

In vitro the effect of intestinal normal flora on some pathogenic bacteria .

Dr.abbass shaker

Ali adel

Leena abd Al-Redha

Abstract:

The effect of two types of intestinal bacterial normal floral (*E.coli* and *klebsiella*) were tested against 50 bacterial isolates collected from different infection of patients and hospital setting ,using inverted disk technique and wells on nutrient agar, the result revealed that the normal flora were affected on 30 bacterial isolates from total 50 isolates .The effect of normal flora were appear on 8 isolates from burns and wounds,5 isolates from ear infections, 9 isolates from gastrointestinal infections (diarrhea), 8 isolates from hospitals delivery rooms *E.coli* affect on 2 isolate for each of *klebsiella* and *pseudomonas* isolated from gastrointestinal infections (diarrhea). In contrast, *klebsiella* was affect on 9 isolates of *pseudomonas aerogenosa* , 6 isolates of *klebsiella* and 3,3,2,2,1 isolates of *staphylococcus aereus* , *serratia* ,*Acinetobacter* ,*Enterobacter* ,*E.coli*, respectively, isolated from ear infections ,delivery rooms ,gastrointestinal infections, burns and wounds.

Introduction:

Normal flora are bacteria, fungi, and protozoa that live on or within the bodies of humans and animals. They do not cause disease in healthy individuals, instead, they are commensalists or mutualists with regard to the host.. In other words, normal flora :that population of microbes that are commonly found (colonized) in or on the body in the absence of disease.(Levinson, 2008)At birth the intestinal tract is sterile, but organisms are soon introduced with food, in breast-fed children, the intestine contains large numbers of lactic acid streptococci and lactobacilli, these aerobic and anaerobic, produce acid from carbohydrates and tolerate pH 5.0.(Jawetz *et al.*,2007)In the normal adult colon, 96–99% of the resident bacterial flora consists of anaerobes ,only 1–4% are facultative aerobes (gram-negative coliform bacteria, *enterococci*, and small numbers of *pseudomonads*, lactobacilli, candidae, and other organisms)(Cebra,1999; Davis,1996) . More than 100 distinct types of organisms, which can be cultured routinely in the laboratory, occur regularly in normal fecal flora, there probably are more than 500 species of bacteria in the colon including many that are likely unidentified. (Schauer , 1997)The microorganisms that are constantly present on body surfaces are commensals, their flourishing in a given area depends upon physiologic factors of temperature, moisture, and the presence of certain nutrients and inhibitory substances. these organisms may yet the resident flora of certain areas plays a definite role in maintaining health and normal function(Hunter *et al*, 1999). On mucous membranes and skin, the resident flora may prevent colonization by pathogens and possible disease through "bacterial interference." The mechanism of bacterial interference may involve competition for receptors or binding sites on host cells, competition for nutrients, mutual inhibition by metabolic or toxic products, mutual inhibition by antibiotic materials or bacteriocins, or other mechanisms. (Levinson, 2008).

The Methods :

1-Specimen collection:

A- A total of 50 isolates of pathogenic bacterial isolates were collected from different infections in order to test the effect of normal intestinal flora on pathogenic bacteria. They were represented by 9 isolates of burns, 7 isolates of ear infection, 24 isolates of intestinal infection (diarrhea) and 10 isolates from delivery room.

B-Normal bacterial isolates :

Ten normal bacterial isolates were collected from healthy persons represented by 5 isolates of *E. coli* and 5 isolates of *Klebsiella*.

2-Identification of bacterial isolates:

All bacterial isolates were identified according to the (Macfaddin, 2000)

using the following criteria :

1-morphological characters

2-biochemical tests were include:

a-Indole test

b-Methyl red test

c-Simmon citrate

d-TSI test

e-Motility test

3-the effect of two type of normal bacteria isolates (*E. coli* and *klebsiella*) were tested against all the bacterial isolated using inverted disk and wells method on Mollure-Hinton agar (Al-Qasab and Al-Kaphagi, 1992).

Result and discussion :

The normal flora of human body particularly that of intestinal normal flora have important role in maintenance of microbial contain especially that pathogenic were invaded intestinal tract, via antagonism them in different ways. For this reasons the effect of two type of normal bacterial flora (*klebsiella* and *E. coli*) was tested on 50 isolates of pathogenic bacteria isolates from different sites, including 9 isolates from burns infections, 24 from gastrointestinal, 7 from ear infections and 10 isolates from delivery rooms of hospitals. *E. coli* was detected in 6 isolates. *Klebsiella* was recorded in 14 isolates, 19 isolates of *Pseudomonas aeruginosae*, 3 isolates of *Serratia sp.* And 2 isolates for each of *Enterobacter sp.* and *Acinetobacter sp.*, While gram positive *Staphylococcus aureus* was detected in 4 isolates (table -1)

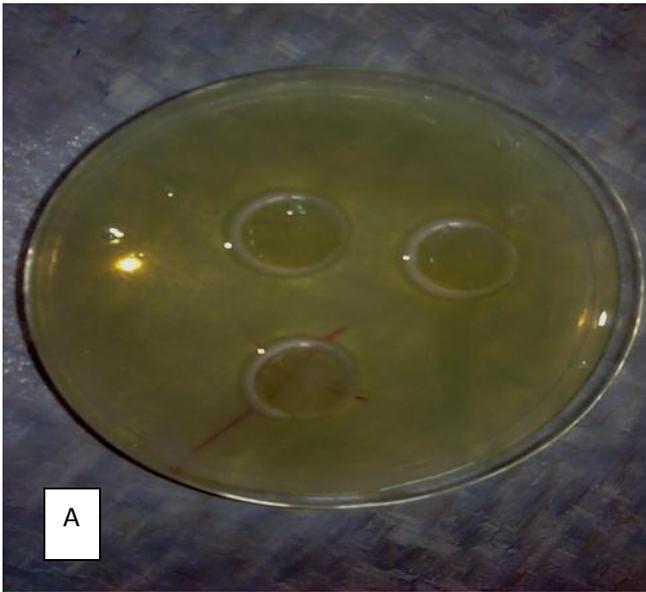
Table-1 :Distribution of bacterial isolates according to infection site

Pathogenic bacteria	Burns	diarrhea	Ear infection	Delivery room	Total number
<i>E.coli .</i>	1	5	/	/	6
<i>Klebsiella spp.</i>	2	10	/	2	14
<i>Pseudomonas aerogenosa</i>	4	6	7	2	19
<i>Enterobacter spp.</i>	1	1	/	/	2
<i>Serratia spp.</i>	/	1	/	2	3
<i>Staphylococcus aureus</i>	1	1	/	2	4
<i>Acinetobacter spp.</i>	/	/	/	2	2

The Inverted disk and wells method on Muller –Hinton agar was used to tested the effect of normal flora on pathogenic bacteria, the result of the study revealed that the effect of normal flora were appear on 30 of bacterial isolates, indicating from inhibition zone around the inverted disk or well, The effect of normal flora were represented on 8 isolates from burns and wounds infections ,5 from ear infections ,9 and 8 isolates from intestinal infections and delivery rooms respectively (Table 2,figure 1)

(table 2) Distribute of cases according to infection site

Site	Number of bacteria	Positive	Negative
Wounds and burns	9	8	1
Ear infections	7	5	2
Diarrhea	24	9	15
Delivery room	10	8	2
Total	50	30	20



Figar-1 :The inhibition zone of normal flora on pathogenic isolates

A- Inverted disk

B- Walls method

Table -3 : Effect of normal intestestinal flora on some of pathogenic bacteriae

Normal flora	Isolated bacteria	Total number	positive	Negative
<i>E.coli</i>	<i>Klebsiella spp.</i>	5	2	3
	<i>E.coli</i>	3	/	3
	<i>Pseudomonas aerogenosa</i>	5	2	3
<i>Klebsiella spp.</i>	<i>Pseudomonas aerogenosa</i>	14	9	5
	<i>E.coli</i>	3	1	2
	<i>Staphylococcus aureus</i>	4	3	1
	<i>Enterobacter spp.</i>	2	2	/
	<i>Serratia spp.</i>	3	3	/
	<i>Acinetobacter spp.</i>	2	2	/
	<i>Klebsiella spp.</i>	9	6	3
	total	50	30	20

The normal flora *klebsiella* spp. was affected on 26 of bacterial isolates , whereas *E.coli* was affected on only 4 of bacterial isolates (table 2)

This results may due to that the normal flora isolates may be produce some microbial substance causing inhibit the growth of the bacterial isolates ,one of the important substance called bacteriocin it is a protein inhibited the growth of similar or related bacteria lacking the gene encoded to produced this substance, bacteriocin production was controlled by gene carried on plasmid, some of bacteria lack the same plasmid mostly are sensitive to some substance (Davis, 2007).On other hand, the resident flora may prevent colonization of pathogens and possible disease through "bacterial interference." The mechanism of bacterial interference may involve competition for receptors or binding sites on host cells, competition for nutrients, mutual inhibition by metabolic or toxic products, inhibition by antibiotic

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