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Original Research

Evaluation of Neutrophil-to-Lymphocyte Ratio and Mean Platelet Volume in Patients with Active and Inactive Thyroid Orbitopathy

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Abstract

Objectives: The aim of this study was to evaluate the neutrophil-to-lymphocyte ratio (NLR) and the mean platelet volume (MPV) in patients with active thyroid orbitopathy (TO) and to compare it with that of both healthy subjects and patients with inactive TO.

Methods: Twenty patients with active TO (Group 1), 25 patients with inactive TO (Group 2), and 35 age- and sex-matched healthy subjects (Group 3) were included in this study. Patients with other systemic and ocular diseases, patients with a history of intraocular or orbital surgery, and patients using systemic drugs were excluded. The VISA (vision, inflammation, strabismus, appearance) classification scheme was used to discriminate between active and inactive TO. The neutrophil and lymphocyte counts and the MPV of all participants were recorded. The NLR was calculated by dividing the neutrophil count by the lymphocyte count, and the result was compared between groups. The optimal cut-off value was determined for NLR and MPV and the data were compared with a one-way analysis of variance test and the 'Bonferroni post-test.

Results: The mean age was 45.4±13.4, 41.0±13.7, and 42.6±14.4 years in Group 1, 2, and 3, respectively (p=0.68). The NLR was 2.11 in Group 1, 1.56 in Group 2, and 1.47 in Group 3 (p=0.03). The 'Bonferroni post-test revealed a difference between Group 1 and Group 2 (p=0.01) and between Group 1 and Group 3 (p<0.001). The MPV was 10.76 fL in Group 1, 9.94 fL in Group 2, and 8.19 fL in Group 3 (p<0.001). The results of the 'Bonferroni post-test showed a difference between Group 1 and Group 2 (p=0.04), between Group 1 and Group 3 (p<0.001), and between Group 2 and Group 3 (p<0.001). The mean cut-off value obtained from receiver operating characteristic (ROC) analysis of NLR was 1.69 (sensitivity: 72%; specificity: 66%). The mean cut-off value obtained from ROC analysis of MPV was 9.95 (sensitivity: 63%; specificity: 66%).

Conclusion: High NLR and MPV values may be indicative of active inflammation in patients with TO."

Keywords: Mean platelet volume; neutrophil-to-lymphocyte ratio; thyroid-associated orbitopathy.

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Thyroid orbitopathy (TO) is an autoimmune inflammatory disease that can threaten vision, cause cosmetic problems, and can eventually lead to serious impairment of quality of life in patients.[1, 2] It is generally seen in patients with Graves' disease (GD), but it is also occasionally observed in Hashimoto's thyroiditis.[3] Although the pathogenesis of TO is not fully understood, the autoimmune characteristics have been generally accepted.[4]

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Determination of the clinical stage and severity of the disease during the examination of patients with TO is important for the selection of the treatment regimen to keep the disease under control. TO has 2 stages: the first stage of the disease is a progressive, active stage, characterized by active inflammation and orbital tissue modeling, which is generally followed by an inactive phase, in which clinical findings are stabilized. Most of the clinical symptoms and signs of TO, such as retraction of the upper eyelid, conjunctival congestion, proptosis, restrictive strabismus, and diplopia, are observed during the active phase.[5]

The immune system plays a vital role in the control and progression of numerous diseases. It is possible to evaluate the response of the immune system in a disease state from a routine analysis of a blood sample. Neutrophils are the active component of inflammation, while lymphocytes are regulatory and protective components.[6] The neutrophil-to-lymphocyte ratio (NLR), calculated by dividing the number of neutrophils in a sample of peripheral blood by the lymphocyte count, is now used as a simple and inexpensive marker in the evaluation of inflammatory response in diabetes mellitus, Alzheimer's disease, lung-colorectal cancers, and renal disorders.[7, 8]

Mean platelet volume (MPV) is the measurement of the mean size of platelets in the blood circulation, and it is also used as a simple and inexpensive parameter. An increased MPV value has been investigated in many diseases, and the potential use of MPV as a marker of systemic inflammation has been highlighted.

Only a few studies have investigated NLR and MPV in thyroid diseases. Some studies have indicated an increase in NLR in thyroid cancers, as in other cancers.[9-12] However, the association between TO and whole blood count has not yet been studied. Though various scoring systems are used to discriminate between active and inactive TO, the whole blood count may be important in patient follow-up. This study was an evaluation of the NLR and MPV as markers of inflammation in patients with active TO, and a comparison of NLR and MPV values in inactive TO patients and healthy individuals.

Methods

A total of 80 individuals were included in this retrospective study. The records of 20 active TO patients (Group 1), 25 inactive TO patients (Group 2), and 35 age- and gender-matched controls (Group 3) examined and diagnosed at the ophthalmology clinic of the Şişli Education and Research Hospital were retrospectively investigated after the study was approved by the ethics committee. The study was performed in compliance with the principles of the Helsinki Declaration.

Patients who were not in the age group of 20 to 65 years, those with other systemic disease or eye disease, those who had undergone intraocular surgery, patients with glaucoma or dry eye disease not related to thyroid disease, and those who had received systemic or local treatment or had undergone surgery for TO were not included in the study. Antithyroid drugs were used by 85% of the patients, and all were found to be euthyroid in an ophthalmological examination. None of the patients had a history of smoking. The best corrected visual acuity, intraocular pressure of the patients as measured with Goldmann aplanation tonometry, and detailed biomicroscopic and dilated fundus examination findings were retrospectively studied and recorded. Hertel exophthalmometer measurements over 21 mm were evaluated as indicative of proptosis. Axial-coronal orbital computed tomography (CT) and magnetic resonance imaging (MRI) obtained during the initial ophthalmologic examination were retrospectively analyzed, and the presence of extraocular muscle hypertrophy due to TO was evaluated, as well as the presence of another lesion occupying the intraorbital space that could cause proptosis. Activity scoring of TO was performed using the VISA (vision, inflammation, strabismus, appearance) classification. Each parameter was scored separately. Deterioration of any parameter was evaluated as active inflammation.[13]

During the first ophthalmological control, blood samples were drawn from an antecubital vein, placed in tubes containing dipotassium ethylenediaminetetraacetic acid, and analyzed with an automated blood counter (Beckman Coulter, Inc., Brea, CA, USA). Neutrophil, lymphocyte, and platelet counts and MPV were recorded. The NLR was calculated for each patient.

Statistical Analysis

PASW Statistics for Windows, Version 18.0 (SPSS Inc., Chicago, IL, USA) was used to analyze the data. Normality of data distribution was determined using the Kolmogorov-Smirnov test and a chi-square test. Significance of intergroup differences was determined using one-way analysis of variance and the 'Bonferroni post-test was applied to confirm the significance. $P<0.05$ was accepted as the level of significance. Receiver operating characteristic (ROC) curve analysis was used to ascertain the diagnostic value of NLR and MPV in TO.

Results

The mean age of the patients in Group 1 (12 women, 8 men), Group 2 (15 women, 10 men), and Group 3 (22 female, 13 men) was 45.14±13.4, 41.0±13.7, and 42.6±14.4 years, respectively. No significant intergroup difference

Table 1. Demographic characteristics and mean blood parameters of all patients

Variables	Group 1 (ATO) n=20 Mean±SD	Group 2 (ITO) n=25 Mean±SD	Group 3 (Control) n=35 Mean±SD	p	P*** (Groups 1&2)	P*** (Groups 1&3)	P*** (Groups 2&3)
Age (years)	45.45±13.47	41.00±13.70	42.60±14.44	0.68*			
Gender (female/male)	12:8	15:10	22:13	0.79**			
Neutrophil (10^9/L)	5.31±2.38	4.20±1.39	3.99±0.92	0.04*	0.05	0.09	1.00
Lymphocyte (10^9/L)	2.51±1.12	2.68±0.69	2.71±0.56	0.01*	1.00	0.01	0.37
Platelet (10^9/L)	289.18±37.94	252.69±46.95	241.71±70.82	0.07*			
MPV (fL)	10.76±0.70	9.94±1.39	8.19±0.81	<0.001*	0.04	<0.001	<0.001
NLR	2.11±1.27	1.56±0.71	1.47±092	0.03*	0.01	<0.001	0.33

ATO: Active thyroid ophthalmopathy; ITO: Inactive thyroid ophthalmopathy; MPV: Mean platelet volume; NLR: Neutrophil-to-lymphocyte ratio.
p<0.05 was considered statistically significant; *One-way analysis of variance; ***'Bonferroni post-test; **Chi-square test.

was detected with respect to age or gender (p=0.68 and p=0.79, respectively).

The neutrophil, lymphocyte, and platelet counts, and the MPV and NLR values of all of the patients are shown in Table 1. The mean neutrophil and lymphocyte counts differed significantly between groups (p=0.04 and p=0.01, respectively). The 'Bonferroni post-test demonstrated a significant intergroup difference between Groups 1 and 3 (p=0.01). The mean platelet count was similar between groups (p=0.07).

The mean MPV value of Group 1, Group 2, and Group 3 was 10.76±0.70, 9.94±1.39, and 8.19±0.81 fL, respectively (p=<0.001). The 'Bonferroni post-test revealed significant intergroup differences between Group 1 and Group 2 (p=0.04), Group 1 and Group 3 (p=0.001), and Group 2 and Group 3 (p=<0.001).

The mean NLR value of Group 1, Group 2, and Group 3 was 2.11±1.27, 1.56±0.71, and 1.47±0.92, respectively. There was a significant difference in the NLR between groups (p=0.03). The 'Bonferroni post-test indicated that Group 1 significantly differed from Group 2 (p=0.01) and Group 3 (p<0.001). In ROC analysis of NLR with a threshold value of 1.69, the sensitivity for defining active inflammation was 72% and the specificity was 66% (Fig. 1). In analysis of MPV with a threshold value of 9.95 for active inflammation, the sensitivity was 63% and the specificity was 66 % (Fig. 2).

Discussion

Graves' disease with associated hyperthyroidism is an autoimmune disease, which may result in pathologies in other organs due to increased blood levels of free thyroid hormones. In Graves' disease, apart from thyroid gland,

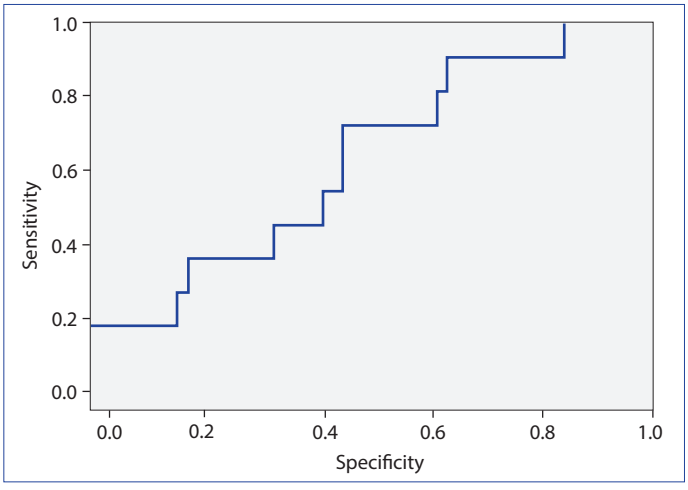

Figure 1. Receiver operator characteristic curve for the neutrophil-to-lymphocyte ratio of patients with thyroid orbitopathy with a cut-off point of 1.69.
(Area under the curve for the neutrophil-to-lymphocyte ratio was 0.630 with a cut-off value of 1.69).

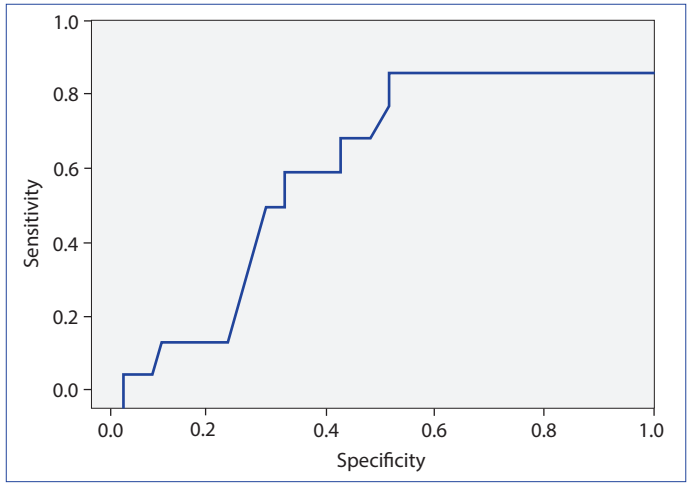

Figure 2. Receiver operator characteristic curve for the mean platelet volume in patients with thyroid orbitopathy with a cut-off point of 9.95.
(Area under the curve for the mean platelet volume was 0.642 with a cut-off point of 9.95).

most frequently the eye is affected.[14] Autoimmunity that develops against some prevalent antigens in the thyroid tissue and the eye orbit is thought to be the mechanism.[1] Thyroid-stimulating hormone (TSH) receptor antigens are the most well-known antigen; however recently, other antigens have also been identified.[4, 15, 16] Reactive T-lymphocytes recognize antigens in the thyroid and the eye orbit and invade the orbital and extraocular muscles, which leads to the release of various cytokines. These cytokines then stimulate CD8+T lymphocytes and beta cells, and reinforce the immune reaction.[17] The cytokines also stimulate the synthesis and release of fibroblasts, due to an increase in the production of glycosaminoglycans (GAGs). The GAGs retain water, and result in periorbital edema, proptosis, and edematous swelling of the extraocular muscles.[18] The orbital structure becomes enlarged, and retroperitoneal fat tissue expands secondary to the proliferation of fibroblasts.[19]

Orbital CT and MRI are important in the diagnosis and follow-up of TO. In some patients, orbital muscle involvement may be visualized on CT or MRI scans even though clinically detectable symptoms and signs are not observed. TO should be followed up regularly. Patient complaints increase during the active phase, when inflammation is a particularly dominant feature, and the disease is also more responsive to treatment during this phase. Therefore, determination of the disease phase should be performed with care, and the appropriate treatment should be provided at the optimal time. Numerous classifications have been developed to score TO activity. Most predominantly evaluate subjective patient complaints; more objective classifications are needed.

Inflammation in TO is actually systemic inflammation, rather than limited to the eye. Therefore, the disease should be evaluated not just from the ophthalmological perspective, but using systemic parameters. Various laboratory tests are already used to evaluate the response of the body to systemic inflammation. The level of C-reactive protein (CRP), which is an acute phase reactant, and the erythrocyte sedimentation rate (ESR) increase during the inflammatory process. However, they reflect only transient inflammatory activity, and they are expensive tests. The total leukocyte count provides a more sensitive assessment of the inflammatory state of the body, and it is less expensive to perform. The NLR is calculated by simply dividing the neutrophil count by the lymphocyte count. The NLR is an inexpensive and user-friendly method to evaluate systemic inflammation.[20] Activated neutrophils stimulate the release of proinflammatory cytokines and active T-lymphocytes, and play a role in the pathogenesis of many diseases, including inflammatory diseases.[21] An increased neutrophil count and a decreased lymphocyte count is associated with a poor prognosis.[22] Platelets also have an important role in inflammation and immune activity. The MPV is a parameter to evaluate thrombocytic function, and it can be easily estimated from the routinely performed whole blood count. Some studies have demonstrated a relationship between inflammatory disease and a high NLR and MPV.[23, 24]

The association between blood parameters and thyroid disease is not well known. Some studies have demonstrated a correlation between the NLR and several types of cancer, especially papillary thyroid cancer.[9-12] Koçer et al.[10] investigated the NLR in papillary thyroid cancer and various benign thyroid cancers, and detected a significantly higher NLR level in patients with papillary thyroid cancer. They suggested that the NLR might aid in the differentiation between benign and malignant thyroid diseases. On the other hand, Keskin et al.[11] found an elevated NLR in patients with euthyroid chronic autoimmune thyroiditis compared with a control group, which they associated with the presence of thyroid antibodies.

Although ocular diseases induce a local inflammatory response, rather than systemic inflammation, an association has been found between the NLR and several eye diseases, including age-related macular degeneration, retinal vein occlusion, nonarteritic anterior ischemic optic neuropathy, vernal keratoconjunctivitis, dry eye, and primary open angle glaucoma.[25-31] In order for the NLR to be significant in local inflammatory diseases, it must change the systemic blood parameters. Inflammation in TO is actually a result of systemic inflammation, and it may change blood parameters.

To the best of our knowledge, no literature study has investigated a correlation between whole blood count and TO. We detected significantly higher NLR and MPV values in active TO patients compared with those of both inactive TO patients and the control group. The MPV values in inactive TO patients were also significantly higher in comparison with the control group. These higher MPV values may be the result of persistent, subclinical systemic inflammation, even though the patients get involved in inactive phase of the disease. In addition to VISA classification, blood NLR and MPV values may help differentiate between the active and inactive phases of the disease.

Our study has some limitations. One of the most important is that we didn't measure well- defined inflammatory markers, such as tumor necrosis factor-alfa, interleukin 6, interleukin 1 beta, CRP, and ESR. Investigation of the correlations between these inflammatory markers and hematological parameters might be important in confirmation of our results. Other limitations of the study are its retrospective design and the relatively small number of patients. Perhaps prospective evaluation of blood parameters in both the

active and inactive phases of the disease in patients with active TO will be the subject of further studies.

Disclosures

Ethics Committee Approval: The study was approved by the Local Ethics Committee.

Peer-review: Externally peer-reviewed.

Conflict of Interest: None declared.

Authorship contributions: Concept – C.U.A., S.Y.S., P.K.; Design – C.U.A., S.Y.S., P.K.; Supervision – C.U.A., S.Y.S.; Materials – C.U.A., S.Y.S.; Data collection &/or processing – C.U.A., M.C., A.A.; Analysis and/or interpretation – C.U.A., M.C., P.K.; Literature search – M.C., A.A.; Writing – C.U.A, P.K.; Critical review – D.G., P.Y.

References

1. Bahn RS. Graves' ophthalmopathy. N Engl J Med 2010;362:726–38.
2. Ponto KA, Hommel G, Pitz S, Elflein H, Pfeiffer N, Kahaly GJ. Quality of life in a german graves orbitopathy population. Am J Ophthalmol 2011;152:483–490.e1. [CrossRef]
3. Lim NC, Sundar G, Amrith S, Lee KO. Thyroid eye disease: a Southeast Asian experience. Br J Ophthalmol 2015;99:512–8. [CrossRef]
4. McGregor AM. Has the target autoantigen for Graves' ophthalmopathy been found? Lancet 1998;352:595–6. [CrossRef]
5. Douglas RS, Gupta S. The pathophysiology of thyroid eye disease: implications for immunotherapy. Curr Opin Ophthalmol 2011;22:385–90. [CrossRef]
6. Bhutta H, Agha R, Wong J, Tang TY, Wilson YG, Walsh SR. Neutrophil-lymphocyte ratio predicts medium-term survival following elective major vascular surgery: a cross-sectional study. Vasc Endovascular Surg 2011;45:227–31. [CrossRef]
7. Imtiaz F, Shafique K, Mirza SS, Ayoob Z, Vart P, Rao S. Neutrophil lymphocyte ratio as a measure of systemic inflammation in prevalent chronic diseases in Asian population. Int Arch Med 2012;5:2.
8. Turkmen K, Guney I, Yerlikaya FH, Tonbul HZ. The relationship between neutrophil-to-lymphocyte ratio and inflammation in end-stage renal disease patients. Ren Fail 2012;34:155–9. [CrossRef]
9. Liu CL, Lee JJ, Liu TP, Chang YC, Hsu YC, Cheng SP. Blood neutrophil-to-lymphocyte ratio correlates with tumor size in patients with differentiated thyroid cancer. J Surg Oncol 2013;107:493–7.
10. Kocer D, Karakukcu C, Karaman H, Gokay F, Bayram F. May the neutrophil/lymphocyte ratio be a predictor in the differentiation of different thyroid disorders? Asian Pac J Cancer Prev 2015;16:3875–9. [CrossRef]
11. Keskin H, Kaya Y, Cadirci K, Kucur C, Ziypak E, Simsek E, et al. Elevated neutrophil-lymphocyte ratio in patients with euthyroid chronic autoimmune thyreotidis. Endocr Regul 2016;50:148–53.
12. Gong W, Yang S, Yang X, Guo F. Blood preoperative neutrophil-to-lymphocyte ratio is correlated with TNM stage in patients with papillary thyroid cancer. Clinics (Sao Paulo) 2016;71:311–4.
13. Dolman PJ, Rootman J. VISA Classification for Graves orbitopathy. Ophthal Plast Reconstr Surg 2006;22:319–24. [CrossRef]
14. Ginsberg J. Diagnosis and management of Graves' disease. CMAJ 2003;168:575–85.
15. Gopinath B, Wescombe L, Nguyen B, Wall JR. Can autoimmunity against calsequestrin explain the eye and eyelid muscle inflammation of thyroid eye disease? Orbit 2009;28:256–61. [CrossRef]
16. Wall JR, Lahooti H. Pathogenesis of thyroid eye disease--does autoimmunity against the TSH receptor explain all cases? Endokrynol Pol 2010;61:222–7.
17. Heufelder AE. Pathogenesis of Graves' ophthalmopathy: recent controversies and progress. Eur J Endocrinol 1995;132:532–41.
18. Korducki JM, Loftus SJ, Bahn RS. Stimulation of glycosaminoglycan production in cultured human retroocular fibroblasts. Invest Ophthalmol Vis Sci 1992;33:2037–42.
19. Sorisky A, Pardasani D, Gagnon A, Smith TJ. Evidence of adipocyte differentiation in human orbital fibroblasts in primary culture. J Clin Endocrinol Metab 1996;81:3428–31. [CrossRef]
20. Papa A, Emdin M, Passino C, Michelassi C, Battaglia D, Cocci F. Predictive value of elevated neutrophil-lymphocyte ratio on cardiac mortality in patients with stable coronary artery disease. Clin Chim Acta 2008;395:27–31. [CrossRef]
21. Wright HL, Moots RJ, Bucknall RC, Edwards SW. Neutrophil function in inflammation and inflammatory diseases. Rheumatology (Oxford) 2010;49:1618–31. [CrossRef]
22. Wu G, Yao Y, Bai C, Zeng J, Shi D, Gu X, et al. Combination of platelet to lymphocyte ratio and neutrophil to lymphocyte ratio is a useful prognostic factor in advanced non-small cell lung cancer patients. Thorac Cancer 2015;6:275–87. [CrossRef]
23. Gasparyan AY, Ayvazyan L, Mikhailidis DP, Kitas GD. Mean platelet volume: a link between thrombosis and inflammation? Curr Pharm Des 2011;17:47–58. [CrossRef]
24. Guthrie GJ, Charles KA, Roxburgh CS, Horgan PG, McMillan DC, Clarke SJ. The systemic inflammation-based neutrophil-lymphocyte ratio: experience in patients with cancer. Crit Rev Oncol Hematol 2013;88:218–30. [CrossRef]
25. Ilhan N, Daglioglu MC, Ilhan O, Coskun M, Tuzcu EA, Kahraman H, et al. Assessment of Neutrophil/Lymphocyte Ratio in Patients with Age-related Macular Degeneration. Ocul Immunol Inflamm 2015;23:287–290. [CrossRef]
26. Dursun A, Ozturk S, Yucel H, Ozec AV, Dursun FG, Toker MI, et al. Association of neutrophil/lymphocyte ratio and retinal vein occlusion. Eur J Ophthalmol 2015;25:343–6. [CrossRef]
27. Polat O, Yavaş GF, İnan S, İnan ÜÜ. Neutrophil-to-Lymphocyte Ratio as a Marker in Patients with Non-arteritic Anterior Ischemic Optic Neuropathy. Balkan Med J 2015;32:382–7. [CrossRef]
28. Karaca EE, Özmen MC, Ekici F, Yüksel E, Türkoğlu Z. Neutrophil-to-lymphocyte ratio may predict progression in patients with keratoconus. Cornea 2014;33:1168–73. [CrossRef]
29. Elbey B, Yazgan UC, Yildirim A, Karaalp U, Sahin A. Mean platelet volume and neutrophil to lymphocyte ratio in patients with vernal keratoconjunctivitis. Journal of Clinical and Experimental Investigations 2015;6:40–3. [CrossRef]
30. Sekeryapan B, Uzun F, Buyuktarakci S, Bulut A, Oner V. Neutrophil-to-Lymphocyte Ratio Increases in Patients With Dry Eye. Cornea 2016;35:983–6. [CrossRef]
31. Ozgonul C, Sertoglu E, Mumcuoglu T, Kucukevcilioglu M. Neutrophil-to-Lymphocyte Ratio and Platelet-to-Lymphocyte Ratio as Novel Biomarkers of Primary Open-Angle Glaucoma. J Glaucoma 2016;25:e815–e820. [CrossRef]