ZITA MEDICAL MANAGEMENT

www.hsmc.gr/en/pharmakeftiki-journal/

PHARMAKEFTIKI, 37, 2S, 2025 | 177-180 RESEARCH

https://doi.org/10.60988/p.v37i2S.169

The correlation between some immunological parameters and causative agent in patients with otitis media

Azhar Althahab[1,*], Khulood Abdul-Majeed[2], Wasan Hameed[3], Wurood Hathal[4]

[1]Department of Biology-Microbiology, College of Science, University of Babylon, Hillah, Iraq
[2]Department of Community Health Techniques, Babylon Technical Institute, Al-Furat Al-Awsat Technical University, Hillah, Iraq
[3]Department of Microbiology, College of Medicine, University of Kufa, Kufa, Iraq
[4]Department of Laboratory Techniques, College of Health and Medical Techniques, Al-Mustaqbal University, Hillah, Iraq

KEY WORDS:
otitis media; bacteria; CRP; IL-6; IL-10

ARTICLE INFO:
Received: October 14, 2024
Revised: January 01, 2025
Accepted: January 14, 2025
Available online: October 10, 2025

*** CORRESPONDING AUTHOR:**
Azhar Althahab, Department of Biology-Microbiology, College of Science, University of Babylon, Hillah, Iraq; e-mail: azharammran@yahoo.com

ABSTRACT

Otitis media, an inflammation of the middle ear, is a common illness in both childhood and adulthood. It arises due to various factors, primarily microbiological and immunological. This study aimed at investigating the microorganisms and fungi associated with otitis media in Iraqi patients, as well as to examine the correlation between causative agents (bacteria and fungi). The study was conducted between November 2017 and April 2018 at the Al-Hilla Teaching Hospital in Babylon City. A total of 90 pus and blood samples were collected from patients aged 1–80 years with otitis media. Samples were taken from discharging ears using sterile transport medium swabs. Of the 90 samples, 86 (95.6%) tested positive for microbial agents, while 4 cases (4.4%) had no identifiable causative agent. Among the infected individuals, 57 were male (63.3%) and 33 were female (36.7%). Samples were analysed in the microbiology laboratory in order to identify microbial species responsible for otitis media. The highest infection rates were observed in the age groups 1–20 years (34.4%) and 41–60 years (32.2%). Immunological parameters from the patients' sera revealed significant differences compared to those of the control group (N=20). While C-reactive protein levels showed no significant difference between patients and controls, serum interleukin-10 concentrations were found to be significantly elevated in patients with bacterial infections.

PHARMAKEFTIKI, 37, 2S, 2025 | 177-180

1. Introduction

Otitis media (OM) is an inflammation of the middle ear, with acute OM representing its acute manifestation. The diagnosis of acute OM should be based on the presence of acute symptoms and middle ear effusion. It is a multifactorial disease, influenced by environmental, bacteriological, host, immunological, and genetic factors[1]. OM is characterized as an inflammatory response to either acute or persistent stimuli, typified by the accumulation of both cellular and chemical mediators in the middle ear. These mediators (glycoproteins, peptides, cytokines, prostaglandins, leukotrienes, and others) play roles in the inflammatory process through mechanisms such as changes in vascular permeability and chemotaxis. OM is frequently caused by bacteria; however, sterile cases are not uncommon. Pathogens typically enter the middle ear from the nasopharynx *via* the Eustachian tube.

In bacterial OM, keratinocytes can produce various soluble inflammatory mediators in response to injury, independent of immune cells. These include interleukin-1 (IL-1), -6 (IL-6), and -8 (IL-8)[2]. IL-6 is a proinflammatory cytokine that plays a key role in the acute phase reaction, including the induction of C-reactive protein (CRP), which is predominantly synthesized by hepatocytes under the transcriptional control of IL-6. IL-6 is considered a sensitive and reliable marker of neonatal bacterial infection[3]. Interleukin-10 (IL-10), a critical immunoregulatory cytokine, is also involved in modulating the middle ear's inflammatory response to bacterial infection[4].

This study aimed at investigating the microorganisms and fungi associated with OM in Iraqi patients, as well as to examine the correlation between causative agents (bacteria and fungi).

2. Methodology

Sterile cotton swabs and gel tubes were used in order to collect ear exudate samples from a total of 90 patients diagnosed with OM. These patients attended private surgical clinics during the five-month period from November 2017 to the end of April 2018, at the ENT Department of the Al-Hilla Teaching Hospital in Babylon City. Our study's control group consisted of 20 healthy and randomly selected volunteers. All collected samples were directly cultured on the following media: (i) blood agar, MacConkey agar, chocolate agar, and mannitol salt agar (in which the cultures were incubated at 37°C, for 24–48 h), and (ii) potato dextrose agar (in which the cultures were incubated at 25°C, for 5–7 days). Direct smears were prepared from samples before and after culturing. Gram staining was performed in order to differentiate Gram-positive and Gram-negative bacteria. The serum levels of CRP, IL-6, and IL-10 were determined using ELISA kits. Data were analysed using Microsoft Excel (2010) and SPSS version 23.

3. Results and Discussion

Out of 90 samples, 86 yielded positive microbial cultures, while 4 showed no growth after 48 h. These 4 cases may involve other types of causative agents (such as viruses, parasites, *Chlamydia*, or *Mycoplasma*), which require more specialized detection techniques[5].

Bacterial infection was the predominant cause (62.2%) of OM, followed by mixed infections involving both fungi and bacteria (28.9%). Cases attributed solely to fungi or unknown agents accounted for 4.4%. A high proportion of the causative organisms of OM were Gram-positive bacteria (69 isolates; 46%); a finding consistent with previous findings[6].

Among the positive OM cases, 57 were male (63.3%) and 33 female (36.7%). The age groups 1–20 years and 41–60 years exhibited the highest infection rates, 34.4% and 32.2% respectively, while individuals aged 61–80 years exhibited the lowest infection rate (10%).

Serum CRP levels (measured in mg/L) were compared between the OM infection and the control groups by using one-way ANOVA with a significance threshold of $p<0.05$. The mean CRP levels were significantly higher in the bacterial (1.61 ± 3.88 mg/L), fungal (1.5 ± 3.0 mg/L), and unknown (1.5 ± 3.0 mg/L) cases compared to those of the control group

PHARMAKEFTIKI, 37, 2S, 2025 | 177-180

Table 1. Correlations among the serum C-reactive protein (CRP), interleukin-6 (IL-6), and interleukin-10 (IL-10) levels in Iraqi otitis media patients. Note: *, signifies a statistically significant correlation at p<0.05.

Pearson correlation coefficient (*r*)	IL-6 levels	IL-10 levels	CRP levels
IL-6 levels	1		
IL-10 levels	-0.078	1	
CRP levels	0.256 *	-0.129	1

(0.10 ± 0.00 mg/L). However, the mixed OM infection group did not exhibit a significant increase in CRP levels (0.92 ± 2.20 mg/L) when compared to those of the control group. The least significant difference (LSD) value was 0.97.

Serum IL-6 levels were found to be significantly elevated (p<0.05) in both the unknown cause case (157.73 ± 46.32 pg/mL) and the bacterial case (144.63 ± 148.78 pg/mL) groups, compared to those of the control group (53.53 ± 11.01 pg/mL). No significant increase was noted in the mixed OM infection (119.69 ± 145.82 pg/mL) or fungal case (101.02 ± 27.67 pg/mL) groups. The LSD value was 70.57.

As far as the serum IL-10 levels are concerned, only the bacterial case group exhibited a statistically significant increase (72.1 ± 22.89 pg/mL) compared to those of the control group (57.69 ± 2.97 pg/mL). Other groups – fungal case (71.35 ± 14.03 pg/mL), mixed case (67.07 ± 11.08 pg/mL), and unknown cause case (67.31 ± 4.97 pg/mL) groups – did not exhibit significant differences in terms of their IL-10 levels when compared to those of the control group. The LSD value was 13.70.

The correlations between the serum levels of the aforementioned parameters are presented in Table 1. Our findings suggest that serum CRP levels have a strong positive correlation with those of IL-6 in OM, thereby reinforcing their diagnostic value in distinguishing OM patients from healthy individuals.

Variations in infection type may be influenced by environmental conditions, geographic factors, and individual health status. Mixed infections likely result from a primary bacterial infection, followed by secondary fungal colonization. Diagnosing the initial *versus* the secondary agent in such cases remains complex, as supported by previous findings[6].

The present study has revealed significant age-related variations in OM incidence, with highest infection rates observed in the 1–20 and the 41–60 year groups (34.4% and 32.2%, respectively). This may be attributed to several predisposing factors in these age ranges, including smoking, diet, environmental exposures, immature immune systems (particularly in children), and weaker physical constitutions[7].

In contrast to a previous study[8] that has identified the highest OM infection rates in children aged 6–12 months, the current study has found a marked sex-based difference, with males (63.3%) being more affected than females (36.7%). This diverges from earlier studies that have reported no significant sex differences, largely because their cohorts focused on children. The higher male incidence in this study may reflect greater exposure to environmental stressors[9].

Finally, the acute stage of an OM infection appears to correlate with elevated CRP levels, especially in the case of bacterial OM. This supports the view that bacterial pathogens are more potent in inducing CRP production than fungal or other / unidentified causes[10].

4. Conclusion

The immunological assessment of Iraqi OM patients has demonstrated significant differences compared to that of controls. Although the serum CRP levels did not reveal significant differences across all OM infection

PHARMAKEFTIKI, 37, 2S, 2025 | 177-180

types, the serum IL-6 levels were found to be markedly elevated in OM patients, and the serum IL-10 levels were found to be significantly increased in those OM patients with bacterial infections, as compared to the respective markers' levels of the control group.

Acknowledgements

The authors wish to express their sincere gratitude to the College of Science of the University of Babylon and to its Advanced Microbiology Laboratory for their valuable support.

Conflicts of interest

None exist.

ORCIDs

0009-0009-4664-8359 (A. Althahab); 0000-0003-2487-3706 (K. Abdul-Majeed); 0000-0002-9092-5116 (W. Hameed); 0009-0002-1048-2325 (W. Hathal)

References

1. Rye M.S., Blackwell J.M., Jamieson S.E. Genetic susceptibility to otitis media in childhood. *Laryngoscope* 122(3), 665–675, 2012. DOI: 10.1002/lary.22506
2. Nofal K., Al Kwatly K. Serum interleukin-6 and C-reactive protein in bacterial otitis media patients in Damascus city. *J. Chem. Pharm. Res.* 7(7), 403–408, 2015.
3. Heikkinen T., Ghaffar F., Okorodudu A.O., Chonmaitree T. Serum interleukin-6 in bacterial and non-bacterial acute otitis media. *Pediatrics* 102(2pt1), 296–299, 1998. DOI: 10.1542/peds.102.2.296
4. Smirnova M.G., Birchall J.P., Pearson J.P. The immunoregulatory and allergy-associated cytokines in the aetiology of the otitis media with effusion. *Mediators Inflamm.* 13(2), 75–88, 2004. DOI: 10.1080/09629350410001688477
5. Block S.L., Hammerschlag M.R., Hedrick J., Tyler R., Smith A., Roblin P., *et al. Chlamydia pneumoniae* in acute otitis media. *Pediatr. Infect. Dis. J.* 16(9), 858–862, 1997. DOI: 10.1097/00006454-199709000-00008
6. Mohammed N.J., Al-Thahab A. Microbial profile from patients with otitis media. *Int. J. Pharm. Res.* 11(1), 483–487, 2019.
7. DeAntonio R., Yarzabal J.P., Cruz J.P., Schmidt J.E., Kleijnen J. Epidemiology of otitis media in children from developing countries: a systematic review. *Int. J. Pediatr. Otorhinolaryngol.* 85, 65–74, 2016. DOI: 10.1016/j.ijporl.2016.03.032
8. Paradise J.L., Rockette H.E., Colborn D.K., Bernard B.S., Smith C.G., Kurs-Lasky M., *et al.* Otitis media in 2253 Pittsburgh-area infants: prevalence and risk factors during the first two years of life. *Pediatrics* 99(3), 318–333, 1997. DOI: 10.1542/peds.99.3.318
9. Kumar R., Srivastava P., Sharma M., Rishi S., Nirwan P.S., Hemwani K., *et al.* Isolation and antimicrobial sensitivity profile of bacterial agents in chronic suppurative otitis media patients at NIMS Hospital, Jaipur. *Int. J. Pharm. Biol. Sci.* 3(4), 265–269, 2013.
10. Tejani N.R., Chonmaitree T., Rassin D.K., Howie V.M., Owen M.J., Goldman A.S. Use of C-reactive protein in differentiation between acute bacterial and viral otitis media. *Pediatrics* 95(5), 664–669, 1995.

HOW TO CITE:
Althahab A., Abdul-Majeed K., Hameed W., Hathal W. The correlation between some immunological parameters and causative agent in patients with otitis media. *Pharmakeftiki* 37(2s), 177-180, 2025. https://doi.org/10.60988/p.v37i2S.169