Kulak Burun Bogaz Ihtis Derg 2017;27(5):226-232 doi: 10.5606/kbbihtisas.2017.58997

Experimental Study / Deneysel Çalışma

The anti-inflammatory effects of thymoquinone in a rat model of allergic rhinitis

Sıçan alerjik rinit modelinde timokinonun anti-enflamatuvar etkileri

Ceren Günel, MD.,[1] Buket Demirci, MD.,[2] İbrahim Meteoğlu, MD.,[3] Mustafa Yılmaz, MD.,[4] İmran Kurt Ömürlü, MD.,[5], Tolga Kocatürk, MD.[6]

[1]*Department of Otolaryngology, Medical Faculty of Adnan Menderes University, Aydın, Turkey*
[2]*Department of Medical Pharmacology, Medical Faculty of Adnan Menderes University, Aydın, Turkey*
[3]*Department of Medical Pathology, Medical Faculty of Adnan Menderes University, Aydın, Turkey*
[4]*Department of Biochemistry, Medical Faculty of Adnan Menderes University, Aydın, Turkey*
[5]*Department of Biostatistic, Medical Faculty of Adnan Menderes University, Aydın, Turkey*
[6]*Department of Opthtalmology, Medical Faculty of Adnan Menderes University, Aydın, Turkey*

ABSTRACT

Objectives: This study aims to investigate the effect of thymoquinone (TQ) on airway inflammation in a rat model of allergic rhinitis (AR).

Materials and Methods: Allergic rhinitis was induced in 42 rats by intraperitoneal sensitization and intranasal challenge with ovalbumin (OVA). The animals were divided into six subgroups (n=7/per cage): healthy controls, AR group, AR group treated with corticosteroid (dexamethasone 1 mg/kg; CS+AR), healthy rats that were given only TQ of 10 mg/kg (TQ10), AR group treated with TQ of 3 mg/kg (TQ3+AR) and AR group treated with TQ of 10 mg/kg (TQ10+AR). We measured the serum levels of interferon-gamma (IFN-γ), interleukin-4 (IL-4), IL-10, and OVA-specific immunoglobulin E (Ig-E). Histopathologic changes in nasal mucosa and expression of tumor necrosis factor-alpha (TNF-α) and IL-1β were evaluated.

Results: Thymoquinone has inhibited the production of the IL-4, OVA-specific IgE, and the expression of TNF-α and IL-1β. It also reduced eosinophil infiltration and edema in the nasal mucosa while it has no increased effect on IFN-γ and IL-10.

Conclusion: We observed that TQ has multiple suppressive effects on allergic response. Thymoquinone may be considered as a supplemental agent in the treatment of allergic rhinitis.

Keywords: Interleukin-10; interleukin-1 beta; interleukin-4; ovalbumin-specific IgE; thymoquinone; allergic rhinitis; tumor necrosis factor-alpha.

ÖZ

Amaç: Bu çalışmada sıçan alerjik rinit (AR) modelinde timokinonun (TQ) havayolu enflamasyonu üzerindeki etkisi araştırıldı.

Gereç ve Yöntem: Ovalbümin (OVA) ile intraperitoneal sensitizasyon ve intranazal uygulama ile 42 sıçanda alerjik rinit indüklendi. Hayvanlar altı alt gruba ayrıldı (kafes başına n=7): sağlıklı kontroller, AR grubu, kortikosteroid ile tedavi edilen AR grubu (1 mg/kg deksametazon; CS+AR), sadece 10 mg/kg TQ verilen sağlıklı sıçanlar (TQ10), 3 mg/kg TQ ile tedavi edilen AR grubu (TQ3+AR) ve 10 mg/kg TQ ile tedavi edilen AR grubu (TQ10+AR). İnterferon-gama (IFN-γ), interlökin-4 (IL-4), IL-10 ve OVA'ya özgü immünoglobülin E'nin (Ig-E) serum düzeyleri ölçüldü. Nazal mukozadaki histopatolojik değişiklikler ve tümör nekroz faktör-alfa (TNF-α) ve IL-1β ekspresyonu değerlendirildi.

Bulgular: Timokinon, IL-4, OVA'ya özgü IgE ve TNF-α ve IL-1β'nın ekspresyonunu engelledi. Aynı zamanda burun mukozasında eozinofil infiltrasyonunu ve ödemi azalttı, ancak IFN-γ ve IL-10 üzerinde artmış bir etki göstermedi.

Sonuç: Timokinonun alerjik reaksiyon üzerinde birden fazla baskılayıcı etkiye sahip olduğu gözlemlendi. Timokinon, alerjik rinit tedavisinde yardımcı bir ajan olarak düşünülebilir.

Anahtar Sözcükler: İnterlökin-10; interlökin-1 beta; interlökin-4; ovalbümine özgü IgE; timokinon; alerjik rinit; tümör nekroz faktör-alfa.

Available online at
www.kbbihtisas.org
doi: 10.5606/kbbihtisas.2017.58997
QR (Quick Response) Code

Received / *Geliş tarihi:* January 15, 2017 Accepted / *Kabul tarihi:* October 07, 2017

Correspondence / İletişim adresi: Ceren Günel, MD. Adnan Menderes Üniversitesi Tıp Fakültesi Kulak Burun Boğaz Anabilim Dalı, 09100 Aydın, Turkey.
Tel: +90 533 - 717 56 93 e-mail *(e-posta):* drgunel@hotmail.com

Allergic rhinitis (AR) is a common illness that has a markedly increasing prevalence.[1] It is characterized by T helper (Th) 2-mediated inflammation through hypersensitivity resulting from specific seasonal or perennial environmental allergens.[2] The inflammatory response is associated with an increase in numbers of eosinophils, mucus secretion and increased production of Th2-type cytokines.[3]

The management of AR involves education, pharmacotherapy, immunotherapy, and surgery. Despite recent advances in understanding the mechanisms underlying allergic inflammation, current therapies can only alleviate the symptoms of disease.[4] Since these therapies are still imperfect, it is important to continue to study the pharmacology of this disease as part of the search to obtain better drugs.

Nigella sativa (N. sativa) is a member of Ranunculaceae family and commonly known as a black seed. The seed of N. sativa is a traditional herbal medicine and has been used for several diseases.[5] Thymoquinone (TQ) (Sigma-Aldrich Chemical Co., St. Louis, Missouri, USA) is a bioactive constituent of the oil of N. sativa. Various biological effects have been linked to the use of N. sativa and some of its active ingredients. In particular TQ has been reported to have antioxidant, anti-inflammatory and immunomodulatory activities.[3,6,7] Recent studies have shown TQ effects on the allergic lung inflammation, rhinosinusitis and sinonasal cilia beat frequency.[3,5,8] However, little is known about the factors and mechanisms underlying these effects. In the light of these findings, TQ may play a potential role on the treatment of allergic rhinitis.

In the present study we investigate the effect of TQ on airway inflammation in a rat model of AR.

MATERIALS AND METHODS

Animals

Forty-two 12-15 month-old female Wistar rats were obtained from the Experimental Animal Center of Medical Faculty and kept in regular cages under standardized conditions (12 h dark/light cycle, 20±1°C room temperature) and allowed free access to food and water. All experiments were performed in accordance with the principles and guidelines of the ADU Animal Ethical Committee (approval; HADYEK 60583101/2014/028). The study was conducted in accordance with the principles of the Declaration of Helsinki.

Production of the AR model and treatment protocol

The rats were randomized into one of six subgroups (n=7/per cage): healthy controls, AR group, AR group treated with corticosteroid (dexamethasone 1 mg/kg; CS+AR), healthy rats that were given only TQ of 10 mg/kg (TQ10), AR group treated with TQ of 3 mg/kg (TQ3+AR), AR group treated with TQ of 10 mg/kg (TQ10+AR).

The allergic rhinitis group was sensitized by intraperitoneal injection of saline of 1 mL containing 30 mg of aluminum hydroxide and 0.3 mg of ovalbumin (OVA) (Sigma A5253, Interlab, Turkey) performed once every other day for 14 days (for a total of seven injections per rat), whereas the control group and only TQ-treated rat group were given 30 mg/kg aluminum hydroxide in saline of 1 mL. Then, they were sensitized by intranasal dripping of 2% OVA, two or three drops per treatment, once a day for following seven days (Figure 1).[9,10] The control group and only TQ-treated rat group were given saline drops. Meanwhile, during the sensitization period, they were also given either dexamethasone or TQ in high and low doses intraperitoneally, once every day. Thymoquinone was dissolved in 100% ethylalcohol (ETOH), aliquoted for bodyweight of per animal and immediately applied within two minutes after second dilution in serum physiologic salt solution. Hence, the final ETOH concentration was 10%. The control group also received 10% ETOH. The CS+AR group served as a comparison of TQ treatment with the standard therapy. Only the TQ group itself clarified the safety of TQ.

Cytokines and OVA-Ig E in serum

To evaluate the allergic reaction, the following were measured by enzyme-linked immunosorbent assay (ELISA; Baoshan District, Shanghai, Chinese); interferon (IFN)-γ for T helper 1 (Th1) immune reaction, interleukin 4 (IL-4) for T helper 2 (Th2) immune reaction, IL-10 for T regulator (Treg) and serum OVA-specific immunoglobulin E (IgE) levels. Procedures were performed according to manufacturer's instructions.

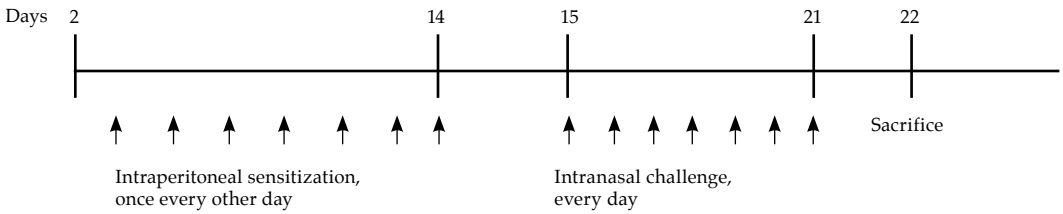

Figure 1. *Allergic rhinitis rat model.*

Histopathological examination

All of the animals were sacrificed at post-experiment day 21 under ketamine and xylasine (50 and 5 mg/kg, respectively) anesthesia. Blood samples were taken by cardiac puncture, centrifuged and the sera kept in -80°C for measurement of interleukin and interferon levels. The nasal respiratory tissues were harvested and fixed in 10% formalin solution overnight. Thereafter, they were decalcified and coronal sections were chosen from the middle segment of the sinonasal cavity (maxillary sinus and olfactory region) as the principal area of the histopathological examination.

Paraffin blocks were formed and anatomically similar sections were collected. Finally, 4 µm

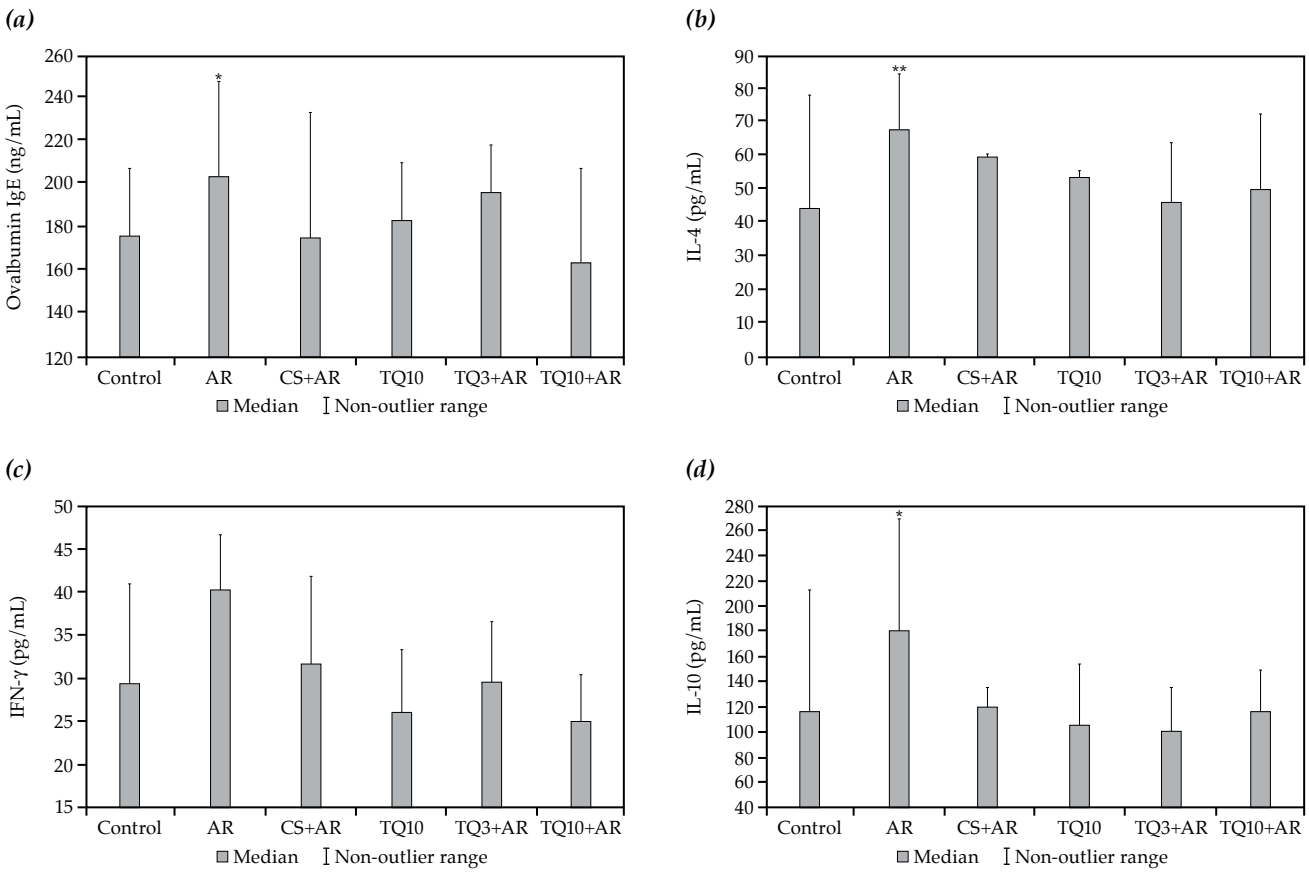

Figure 2. ***(a)*** *The ovalbumin specific IgE levels in the serum of the rat groups. * The ovalbumin specific IgE levels were significantly higher in the AR group than control, CS+AR and TQ10+AR groups (p<0.05).* ***(b)*** *The interleukin-4 (IL-4) levels in the serum of the rat groups. ** There were significantly differences in the IL-4 levels between AR and control, TQ3+AR, TQ10+AR groups (p<0.05).* ***(c)*** *The interferon (IFN)-γ levels in the serum of the rat groups.* ***(d)*** *The interleukin-10 (IL-10) levels in the serum of the rat groups. * IL-10 levels were significantly higher in the AR group than TQ10, TQ3+AR and TQ10+AR groups (p<0.05).* *AR: Allergic rhinitis; CS+AR: Corticosteroid+AR, TQ: thymoquinone 10 mg/kg; TQ3+AR: Thymoquinone 3 mg/kg +AR; TQ10+AR: Thymoquinone 10 mg/kg+AR.*

thick sections were stained with hematoxylin and eosin (H-E) to evaluate nasal mucosa edema, goblet cell, and eosinophil counts in the nasal mucosa. The goblet cell count was indicated through (0) normal, (1) slight increase and (2) severe increase; eosinophil through (0) <10/high, high power field (HPF) and (1) >10/HPF; edema through (0) absence, (1) mild, (2) moderate and (3) severe.

Immunohistochemistry

Immunohistochemical staining was done by using DAKO Autostainer Universal Staining System (Autostainer Link 48 DAKO, Glostrup, Denmark). Firstly, 4 μm sections were mounted on positive-loaded glass slides. Then the sections were deparaffinized with xylol and dehydrated by passing through ethyl alcohol series. Subsequently, antigen retrieval was performed in a thermostatic bath (PT Link) at 96°C (10 mM/L citrate buffer, pH 6) for 40 minutes. The sections were incubated with anti-tumor necrosis factor-α (TNF-α) (cat. No: NB600-587, Novus Biologicals, CO/USA) and anti-IL-1β (cat. No. SC-7884, Santa Cruz Biotechnology, USA) primary antibodies for 60 minutes. Using streptavidin-biotin immunoperoxidase technique (K8000 Envision Flex, DAKO, Glostrup, Denmark) an automatized

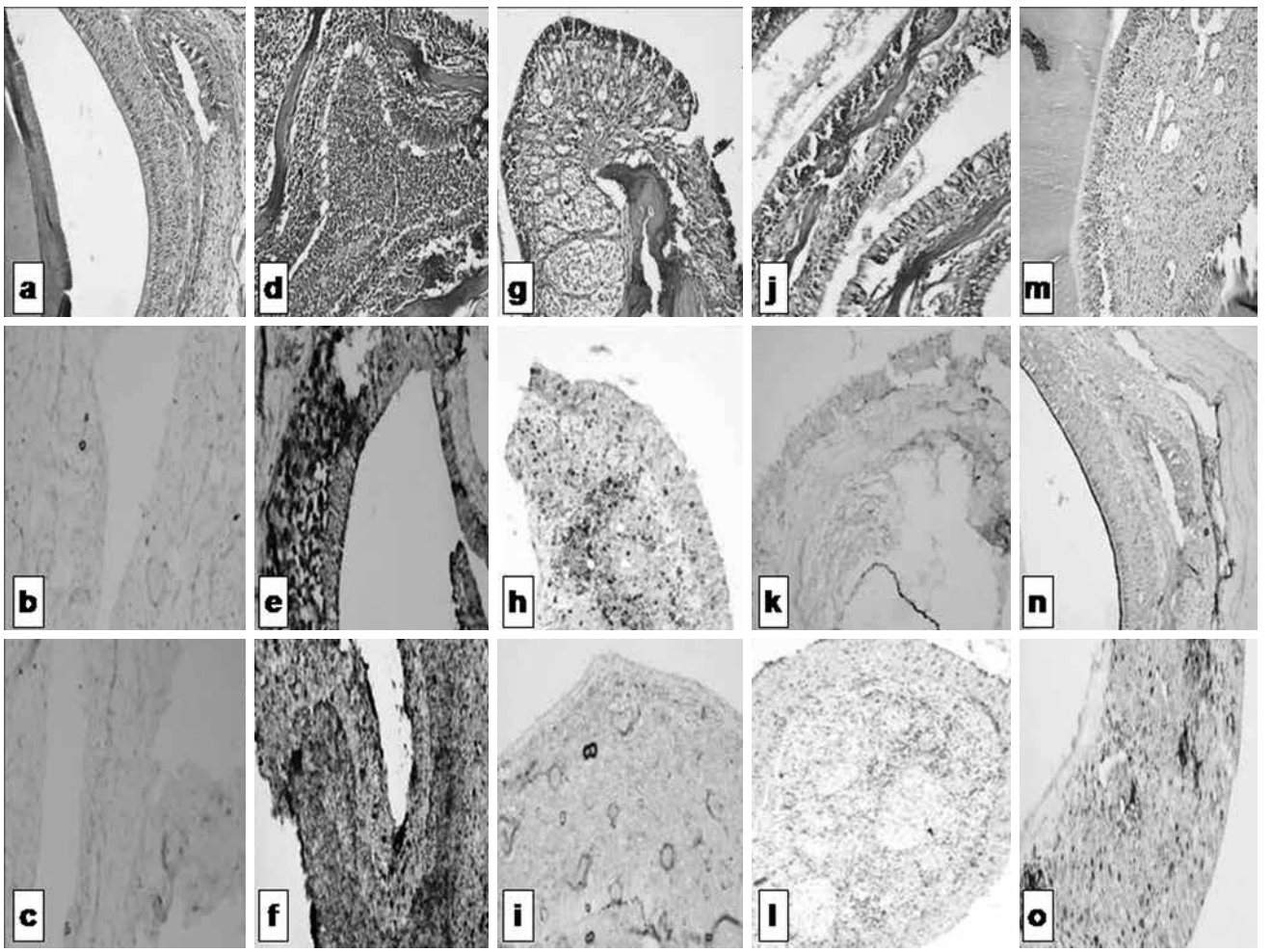

Figure 3. *There is a normal appearance in the control group* ***(a)*** *(H-E×100), there are no immunohistochemical staining with TNF-α* ***(b)*** *(TNF-α, x100) and IL-1β* ***(c)*** *(IL-1 ×100); There are severe eosinophil infiltration, and edema* ***(d)*** *(H-E×100), severe immunohistochemical staining with TNF-α* ***(e)*** *(TNF ×100) ve IL-1β* ***(f)*** *(IL-1×100) in the allergic rhinitis group. There is mild edema* ***(g)*** *(H-E×100), mild immunohistochemical staining with TNF-α* ***(h)*** *(TNF-α ×100) and no staining with IL-1β* ***(i)*** *(IL-1, ×100) in the CS+AR group; There is mild edema* ***(j)*** *(H-E×100), mild immunohistochemical staining with TNF-α* ***(k)*** *(TNF-α ×100) and IL-1β* ***(l)*** *(IL-1 ×100) in the TQ3+AR group; There is mild edema* ***(m)*** *(H-E×100), mild immunohistochemical staining with TNF-α* ***(n)*** *(TNF-α ×100) ve IL-1β* ***(o)*** *(IL-1 ×100) in the TQ10+AR group. CS+AR: Corticosteroid+ allergic rhinitis; TQ3+AR: Thymoquinone 3 mg/kg+ allergic rhinitis; TQ10+AR: Thymoquinone 10 mg/kg+ allergic rhinitis.*

Table 1. The eosinophil count in the nasal mucosa of the rat groups

	Groups						
Eosinophil count	Control	AR**	CS+AR	TQ10	TQ3+AR	TQ10+AR	*p*
0	6	2	5	6	6	7	0.038
1	1	5	2	1	1	0	
Total	7	7	7	7	7	7	

(0) Eosinophil count <10/hpt, (1) eosinophil count >10/hpt. * AR: Allergic rhinitis was significantly different from control, TQ10, TQ3+AR, TQ10+AR groups. AR: Allergic rhinitis; CS+AR: Corticosteroid+AR; TQ: Thymoquinone 10 mg/kg; TQ3+AR: Thymoquinone 3mg/kg +AR; TQ10+AR: Thymoquinone 10 mg/kg+AR.

system was used. In order to provide a colored image, immunoreactions with diaminobenzidine tetrachloride (DAB) were displayed. For background staining, the sections were oppositely stained with hematoxylin. For dehydration, the sections were passed through alcohol series with increasing strength and after being cleared in xylol, lined with balsam. After the staining stage, the sections were examined under Olympus BX51 light microscope (Olympus Optical, Tokyo, Japan) with 4, 10, 20, and 40 magnifications by a single pathologist blinded to subject groups. A minimum 200 cells were counted in the mostly stained areas (hot spots). They were scored as; (0) <5% staining, (1) 5-10% staining, (2) 11-20% staining and (3) >20% staining.

Statistical analysis

Comparisons between groups were analyzed using the Kruskal Wallis test. Dunn's post-hoc test was used to compare between selected groups. Descriptive statistics were showed as median (25-75 percentiles) or frequency. P-values <0.05 were considered significant.

RESULTS

Serum levels of OVA-IgE and cytokines

We evaluated the effects of the TQ on OVA-induced allergic responses in rats, the levels of OVA-specific IgE, IFN-γ, IL-4, and IL-10 were measured by ELISA. Ovalbumin-specific IgE levels were increased by sensitization (p=0.048). The OVA-specific IgE levels were significantly decreased in the TQ10+AR and CS+AR groups than the OVA-sensitized rats (p=0.038, p=0.048), (Figure 2a). The IL-4 levels significantly increased in the AR group (p=0.004) compared with the control group. Administration of TQ at dose of 3 mg/kg and 10 mg/kg, significantly inhibited IL-4 production (both p=0.013) compared with the AR group (Figure 2b). Although, IFN-γ in serum did not differ significantly among the AR, control, and treatment groups, its production tended to decrease in treatment groups (Figure 2c). Compared to the AR group, the levels of the Treg cytokine, IL-10, significantly decreased in the TQ group at doses of 3 mg/kg and 10 mg/kg (p=0.033, p=0.002) respectively (Figure 2d).

Histopathological changes in nasal mucosa

The histopathological examination of the nasal mucosa was normal in the control group while edema, eosinophilic infiltration and goblet cell count increased in the AR group (Figure 3a-o). In the AR group the eosinophil count in the nasal mucosa was significantly higher (p=0.013) than the control group, while the eosinophil count significantly decreased in the TQ3+AR and TQ10+AR groups when compared to AR group (p=0.002, p=0.013) respectively (Table 1). Edema was significantly greater in the AR group compared to the control group (p<0.001). Administration of TQ at doses of 3 mg/kg and 10 mg/kg significantly decreased edema (both, p<0.001) (Table 2). Interestingly, edema and eosinophilic inflammation decreased slightly in the corticosteroid group. However, goblet cell counts did not differ significantly among the AR, control, and treatment groups (Table 3).

Expression of TNF-α and IL-1β in nasal mucosa

The expression of TNF-α significantly decreased in the CS+AR, TQ3+AR, TQ10+AR groups (p=0.05, p=0.015, p=0.004, respectively), while sensitization of OVA significantly increased the expression of TNF-α and IL-1β (both, p<0.001) (Figure 3b-o). The expression level of IL-1β significantly decreased in the CS+AR and

Table 2. The degree of edema of the nasal mucosa of the rat groups

	Groups						
Eosinophil count	Control	AR**	CS+AR	TQ10	TQ3+AR	TQ10+AR	*p*
0	5	0	1	5	5	5	0.001
1	2	3	6	2	2	2	
2	0	4	0	0	0	0	
Total	7	7	7	7	7	7	

(0) Absence, (1) mild, (2) moderate, (3) severe. ** AR was significantly different from control, TQ10, TQ3+AR, TQ10+AR groups. AR: Allergic rhinitis; CS+AR: Corticosteroid+AR; TQ: Thymoquinone 10 mg/kg; TQ3+AR: Thymoquinone 3 mg/kg +AR; TQ10+AR: Thymoquinone 10 mg/kg+AR.

Table 3. The goblet cell count in the nasal mucosa of the rat groups

	Groups						
Goblet cell	Control	AR	CS+AR	TQ10	TQ3+AR	TQ10+AR	*p*
0	5	3	5	5	6	1	0.38
1	2	4	2	2	1	6	
Total	7	7	7	7	7	7	

(0) normal, (1) slight increase, (2) severe increase. AR: Allergic rhinitis, CS+AR: corticosteroid+AR, TQ: Thymoquinone 10 mg/kg, TQ3+AR; Thymoquinone 3 mg/kg +AR; TQ10+AR: Thymoquinone 10 mg/kg+AR.

TQ10+AR groups ($p<0.001$, $p=0.006$, respectively) (Figure 3b-o). Conversely, administration of TQ at dose of 3 mg/kg had no effect on IL-1β ($p=0.072$).

DISCUSSION

Allergic rhinitis is characterized by an Ig-E mediated disease. Treatment protocols for AR aim at either reducing the effect of chemical mediators from activated mast cells and eosinophils, or tissue.[11] Although treatment protocols including antihistamines and corticosteroids relive the symptoms, drug studies continue to obtain better outcomes.

The production of T_2 cytokines such as IL-4 and IL-13 is known to play an important role in AR pathophysiology.[3] These cytokines are produced by mast cells, T cells, macrophage and epithelial cells.[12] Interleukin-4 is necessary for differentiation of T cells from the Th2 type. Besides, it plays a role in the late-phase response, including eosinophil migration and mucus hypersecretion. Both the IL-4 and IL-13 regulate IgE isotype switching in B cells and eosinophil function, and increase mucus production.[13] Conversely, IFN-γ and IFN-γ inhibit IgE production.[11] In this study, TQ significantly inhibited the production of IL-4 and OVA-Ig-E. However, TQ had no effect on the induction of IFN-γ. These results demonstrate that TQ may inhibit IgE production by the decreasing IL-4 in allergic responses. In addition, there were no significant difference in IL-4 levels between TQ doses, while TQ significantly decreased the release of the Th2 cytokine, IL-4, compared with corticosteroid. These results suggest that TQ could control the Th2 cytokine response in allergic conditions and may explain the apparent therapeutic potential of TQ in AR.

Regulatory T cells have a suppressive effect on allergic inflammation. Interleukin 10 which is one of the regulatory T cells, inhibits the proinflammatory effect of the mast cell and eosinophil.[14] In our study, IL-10 level was significantly decreased by TQ treatment at doses of 3 mg/kg and 10 mg/kg. This result shows that TQ had no increased effect on the regulatory T cells and the production of IL-10. We thought this impact was due to TQ's inhibitor effect.

In AR, the presence of eosinophils in nasal mucosa is characteristic and edema in nasal mucosa develops with inflammatory cells as a secondary reaction.[14] Proinflammatory cytokines such as TNF-α and IL-1β play an important role in the pathogenesis of AR and are produced by the IgE-mediated activation of mast cells and

eosinophils.[15,16] We demonstrated TQ is a more potent inhibitor on the eosinophil infiltration and edema than corticosteroids. In the present study, TQ strongly inhibited the expression of TNF-α and IL-1β in the nasal mucosa of AR rats. There was no significant difference between TQ doses and corticosteroids in terms of TNF-α concentration. When we looked at the IL-1β results, we found that corticosteroids significantly inhibited the IL-1β concentration compared to TQ. These results suggest that TQ can attenuate allergic inflammation by suppressing the expression of TNF-α and IL-1β in AR.

In conclusion, our observations suggested that in a rat model of AR, TQ had an anti-allergic effect on allergic parameters. Thymoquinone inhibited IL-4 release from T cells and OVA-specific IgE secretions from B cells. Furthermore, TQ has anti-inflammatory activity by suppressing the production of the inflammatory cytokines, TNF-α and IL-1β and by reducing eosinophil infiltration into and edema in the nasal mucosa. Additionally, its effect on eosinophil count, edema and IL-4 level was significantly higher than that of corticosteroids. These multiple effects may synergize to reduce allergic symptoms. It seems that TQ at doses of 10 mg/kg is more effective than a TQ at doses of 3 mg/kg. Thus, TQ may potentially be considered a supplemental agent in the treatment of AR.

Declaration of conflicting interests

The authors declared no conflicts of interest with respect to the authorship and/or publication of this article.

Funding

The authors received no financial support for the research and/or authorship of this article.

REFERENCES

1. Bousquet J, Khaltaev N, Cruz AA, Denburg J, Fokkens WJ, Togias A, et al. Allergic Rhinitis and its Impact on Asthma (ARIA) 2008 update (in collaboration with the World Health Organization, GA(2)LEN and AllerGen). Allergy 2008;63:8-160.
2. Kleinjan A, van Nimwegen M, Leman K, Hoogsteden HC, Lambrecht BN. Topical treatment targeting sphingosine-1-phosphate and sphingosine lyase abrogates experimental allergic rhinitis in a murine model. Allergy 2013;68:204-12.
3. El Gazzar M, El Mezayen R, Marecki JC, Nicolls MR, Canastar A, Dreskin SC. Anti-inflammatory effect of thymoquinone in a mouse model of allergic lung inflammation. Int Immunopharmacol 2006;6:1135-42.
4. Kim BY, Shin JH, Park HR, Kim SW, Kim SW. Comparison of antiallergic effects of pneumococcal conjugate vaccine and pneumococcal polysaccharide vaccine in a murine model of allergic rhinitis. Laryngoscope 2013;123:2371-7.
5. Cingi C, Eskiizmir G, Burukoğlu D, Erdoğmuş N, Ural A, Ünlü H. The histopathological effect of thymoquinone on experimentally induced rhinosinusitis in rats. Am J Rhinol Allergy 2011;25:e268-72.
6. Gali-Muhtasib H, Roessner A, Schneider-Stock R. Thymoquinone: a promising anti-cancer drug from natural sources. Int J Biochem Cell Biol 2006;38:1249-53.
7. Salem ML. Immunomodulatory and therapeutic properties of the Nigella sativa L. seed. Int Immunopharmacol 2005;5:1749-70.
8. Uz U, Chen B, Palmer JN, Cingi C, Unlu H, Cohen NA. Effects of thymoquinone and montelukast on sinonasal ciliary beat frequency. Am J Rhinol Allergy 2014;28:122-5.
9. Wang W, Zheng M. Nuclear factor kappa B pathway down-regulates aquaporin 5 in the nasal mucosa of rats with allergic rhinitis. Eur Arch Otorhinolaryngol 2011;268:73-81.
10. Xu YY, Liu X, Dai LB, Zhou SH. Effect of Tong Qiao drops on the expression of eotaxin, IL-13 in the nasal mucosa of rats with allergic rhinitis. J Chin Med Assoc 2012;75:524-9.
11. Jung HW, Jung JK, Park YK. Antiallergic effect of Ostericum koreanum root extract on ovalbumin-induced allergic rhinitis mouse model and mast cells. Asian Pac J Allergy Immunol 2011;29:338-48.
12. Settipane RA, Schwindt C. Chapter 15: Allergic rhinitis. Am J Rhinol Allergy 2013;27:52-5.
13. Ying S, Durham SR, Corrigan CJ, Hamid Q, Kay AB. Phenotype of cells expressing mRNA for TH2-type (interleukin 4 and interleukin 5) and TH1-type (interleukin 2 and interferon gamma) cytokines in bronchoalveolar lavage and bronchial biopsies from atopic asthmatic and normal control subjects. Am J Respir Cell Mol Biol 1995;12:477-87.
14. Osguthorpe JD. Pathophysiology of and potential new therapies for allergic rhinitis. Int Forum Allergy Rhinol 2013;3:384-92.
15. Zhang HQ, Sun Y, Xu F. Therapeutic effects of interleukin-1 receptor antagonist on allergic rhinitis of guinea pig. Acta Pharmacol Sin 2003;24:251-5.
16. Jung HW, Jung JK, Park YK. Comparison of the efficacy of KOB03, ketotifen, and montelukast in an experimental mouse model of allergic rhinitis. Int Immunopharmacol 2013;16:254-60.