Study of the inhibitory effects of some medicinal plants extracts on growth of *Listeria monocytogenes* isolated from human, cow and sheep in Al-Qadisiyah governorate

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Summary

This study aimed to evaluate the inhibitory effects of ethanolic and chloroformic extracts of local medicinal plants as Oak *(Quercus acuta*), Thyme *(Thymus vulgaris*) and Cinnamon *(Cinnamomum zeylanicum*) prepared in different concentrations (50, 100, 200 and 400 mg/ml) against the growth of *Listeria monocytogenes* isolated from infected humans and animals and comparing their activity with effectiveness of the standard antibiotics. The results showed detection of *Listeria monocytogenes* in 3 of (50) blood samples collected from aborted woman making 6% samples, while the detection rates of *Listeria-monocytogenes* in milk samples collected from sheep and cattle were 4% (4/100) and 9.16% (11/120) respectively. The ethanolic and chloroformic extracts of Thyme as well as, chloroformic extracts of Oak and Cinnamon at concentrations (50 and 100 mg/ml) showed significant antibacterial activity against the growth of *Listeria monocytogenes* isolates from humans, while the ethanolic extracts of Oak and Cinnamon did not show any antibacterial activity against the growth of same bacterial isolates. The ethanolic and chloroformic extracts of Thyme, as well as chloroformic extracts of Cinnamon at concentrations (50 and 100 mg/ml) showed antibacterial activity against growth of *Listeria monocytogenes* isolates of cattle, while ethanolic and chloroformic extracts of Oak and ethanolic extracts of Cinnamon did not show any activity against growth of the same isolates. The results showed that all extracts have antibacterial activity against growth of *Listeria monocytogenes* isolates of sheep, except ethanolic extracts of Cinnamon at all tested concentration; as well, the chloroformic extracts of Thyme at concentrations (5o and 100 mg/ml) didn't show any inhibitory activity for the growing of the same *Listeria monocytogenes* isolates. Most of the results showed high antibacterial activity against growth of all *Listeria monocytogenes* isolates from human and animals compared with negative control and this depends on their inhibition zones. In this study, we used six standard antibiotics as a positive control for *Listeria monocytogenes*, where rifampin (5mcg), chloramphenicol (10mcg), streptomycin (25 mcg) and amoxicillin/clavulanic acid (20/10mcg) were effective in inhibition the growth of all *L. monocytogenes* isolates from human and animals, while cefotaxime (10 mcg) and novobiocin (30 mcg) showed no inhibitory effects against growth of all *L. monocytogenes* isolates.

Keywords: Antibacterial activity, Medicinal plants, *Listeria monocytogenes*, Humans.

Introductions

Listeria is a Gram positive, facultative, non-spore forming and anaerobic bacterium; it is important food-borne pathogen because of its prevalent distribution in environment (1). Symptoms caused by listeriosis in humans and animals include encephalitis, meningitis, abortion and septicemia (2). Also, *Listeria monocytogenes* can contaminate milk of animals with mastitis. Furthermore to the quality of silage, other hygiene parameters, confirmed by contribute deeply to the microbiological quality of the milk and good herd health management (3). The studies are suggested that the increased use of antibiotics for therapeutic purposes in humans and animals can cause the development of antibiotic resistance. The factor effective on the level of resistance is the geographical changes; it is originally essential to screen the antibiotic resistance patterns of *Listeria monocytogenes* in environmental sources and food from dissimilar geographic regions (4). The expansion of microbial resistance towards antibiotics makes it needed to search for new potential effective compounds against pathogenic bacteria (5 and 6).

Plants are rich in extensive change of secondary metabolites, for example tannins terpenoids, alkaloids and flavonoids, etc, whereas, they have been made *in vitro* to have antimicrobial properties (7). Many reports are available on the antiviral, antibacterial, antifungal and anti-inflammatory properties of medicinal plants and certain of these records have assisted in recognizing the active principle accountable for such activities and in the developing drugs for the therapeutic use (8). The main antimicrobial components of the medicinal plants in the study included Tannins and Quercetin for Oak*;* it's used to treat acute diarrhea and inflammation. Oak (*Quercus. acuta*) have antibacterial-activity against gastrointestinal bacterial pathogens (9). The green parts of *Thymus vulgaris* is closely general herbal medicine used worldwide, its photochemical has been used as antibacterial (10). The extracts of Cinnamon (*Cinnamomum zeylanicum*) showed notable antibacterial activity against Gram positive bacteria (11). Plants strength signify another treatment in non-sever cases of infectious disease, medicinal plants in general contain numerous bioactive ingredients that can be of interest in therapeutic as well as their low toxicity related with chemical drugs , they can also be likely source for new strong antibiotics to which pathogenic strains are not resistant (12). The present study designed to evaluate the inhibitory effects of ethanolic and chloroformic extracts of three local medicinal plants, included: Oak trunks (*Quercus acuta*), Thyme fruit (*Thymus Vulgaris)* and Cinnamon tree cortex *(Cinnamomum zeylanicum*) (prepared in different concentrations) against the growth of *Listeria monocytogenes* isolated from infected human and animals and compared their activity with efficiency of standard antibiotics.

Materials and Methods

The samples were collected during the period from November 2014 to April 2015 and included: Fifty blood samples were collected from aborted woman under the supervision of specialist medical in maternal and pediatrics hospital in Al-Qadisiyah governorate. Animal samples were collected randomly from infected animals with mastitis, and included **(**100) milk samples of sheep and (120) milk samples of cattle and these samples were collected from different areas in Al-Qadisiyah governorate; all samples were collected and preserved according to (13).

Oxford Listeria Selective agar was used for isolating of *L. monocytogenes* from samples of human and animals; the isolates were confirmed by microscopical examination and motility test (14) and bio-chemical tests (Oxidase test and Catalase test) (15). All bacteria isolates were cultured on nutrient broth then incubated for 24 hr. at 37ºC before use. Oak trunks (*Quercus acuta*), Thyme fruit (*Thymus Vulgaris)* and Cinnamon tree cortex *(Cinnamomum zeylanicum*) were dried and good grinded to be used. These medicinal plants have been obtained from the local market, at Al-Qadisiyah governorate. The preparation of both ethanolic and chloroformic extracts were accomplished according to (16). Whereas, 50 gm. each of powder of plant samples were mixed with 250 ml. each of ethanol (96%) and chloroform; the mixtures placed in tightly closed containers, then they were kept for 2-5 days at room temperature and were shaken by magnetic stirrer times daily. The mixtures were filtered through filter paper to eliminate the coarse plant materials, additional extraction of the residue was recurrent 3-5 times, after a clear supernatant extraction liquid was acquired. The filtrated medicinal plants were evaporated to dryness using a rotary evaporator at 40ºC. Finally, the dried samples were weighed and then they were stored at -20ºC.

Six of the standard antibiotics were selected depending on their broad-spectrum activity and were used as positive controls against *Listeria monocytogenes* isolates**.** The standard antibiotics include: Rifampin (5 mcg), Chloramphenicol (10 mcg), Streptomycin (25 mcg), Amoxicillin/clavulanic acid (20/10 mcg), Cefotaxime. (10 mcg) and Novobiocin (30 mcg) (Bioanalyse). The serial dilutions of each extract was prepared by diluting 2 gm. each of dried plant extract with 5 ml. each of ethanol (96%) and chloroform to get stock solution at a concentration (400 mg/ml) and rest of concentrations were made from this stock solution including: concentration (200) mg/ml (17).

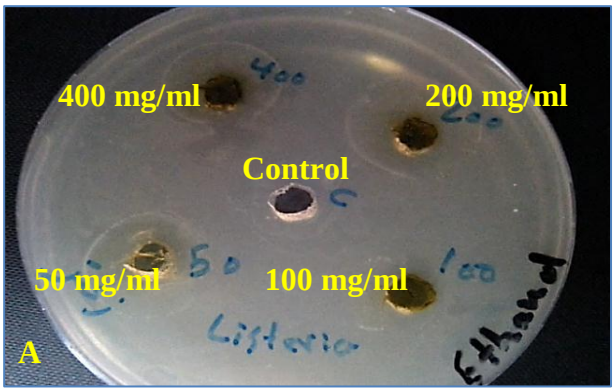
Inhibition effects of tested medicinal plants extracts on growth of *Listeria monocytogenes* was detected by the agar well diffusion method according to (18). *Listeria monocytogenes* isolates were cultured in nutrient broth prepared according to the instructions given by the manufacturing company (HIMEDIA/ Laboratories, and MUMBAI/India). The colonies of *L. monocytogenes* were postponed by sterile cotton swab, then they were placed in sterile tube having 10 ml of nutrient broth, then incubated for 24 hours at 37ºC to produce bacterial suspension exposed by the attendance of turbidity. The turbidity of the culture was compared with 0.5 McFarland Nephelo-meter. The consistent inoculum suspension has been inoculated within (15-20) min. The Mueller-Hinton Agar was used for testing standard antibiotics and the selected plants extracts susceptibility of *L. monocytogenes* isolates and was prepared according to the producer guide (HIMEDIA/ Laboratories, and MUMBAI/ India). The media were poured at 45^0C, a standard borer of 5 mm. The diameter has been used to cut 5 constant wells on the surface for each agar plate. Each plate contained four different concentrations of each plant extract, and included (50, 100, 200, and 400) mg/ml. Also, each of ethanol (96%) and Chloroform as (negative controls) more dropped 0.1 ml in one well on the similar extract plate. *L. monocytogenes* isolates inoculated on the Mueller-Hinton agar surface by streaking of the swab over its, then the inoculums were dried; 0.1 ml of each concentration of each plant extract was dropped into the wells of its inoculated plates. One disc of each standard antibiotic as (positive controls) was located with a sterile forceps over the surface of plate. The plates were incubated for 24 hrs. at 37^0C. The zone of inhibition around each well was measured in MM. according to (19).

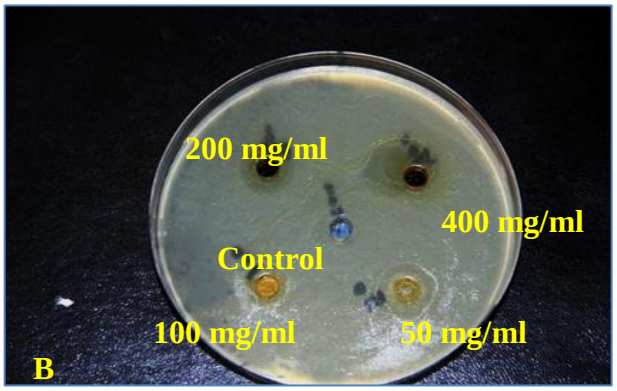
Data were analyzed by (ANOVA) test at significant differences level of ($P<0.05$) by using SPSS program (Version. 10). The values were expressed as mean ± SE (20).

Results and Discussions

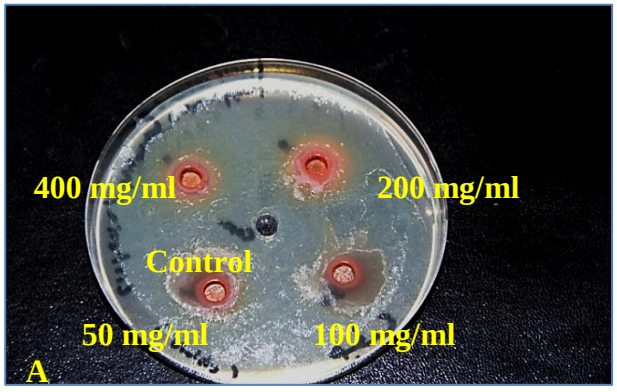
Listeriosis is the highest common zoonotic disease that causes many health problems in humans and leads to significant economic losses in animals. One of the important causes of this disease is *L. monocytogenes* which is extensive in the environment where it moves through the food chain to humans and animals; this cause epidemics or individual cases of infection (21). The results in present study of the investigation of *L. monocytogenes* in human and animals showed that the detection rate of *L. monocytogenes* isolates in blood samples collected from aborted woman was 6% (3/50), while the detection rates in milk samples collected from sheep and cattle were 4% (4/100) and 9.16% (11/120) respectively. These results may be attributed to *L. monocytogenes* infections of animals and people are transmitted between the environment and contaminated food (22). Also, some studies have associated contaminated foods, for example cheese, milk, and beef with the transmission of *L. monocytogenes* to human (23 and 24).

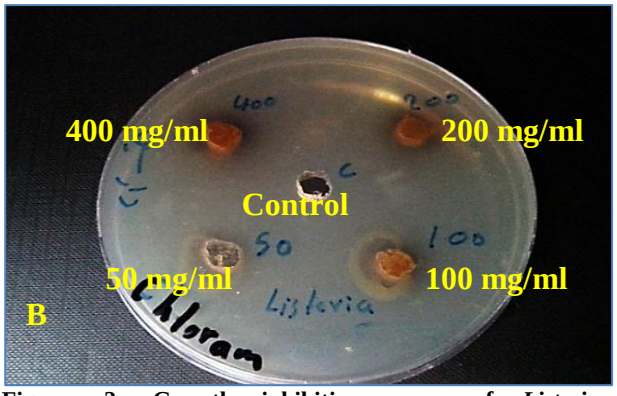
In this study, the antimicrobial activity of various concentrations of ethanolic and Choroforomic extracts of Oak, Thyme and Cinnamon against *L. monocytogenes* isolated from humans and animals was determined by agar well diffusion method. Agar well diffusion methods are widely used because of their simplicity on one hand and low cost on the other hand; in addition, it may help to detect if there is any resistance from the bacteria to any drug, medicinal plant or agents that may be used to study (25). The antibacterial activity of the tested plants extracts against isolates from human is shown in (Table, 1) and (Fig. 1 and 2). These results showed good inhibitory effect for ethanolic and chloroformic of Thyme at all tested concentrations, as well as the chloroformic extracts of Oak and Cinnamon at concentrations (50, 100 mg/ml) against *L. monocytogenes* isolated from blood samples of aborted women, while ethanol extracts of Oakand cinnamon did not show any antibacterial activity against these isolates. The statistical analysis by using ANOVA at ($P<0.05$) displayed that there were significant differences between the effect of the studied concentrations of Oak, Cinnamon and Thyme extracts.





Figure, 1: Growth inhibition zones of *Listeria monocytogenes* isolates from human on Mueller-Hinton agar caused by using (A) ethanolic extract and (B) Chloroformic extracts of thyme *(Thymus Vulgaris)*, The central well was contained (0.1 ml) each of chloroform and ethanol (96%) and other four wells were contained on the extract concentrations.





Figure, 2: Growth inhibition zones of *Listeria monocytogenes isolates* from human on Mueller-Hinton agar caused by (A) Chloroformic extracts of oak and (B) chloroformic extracts of Cinnamon *(Cinnamomum zeylanicum*). The central well was containing (0.1 ml) each of chloroform and ethanol (96%) and other four wells were contained on the tested extracts concentrations.

Table, 1: Effects of some medical plant extracts at different concentration on zone of growth inhibition (mm) of *Listeria monocytogenes* isolates from blood samples of human.

Concen. of extracts	Growth inhibition zones (mm) /Concentration (mg/ml)					
	Oak (*Quercus acuta*)		Thyme *(Thymus Vulgaris)*		Cinnamon *(Cinnamomum zeylanicum*)	
	Ethanol	Chloroform	Ethanol	Chloroform	Ethanol	Chloroform
50	0 ± 0^{Aa}	19 ± 1.25^{Ab}	14.66 ± 0.66^{Ac}	14.83 ± 0.3^{Ac}	0 ± 0^{Aa}	13.5 ± 0.61^{Bd}
100	0 ± 0^{Aa}	19.83 ± 0.65^{Ac}	12.33 ± 0.66^{Bb}	11 ± 0.36^{Bb}	0 ± 0^{Aa}	11.83 ± 0.3^{Cb}
200	0 ± 0^{Aa}	0 ± 0^{Ba}	11.83 ± 0.47^{Bb}	11.83 ± 0.47^{Bb}	0 ± 0^{Aa}	0 ± 0^{Aa}
400	0 ± 0^{Aa}	0 ± 0^{Ba}	15.83 ± 0.4^{Ab}	19.16 ± 1.13^{Cc}	0 ± 0^{Aa}	0 ± 0^{Aa}
Negative Control	0 ± 0^{Aa}	0 ± 0^{Ba}	0 ± 0^{Ca}	$0\pm 0^{Da.}$	$0\pm 0^{Aa.}$	$0\pm 0^{Aa.}$

Dissimilar small letters mean significant difference of horizontal values at (P<0.05). Dissimilar capital letters mean significant difference of vertical values at (P<0.05). The sample volume was 100 µl in each well. Data of the results of inhibition zone in mm diameter including the diameter of well (6mm).The results were expressed as mean ± SE for 3 bacterial isolates.

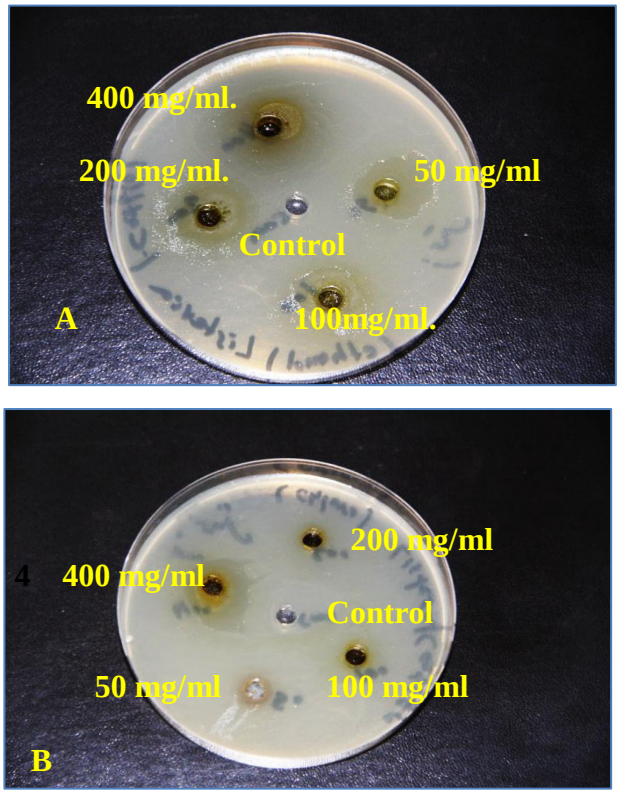
The standard Antibiotic sensitivities of *Listeria monocytogenes* isolates from blood samples of aborted women were confirmed with disc diffusion assay against five standard antibiotics. *Listeria monocytogenes* isolates were susceptible to rifampin (5 mcg), amoxicillin/ clavulanic acid (20/10 mcg), chloramphenicol (10 mcg) and streptomycin (25 mcg) and gave zones of growth inhibitions (9.83±0.4, 11.33±0.42, 16.83±0.87 and 18±0.36 mm) respectively, while the same *L. monocytogenes* isolates were resistant to the antibiotic including Cefataxime (10 mcg) and Novobiocin (30 mcg) as in (Table, 2).

Table, 2: The effects of standard antibiotics on *Listeria monocytogenes* isolated from humans.

Antibiotics	Zone of growth inhibition (mm)
Cefotaxime (CTX) 10 mcg	0 ± 0^{a}
Rifampin (RA) 5 mcg	9.83 ± 0.4^{b}
Chloramphenicol (C)10 mcg	16.83 ± 0.87^{c}
Amoxicillin/clavulanic acid (AMC) 20/10 mcg	11.33 ± 0.42^{b}
Streptomycin (S)	18 ± 0.36^{d}
Novobiocin (NV) 30 mcg	0 ± 0^{a}

Dissimilar small letters mean significant difference of the values at (P<0.05). The results were expressed as mean ± SE for 3 bacterial isolates.

The results showed the anti-bacterial activity of four concentrations of ethanolic and chloroformic extracts of three medicinal plants and five standard antibiotics in the inhibition of the growth of *L. monocytogenes* isolates from milk samples of sheep and cattle showed by measuring the size each of bacterial growth inhibition zone as abridged in (Tables, 3-5). It varied in dependence on the type of plant and the used concentration. Results presented in (Table, 3) show that ethanolic extracts of Thyme were the most active agents against isolates of cattle and gave inhibition zones (18.66±0.61, 16.66±0.61, 15.5±0.42 and 10.66±0.33 mm) at concentrations (50, 100, 200 and 400) mg/ml respectively. The chloroformic extracts of Thyme showed low antibacterial activity against *L. monocytogenes* isolates from cattle and gave inhibition zones (11.16±0.3, 10.33±0.42 and 9.16±0.47 mm) at a concentrations (50, 100 and 200 mg/ml.) respectively, except chloroformic extracts of Thyme that showed a high antibacterial activity and gave inhibition zones 20.5±0.76 mm. at a concentration (400 mg/ml) (Fig. 3). The results revealed that chloroformic extracts cinnamon exhibited antibacterial activity against *L. monocytogenes* isolates from cattle. As the mean of the diameter of inhibition zone were10.83±0.3 and 12.66±0.42 mm at the concentrations 50 and 100 mg/ml respectively, ethanolic extracts of cinnamon, as well as ethanolic and chloroformic extracts of Oak had no effect on all isolates from cattle at all the tested concentrations.



Figure, 3: Growth inhibition zones of *L. monocytogenes* isolates from cattle on Mueller-Hinton Agar caused by using (A) ethanolic Extract and (B) chloroformic extracts of thyme *(Thymus vulgaris)*, the central well contained (0.1 ml) each of chloroform and ethanol (96%), while other four wells contained on the tested extract concentrations.

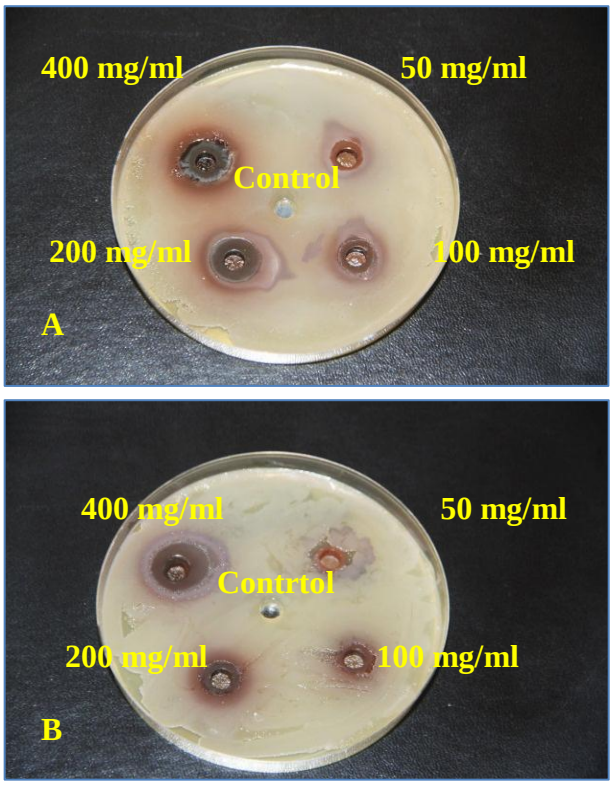
Table, 3: The effects of some medical plant extracts at different concentration on growth inhibition zone (mm) of *Listeria monocytogenes* isolates from cattle.

Concentrations of extracts	Growth inhibition zones (mm.)/Concentration (mg/ml.)					
	Oak (*Quercus acuta*)		Thyme (*Thymus Vulgaris*)		Cinnamon (*Cinnamomum zeylanicum*)	
	Ethanol	Chloroform	Ethanol	Chloroform	Ethanol	Chloroform
50	0± 0^{Aa}	0± 0^{Aa}	18.66±0.61^{Ab}	11.16±0.3^{Ac}	0± 0^{Aa}	10.83±0.3^{Ac}
100	0± 0^{Aa}	0± 0^{Aa}	16.66±0.61^{Bb}	10.33±0.42^{ABc}	0± 0^{Aa}	12.66±0.42^{Bd}
200	0± 0^{Aa}	0± 0^{Aa}	15.5±0.42^{Bb}	9.16±0.47^{Bc}	0± 0^{Aa}	0±0^{Ca}
400	0± 0^{Aa}	0± 0^{Aa}	10.66±0.33^{Cb}	20.5±0.76^{Cc}	0± 0^{Aa}	0± 0^{Ca}
Negative Control	0± 0^{Aa}	0± 0^{Aa}	0± 0^{Da}	0± 0^{Da}	0± 0^{Aa}	0± 0^{Ca}

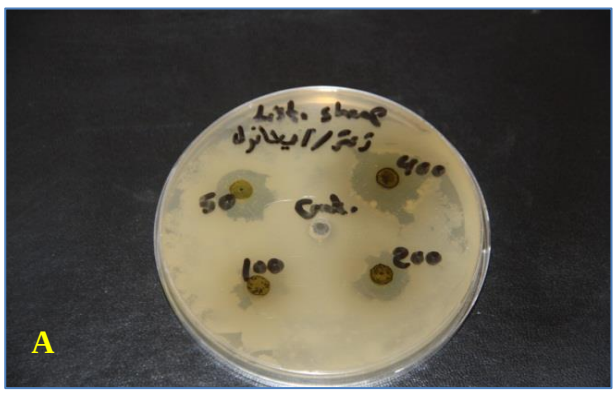
Dissimilar small letters mean significant difference of horizontal values at (P<0.05). Dissimilar capital letters mean significant difference of vertical values at (P<0.05). The sample volume was 100 µl in each well. Data of the results of inhibition zone in mm. diameter including the diameter of well (6 mm). The results for 3 bacterial isolates as mean ± SE.

Listeria-monocytogenes isolated from milk samples of sheep showed moderate sensitivity to ethanolic and chloroformic extracts of Oak, they produced inhibition zones at concentrations (50, 100, 200, 400 mg/ml) (Fig. 4). The ethanol extracts of Thyme showed good anti-bacterial activity and gives inhibition zones (15.16±0.3, 12.16±1.19, 13.16±0.54 and 20±1 mm) at a concentrations (50, 100, 200 and 400 mg/ml) respectively, while chloroformic extracts of Thyme showed low anti-bacterial activity and gave inhibition

zones (11.33±0.66 and 9.33±0.33 mm) at concentration 200 and 400 mg/ml respectively against the isolates from sheep; also the same isolates showed the highest sensitivity to chloroformic extracts of Cinnamon and gave inhibition zones (10.66±0.55, 16.33±0.55, 25.16±1.44 and 26.16±1.4 mm) on concentrations (50, 100, 200 and 400 mg/ml) respectively (Fig. 5). While ethanol extracts of Cinnamon did not show any effect on these tested isolates, and the results are described in (Table, 4).



Figure, 4: Growth inhibition zones of *L. monocytogenes* isolates from sheep on Mueller-Hinton agar caused by (A) ethanolic extract and (B) chloroformic extracts of Oak (*Quercus acuta)*. The central well was contained (0.1 ml.) each of chloroform and ethanol (96%), while other four wells were contained on the tested extract concentrations.



Figure, 5: Growth inhibition zones of *L. monocytogenes* isolates from sheep on Mueller-Hinton agar caused by (A) Ethanolic extract of thyme *(Thymus Vulgaris)* (B) Chloroformic extracts of Cinnamon *(Cinnamomum zeylanicum*). The central well was contained (0.1 ml) each of chloroform and ethanol (96%), and other four wells were contained on the extract concentrations.

Listeria monocytogenes isolates from milk samples of cattle were susceptible to the antibiotics, including: (Rifampin 5 mcg, Chloramphenicol 10 mcg, Streptomycin 25 mcg., Amoxicillin/clavulanic acid 20/10mcg. with the growth inhibition zones 19.22 ± 0.32, 10.55 ± 0.37, 30.11± 0.48 and 10±0.23 mm. respectively, while the isolates from milk samples of sheep were low susceptible to the standard antibiotics, including (Rifampin 5 mcg, Chloramphenicol and 10 mcg, Streptomycin 25 mcg and Amoxicillin/ clavulanic acid 20/10 mcg, with the inhibition zones 14.88±0.53, 10.80±0.30, 21.77±0.57 and 10.77±0.22 mm, respectively. All the isolates from cattle and sheep were resistant to Novobiocin 30 mcg, and Cefotaxime 10 mcg (Table, 5).

The different extracts and standard antibiotics showed various degrees of growth inhibition zone (mm) in the culture media depending mainly on a difference in source of *Listeria monocytogenes* isolates, type of solvents used for extraction process in addition to concentration of extracts. Through, analysis of variance showed a significant difference for the values at ($P<0.05$) inefficacy between antimicrobial agents which could have arisen as a result of genetic differences in the sensitivities of the isolates to the anti-microbial agents and differences in the modes of action of the antibiotics, some results of this study were significantly greater than those produced by these antibiotics, where these results gave an indicator of a possible broad-spectrum mode of action for the different plant

extracts. The anti-microbial activity found in the plant extracts have been attributed to some of the secondary metabolites that provides its presence in the plant extracts as flavonoids, tannins and terpenes. The anti-bacterial effectiveness of different extracts of the oak may due to the synergistic effect of a number of compound which are present in this plant especially to the tannins and polyphenolic compounds (26). Ethanolic extract of Oak have anti- microbial activity against together Gram positive and Gram negative bacteria in culture media (27). Although the effect of Oak on the growth and multiplication of number of gram positive and negative bacteria *in vitro* in local study is promising, further microbiological, pharmacological and clinical trials are required.

Also Thyme extracts have active compounds that produce anti-bacterial activity *in vitro* similar to thymol (the main constituent of thyme 46.2%) carvacrol, tannin, saponin, triterpenic acids and flavonoides. The presence of aromatic nuclei containing polar functional groups, specially the thymol phenol group is the source of good antimicrobial properties (28). The dissimilar components dispersing at different rates may have been responsible for the variable zones of inhibition obtained in against the susceptible bacteria. *Thymus vulgaris* is expected to have compounds which have a potential antibacterial activity; its significant compounds are flavonoids. Also flavonoid's action is probably due to their capacity to complex with extracellular and soluble proteins and to complex with bacterial cell walls and lipophilic flavonoids may also upset bacterial membranes (11). The antibacterial activity of Cinnamon may be due to their main component, and cinnamaldehyde, and their properties could be manifold, an significant characteristic of plant extracts and their components are their hydrophobicity, which allows them to put in to partition the lipids of the bacterial cell membrane and mitochondria, disturbing the cell structures and version them more permeable, extensive leak from bacterial cells or the departure of dangerous molecules and ions will cause death (29).

Table, 4: The effects of some medicinal plant extract on growth inhibition zone (mm) of *Listeria monocytogenes* isolates from sheep.

Concentration of plant extracts	Growth inhibition zones (mm.)/Concentration (mg/ml.)					
	Oak (*Quercus acuta*)		**Thyme *(Thymus Vulgaris)***		**Cinnamon *(Cinnamomum zeylanicum*)**	
	Ethanol	**Chloroform**	**Ethanol**	**Chloroform**	**Ethanol**	**Chloroform**
50	**10.5 ± 0.42^{Aab}**	**11.16 ± 0.6^{ABb}**	**15.16 ± 0.3^{Ac}**	**0 ± 0^{Ad}**	**0 ± 0^{Ad}**	**10.66 ± 0.55^{Aab}**
100	**12 ± 0.44^{Aba}**	**10.16 ± 0.4^{Ab}**	**12.16 ± 1.19^{Aa}**	**0 ± 0^{Ac}**	**0 ± 0^{Ac}**	**16.33 ± 0.55^{Bd}**
200	**14.5 ± 0.5^{BCa}**	**11.83 ± 0.3^{Bb}**	**13.16 ± 0.54^{Ba}**	**11.33 ± 0.66^{Bb}**	**0 ± 0^{Ac}**	**25.16 ± 1.44^{Cd}**
400	**15.66 ± 0.84^{Ca}**	**16.33 ± 0.42^{Cb}**	**20 ± 1^{Cc}**	**9.33 ± 0.33^{Cd}**	**0 ± 0^{Ae}**	**26.16 ± 1.4^{Cf}**
Negative Control	**0 ± 0^{Da}**	**0 ± 0^{Da}**	**0 ± 0^{Da}**	**0 ± 0^{Aa}**	**0 ± 0^{Aa}**	**0 ± 0^{Da}**

Dissimilar small letters mean significant difference for horizontal values at (P<0.05). Dissimilar capital letters mean significant difference for vertical values at (P<0.05). The sample volume was 100 µl in each well. Data of the results of inhibition zone in mm. diameter including the diameter of well (6mm.). The results for 3 bacterial isolates as mean ± SE.

Table, 5: The effects of standard antibiotics on growth inhibition zone (mm.) of *L. monocytogenes* isolates from cattle and sheep.

Antibiotics	The source of *L. monocytogenes* isolates	
	Cattle	**Sheep**
	Zone of growth inhibition (mm)	
Cefotaxime (CTX)10mcg	**0 ± 0^{a}**	**0 ± 0^{a}**
Rifampin (RA) 5mcg	**19.22 ± 0.32^{b}**	**14.88 ± 0.53^{b}**
Chloramphenicol (C) 10 mcg	**10.55 ± 0.37^{c}**	**10.80 ± 0.30^{c}**
Amoxicillin/clavulanic acid (AMC) 20/10 mcg	**10 ± 0.23^{c}**	**10.77 ± 0.22^{c}**
Streptomycin (S) 25 mcg	**30.11 ± 0.48^{d}**	**21.77 ± 0.57^{d}**
Novobiocin (NV) 30 mcg	**0 ± 0^{a}**	**0 ± 0^{a}**

Dissimilar small letters mean significant difference of values at (P<0.05). The results expressed as mean ± SE for 3 bacterial isolates.

The above studies showed that the medicinal plants may signify new sources of antibacterial with steady, biologically active components that can found a scientific base for the use of medicinal plants. However, the higher efficacy of Oak (*Quercus acuta*), Thyme (*Thymus vulgaris*) and Cinnamon *(Cinnamomum zeylanicum*) which have been recommended for usage in short-term storage of products but additional studies are needed before these extracts can be applied to this purpose.

Extracts and antibiotics had similer levels of anti-microbial activity against *L. monocytogenes*. This was credited to a similarity in the mechanism of action of these plant and antibiotics. While some extracts were better antimicrobial activity than antibiotic. These plants are active even against organisms that have become resistant to antibiotics. The ethanolic and chloroformic extract of thyme *(Thymus Vulgaris)* had a good anti-listeria activity, signifying its possible use in treatment of infections caused by *L. monocytogenes* isolated from humans and animals. The results of present study support the facts that more research needs to be done on the purification, identification and quantification of the active of extract components with the estimation of their use for the *invivo* studies.

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دراسة التاثيرات التثبيطية لمستخلصات بعض النباتات الطبية في نمو اللستيريا المستوحدة المعزولة من الإنسان والأبقار والأغنام في محافظة القادسية

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الخلاصة

تهدف هذه الدراسة الى تقييم التاثيرات التثبيطية لمستخلصات الايثانولية والكلوروفورم لبعض النباتات الطبية المحلية وهي قشرة جذوع البلوط وثمرة الزعتر وقشور شجرة القرفة التي حضرت بتراكيز (٥٠ و100 و200 و400) ملغم/مل ضد نمو اللستيريا المستوحدة المعزولة من الإنسان والحيوانات ومقارنة نشاطها مع فعالية المضادات الحيوية القياسية.أظهرت نتائج تشخيص اللستيريا المستوحدة في 50 عينة دم جمعت من النساء المصابات بالاجهاض كانت 3 (6%) عينات موجبة، في حين نسب التشخيص في عينات الحليب التي جمعت من الأغنام والأبقار فكانت 4% (100/4) و 9.16 % (120/11) على التوالي. وقد أظهرت نتائج المستخلصات الايثانولية والكلوروفورم لنبات الزعتر وكذلك مستخلصات الكلوروفورم لنبات الجفت والقرفة للتراكيز (٥٠ و١٠٠) ملغم/مل تاثيرات تثبيطية مهمة ضد عزلات اللستيريا المستوحدة من الإنسان في حين مستخلصات الايثانولية لنبات الجفت والقرفة لم تبين اي فعالية مضادة للبكتيريا ضد نمو نفس العزلات البكتيرية. كما بينت مستخلصات الايثانولية والكلوروفورم لنبات الزعتر وكذلك مستخلصات الكلوروفورم لنبات القرفة للتراكيز (٥٠ و١٠٠) ملغم/مل فعالية مضادة للبكتيريا ضد نموعزلات اللستيريا المستوحدة من الابقار، بينما لم تبين المستخلصات الايثانولية والكلوروفورم لنبات الجفت والمستخلص الايثانولية لنبات القرفة اي فعالية مضادة للبكتيريا ضد نمو نفس العزلات البكتيرية. أظهرت النتائج لهذه الدراسة ان جميع

المستخلصات لها فعالية مضاد للبكتيريا ضد عزلات اللستيريا المستوحدة من الاغنام، ماعدا مستخلصات الايثانولية لنبات القرفة بجميع تراكيزها، وكذلك مستخلصات الكلوروفورم لنبات الزعتر في التراكيز (٥٠ و١٠٠) ملغم/مل لم تبين اي فعالية تثبيطية ضد نمو نفس عزلات اللستيريا المستوحدة. ومعظم النتائج بينت تاثيرات تثبيطية مرتفعة ضد نمو جميع عزلات اللستيريا المستوحدة من الإنسان والحيوانات، مقارنة مع السيطرة السالبة وهذا يعتمد على النطاق التثبيطي. وفي هذه الدراسة استعملت ست مضادات حيوية قياسية كسيطرة موجبة اللستيريا المستوحدة وهي ريفامبين (٥ مايكروكرام) وكلورامفنكول (١٠ مايكروكرام) وستربتومايسين (٢٥ مايكروكرام) واموكسيسلين/كلوفانك أسد (١٠/٢٠ مايكروكرام) التي كان لها فعالية في تثبيط نمو جميع عزلات اللستيريا المستوحدة من الإنسان والحيوانات، بينما سيفوتاكسايم (١٠ مايكروكرام) و نوفوبايوسين (٣٠ مايكروكرام) لم تبين أي تاثيرات مثبطة ضد نمو جميع عزلات اللستيريا المستوحدة.

الكلمات المفتاحية:الفعالية المضادة للبكتيريا، النباتات الطبية، اللستيريا المستوحدة، الإنسان.