Prevalence of Nosemosis and Varroosis in Honey Bees in Sinop Province

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Abstract

Aim of study: Sinop province has a potential to progress in beekeeping. In this study, it was aimed to investigate the presence and distribution of nosemosis and varroosis in honey bees in Sinop province.

Area of study: 12 villages from 7 districts (Ayancık, Boyabat, Dikmen, Durağan, Erfelek, Gerze and Saraydüzü) in Sinop province were determined as study area.

Material and methods: A total of 874 adult bee samples were dissected in Ringer's solution and examined for nosemosis. A total of 1.640 adult bee samples were examined for varroosis.

Main results: Nosemosis was the most common disease. Average of the nosemosis infection was 7.6%. On the other hand, varroosis was observed in the eight of 12 localities examined in Sinop province. Total infestation ranged from 0.6 to 8.2%.

Highlights: Nosemosis and varroosis were observed in eight (66.7%) of the 12 examined localities in Sinop province. These data are considered as proof that honey production is negatively affected. This study also confirms that the beekeepers in the Sinop province should be informed in detail about the precautions to be taken in the monitoring and controlling varroosis and nosemosis.

Keywords: Honey bee, Nosemosis, Varroosis, Sinop, Türkiye

Sinop Yöresi Bal Arılarında Nosemosis ve Varroosis'in

Yaygınlığı

Öz

Çalışmanın amacı: Sinop ili zengin flora ve bitki örtüsü nedeniyle arıcılıkta gelişme potansiyeline sahiptir. Bu çalışmada, Sinop ilinin farklı ilçelerindeki bal arısı örneklerinde varroosis ve nosemosis varlığının belirlenmesi amaçlanmıştır.

Çalışma alanı: Sinop bölgesindeki 7 ilçeden (Ayancık, Boyabat, Dikmen, Durağan, Erfelek, Gerze ve Saraydüzü) 12 köy çalışma alanı olarak belirlenmiştir.

Materyal ve yöntem: Toplam 874 ergin arı örneği Ringer solüsyonunda disekte edilerek nosemosis yönünden incelenmiştir. Aynı zamanda, 1.640 ergin arı örneği varroozis yönünden incelenmiştir.

Temel sonuçlar: Nosemosis en yaygın hastalık olarak belirlenmiştir. Nosemosis enfeksiyonunun ortalaması % 7.6 olarak belirlenmiştir. Öte yandan, incelenen 1640 arı örneğinden 33'ü Varroa akarı ile enfekte olmuştur. Toplam enfeksiyon % 0.6 ile % 8.2 arasında değişmiştir.

Araştırma vurguları: Bu çalışma, Sinop bölgesinde Varroa akarı ve Nosema spp. patojenleri ile bulaşıklığın yüksek bir düzeyde olduğunu göstermekte ve Sinop bölgesindeki arıcıların varroosis ve nosemosisin izlenmesi ve kontrol altına alınmasında gereken önlemler hakkında ayrıntılı olarak bilgilendirilmesi gerektiğini ortaya koymaktadır.

Anahtar Kelimeler: Bal arısı, Nosemosis, Varroosis, Sinop, Türkiye

Introduction

Different types of microorganisms cause diseases in agriculturally valuable animal and plant species (Oso et al., 2021; Tatli et al., 2021). While some of these diseases are desired diseases, some are undesirable diseases. Nosemosis and varroosis are undesirable disease in honey bees. Several nosemosis develops slowly in bee colonies and causes the death of many bees (Korkmaz, 2015). The disease is caused by two different species, *Nosema apis* and *Nosema ceranae*, which are included in the genus *Nosema* in Microspora (Fries, 1988; Paxton, 2010). Varroosis is another parasitic disease that is widely known in Türkiye and causes

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weakening of the immune systems of bees, decrease in body weight and shortening of life span (Kumova, 2003; Yücel, 2005; Güler, 2017). These two parasitic and pathogenic diseases seriously reduce honey production and cause colony losses.

Sinop province has an important place in honey production (Erka Alkan, 2020). It is a very suitable location for beekeeping with its cool winters and warm summers, diverse vegetation and rich forests (Albayrak, 2019). Sinop province has a significant honey forest potential. At least 13 nectar, secretion and pollen-producing plant species that can be found in honey forests are found in Sinop province (Ayan et al. 2014). One of them, chestnut is an important tree for honey production. Chestnut honey is a valuable honey and it has high antioxidant activity.

While Sinop province has 29.731 bee hives, honey production is 257 tons and Sinop province ranks 65th in honey production in Türkiye according to 2022 data (TÜİK, 2022). Diseases in bee colonies are the main factor effecting honey production. In this study, the presence and distribution of nosemosis and varroosis diseases were investigated in honey bees in Sinop province for the first time.

Materials and Methods

Bee Samples

Adult bee samples were collected from 12 villages from 7 districts (Ayancık, Boyabat, Dikmen, Durağan, Erfelek, Gerze and Saraydüzü) in Sinop province. Sample collection was carried out in September 2022.

Investigation for Nosemosis

A total of 874 adult bee samples bees were dissected in Ringer's solution and wet smears were prepared. Host fat body, Malpighian tubules, gut epithelium and hemolymph were examined for the presence of pathogens under a light microscope at \times 400–1000 magnification (Yaman, 2020). When an infection with the pathogen was observed, a part of the material was used for the preparation of Giemsa-stained smears. For this, the slides were air-dried and fixed with methanol, then stained with a freshly prepared 5% solution of Giemsa stain and reexamined under the microscope by using the oil immersion lens. The spores detected by the light microscopy were measured and photographed using a microscope with a digital camera and Soft Imaging System. The spores of the detected pathogens were examined in terms of shape and spore sizes and measured as width and length of the spores in both fresh preparations and Giemsa dyed preparations.

Investigation for Varroosis

A total of 1.640 adult bee samples were examined for varroosis. The dead and living bees were flooded with 70% alcohol and shaken on a shaker for 30 minutes. After shaking, it waited for 10-15 minutes for the bee samples and other particles to fall out. Then, it was filtered with filter paper and bee and Varroa samples were transferred to a new filter paper on a bright plate, where the mites can be easily identified and counted under stereo microscope (Kuvancı et al., 2013). The diagnostic method for Varroa parasite was based on morphological identification by alcohol wash method according to Oliver (2020). The percentage of a mite infestation level was calculated by the following formula: mite infestation (%) = (Number of varroa mites/number of adult bees) \times 100 (Salkova and Gurgulova, 2022).

Results and Discussion

In this study, the presence and distribution of nosemosis and varroosis, the most destructive diseases of honey bees, were investigated in bee colonies from 12 villages of the 7 districts in Sinop province for the first time. Nosemosis was the most common disease. Spore of the pathogen is the most characteristic stage in the examined bees. Light microscopic examinations of the living and dead bees revealed the presence of the pathogen spores in the intestine and the body cavity. Signs of infection spores were confirmed using the Giemsa staining technique (Figure 1). Mature fresh and Giemsa stained spores were measured. Mature fresh spores measured 4.46 \pm 0.22 x 2.53 \pm 0.21 (n=25) and Giemsa stained spores $4.12 \pm$ $0.49 \text{ x } 2.33 \pm 0.21 \text{ } \mu\text{m}$ (n=25).



Nosemosis was observed in 66 of 874 adult worker bees examined in Sinop province, and the average infection rate was determined as 7.6% (Table 1).

During the study, the disease was observed in eight (66.7%) of the examined 12 localities in Sinop province. While nosemosis was observed in Erfelek/Değirmencili and İnesökü, Ayancık/Abdülkadirköy and Atbaşı, Boyabat/Maruf, Durağan/Yandak, Çaktucak and Akçabük, the disease could not be observed in Dikmen/Karakoyun, Saraydüzü/Arım, Boyabat/Çarsak and Gerze/Kışla. The highest infections were determined Ayancık/Abdüladirköy, in Erfelek/İnesökü and Boyabat/Maruf with the infection rates, 37%, 17.4% and 6.0%, respectively. The highest infections in both live and dead bees was determined in Ayancık/Abdülkadirköy with the infection rates 36.5 and 37.5%, respectively (Table 1).

Figure 1. Nosemosis infection

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|----------|------------|--------------|----------|------------|-----------|----------------|
| Table L | Occurrence | of Nosem | 0515 111 | Sinon | nrovince | Tirkive |
| rable r. | Occurrence | 01 1 1050111 | 0515 111 | omop | province, | I unkiye. |

| | Living | | | Dead | | | Total | | |
|-----------------------|---------|-----------|---------|---------|-----------|---------|----------|-------|---------|
| Locality | bee | Nosemosis | (%) | bee | Nosemosis | (%) | Examined | Total | (%) |
| | samples | | | samples | | | Sample | | |
| Erfelek/Değirmencili | 56 | 1 | (1.8%) | 31 | 3 | (9.7%) | 87 | 4 | (4.6%) |
| Erfelek/İnesökü | 50 | 7 | (14%) | 19 | 5 | (26.3%) | 69 | 12 | (17.4%) |
| Dikmen/Karakoyun | 26 | 0 | - | 40 | 0 | - | 66 | 0 | - |
| Ayancık/Abdülkadirköy | 52 | 19 | (36.5%) | 40 | 15 | (37.5%) | 92 | 34 | (37%) |
| Ayancık/Atbaşı | 50 | 1 | (2%) | 24 | 1 | (4.2%) | 74 | 2 | (2.7%) |
| Saraydüzü/Arım | 58 | 0 | - | 10 | 0 | - | 68 | 0 | - |
| Boyabat/Maruf | 34 | 4 | (11.8%) | 50 | 1 | (2%) | 84 | 5 | (6.0%) |
| Boyabat/Çarsak | 52 | 0 | - | 4 | 0 | - | 56 | 0 | - |
| Durağan/Yandak | 50 | 0 | - | 52 | 6 | (11.5%) | 102 | 6 | (5.9%) |
| Durağan/Çaltucak | 54 | 1 | (1.9%) | 5 | 1 | (20%) | 59 | 2 | (3.4%) |
| Durağan/Akçabük | 54 | 0 | - | 7 | 1 | (14.3%) | 61 | 1 | (1.6%) |
| Gerze/Kışla | 52 | 0 | - | 4 | 0 | - | 56 | 0 | - |
| Total | 588 | 33 | (5.6%) | 286 | 33 | (11.5%) | 874 | 66 | (7.6%) |

Several studies have been carried out on the presence and distribution of nosemosis disease in different regions of Türkiye as well as in different parts of the world. However, there is no detailed study for the Sinop province. In the studies on the occurrence and distribution of nosemosis in Türkiye, different infection rates were found in the different localities. Recently, nosemosis was investigated in Bolu province (Yaman & Sağlam, 2023). In previous similar studies, the disease was found as 26% in Bursa province (Aydın et al., 2001), 15.74% in Kars province (Topçu & Aslan, 2004), 8.7% in Elazığ province (Şimşek, 2005), 6.5% in the Thrace region (Doğaroğlu & Sıralı, 2005), 100% in Muğla province (Şimşek, 2007), 10% and 21% in the Hatay province (Muz et al., 2012, Zerek, 2022), 44% in Ordu province (Yaman et al., 2015) and 43.18% in Siirt and Sanlıurfa provinces (Tel et al., 2021). In our study, we found that the average infection rate was 7.6% (Table 1), however it reaches to 37.5% in some bee colonies. When compared the highest infection rates of nosemosis recorded from some different regions before such as Ordu province (44%, Yaman et al., 2015), Hatay province (21%, Zerek, 2022), in Siirt and Şanlıurfa provinces (43.18%, Tel et al., 2021) and Muğla province (100%, Simsek, 2007), the nosemosis found in Sinop province is considerable low (7.6%). Similar result was found for Bolu province as 6.3%. Yaman and Sağlam (2023) stated that the low infection rate in Bolu province was due to climatic conditions. Two Nosema species, Nosema apis and Nosema ceranae that cause nosemosis are pathogens that are highly affected by climatic conditions, and their effects vary. This situation plays a decisive role in the spread of the disease in bee colonies. At high temperatures, N. ceranae show more virulence in honey bees than N. apis (Martin Hernandez et al., 2007).

On the other hand, varroosis was observed in the eight of 12 localities examined in Sinop province. Thirty-tree of the 1640 examined bee samples were infested by *Varroa* mites. Total infestation ranged from 0.6 to 8.2% (Table 2). The *Varroa* parasite in honey bees in Sinop province was identified as *Varroa destructor* (Figure 2).



Figure 2. Varroa destructor

The highest varroosis was determined in Saraydüzü/Arım, Dikmen/Karakoyun and Durağan/Çaltucak with the 8.2, 5.9 and 5.6% rates, respectively. The highest infestation in live bees was detected in Dikmen/Karakoyun with the 15.4% rate, and the highest infestation in dead bees was detected in Dikmen/Yandak with the 2.1% rate.

| | T • • | | F F | D 1 | j •• | | T 1 | | |
|-----------------------|--------------|-----------|---------|---------|-----------|--------|------------|-------|--------|
| | Living | | | Dead | | | Total | | |
| Locality | bee | Varroosis | (%) | bee | Varroosis | (%) | Examined | Total | (%) |
| | samples | | | samples | | | Sample | | |
| Erfelek/Değirmencili | 87 | 0 | - | 31 | 0 | - | 118 | 0 | - |
| Erfelek/İnesökü | 86 | 1 | (1.2%) | 19 | 0 | - | 105 | 1 | (0.9%) |
| Dikmen/Karakoyun | 26 | 4 | (15.4%) | 42 | 0 | - | 68 | 4 | (5.9%) |
| Ayancık/Abdülkadirköy | 146 | 0 | - | 167 | 2 | (1.2%) | 313 | 2 | (0.6%) |
| Ayancık/Atbaşı | 97 | 0 | - | 24 | 0 | - | 121 | 0 | - |
| Saraydüzü/Arım | 86 | 8 | (9.3%) | 12 | 0 | - | 98 | 8 | (8.2%) |
| Boyabat/Maruf | 41 | 0 | - | 98 | 0 | - | 139 | 0 | - |
| Boyabat/Çarsak | 160 | 2 | (1.3%) | 2 | 0 | - | 162 | 2 | (1.2%) |
| Durağan/Yandak | 132 | 8 | (6.1%) | 97 | 2 | (2.1%) | 229 | 10 | (4.4%) |
| Durağan/Çaltucak | 84 | 5 | (6.0%) | 6 | 0 | - | 90 | 5 | (5.6%) |
| Durağan/Akçabük | 85 | 1 | (1.2%) | 8 | 0 | - | 93 | 1 | (1.1%) |
| Gerze/Kışla | 100 | 0 | - | 4 | 0 | - | 104 | 0 | - |
| Total | 1.130 | 29 | (2.6%) | 510 | 4 | (0.8%) | 1.640 | 33 | (2.0%) |

Table 2. Occurrence of Varroosis in Sinop province, Türkiye.

Albayrak (2019) conducted a survey with beekeepers in Sinop. According to the results of the survey, while varroa was the most common pest in Sinop, other diseases were very rare. However, in our study, it was found that 66 of 874 bees examined in Sinop province were infected with the Nosema pathogen. In addition, infection rate was 7.6%. Albayrak (2019) reported that 71% of beekeepers only encountered varroa pests. In our study, varroosis was observed in eight of the 12 examined locations. It could not be observed locations in 4 (Erfelek/Değirmencili, Ayancık/Atbaşı, Boyabat/Maruf and Gerze/Kışla). It was determined that the lack of care in the colonies in which the presence of varroosis was observed may cause an increase in Varroa mite infestation. In addition, it was observed that the bees infested with Varroa parasite had a decrease in body weight and deformations in their abdomen and wings.

The ectoparasite mite, V. destructor has caused severe damage to honeybee colonies (Salkova & Gurgulova, 2022). One of the most common and obvious reasons of colony collapse is varroosis with the 38% infestation (Meixner & Büchler, 2015). As in nosemosis disease, the occurrence and distribution of varroa mites have been investigated in different regions of Türkiye. In last two decades, the diseases was found as 13.32% in Cukurova region (Kumova, 2001), 89% in Blacksea region including Sinop, Samsun, Ordu, Giresun, Trabzon, Rize ve Artvin provinces (Yaşar et al., 2002), 1.2-13.5% in Ankara province (Kar et al., 2006), 5% in Ordu province (Kuvancı et al., 2013), 23.8% in Düzce province (Kekeçoğlu et al., 2013), 100% in Kars province (Önk & Kılıç, 2015), 38% in Marmara Island-Balıkesir province (Seven Çakmakçı, 2017) and 33.92% in Bingöl province (Kesik et al., 2022). Our study revealed that varroosis occurs in similar infection rates with all investigated regions of Türkiye with the exceptions, Kars and Bingöl provinces. Varroosis was found significantly high rates in Kars and Bingöl provinces. These high infestations in the two provinces can be explained by with geographic location differences. Meixner and Büchler (2015), stated that when compared to Varroa infestations in different apiaries, infestation was significantly affected by the apiary location rather than the genetic origin of the colony.

Conclusion

During the study, nosemosis and varroosis were detected in eight (66.7%) of the examined twelve localities in Sinop province. These data are considered as proof that honey production is negatively affected. The effect of N. ceranae varies considerably according changing environmental conditions. to Therefore, the nosemosis agent in Sinop province should be identified at the species level. This study also revealed that the beekeepers in the Sinop province should be informed in detail about the precautions to be taken in the monitoring and controlling varroosis and nosemosis.

Ethics Committee Approval

N/A

Peer-review

Externally peer-reviewed.

Author Contributions

Conceptualization: M.Y.; Investigation: M.Y. S.Ü.; Material and Methodology: M.Y., T.S.; Writing-Original Draft: M.Y., T.S.; Writing review & Editing: M.Y. S.Ü.; Other: All authors have read and agreed to the published version of manuscript.

Conflict of Interest

The authors have no conflicts of interest to declare.

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