INTERNATIONAL JOURNAL OF CURRENT RESEARCH IN CHEMISTRY AND PHARMACEUTICAL SCIENCES

(p-ISSN: 2348-5213: e-ISSN: 2348-5221)
www.ijcrcps.com

Research Article

STUDY ON ANTIBIOTIC SENSITIVITY AND DISSEMINATION OF *Shigella dysenteriae* TYPE1 IN WHITE MICE FOLLOWING EXPERIMENTAL INFECTION

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Abstract

Shigellosis is a highly infectious disease affecting man and animals. For the importance of the bacteria causing this disease (*Shigella dysenteriae* type 1) , this study aimed to identify their dissemination in white mice and antibiotic sensitivity. For this reason strain, of *Shigella dysenteriae* type1 obtained from the central public health laboratory, following their identification to be sure *Shigella dysenteriae* type 1 .Using cultural , biochemical , API-20 and sereny test (induce keratoconjunctivitis) The LD50 for this strain was $8x10^3$CFU in (0.25 ml bacterial suspension) then I/p inoculated into white mice. Other group of white mice given $1x10^7$ CFU/ ml bacterial suspension orally. Three mice were sacrificed every two days intervals for 28 days .*Shigella dysenteriae* type 1 were persisted for 2- 3 weeks in liver, spleen and for 1-2 weeks on other organs. Following oral route, the bacteria persisted for 2-3 weeks in intestine and for 1-2 weeks in other organs. *Shigella dysenteriae* type 1 were sensitive to Gentamycin,Nalidix acid , Neomycin and Tobramycin whereas, resistant to streptomycin , Amoxicillin and Trimethoprim and the bacteria showed variable sensitivity to other antibiotics such as Erythromycin , Chloramphenicol and Clindamycin .

Keywords: Shigellosis, *Shigella dysenteriae* type 1, keratoconjunctivitis, sensitivity to antibiotics.

Introduction

Shigellosis is an infectious disease affecting children at different ages, associated with a dysenteric diarrhea admixed with mucous and blood and abdominal pain and fever. It distributed all over the world accompanied by high mortalities in children (1) especially in poor and crowded countries (2).The infection with *Shigella dysenteriae* type 1 considered to be highly fatal associated with the different inflammatory lesions and microabscesses occurred in the intestinal mucosa induced by this microbe a highly pathogenic Shigella species in addition to hemorrhage and pseudodiphtheric membrane on intestinal mucosal surface (3). Monkeys considered to be the natural host for shigella in addition to human being, also Guinea pigs and other laboratory animals might be affected by this microbial agent (4). From the importance of this disease caused by*Shigella dysenteriae* type1 in human and animals, this study aimed to study the dissemination of *Shigella dysenteriae* type 1 - in the white mice organs and identify their antibiotic resistance.

Materials and Methods

Alocal strain of *Shigella dysenteriae* type1 obtained from central public health laboratory. The strain were re-identified to be sure *Shigella dysenteriae*type1 (5) using cultural, biochemical test ,API -20 tests , slide agglutination test and serenytest in Guinea pigs causing keratoconjunctivitis (6) . The LD 50 for this organism were 8 $x10^3$ CFU (7),then 45 mice were taken and injected 1/p with $8x10^3$ CFU (0.25ml of bacterial suspension) (8) Three mice were sacrificed at 2 days intervals and bacterial isolation were done from the different organs to identify *Shigelladysenteriae* type1 dissemmation (5). Also the similar bacterial isolate (*Shigella dysenteriae* type1) were given orally into 45

mice with a dose of $1x10^7$ CFU/ml bacterial suspension containing 5mg streptomycin and 50 mg $CaHCo_3$ per ml. (8). Also 3 mice were sacrificed 2 days intervals and bacterial isolation were done from the different organs following oral administration of *Shigella dysenteriae* type1 to identify their dissemination in mice organs comparable to 1/p route. Antibiotic sensitivity test were done (9) using 10 types of antibiotic discs (Streptomycin, Erythromycin, Amoxicillin, Chloramphenicol, Gentamycin, Nalidix acid, Tobramycin, Trimethoprim, Clindamycin and Neomycin.

Results

The dissemination of *Shigella dysenteriae* type1in mice organs following 1/p route of infection

The microbial agent were persisted in liver and spleen for 19 and 23 days respectively (Tabe-1) and in heart blood, lungs and mediastinal lymph node for 15 days and in gall bladder and kidneys for 13 days. In periton for 11 days and in intestine and brain for 9 days.

Table-1 : The dissemination of *Shigella dysenteriae* type1 in mice organs following 1/p infection

periton	Intestine	brain	Lungs	Kidney	Mediastinal L. node	Heart blood	Gall bladder	spleen	Liver	Period
+	—	—	—	+	+	—	—	+	—	1 day
+	—	—	+	+	+	+	—	+	—	2day
+	+	+	+	+	+	+	—	+	—	3 day
+	+	+	+	+	+	+	+	+	+	5 day
+	+	+	+	+	+	+	+	+	+	7 day
+	+	+	+	+	+	+	+	+	+	9 day
+	—	—	+	+	+	+	+	+	+	11day
—	—	—	+	+	+	+	+	+	+	13 day
—	—	—	+	—	+	+	—	+	+	15day
—	—	—	—	—	—	—	—	+	+	17day
—	—	—	—	—	—	—	—	+	+	19day
–	–	–	–	–	–	–	–	+	—	21day
–	–	–	–	–	–	–	–	+	—	23day
—	—	—	—	—	—	—	—	—	—	25day
—	—	—	—	—	—	—	—	—	—	28day

No. of animals =45 ,No. of sacrificed animals =3 each period , The dose = 8×10^3 CFU

The dissemination for *Shigella dysenteriae* type 1 in mice organs following oral route of infection

The microbial agents were persisted (Table 2) in intestine for 19 days, in spleen for 15 days , in liver and kidneys for 13 days , in lungs for 11 days , in heart blood and brain for 9 days and not isolated from periton all over the period of infection.

Antibiotic sensitivity test

The results showed that there was variation in the sensitivity and resistance of *Shigella dysenteriae* type 1 against different 10 types of antibiotics (table 3). The microbial agent was sensitive to Gentamycin ,Nalidix acid , Neomycin and Tobramycin , whereas , resistant to Streptomycin ,Amoxicilliy and Trimethoprim and the bacterial agent were variable between resistance and sensitive to Erythromycin , Chloramphenicol and Clindamycin.

The similar microbial agent were highly virulent through sereny test (Keratoconjunctivitis) in infection eye and negative other control eye .

Table (2) : The dissemination of *Shigella dysenteriae* type1 in mice organs following oral route of infection

periton	Intestine	brain	Lungs	Kidney	Heart blood	spleen	Liver	Period
—	+	—	—	—	—	—	—	1 day
—	+	—	—	+	—	+	—	2day
—	+	—	—	+	—	+	—	3 day
—	+	—	—	+	+	+	+	5 day
—	+	+	+	+	+	+	+	7 day
—	+	+	+	+	+	+	+	9 day
—	+	—	+	+	—	+	+	11day
—	+	—	—	+	—	+	+	13 day
—	+	—	—	—	—	+	—	15day
—	+	—	—	—	—	—	—	17day
—	+	—	—	—	—	—	—	19day
—	—	—	—	—	—	—	—	21day
—	—	—	—	—	—	—	—	23day
—	—	—	—	—	—	—	—	25day
—	—	—	—	—	—	—	—	28day

No of animals =45 ,No of sacrificed animals =3 each period , The dose = 1 x 10^7 CFU

Table -3 : Antibiotic sensitivity test against *Shigella dysenteriae* type1

Type of resistance	abbreviation	Antibiotic type	Number
R	S	Streptomycin	1
I	E	Erythromycin	2
R	Amx	Amoxicillin	3
I	C	Chloramphenicol	4
S	GN	Gentamycin	5
S	NA	Nalidixic acid	6
S	Tob	Tobramycin	7
R	T	Trimethoprim	8
I	CI	Clindamycin	9
S	N	Neomycin	10

R=Resistance, S= sensitive , I= Intermediate

Discussion

Following 1/p inoculation of *Shigella dysenteriae* type1, the microbial agent were continuously proliferated in periton and mediastinal lymph node and through the thoracic duct reach into heart blood and disseminated all over the organs and persisted for along time (2-3 weeks) in liver, spleen and in mediastinal lymph node due to that these microbial agents prefer the reticudoendothelial tissue enriched in these organs (10) whereas , in other organs such as brain , kidneys and lungs were persisted for 1-2 weeks. Their persistence in these organs were occurred following reaching the microbial agents into the blood and the bacterial emboli disseminated all over the organs (8) similarly with other microbial agents dissemination (*Salmonella typhi*) in mice.

Following oral route of the infection the bacterial agents (*Shigella dysenteriae* type1) were disseminated slightly in the mice organs comparable to 1/p route of inoculation which showed extensive bacterial proliferation and dissemination into other organs. The slight dissemination of the microbial agent following oral route of infection belong to mild proliferation and growth of the microbial agents in the intestine, in addition to the bacterial flora in intestine had a role in bacterial clearance from this organ,so the microbial agent persisted in intestine for 2-3 weeks comparable to the 1/p route of infection together with the reticuloendothelial tissue a preferable site for the bacteria in the other organs such as liver, spleen and mediastinal lymph node (10 , 11).

This study revealed that the *Shigella dysenteriae* type 1 were sensitive to some antibiotics and resistant to others , the resistance against the antibiotics by these microbial agent due to transmission of genes carried on plasmids between these bacterial agents from the resistant to the sensitive bacteria (12,13) also some bacterial agents had R-factor which important for transmission of resistant feature to sensitive bacteria against antibiotics (13,14,15) .

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