ABSTRACT

Mycotoxins are toxic secondary metabolites, produced by *Fusarium* species are a worldwide problem, especially in cereals and cereal-based food and feed products. The most prevalent *Fusarium*-species in feedstuffs produced in Karnataka, India is *Fusarium verticillioides*. As rather little is known about the diversity of *Fusarium* species and occurrence of fumonisin levels, this study was conducted to devise new microbiological methods to detect and assess the significance of *Fusarium* species as natural contaminants. A total number of 120 animal feedstuffs and 80 poultry feed mixtures were analyzed for per cent incidence, diversity and frequency of *Fusarium* species by serial dilution and agar plating method. The study revealed the occurrence of nine diverse species of *Fusarium* with varying degree of per cent incidence and frequency. The *Fusarium* species include *Fusarium verticillioides*, F. proliferatum, F. anthophilum, F. oxysporum, F. pallidoroseum, F. solani, F. lateritium, F. graminearum and F. decemcellulare. Manual identification of *Fusarium* species was confirmed with molecular detection by PCR method using genus specific primers. The toxigenic potential of potential fumonisin- producing strains was also determined using genus-specific and species-specific primer sequences. The genus-specific primers included (ITS-F/ ITS-R), *fum1* gene specific (FUM1-F/ FUM1-R), *F. verticillioides* species specific (VERT-1/ VERT-2) and fumonisin-producing *F. verticillioides* species specific (VERTF-1/ VERTF-2) set of primers. A total number of 48 *Fusarium* isolates such as *Fusarium verticillioides* (20), F. oxysporum (2) and F. lateritium (1) from animal feeds, and *F. verticillioides* (22), F. oxysporum (2) and F. lateritium (1) from poultry feed mixtures were analyzed by PCR for their potential ability to produce fumonisins using two primer sets namely (VERT-1/ VERT-2) and (VERTF-1/ VERTF-2). A total of 375 *Fusarium* isolates such as *F. verticillioides* (157), F. proliferatum (8), F. anthophilum (8), F. pallidoroseum (5), F. oxysporum (10), F. graminearum (5), F. lateritium (4) and F. solani (1) from animal feedstuffs, while those isolated from poultry feed mixtures include *F. verticillioides* (145), F. proliferatum (3), F. anthophilum (6), F. pallidoroseum (17), F. oxysporum (2), F. lateritium (1), F. graminearum (1), F. solani (1), and *F. decemcellulare* (1), were analyzed by using *Fusarium* genus-specific (ITS-F/ ITS-R), and *fum-1* gene specific primers (FUM1-F/ FUM1-R). The
analytical method namely High Performance Liquid Chromatographic technique was used to detect the levels of fumonisins in 24 animal and poultry feed samples and 18 toxigenic *Fusarium verticillioides* (12), *F. proliferatum* (04) and *F. anthophilum* (02) isolates respectively. This method was applied to Indian animal feedstuffs and poultry feed mixtures that are predominantly maize-based products, to establish the occurrence and levels of fumonisins in samples collected from the region of Karnataka. The study revealed the presence of high levels of fumonisins in the animal feedstuffs and poultry feed mixture samples positive for fumonisin B₁ (FB₁) contamination. The study warrants the need for proper surveillance and monitoring of contaminated feeds before they are channeled for poultry or feeds mixtures for animal consumption.