Xerophiles and other fungi associated with cereal baby foods locally produced in Uganda

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Fifty samples from five baby food products mainly made of cereal flour(s) were analyzed. The moisture contents of these products were between 11.14% and 11.9%, a level below 14.0%, the recommended level for safe storage of cereal grains and their products. The mycological analysis was carried out using the dilution plate method and two isolation media (DG18 for isolation of xerophilic fungi and DRBC for fungi in general). A total of 80 species related to 37 genera in addition to some unidentified fungal and yeast species were recorded on both media from the five products. The products were contaminated abundantly by xerophilic fungi which were occurring in 88% of food samples and accounting for 18.1% of the total CFU as recorded on DG18. The highest contamination level by xerophiles was registered in Mwebaza rice porridge (a component of rice flour) and the lowest in Mukuza (a product of maize, soyabean and sorghum flours). 11 xerophilic species were recorded of which Aspergillus and Eurotium (4 species each) were the predominant giving rise to 9.1% and 8.9% of the total CFU, with A. wentii, A. candidus, E. cristatum and E. repens were the most contaminating species. Of the fungi recorded other than xerophiles, species of Aspergillus (particularly A. flavus followed by A. niger), Penicillium (P. citrinum, P. oxalicum), Fusarium (F. solani, F. tricinctum), Cladosporium (C. sphaerospermum) and yeasts were the most predominant. Contamination of such foods is a matter of health hazard as these foods are for babies. So, the use of fresh, well-dried and uncontaminated flours for production of such foods is recommended.

Key words: mycobiota, spoilage, contamination, food products, DG18, DRBC

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INTRODUCTION

Baby foods rich in carbohydrates and proteins are being produced from dried cereal and leguminous grains/seeds. The manufacture of baby foods involves mixing the cereal flour with some additional ingredients such as powders of soya beans, dried fish or fruits. Since these dried foods possessed low moisture content levels, they are subject to contamination and spoilage by microorganisms. Fungi probably contaminate and spoil more foods than any other group of microorganisms. They render contaminated foods not only unpalatable, but also unsafe for consumption by producing mycotoxins (Munimbazi, Bullerman 1996). Sanchis et al. (1982) noted that certain species if humidity reaches a sufficient level would start producing toxic metabolites. The presence of aflatoxins in foods is of great concern in terms of food safety since they are among the most active ingested carcinogens (Pitt, Hocking 2009). Spoilage of such foods is due to a group of fungi termed as xerophiles that are capable of rapid growth above 0.77 a, and of slow growth at 0.75 a, and below-down to about 0.68 a,. Taligoola et al. (2004); Pitt, Hocking (2009) stated that the most common causes of spoilage of dried cereals are species of Eurotium particularly E. chevalieri, E. repens, E. rubrum and E. amstelodami L. Mangin. A variety of yeasts are also common on cereal grains and flour (Kurtzman et al. 1970; Aran, Eke 1987; Pitt, Hocking 2009). Other ingredients such as powders of soya beans, milk, dried fish and fruits were also found to be contaminated with a wide variety of fungal species (Mislivec, Bruce 1977; Sutic et al. 1979; Bullerman 1979; Jarchovska et al. 1980; Ito, Abu 1985; Pitt et al. 1994; Ismail, Saad 1997; Pitt, Hocking 2009).

Many reports have been published earlier at different localities of the world on the microbiological quality of baby foods (Jarchovská et al. 1980; Moustafa et al. 1984; Abdel-Sater, Ismail 1993; Zohri et al. 1995; Munimbazi, Bullerman 1996) or their ingredients such as wheat (Aran, Eke 1987; Mills et al. 1995), milk powder (Ismail, Saad 1997), rice (Pitt et al. 1994; Taligoola et al. 2004, 2010), maize (Ismail et al. 2003), sorghum (Pitt et al. 1994), millet (Mishra, Daradhiyar 1991), corn chips, breakfast cereal (Bullerman, Tsai 1994; Zohri et al. 1995), and baby foods imported into Uganda (Ismail et al. 2008, 2010).

In Uganda, knowledge about fungi contaminating locally produced foods is needed. Henceforth, this work was designed to survey the xerophilic and other fungi associated with baby foods locally manufactured in Uganda.

MATERIALS AND METHODS

Fifty samples of five baby food products manufactured locally (10 packets each) were randomly collected from different shops at five towns of Uganda (Kampala, Jinja, Mbarara, Masaka and Mbale). Most of these products are made in these towns. Each of these foods contained at least one or more cereal flour. The names, components and the producing companies of these products are shown in Table 1.

Determination of moisture content. Three sub-samples of 50 g each were taken from each food sample and put in aluminium foil dish. These were dried in an oven

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No	Product	Ingredients	Producing company	Mean moisture content (n=10)
1	Baby soya	Maize flour, soya bean flour, carrot flour	East African Basic Food, Ltd	11.47±0.23
2	Kayebe	Maize flour, powdered Enkejje (<i>Haplochromis</i> fish), soya bean flour	Kayebe Sauce Packers, Ltd	11.14±0.38
3	Mwebaza rice porridge	Rice flour	Ebenezer Packers, Ltd	11.9±0.23
4	Jacinta millet flour	Millet flour	Mahimba Company, Ltd	11.6±0.09
5	Mukuza	Maize flour, Sorghum flour, soya bean flour	Hodeco, Ltd	11.52±0.09

Table 1 Food products investigated, their ingredients, producing companies and their mean moisture contents

at 110°C for 24 hours, and reweighed (Magan, Lacey 1985; Pitt, Hocking 2009). The moisture content of each sample was expressed as the average percentage of the weight loss of the three replicates.

Mycological analysis. All food samples were analyzed mycologically on two isolation media: (1) Dichloran 18% glycerol agar (DG18) which contains glycerol and glucose needed for the growth of xerophilic fungi (Hocking, Pitt 1980) and (2) dichloran rose bengal chloramphenicol agar, DRBC (King et al. 1979). Dilutions were prepared by shaking 10 g of each sample in 90 ml diluent of 0.1% peptone water (Kurtzman et al. 1971). Serial tenfold dilutions were prepared and 1 ml aliquots of the appropriate dilution were placed in sterile Petri dishes. Eight plates were used for each food sample (4 plates for each isolation medium) i.e. 80 plates for each type of food products and in total 400 plates for the fifty food samples. Plates were incubated at room temperature (25-27°C) in day and night cycle of light conditions for 10-14 days for DG18 and for 10 days for DRBC plates. The growing fungi were enumerated, isolated and identified.

Identification of fungi. The identification of different fungal groups was carried out based on their macroscopic and microscopic features using the methods and keys described by Raper, Fennell (1965) for *Aspergillus* and its teleomorphs; Booth (1971) and Leslie, Summerell (2006) for species of *Fusarium*; Pitt (1979) for species of *Penicillium*; Ellis (1971), Moubasher (1993), Domsch et al. (2007), and Pitt & Hocking (2009) for other fungi.

Statistical analysis. Data were subjected to analysis of variance (ANOVA), using the Statistical Analysis System, (SAS institute Inc., 1996). Means were compared with L.S.D. test at $P \le 0.05$ levels.

RESULTS AND DISCUSSION

Each of the five baby foods investigated contained at least one or more cereal products (Tab. 1).

Moisture content of the products investigated. All samples of the five baby food products were characterized by the average moisture contents ranging from 11.14%

 \pm 0.38 % in Kayebe to 11.9 \pm 0.23 % in Mwebeza rice porridge (Tab. 1). However, all the products had moisture contents below 14.0%, the recommended level for safe storage of cereal grains and their products (Christensen, Kaufman 1974; Taligoola et al. 2004, 2010; Ismail et al. 2003, 2008, 2010).

Incidence of xerophilic fungi in baby food products (recovered mainly on DG18%). Baby foods locally manufactured in Uganda were abundantly contaminated by xerophilic fungi which occurred in 88% of food samples and accounted for 18.08% of the total CFU as recovered on DG18 (Tab. 2). The contamination level with xerophiles ranged from 2.62% of the total CFU in Mukuza to 60.44% in Mwebaza rice porridge (Tab. 3). These high contamination levels with xerophiles indicate that the baby foods may have stayed in shops and supermarkets for a long time where they might have been invaded by storage fungi. Bullerman (1979) reported that, food stuffs in shops and markets are actually under storage, hence fungal contamination is likely to occur. Ten xerophilic species belonging to four genera were recovered from the five products. Aspergillus (72% of 50 food samples) and Eurotium (64%) were isolated in high incidences while Wallemia Johan-Olsen and Xeromyces L. R. Fraser were infrequent (Tab. 2). The levels of contamination by xerophilic fungi in locally produced foods (88% of the food samples) were higher than those reported by Ismail et al. (2010) in imported ones (54%), however similar species of the genera Aspergillus, Eurotium and Wallemia were reported from both foods though in different frequencies.

Eurotium (4 species) occurred in 64% of the food samples and constituted the majority of xerophilic CFU (48.89%) from the 5 products. The highest level of contamination with Eurotium species was found in Mwebaza rice porridge (24.56% of the total CFU), and the lowest in Mukuza (1.58%) (Tab. 3). Eurotium species have been reported on cereal baby foods imported into Úganda (Ísmail et al. 2010), maize, rice, soya beans and dried fish (El-Kady, Youssef 1993; Pitt et al. 1994; Taligoola et al. 2004). Among the Eurotium species reported, E. cristatum and E. repens were the most predominant, accounting for 21.17% and 17.98% of the total xerophiles CFU. This finding agrees with earlier reports by Kurata et al. (1968) who found these two species to be common spoilage fungi in many cereals (rice, maize, sorghum, wheat and barley) in storage. The observation of E. repens in high occurrence on the foods analysed is inconsistent with the findings whereby E. repens and other unidentified Eurotium species were recovered in rare frequency on Turkish cereals and cereal products (Åran, Eke 1987). E. chevalieri and E. rubrum were infrequently encountered, however, these two species were reported earlier as frequent on maize and rice (Kurata et al. 1968; Taligoola et al. 2004) and on baby foods imported to Uganda (Ismail et al. 2010).

Xerophilic aspergilli (4 species) accounted for 50.13% of the xerophiles CFU and 9.06% of the total fungi CFU (Tab. 2). They were recovered in 72% of food samples from the five products. The highest level of contamination was found in Mwebaza rice porridge (35.87% of the total CFU) and the lowest was registered in Mukuza (1.04%). *A. wentii* was the most prevalent species, accounting for 25.95% of the xerophiles CFU. It was found most heavily contaminating Kayebe and Baby soya (major components: maize and soyabeans). In this respect, low levels of infection by *A. wentii* were recorded on soya beans, maize, paddy rice and sorghum in Thailand and Indonesia (Pitt et al. 1994) and on maize (Ismail et al. 2003) and rice (Taligoola

Таха			DG18					DRBC		
TURNE	CFU	CFU%	F%	OR	Source	CFU	CFU%	F%	OR	Source
Xerophilic fungi	28225	18.08	88	H	1.2.3.4.5	1825	1.08	25	W	1.2.3.5
Aspergillus (Total)	14150	9.06	72	H	1,2,3,4,5	1250	0.74	20	Г	
A. candidus Link	6550	4.20	16	Г	1	150	0.09	9	Я	5
A. penicillioides Spegazzini	175	0.11	8	ж	1,3,5	50	0.03	4	ч	1,2
restrictus Smith	100	0.06	8	Я	1,2,3	75	0.04	4	Ч	1,2
A. wentii Wehmer	7325	4.71	72	Н	1,2,3,4,5	975	0.58	14	Γ	0
Eurotium (Total)	13800	8.87	64	Н	1,2,3,4,5	500	0.30	10	ч	1,2,3
E. chevalieri Mangin	2475	1.59	18	Г	2,3,4,5	200	0.10	9	ч	m
E. cristatum (Raper & Fennell) Malloch & Cain	5975	3.84	56	Н	1,2,3,4,5	300	0.20	4	ч	1,2
E. repens de Bary	5075	3.26	34	M	1,2,3,4,5					
E. rubrum Konig, Spieckermann & Bremer	275	0.18	8	ч	1,3					
Polypaecilum pisce A. D. Hocking & Pitt						25	0.014	2	Я	1
Wallemia sebi (Fries) von Arx	150	0.09	4	ж	-	50	0.03	2	ч	
Xeromyces bisporus L. R. Fraser	125	0.08	2	R	1					
Xerotolerant fungi	127900	81.92	100	H	1,2,3,4,5	167415	98.92	100	Н	1,2,3,4,5
Acremonium strictum W. Gams	25	0.02	5	ч	S	200	0.12	10	z	1,3
Acrophialophora sp.						7525	4.4	4	Я	ε
Alternaria alternate (Fries) Keissler						75	0.04	2	ч	0
Aspergillus (Total)	32700	20.95	82	Н	1,2,3,4,5	39010	23.05	84	Н	1,2,3,4,5
A. aegyptiacus Moubasher & Moustafa						50	0.03	2	ч	
A. carbonarius (Bainier) Thom	125	0.08	2	Я	1	75	0.04	2	Я	1
A. deflectus Fennell & Raper						25	0.014	2	Ч	-
A. flavus Link	23725	15.19	64	Н	1,2,3,4,5	30435	18.0	74	Н	1,2,3,4,5
A. fumigates Fresenius	1000	0.64	20	Г	1	526	0.5	16	Г	2,3
A. niger van Tieghem	1575	1.01	42	Σ	1,2,3,4,5	1800	1.1	32	М	1,2,3,4,5
A. nomius Kurtzman, Horn & Hesseltine	25	0.02	2	Я	1					
A. ochraceus Wilhelm	800	0.51	30	Μ	1,2.3,4	450	0.3	12	R	2,3,5
A. oryzae (Ahlburg) Cohn	1800	1.15	22	Г	2,3,4	525	0.3	14	Г	L,2,3
A. parasiticus Speare	50	0.03	4	R	2	225	0.13	10	R	1,2
A. phoenicis (Corda) Thom						25	0.014	2	R	3
A. sydowii (Bainier & Sartory) Thom & Church	725	0.46	12	R	1,3,5	375	0.2	8	R	2,3
A. tamari Kita	2075	1.33	22	L	2,3,4	3050	1.8	24	L	1,2,3
A. terreus Thom	125	0.08	9	R	1,3	550	0.3	8	R	2,3
A. versicolor (Vuillemin) Tiraboschi	675	0.43	22	Γ	1.2.3.5	450	0.3	18	Γ	1,3

Incidence of fungi in local baby food products on dichloran 18% glycerol agar (DG18) and dichloran rose Bengal chloramphenicol agar (DRBC) Table 2

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1	1 5	C, I	1,2,3,5	1,3,5		1,2,3,4,5	1	e	e	e,	1	2,5	1	1	1,2,3,4,5	4				1,2,3,4,5	1,5	1,3,4,5		5	1	1	1,3	1	1,2,3,5	1,2,3,5	2,3	2,3	3	2,3,4,5	1.2.3
R	-	× ;	X	R		Μ	R	R	Я	ч	ч	Я	Я	R	Η	Я				Μ	Г	М		Я	R	R	R	R	Г	R	R	L	R	Μ	Γ
2	ç	7	46	8		40	4	2	0	2	2	×	7	2	56	4				26	20	32		7	4	7	9	2	20	12	9	14	2	28	12
0.03	0.014	0.014	1.6	0.14		1.3	0.03	0.014	0.014	0.09	0.014	0.09	0.014	0.03	25.6	0.03				14.25	10.5	0.8		0.014	0.04	0.03	0.07	0.1	2.5	1.2	0.09	1.3	2.6	4.8	0.2
50	30	67	2655	425		2230	50	25	25	150	25	150	25	50	43300	50				24100	17775	1375		25	75	50	125	200	4275	1950	150	2175	4325	8175	350
	n	1	1,2,3,4,5	1, 2, 3, 5	1	1, 3, 4, 5									1,2,3,4,5		4	1	n	4	5	2,3	e G				s				1,3		3	3,4,5	1.3
¢	×	;	Η	Μ	R	M									X		L	Я	Ч	R	Я	Я	Я				Я				Я		R	Μ	Я
	٥	4	68	34	2	40									42		16	6	7	2	12	10	6				9				×		2	30	10
	0.34	100	3.95	2.37	0.02	1.57									8.68		2.0	0.02	0.02	0.02	6.31	0.02	0.05				0.09				0.13		0.06	15.04	0.26
101	32	1 1 1 1	6175	3700	25	2450									13550		3125	25	25	25	9850	425	75				150				200		100	23475	400
Botryotrichum piluliferum Saccardo & Marchal	Byssochtamys futive S. L. Ullivr & G. M. Smith	Chaetomum sp.	Cladosporium (Total)	C. cladosporioides (Fresenius) de Vries	C. herbarum (Persoon) Link	C. sphaerospermum Penzig	Cochliobolus lunatus R. Nelson & Haasis	Curvularia sp.	Doratomyces sp.	Emericella nidulans (Eidam) Vuillemin	Epicoccum nigrum Link	Eupenicillium sp.	Fennellia flavipes Wiley & Simmons	Fusariella sp.	Fusarium (Total)	E equiseti (Corda) Saccardo	^r . lateritium Nees	^c . oxysporum Schlechtendal	F. poae (Peck) Wollenw.	⁷ solani (Martius) Saccardo	F. tricinctum (Corda) Saccardo	⁷ . verticillioides (Saccardo) Nirenberg	fusarium sp.	Geotrichum candidum Link	Hypomyces chrysospermus Tul.	Lasiodiplodia theobromae (Pat.) Griffon & Maubl.	Microdochium nivale (Fries) Samuels & Hallett	<i>Monilia</i> sp.	Mucor (Total)	M. plumbeus Bonord.	M. racemosus Fresenius	Mucor sp.	Veosrtorya fischerii (Wehmer) Malloch & Cain	Veurospora crassa (Shear & Dodge) v. Arx	Paecilomyces variotii Bainier

Table 2 – cont.

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1,2,3,4,5			1,2,3,4,5	1,2,3		2,3,4	e	e		1,3	1,2			1, 3, 4, 5	1	7	1,2	1,3,5	1,2,3,4,5	2,3	1,4	3,4,5	e	1,2,3,4,5		
Н			Σ	R		Г	2	2		2	Г		2	M	Я	ч	R	Г	Μ	Г	Я	Г	2	Н		
58			40	8		24	9	0		4	16		7	36	7	7	4	18	34	20	4	14	7	100	36	65
7.9			3.5	0.15		3.50	0.04	0.014		0.04	0.5		0.2	3.0	0.03	0.014	0.03	0.74	21.2	0.6	0.07	20.5	0.014	100		
13400			5950	250		6075	75	25		75	950		325	5075	50	25	50	1250	35775	975	125	34750	25	169240		
1.2.3.4.5	5	2	1,2,3,5	2,3,4,5	n	1,2,3,4,5			1			m		3,4,5					1,2,3	2		1,2,3,5		1,2,3,4,5		
Η	ч	ч	Σ	L	ч	Σ			ч			z		Γ					Г	Я		L		Η		
80	6	8	28	18	7	44			7			7		22					22	7		22		100	16	48
10.65	0.09	0.42	2.58	4.82	0.03	2.64			0.03			0.03		2.63					18.81	0.16		18.65		100		
16625	150	650	4025	7525	50	4125			50			50		4100					29375	250		29125		156125		
enicillium (Total)	chermesinum Biourge	chrysogenum Thom	citrinum Thom	corylophilum Dierckx	islandicum Sopp	: oxalicum Currie & Thom	: pinophilum Hedgcock	: puberulum Bainier	: purpurogenum Stoll	<i>Pariabile</i> Sopp	? viridicatum Westling	enicillium sp.	homa sp.	hizopus stolonifer (Ehrenberg) Vuillemin	copulariopsis candida (Gueguen) Vuillemin	hermoascus aurantiacus Miehe	richoderma harzianum Rifai	Other unidentified fungi	easts	Chodotorula mucilaginosa (Jörgensen) Harrison	(easts (brown)	(easts (white)	(easts (yellow)	otal fungi	Io. of genera (37)	Io. of species (80)

Abbreviations: CFU = Colony forming units (calculated /g baby food product in 50 samples); CFU% = Percentage colony forming units (calculated per total tungal CFU); F% = percentage frequency (calculated per 50 samples investigated); OR = occurrence remarks; High (H) = 50-100 %, Moderate (M) = 25-49 %, Low (L) = 13-24 %, Rare (R) = less than 13 %; Source: 1 = Baby soya, 2 = Kayebe, 3 = Mwebaza rice porridge, 4 = Jacinta millet flour, 5 = Mukuza.

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Table 3	Colony forming units and percentage frequency of most common fungi, and the number of genera and species recovered from the five baby food	products on DG18 and DRBC
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	BC	F%	20				100	70	20	10	10	40	10	40	10	10		40	40	100		
tuza.	DR	CFU	100				46350	1050	50	25	25	300	75	225	175	150		150	34075	46450	8	16
Muk	18	F%	80	09	20	80	100	60	30		10	70			20	40	20	20		100		
	DG	CFU	1075	225	125	300	39975	850	125		75	475			125	500	50	50		41050	7	50
н	BC	F%					100	70	10			10	100	20	100	40	20	90	20	100		
illet flor	DR	CFU					47275	575	25			550	23925	275	7825	175	50	4550	725	47275	9	10
cinta m	18	F%	90	40	20	80	100	40	09	20		99	10		100		100	50	10	100		
Jac	DG	CFU	5175	300	3100	1525	42000	1125	625	75		1100	25		23050		2625	3725	125	47175	7	16
dge	3C	F%	30				100	70	30	30	20	30		60	20	30	10	40	50	100		
ce porri	DRI	CFU	200				21400	1750	800	350	350	500		350	100	1250	150	300	350	21600	18	34
baza ri	DG18 DG18 CFI1 F02	F%	100	50	40	40	100	70	30	50	60	40		40	30	30	30	40		100		
Mwe		CFU	11200	2150	275	150	7325	400	175	375	250	600		300	300	575	900	325		18525	12	35
	ЗС	F%	50	10		40	100	100	60	20		40		10	10	80	90		50	100		
sbe	DRI	CFU	1125	275		825	47910	26660	700	75		75		200	75	4125	5875		600	49035	11	27
Kay	18	F%	100	60	30	100	100	100	70	50	60			10		50	50			100		
	DG	CFU	9275	2925	1200	5025	35200	21075	525	250	2750			125		2850	475			44475	4	19
	ŝ	F%	50	10		30	100	60	40		10	80	20	30		40		10	10	100		
soya	DRI	CFU	375	25		150	4505	400	225		50	805	100	325		250		25	52	4880	24	39
Baby	18	F%	70	70	70	60	100	50	20	30	40	30				20	20		10	100		
	DG	CFU	1500	375	375	325	2900	275	125	100	625	275				100	75		75	4400	8	25
Product	Medium	Common fungi	Xerophiles	Eurotium cristatum	E. repens	Aspergillus wentii	Xerotolerants	A. flavus	A. niger	A. ochraceus	Cladosporium cladosporioides	C. sphaeros- permum	Fusarium solani	F. verticil- lioides	Neurospora crassa	Penicillium citrinum	P. oxalicum	Rhizopus stolonifer	Yeasts (Total)	CFUs of all fungi	No. of genera	No. of species

For abbreviation see Table 2; A = Product, B = Medium, C = Common fungi; A*B*C L.S.D at P \leq 0.05 = 210.52

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et al. 2004) in Uganda. A. candidus was the second most common xerophilic Aspergillus species. It was recovered in 16% of the samples, however giving rise to high percentages of CFU (23.21% of xerophiles) and found only in Baby soya. A. penicillioides and A. restrictus occurred infrequently in Baby soya, Kayebe, Mwebaza rice porridge or Mukuza. These two species were also uncommon in maize and rice in Uganda (Ismail et al. 2003; Taligoola et al. 2004), and in baby foods imported to Uganda (Ismail et al. 2010), while A. restrictus was predominant on cereal grains in Iran (Lacey 1988) and A. penicillioides was isolated in high frequency from soya beans in Thailand (Pitt et al. 1994) and dried fish in Indonesia (Wheeler et al. 1986). The presence of A. restrictus on Baby soya and Kayebe (products of soya beans), is in agreement with the finding where A. restrictus was found invading soya beans whose moisture contents were between 12.5%-13% (Christensen, Kaufmann 1965). This fungus was first reported in stored grains by Tuite, Christensen (1955) and since then has been found to be a common cause of deterioration in all kinds of stored grains and seeds (Christensen, Kaufmann 1965).

Wallemia sebi and *Xeromyces bisporus* were infrequently encountered and only from Baby soya. *W. sebi* was earlier reported in maize and soya beans from Thailand, and on peanuts, maize, paddy and milled rice, and soya beans from Indonesia (Pitt et al. 1994) and on Pakistani and Ugandan rice (Taligoola et al. 2004), while *X. bisporus* was reported from dried prunes, spice powders, nutmegs, dates, fruit cakes and cookies (Pitt, Hocking 2009).

It is worthy to mention that 7 of these xerophilic species in addition to *Polypaecilum pisce* were reported on DRBC but infrequently, accounting for a minor proportion of total CFU (1.08%).

Incidence of fungi other than xerophiles in baby food products (recovered on both DG18 and DRBC). Apart from the 11 xerophilic species, baby food products yielded a total of 33 genera and 69 species of xerotolerant fungi on both DG18 (38 species and 12 genera) and dichloran rose Bengal chloramphenicol agar (DRBC) (57 species belonging to 32 genera). The broadest spectrum of species was recorded in Mwebaza rice porridge (35 species on DG18) and in Baby soya (39 species on DRBC), while the narrowest was recorded in Jacinta millet flour (16 species on DG18 and 10 species on DRBC). The current results revealed that the diversity of fungi was higher in locally produced foods (36 genera and 65 species on DRBC) than in imported ones (21 and 42) analysed by Ismail et al. (2008). Aspergillus, Penicillium, Fusarium and Cladosporium were the most predominant genera on both isolation media (Tab. 2). Penicillium and Aspergillus were reported earlier as the most commonly isolated genera on starches (potatoes, rice, maize and wheat) intended for human consumption (Suarez et al. 1981), on wheat, maize, sorghum and barley in Egypt (Moubasher et al. 1972; El-Maghraby 1989). In a study on baby foods imported into Uganda, the most common fungal genera were found to be similar to those reported in the current study from the local foods, however, species of Aspergillus and Penicillium were more dominant in locally produced foods, while species of Cladosporium and Fusarium were more common in imported ones (Ismail et al. 2008).

Aspergillus was the most frequent genus. It emerged from 82% and 84% of food samples accounting for 20.95% and 23.05% of the total CFU on DG18 and DRBC respectively. It was represented by 15 species of which A. flavus was the most common. A. flavus accound for 15.19% and 18.0% of the total fungi CFU on DG18 and

DRBC, respectively (Tab. 2). It was recovered in high frequency from all products on both media but had its highest level of contamination in Kayebe (Tab. 3). The high incidence of *A. flavus* on Kayebe whose major components are maize, soya beans and fish flours, is in agreement with earlier reports on maize (El-Maghraby 1989; Sebunya, Yourtee 1990, Munimbazi, Bullerman 1996, Ismail et al. 2003), on dried fish (Ito, Abu 1985) and on soya beans (El-Kady, Youssef 1993). Contrary to the above findings, *A. flavus* was reported with low occurrence in maize and sorghum in Egypt (El-Kady et al. 1982) and soya beans in Uganda (Sebunya, Yourtee 1990).

A. niger came second and was found contaminating 42% and 32% of food samples from the five products, accounting for 1.01% and 1.1% of the total fungi CFU on DG18 and DRBC, respectively. It was recovered in moderate frequency and most heavily contaminating Kayebe and Baby soya (products of maize and soya beans). This is in agreement with the findings of Suarez et al. (1981), who recovered A. niger from starches intended for human consumption, where rice and maize were among these starches. Also, Sanchis et al. (1982) fond that A. niger caused severe deterioration to corn and sorghum. Other four Aspergillus species were recovered in low frequency of occurrence on both media, accounting collectively for 3.55% and 4.75% of the total fungi CFU on DG18 and DRBC media, respectively, and these were: A. fumigatus, A. oryzae, A. tamarii and A. versicolor (Tab. 2). Earlier reports mentioned these species to occur less commonly on freshly ground mouldy maize meal (Marasas, Smalley 1972). A. ochraceus was reported moderately on DG18 but rarely on DRBC giving rise to 0.51% and 0.3% of the total CFU respectively. On the other hand, 8 species of Aspergillus were isolated in rare incidence on one or both media: A. aegyptiacus, A. carbonarius, A. deflectus, A. nomius, A. parasiticus, A. phoenicis, A. sydowii and A. terreus (Tab. 2). A. carbonarius and A. parasiticus have been reported as rare species in food products imported into Uganda (Ismail et al. 2008, 2010), though some of the above species have been reported to be common on barley e.g., A. sydowii, A. ochraceus, A. terreus (Moubasher et al. 1972) and A. terreus on maize (Ismail et al. 2003), paddy rice (Abdel-Hafez et al. 1987) and flour (Augustine et al. 1984).

Penicillium was the second most frequent genus, recovered from 80% and 58% of the total food samples on DG18 and DRBC respectively, accounting for 10.65% and 7.9% of the total CFU. The highest levels of contamination with penicillia were recorded in maize flour-containing products (Kayebe and Baby soya), and Mwebaza rice porridge and the least was found in Mukuza (Tabs 2 and 3). The above findings agree with earlier observation where Penicillium was among the most common genera recovered from corn snacks (Zohri et al. 1995) and starches (Suarez et al. 1981). Similarly, corn was found to be highly contaminated with Penicillium species, while rice was found to be penicillia free (Munimbazi, Bullerman 1996). In contrast to our finding, whereby Mukuza (a product of sorghum) had a low contamination level with Penicillium species, Diener et al. (1981) found Penicillium to be among the most dominant genera on sorghum. P. citrinum (2.58% and 3.5%) and P. oxalicum (2.64% and 3.5% of the total CFU on DG18 and DRBC respectively), the most common species in the present study, have been reported earlier to occur on starches (Suarez et al. 1981), and barley (Abdel-Kader et al. 1979). P. oxalicum was also one of the chief species isolated from unstored corn kernels (Mislivic, Tuite 1970) and in preharvest corn from Valencia, Spain (Jimenez et al. 1985). P. citrinum,

a nephrotoxigenic fungus, is known as citrinin-producer (Mislivec, Tuite 1970; Frisvad 1983). *P. viridicatum*, a well known nephrotoxigenic species (Frisvad 1983) and *P. corylophilum* were also isolated though in low frequency, respectively on DRBC and DG18, constituting 0.5% and 4.82% of the total CFU. *P. viridicatum* was earlier recovered from starches intended for human consumption (Suarez et al. 1981). Carlton et al. (1968) found that *P. viridicatum*, *P. oxalicum* and *P. multicolor* Grig.-Manoil. & Porad, all isolated from corn kernels in Indiana were toxic to mice when included in their diets. Other 7 *Penicillium* species: *P. chermesinum*, *P. chrysogenum*, *P. islandicum*, *P. pinophilum*, *P. puberulum*, *P. purpurogenum* and *P. variabile* were rarely isolated from only one or two products (Tab. 2). Of these, *P. variabile* has been reported to cause severe deterioration in wheat, corn and sorghum (Moubasher et al. 1972) and *P. aurantiogriseum*, *P. chrysogenum*, *P. viridicatum* from baby foods imported into Uganda (Ismail et al. 2008, 2010).

Fusarium occupied the third place with regard to its frequency, occurring in 42% and 56% of the samples on DG18 and DRBC respectively. However its count (25.6% of the total CFU) was more than that of Aspergillus and Penicillium on DRBC while lower than that of both genera on DG18 (Tab. 2). This result agrees with the earlier finding that species of Fusarium are field fungi (Moubasher et al. 1972; Christensen 1987) less tolerating the low water activity medium, DG18. The results revealed that Fusarium represented the highest and the major food-contaminant. Among the five products investigated, Jacinta millet flour and Mukuza were the most heavily contaminated having Fusarium CFU in all their samples. It is possible that fusaria infected the cereals while still in the field and persisted even after the cereals were processed and stored. Of eight species encountered, F. solani (0.02% and 14.24%) of the total CFU) and F. tricinctum (6.31% and 10.5%) were the most contaminating on both DG18 and DRBC respectively, though these were recovered in rare to moderate frequency. F. solani was found most heavily contaminating Jacinta millet flour while F. tricinctum in Mukuza (a product of sorghum and maize). This finding disagreed with earlier reports where F. solani and F. tricinctum were absent on millet and sorghum (Diener et al. 1981; Munimbazi, Bullerman 1996). F. verticillioides constituted low percentages of the total CFU and was found most heavily contaminating Baby soya and Mwebaza rice porridge (products of maize and rice flour, respectively). F. verticillioides and F. tricinctum were registered earlier in maize meal (Marasas, Smalley 1972), maize stalks and grains (Logrieco et al. 1988), and sorghum (Diener et al. 1981). F. verticillioides is a major producer of moniliformin and fumonisins toxins that cause liver cancer in rats and oesophageal cancer in humans (Lacey 1988, Logrieco et al. 1988; Sydenham et al. 1990). The remaining Fusarium species were recorded infrequently either on DG18 (F. lateritium from 8 food samples from Jacineta millet flour, F. oxysporum from one Baby soya sample, F. poae from one sample of Mukuza and unidentified Fusarium species from one sample of Mwebaza) or on DRBC (F. equiseti from only 2 samples of Baby soya).

Cladosporium (3 species) was also isolated from all products in high frequency (68% of food samples) on DG18 and moderate frequency (46%) on DRBC, constituting 3.95% and 1.6% of the total CFU on DG18 and DRBC, respectively. *C. sphaerospermum* was moderately isolated on both media with 1.57% and 1.3% of the total CFU while *C. cladosporioides* was moderate on DG18 and rare on DRBC

(Tab. 2). *C. herbarum* was rare and recovered only from 1 sample of Kayebe. In this respect, Mazen et al. (1984) found *C. sphaerospermum* in low frequency on maize.

Neurospora crassa, Rhizopus stolonifer and yeasts (with *Rhodotorula mucilaginosa* being the most common) were all isolated in moderate or low frequency on DG18 and in moderate frequency on DRBC. These species were reported earlier as food spoilage fungi (Pitt, Hocking 2009). *R. stolonifer* had also been reported earlier from soya beans (El-Kady, Youssef 1993) and barley (Abdel-Kader et al. 1979).

Some other fungal species were isolated infrequently either in low or rare frequency from one or more food products on DG18 or DRBC or both (Tab. 2).

Analysis of variance. Analysis of variance was computed on baby food products using Anova test at 5% significance level (Tab. 3). The calculated value of $F_{test} = 8.84$ at df 4. This is greater than the tabulated value $F_{critical} = 1.96316$, hence there is a significant difference in the total count of the different species recovered from the different food products on DRBC and DG18. The type and the CFU of most fungal species recovered on DRBC and DG18 from food products are probably dependent on the type of that product. This may also be due to the different ingredients including cereal flours involved in these products.

CONCLUSIONS

The current results revealed that Kayebe (a product of maize, fish and soya bean) and Jacinta millet flour (a product of millet) were the most heavily contaminated by fungi CFU of the five products investigated, while Baby soya was the lowest as determined on both isolation media. However, the highest contamination level by xerophiles was registered in Mwebaza rice porridge and the lowest in Mukuza. Among eleven xerophilic species recorded on these baby foods, species of Aspergillus and Eurotium were the most common. In addition, a high incidence of Aspergillus flavus, Yeasts, Fusarium solani, F. tricinctum, Penicillium citrinum, P. corylophilum, P. oxalicum and Cladosporium sphaerospermum on one or both isolation media were also recorded. Many of these fungi are capable of producing mycotoxins. Contamination of such foods (especially those for babies) is a matter of health hazard for human consumption. However their safety can be insured and improved greatly by using quality raw materials. As contamination occurs for cereal grains before, during or after harvesting, during drying process, or even during food production and this contamination could also be due to long-term storage, marketing under non-hygienic conditions of the food products. We suggest that monitoring fungal contaminations as well as mycotoxins should be carried out periodically and procedures to prevent mould contamination should be developed.

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REFERENCES

- Abdel-Hafez S.I.I., El-Kady I.A., Mazen M.B., El-Maghraby O.M. 1987. Mycoflora and trichothecene toxins of paddy grains from Egypt. Mycopathologia 100 (2): 103–112.
- Abdel-Kader M.I.A., Moubasher A.H., Abdel-Hafez S.I.I. 1979. Survey of the mycoflora of barley grains in Egypt. Mycopathologia 69 (3): 143–147.
- Abdel-Sater M.A., Ismail M.A. 1993. Enzymatic studies on fungi associated with biscuits in Egypt. International Biodeterioration and Biodegradation 31 (4): 277–292.
- Aran N., Eke D. 1987. Mould mycoflora of some Turkish cereals and cereal products. MIRCEN Journal of Applied Microbiology and Biotechnology 3 (3): 281–287.
- Augustine B., Parvathi M., Kalyanasundaram I. 1984. Potential mycotoxigens in wheat and wheat products. Journal of Food Science and Technology, India 21 (5): 312–316.
- Booth C. 1971. The genus *Fusarium*. Commonwealth Mycological Institute, Kew, Surrey, England, pp. 237.
- Bullerman L.B. 1979. Significance of mycotoxins to food safety and human health. Journal of Food Protection 42 (1): 65–86.
- Bullerman L.B., Tsai W.Y.J. 1994. Incidence and levels of *Fusarium moniliforme, Fusarium proliferatum* and fumonisins in corn and corn-based foods and feeds. Journal of Food Protection 57 (6): 541–546.
- Carlton W.W., Tuite J., Mislivec P. 1968. Investigations of the toxic effects in mice of certain species of *Penicillium*. Toxicology and Applied Pharmacolology 13 (3): 372–387.
- Christensen C.M. 1987. Field and storage fungi. In: Food and Beverage Mycology, 2nd edition, Beuchat L.R. (Ed), Van Nostrand Reinhold, New York.
- Christensen C.M., Kaufmann H.H. 1965. Deterioration of stored grains by fungi. Annual Review of Phytopathology 3 (1): 69–84.
- Christensen C.M., Kaufman H.H. 1974. Microflora: In Storage of cereal grains and their products. C.M. Christensen (ed.). American Association of Cereal Chemists, St. Paul Minnesota, pp. 159–191.
- Diener U.L., Morgan-Jones G., Wagener R.E., Davis N.D. 1981. Toxigenicity of fungi from grain sorghum. Mycopathologia 75 (1): 23–26.
- Domsch K.H., Gams W. and Anderson T.-H. 2007. Compendium of soil fungi. 2nd edition IHW-Verlag, Eching, Germany, 672 pp.
- El-Kady I.A., Youssef M.A. 1993. Survey of mycoflora and mycotoxins in Egyptian soybean seeds. Journal of Basic Microbiology 33 (6): 371–378.
- El-Kady I.A., Abdel-Hafez S.I.I., El-Maraghy S.S. 1982. Contribution to the fungal flora of cereal grains in Egypt. Mycopathologia 77 (2): 103–109.
- Ellis M.B. 1971. Dematiaceous Hyphomycetes. Commonwealth Mycological Institute, Kew, Surrey, England, 608 pp.
- El-Maghraby O.M. 1989. The fungal flora and natural occurrence of mycotoxins in cereal grains varieties and hybrids. Bulletin of the Faculty of Science, Assiut University, Egypt 18 (1-D): 131–142.
- Frisvad J.C. 1983. A selective and indicative medium for groups of *Penicillium viridicatum* producing different mycotoxins in cereals. Journal of Applied Bacteriology 54 (3): 409–416.
- Hocking A.D., Pitt J.I. 1980. Dichloran glycerol medium for enumeration of xerophilic fungi from low moisture foods. Applied and Environmental Microbiology 39 (3): 488–492.
- Ismail M.A., Saad N.M. 1997. Studies on the mycological quality of milk powder. Journal of Food Science and Technology, India 34 (6): 488–493.
- Ismail M.A., Taligoola H.K., Ssebukyu E.K. 2003. Mycobiota associated with maize grains in Uganda with special reference to aflatoxigenic Aspergilli. Journal of Tropical Microbiology 2 (1): 17–26.
- Ismail M.A., Taligoola H.K., Rebecca Nakamya 2008. Mycobiota associated with baby food products imported into Uganda with special reference to aflatoxigenic aspergilli and aflatoxins. Czech Mycology 60 (1): 75–89.
- Ismail M.A., Taligoola H.K., Rebecca Nakamya 2010. Incidence of xerophilic/xerotolerant mycobiota, fusaria, and nephrotoxigenic penicillia in some cereal baby foods imported to Uganda. Journal of Basic and Applied Mycology, Egypt 1 (1): 23–33.
- Ito H., Abu M.Y. 1985. Study of microflora in Malaysian dried fishes and their decontamination by gamma-irradiation. Agricultural and Biological Chemistry 49 (4): 1047–1051.
- Jarchovska H., Koudelka J., Lukasova J. 1980. Microbiological quality of infant foods made from dried milk in Czechoslovakia. Veterinarni Medicina 25 (11): 691–695.

- Jimenez M., Sanchis V.M., Santamarina P., Hernandez E. 1985. *Penicillium* in pre-harvest corn from Valencia (Spain): Influence of different factors on the contamination. Mycopathologia 92 (1):53–57.
- King A.D., Hocking A.D., Pitt J.I. (1979): Dichloran-rosebengal medium for enumeration and isolation of moulds from foods. Applied and Environmental Microbiology 37 (5): 959–964.
- Kurata H., Udagawa S., Ichinoe M., Kawasaki M., Takoda M., Tazawa M., Koizurni A., Tanabe H. 1968. Studies on the population of toxigenic fungi in foodstuffs. III. Mycoflora of milled rice harvested in 1965. Journal of the Food Hygienic Society of Japan 9: 23–28.
- Kurtzman C.P., Wickerham L.J., Hesseltine C.W. 1970. Yeasts from wheat and flour. Mycologia 62 (3):542–547.
- Kurtzman C.P., Rogers R., Hesseltine C.W. 1971. Microbiological spoilage of mayonnaise and salad dressings. Applied Microbiology 21 (5): 870–874.
- Lacey J. 1988. The microbiology of cereal grain from area of Iran with high incidence of oesophageal cancer. Journal of Stored Production and Research 24 (1): 30–50.
- Leslie J.F., Summerrell B.A. 2006. The Fusarium laboratory manual. Blackwell Publishing, pp.388.
- Logrieco A.M.A., Bottalico A., Altomare C. 1988. Chemataxonomic observations on zearalenone and trichothecene production by *Gibberella zeae* from cereals in Southeren Italy. Mycologia 80 (6): 892–895.
- Magan N., Lacey J. 1985. Interactions between field and storage fungi on wheat grains. Transactions of the British Mycological Society 85 (1): 29–37.
- Marasas W.F.O., Smalley E.B. 1972. Mycoflora, toxicity and nutritive value of mouldy maize. Onderstepoort Journal of Veterinary Research 39 (1): 1–10.
- Mazen M.B., Abdel-Hafez S.I.I., Shaban G.M.M. 1984. Survey on the mycoflora of Egyptian wheat grains and their lemmae and paleae. Mycopathologia 85 (3): 155–159.
- Mills J.T., Seifert K.A., Frisvad J.C., Abramson D. 1995. Nephrotoxigenic *Penicillium* species occurring on farm-stored cereal grains in western Canada. Mycopathologia 130 (1): 23–28.
- Mishra N.K., Daradhiyar S.K. 1991. Mold flora and aflatoxin contamination of stored and cooked samples of pearl millet in the Paharia tribal belt of Santhal Pargana, Bihar, India. Applied and Environmental Microbiology 57 (4): 1223–1226.
- Mislivec P.B., Tuite J. 1970. Species of *Penicilium* occurring in freshly harvested and in stored dent corn kernels. Mycologia 62 (1): 67–74.
- Mislivec P.B., Bruce V.R. 1977. Incidence of toxic and other mold species and genera in soyabeans. Journal of Food Protection 40: 309–312.
- Moubasher A.H. 1993. Soil fungi of Qatar and other Arab countries. The Sientific and Applied Research Centre, University of Qatar, Doha, Qatar, 566 pp.
- Moubasher A.H., Elnaghy M.A., Abdel-Hafez S.I. 1972. Studies of fungus flora of three grains in Egypt. Mycopathologia et Mycolgia Applicata 47 (3): 261–274.
- Moustafa M.K., Ahmed A.A.H., El-Bassiony T.A., Aboul-Khier F. 1984. Microbiological quality of infant milk foods. Assiut Veterinary Medical Journal 12: 163–167.
- Munimbazi C., Bullerman L.B. 1996. Moulds and mycotoxins in foods from Burundi. Journal of Food Protection 59 (8): 869–875.
- Pitt J.I. 1979. The genus *Penicillium* and its teleomorphic states *Eupenicillium* and *Talaromyces*. Academic Press, London, 634 pp.
- Pitt J.I., Hocking A.D. 2009. Fungi and food spoilage. 3rd ed, Springer, 519 pp.
- Pitt J.I., Hocking A.D., Bhudhasamai K., Miscamble B.F., Wheeler K.A., Tanboon-Ek P. 1994. The normal mycoflora of commodities from Thailand. 2. Beans, rice, small grains and other commodities. International Journal of Food Microbiology 23 (1): 35–53.
- Raper K.B., Fennell D.I. 1965. The genus Aspergillus. The Williams and Wilkins Co., Baltimore, pp. 686.
- Sanchis V., Vinas I., Jimenez M., Calvo M.A., Hernandez E. 1982. Mycotoxin-producing fungi isolated from bin-stored corn. Mycopathologia 80 (2): 89–93.
- SAS Institute Inc. 1996. SAS/C Compiler and Library User's Guide, Fourth Edition, Release 6.00, Cary, NC: SAS Institute Inc., 433 pp.
- Sebunya T.K., Yourtee D.M. 1990. Aflatoxigenic Aspergillus in foods and feeds in Uganda. Journal of Food Quality 13 (2): 97–107.
- Suarez G., Guarro J., Calvo M.A. 1981. Toxicological study of fungi isolated from starches intended for human consumption. Mycopathologia 75 (1): 27–31.
- Sutic M., Mitic S., Svilar N. 1979. Aflatoxin in milk and milk products. Mljekarstvo 29 (4): 74-80.

- Sydenham E.W., Theil F.G., Marasas W.F.O., Shepard G.S., van Schalkwyk D.J., Koch K.R. 1990. Natural occurrence of some *Fusarium* mycotoxins in corn from low to high esophageal cancer prevalence areas of the Transkei, Southern Africa. Journal of Agricultural and Food Chemistry 38(10): 1900-1903.
- Taligoola H.K., Ismail M.A., Chebon S.K. 2004. Mycobiota associated with rice grains marketed in Uganda. Journal of Biological Science 4 (1): 271–278.
- Taligoola H.K., Ismail M.A., Chebon S.K. 2010. Toxigenic fungi and aflatoxins associated with marketed rice grains in Uganda. Journal of Basic and Applied Mycology, Egypt 1 (1): 45–52.
- Tuite J.F., Christenseen M. 1955. Grain storage studies. 16: Influence of storage conditions upon the fungus flora of barley seed. Cereal Chemistry 32: 1–11.
- Wheeler K.A., Hocking A.D., Pitt J.L., Anggawati A. 1986. Fungi associated with Indonesian dried fish. Food Microbiology 3 (4): 351–357.
- Zohri A.A., Abdel-Sater M.A., Ismail M.A. 1995. Incidence of aflatoxins and mould flora in corn snacks. Journal of Food Science and Technology, India 32 (4): 289–294.