Antimicrobial Activities of Some Herbs Extracts on Food Borne Bacteria.

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Abstract: Food poisoning illness outbreaks caused by some pathogenic bacteria and / or their toxins is still a concern for both consumers and the food industry.The increasing antibiotic resistance of some pathogens that are associated with food borne illness is another concern. There is growing interest in using natural antibacterial compounds, such as extracts of spices and herbs for food preservation. The present investigation was planned out to throw a light on the presence of food poisoning bacteria in different food samples and study antibacterial activity of ethanolic extracts of some medicinal plant. ***Salmonella*** was detected in chicken thighs meat, chicken wings and chicken livers with 66.7%, 37.5%, and, 40% respectively while were not isolated from chicken frankfurters, beef sausages or peanut butter. ***Staphylococcus aureus*** was isolated from chicken thighs, chicken wings. Chicken livers, chicken frankfurters and fresh beef sausage with 25%, 37.5%, 13.3%, and, 25% respectively. ***Bacillus cereus*** found in chicken livers, chicken frankfurters, fresh beef sausages, frozen beef sausage and peanut butter with 20%, 75%, 83.3%, 60%,and, 42% respectively. ***Pseudomonas*** was isolated from chicken thighs, and chicken wings with 25% and 37.5%. ***Klebsiella*** was isolated from chicken livers, and chicken wings with 40% and 37.5%. ***Citrobacter*** was isolated from chicken livers, and chicken thighs with 20% and 50%. ***Enterobacter*** was isolated from chicken thighs, chicken wings, with 8.4% and 25% respectively. Seven spice (Cloves, Cinnamon, Black cumin seeds, Cumin, Black and White pepper and Ginger) were shown to have an inhibitory effect against ***Salmonella, S. aureus*** and ***B. cereus*** which are an important pathogen in food poisoning. Cloves, Black cumin seeds extracts were found to be the most effective plant against almost tested microorganisms. Fennel, Garlic, Cardamom and Red chili pepper ware found to be ineffective against the isolated bacteria.
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1. Introduction:

Food borne illness resulting from consumption of contaminated food with pathogenic bacteria and for their toxins has been of vital concern to public health. More than 250 different food borne diseases (FBD) have been described, and the bacteria are the causative agents of two thirds of food borne disease outbreaks (Olsen *et al.,* 2000).

The most common bacteria causing food borne illness are ***Escherichia coli, Staphylococcus aureus, Salmonella spp., Listeris monocytogenes, clostridium botulinus, vibrio parahaemolyticus*** and others (Van *et al.,* 2007). Food borne illness can cause symptoms that ranged from on upset stomach to more serious symptoms such as diarrhea, fever, Vomiting, abdominal cramps and dehydration despending on the etiological agents.

Food borne illnesses not only affects the health of individuals, but it can also have dramatic economic impact. The economic losses from various factors, such as medical treatment, lost wages and productivity, loss of business, recall and destruction of products, and investigation of the outbreaks, can be very high. (Doores, 1999).

Food poisoning is still a concern for both consumers and the food industry despite the use of various preservation methods. Food processors, food safety researchers and regulatory agencies are continuously concerned with the high and growing number of illness outbreaks caused by some pathogenic and spoilage microorganisms in foods.The increasing antibiotic resistance of some pathogens that are associated with food borne illness is an other concern (Stermitz *et al.,* 2000). Uncontrolled use of chemical antimicrobial preservatives has been inducing factor for appearance of genetically modified microbial strains more resistant to classic antimicrobial agents.

Difficult to control the modified microbial population showed multi- resistant, has been reported all over the world (Levy, 1997).

Food borne strains of resistant pathogens to avariety of antimicrobials have become a major health concern (Kiessling *et al.,* 2002) and it could decrease the successful application of control measures on spoilage and pathogen microorganisms, many times leading for use of less safe, ineffective or expensive alternatives (Levy, 1997).

Antimicrobial resistance is an increasingly global problem and emerging has become a public health issue worldwide (kaye *et al.,* 2004). A variety of foods and environmental sources harbor bacteria that are resistant to one or more antimicrobial drugs used in human or veterinary medicine and in food – animal production (Schroeder *et al.,* 2004).

Recently, there has been increasing interest in discovering new natural antimicrobials. Plant products with antimicrobial properties notably have obtained emphasis for a possible application in food production in order to prevent bacterial and fungal growth (Lanciotti *et al.,* 2004).

Plant products are characterized for a wide range of volatile compounds, some of which are important flavor quality factors (Utama *et al.,* 2002). Moreover, plant volatiles not only considered safe but also not leading to antimicrobial resistance (Newberne *et al.,* 2000).

Consumers are also concerned about the safety of food containing synthetic preservatives. Therefore, there has been increasing interest in the development of new types of effective and nontoxic antimicrobial compounds. There is growing interest in using natural antibacterial compounds, such as extracts of spices and herbs, for food preservation (Smid and Corris, 1999).

Spices and herbs have been added to foods since ancient times, not only as flavoring agents, but also as folk medicine and food preservatives (Cutler, 1995).

In addition to importing characteristic flavors, certain spices and herbs prolong the storage life of foods by preventing rancidity through their antioxidant activity or through bacteriostatic or bactericidal activity (Beuchat and Golden., 1989).

The presence of ***S.typhi*** and ***paratyphi*** in meat products indicates human origin and therefore poor personal hygiene during handling of the meat products (Mrema *et al.,* 2006). ***Stapheloccus aureus*** contamination in meat and meat products was 10 %. This contamination rate is lower than that observed in the survey previously conducted in Italy that revealed a prevalence of **coagulase positive *staphylococci*** rates in meat products ranging from 17.1% to 48% (Normanno *et al.,* 2007. Saikis and Joshi., 2010) reported that out of 110 samples of raw chicken meat, collected from local meat markets of North east India, were contaminated with ***Enterobacter aerogenes*** (100%), ***Escherichia coli*** (98%), ***Klebsiella pneumonia*** (98%), ***Proteus spp*** (44%), ***Staphylococcus spp*** (20%), ***yersinea enterocolitica*** (23%), ***Salmonella typhi*** (20%).

Microbiological analysis of three opened Jars of peanut butter from case households and three unopened jars from ratail outlets revealed the presence of ***salmonella mbandaka*** at populations as low as three cells per gram. ***Salmonella Senftenberg*** was also isolated during subsequent testing of the peanut butter (killalea *et al.,* 1996).***Salmonella*** can survive in peanut butter for at least 24 weeks at 5 or 21 C (Burnett *et al.,* 2000). A second major ***Salmonella*** outbreak in the U.S attributed to peanut butter and peanut butter – containing products (FDA, 2009).

Raw meat products (in particular poultry) have frequently been associated with the presence of ***Salmonella*** (Bryan and Doyle., 1995). ***Salmonella*** positive animal at the time of slaughter have high numbers of organisms in their intestines as well as on external surfaces (faecal, contamination of hides, fleece, skin, or feathers). Cross contamination during processing may also lead to increase prevalence of ***Salmonella*** in finished products.

Staphylococcal food poisoning is caused by the consumption of food containing enter otoxins produced by certain strains of ***S.aureus***. Despite the wide - Spread association of ***S.oureus*** with animals, humans are the main reservoir for ***S.aureus*** involved in human disease (Jablonski and Bohach, 1997). Hand contact with ready – to – eat foods is an important means by witch ***S.aureus*** may enter food supply by food handlers. Foods that present the greatest risk of causing illness are those in which the normal flora has been destroyed (e.g. cooked meats) or inhibited (e.g. cured meat containing high salt content) (Stewart, 2003).

The ethanolic extracts of medicinal plants and spices for inhibition of bacteria and fungi in culture media at different PH. Clove exhibited remarkable antibacterial activity against all organisms tested. Oregano and cinnamon extract showed a wide inhibitory spectrum against Gram- positive bacteria while they were less potent against gram – negative bacteria. Black pepper has bee shown to have antimicrobial activity (Dorman and Deans, 2000). Both aqueous and ethanol extract of black pepper screened for antibacterial activity against a penicillin G resistant strain of ***Staphylococcus aureus***, showed antibacterial activity, which was determined by the agar – well diffusion methods.

2. Material and methods:

A total of 93 samples of different food products including 19 peanut butter, 12 fresh beef sausages, 15 frozen beef sausages and 74 meat chicken sample (12 frankfuters, 12 thighs, 15 livers, and 8 wings) were collected from markets in Mansoura city.

Each sample was obtained aseptically and transported in iceboxes packed with ice to the microbiological lab within 2 hours. 25 grams of each sample were taken aseptically and placed in a sterile

blender Jar containing 225 ml of sterile buffered peptone water (PW). A loopfull of 24 hours buffered peptone water media was streaked onto blood agar.Bacterial isolates were indentified according to the colonial characteristic appearance, haemolytic patterns, Microscopically by Gram`s stain and biochemically according to Bergey`s Manual of Determinative Bacteriology (Roberts and Green wood, 2003).

Eleven types of medicinal plants including Cardamom (*Elettaria carfomoun*), Cinnamon (*Cinnamomum zeylamicm*), Clove (*Syzygium aromaticum*), Cumin (*Cuminum syminum*), Fennel (*Foeniculum vulgare*), Garlic (*Alium sativum*), Ginger (*Zingiber officinalis*), Pepper (block and white) *Pipre nigrum*, Red chilli (*Capsicum frutescens*) and Black Cumin (*Nigella sativa*) were purchased from local retail markets in Mansoura city.

All the spices were first cleaned using tap water and using sterile distilled water.

They were dried in laminar flow biological safety cabinet. Concerning garlic, it was skimed manually then placed in a hot air oven for drying, at the temperature of 65^{o}C for 72 hours (Ekwenye and Eleglam., 2005). The dried plants were crushed immediately before assay using an electric grinder.

Organic extracts were prepared by soaking 50 gm of the dried powder separately in 200ml of analytical organic solvents (Ethanol 70%), using a conical flask plugged. The mixture was kept at 20^{o}C over night under continuous shaking at 130 rpm, and were filtered through Whatman filter paper (No.2). The filtrates were evaporated using vacuum rotary evaporator. Stock solutions of crude ethanolic extracts were prepared by diluting the dried extracts with 10% Dimethyl Sulphoxide (DMSO) solution to obtain a final concentration of 10 mg / ml (Hoque *et al.,* 2008).

Three – five colonies from the pure culture were suspended in 5-10 of sterile nutrient broth. The turbidity of the test suspension was compared with 0.5 Mcfarland turbidity standard (10^{8} CFU / ml) (Sofia *et al.,* 2007).

An agar well diffusion method was employed for determination of antibacterial activities. 0.1ml of bacterial suspension was spread onto the surface of nutrient agar medium. Wells of 8 mm diameter were cut from the agar with sterile cork borer. The base of each well was sealed with 50 ul of sterilized molten nutrient agar. The wells were filled by adding 300 ul of the different plant extracts, while DMSO was used as control. The plates were incubated for 24 h at 37^{o} C. After incubation, the inhibition zones around each cup were measured with a caliper and recorded.

3. Results:

93 different food samples including 47 chicken sample (12chicken thighs, 15 chicken livers, 8 chicken wings, 12 chicken frankfurter), 27 beef samples (12 fresh beef sausage, 15 frozen beef sausage), and 19 peanut butter samples.

A number of 96 bacterial isolates have been isolated from examined samples. These isolates were identified according their morphological features and Gram stain into three divisions which proved to be:

- Fourty four bacterial isolates are Gram negative rods:
- Six isolates that show positive oxidase test, positive catalase test,negative lactose fermentation, positive pigment production, positive motility test, B haemolysis, growth on SS ager medium show they are ***Pseudomonas aeruginosa***.
- Thirty eight isolates negative oxidase test:

*Twenty one isolates that show positive result of lactose fermentation test:

*Twelve isolates positive motellity test (9 isolates positive Methyl red test and Negative sucrose test that show they are ***Citrobacter*** and 3 isolates Negative Methyl red test, Positive sucrose test that show they are ***Entrobacter,*** and:

9 isolates Negative motellity test they are ***Klebsiella***.

*Seventeen isolates that show Negative result of lactose fermentation test, growth on SS agar medium showed colorless colonies, and, positive Motility test, they are ***Salmonellae.***

- Thirteen isolates Gram positive cocci:

Concerning the morphological feature they are forming irregular clusters and tetrads (grape like appearance), strong catalase positive test, ferment mannital sugar, B- haemolysis, coagulase positive test, Negative oxidase test, and, positive glucose fermentation. from the previous tests they are ***Staphylococcus aureus***

- 39 isolates Gram Positive spore forming bacilli:

Colonies of isolated appear turquoise peacock blue on PEMBA agar, B-haemolysis, negative mannitol test, negative xylose fermentation test, positive motility test, positive glucose fermentation, they are ***Bacillus cereus***.

Incidence of Bacterial isolates from different food samples:

The obtained results showed 17 samples (18.3%) were positive to bacteriological examination for ***Salmonella*** isolate distributed as; 8 samples (66.7%) of chicken thighs, 6 samples (40%) of chicken livers, 3 samples (37.5%) of chicken wings. On the other hand, ***Salmonella*** were not isolated from chicken frankfurters, beef sausages or peanut butter. 13 samples (14%) ***Staphylococcus aureus***

isolates distributed as; 3 samples (37.5%) of chicken wings, 3 samples (25%) of chicken thighs, 3 samples (25%) of chicken frankfurters, 2 samples (16.7%) of beef sausage, and, 2 samples (13.3%) of chicken livers. ***S.aureus*** was not isolated from frozen beef sausages or peanut butter. ***Bacillus*** cereus was isolated from 10 samples (83.3%) of fresh beef sausage, 9 samples (75.0%) of chicken frankfurter, 9 samples (60.0%) of frozen sausage, 8 (42.0%) of peanut butter, and, 3 (20.0%) of chicken livers, while, ***B. cereus*** was not isolated from checking thighs or wings. ***Pseudomonas*** was isolated from 3 isolates (25%) from chicken thighs and 3 isolates (37.5%) from chicken wings, while not isolated from chicken liver, chicken frankfurter, fresh beef sausage, frozen beef sausage, or, peanut butter.

Klebsiella was isolated from 6 samples (40. 0 %) of chicken livers, and 3 samples (37.5%) of chicken wings. ***Citrobacter*** was isolated from 6 samples (50.0%) of chicken thighs and 3 samples (20.0%) of chicken livers. ***Enterobacter*** was isolated from one samples (8.3%) of chicken thighs and 2 sample (25.0%) of chicken wings (Table 2).

Screening of antibacterial activities of medicinal plant extracts:

The results of the well diffusion test (Table 3, Fig 2) indicated that ethanolic extracts of medicinal plants 10 mg/ml concentration as fellow: Clove extract showed antibacterial activities against ***Salmonella*** with inhibition zone (22 mm), ***B.cereus***, (26mm),***Klebsiella*** (21mm), ***Citrobacter*** (22mm) and ***Enterobacter*** (17mm). Ginger inhibited the growth of ***Salmonella*** (26mm), and, ***Staphylococcus aureus*** (30mm). Cinnamon showed inhibition growth of ***Salmonella*** (15mm), and ***Pseudomonad aeruginosa*** (30mm). Black and white pepper extracts were active against ***B.cereus*** giving inhibition zone (16.5 and 25.5 mm respectively). Black cumin seed extracts inhibited the growth of ***B. cereus*** (39mm), ***Citrobacter*** (35m), and, ***Enterobacter*** (37.5m), but, Cumin extracts inhibited the growth of ***Staphylococcus aures*** (25mm) and ***Salmonella*** (27mm).

Ethanolic extracts of Cardamom, Garlic, Fennel and, Red chili were inactive against all bacterial strains tested (Table 3, Fig.1).

Table (1): Bactria Isolated from tested samples

Bacterial isolates	NO. of test samples
Pseudomonad aeruginosa	6
Citrobacter	9
Entrobacter	3
Klebsiella	9
Salmonellae	17
Staphylococcus aureus	13
Bacillus cereus	39
Total	**93**

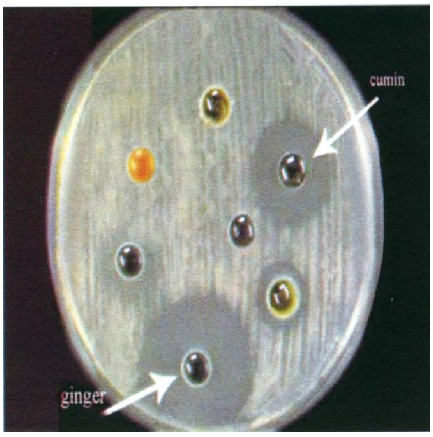

Photo (1): **Sensitivity test of *S.aureus* to medicinal plant extracts**
(Well Diffusion Method)

Table (2): Prevalence of food borne bacteria isolated from different food samples

microorganism and Site of isolation	Salmonellae		*S. aureus*		*B. cereus*		pseudomonas		Klebsiella		citrobacter		Enterobacter	
	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
Chicken thighs (12)	8	66.7	3	25.0	0	0	3	25.0	0	0	6	50.0	1	8.3
Chicken Livers (15)	6	40.0	2	13.3	3	20.0	0	0	6	40.0	3	20.0	0	0
Chicken wings (8)	3	37.5	3	37.5	0	0	3	37.5	3	37.5	0	0	2	25.0
Chicken frankfurter(12)	0	0	3	25.0	9	75.0	0	0	0	0	0	0	0	0
Fresh beef sausage(12)	0	0	2	16.7	10	83.3	0	0	0	0	0	0	0	0
Frozen beef sausage(15)	0	0	0	0	9	60.0	0	0	0	0	0	0	0	0
Peanut butter (19)	0	0	0	0	8	42.0	0	0	0	0	0	0	0	0
Total (93)	17	18.3	13	14.0	39	41.9	6	6.5	9	9.7	9	9.7	3	3.2

% calculated according to the number of examined samples
(): number of examined samples

Table (3):Antibacterial activity of ethanolic extracts of plants against some foodborne bacterial strains using well diffusion test expressed by its Diameter of inhibition zone(DIZ)/mm:

microorganism / Plant used	Salmonella (DIZ) mm	*S. aureus* (DIZ) mm	*B. cereus* (DIZ) mm	*P. aureogonosa* (DIZ) mm	Klebsiella (DIZ) mm	Citrobacter (DIZ) mm	Enterobacter (DIZ) mm
Ginger	26	30	–	–	–	–	–
Clove	22	–	26	–	21	22	17
Cinnamon	15	–	–	30	–	–	–
Cardamom	–	–	–	–	–	–	–
Garlic	–	–	–	–	–	–	–
Cumin	27	25	–	–	–	–	–
Blck pepper	–	–	16.5	–	–	–	–
White pepper	–	–	25.5	–	–	–	–
Fennel	–	–	–	–	–	–	–
Black cumin seed	–	–	39	–	–	35	37.5
Red chili pepper	–	–	–	–	–	–	–
10%DMSO	–	–	–	–	–	–	–

Values calculated as means of duplicates
DMSO: Dimethyl Sulfoxide
Concenteation of all of the plant extreacts were 10mg/ml.
(-) no inhibition zone

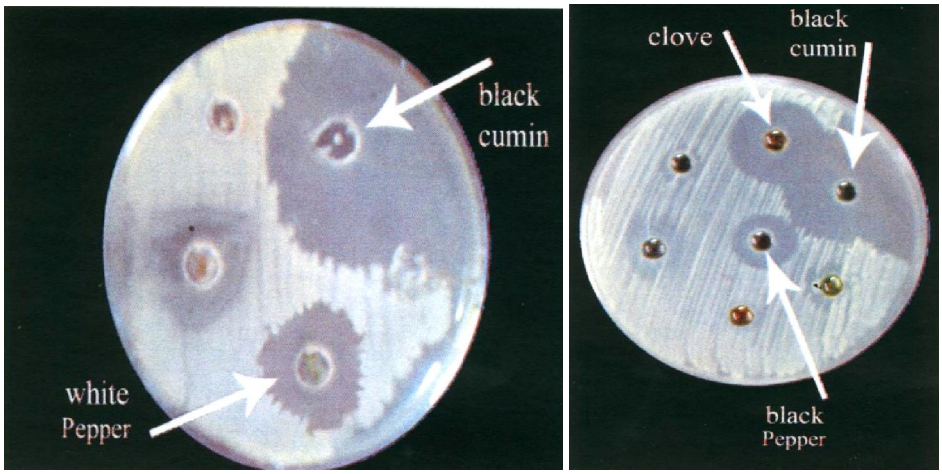

Photo (2): **Sensitivity test of *Bacillus cereus* to medicinal plant extracts *(Well Diffusion Method)***

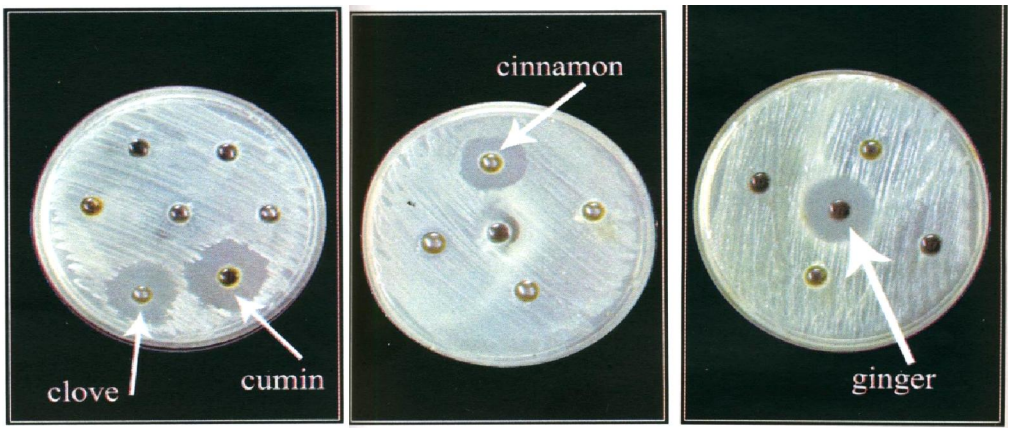

Photo (3): **Sensitivity test of *Salmonella* To medicinal plant extractas (Well Diffusion Method)**

4. Discussion

Food born infections are in important public health concern worldwide. According to reports of WHO (2003), every year a large number of people are affected by diseases due to contaminated food consumption. Wide spectrums of pathogens play a role in food borne disease. Foods of animal origin are considered major vehicles of food borne infections (Todd, 1997).

According to the present study, ***Salmonella*** was detected in 66.7% of chicken thighs meat, 40% of chicken livers, and 37.5% chicken wings. However, it could not be detected in chicken frankfurter, fresh and frozen beef sausage, and peanut butter samples

Concerning chicken thighs meat, the obtained results agree with those reported by Jarngklincham *et al.* (1994) who isolated ***Salmonella*** in a rate of 66% in chicken meat. Nearly similar results of ***Salmonella*** in chicken wings were reported by Capita *et al.* (2003) whose result was 40% and a similar result of chicken liver samples was 40%.

Regarding chicken frankfurter samples as showed in table (2), similar results was recorded by Alexandre de Freitas luiz *et al.* (2004). Also, Sharma *et al.* (1995) failed to detect ***Salmonella*** in chicken sausage. Capita *et al.* (2003) who isolated ***Salmonella*** in rate of 40% and 60% in red and white sawsage samples respectively.

Staphyloccus microorganisms are also widely distributed wherever people handle food. Contamination is so easy as the ***Staphylococci*** are always present in human skin and in respiratory tract. It is evident from table (2) that the incidence of isolated ***S. aureus*** organism from chicken thighs, livers, wings, frankfurter and fresh sausage samples were 25%, 13.3, 37.5%, 25% and 16.7% respectively. Concerning chicken samples, the results were disagreed with that reported by Capita *et al.* (2001) who detected ***S. aureus*** in chicken legs, giblets, wings and sausages with percentage 40%, 60%, 60%, and 100% respectively. Altay et al,(2003) isolated ***S. aureus*** from chicken giblets in fate of 23.3%, this result is higher than obtained result (13.3%). Nearly similar results of ***S. aureus*** in beef sausage were reported by Abdel-Aziz *et al.* (1996) whose result was 15%. According to kuku (1985), the presence of ***S. aureus*** could be as a result of it being a common organism on the skin, hands and boil and hence their presence in sausage may be as a result of contamination due to handling, processing, transportation and storage. Its presence in high numbers is a good indication of poor hygiene and temperature control. No ***S. aureus*** could be detected in frozen sausage samples table (2).

According to the data in table (2), ***Bacillus cereus*** was detected in 20% of chicken livers, 25% of chicken frankfurters, 83.3% of fresh sausages, 60% of frozen sausage.Many studies have been conducted to determine the prevalence of ***B.cereus*** in sausage. Similar results were recorded by Saleh *et al.* (1993). On the other hand low prevalence was reported by El-sayed *et al.* (1999) whose percentages was 28%, Also, Ouf (2004) detected ***B.cereus*** in a percentage 30%. High prevalence (80%) was reported by El-Ghamry (2004).

No ***B.cereus*** could be detected in the examined chicken thighs and wings samples. These results were in acceptance to findings reported by Saika and Hoshi (2010) could isolated ***B.cereus*** from chicken meat in percentage of 10%.

From the obtained results in table (2) ***Pseudomonas, Klebsiella, Citrobacter and Enterobacter*** could be isolated from chicken meat and livers. This result agrees with kilonze-Nthenge *et al.* (2008) who isolated ***Klebsiella, Pseudomonas***, and ***Citrobacter*** from chicken carcasses and livers. Cardose *et al.* (2006) verified the following frequency of ***Enterobacteria*** in broiler carcasses: ***Enterobacter SPP***. 15.7%; ***Citrobacter SPP.*** 2%; ***Klebsiella SPP***.11.8% from examined broiler carcasses.

No ***Pseudomonas, Klebsiella, Citrobacter or Enterobacter*** could be isolated from chicken frankfurter, beef sausage samples. This result was in acceptance to the findings reported by Olueafemi and Simisaye, (2006) could isolate them from sausage samples.

In the present study, ***Salmonella*** could not be isolated from peanut butter samples, this is may be attributed to the fact that peanut butter treated with high temperature during processing, and it has been generally assumed that this step would in killing ***Salmonella*** that has contaminated the raw product. However, there have been several outbreaks associated with peanut butter, with the earliest outbreak occurring in 1996. This outbreak was due to ***Salmonella*** from peanut butter products in Australia (Burnett *et al.*, 2000). In 2006, the U.S. experienced its first peanut butter outbreak when Pulse Net discovered and increasing trend in ***Salmonella*** isolates.Opened and unopened Jars of peanut butter tested positive for ***Salmonella*** (CDC,2007). The reason lies in the neglect of the protective practice which must be adapted after time of processing i.e. storage, marketing and during home preservation.

There is no available data about prevalence of ***Staphylococcus aureus*** in peanut butter. ***S.aureus*** could not be isolated from peanut butter. According to the data in table (2) ***Bacillus cereus*** was isolated from 42.0% of peanut butter samples. ***B. cereus*** is widely distributed in the air, water, soil, and faces Drobniewski (1993). The presence of ***B. cereus*** in

peanut butter may be attributed to ***Bacillus*** spores can survive high heat treatment used in many cooking procedures (Acheson, 1999).

The antimicrobial quality of medical plant and their extracts have been recognized since antiquity while attempt to characterize these properties in the laboratories data back to the early 1900(Martindale, 1910).In this study antimicrobial properties of some plants have been assessed.

Investigation on the crude ethanol extracts of Clove, Black Cumin seed, Black and White Pepper, Ginger, Cumin and Cinnamon showed different degrees of growth inhibition, by using the well diffusion test (table 3) and figure (1). Clove extract proved to be the most inhibitor against almost tested bacteria, followed by Black seed then Ginger, Cumin and Cinnamon extracts.

Clove was detected to exhibit an inhibitory effect against ***Salmonella, B. cereus, Klebsiella SPP., Citrobacter*** and ***Enterobacter***. The antimicrobial effect of Cloves reported by Azzouz and Bullerman (1982). Similar result of Clove was reported by Nanasombat and Lohasupthawee(2005) who found that the ethanolic extract of Clove showed the highest antibacterial activity against ***S.typhimurium*** (15mm), and ***S.morcescens*** (22mm). Tayel and El-tras (2009) reported that ethanolic extract of Clove inhibited the growth of ***Bacollus subtilis, P.aerugmosa and S. aureus***.

Cinnamon extract exhibited notable antibacterial activities toward ***S. paratyphi*** and ***P. aeruginosa*** table (3). Tayel and El- tras (2009) reported that Cinnamon extract proved to be the most inhibitor against all of tested bacteria. While, Hoque *et al.* (2008) found the ethanol extract of Cinnamon, was active only against ***S. aureus*** and ***Vibrio parahoemloyticus.***

Ginger was shown to have an inhibitory effect against ***S. aureus*** and ***S. kentvcky***, However, a similar effect against other microorganisms was not found. These results compare with the findings of Tayel and El-tras (2009) who reported the antimicrobial activity of Ginger against ***S. aureus*** and ***salmonella SPP***. On the other hand Onyeagba *et al.* (2004) demonstrated that the ethanolic extracts of Ginger singly did not inhibit ***S. aureus*** and ***Salmonella SPP***.

Al Zoreky and Nakahara (2003) showed that ***B. cereus*** was the most sensitive microorganism to extracts from Cinnamon and Ginger. The antimicrobial activity of Ginger may be attributed to the fact that it contains antimicrobial substances (Michael derrida, 1999).

The result of the present study is similar to those of Tayel and E-tras (2009) who mentioned that Black Sumin was active against gram positive bacteria and gram negative. Singh *et al.* (2005) found that Black Cumin extract possess antibacterial activity against ***B.cereus.***

Black Sumin seed extract was active against ***B.cereus, Citrobacter*** and ***Enterobacter***, where ***B.cereuse*** is the most susceptible strain to Black Cumin. On the other hand, Shan *et al.* (2007) found that Cumin had no active against ***S.aureus*** and ***Salmonella.***

Piper Nigrum showed an inhibitory effect only against ***B. cereus***. Hema *et al.* (2009) who mentioned that Piper Nigrum showed the highest antimicrobial zone against ***B. cereus***. Also, Singh *et al.* (2005) found that Black Pepper extracts showed complete reduction of colonies against ***B. cereus***.

No effects of Garlic were observed to all bacterial tested in this study. Similar results of Garlic were reported by Nanasomdat and Lohasupthawee(2005). Fennel showed no antimicrobial effect against test strains. These findings are not accordance with findings of other investigation (Mandeel *et al.*.,2003). Tayel and El-tras (2009) who found that Fennel had inhibitory effect against bacterial growth. Agaoglu *et al.* (2007) demonstrated that Fennel had a weakest antibacterial effect, where Fennel showed an inhibitory effect only against ***S.aureus.***

Cardamom and Red Chili (pepper) showed no antimicrobial effect against test strains. Similar result was reported by Stonsaovapak *et al.* (2000) who mentioned that Cardamom extract had no inhibitory effect against tested bacteria. In a study on antimicrobial activity of cardamom, Agaoglu *et al.* (2005) demonstrated that Cardamom had inhibitory effect against ***S.aureus.***

Seven spice (Cloves, Cinnamon, Black Cumin seeds, Cumin, black and white Pepper and Ginger) were shown to have an inhibitory effect against ***salmonella***, ***Staphylococcus aureus, B.cereus*** which are an important pathogen in food poisoning. The most effect activity against ***B.cereus*** was exhibited by Black Cumin seed and Cloves; White Pepper showed less activity; and the effects of Black Pepper was the lowest. The most effective spice against ***S.aureus*** and ***salmonella*** was Ginger. The most effective spice against ***Klebsiella*** was Clove, while the most potent spice against ***Citrobacter*** and ***Enterobacter*** was Black Cumin. The other test strains, ***P.aeruginosa*** and ***salmonella*** were effect only by Cinnamon.

Increased knowledge of concern over leading to raise pointer to the effect of medical plant, herbs and their extracts in improving keeping quality of food as a preservative and their bacteriostatic and bactericidal against food borne microorganisms, they also contain two principle aromatic and flavoring components of

herbs and spice, if added to food stuff, would cause no less of organoleptic properties, will retard microbial contamination and there for reduce onset of spoilage. To fulfill this only a small quantities would be required for this effect.The results of the present study are quite encouraging as some spices exhibited antimicrobial activity against most of the pathogens, but the antimicrobial activity varies widely, depending on the type of spices and microorganism. This study opens up the possibility for the search of new antimicrobials as an alternative to the antibiotics. It is hope that this study positively participate in solving the problem of food contamination.

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