

Microbiological Quality of Various Medicinal Herbal Teas and Coffee Substitutes

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Abstract: Various herbal teas including German chamomile, *Chrysanthemum* Vascuflow herb tea, hop, jasmine and orange flowers, sweet marjoram, spearmint and thyme leaves, and papaya-mint tea as well as coffee substitutes (Bambu instant Swiss, Teeccino chocolate-mint, and Teeccino Mediterranean Espresso) were analyzed for fungal contamination and the presence of aerobic mesophilic bacteria (APC). The results of this investigation showed that fungal counts reached levels as high as 5.8×10^5 colony forming units (cfu) per gram. German chamomile harbored the highest fungal contamination. The most common fungi found in herbal teas were *Aspergillus niger*, *Penicillium* spp., *Eurotium rubrum*, *E. chevalieri*, *A. flavus*, *Fusarium* spp., *Alternaria alternata*, and yeasts. Among the coffee substitutes, only the chocolate-mint coffee was contaminated with low numbers ($<1.0 \times 10^3$ cfu g⁻¹) of *E. rubrum*, *Ulocladium* spp. and *Phoma* spp., and with yeasts (<100 – 6.8×10^3 cfu g⁻¹). Aerobic mesophilic bacteria were recovered from 100% of the herbal tea, chocolate-mint and Mediterranean Espresso, and from 50% of the Bambu instant Swiss coffee samples. The highest APC counts of 1.2×10^7 cfu g⁻¹ were observed in spearmint leaves.

Keywords: moulds, yeasts, APCs, herbal teas, coffee substitutes

Introduction

Hot water infusions of botanicals have been used as drinks for thousands of years in the Far East and Mediterranean countries for their taste and flavors. Many of them are believed to have medicinal properties and they are used to treat minor illnesses and disturbances. German chamomile (*Matricaria chamomilla*), for instance, has been used since ancient times in the region around the Mediterranean Sea as a stomach-soothing agent, mild sedative and as a diuretic. Recent reports by Gyllenhaal and his co-workers (2000) described some sedative attributes of German chamomile, while research by Macchioni et al. (2004) demonstrated a significant acaricidal activity of extracts from chamomile flowers. *Chrysanthemum* tea has been used in Chinese medicine as a detoxifier, to maintain healthy cholesterol levels and to improve blood flow. This herb contains health-promoting compounds such as choline, vitamins A, B₁ and ascorbic acid, adenine, amino acids, flavonoids, glycosides and volatile oil (Gins et al. 2000), possesses anti-tumor properties (Ukiya et al. 2002), and has inhibitory effects against several bacteria and against the yeasts *Saccharomyces cerevisiae*, *Candida* spp. and *Hansenula anomala* (Shunying et al. 2005).

Infusions of spearmint (*Mentha spicata*), marjoram (*Origanum marjorana*), thyme (*Thymus vulgaris*) and papaya-mint tea are primarily consumed to treat digestive ailments, to reduce fever and for their anti-inflammatory, anti-oxidant and antimicrobial properties (Abe et al. 2004; Dorman et al. 2003; Mahady et al. 2005; Triantaphyllou et al. 2001; Dorman et al. 2004; Morton, 1987; Bagamboula et al. 2001). Marjoram, mint and other members of the *Lamiaceae* family contain phenolic compounds and flavonoids to which they probably owe their antioxidant attributes.

Hops (*Humulus lupulus*) and orange (*Citrus sinensis*) flowers are mainly used to relieve nervous tension (Gyllenhaal et al. 2000; Morton, 1987). Jasmine (*Jasminum grandiflorum*) flowers are used as an ingredient to add aroma and flavor to *Chrysanthemum* Vascuflow, green and other teas. This herb is also used as an alternative treatment for cancers and for viral and bacterial infections. Research by Kolanjiappan and Manoharan (2005) demonstrated a strong chemoprevention against experimental mammary carcinogenesis in rats.

Herbal coffee substitutes have the advantage over coffee that they do not contain caffeine and, depending on their ingredients, could contain various concentrations of health-promoting phytochemicals while they

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imitate coffee flavors. Therefore, an increasing number of health-conscious individuals are substituting coffee with these formulations. Several such products including chocolate-mint, Mediterranean Espresso and instant Swiss coffee are readily available in the U.S. market, mostly found in health stores and dietary supplement companies.

Although many herbal tea remedies are known and utilized for centuries, there is an increase of their consumption in recent years due to a trend to use natural therapies. Therefore, it is essential that these products are microbiologically safe. A few reports demonstrating microbial contamination of medicinal herbs from various parts of the world exist in the literature. Rizzo et al. (2004) indicated that medicinal plants in Argentina harbored toxigenic fungi such as *A. flavus*, *A. parasiticus* and several members of the Genus *Fusarium*; Efuntoye (1996) showed that dried medicinal plants from Nigerian herb markets contained *A. flavus*, *A. parasiticus* and *A. ochraceus*. The same investigator (1999) reported that the above fungal isolates were capable of elaborating mycotoxins when grown on semi-synthetic media. Martins et al. (2001), after evaluating several medicinal herbs obtained from Portuguese markets, reported that these commodities were infested with a variety of moulds such as *Aspergillus* and *Fusarium* spp., whereas Halt (1998) isolated a wide spectrum of fungi (including *Aspergillus*, *Penicillium*, *Alternaria*, *Cladosporium*, *Rhizopus* and *Mucor* species) from Croatian herbal teas and medicinal plants. Czech et al. (2001) reported bacterial and fungal contamination of medicinal herbs in Austria, while Skorska et al. (2005) showed that the air in chamomile and peppermint processing farms in Poland contained high levels of *Pantoea agglomerans* and other gram negative bacteria. Such reports indicate the existence of a ubiquitous problem. Consequently, this research was designed to determine if various medicinal herbal teas and coffee substitutes available in the U.S. market are contaminated with toxigenic fungi and if they contain high levels of aerobic mesophilic bacteria.

Materials and Methods

Materials

A total of 69 herbal tea and coffee substitute samples comprised of German chamomile, *Chrysanthemum* Vascuflow tea, hop, jasmine and

orange flowers, papaya-mint tea, sweet marjoram, spearmint and thyme leaves, and Bambu instant Swiss, Teeccino chocolate-mint and Mediterranean Espresso coffee substitutes were tested during the course of this experiment. Fifty-gram samples of spearmint, thyme, jasmine and hop flowers were obtained from bulk jars in local supermarkets; German chamomile, sweet marjoram, orange flowers and papaya-mint tea were purchased from commercial sources in their original, individual 4.0-oz (113 g) packages; *Chrysanthemum* Vascuflow tea was obtained from the same companies in intact boxes containing 20 tea bags; the coffee substitutes (Bambu instant Swiss, Teeccino chocolate-mint and Mediterranean Espresso) were also purchased from commercial sources in their individual, intact containers {8.5-oz (241 g) Teeccino chocolate-mint, 3.5-oz (99 g) Bambu instant Swiss and 0.85-oz (24 g) Teeccino Mediterranean Espresso}. Samples were purchased and maintained at room temperature until analysis; analysis was conducted within 1–4 days from the day of purchase.

Mycological analysis

All supplements were tested as follows: Ten grams of each sample were aseptically removed and transferred to sterile blender jars. Subsequently, each sample was blended in 90 ml of 0.1% peptone (Tournas et al. 2001) for 45 sec. Serial dilutions of the homogenate (in 0.1% peptone) were surface plated in duplicate potato dextrose agar (PDA) (DIFCO, Detroit, MI, U.S.A.) containing 0.01% chloramphenicol (0.1 ml/plate) and plates were incubated for 5 days at 25 °C. Then, colonies were counted and counts were expressed as colony forming units per gram (cfu g⁻¹); mould isolates were purified on PDA and further sub-cultured on malt extract agar (MEA), czapek yeast extract (CYA), 25% glycerol nitrate (G25N) and czapek yeast extract 20% sucrose (CY20S) agar for microscopic examination and identification. Identification was performed according to the methods and keys described by Pitt and Hocking (1997) and Nelson et al. (1983). The compositions of CYA, MEA, G25N and CY20S media are described in “Fungi and Food Spoilage” (Pitt and Hocking, 1997).

Aerobic plate count determination

The aerobic plate counts were determined as follows: Ten-gram sample portions were

aseptically transferred into sterile blender jars and treated as stated above except that Butterfield's phosphate buffer (Maturin and Peeler, 2001) was used as diluent instead of 0.1% peptone water. Serial dilutions were surface plated in duplicate plate count agar (PCA) plates. Plates were incubated at 35 °C for 48 hrs. Plate reading and colony counts determination were done according to the method described by Maturin and Peeler (2001).

Results and Discussion

Herbal teas

The mould and yeast (MY) counts from the various samples analyzed during the course of this investigation are shown in Table 1. One hundred per cent of the marjoram, spearmint and thyme leaves, papaya-mint tea, hop and jasmine flowers, 88% of the orange flowers and German chamomile, and 17% of the *Chrysanthemum* Vascuflow tea samples were contaminated with fungi. The highest contamination (5.8×10^5 cfu g⁻¹) was found in German chamomile and the lowest (1.0×10^2 cfu g⁻¹) was observed in jasmine

flowers. Fungal profiles of the various herbal teas tested are summarized in Table 2. German chamomile was contaminated with *A. ochraceus*, *E. chevalieri*, *Penicillium* spp., yeasts and low numbers ($<1.0 \times 10^3$ cfu g⁻¹) of *A. niger*, *A. flavus*, *A. alternata*, *E. rubrum* and *Rhizopus* spp. *A. niger* was the most frequently encountered mould found in 50% of the samples. Spores of *A. niger*, *A. flavus*, *A. alternata* and *Rhizopus* are common air contaminants probably present in the drying and packing areas (Lee and Jo, 2006; Zhang et al. 2005). At such low numbers, however, there is no indication that these organisms were growing on the product. If the storage conditions were to change (e.g. if the moisture of the product was to increase), they could proliferate and spoil the product, and possibly produce mycotoxins. *Penicillium* spp., *E. chevalieri* and *A. ochraceus* were found in higher numbers and that could mean that there was some growth of these organisms established before complete drying of the commodity. These organisms have the potential of producing mycotoxins under the right conditions. *A. niger*, *A. flavus*, *Fusarium* spp., *Penicillium* spp. and low levels

Table 1. Mould and yeast (MY) counts in various herbal teas and coffee substitutes.

Product	Number of samples tested	MY counts (cfu g ⁻¹) ^a (Range)	Frequency (% contam. ^b samples)
Herbal teas			
German Chamomile	8	<100– 5.8×10^5	88
<i>Chrysanthemum</i> Vascuflow tea	6	<100– 4.1×10^3	17
Hop flowers	2	4.0×10^2 – 1.1×10^4	100
Jasmine flowers	2	1.0×10^2 – 1.0×10^2	100
Marjoram leaves	10	2.0×10^3 – 1.8×10^4	100
Orange flowers	8	<100– 2.9×10^4	88
Papaya-mint tea	10	1.0×10^3 – 5.6×10^3	100
Spearmint leaves	3	1.3×10^4 – 1.4×10^5	100
Thyme leaves	2	7.0×10^3 – 1.6×10^4	100
Herbal coffee substitutes			
Bambu instant Swiss	6	<100	0
Teeccino Chocolate-mint	6	<100– 7.0×10^3	83
Teeccino Mediterranean Espresso	6	<100	0

^acfu, colony forming units; ^bcontam, contaminated.

Table 2. Fungal species and APCs^a found in herbal teas and coffee substitutes.

Organism	cfu g ⁻¹ (range) ^b	Frequency (% contam. ^c samples)
Herbal teas		
German chamomile		
<i>Aspergillus niger</i>	<100– 8.0×10^2	50
<i>Aspergillus flavus</i>	<100– 3.0×10^2	12
<i>Penicillium</i> spp	<100– 2.0×10^3	25
<i>Eurotium chevalieri</i>	<100– 1.0×10^3	25
<i>Eurotium rubrum</i>	<100– 5.0×10^2	12
<i>Aspergillus ochraceus</i>	<100– 1.0×10^3	12
<i>Alternaria alternata</i>	<100– 3.0×10^2	12
<i>Rhizopus</i> spp	<100– 7.0×10^2	12
Yeasts	<100– 5.8×10^5	12
APC	6.6×10^3 – 1.6×10^6	100
Chrysanthemum Vascuflow tea		
Yeasts	<100– 4.1×10^3	17
APC	8.4×10^3 – 3.2×10^4	100
Hop flowers		
<i>Cladosporium</i> spp	<100– 2.0×10^2	50
<i>Aspergillus niger</i>	2.0×10^2 – 2.5×10^2	100
Yeasts	<100– 1.1×10^4	50
APC	1.6×10^3 – 2.2×10^3	100
Jasmine flowers		
<i>Alternaria alternata</i>	1.0×10^2 – 1.0×10^2	100
Yeasts	<100	0
APC	3.4×10^3 – 9.0×10^3	100
Marjoram leaves		
<i>Aspergillus niger</i>	<100– 1.4×10^4	80
<i>Aspergillus flavus</i>	<100– 1.0×10^3	30
<i>Aspergillus</i> sp	<100– 1.2×10^3	40
<i>Penicillium</i> spp	<100– 1.0×10^3	30
<i>Eurotium chevalieri</i>	<100– 7.0×10^3	30
<i>Fusarium</i> spp	<100– 2.0×10^2	10
<i>Rhizopus</i> spp	<100– 7.0×10^2	20
<i>Aspergillus versicolor</i>	<100– 1.0×10^3	10
<i>Aspergillus carbonarius</i>	<100– 3.0×10^3	10
<i>Ulocladium</i> spp	<100– 6.0×10^2	10
Yeasts	<100– 3.9×10^3	20
APC	2.2×10^4 – 2.1×10^6	100
Orange flowers		
<i>Aspergillus niger</i>	<100– 3.0×10^3	88

(Continued)

Table 2. (Continued)

Organism	cfu g ⁻¹ (range) ^b	Frequency (% contam. ^c samples)
<i>Aspergillus flavus</i>	<100–1.0 × 10 ²	12
<i>Aspergillus</i> spp.	<100–1.0 × 10 ³	25
<i>Penicillium</i> spp.	<100–2.0 × 10 ²	12
<i>Alternaria alternata</i>	<100–2.0 × 10 ³	12
<i>Fusarium</i> spp.	<100–4.0 × 10 ²	12
Yeasts	<100–2.6 × 10 ⁴	12
APC	6.2 × 10 ⁴ –9.4 × 10 ⁵	100
Papaya-mint tea		
<i>Aspergillus niger</i>	<100–2.1 × 10 ³	90
<i>Aspergillus</i> spp.	<100–3.5 × 10 ²	20
<i>Aspergillus flavus</i>	<100–1.0 × 10 ³	10
<i>Eurotium</i> spp.	<100–1.3 × 10 ³	10
<i>Penicillium</i> spp.	<100–2.0 × 10 ³	40
<i>Fusarium</i> spp.	<100–1.0 × 10 ³	30
<i>Rhizopus</i> spp.	<100–2.0 × 10 ²	20
Yeasts	<100–3.8 × 10 ³	20
APC	7.8 × 10 ⁴ –7.1 × 10 ⁶	100
Spearmint leaves		
<i>Alternaria alternata</i>	8.1 × 10 ³ –7.4 × 10 ⁴	100
<i>Aspergillus</i> spp.	4.0 × 10 ² –5.5 × 10 ⁴	100
<i>Phoma</i> spp.	<100–1.0 × 10 ⁴	67
Yeasts	<100–4.8 × 10 ³	33
APC	3.1 × 10 ⁵ –1.2 × 10 ⁷	100
Thyme leaves		
<i>Aspergillus niger</i>	2.3 × 10 ³ –4.0 × 10 ³	100
<i>Aspergillus</i> spp.	1.0 × 10 ³ –7.8 × 10 ³	100
<i>Penicillium</i> spp.	4.0 × 10 ³ –4.0 × 10 ³	100
Yeasts	<100	0
APC	9.2 × 10 ⁴ –6.4 × 10 ⁵	100
Herbal coffee substitutes		
Bambu instant Swiss		
APC	<100–1.0 × 10 ³	50
Chocolate-mint		
<i>Eurotium rubrum</i>	<100–1.0 × 10 ²	17
<i>Phoma</i> spp.	<100–7.0 × 10 ²	33
<i>Ulocladium</i> spp.	<100–2.0 × 10 ²	17
Yeasts	<100–6.8 × 10 ³	67
APC	2.2 × 10 ³ –8.2 × 10 ³	100
Mediterranean Espresso		
APC	1.6 × 10 ³ –9.4 × 10 ³	100

^aAPCs, aerobic plate counts; ^bcfu, colony forming units; ^ccontam., contaminated.

(20–70 ppb) of fumonisin B₁ were found in chamomile by Martins et al. (2001a, 2001b). Since aspergilli and especially eurotia are capable of growing at low water activities, in order to avoid such growth and possible production of toxic metabolites, care should be taken to dry the product quickly before these moulds have the chance to establish any significant growth. Yeasts were present in 12% of the samples but they reached numbers as high as 5.8×10^5 cfu g⁻¹. Yeasts such as *Rhodotorula glutinis* and *Cryptococcus* spp. were reported as contaminants of chamomile by Martins et al. (2001a).

Seventeen per cent of the *Chrysanthemum* Vascuflow tea samples were contaminated with yeasts. No live moulds were found in this commodity (Table 2). Mould spores originally present in the herb were probably killed during processing. The isolated yeasts possibly originated from the personnel handling the materials after processing. Low numbers of *A. niger* were present in both hop flower samples, whereas one of the samples also contained *Cladosporium* spp. at levels not exceeding 2.0×10^2 cfu g⁻¹ and yeasts (1.1×10^4 cfu g⁻¹) (Table 2). Such low numbers of moulds are probably random air contaminants found in herb processing plants (Dutkiewicz et al. 2001). The higher incidence of yeasts could be due to contamination during handling after drying. Low incidence of *A. alternata* in jasmine flowers (1.0×10^2 cfu g⁻¹) seems insignificant and it could be attributed to random contamination from the air in the processing area (Dutkiewicz et al. 2001). No other fungi were isolated from this product.

All marjoram samples tested contained live fungi. MY levels ranged between 2.0×10^3 and 1.8×10^4 cfu g⁻¹ (Table 1). The most frequently isolated fungus was *A. niger* found in 80% of the samples at levels reaching as high as 1.4×10^4 cfu g⁻¹. One third of the samples contained *A. flavus*, *E. chevalieri* and *Penicillium* spp. at numbers reaching or exceeding 1.0×10^3 cfu g⁻¹. *Aspergillus* spp. other than *A. niger* and *A. flavus* were isolated from 40% of the tested samples; one sample contained *A. carbonarius* (3.0×10^3 cfu g⁻¹) and another was contaminated with *A. versicolor* (1.0×10^3 cfu g⁻¹) (Table 2). The isolation of various aspergilli, especially *A. flavus* and *A. carbonarius*, is of the highest concern because these organisms are known to produce aflatoxins and ochratoxin, respectively (Pitt and Hocking, 1997; Riba et al.

2008). *E. chevalieri*, *A. niger*, *A. versicolor* and *Penicillium* spp. also have the potential for toxigenesis (El-Kady et al. 1994; Perone et al. 2006). El-Kady et al. (1995) reported the presence of aflatoxin (9 ppb) and sterigmatocystin (17 ppb) in marjoram. Yeasts were found in 20% of the samples; their counts ranged between <100 and 3.9×10^3 cfu g⁻¹ (Table 2).

The main fungal contaminants found in orange flowers were *A. niger*, *Aspergillus* spp., *A. alternata* and yeasts. *A. niger* was the most frequently encountered fungus isolated from 88% of the samples, whereas yeasts were found at the highest levels reaching up to 2.6×10^4 cfu g⁻¹. *A. alternata* and yeasts were isolated from 12% while *Aspergillus* spp. other than *A. niger* were recovered from 25% of the tested samples (Table 2). Several *Aspergillus* species including *A. niger* were also isolated from orange tree leaves by Martins et al. (2001a).

Ninety per cent of the papaya-mint tea samples contained *A. niger*, 10% had *A. flavus* and *Eurotium* spp., 40% had *Penicillium* spp., 30% were contaminated with *Fusarium* spp. and 20% contained yeasts. The levels of these contaminants were below 4.0×10^3 cfu g⁻¹ (Table 2). Morton (1987) also reported the isolation of *F. solani* from papaya plants. This organism is a plant pathogen attacking papaya plants in the field. Spores of *Fusarium* could survive drying conditions and remain dormant for several months possibly years on the dried herb. During that time, if the moisture of the product would increase to levels allowing spore germination, significant mould growth and possibly mycotoxin production could occur. *Fusarium* could also grow on mint herb and produce toxins. The presence of fumonisin B₁ (160 ppb) in mint was reported by Omurtag and Yazicioglu (2004). Other organisms found in this product could be originating from its mint component.

All spearmint samples contained *Aspergillus* spp. and *A. alternata* at levels reaching 5.5×10^4 and 7.4×10^4 cfu g⁻¹, respectively. Sixty seven per cent of the analyzed samples were contaminated with *Phoma* spp. (up to 1.0×10^4 cfu g⁻¹) and 33% had live yeasts reaching levels as high as 4.8×10^3 cfu g⁻¹ (Table 2). *Alternaria* and *Phoma* are plant pathogens occasionally causing disease in mint plants in the field (PMSP 2002). Infection and colonization of the mint plants by these organisms could have started from the field and spoilage could become rapid and extensive after

harvest, when the plant defenses are weakened or eliminated. Alternatively, a few spores carried on healthy mint leaves from the field could also spoil the product after harvest during transport and storage if the moisture level were sufficient for fungal growth. Both, *Alternaria* and *Phoma*, have the ability to produce mycotoxins (Canafoglia et al. 2007; Lugauskas et al. 2006). Therefore, their presence in this herb should be kept as low as possible and the moisture of the product should be maintained at levels that do not allow fungal growth.

Both thyme leaf samples tested contained live *Aspergillus* spp., *A. niger* and *Penicillium* spp. at levels of 1.0×10^3 cfu g⁻¹ or higher. No yeasts were isolated from this product (Table 2). Some thyme oil constituents such as thymol have fungicidal properties, and they could be detrimental to yeast cells. Inhibition of the yeast *Candida albicans* by thyme oil was demonstrated in research conducted by Azaz et al. (2004).

The aerobic plate counts from herbal teas are shown in Table 2. One hundred per cent of the analyzed samples were contaminated with aerobic mesophilic bacteria. The highest APCs (1.2×10^7 cfu g⁻¹) were recovered from spearmint leaves and the lowest (1.6×10^3 cfu g⁻¹) were found in hop flowers. Higher numbers of bacteria could be explained by the fact that some of these organisms (e.g. *Bacillus* and *Clostridium* spp.) produce spores which are resistant to harsh processing, elevated heat and dry conditions. Therefore, they can survive for a long time on the product in a dormant state. *B. cereus* and *C. perfringens* were isolated from chamomile and other herbs by Martins (2001a). Also, part of the bacterial bioburden may have originated from the personnel handling the tea materials after processing, especially if strict GMPs and hygienic conditions were not followed, and from the processing plants' environment. The air and settled dust in herb processing plants are often highly contaminated with bacteria, which could add to the microbial burden already present in the commodities brought in from the field. Dutkiewicz and his coworkers (2001) reported the presence of *B. cereus*, *B. subtilis*, *Alcaligenes faecalis*, *Pseudomonas fluorescens*, *P. agglomerans*, *Staphylococcus epidermidis*, various fungi, and actinomycetes from herb (mint, marjoram, sage, etc) processing plants. Some of these organisms are capable of causing human infections, allergies and/or producing endotoxins, which makes them health risks (Dohmae et al. 2008; Obi et al. 1995). Therefore,

care should be taken to reduce such contaminants, mainly by following strict GMPs at all stages of processing.

Herbal coffee substitutes

Mould and yeast contamination of herbal coffee substitutes are summarized in Table 1. Fungi were isolated only from 83% of the chocolate-mint coffee samples. MY counts ranged from <100 to 7.0×10^3 cfu g⁻¹. The vast majority of the isolated organisms were yeasts contaminating 67% of the analyzed samples; moulds only comprised a small percentage of the total fungal population. *E. rubrum*, *Phoma* and *Ulocladium* spp. were recovered from this commodity in numbers not exceeding a few hundred cfu g⁻¹ (Table 2). Generally, such low numbers indicate random contamination from the environment. However, in this case, the mould contamination could be originating from the mint component of the product. Mint, as shown in previous pages, supports good growth of a variety of microfungi. Higher yeast populations could be the result of non-strict GMP and hygienic conditions during preparation and packing of the product. Bambu instant Swiss and Mediterranean Espresso coffee substitutes showed no fungal growth. These products possibly undergo harsher processing which inactivates any fungal entities present in the raw ingredients.

One hundred per cent of the chocolate-mint and Mediterranean Espresso and 50% of the Bambu instant Swiss coffee substitute samples contained aerobic mesophilic bacteria. APCs ranged from <100 to 9.4×10^3 cfu g⁻¹ (Table 2). Bacteria isolated from samples that had no live fungi could indicate that these organisms belong to processing-resistant species probably spore-formers such as *Bacillus* and *Clostridium* spp. Some of these species (e.g. *B. cereus* and *C. perfringens*) are known to cause human illness. Although hot water is added to the product during preparation of the coffee drink and before consumption, *Bacillus* spores are not killed by immersion in boiled water. Consumption of live spores could cause illness especially in individuals with compromised immune system. Therefore, care should be taken to rid these products of such bacteria.

Conclusions

Potentially toxigenic moulds including *A. flavus*, *A. niger*, *A. carbonarius*, *A. versicolor*, *A. ochraceus*,

A. alternata, *Eurotium*, *Fusarium* and *Penicillium* spp. were isolated from herbal teas at levels sometimes exceeding 1.0×10^4 cfu g⁻¹. Such levels indicate that these organisms could grow in the commodities. *A. niger* was the most frequently encountered mould. Yeasts were isolated from the majority of the samples and comprised a high percentage of the total fungal bioburden. Part of the yeast populations could be originating from the personnel handling the products after drying or other forms of processing. Therefore, strict GMPs and hygienic practices should be followed in order to minimize added contamination. Aerobic mesophilic bacteria were recovered from all herbal tea samples tested at numbers sometimes reaching or exceeding 1.0×10^6 cfu g⁻¹. Such high bacterial contamination could constitute a health hazard if the bacteria present are pathogenic or capable of producing toxin.

The herbal coffee substitutes analyzed were of better mycological quality than the herbal teas. Only the chocolate-mint coffee substitute contained live fungi. These fungi probably originated from the mint component of the formulation. Aerobic mesophilic bacteria ($\geq 1.0 \times 10^3$ cfu g⁻¹) were isolated from 83% of the herbal coffee substitute samples tested. More extensive surveys are needed in order to establish fungal profiles and APC levels in herbal teas and coffee substitutes. Additionally, these commodities should be tested for the presence of pathogenic bacteria.

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Disclosure

The authors report no conflicts of interest.

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