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EFFECT OF PLANT EXTRACT IN SALMONELLA TYPHI ISOLATED FROM TYPHOID PATIENT

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Abstract*: Salmonella typhi* is a member of *S. serovar* which cause "typhoid fever" , consider common health difficult in developing countries. The purpose of the research was to study plant extract of *S. typhi* in typhoid patient by molecular and bacteriological assay .A total of 90 blood samples aged between (10-60) years ,were involved in this study. blood samples were cultured directly on brain heart infusion broth .After that sub cultured of isolates on MacConkey agar and Hekton enteric agar and S.S agar to find the *Salmonella typhi* then identified by biochemical, molecular assay and antibiotic sensitive test. After that preparation of plant extracts from zingiber and acacia , fractionation with soxholet extractor and isolate the active material by HPLC , evaluation its potency on *Salmonella typhi* isolates by well diffusion and disk diffusion test. Results showed that there was 4 *Salmonella typhi* isolates from blood culture, as well as, *Flic* gene success in amplification of 450bp fragment for amino glycoside resistant , wherever Plant extracts may be show highest inhibition zone diameter of *S. typhi* reached by action of zingiber extract . while acacia extract was accompanied the lowest inhibition effect for *S. typhi* growth
KEYWORDS: *effect of plant extract inSalmonella typhi isolated from typhoid patient*

I. INTRODUCTION

Salmonella is a gram negative, facultative anaerobes essential genus of Enterobacteriaceae. live in the intestinal tract of animals and human also broad range of hosts such as human, swine, poultry, environment and foods . Members of the genus Salmonella might be pathogenic to domestic or wild human and animals (1). It's significant pathogen to the food manufacturing and has been commonly recognized as the etiological cause of food borne out breaks (2).This bacteria cause systemic infection, enteritis and enteric fever (3).

The illness may well last from 3 to 4 weeks and death rate range between (12% and 30%). Although the worldwide burden of typhoid fever has less , emergence of multi drug resistant Salmonella typhi (MDRST) is still a threat to public health . Currently ,almost 107 strains of it isolated; with varying metabolic characteristics, levels of virulence, and multi-drug resistance genes that complicate treatment in areas that resistance are widespread (4).

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II. MATERIALS AND METHODS

Patients: The study included 100 patient, admitted at the hospital and Clinical signs of typhoid patient were recorded by physician and firstly diagnosed with Widal test

***Bacterial isolation*:** specific isolation of *S.typhi* from blood specimens, 5 ml of blood were cultured on Brain-heart infusion broth (BHI broth) then incubated at 37C°/24 hr. , then purified via sub cultured on blood and MacConkey agar . The identification tests for the isolate, including cultural, morphological and biochemical characteristics was done for each isolate.

Molecular Identification for genes by PCR-based assay:

The system used was Flic primers system. As advised by(5,6). And provided by (Bioneer/ Korea) company, as described

Primer	Sequence		Amplicon
fliC 18	F	5’ ACT GCT AAA ACC ACT ACT 3’	363
	R	5’ TGG AGA CTT CGG TCG CGT AG3’	

The PCR amplification process.

The reaction solution : Enzymatic amplification of DNA was approved out in a final volume of (25µl) according to the recommendations of manufacture

• Cycling condition : the reaction was execute in a DNA thermal cycler with no mineralize oil .following numerous tests ,the subsequent program was rely, PCR composed of a pre-heating in 95C for 5min following the start denaturation step ,the mixture was undergo to 40 amplification cycles.

Plants materials :

Plant that used in our studies are completely collected from the market are dried , it is cutted to small pieces and then ground to powder form. Preparation of extracts according to Abo-shanab *et al* (7), dilutions are done by dimethyl sulphoxide10% to different concentration (15,30 and 50 %) of extracts by dissolving (150, 300 and 500) mg of extract with 1 ml of organic solvent as mentioned by Nanasombat and lohasupt-hawee(8).

III. RESULTS AND DISCUSSION

One hundred blood samples collected from patients with suspected typhoid fever 4 isolates (4.44%) recovered from blood of patient. typhoid fever continues to be major cause of mortality and morbidity . in Iraq , rare studies on typhoid fever were approved out .these studies give emphasis to on the epidemiological of *S. typhi* , has studied the out breaks of typhoid fever and reported that the distribution of cases over a large zone within a small time favored the possibility of water-borne disease (9).

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In present study was used flic gene to detect the *S. typhi* form *Salmonella* isolates. mean while the DNA extracted from cultured harboring *S. Typhi* DNA , so that they yielded predicated 363pb fragment .

Holt *et al.*,(10) used PCR in the diagnosis of *S. typhi* and described it as a very specific, sensitive and simple and could become a routine diagnosis test for salmonellosis. since then many studies described the PCR method for detection of the *S.typhi* in human blood samples provide better results than the conventional culture techniques for the diagnosis of both primary infection and relapses, as well as for focal complication of the disease . because of high specificity (95%) and extreme sensitivity (98%) , PCR was used for detection of *S.typhi* from blood ,stool and cultures

Zingiberofficinale extract develop the highest inhibition zone diameter reached 21.36mm ,regarding Arabica ***Acacia Arabica*** accomplished the lowest inhibitory effect(6.5mm) the highest inhibitory zone effected was attained with zingiber at 50% concentration (20.76mm) whilst the lowest effect was at 15 % concentration (18.80mm) as table(1)

Results that obtained from our studies, showed that *Zingiberofficinale* extract is more effective as antimicrobial drugs against *S.typhi* were acacia Arabicais less effective against *S.typh* (table 1). This result same as like reported by al-bayati *et al.*, (11) whom mentioned that *Zingiberofficinale* extracts was more effective against most pathogenic bacteria like *S .aureus* , *E.coli* , *Klebsiella pneumonia*, *St.pyogenes. Enterococcus.faecales* and *pseudomonas .aeruginosa* when tested by well diffusion method. Present study was non confirmed with **Ajayi and Akintola** (12) whom proved that *Acacia Arabica* (Bark) showed that the plant possess antibacterial activities against various organisms, this extracts showed antibacterial activity against *S. aureus, S. mutans, S. sanguis, S.salivarius, L. acidophilus* and *C. albicans*. Activities were also found against *P. aeruginosa, E. coli, B.licheniformis, S.aureus, Enterobacter sp., E. coli, P. intermedia* and *P. gingivalis* .

Table(1) :inhibition zones (mm in diameter) caused by plant extracts against *S.typhi* isolated from typhoid fever patient .

Treatment extract	Concentration(mg/ml)			Mean
	150	300	500	
Zingiber	18.80	20.76	22.72	20.76
Acacia	5.0	5.5	6.5	5.5

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