ISSN: 0975 -8542

Journal of Global Pharma Technology

Available Online at: www.jgpt.co.in

RESEARCH ARTICLE

Isolation and Identification of Bacterial Causes of Acute Tonsillitis

Wurud Ali Hathal, Frial Gemeel Abd*, Shaimaa Jassim Mohemeed

University of Babylon, College of Sciences, Dept. of Biology, Hilla. Iraq.

Abstract

Acute tonsillitis was an inflammatory process of the tonsillar tissues and was usually infectious in nature. Acute tonsillitis starts suddenly and usually goes away again within one to two weeks. This infection was often bacterial, or sometimes viral. A hundred of blood and swabs specimens were collected from patients who were suffering from tonsillitis whose did not take any drugs for both sex with age ranging from 1-60 years in AL-Hilla-Teaching hospital and hospital Babylon for birth and children during a period from October 2017 through January 2018 . The causative agent for tonsillitis was isolated and diagnostic of microorganism according to morphological and biochemical test by using VITEK compact 2 .The results appeared the number of gram positive bacteria about 100 and 23 isolate from gram negative bacteria .The most common causative agent was *Streptococcus pyogenes* and *Staphylococcus aureus*. The sensitivity of bacterial to antibiotic was appeared most of bacterial resistance to amoxicillin, Cefotaxime and Aztreonam) but sensitive to other antibiotic such as Gentamycin, Chloramphenicol and Ciprofloxacin kit. Qualitative and semi quantitative ASO determination was done using the ASO latex agglutination kit. 24%of patient was ASO positive and 67% was ASO negative The level of ASO in patients that tested positive for the presence of ASO ranged from 1600IU/ml to 25600IU/ml. 12 sample were the highest concentration of 25600IU/ml .The conclusion of this study found that *Streptococcus pyogenes* was most common causative agent for tonsillitis and the bacteria resist amoxicillin.

Keywords: *Tonsillitis, Amoxicillin, Streptococcus, ASO.*

Introduction

Tonsillitis is inflammation of the tonsils, typically of rapid onset. It is a type of pharyngitis. Symptoms may include sore throat, fever, enlargement of the tonsils, trouble swallowing, and large lymph nodes around the neck. Complications include peritonsillar abscess [1] Tonsillectomy is still one of the most common surgical procedures for the treatment of tonsillitis in children. Tonsillitis and tonsillectomy are likely to have a distinct link where the incidence of one should reflect that of the other.

While the entire incidence of tonsillectomy in a population may be extreme less than the total incidence of tonsillitis, a healthcare system should be capable to familiarize to upsurge rates of a specific surgical treatment when the specific sign for that treatment upsurges [2].The tonsils contain immune-cells such as, germinal centers of B-lymphocytes, T-lymphocytes and other antigen presenting cells such as macrophages, which serves immune acquisition and defense [3, 4].

The palatine tonsils are dense compact bodies of lymphoid tissue that are located in the lateral wall of the oropharynx, bounded by the palatoglossal muscle interiorly and the palatopharyngeus and superior constrictor muscles posteriorly and laterally Both tonsils and adenoid are part of the Waldeyer ring, which is a ring of lymphoid tissue found in the pharynx.

The lymphoid tissue in this ring provides defense against pathogens. The Waldeyer ring is involved in the production of immunoglobulins and the development of both B cells and T cells [5]. There are many types of tonsillitis acute, recurrent, and chronic tonsillitis, and peritonsillar abscess.

Acute Tonsillitis

Is inflammation of the tonsils, which is caused by bacteria or virus such as double-stranded DNA viruses (human adenoviruses, Epstein Barr Virus), single-stranded DNA viruses (Human Boca Virus), single-stranded RNA viruses (influenza and para-influenza

viruses; rhino-viruses; entero-viruses including Coxsackie viruses; corona viruses; respiratory syncytial virus (RSV); human meta-pneumonia-virus), retroviruses [human immunodeficiency viruses (HIV)] The most important microorganisms that create bacterial tonsillitis are GABHS, i.e. *S. pyogenes*. The illness transmission typically happens using droplet infection transferred by various other patients with acute GABHS tonsillitis, very rarely by asymptomatic service providers [6].

Chronic Tonsillitis

Is one of the most common otorhinolaryngological diseases and tonsillectomy continues to be one of the most common surgical procedures as well as the standard for treating adults with this clinical entity[7].

Recurrent Tonsillitis

A poly microbial flora containing both aerobic and anaerobic bacteria has been perceived in core tonsillar cultures in cases of recurrent pharyngitis, and children with recurrent GABHS tonsillitis have different bacterial populations than children who have not had as many infections. Other competing bacteria are reduced, offering less interference to GABHS infection. *S. pneumoniae, S. aureus,* and *H. influenzae* are the most common bacteria isolated in recurrent tonsillitis, and *Bacteroides fragilis* is the most common anaerobic bacterium isolated in recurrent tonsillitis [8] usually caused by many different bacterial pathogens and flare up again a few weeks after cessation of an antibiotic therapy. Depending on the frequency and severity of such episodes, there is an indication for tonsillectomy [9].

Peritonsillar Abscess

Are collections of purulent material between the tonsil fibrous capsule and the pharyngeal constrictor muscles that usually develop near the superior pole [1011].The formation of the PTA is probably an evolution from acute tonsillitis to peritonsillar tonsillitis to peritonsillar abscess [11].

The main causes of tonsillitis are caused by bacterial especially beta-hemolytic and other streptococci. However, in tonsillitis related to infectious mononucleosis, the most common virus is the EBV, which present in 50% of children. CMV, hepatitis A, HIV, rubella and toxoplasmosis infections may also result in the clinical picture of infectious mononucleosis, which requires differential diagnosis *S. aureus* appears to be the main causative agent in the pathogenesis of acute tonsillitis **[**12]. Tonsilitis is managed by means of treatment according to viral and bacterial infection. Viral tonsillitis treatment involves rest, recovery and symptom relief. It is also important to drink plenty of fluids and have regular meals (soft foods and smoothies are best) whereas bacterial infection is confirmed by throat culture and antibiotics will be prescribed to prevent complications [13].

It is important to take the full course of antibiotics as prescribed to prevent the infection. Antibiotics will not be prescribed for viral tonsillitis because antibiotics are not effective against viruses. Viral tonsillitis will usually get better without treatment. Pain relief and reduction of fever can be achieved with over-the-counter paracetamol (e.g. Panadol) and ibuprofen [14].

Antibiotics are kept for secondary bacterial pharyngitis. Due to the danger of a generalized papular rash, prevent ampicillin and associated compounds when infectious mononucleosis (MN) is suspected. Related reactions from oral penicillin-based antibiotics (eg, cephalexin) have been described. Hence, initiate treatment with alternative anti streptococcal antibiotic, for example, erythromycin.

Manage antibiotics if situations support a bacterial etiology, for example, the incidence of tonsillar exudates, occurrence of a fever, leukocytosis, contacts who are ill, or contact with a person who has a documented group A beta-hemolytic S. pyogenes (GABHS) infection. In several cases, bacterial and viral pharyngitis are clinically indistinguishable.

Waiting 1-2 days for throat culture consequences has not been shown to reduce the practicality of antibiotic treatment in avoiding rheumatic fever [15] Treatment of acute tonsillitis is largely supportive and focuses on maintaining adequate hydration and caloric intake and controlling pain and fever. Corticosteroids may shorten the duration of fever and pharyngitis in cases of infectious mononucleosis. In severe cases of mononucleosis, corticosteroids or gamma globulin may be helpful. GABHS infection obligates antibiotic coverage [16].

Tonsillitis treatment started with antibiotics, Nsaids, fluoroquinolone antibiotics and penicillin's but if disease continues or patient is unresponsive to medication, surgery is recommended. Antibiotics were given to patient after surgery to prevent any further infection and care should be taken. Here, pharmacist should advise patient about his /her medication and should cross check medication errors, if any (multiple prescribing). This will lead to great improvement in patient care [17].

The use of immunological assays such as Anti-Streptolysin O (ASO) would provide useful in the diagnosis of streptococcal infections and their complications, and during follow-up, as well as in evaluating the effectiveness of treatments [18] as well as in situations when the throat culture technique is ineffective or when the patient has commenced antibiotics therapy. Significant findings have shown that an ASO-positive measurement might be used in conjunction with throat culture to identify GAS) [19].

The presence of an immune response to either GAS somatic or extracellular antigens remains the most reliable means for documentation of bonfire infection [20]. Aim of this study was isolation of gram negative and positive bacteria and sensitivity of bacterial to antibiotic and study of some immunological parameter such as ASO test.

Material and Method

Method

Study Group

A total number of 92 patients (41female and 52 male).The age of the study group ranged from1 year-60years

Bacterial Culture and Isolation

Samples

A total number of 92tonsillar swabs were collected from children (41female and 52 male) The age of the patient ranged from 1 years to 60 years.

Bacteriological Examination

Isolation of Bacteria

Tonsillar swabs were cultured on the following media

- Blood agar: to show the hemolytic properties of micro-organisms.
- Mac Conkey's agar: for isolation of *Enterobacteriacea.*
- Mannitol salt agar :for isolation of *staphylococcus* species
- Muller Hinton agar: for bacterial antibiotic selectivity

Samples were cultured on these three media were then incubated overnight aerobically at 37°C. The organisms were identified according to the method of [21]

Identification of the Isolates

Microscopical Examination

Smears from the colonies were stained with Gram's stain and examined microscopically. According to Gram Staining reaction, shape and cell arrangement the isolated microorganisms were divided into:

- Gram positive cocci.
- Gram negative bacilli

Disk Diffusion Test

Susceptibility of S. *aureus* isolates was determined by the KirbyBauer disc diffusion method on Mueller-Hinton agar. Bacterial colonies from each isolate were transferred into a suspension medium adjusted to 0.5 McFarland turbidity standards (1.5 × 108 CFU/ml). Inoculums were swabbed on the entire surface of agar plates followed by the application of six selected commercially available antibiotic disks of methicillin (30 μm), gentamicin (10 μm), erythromycin (15 μm), co-trimoxazole (1.25 + 23.75 μm), clindamycin (30 μm) and fusidic acid (10 μm) using sterile forceps (5 disks per plate).

Plates were inverted and incubated for 18 to 24 h at 37°C. The antibiotic vancomycin was not included in the susceptibility testing of methicillin-resistant S. aureus (MRSA) isolates. Zones of inhibition were determined according to the standards outlined by the Clinical and Laboratory Standards Institute (CLSI) [22].

Vitek 2 System

The Vitek 2 System was used to confirm the biochemical test and antibiotics; assay had been performed according to the manufacturer's instructions. This system consists of personal computer, reader incubator that prepared up of many inner constituents including: card filler mechanism, card cassette,

cassette loading processing mechanism, bar code reader, card sealer, cassette carousel and incubator.

In addition to transmittance optics, waste processing, instruments control electronics and firm ware. The system was equipped with an extended identification data base for all routine identification tests that provide an improved efficiency in microbial diagnosis which reduce the need to perform any additional tests, so that will increase safety for both the test and the users. All the following steps were prepared according to the manufacturer's instructions.

Three ml of normal saline were placed in plane test tube and inoculated with a loop full of isolated for standardization of colony to McFarland's standard solution (1.5×10^8cell/ml). The standardized inoculums were placed into the cassette and a sample identification number entered into the computer software via barcode. The VITEK 2 card thus connected to the sample ID number. Then the cassette was placed in the filler module, when the cards were filled, transferred the cassette to the reader incubator module. All following steps handled by the instrument, the instrument controls the incubation temperature, the optical reading of the cards and continually monitors and transfers test data to the computer for analysis.

Serological Test

Antistreptolycin ASO test was done according [23]. The ASO-latex reagent of Hannover Company, Germany was a suspension of polystyrene particle sensitized with streptolysin O. Slide agglutination test for the qualitative and semi-quantitative detection of anti-streptolysin O antibodies. Latex particles coated with streptolysin O were agglutinated when mixed with samples containing ASO. Each serum sample was tested for ASO by a commercial kit using their instructions. Equal volumes (50µl) of serum and the reagent were mixed on a microscopic slide for a few minutes and observed for agglutination .Development of visible agglutination indicated a positive qualitative.

ASO test and represents 200 IU/ml as per the manufacturer's instructions. If positive, the end titer was determined semi-quantitatively. For this, the positive serum was diluted 1:2, 1:4, 1:8, 1:16, 1:32 ….etc. with sterile normal saline. Each dilution was tested as for the qualitative test. To get the end titer, the dilution factor was multiplied by 200. Thus if the serum sample is positive at 1:4 dilutions, the titer is taken as 200 IU x 4 = 800 IU/ml.

The Results

Bacteriological Study

Diagnostic of Bacteria that Isolated from Tonsillitis

A 92 patients who met the study criteria and their samples were analyzed of which 41% were females and 51% were males .the mean aged were 47.8% aged between (1-5) years .17.3% of patient from (6 -20)years 28.2% patients from (21-60) (Table1).

Table 1: Distribution of tonsillitis patients according the age

Age	Percentage %
1-5 years	48%
6-10 years	17%
11-20 years	7%
21-35 years	15%
36-60 years	13%

Table 2: Distribution of tonsillitis patients according the gender

Gender	Percentage %
Male	55%
Female	45%

This table show that the infection occurs were high in aged group (1-5) years and the infection occur in males were high than female in all age groups Growths of *S. pyogenes*, *S. pneumoniae*, *S .aureus*, *Klebsiella* pneumonia, *Pseudomonas aeruginosa*, and *Escherichia coli* were identified using enriched and differential media (Blood agar and MacConkey agar ,Eosin methylene blue , mannitol salt agar), Gram stain technique, as well as biochemical tests (catalase test, oxidase test, coagulase test) Using Blood agar Eosin methylene blue and MacConkey agar; *S. pyogenes* was exhibited a clear and complete hemolysis on blood agar while *S. pneumonia was* appeared as a mucoid stain on the blood agar appearing as a central depression, indicating

alpha hemolysis. On MacConkey agar, colonies of *E. coli* were seen as non-mucoid pink colonies, and on Eosin methylene blue were seen metallic sheen those of *K.pneumonia* were seen as large, pink and mucoid colonies on Mac Conkey agar, *P. aeruginosa* was seen as large green-brown colonies.

On MacConkey agar. After isolation of the organisms from tonsil swab and culture on different agar used, the organisms were Gram stained and *S. pyogenes* and *S. pneumoniae* were seen as Gram positive cocci some appearing in chains and others in pairs, *S.*aureus was also seen as Gram positive cocci but appearing in clusters. *E.coli*, *K. pneumonia*, and *P. aeruginosa* species were seen as Gram negative rods. Under the light microscope the catalase and oxidase test were done indicated that *S. pyogenes* was a negative catalase activity and positive oxidase while *E. coli*, *K. pneumonia,* and *P. aeruginosa* were positive catalase activity. And *E. coli* was negative oxidase. *P. aeruginosa* was positive oxidase and *K. pneumonia was* negative oxidase. *Staphylococcus* was positive catalase and negative oxidase .coagulase test done indicated that. *S. aureus* was positive coagulase test but *S. Epidermis* was negative for this test Gram negative bacteria were diagnostic by biochemical test such as in dole .methyl red, Vogas-proskauer, Simmon Citrate test and Kligler iron agar test the result positive. result of in dole formation of red ring after added Kovacs' reagent, Simmon Citrate positive result convert of color media after incubation 24 hr from blue color to green color Negative result remain the blue color.

Vogas-proskauer test positive result formation of red ring after added of reagent Table (3) Show bacteria diagnostic by Vitec 2 compact system and Table (4) the percentage of bacteria isolated from sore throat.

Table 3: Some biochemical test (IMVC) for gram negative bacteria

Bacteria	Indole	methyl red	Vogas procurer	Cimon citrate
Acinetobacter ursingii	-	+	-	+
Pseudomonas aeruginosa	-	+	-	+
Klebsiella peumoniae	-	+	-	+
Acinetobacter iwoffii	-	+	-	+
Pantoea spp	-	+	-	+
Acinetobacter baumannii complex	-	-	-	+
Klebsiella oxytoca	+	-	-	-
Buttiaurella agrestic	-	+	-	+
Oligella ureolytica	-	+	-	+
Pseudomonas fluoresces	-	-	-	+
Escherichia coli	+	+	-	-

Isolation of Bacteria that Causes of Tonsillitis

Isolation of bacteria causes of acute tonsillitis from the 92 clinical specimens .only 23 specimen isolated were belonged to gram negative bacteria and 92 specimens isolated were belong to gram positive bacteria The result were show in Figure (1).

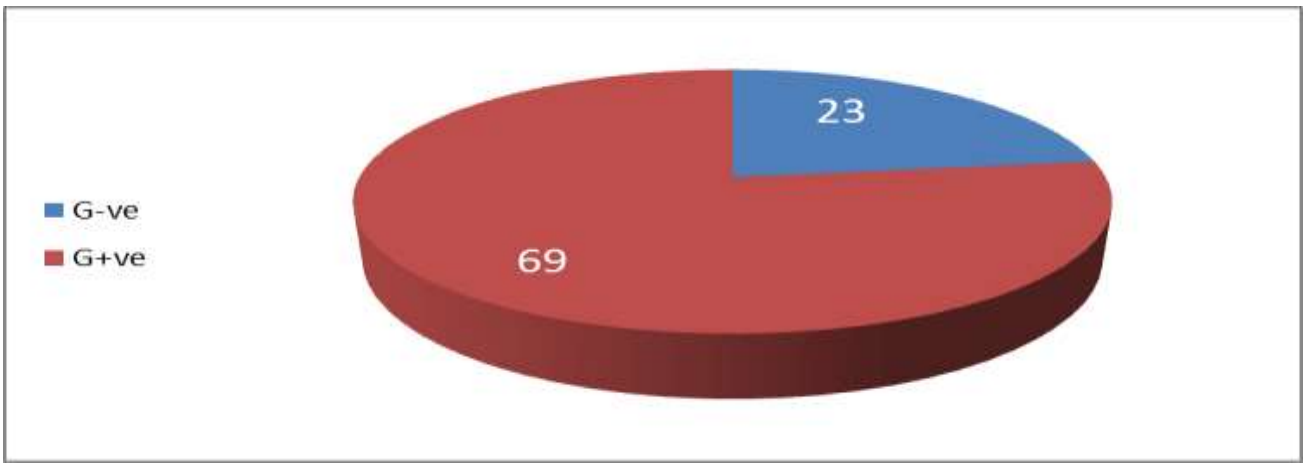

Figure 1: Profile of bacteria that isolated from tonsillitis swabs

In this study included isolation of different species of gram negative bacteria five isolates of *pseudomonas* ,six from *E. coli* ,two of *Klebsilla* and three isolates from

Acinetobacter and other of bacteria isolated from patient was suffer from sore throat.
The gram positive bacteria isolated from sore throat *S. aureus* ,*S. pyrogens* and other of microorganism and diagnostic by different test such as macroscopic and microscope, biochemical test .and Vitec 2compact system.

Table 4: The percentage of bacteria that isolated from sore throat

	Bacteria	Number	Percentage %
1	*Staphylococcus aureus*	18	19.5%
2	*Staphylococcus lantus*	5	5.4%
3	*Staphylococcus epidermis*	17	18.4%
4	*Streptococcus pyogenes*	24	26.1%
5	*Bacillus*	6	6.5%
6	*Pseudomonas aeruginosa*	4	4.3%
7	*pseudomonas fluorescence*	1	1.1%
8	*Acinetobacter iwoffii*	1	1.1%
9	*Acinetobacter urisingii*	1	1.1%
10	*Acinetobacter baumonia*	1	1.1%
11	*Pantoea spss.*	1	1.1%
12	*Buttiaurella agrestic*	1	1.1%
13	*Oligella urelytica*	1	1.1%
14	*Salmonella enteritis*	1	1.1%
15	*Sallmonella typhi*	1	1.1%
16	*Enterobacter acrogens*	1	1.1%
17	*E .coli*	6	6.5%
18	*Klebsiella pneumonia*	1	1.1%
19	*Klebsiella oxytoca*	1	1.1%
	Total	92	

This study included antibiotic sensitivity for bacteria causes of tonsillitis. The result was analyzing sensitivity pattern of18 isolates of *S. aureus* and 24 isolated of *S .pyogenes* were, sensitivity of 100%with gentamycin, 50%with Amikacin and 77.7 % with Ciprofloxacin and 88.8 % with Chloramphenicol and 72.2 % with Tetracycline and 72.7 % with Erythromycin.

The resistance of 100% with Cefotaxime, 100% Cefotaxime and 55.5% Amoxicillin .*E .coli* were showed 16.6% resistance to Amoxicillin, Cefotaxime .Amikacin ,Aztreonam, Tetracycline, Gentamycin and sensitive to ciprofloxacin and streptomycin. *Pseudomonades were* found sensitive 33.3%with Aztreonam and 66.6 % Gentamycin and Amikacin and 100% sensitive ciprofloxacin. Resistance 33.3 % with Gentamycin and Amikacin. *Acinetobacter* were sensitive 100 %with ciprofloxacin and 66.6%with Amikacin and resistance 66.6%with Gentamycin and 33.3 % with Amikacin. *Klebsilla* were sensitive 100% ciprofloxacin and 33.3 % Aztreonam and 66.6% Gentamycin and tetracycline, while resisted 100 % with Amoxicillin and Cefotaxime. 66.6 % Aztreonam 33.3 % Gentamicin and 33.3% amikacin, streptomycin and tetracycline. *Oligella ureolytica* appeared resistance 100 % with (vancomycin, Cefotaxime. Amoxicillin, streptomycin, Gentamycine and erythromycin). *Pantoea spp*. was resistance 100%with vancomycin and erythromycin .*Buttiaurellaagrestis* was resistance 100% with (vancomycin, erythromycin and Cefotaxime Table (5).

Table 5: Antibiotic sensitivity to some bacterial isolates from sore throat

Bacteria	CN	VA	C	AK	CN	CTX	CIP	TE	E	AMC	S	ATM
Streptococcus pyogenes	S	-	S	S	S	R	S	S	S	R	-	-
E.coli	R	-	S	R	R	R	S	R	-	R	I	R
Klebsiella pneumoniae	-	-	R	I	S	R	S	S	-	R	I	S
Klebsiella oxytoca	-	-	I	I	S	R	S	S	-	R	I	R
Oligella ureolytica	-	R	-	-	-	R	-	R	R	R	R	-
Acinetobacter ursingii	-	-	-	S	S	R	S	I	-	-	-	-
Acinetobacter iwoffii	-	-	-	S	S	R	S	R	-	-		-
Acinetobacter baumannii complex	-	-	-	R	I	I	S	S	-	-	-	-
Buttiaurella agrestis	-	S	-	-	-	S	-	-	S	-	-	-
Staphylococcus aureus	S	-	S	S	S	R	S	S	S	R	-	-

S=sensitive R= resistance I=inter mediaAK= Amikacin CIP= Ciprofloxacin CN= Gentamycin TE= Tetracycline VA=vancomycin E= Erythromycin C=Chloramphenicol AMC= Amoxicillin CN= Gentamycin S= streptomycin CTX=Cefotaxime ATM= Aztreonam

Immunological Results

Antistreptolysin O (ASO) test from one hundred patients only 24 patients had positive to ASO test, and this confirmed by culture, in this study showed that 24(26.1%) Streptococcus isolates.

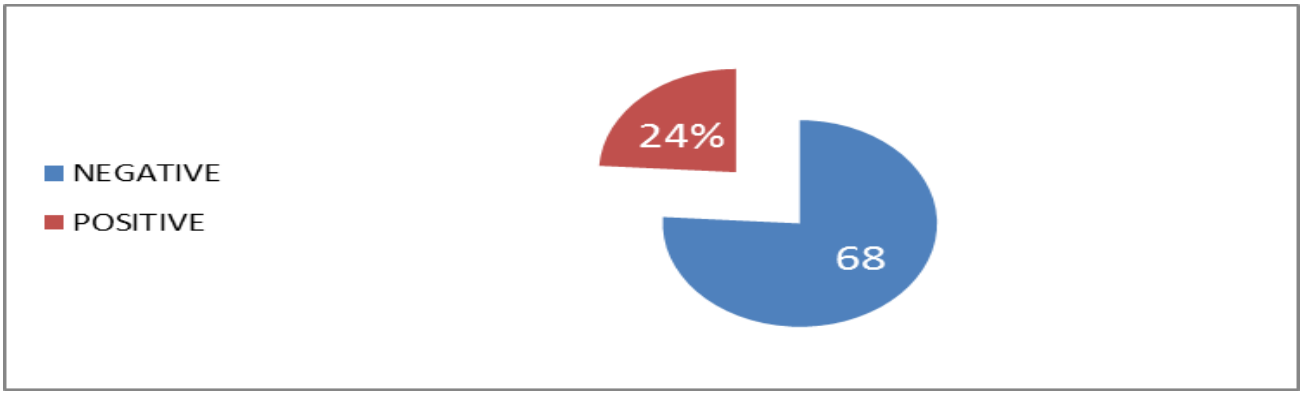

Figure 2: the results of antistreptolysin O in this study

Table 6: the level of ASO in serum of patients that appeared positive for the presence of ASO Antibody

NO. of bacteria	Titer	Concentration (IU/ml)
12	1/128	25600
6	1/16	3200
1	1/32	6400
4	1/8	1600
1	1/64	12800

24 ASO+ and 66 ASO- less than 200IU/ml leaflet of kit

Discussion

Microbiological study for patients suffered from acute tonsillitis showed that *Streptococcus* and *Staphylococcus* were the most common of bacteria that isolated from tonsillitis 19.5 %of *S. aureus* and 26.1 % of *S. pyogens* isolated from tonsillitis and other mixture of bacteria isolated such as *E. coli* and *P. aeruginosa* and *Acinetobacter*.

The results of present study showed that the GABHS was a major cause of tonsillitis as confirmed by positive culture of these microorganisms in 92 of samples as can be observed in these results were in agreement with the findings from several studies were done in many countries, [24] this study agree with [25] who also reported a predominant incidence of *Staphylococcus* over the *Streptococci*. In our study the result was same to the result obtain by [26].

S. aureus was the most common organism streptococcus *pyogenes* this agree to the result obtain from [27] *E. coli* and *K. pneumonia* were obtain in our study was agree with other study obtain by [28] and agree with [29].*P. aeruginosa* obtain as [30] .And other gram negative and positive obtain from tonsillar swab a 11 type of antibiotic study of sensitivity of bacteria for this antibiotic such as Vancomycin, Cefotaxime, Amoxicillin, streptomycin, Amikacin, Ciprofloxacin, Gentamycin, Tetracycline, Erythromycin, Aztreonam and Chloramphenicol. The result of *E. coli* strain sensitive to ciprofloxacin was agreed with other study obtains by [31].

P.aeruginosa appear in our study was Resistance Gentamycin was agree with study obtain by [31]. *Staphylococcus* sensitive to gentamycin, Ciprofloxacin, Erythromycin show in our study also agree with study by [31].

In this study appear all gram negative bacteria were sensitive to ciprofloxacin related to different of factor and resistance to amikacin antibiotic Gram positive bacteria appear high resistance to Cefotaxime and Amoxicillin and 100% sensitive to gentamycin and Cefotaxime ASO remains a useful in the diagnosis of streptococcal infections and their complications, follow-up, as well as in evaluating the effectiveness of treatments [32, 33]. ASO is helpful when the throat culture technique is ineffective or when the patient has already taken antibiotics.

Since impoverished societies cannot afford other tests, such as throat culture, ASO is the only available test for diagnosing streptococcal infection. Significant findings have shown that an ASO-positive

measurement might be used in conjunction with throat culture to identify Group A Streptococcus (GAS) carriers [34] level of ASO test in our study over 200 IU the result agree with [35]. ASO titers have been deployed exclusively for epidemiological studies and the clinical diagnosis of *S. pyogenes* infections and its sequelae, such as rheumatic fever, glomerulonephritis, and reactive arthritis after throat infections [36]. Some individuals are reported to harbor the same strain of GAS in the upper respiratory tract for long periods of time without symptoms and without an increase in ASO titers. This "carrier state" phenomenon remains poorly understood posing some limitations to clinicians and laboratory scientists studying the pathogenesis of streptococcal infections and their sequelae [37].

References

1. Klug TE, Rusan M, Fuursted K, Ovesen T (2016) Peritonsillar Abscess Complication of Acute Tonsillitis or Weber's Glands Infection or Weber's Glands Infection .Otolaryngol. Head Neck Surg., 155(2):199-207.
2. Millington AJ, Phillips JS (2014) Current trends in tonsillitis and tonsillectomy. The Annals of the Royal College of Surgeons of England. 96(8): 586-589.
3. Westermann J (2010) Organe des Abwehr systems. In Anatomie., 355-375.
4. Brandtzaeg P (2011) Immune functions of nasopharyngeal lymphoid tissue. In Recent Advances in Tonsils and Mucosal Barriers of the Upper Airways, 72: 20-24.
5. Windfuhr JP, Toepfner N, Steffen G, Waldfahrer F, Berner R (2016) Clinical practice guideline tonsillitis I. Diagnostics and nonsurgical management. Eur. Arch. Otorhinolaryngology, 273(4):973-987.
6. Kenna MA, Amin A (2009) Anatomy and physiology of theoral cavity. Snow JB, Wackym PA. Ballenger's Otorhinolaryngology Head and Neck Surgery.17th ed. Shelton. BC Decker Inc., 769-774
7. Scholz H, Berner R, Duppenthaler A, Forster J, Topfner N (2013) Deutsche Gesellschaftfu¨rPa¨diatrischeInfektiologie (DGPI), DGPI Handbuch: InfektionenbeiKindern und Jugendlichen, 6. u¨berarbeitete Auflage, Infektionendurchß-ha¨molysierendeStreptokokken der Gruppe A. Georg Thieme Verlag, Stuttgart:509-516.
8. Schwentner I, Hofer S, Schmutzhard J, Deibl M, Sprinzl GM (2007) Impact of tonsillectomy on quality of life in adults with chronic tonsillitis. Swiss Med Wkly, 137(31-32): 454-61.
9. Witsell DL, Orvidas LJ, Stewart MG (2008) To treat study investigators: quality of life after tonsillectomy in adults with recurrent or chronic tonsillitis. Otolaryngol. Head Neck Surg., 138:1-8.
10. Blair AB, Booth R, Baugh R (2015) A unifying theory of tonsillitis intratonsillar abscess and peritonsillarabscess. American Journal of Otolaryngology Head and Neck Medicine and Surgery, 36(4):517-520.
11. Papacharalampous GX, Vlastarakos PV, Vlastarakos G, Kotsis D, Davilis Manolopoulos L (2011) Bilateral peritonsillar abscesses: a case presentation and review of the current literature with regard to the controversies in diagnosis and treatment. Case Reports in Medicine. Article ID 981924, 4.
12. Stelter K (2014) Tonsillitis and sore throat in children. GMS Curr Top Otorhinolaryngol Head Neck Surg., 13:Doc07.
13. Ezzeddini R, Darabi M, Ghasemi P, Jabbari Y, Abdollahi (2012) Circulating phospholipase A2 activity in obstructive sleep, apnea and recurrent tonsillitis. International Journal of Paediatric Otorhinolaryngology, 76:471-47
14. Khoo M, Leong C, Nurjahan I, Shajahan Y, Teng L (2009) The management of upper respiratory tract infection Medical. Journal of Malaysia. 56:260-266.
15. Murphy J, Murphy JT, Sama A (2007) Qunisy trainer. J. Laryngol. Otol., 121:1194-6.
16. Alasmari *N* (2017) Causes and Treatment of Tonsillitis the Egyptian Journal of Hospital Medicine, 69 (8):2975-2980
17. Farooqi IA, Akram T, Zaka M (2017) Incidence and empiric use of antibiotics therapy for tonsillitis in children.

International Journal of Applied Research, 3(12): 323-327.

18. Mahajan GD, MayurIngale (2017) Study of common bacterial isolates in acute tonsillitis in India. Indian Journal of Basic and Applied Medical Research – Otorhinolaryngology special 6 (2): 59-62
19. Spinks A, Glasziou PP, Del Mar CB (2013) Antibiotics for sore throat. Cochrane Database Syst. Rev., 511:CD000023.
20. Mahendrappa KBR (2010) Upper limit of normal antistreptolysin-O titre in healthy schoolchildren. Indian pediatr., 4 (7):629.
21. Manandhar A, Shah Y, Shrestha J (2013) Study on the Prevalence of Beta Haemolytic *Streptococcus* among School Children. Journal of nepalpaediatric society 33(1):45 – 47
22. Johnson DR, Kurlan R, Leckman J, Kaplan EL (2010) The human immune response to streptococcal extracellular antigens: Clinical, Diagnostic, and Potential Pathogenetic Implications Clinical infectious diseases, 50:481-90.
23. Collee J, Fraser AG, Marmian BP, Simmon SA (1996) Mackie and McCartney practical Medical microbiology .4thed.churchill cancer and living stone ,INC.USA
24. CockerillF,WiklerM,BushK,DudleyM,EliopoulosG,HardyD,HechtD,HindlerJ,PatelJ,PowellM,ThomsonR,TurnidgeJ,WeinsteinM, ZimmerB,FerraroM,SwensonJ(2010)Performance standards for antimicrobial susceptibility testing .Twentieth informational, 30:1-153.
25. Haffejee (1992) Streptococcal antibody tests: measurement of antistreptolysin O titer (ASOT). *Quarterly Journal of Medicine*, 84(305):641-658.
26. Al-Gebori ARQ (2007) Bacterial study of beta-hemolytic streptococci causing tonsillopharyngitis and correlation of its MIC and MBC with hemolysin production. M.Sc. Thesis, College of Medicine, Baghdad University, Iraq
27. Martin DR (2004) The laboratory diagnosis of Streptococcal pharyngitis. Infect .Dis., 83:75-84
28. Mitchel more IJ, Reilly PG, Hay AJ (2007) Recurrent tonsillitis .The Journal of laryngology and otology.91:911-31
29. Salman M, Shaikh Mahboob, Afzal J, Naila TM (2009) Umar Farooq Bacteriology Oftonsilar Surface and Core in Patients with Recurrent Tonsillitis, Undergoing Tonsillectomy. Medical channel, 6 (4):95-97.
30. Sarah Yousef Abd El Galil1ab, SabryAbd El Gawad1cd, Esteglal El Ateeq2a (2014) Isolation and Identification of Microorganisms Causing Tonsillitis among Children of Hail Region Isolation and Identification of Microorganisms Causing Tonsillitis among Children of Hail Region. International Journal of Health Sciences & Research (www.ijhsr.org) .125 (4):125-129.
31. Kurien M, Stanis A, Job A, Brahamadathan Thomas K (2000) Throat swab in the chronic tonsillitis: How reliable and valid is it? Singapore Med. J., 41 (7): 324-6.
32. A Agrawal, D Kumar, A Goyal, R Gupta, Suneel Bhooshan (2014) Bacteriological Evaluation and Their Antibiotic SensitivityPattern in Tonsillitis .IOSR Journal of dental and medical Sciences (IOSR-JDMS)e-ISSN: 2279-0853, p-ISSN: 2279-0861. 13 (3): Ver. V: 51-55.
33. Periwal KL, Gupta BK, Panwar RB, Khatri PC, Raja S, Gupta R (2006) Prevalence of rheumatic heart disease in school children in Bikaner: an echocardiographic study. Assoc Physicians India. 54:279-82.
34. Danchin MH, Rogers S, Kelpie L, Selvaraj G, Curtis N, Carlin JB, Nolan TM, Carapetis JR (2007) Burden of acute sore throat and group A streptococcal pharyngitis in school-aged children and their families in Australia. Pediatrics, 120 (5):950-7.
35. Manandhar A, Shah Y, Shrestha J (2013) Study on the Prevalence of Beta *Haemolytic Streptococcus*Among School Children. Journal of nepalpaediatric society .33(1):45-47.
36. Ella, Elijah Ekah, Okafor Rita Blessing (2015) Antistreptolysin O titer in comparism to positive blood culture in determimining the prevalence of group A*streptococcus* infection in selected patients in zaria Nigeria. International journal of microbiology, Genetics and Monocular Biology Research, 1-9.
37. Uc¸kay I, Ferry T, Stern R, Ambrosioni J, Gamulin A, Andrey D, Djordjevic M, Bernard L, Hoffmeyer P, Lew D (2009) Use of serum antistreptolysin O titers in

the microbial diagnosis of orthopedic infections. International journal of infectious diseases, 13: 421-424.

38. Johnson DR, Kurlan R, Leckman J, Kaplan EL (2010) The Human Immune Response to Streptococcal Extracellular Antigens: Clinical, Diagnostic, and Potential Pathogenetic Implications .Clinical infectious diseases.50:481-90.

*Corresponding Author Email: *frialabd@yahoo.com*

