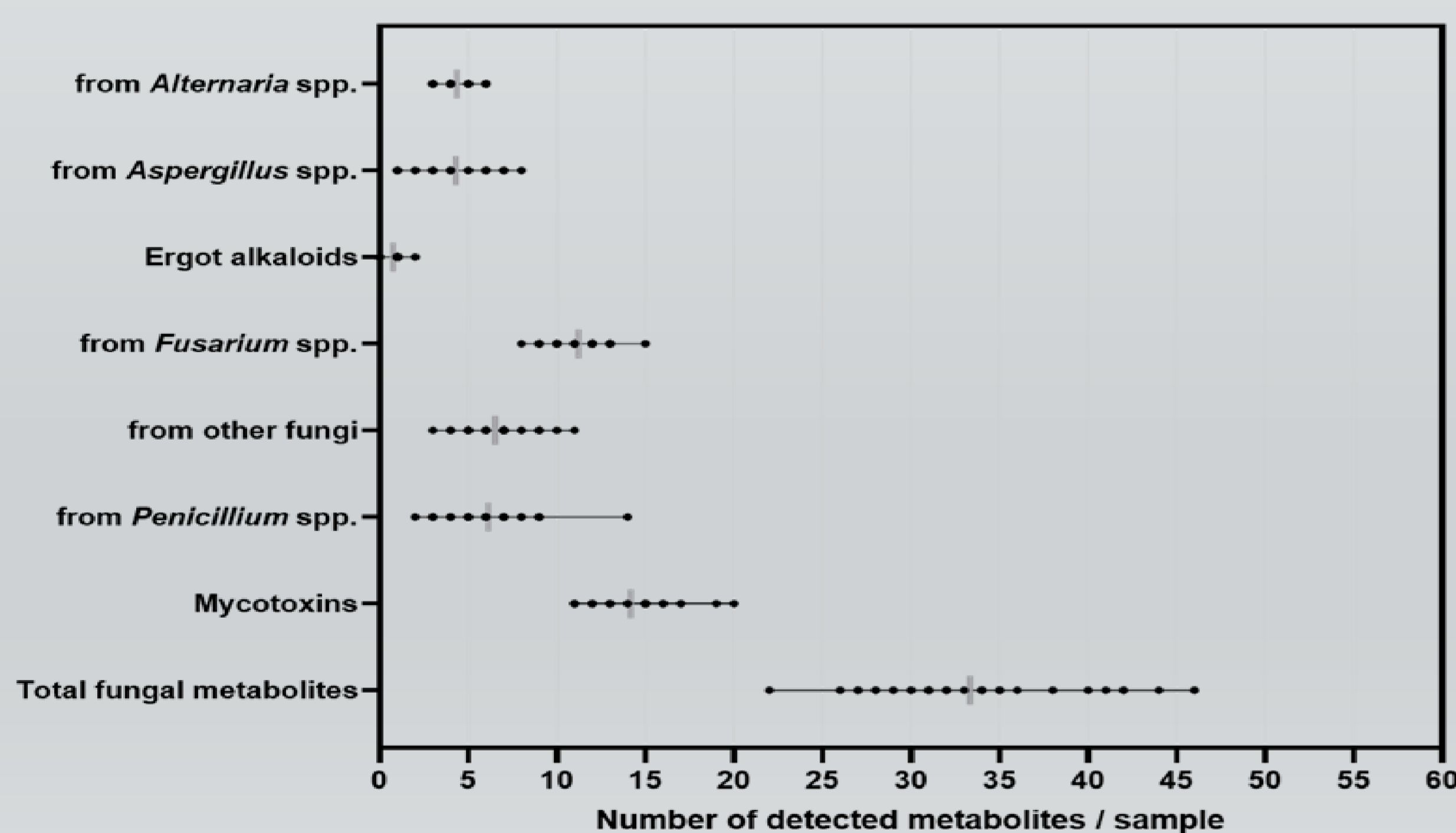
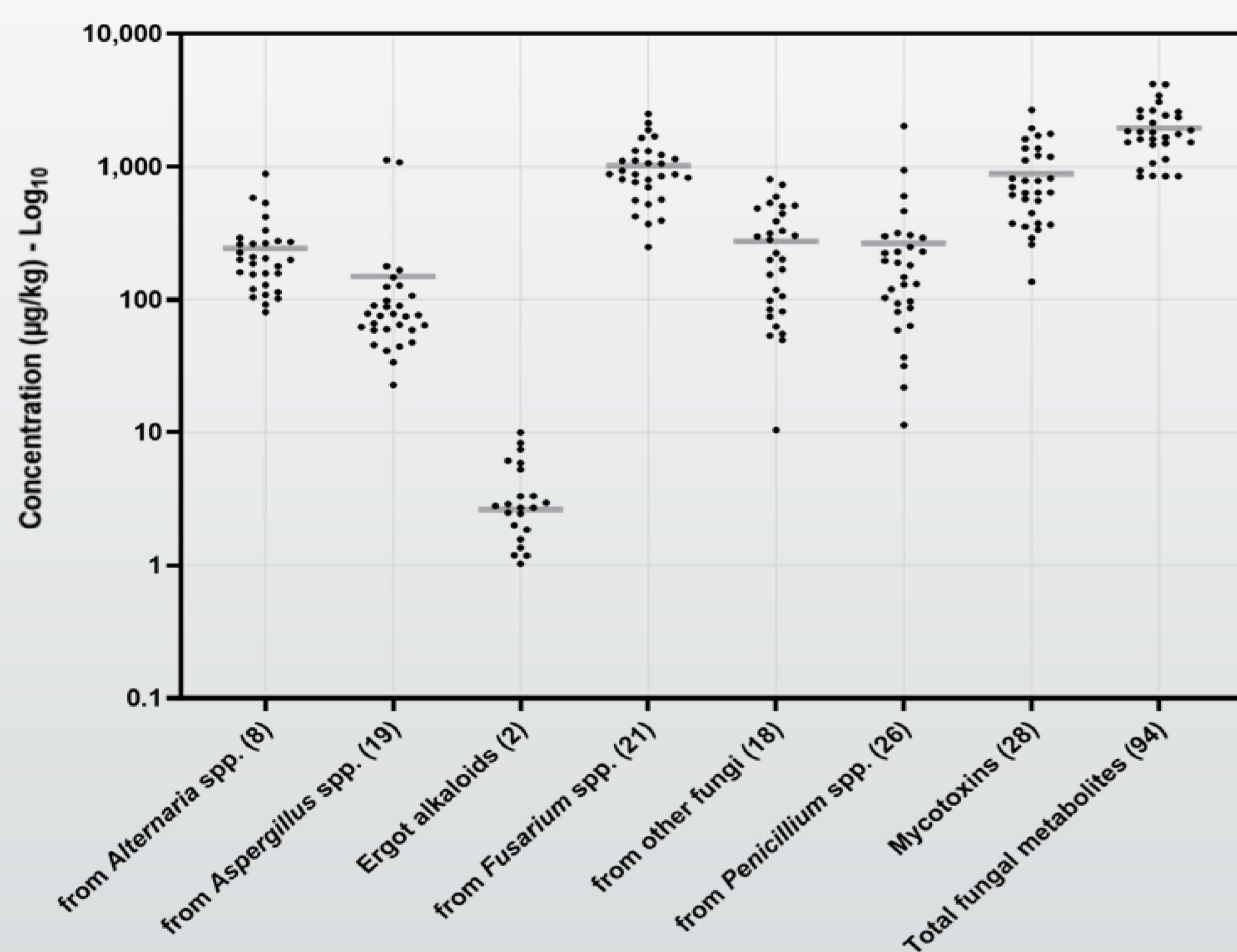
## Introduction

Pakistan is third in worldwide dairy production ranking, behind India and USA. The dairy cows are usually fed total mixed rations (TMR) containing various ingredients, including roughages, cereal grains, and agro-industrial by-products (1). Due to favorable climatic conditions in South Asia, feedstuffs are vulnerable to mould infection and colonization with successive mycotoxin contamination leading to animals and public health problems. This is indicated by far higher levels of aflatoxin (AF) M1 observed than those of recommended level by European Union (2). As feed is the central source of AFM1-precursor (AFB1), contamination of in dairy cattle diets should also be monitored to kept the risk at minimum. This exploratory study aimed to determine via a LC-MS/MS method the presence, co-occurrence, and concentrations of mycotoxins and other secondary metabolites in 30 dairy farms of Punjab-Pakistan.

## Methods

The samples were collected during June - July 2020 from 30 dairy farms (each with minimum 200 animals herd size) located in Punjab-Pakistan (n = 30) in such a way that each administrative division contributed equal farms. A total of 30 incremental samples from feed bunk right after serving were pooled and 1 - 1.5 kg sample was vacuum-packed, and stored at -20 °C. The frozen TMR were thawed at room temperature for 24 hours and air-dried at 65 °C for 48 hours. Then, the dried TMR were milled to a final particle size of ≤ 0.5 mm, using a cutting mill following the previously reported procedures (3). Finally, the samples were analyzed via LC-MS/MS (Spectrum 380®) for mycotoxin analysis. The concentration values are presented on a dry basis in parts per billion (ppb).

## Results



Mycotoxin / Metabolite	Proportion of ingredient (% DM)						Commercial concentrate
	Rhodes grass hay	Wheat straw	Corn grain	Soybean meal	Canola meal	Molasses	
Aflatoxin B1	0.26	0.03	0.06	0.00	0.07	0.41*	-0.26
Kojic acid	-0.12	-0.07	0.10	0.06	0.07	0.38*	-0.23
Ergometrinine	-0.23	0.26	0.39*	0.31	0.38*	0.54**	-0.36
Ergot alkaloids	-0.23	0.27	0.40*	0.31	0.38*	0.53**	-0.35
Bikaverin	0.08	0.13	0.47*	0.47*	0.49**	0.31	-0.58**
Fumonisin B1	-0.16	0.22	0.54**	0.54**	0.52**	0.36	-0.56**
Fumonisin B2	-0.09	0.21	0.55**	0.56**	0.57**	0.32	-0.47**
Fumonisin B3	0.02	0.18	0.47	0.42*	0.35	0.27	-0.50**
Moniliformin	0.16	-0.36	0.14	0.13	0.17	0.36	-0.56**
Neoechinulin A	0.38*	-0.26	-0.12	-0.03	-0.06	0.03	-0.40*
Questiomycin Derivate	-0.06	0.41*	-0.29	-0.23	-0.19	-0.12	-0.12
from Penicillium	0.23	0.10	-0.19	-0.18	-0.11	0.32	-0.05

## Conclusions

Our results indicated ubiquitous contamination of TMRs with multiple mycotoxins. The most prevalent were *Fusarium* toxins like fumonisin B1 (FB1) (93%), B2 (FB2) (100%), B3 (FB3) (77%). Aflatoxin B1 (AFB1) was evidenced in 40% of the samples, and 7% exceeded the EU maximum limit for feeding dairy cattle (5 µg/kg). The dietary ingredients like corn grain, soybean meal, and canola meal were related to increased contamination of some mycotoxins (like FB1, FB2, and FB3) in TMRs from the Province of Punjab, Pakistan.

## References

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