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Evaluation of Chemokine Ligand 4 and Matrix Metalloproteinase-2 as Inflammatory Biomarkers in the Serum of Patients with Cataracts

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***Abstract*—Cataracts are a progressive health issue with significant implications for individuals and public health systems. They are considered a critical global health issue, particularly affecting older adults. MMP-2 and CCL4 are both chemokines that are known to play a critical role in inflammation and tissue remodeling. The collected data were analysed by using SPSS and visualization tools. Our results indicate a significant correlation between MMP-2 and CCL4 among cataract patients. There is a substantial relationship between MMP-2 and other factors, such as gender and residency state, unlike CCL4. No significant correlation exists between the two serum biomarkers (MMP-2 and CCL4) and smoking cataract patients. By exploring the expression of MMP-2 and CCL4, this study aims to enhance cataract pathophysiology and identify inflammatory biomarkers associated with cataract progression. measuring serum levels of MMP-2 and CCL4 in cataract patients to assess their role in tissue remodeling, inflammation, and post-operative complications.**

***Keywords*— Cataract, MMP-2, CCL4, Eye diseases, Ocular inflammation.**

I. INTRODUCTION

Cataracts are a progressive health issue with significant implications for individuals and public health systems. They are considered an important global health issue, particularly affecting older adults [1- 2].

It is characterised by the opacification of the crystalline lens [3]. Primarily due to aging [4]. Moreover, it is influenced by other factors like smoking [5] and gender [6], drug induced cataract(corticosteroid) [7].

Moreover, the involvement of oxidative stress in the development of cataract [8] is well established.

MMP-2 constitutes a family of zinc-dependent endopeptidases, which are secreted mainly by myofibroblasts, cardiomyocytes, and fibroblasts, and are also known as gelatinase [9], MMP-2 demonstrated a notable correlation with cataracts, as evidenced by a study conducted by Lin, Y.F., which revealed elevated levels of MMP-2 in the aqueous humour of cataract patients compared to healthy controls [10]. A study conducted by Ying Chen in 2021 found that MMP-2 and other cytokines were significantly higher in patients with Acute Primary Angle Closure [11]. Likewise, CCL4 (formerly known as macrophage inflammatory protein [MIP]-1β) is a small cytokine that belongs to the cc chemokine subfamily. It acts as a chemoattractant for natural killer (NK) cells, monocytes, and various other immune cells in sites of inflammation or damage [12].

The precise role of inflammation in the aetiology of cataracts remains to be elucidated [13]; however, a study by Jabs et al. (2023) examined the correlation between systemic inflammatory biomarkers, including CCL4, and ocular conditions in individuals with AIDS. This investigation revealed that elevated plasma levels of multiple systemic inflammatory markers were associated with a reduced risk of cataracts within this population. This observation suggests a potential link between elevated systemic inflammation, as indicated by increased CCL4 levels, and a reduced incidence of cataract formation in individuals with AIDS [14].

Another study conducted by Adamus, G.2001 revealed that CCL4 plays a key role in attracting inflammatory cells to the eye, and has a role in disease progression [15].

The existing literature on the subject indicates a paucity of studies investigating the association between serum levels of MMP-2 and CCL4 and the development of cataracts.

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II. MATERIALS AND METHODS

A. Study Design and Participants

Acase-control study was conducted at the ophthalmology department of Al-Nasiriyah Teaching Hospital in Thi-Qar governorate, Iraq, on patients diagnosed with cataracts between September 2024 and January 2025.

The cataract group consists of 93 males and 107 females, while the control group consists of 100 males and 100 females.

B. Ethical Approval

Ethical approval was obtained from the following institutions and individuals: the University of Basrah, College of Medicine, Department of Microbiology, Al-Nasiriyah Teaching Hospital, the primary health care centre manager, and the patients.

All information concerning the study was clearly explained to the patients.

C. Sample Collection and Processing

Two milliliters of venous blood were obtained using a sterile disposable syringe for each participant (patient and control), as in [16] Then, they were transferred to a sterile gel tube to measure MMP-2 and CCL4.

D. Measurement of the MMP-2 and CCL4

Serum was analysed to determine the total MMP-2 and CCL4 concentration by using sandwich ELISA kits for MMP-2 and CCL4, using a commercially available kit (MMP-2 ELISA Kit (cat. No. SL115Hu), SunLong Biotech Company (China), and the CCL4 ELISA Kit (cat. No. SL1124Hu). SunLong Biotech Company (China)).

E. Statistical Analysis

The collected data was statistically analyzed using SPSS and other visualization tools like DTA tab and Geneious.

III. RESULTS

A. Comparison between Two Markers (MMP-2 and CCL4) among Patients and the Control Group

Table (1) shows that MMP-2 and CCL4 were significantly higher in patients with cataract than in the control group, showing that the mean concentration of the two markers, respectively, was (1387.019± 158.17 pg/mL, 18.348± 5.21 pg/mL) in patients. In contrast, the mean in the control group for the two markers was equal to (1195.199 ± 175.89 pg/mL, 16.897± 1.63 pg/mL). The p-value for the two markers = p≤ 0.01, so it's statistically significant.

B. Comparison between Two Markers (MMP-2 and CCL4) According to Gender among Patients and Control Groups

Table (2) shows that MMP-2 was significantly higher in the patient group among males and females than in the control group; the mean for males in the patient group was equal to 1390.166± 183.665 pg/mL, and in females it was equal to 1384.876± 138.963 pg/mL. While CCL4 showed no significant difference, the mean for males was 18.946±7.322g/mL, and for females, the mean was equal to 1717.941±2.996g/mL.

Table (1): Serum concentration of MMP-2 and CCL4 among patients and control group

	Patient (Mean±SD)pg/mL	**No.**	**Control (Mean±SD)pg/mL**	**No.**	**P -value**
MMP-2**	1387.019 ± 158.17	200	1195.199 ± 175.89	200	**=p≤0.01
CCL4**	18.348 ± 5.21	200	16.897 ± 1.63	200	**=p≤0.01

Table (2) Serum concentration of MMP-2 and CCL4 according to gender among patients and the controls group

	Male		Female		P Value
	Patients (Mean± SD)pg/mL	Control (Mean±SD)pg/mL	Patients (Mean±SD)pg/mL	Control (Mean±SD)pg/mL	
No.	81	96	119	104	
MMP-2*	1390.166 ± 183.665	1153.664 ± 180.396	1384.876 ± 138.963	1233.538 ± 163.302	*=p≤0.05 p=0.026
CCL4ns	18.946 ± 7.322	16.850 +± 1.961	17.941 ± 2.996	16.940 ± 1.255	No significant t=0.24

C. Relation between MMP-2 and Smoking and Nonsmoking among Patients and Control Groups

The result of this table (Table3) shows that there's no significant relationship between MMP-2 among smokers and non-smokers. It's statistically not substantial, p-value was of 0.23, the mean for smokers in patient group was equal to 1339.608 ±181.355 pg/mL, and for non-smokers was equal to 1409.330 ±141.291 pg/mLfor the control group, the mean for smokers was 1120.896±29.269 pg/mL, and for non-smokers 1195.949±176.612 pg/mL.

Table (3) serum concentration of MMP-2 and its relation with smoking and non-smoking among patients and control groups

	Smokers		Non-smokers		P Value
	Patients (Mean± SD)pg/mL	Control (Mean±SD)pg/mL	Patients (Mean±SD)pg/mL	Control (Mean±SD)pg/mL	
No.	64	2	136	198	
MMP-2ns	1339.608 ± 181.355	1120.896 ± 29.269	1409.330 ± 141.291	1195.949 ± 176.612	Smoking P=0.23 Ns = no significance

D. Relation between CCL4 and Smoking and Nonsmoking among Patients and Control Groups

(Table 4) shows no statistically significant correlation between the CCL4 marker and smokers-nonsmokers among the patient and control groups, with a p-value of 0.97. The mean for smokers and nonsmokers among patients,

respectively, was 18.552±6.068 pg/mL, 18.252 ±4.772 pg/mL, while for the control group, the mean for smokers and non-smokers, respectively, was equal to 16.696±2.398 pg/mL, and16.899±1.6288 pg/mL.

E. ROC Curve

Figure (1) represents the ROC curve analysis for MMP-2 and CCL4. The AUC (area under the curve) values indicate that MMP-2 (AUC=0.789) has good diagnostic accuracy, while CCL4 (AUC = 0.607) showed the weakest

F. MMP-2 Serum Biomarker Distribution: Assessing the Degree of Data Dispersion ROC Curve

Figure (2) shows MMP-2 serum marker levels display significant variability, with some extremely high values. In contrast, others are considerably low, indicating a wide dispersion in the dataset.

G. CCL4 Serum Biomarker Distribution: Assessing the Degree of Data Dispersion

Figure (3) shows that CCL4 serum marker levels display variability in this dataset, which is relatively moderate and evenly distributed. However, both groups show the same degree of variability; the patient group shows a slightly greater spread has been observed.

Table (4) Serum concentration of CCL4 and its relation with smoking and non-smoking among patients and control groups.

	Smokers		Non-smokers		P Value
	Patients (Mean±SD)pg/mL	Control (Mean±SD)pg/mL	Patients (Mean±SD)pg/mL	Control (Mean±SD)pg/mL	
No.	64	2	136	198	
CCL4^{ns}	18.552 ± 6.068	16.696 ± 2.398	18.252 ± 4.772	16.899 ± 1.628	Smoking P=0.97 Ns = no significance

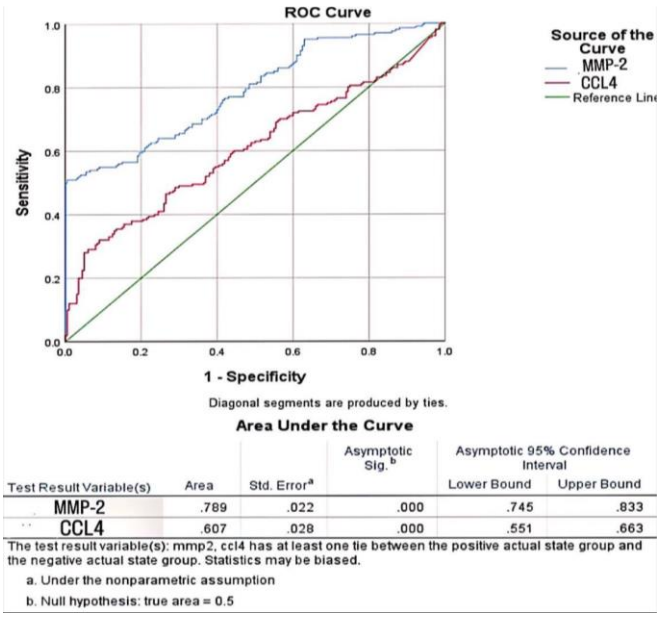

Fig. (1): ROC curve for the two markers (MMP-2 and CCL4). The blue curve represents the MMP-2 serum biomarker, and the red curve represents the CCL4 serum biomarker.

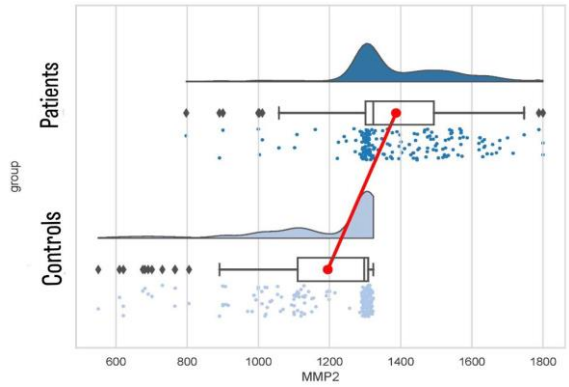

Fig. (2): The dispersion of the data of the MMP-2 serum biomarker among patients and controls.

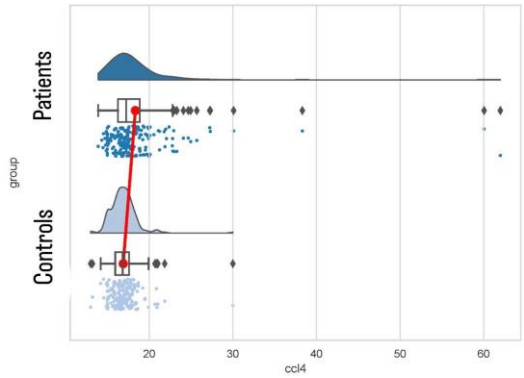

Fig. (3): The dispersion of the data on CCL4 serum biomarkers among patients and controls

IV. DISCUSSION

The results of this study show that the mean levels of the two markers (mmp-2 and CCL4) were highly significant in patients compared to control groups, pointing to their potential role in disease progression.

Excessive MMP-2 activity contributes to lens opacification; high levels of MMP-2 lead to basement membrane degradation and disrupt lens epithelial cell integrity.

Our findings agreed with Alapure, B.V., 2012 findings, which reveal that levels of MMP-2 were high in the serum of cases with steroid-induced posterior subcapsular cataract (PSC) [17]

This study also matched with Tamiya, S.2000, which found that the level of MMP-2 is associated with the development of cataract [18].

CCL4 was more significant in patients diagnosed with cataract than in those in the control group. This can be attributed to increased levels of MMP-2, which influence the expression of other cytokines like CCL4. There are limited studies that link CCL4 and cataracts.

A study conducted by Xinbo Gao in 2016 found that the CCL2 and CCL7 concentration levels are high after an acute glaucoma attack [19].

This study shows a statistically significant relationship between MMP-2 and sex in cataract patients. Although statistical significance was achieved, the absolute difference between sex groups remains small, indicating that gender may influence MMP-2 levels. This can be explained by

biological factors, including hormonal imbalance (estrogen–testosterone).

The relation between MMP-2 and gender remains unclear, with some studies suggesting there's no relation and exhibiting a hormonal effect.

This study did not match the result of Tayebjee, M.H. (2005), who found that there is no relation between gender and the level of MMP-2 [20]

The other marker (CCL4) shows no significant difference, with a p-value of 0.16. Studies for CCL4 are still limited.

MMP-2 shows no statistically significant relationship between the level of this marker and smoking, with a p-value of 0.23, which can be explained by the fact that the immune system regulates this marker to maintain homeostasis.

Another explanation is that smoking might influence other pro-inflammatory markers (such as IL-6 or CRP) more significantly than MMP-2.

CCL4 shows statistically no significant relationship between the level of this marker and smoking, with a p-value of 0.97. This can be explained by the fact that the immune system regulates this marker to maintain homeostasis, or this marker is more stable than others, so it cannot be affected by another factor, such as smoking.

There is a paucity of studies linking MMP-2 and CCL4 serum biomarkers to smoking.

The findings of the ROC curve suggest that MMP-2 could serve as a more reliable serum biomarker compared to CCL4 in the context of cataract detection.

In the present study, it was shown that the serum level of MMP-2 varies from one individual to another, varying from extremely high levels to low levels, several factors can be behind this distribution increase, including the severity of the cataract. At this stage, it was unclear whether the cataract was a result of trauma or the aging process. Other factors include lifestyle and other underlying health conditions.

We noticed the distribution of the CCL4 serum biomarker were slightly higher in the patient group; several factors can be behind this, including environmental, underlying health conditions, or can be influenced by elevated levels of MMP-2.

V. CONCLUSIONS

In conclusion, MMP-2 and CCL4 were highly significant in cataract patients compared to healthy individual. Also, our study shows that unlike CCL4, MMP-2 showed highly significant relationship with gender differences. Furthermore, it shows no considerable correlation between MMP-2 and CCL4 serum biomarker and smoking patients with cataract.

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CONFLICT OF INTEREST

Authors declare that they have no conflict of interest.

REFERENCES

[1]A. Pollreisz and U. Schmidt-Erfurth, "Diabetic cataract—pathogenesis, epidemiology, and treatment.," *Journal of ophthalmology*, vol. 2010, no. 1, p. 608751, 2010.

[2]S. Z. Al-Asadi, W. Nazar, and S. Moayeed, "Intraoperative Assessment of Interleukin-2, Interleukin-6, and Tumor Necrosis Factor-* in Aqueous Humor of Diabetic and Non-diabetic Patients Undergoing Phacoemulsification Surgery.," *Iraqi National Journal of Medicine*, vol. 2, no. 2.

[3]G. Davis, "The evolution of cataract surgery.," *Missouri medicine*, vol. 113, no. 1, p. 58, 2016.

[4]R. Fang *et al.*, "Global, regional, national burden, and gender disparity of cataract: findings from the global burden of disease study 2019.," *BMC Public Health*, vol. 22, no. 1, p. 2068, 2022.

[5]R. Hiller *et al.*, "Cigarette smoking, and the risk of development of lens opacities: the Framingham studies.," *Archives of ophthalmology*, vol. 115, no. 9, pp. 1113-1118, 1997.

[6]V. L. E. G. o. t. G. B. o. D. Study, "Global estimates on the number of people blind or visually impaired by cataract: a meta-analysis from 2000 to 2020.," *Eye*, vol. 38, no. 11, p. 2156, 2024.

[7]P. A. Asbell, I. Dualan, J. Mindel, D. Brocks, M. Ahmad, and S. Epstein, "Age-related cataract.," *The Lancet*, vol. 365, no. 9459, pp. 599-609, 2005.

[8]H. Lesiewska, A. Woźniak, P. Reisner, K. Czosnyka, J. Stachura, and G. Malukiewicz, "Is cataract in patients under 60 years associated with oxidative stress?." *Biomedicines*, vol. 11, no. 5, p. 1286, 2023.

[9]Y. Ma *et al.*, "Cross talk between inflammation, and extracellular matrix following myocardial infarction.," in *Inflammation in heart failure*: Elsevier, 2015, pp. 67-79.

[10]Y. F. Lin, J. X. Xie, and X. L. Chen, "Changes in PEDF, MMP-2, and TGF-β2 levels in the aqueous humor of cataract patients, and their correlation with disease severity.," *Clinics*, vol. 79, p. 100402, 2024.

[11]Y. Chen, Yan, H., Li, G. and Zhang, Y., , "Higher TGF-β1, TGF-β2, MMP-2, and TIMP-1 levels in the aqueous humor of patients with acute primary angle closure," *Ophthalmic Research*, vol. 64, no. 1, pp. 62-67, 2021.

[12]G.-N. Hu *et al.*, "Correlation between CCL4 gene polymorphisms, and clinical aspects of breast cancer.," *International journal of medical sciences*, vol. 15, no. 11, p. 1179, 2018.

[13]S. Moayeed, W. Nazar, and S. Z. ALAsadi, "Analysis of TNF, IL2, and IL6 in the serum, and aqueous humour of patient with cataract in Ba-sra/Iraq.," *International Journal of Scientific Engineering Research,* vol. 7, no. 9, pp. 648-651, 2016.

[14]D. A. Jabs, M. F. Schneider, J. W. Pak, and P. Hunt, "Systemic inflammatory biomarkers are associated with increased risk of age-related macular degeneration but a decreased risk of cataract in people with AIDS.," *Investigative Ophthalmology Visual Science,* vol. 64, no. 8, pp. 534-534, 2023.

[15]G. Adamus, M. Manczak, and M. Machnicki, "Expression of CC chemokines and their receptors in the eye in autoimmune anterior uveitis associated with EAE.," *Investigative Ophthalmology & Visual Science,* vol. 42, no. 12, pp. 2894-2903, 2001.

[16]W. Ibraheim, H. Jasim, and A. Abdullah, "The impact of interferon-gamma level on the health status of patients with sickle cell disease in Basrah.," *Вопросы гематологии/онкологии и иммунопатологии в педиатрии,* vol. 22, no. 3, pp. 65-67, 2023.

[17]B. V. Alapure, M. R. Praveen, D. U. Gajjar, A. R. Vasavada, T. J. Parmar, and A. I. Arora, "Matrix metalloproteinase-2, and-9 activities in the human lens epithelial cells, and serum of steroid induced posterior subcapsular cataracts.," *Molecular Vision,* vol. 18, p. 64, 2012.

[18]S. Tamiya, I. M. Wormstone, J. M. Marcantonio, J. Gavrilovic, and G. Duncan, "Induction of matrix metalloproteinases 2, and 9 following stress to the lens.," *Experimental eye research,* vol. 71, no. 6, pp. 591-597, 2000.

[19]H. W. Gao X, Zhang X, Du S, Wang J, Wang W, Zhou M, Chen S, Li X, Jonas JB., "Chemokine (C-C motif) ligand 2 and chemokine (C-C motif) ligand 7 in angle-closure glaucoma," *Acta Ophthalmologica,* 2016, doi: doi: 10.1111/aos.12696.

[20]M. H. Tayebjee, G. Y. Lip, A. D. Blann, and R. MacFadyen, "Effects of age, gender, ethnicity, diurnal variation, and exercise on circulating levels of matrix metalloproteinases (MMP)-2, and-9, and their inhibitors, tissue inhibitors of matrix metalloproteinases (TIMP)-1, and-2.," *Thrombosis research,* vol. 115, no. 3, pp. 205-210, 2005.