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Isolation and Identification of Pathogenic Bacteria and Fungi from Some Saudi Bank Note Currency

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Saudi Bank notes were evaluated to detect the level of contamination with pathogenic bacteria and fungi using standard techniques. The genera of bacteria that isolated were Escherichia coli, Citrobacter sp., Klebsiella sp., Proteus sp., Streptococcus sp., Bacillus sp., Corynebacterium sp. and Staphylococcus sp. The fungal species Trichophyton sp., Microsporum sp., Epidermophyton sp., Aspergillus sp., Alternaria sp., Penicillium sp., Candida sp., Phycomyces sp., Saccharomyces sp. and Cladosporium sp. were isolated and identified. Potential risk of Saudi bank notes to serve as environmental vehicles for transmission of pathogenic bacteria and fungi was evaluated. Lower values of bank notes were highly contaminated and there is a negative correlation between currency value and microbial contamination. The results suggested that bank notes may be contaminated with infectious bacteria and enteric microbes and may act as a source of serious infection. Personal hygiene, plastic currency notes and electronic or credit banking to reduce the risk of infection were highly recommended.

Keywords: Bank notes, currency notes, air contamination, bacteria, fungi, Saudi Arabia.

Air-borne human pathogens are considered to be serious environmental hazards. Currency notes are handled and circulated by people with different health background and life styles. The level of contamination and risk of microbial transmission via currency notes are associated with the levels of community hygiene and economic status of the country[20, 15]. Despite the fact that paper bank notes that handled by large numbers of people increase possibility of acting as environmental vehicles for transmission of pathogenic bacteria and fungi[1]. The infected currency is therefore, considered to be potential public health hazards as pathogens spread by circulating bank notes[13]. Immuno-compromised people stand the risk of acquiring opportunistic infection, through handling of contaminated bank notes papers which can occur when immune system is not competent to defend bacteria and fungi that usually harmful and cause diseases[4,8].

In literature there is a few published data that obtained from bank notes currency that cause microbial contamination. For example in Ethiopia, Alemu[2] investigated the prevalence and occurrence of microorganisms in bank notes of the country and concluded that currency notes of lower denominations were the most contaminated because lower notes values pass through more hands of people in their lifetime than the higher ones. In Nigeria Emikpe and Oyero[13] isolated *Entrobacter* sp., *Staphylococcus* sp., *Citrobacter* sp., *Klebsiella* sp. and *Proteus* sp. as pathogenic bacteria resistant to Tetracycline and Cotrimoxazole and also sensitive to Amoxoftine, Gentamicin, Nalidixic acid and Ofloxacin. Similar results were also obtained from Nigeria by Enemuor *et al*[14] who

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identified *Salmonella typhi*, *E. coli*, *Staphylococcus* sp. and *Enterococcus* sp. and four fungal isolates from currency counting machines in selected banks of the country. In Nigeria also, a study revealed that paper bank notes could cause urinary tract infection with diarrhea besides skin burn and septicemia infection[17]. Their research findings focused on the economic importance of currency paper notes as a source of microbial contamination. In Ajmer, Rajasthan (India) different isolates of *E. coli* grown on EMB from samples of currency from different social status were isolated and identified[5]. In India, Elumalai *et al*[12] similarly isolated *E. coli*, *S. aureus*, *Bacillus* sp., *Klebsiella* sp., *Salmonella* sp., *Pseudomonas* sp., *P. mirabilis* and *Bacillus* sp. in paper bank note samples. In Sudan (Africa) serious pathogenic fungi that cause mycoses have been isolated and identified from lower values of bank note currency of the country[21]. In USA, Fonseca *et al*[15] identfied *S. aureus*, *Klebsiella pneumoniae*, *Enterobacter* sp., and *Pseudomonas sp.* with a 72% sample contamination.

In Saudi Arabia, there is a very limited documented data concerning the pathogenic bacteria which transmitted by currency notes[3].The data of this publication concentrated on Riyal value only and not included fungal pathogens. Therefore, the objectives of the present research work are to investigate whether Saudi bank notes currency save as barriers for microbial contamination, highlight some environmental pollution due to currency microbial contamination and suggest some control measures to minimize contamination in Saudi paper bank notes.

MATERIALS AND METHODS

Survey

Paper banknotes from different marketing sources such as meat seller (meat market) or butchers, vegetable markets and fish market were obtained. The values of banknotes were: one, five and ten Saudi Riyals, which were collected randomly in sterile polyethylene bags. They were immediately transferred to the microbiology laboratory of clinical laboratory sciences department of Shaqra University to apply all microbiological examinations for the different values of collected banknotes.

Isolation of microorganisms

Paper bank notes in different values were rinsed and soaked separately in 100 ml normal saline in 250 ml beaker for 24 h[10]. Streaking and swabbing were done using sterile loop and swab[6, 20] onto Nutrient Agar (Himedia laboratories, India) and Mac Conkey Agar(Himedia laboratories, India)[11] to obtain bacterial isolation for 48 h incubation period at 37p C. While, Sabouraud Dextrose Agar (SDA) (Himedia laboratories, India)[6], was used for fungal isolation for 1 week at 28p C.

Identification of bacteria

Cultures in solid media were visually inspected for growth rate and colony characteristics[6, 20]. Different colonies were sub- cultured on Nutrient Agar and MacConkey Agar and incubated similarly as before. Different API tests were carried out for identification of bacteria such as *Staphylococcus* spp.,[18], Enterobacteriaceae[16] and for other Gram negative bacteria[22].

Identification of fungi

Fungal growth on SDA was critically examined after one week, using prepared microscope slides. The prepared slides were mounted on Lacto phenol cotton blue and identification of the fungal species was performed with aid of binocular compound microscope using fungal taxonomy keys[9].

RESULTS AND DISCUSSION

Microbiological techniques which were adopted in this study[7, 20] indicated that various species of bacteria (Figure 1) and fungi have been isolated from different Saudi bank notes. The genera of isolated bacteria are the members of the family Enterobacteriaceae i.e. *Citrobacter frendii, Citrobacter koseri, Escherichia coli, Klebsiella ozaeni, Klebsiella pneumoniae, Klebsiella rhinosclrematis, Proteus vulgaris, Shigella flexneria* and *Shigella dysenteryiae* which were shown in Table 1 and Table 2. The identified Gram positive bacteria were *Staphylococcus aureus*, *Staphylococcus epidermidis, Staphylococcus saprophyticus* and spore- forming bacteria such as *Bacillus subtilis, Bacillus pemulus, Bacillus megaterium, Bacillus firmus, Corynbacterium haemolyticum, C. hafmainaii* and *Lactobacillus*

caserii (Tables 1, 2 and 3). Bacteria which isolated from different bank notes that showed high rate of occurrence were *Staphylococcus aureus* and *E. coli.* While species such as *Bacillus* sp., *Klebsiella* sp., *Citrobacter* sp., *Proteus* sp. and *Shigella* sp. were found in a limited counting numbers especially in bank notes value of five and ten Riyals (Tables 1, 2 and 3).

The most predominant fungal species isolated were *Epidermophyton sp., Saccharomyces sp.*, *Cladosporium sp., Microsporum sp., Candida sp. & Aspergillus sp.* those found abundantly in values of one Saudi Riyals. The genera such as *Penicillium sp.,* and *Aspergillus niger* were isolated from a very limited numbers of samples. The presence of some species such as *Microsporum sp*, was somewhat negligible and in limited manner of occurrence with percentage of less than 25%. (Figure 2). Moreover, some species of the genus *Aspergillus sp.* were found very low as compared to the other isolated genera of fungi (Table 4).

From the present investigation, it was clearly noticed that there was a negative correlation between value of bank notes decrease and increase in pathogenic microorganisms that were found[13], regardless of the species found. Areas within Shaqra province are more contaminated with pathogenic microorganisms i.e. bacteria and fungi

Table 1. Total counts and preliminary microbiological tests of bacteria isolated from some Saudi currency notes collected from different locations

Location	Sample (currency value)	Bacterial isolate		
		Total viable count	Lactose fermenter	Non-Lactose fermenter
Vegetable market	1	2	2	1
	5	4	1	1
	10	2	1	1
Meat market	1	21	1	1
	5	3	2	1
	10	4	2	1
Restaurant	1	9	1	1
	5	10	1	1
	10	4	1	3
Fish market	1	9	1	4
	5	3	1	1
	10	4	1	1

Table 2. Gram positive pathogenic bacteria isolated from some Saudi currency notes with different values

Location	Sample (currency value)	Bacteria
Vegetable market	1	*Staphylococcus aureus*
	5	*Bacillus* sp.
	10	*Staphylococcus aureus*
Meat market	1	*Staphylococcus aureusCorynebacterium spStreptococcus* sp.
	5	*Staphylococcus aureus*
	10	*Staphylococcus aureus*
Restaurant	1	*Bacillus* sp.
	5	*Bacillus* sp.*Corynebacterium sp*
	10	*Bacillus* sp.
Fish market	1	*Bacillus* sp.
	5	*Bacillus* sp.
	10	*Streptococcus* sp.

because it is near Riyadh area a capital city of Saudi Arabia and involve a large human population, which means more handling, more frequent exchange of paper bank notes currency leading to more microbial contamination. There were also a large number of pathogenic bacteria and fungi that isolated from samples obtained from meat markets. The bacteria isolated belong to Enterobacteriaceae family and this family is more frequency found in the air (air-borne) and was found also in large quantity in faces[7].This reflects that faucal pollution appears as a result of poor hygienic attitude in the community of the market. Moreover, the species belong to Enterobacteriaceae family are usually dangerous such as *Salmonella typhi* and *Shigella dysenteryiae* that are pathogenic to human and animals. Furthermore, it was observed that high values of currency are less contaminated than a low values currency, because a low values are more wide spread and exchangeable between people in population[13, 20]. Our results are closely related and similar to those obtained in Saudi Arabia Riyadh area[3] and in Nigeria who identified *Enterobacter* sp., *Staphylococcus* sp., *Citrobacter* sp., *Klebsiella* sp. *and Proteus* sp. from different samples of Nigerian bank notes currency[14]. Similarly from this investigation and others[19], we have reached the point that Saudi paper bank notes currency gives a positive appearance of some species of pathogenic bacteria and fungi due to the absence of antimicrobial agent in row material used for manufacture[6]. We also detected bacterial contamination in significant number of samples from vegetable markets that also reflected lack of hygienic attitude within labors. Therefore, a recommendation should be pointed out such as awareness of people is how to handle money

Table 3. Gram negative pathogenic bacteria isolated from some Saudi currency notes with different values.

Location	Sample (currency value)	Bacteria
Vegetable market	1	*Escherichia coli*
	5	*Citrobacter*sp
	10	*Klebsiellasp*
Meat market	1	*Proteus sp*
	5	*Escherichia coli*
	10	*Escherichia coli*
Restaurant	1	*Proteus sp*
	5	*Klebsiellasp*
	10	*Proteus sp*
Fish market	1	*Proteus sp*
	5	*Klebsiellasp*
	10	*Klebsiellasp*,

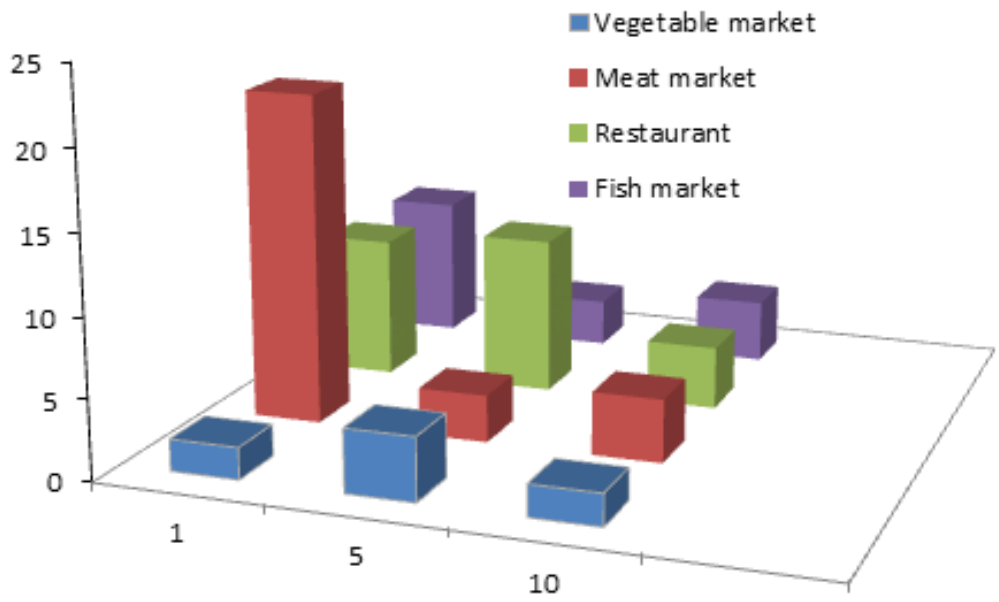

Fig. 1. Total viable counts of bacteria isolated from currency notes collected from different locations

emphasizing to reduce their hand contamination, so reducing currency notes pollution. Emphasis is also to be stressed in the handling of bank notes by children so as to keep them safe from infectious pathogens because they may enter all type of paper bank notes on their mouth. It is also recommended to promote and develop bank notes manufacture by adding antimicrobial agents as a row material

Table 4. Identification of pathogenic fungi isolated from some Saudi currency notes with different values collected from different sources

Surveyed area/ currency value	1(Riyal)	5(Riyal)	10(Riyal)
Vegetable market	*Aspergillus flavus*++ *A. fumigatus* + *A. niger*+++ Penicillium++ Candida ++ Mucor + Cladosporium ++ Trichophyton + Microsporum +	*A. niger*++ *Aspergillus terreus* + *Penicillium spp*++	-*A.niger* +++ -*Penicillium spp* -*Phycomyces sp.*
Meat market	*A. niger* + *A. fumigatus*++ Penicillium spp + Cladosporium spp. +	*A. niger* +++ *Penicillium notatum* + Penicillium spp +	*Penicillium spp* ++ *Microsporum* + A. niger +++ Aspergillus flavus + *A. fumigatus*+++ *Penicillium notatum* ++ Epidermophyton ++ Penicillium roqueforti
Restaurant	*Penicillium notatum* + Cladosporium spp ++ *A. fumigatus*++	*A. fumigatus*++ A. niger + *Aspergillus flavus*+ Candida albicans +	*Cladosporium* ++ A. niger ++

* += low occurrence, ++=medium occurrence ,+++= high occurrence

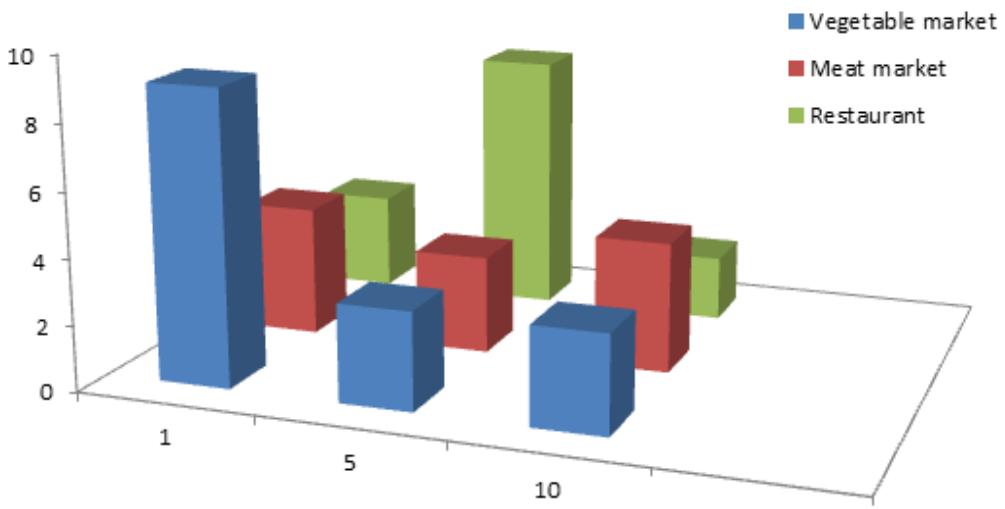

Fig. 2. Total viable counts of fungi isolated from currency notes collected from different locations

during processing. Re -sterilization of paper bank notes currency when it is dirty and returns it back to reduce the risk of infection especially for children. Plastic bank notes currency is strongly recommended, along with coin currency which is sometimes needed.

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