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# RESEARCH ARTICLE

## Serologic prevalence of *Ornithobacterium rhinotracheale* infection in commercial layers

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## Ticari yumurtacı tavuklarda Ornithobacterium rhinotracheale enfeksiyonunun serolojik prevalansı

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**Öz**

**Amaç:** Bu çalışmada ticari yumurtacı tavuk işletmelerinde bulunan tavuklarda *Ornithobacterium (O.) rhinotracheale* enfeksiyonunun serolojik prevalansının belirlenmesi amaçlandı.

**Gereç ve Yöntem:** Konya, Aksaray, Karaman, Ankara ve Gaziantep illerinde bulunan 26 farklı kümesteki yumurtacı tavuklardan 650 kan serum örneği toplandı. Örnekleme yapılan tavukların hiç birisi daha önceden *O. rhinotracheale* enfeksiyonuna karşı aşılanmamışlardı. *O. rhinotracheale*'ye karşı oluşmuş antikorların varlığı ELISA testi ile belirlendi.

**Bulgular:** Toplam 650 kan serum örneğinden 113 (%17.4)'ünde *O. rhinotracheale* antikoru tespit edildi. Bu 113 pozitif serum örneği örnekleme yapılan 26 kümesin 12 (%46.2)'sinden toplanmıştı. Enfekte 12 kümes, Konya, Gaziantep, Ankara ve Karaman illerindeki 12 farklı çiftliğe aitti.

**Öneriler:** Bu çalışmanın sonuçları göstermiştir ki, Türkiye'deki ticari yumurtacı tavuk kümeslerinde *O. rhinotracheale* enfeksiyonu yüksek bir prevalansa sahiptir.

**Anahtar kelimeler:** *Ornithobacterium rhinotracheale*, yumurtacı tavuk, serolojik prevalans.

**Abstract**

**Aim:** The purpose of this study was to determine the serological prevalence of *Ornithobacterium (O.) rhinotracheale* infections in commercial layers.

**Materials and Methods:** A total of 650 blood serum samples were collected from 26 different layer flocks located in Konya, Aksaray, Karaman, Ankara, and Gaziantep provinces. None of the chickens have been vaccinated against *O. rhinotracheale* prior to sampling. The presence of antibodies against *O. rhinotracheale* in each sample was determined by ELISA.

**Results:** Of the 650 serum samples, 113 (17.4%) were found to be positive for *O. rhinotracheale* antibodies. These 113 positive sera were collected from 12 (46.2%) out of 26 flocks. The twelve infected flocks belonged to 12 different farms form Konya, Gaziantep, Ankara and Karaman.

**Conclusion:** The results of this study indicated that *O. rhinotracheale* infection is at high prevalence in the commercial layer flocks from this part of Turkey.

**Keywords:** *Ornithobacterium rhinotracheale*, layer, serological prevalence.

## Introduction

*Ornithobacterium rhinotracheale*, which cause a highly contagious respiratory infection in turkeys and chickens, is a Gram-negative, pleomorphic, nonmotile, rod-shaped, nonspore forming bacterium in the rRNA superfamily V (Van Empel et al 1997). This bacterium has 18 serotypes and is frequently associated with other respiratory diseases (Van Empel and Hafez 1999). Clinical signs of *O. rhinotracheale* infections are tracheitis, airsacculitis, pericarditis, sinusitis, exudative pneumonia, drop in egg production, decreasing in hatchability, increased condemnation restes, osteitis, and meningitis (Van Veen et al 2000). *O. rhinotracheale* has been isolated from chicken, turkey, duck, chukar, goose, quail, ostrich, guinea fowl, rooks, pheasant, pigeon, and can possibly be found in other species of birds as well (Chin and Droual 1997).

*O. rhinotracheale* infections have been reported from Belgium, Iran, Germany, United States, South Africa, Netherlands, France, Israel, Hungary, Japan, United Kingdom and Turkey (Charlton et al 1993, Hinz et al 1994, Van Beek 1994, Dudouyt et al 1995, Travers 1996, Hafez 1998, Sakai et al 2000, Banani et al 2002, Turan and Ak 2002, Allymehr 2006, Rahimi and Banani 2007). In Turkey, presence of *O. rhinotracheale* infections in broiler, layer and turkey flocks has been announced over the last decade by some researchers (Erganiş et al 2002, Turan and Ak 2002, Özbey et al 2004, Türkyılmaz 2005, Türkyılmaz and Kaya 2005, Erganiş et al 2013).

The diagnosis of *O. rhinotracheale* infection is made by isolation and identification of agent by cultural, serological and molecular methods such as polymerase chain reaction. The culture of *O. rhinotracheale* is difficult because the bacterium is slowly grown in the liquid media (Van Empel et al 1997). Serological tests are routinely used to test large population and in surveillance studies (Back et al 1998). ELISA test could detect the presence of antibodies against *O. rhinotracheale* in samples from poultry blood and egg-yolk (Van Empel et al 1997).

There are limited studies on determining of serological prevalence of *O. rhinotracheale* in commercial layers. The aim of this study was to measure the serological prevalence of *O. rhinotracheale* infections in layers from an area known for its layer poultry practices.

## Materials and methods

*Study samples*

A total of 650 blood serum samples were collected from 26 different layer flocks located in Konya, Aksaray, Karaman, Ankara, and Gaziantep provinces in Turkey, during the later six months of 2014. As the farms of the main egg production companies in Turkey are located in these regions, they are important cities for poultry sector of Turkey. The total number of sample was based on an expected prevalence of 10% with an accuracy of 5% and 95% confidence limits. Twenty-five blood serum samples were randomly collected from each flock for serological testing. Sera were kept in aliquots at -20°C until analyzed.

That flock was considered as *O. rhinotracheale* positive if at least one serum sample from the flock gave a positive result. Flocks were defined as hens of same age group (25-50 weeks) and same origin. All of the 26 flocks visited in this study were belong to different farms. None of the chickens sampled has been vaccinated against *O. rhinotracheale* prior to sampling.

*Enzyme-Linked Immunosorbent Assay (ELISA)*

The antibodies against *O. rhinotracheale* in serum samples were investigated by a commercial ELISA test kit (Flock Chek *O. rhinotracheale*, IDEXX, Switzerland). ELISA procedure was performed as recommended by manufacturer. The absorbance at 650 nm was read by a spectrophotometer (Lambda scan 200 Bio-Tek Inst Inc USA). The level of antibody in serum samples was detected by calculating sample to positive

Table 1. Serologic prevalence of *Ornithobacterium rhinotracheale* in layers.

| Province | Age (weeks) | Number of flocks | Number of samples | Number of positive flock (%) | Number of positive sample (%) |
|---|---|---|---|---|---|
| Konya | 25-48 | 11 | 275 | 6 (54.5) | 49 (17.8) |
| Aksaray* | 32-41 | 2 | 50 | 0 (0) | 0 (0) |
| Ankara | 25-50 | 7 | 175 | 3 (42.9) | 31 (17.7) |
| Karaman | 26-28 | 2 | 50 | 1 (50) | 12 (24) |
| Gaziantep | 29-50 | 4 | 100 | 2 (50) | 21 (21) |
| Total | | 26 | 650 | 12 (46.2) | 113 (17.4) |

*This difference was statistically significant (P<0.05).

control (S/P) ratio. The result was considered as positive for *O. rhinotracheale* when the sample having S/P ratio of greater than 0.4.

*Statistical analyzes*

The statistical differences among groups were determined by Chi-square test. SPSS software version 12 was used for the statistical analyses. P<0.05 level was accepted statistically significant.

## Results

Of the 650 serum samples collected from layers, 113 (17.4%) were found to be positive for *O. rhinotracheale* by ELISA (Table 1). The positive samples were collected from the 12 (46.2%) flocks which located in Konya, Karaman, Ankara, and Gaziantep provinces. The serological prevalence of O. rhinotracheale for Konya, Karaman, Ankara, Gaziantep, and Aksaray provinces were found as 17.8%, 24%, 17.7%, 21%, and 0% respectively. The prevalence of disease in Aksaray was statistically lower than other provinces (P<0.05).

## Discussion

*O. rhinotracheale* causes contagious respiratory infections in turkeys, broilers and layers. Symptoms and mortality rates of disease may be changed according to animal species (Hafez 1998). Anti *O. rhinotracheale* antibodies can be determined on the 5th day post aerosol *O. rhinotracheale* exposure (Eroksuz et al 2006). It was showed that ELISA test has a detection capacity up to 100% of chickens infected throughout the 8 weeks post exposure (Lopes et al 2000).

Thus, in this study, presence of *O. rhinotracheale* was investigated serologically by ELISA in Konya, Aksaray, Karaman, Ankara, and Gaziantep provinces, located in central and south of Turkey. Of the 650 serum samples collected from commercial layers, 113 (17.4%) serum samples were found to be positive for *O. rhinotracheale* (Table 1). Turan and Ak (2002) reported that seroprevalence of *O. rhinotracheale* in different broiler and layers was determined as 64.4% in Marmara and Western Black Sea regions of Turkey. Türkyılmaz and Kaya (2005) worked on the prevalence of *O. rhinotracheale* infection in Aydın province of Turkey and announced that 66.3% of the sera samples from 21 broiler and layer flocks were positive. Özbey et al (2004) found that 10.2% serum samples from broiler flocks in Elazığ province located in the East of Turkey were positive for *O. rhinotracheale* antibody. Seroprevalence of *O. rhinotracheale* in different layer serum samples was reported to be 25.8% in pooled samples from Ankara and Afyon provinces of Turkey (Türkyılmaz 2005). Findings of our work are lower than those results of Turan and Ak (2002) and Türkyılmaz and Kaya (2005), but are in partially agreement with the findings of Özbey et al (2004) and Türkyılmaz (2005).

Seroprevalence of *O. rhinotracheale* in chickens was investigated in different parts of the world. Hafez and Sting (1996) announced that *O. rhinotracheale* specific antibodies were detected in 26% of broiler serum samples in Germany. Also, antibodies against *O. rhinotracheale* were detected in 52% of the tested sera of layers by Heeder et al (2001) in United State of America; 12.7% by Sakai et al 2000 in Japan; 20.3% of tested 363 serum samples from different broiler flocks by Refai et al. (2005) in Egypt; 44.2% and 50.1% of broiler chickens in Iran by Allymehr (2006) and Rahimi (2014), respectively. However, in Thailand, 19.6% of tested broiler serum samples were found positive for *O. rhinotracheale* (Chansiripornchai et al 2007). In our study, seroprevalence of *O. rhinotracheale* in layers was detected as 17.4% in central and south of Turkey. The variety in the findings of the works may be due to the regional differences, screening tests used, and the different age of chickens sampled.

In the present study, 113 positive serum samples were obtained from 12 (46.2%) of 26 layer flocks. Rahimi (2014) reported that 18 (75%) of 24 examined broiler flock were positive for *O. rhinotracheale* infection. In Germany, 79% of tested broiler flocks were found to be *O. rhinotracheale* positive (Hafez and Sting 1996). Result of present work is lower than those findings of Rahimi (2014) and Hafez and Sting (1996).

## Conclusion

Presence of *O. rhinotracheale* was serologically introduced for the first time in layers in Karaman and Gaziantep provinces. The results of this study indicated that prevalence of *O. rhinotracheale* infection is high in the commercial layer flocks in the central and south of Turkey.

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