

## Relationship between ADA level and type of bacterial causative agents of chronic Suppurative Otitis media

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### ABSTRACT

A total of 344 ear swabs were obtained from 300 outpatients. ( 152 were female and 148 were male ) .256 of them had unilateral and 44 had bilateral otorrhea , attending to the ENT-Department at general teaching hospital in Najaf governorate for the period from 1st December 1999 to of July 2000 .all patients submitted to ENT-examination by physicians .in addition to that 3ml of venous blood were aspirated from 41 patients with this disease and 45 normal healthy individuals carefully selected as a control for assessment the adenosine deaminase (ADA) enzyme specific activity . *Pseudomonas aeruginosa* ( the predominant pathogens in this study ) represented in 21.6 % from all isolates followed by other pathogens which include *staphylococcus aureus*(18%) *Proteus* species (14.2%) *Echerichia coli*(10%) *Klebsiella pneumoniae*(8.4%) anaerobic bacteria which includes bacteriodes species (3.1%) and peptostreptococcus species. (2.7%) were also of 41 patients. This activity showed a significant decrease in patients ( $69. \pm 5.4$  U/mg) then in the between three patterns of infection and the level of specific activity of immunoenzyme ADA specific activity were also found between three patterns of infection .in which the Mono – inaction group has a higher activity .there is an association between bacterial infection and the level of specific activity of immunoenzyme ADA.

### INTRODUCTION

Otitis media (OM) is one the commonest infectious diseases in infants and young children .and the complications and sequelae of the disease persist in some individual into adult years (1) chronic suppurative otitis media (CSOM) is a from of OM which is still one of the commonest medical problems. and it is damage .it also afflicts

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a significant percentage of the population and causes long – term morbidity clinically manifests with deafness and discharge (2,3) .CSOM may be a squelae of severe combined immunodeficiency (SCID) diseases like others chronic diseases. So that, the specific activity of

adenosine deaminase (ADA) enzyme assay is an important factor to give a clear picture of the human defense system, since ADA enzyme is one of the most essential immune enzyme.

### Materials & methods

The study was carried out on 300 consecutive outpatients with CSOM during the period between December 1999 to July 2000 at ENT – Department in general teaching hospital in Najaf governorate. They included both sexes (152 females and 148 were males) . with a different age groups . they had not received antimicrobial agents systemically or locally for at least one week before examination . with a history of ear disease more than three months . no dry ears were included in this study .all patients submitted to ENT-months before examination. After cleaning the external auditory canal (EAC) from cerumen and pus with a sterile swab moistened by 70% ethyl alcohol, 344 ear swab were obtained from 300 patients 50 patients were randomly selected for nasopharynx swabs (to detect the role of ET in ear infection ) to determine the activity of ADA enzyme in the infected individuals .3ml of venous blood was aspirated from 41 patients .

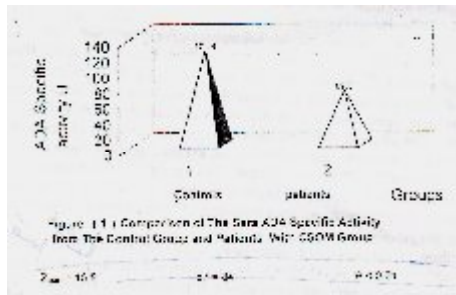
### Results

Out of 344 specimens ( ear swabs ) collected from 300 patient suffering from CSOM 330 ( 9% ) specimen yielded microbial growth presented in 450 microbial isolated .while 14(3%) specimens yielded no growth gram – negative bacteria were the commonest bacterial isolates detected in 271 (60.2%) following by gram – positive bacteria in 166 (36.9%) and unidentified pathogens in 13 (2.9%) *P.aeruginosa* was the dominant microorganism among the gram – negative bacteria (21.6%) following by proteus species (14.2%) *E.coli* ( 10%) and *K.pneumoniae* (8.4%) .other gram-negative bacteria showed low frequency or isolation ranged between 3.1 and 0.2% *S.aureus* was the most frequent microorganism among Gram – positive rest ranged between 2.7 and 0.7 % as presented in table (1) .

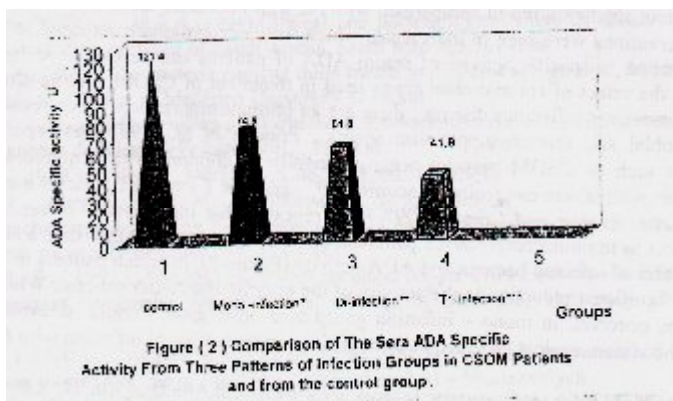
**Table (1) bacterial isolates and their frequency from 300 patients with CSOM.**

Type of isolates	No. of isolates	%
<b>Gram negative ( n= 271 ) aerobic &amp; facultative bacteria (n= 257)</b>		
<b>Pseudomonas aeruginosa</b>	<b>97</b>	<b>21.6</b>
<b>Proteus sp.</b>	<b>64</b>	<b>14.2</b>
<b>Escherichia coli</b>	<b>45</b>	<b>10</b>
<b>Klebsiella pnunioniae</b>	<b>38</b>	<b>8.4</b>
<b>Enterobacter sp.</b>	<b>7</b>	<b>1.6</b>
<b>Moraxella catarrhalis</b>	<b>5</b>	<b>1.1</b>
<b>Citrobacter sp.</b>	<b>1</b>	<b>0.2</b>
<b>Anaerobic bacteria (n= 14) bacteroides sp.</b>	<b>14</b>	<b>3.1</b>
<b>Gram – positive bacteria (n=166) aerobic &amp; facultative bacteria (n=154)</b>		
<b>Staphylococcus aureus</b>	<b>81</b>	<b>18</b>
<b>Staphylococcus epidermidis</b>	<b>30</b>	<b>6.7</b>
<b>Sterptococcus pneumoniae</b>	<b>21</b>	<b>4.7</b>
<b>Bacillus sp.</b>	<b>19</b>	<b>4.2</b>
<b>Diphtheroid bacilli</b>	<b>3</b>	<b>0.7</b>
<b>Anaerobic bacteria (n=12) Peptoreptococcus sp.</b>	<b>12</b>	<b>2.7</b>
<b>Unidentified pathogens</b>	<b>13</b>	<b>2.9</b>
<b>Total</b>	<b>450</b>	<b>100</b>

Results of the ADA enzyme specific is important to give us a clear picture of the human immune defense system .the data of ADA specific activity in sera of control and chronically ear infected individual are presented in figure ( 1 ). The mean activity of the enzyme for the CSOM patients group was (69.5±2.8U/mg) which is significantly lower (P<0.01) than the value of the control sera (121.4± 1.8U/mg)



The sera ADA specific activity for patients form three patterns of bacterial infection is also measured the specific ADA activity of the mono-infection group was  $(74.9 \pm 3.3 \text{U/mg})$  which is significantly ( $P < 0.01$ ) lower than the vlue of the control sera ( $121.4 \pm 1.8 \text{U/mg}$ ) .while at the same time .it is higher than the value of the di-infection group ( $61.8 \pm 4.5 \text{U/mg}$ ) and the tri-infection group ( $41.9 \pm 0.3 \text{U/mg}$ ) sera as shown in figure (2) .



### Discussion :

The present study confirmed that gram – negative bacteria are an important component of the bacterial flora in chronic otitis media P.aeruginosa is the most common organism isolated from chronic ear disease. It was isolated in 21.6%, following by S. aureus (18%) and proteus species (14.2%) as shown in table 1 . this result was expected for this organism ( P.aeruginosa) , due to many reasons , firstly ,P.aeruginosa is the most secondary invaders when resistance of the middle ear is lowered (8) , secondly , it easily spreads to the compromised patients via EAC from healthy carriers ( by nurses or physicians hands ) and / or environmental sites (9) thirdly the high incidence of P.aeruginosa indicates more general antibiotic resistance than in the case with gram –positive strains ( 10 ) and fourthly , it is resistance for phagocytosis and opsonization by producing a large number of intracellular products such as , alkaline protease , elastases and exotoxin A which can cleave both IgG and complement which then lead to inhibit the function of the cells of the immune system (11,12) from the other hand , it has been recognized by Berman (1995)(13) that the first stage of pseudomonas infection in CSOM in initialed by the attachment of bacteria to the mucosal cells of MEC , and this usually occurs because it possesses an adherence factors (Alginate) , since the attachment of bacteria to the mucosal cells is pre- requisite for bacterial proliferation , colonization , and the release of toxins enzymes which

can injure the underlying cells and host and , in consequence , cause an invasive disease (14) the results presented in this study regarding *P. aeruginosa* was found to be in agreement with some studies with of Karma et al 1978 (17) and AL-Faris et al 1998 (18) in which our results disagree particularly *S.aureus* were the predominant pathogen in CSOM . these disagreement may be due to the geographical differences , *Proteus* species were predominantly secondary invaders from EAC and when the resistance of the ME is lowered (8) .in this study proteus sp. Isolated in 14.2%( table 1) this finding in compatible with other studies that found proteus sp. In 20% (19) and 3.6 %(16) .in this study *S. pneumoniae* (4.7%) and *M.catarrhalis* (1.1%) were also isolated .although these organisms are the most common bacterial species causing AOM , our results these bacteria are in agreement with many studies such as Khalil (1980)(20) which reported that these organisms were isolated from CSOM , but in low frequency these organisms are usually associated with acute inflammation , so their isolation was thought to be associated with an acute exacerbation of CSOM ADA enzyme is one of the most essential immune enzyme . its function gives a picture of immune status of the body .it was found that , it plays a critical role in the normal development of the immune system . this enzyme is also essential for proper development of T-and B-lymphocytes in mammals (21) .any reduction in this enzyme will make the patients more susceptible to infection by bacteria or virus (22) .since its function which catalyzes the diminution of adenosine and dexoy-adenosine to inosine and deoxyinosine, so any reduction in this enzyme will lead to be accumulation of adenosine nucleosides (dATP) which acts as an allosteric inhibitor of ribonucleoside Reductase arresting in vitro DNA synthesis (23).

Al- Shawk(1999) (24) found a low ADA activity in women with chronic trichomoniasis infection compared to non – infected women A reduction in ADA specific activity were also observed in moderately and severely malnourished children in comparison with the well nourished children (25) and in addition to that similar observations were seen in individuals which were occupationally exposed to ionizing radiation (26).the reduction in specific activity of serum ADA of patients can be explained by the effect of the disease it self and effect of antimicrobial drugs used in treatment of CSOM as an elucidation for the used of antimicrobial; and immunosuppression agents Principi et al (1991) (27) reported a less severe bacterial diseases such as CSOM may by occur frequency in children with acquired immunodeficiency syndrome . patients with severe congenital or acquired B- and / or T-cell deficiencies may develop CSOM infection (28) since , knappe and

Gregor (1997) (29) revealed that the decrease of resistance to common pathogen may occur in immunocompromised patients .the present study also established a negative correlation between the numbers of infected bacteria and ADA activity (Figure 2) .in which patients in tri-infection group displayed a high significant reduction in the activity of the enzymes than other groups .while , the increase in ADA activity was observed in mono – infection group than other group could be explained as a healthy stimulated immune system against infection in some patients in early time of infection with first pathogen.

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