

HONEY BEE VIRUSES, DISEASES AND HIVE MANAGEMENT IN THE MIDDLE EAST AND THEIR RELATION TO THE COLONY COLLAPSE DISORDER AND BEE LOSSES

Orta Doğuda Balarısı Virüsleri, Balarısı Hastalıkları, Koloni Yönetimi ve Bunların Koloni Kayıpları ve Koloni Çökme Bozukluğu ile İlişkisi

(Genişletilmiş Türkçe Özet Makalenin Sonunda Verilmiştir)

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ABSTRACT

Beekeeping industry plays a pivotal role in the agricultural, food security, biodiversity and national economies, not only by giving various hive products but most importantly is the high impact that the honeybees are making in the crops and wild flora pollination. Since the year 2007 most of the beekeepers around the world had face a very big colony losses and unusual incident of disappearance of the bees "CCD", this article is trying to spotlight some reasons behind both CCD and bee losses in the Middle East area. No clear correlation between a single reason and CCD was found, but several vectors did have a clear relationship with the bee losses in the Middle East area. It is clear that beekeepers suffered big bee losses over the last few years, and until now there is no clear and scientifically proven explanation to CCD.

INTRODUCTION

Status of the CCD and bee losses in the Middle East area.

The beekeeping industry plays a pivotal role in the agricultural sector; its importance is not only hive products such as honey, pollen, royal jelly, venom, production of queen bees, package bees and value added products. The main and the vital role of beekeeping is its fundamental importance in increasing the yield and improving the quality of agricultural crops via pollination, where honeybees transport the pollen from the another of the flower of one plant to the flowers of a different plant, which is known as cross-pollination. It is very important to emphasize that honey bees play a crucial role in the conservation and preservation of the wild plants' biodiversity since they pollinate most of the

cross-pollinating wild flora, in light of the decline in the wild insect pollinators such as carpenter bees (*Xylocopa ssp.*), leaf cutter bees (*Osmia spp.*), bumble bees (*Bombus ssp.*), wild solitary bees,

butterflies, wasps, other insects and wild animals. The increase in urban expansion, practices of intensive farming and misuse of pesticides and herbicides made the spread of wild insect pollinators very limited and confined on the non-cultivated areas, farms edges and marginal areas. This restricts the spread of other pollinators, except honeybees far from the targeted crops. The results of a recent study of the Bee Research Unit (BRU) have shown that the value of total production of the twelve crops pollinated by honey bees, reached \$ 117.5 million in 2005, and increased production due to the direct inoculation of plants with a value of \$ 50.7 million annually. This increase is more than 16

times the value of the annual domestic production of honey (\$ 3.1 million). In 2000, Drs. Roger Morse and Nicholas Calderone of Cornell University, attempted to quantify the effects of one pollinator, the Western honeybee, on only US food crops. Their calculation came up with a figure of US \$14.6 billion in food crop value.

The winter of 2006/2007 witnessed large-scale losses of managed honey bee (*Apis mellifera* L.) colonies in the United States. Those losses continued into the winter of 2007/2008, much less in 2008/2009 and it seems the CCD is again massive in 2009/2010. In the U.S., a portion of dead and dying colonies were characterized "post hoc" (by a rapid response group comprised of academic, private, and Federal scientists), by a common set of specific symptoms: (1) the rapid loss of adult worker bees from affected colonies as evidenced by weak or dead colonies with excess brood populations relative to adult bee populations; (2) a noticeable lack of dead worker bees both within and surrounding the affected hives; and (3) the delayed invasion of hive pests (e.g., small hive beetles and wax moths) and kleptoparasitism from neighbouring honey bee colonies. Subsequently, this syndrome has been termed Colony Collapse Disorder, or CCD.

This phenomenon had an extensive media coverage that led to a big reaction by the US Congress. Because the losses exceeded the normal thresholds of honeybee mortality, millions of dollars were allocated to support specialized research in the field of bees. Concern ranged from the decrease of bee products and honeybee populations down to major problems in the production of field crops production, dependent wholly or partially upon bees as pollinating agents. Almond growers in California concerted to put pressure on the U.S. Congress, which led to the preparation of research programs supported by exceptionally attention from the U.S. government.

In parallel and in the same context, the European Union supported the establishment of an International European Network "COLOSS" to study this phenomenon. Researchers involved in different fields of science, such as Biotechnology, Microbiology, Virology, Plant Protection and many other related fields, gave the network a very strong structure and very wide background. This network preferred to use the term "Bee Losses" since not every dying or dead colonies in the European

Continent had the same three common CCD syndrome symptoms.

In the Arab world, especially in the Middle East, some massive bee death was noted for the years 2007–2009. These were the most difficult years for beekeepers, since tremendous climatic changes had happened during these three years: the area faced very cold winters and a big drop in the rainfall. In addition, some unusual declines occurred to the local honeybee populations in many areas. Some of these were directly attributed by the beekeepers as CCD, because of the role that the media had played, in addition to the big effect of unscientific rumours that were spread between beekeepers throughout the region. According to many research experiments and cases monitoring, the Middle East faced a big drop in the bee population, but not all losses were identical to the USA CCD syndrome.

Many hypotheses were proposed to explain this phenomenon. Some of them got huge public support even though they were not the results of scientific research. However, the media had played a crucial role in this issue. Some of the suggestions proposed that GMO "genetically modified crops" are responsible for this phenomena, others blamed cell phones microwaves and antennas, others suggested that the nanotechnology is responsible, while others proposed that climatic change is the driver of this problem.



Foto 1:Varroa mite infestation, **Foto 2:** Colony losses

Several studies were investigating the potential causes of this specific syndrome (CCD). Among these some studies, you can find a statistical relationship between CCD and nosema, varroa, chemi-

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cals and more. One study, which blindly compared all of the nucleic acids extracted from CCD and non-CCD hives, concluded that IAPV is strongly associated with CCD (Cox-Foster, et al. 2007).

Still, it is not clear what the direct cause of CCD is and whether all the suspects are markers or causative agents. According to our review of the CCD published research and reports, we think that there are few reasonable hypotheses which can explain the causative of CCD, and the hypotheses are built on the accuracy of the conclusions of the study published by Cox-Foster, *et al.* 2007.

If Cox-Foster paper is correct, a specific strain of IAPV infectious virus is the cause of CCD. If this hypothesis is accurate, then IAPV virus can be distinguished from different strains, which do not cause CCD. Therefore, further research is needed to study whether the IAPV virus that was found in the CCD colonies belongs to one or more strains. This paper didn't investigate whether the IAPV sequence is viral or integrated into the bee genome. Thus, it has found that IAPV sequence is strongly associated with CCD. Therefore, it may be that the IAPV integrated segment in the bee's genome was detected and this is the cause of the CCD. Integration into an important immune-gene or immune regulation gene or navigation gene-may harm the immune response and cause CCD. In that case, integration of IAPV will cause a deficient immune response and any stress (nosema, varroa...) will cause CCD. This hypothesis questions whether an integration of IAPV may harm the bee's immune response (or navigation rather than immune-gene?) and cause CCD.

If Cox-Foster paper is not correct, then, we can conclude that 1) CCD is a dangerous disease which is triggered in some bees by any stress (varroa, IAPV, nosema, chemicals) and then the disease emerges—CCD, or 2) a complex of pathogens interacting together will cause a unique condition for a syndrome-CCD.

Focus should not only be on the CCD but also on bee losses, since not every dead colony had CCD. During the Bee Research Unit search for explanations for the bee losses in the Middle East, we were able to find several vectors that are clearly correlated with the dying colonies, but not necessarily with the colonies that have the CCD symptoms. The results of our monitoring studies had shown some logical explanations to the bee population decline but not directly to the CCD phenomena. All

the research on the CCD colonies is coming post the problem while the declining bee's populations inside the bee colonies come before the CCD and before the colony mortality. Surveying and questioning apiaries and beekeepers in the Middle East had led us to gain very important information about the obstacles facing beekeeping in the Arab world. It is very important to state that there is a lack of coordination between the research centres, and the ministries of agriculture within a country and between countries. But it was very clear that most of the beekeepers of all the Middle Eastern countries have been facing very similar problems.

The mortality level can be considered very high during the last few years, and it has been at this high of a level only during 1985–1987, when the varroa mite was discovered and recognized in the Middle Eastern countries. Over 50% of the colonies died in the 80s because no varroa treatment was available in that time, and some of the treatments themselves led to the death of the colonies in the 80's.

In the years 2007-2009 official data from the Ministry of Agriculture in Lebanon showed that beekeepers in Southern Lebanon lost over 90% of their colonies in Rashayya and the western Bekaa. Beekeepers had more than 3600 beehives in this region during the 2007-2008 period (Report of the Lebanese daily As-Safir) but no scientific research was done on these bees. Some reporters attribute the loss of colonies to the war in Southern Lebanon, which prevented beekeepers from inspecting their colonies for over three months, in addition to the chemical pollution that appeared in the area during and after the war. Losses in Syria in 2007 were 50% (Dr. Alburaqi A., Damascus University). Perhaps the biggest losses occurred in Iraq, where the city of Halabka has lost more than 90% of its bee colonies (Mustafa I., Arabiel). He also reported that some beekeepers in Alnagaf and Al-Dewaneah provinces lost approximately 75% of their bees during 2008 (Hasnawi M., Al-Dewaneah). The Iraqi beekeeping experts did not give any explanations for these losses except for areas that had shown a high level of hive mortality, and had a high level of noise pollution because of the ongoing war in Iraq. The Bee Research Unit has received numerous contacts to assist in the interpretation of this phenomenon, which was repeated in most areas of northern Iraq where they had no war and they had 25%-30 % bee losses. However, the lack of research networks across

Arab countries limited our abilities to give clear explanations to each of the cases of colony losses in the region.

Reviewing the results of the research during the past few years, we can consider several vectors as drivers of the bee losses some of which are unique to the Middle East.

1. Viral diseases: Honey bees are infected with more than 18 viruses, BRU research in 2007-2008 found that most of the dying and dead colonies were infected with Deformed Wing Virus (DWV), sac brood virus (SBV), acute bee paralysis virus (ABPV), Israeli acute paralysis virus (IAPV). It has been found that the infection with DWV was the highest compared to all other viruses, but the virus IAPV was found in some of the dying colonies, we cannot say that any of these viruses or all of them together are responsible for the colony losses, since surveys of some of the healthy looking colonies showed the infection with these viruses, but almost every weak or dying colony did show a complex of multiple viruses infection.

2. Varroa mite: The varroa mite, *Varroa destructor*, is currently considered the major pest of honeybees in most parts of the world. The pathology it causes is commonly called varroosis (also called varroatosis or varrosis). Initially discovered in Java, varroa was originally confined to Southeast Asia where it parasitizes the Asian honeybee, *Apis cerana*. This bee has probably coevolved with the parasite, and adapted to keep the mite under control. A post-World War II increase in international travel and commerce has facilitated the worldwide dispersal of varroa. Once established, the mite spreads on drifting, robbing, and feral bees, or swarms. Varroa mite was recorded in Israel in 1980 and officially in Jordan in 1986, and as of 1987 has become an economic concern in all the Middle Eastern countries. Jordanian beekeepers lost over 50% of their bee population. In 1990, varroa mite was reported in all the Arab countries both in the Middle East and in North Africa.

Because it is impossible to eradicate varroa even from a closed population (Sampson & Martin 1999), beekeepers must manage the mite populations within their own colonies. Keeping its level to the minimum has become the main goal of its control. The results of the Bee Research Unit of the National Centre for Agricultural Research and Extension show that in the years 2005-2008, a

large proportion of varroa in Jordan has become immune to varroa chemical treatments available in the local markets. These results were very similar to results of research done by Dr. Al Rose Hisham, Damascus University, Syria. Treatment of varroa mites with the active-loaded "coumaphos" proved to be effective, however, clear evidence of wax and honey contamination make its use illegal according to standards of the European Union. Therefore, great scientific debates have arisen between United States and European experts on the legality of its use, since this pesticide is used in the USA in the control of both varroa mites and small hive beetle. Some oils and acids were used such as thymol, but it is very hard to apply these treatments in areas with high temperature, which is the case in most Arab countries.

We can not conclude that varroa is a direct reason for CCD and bee losses in the Middle East, since it was there for a long time, but we can say it is a very important factor that disturbs the health status of the bee colony.

3. Nosema: Nosema disease (*nosemosis*), the original causative organism of which was identified as the unicellular microsporidium *Nosema apis* about a 100 years ago (Zander, 1909), is considered to be one of the most economically damaging of diseases of the *Apis mellifera*. However, because of its microscopic size, it is very difficult for beekeepers to determine the disease infection level except in severe cases when the symptoms of the nosema disease are seen by the bee's defecations on the hive surface. Usually *N. apis* appears and disappears unnoticed, especially in hot climates, except in the rare of severe infection, which leads to the death of a diseased colony. During the last decade or so, *Nosema ceranae* emerged as a pathogen of the honey bee (*Apis mellifera*). Until now, its origin and date of spread are unclear. Though it has been dismissed as a cause of CCD in the USA based on correlation analyses of snapshot sampling of diseased hives, observations of naturally infected colonies suggest that it leads to colony collapse in Spain.

Robert J. Paxton (2010) gives a very important discussion of this issue in his article entitled "Does infection by *Nosema ceranae* cause Colony Collapse Disorder in honey bees *Apis mellifera*" where he noted that the detailed metagenomic survey of CCD affected colonies of *A. mellifera* in the USA (Cox-Foster et al., 2007) recognized *N.*

ceranae as a potential causative agent of CCD but statistically ruled it out as the primary agent responsible for CCD. It is worth considering the results of this study in more detail. Of 30 CCD affected colonies, all were positive for *N. ceranae*. 10 of 21 (47%) non-CCD affected colonies were also positive for *N. ceranae* (see Table 2 of Cox-Foster et al., 2007). Statistically, the presence of *N. ceranae* in a colony was not a good predictor of whether the colony had collapsed. As the authors themselves are, careful to point out, however, their metagenomic survey may be inappropriate for determining the cause of CCD (Cox-Foster et al., 2007). Firstly, it was a correlational study and, secondly, it only took a "snapshot in time" of the prevalence of disease organisms in colonies. Disease organisms build up over time (i.e. increase in larval / adult incidence of infection) before causing colony mortality, and generally do not act instantaneously. As *N. ceranae* has been reported to build up in prevalence within a colony over an 18 month period before causing colony demise (Higes et al., 2008; 2009b), the dynamic nature of this and other infectious agents cannot be captured by a snapshot analysis of disease organisms in colonies at one point in time. The study of Cox-Foster et al. (2007) therefore still leaves open the possibility that *N. ceranae*, alone or in combination with other factors, causes CCD. According to the clear discussion of the issue above, we can not point to nose-ma as a direct reason for CCD. No correlation was found in the Jordan survey between the colonies with CCD symptoms and nose-ma disease, since we were able to find it in collapsing, weak and healthy looking colonies. Fewer than 20% of Jordanian beekeepers are using Fumidil as a prophylactic treatment, but both beekeepers who use and those not using it had collapsed colonies.

4-Management: A common maxim among beekeepers says, "The main pest of honey bees is the beekeeper". This maxim gives a very true explanation for bad and poor management since many of the bee diseases are transferred and caused by the beekeepers themselves. We can not say that the CCD happens because of the beekeepers management since it may happen in the same apiary with both healthy and dying colonies. In the following points we present the main management problems that had shown an impact on the colony losses in Jordan.

a. Imported bees: Jordanian beekeepers import packaged bees and nuclei of bees from countries

like Egypt, because of the low prices of the honeybees in Egypt in comparison with Jordan. It was very clear that the Egyptian honeybee (*Apis mellifera lamarckii*) cannot adapt to the Jordanian local conditions, and about 60% of the imported packaged bees die within 3 to 4 months of importation. The local breeding of local strains in each of the Arab countries will prevent the transportation of the honeybee diseases between the countries and prevent the disappearance of local strains that are adapted to the local conditions. Local strains in the Arab world are Yemeni honeybee (*Apis mellifera yemenica*), Syrian bees (*Apis mellifera syriaca*), Nubian bees (Sudan) (*Apis mellifera nubica*), Tellian honeybee (*Apis mellifera intermissa*), Egyptian honeybee (*Apis mellifera lamarckii*), and the African honeybee (*Apis mellifera scutellata*). The most imported bee races in many of the Arab countries are the Italian honeybee (*Apis mellifera ligustica*), and Carniolan honeybee (*Apis mellifera carnica*).

b. Pollen supplements and substitutes: The months of July–September are dry and hot in the Middle East. This affects pollen availability. Beekeepers use some pollen patties to supply honeybees with protein. Our experiments in 2006 and 2007 had shown that many of the beekeepers feeding their colonies on pollen, which was not irradiated with gamma radiation, complained of high levels of infection with American foulbrood disease and colony losses.

c. Queening: There is a direct correlation between the colony performance and yearly replacement of the queens. We found that beekeepers who replace the queens yearly had less of a problem with colony losses.

d. Dark frames management: There is a direct correlation between the old frames and the colony losses. 40% of the beekeepers who did not replace the old frames encountered high levels of mortality and weak colonies.

CONCLUSION

Over a million electronic documents related to CCD are available online via the Google internet search engine; those, fewer than a thousand documents are available on the Google Scholar search engine. This gives a clear indication that most of the available information online is from the media. It is clear that the media has exaggerated the CCD syndrome, but this does not mean that the problem did

not exist. However, this exaggeration has benefited the environment, beekeepers, researchers, scientists and even bees themselves. Big financial support in different countries of the world was allocated to study this issue. An interest from environmentalists and ordinary people is heightened due to the media reports. It is clear that beekeepers suffered big bee losses over the last few years, and until now there is no clear and scientifically proven explanation to CCD. However, the conducted research to explain this syndrome has helped bee researchers understand what may affect the health of the honeybee. Therefore, further cross-border and cross-continent research projects need to be conducted in order to find clear explanations for this syndrome.

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GENİŞLETİLMİŞ ÖZET

Amaç: Bu derlemede Orta Doğu'daki arı ölümleri araştırılmış ve geniş kapsamlı literatür çalışması ile bilimsel bulgular ortaya konmaya çalışılmıştır.

Giriş: Arıcılık endüstrisi sadece kovan ve kovan ürünlerinden ibaret değildir. Arıların ekolojide oynadıkları en önemli rollerden birisi bitkilerin ya da tarımsal ürünlerin tozlaştırılmasında oynadıkları rollerdir. Özellikle diğer biyolojik arı zenginliğinin - marangoz arılar (*Xylocopa* türleri), yaprak kesici

arılar (*Osmia* türleri) ve bombus arıları (*Bombus* türleri)-azalması, balarılarının önemini daha da arttırmıştır. Özellikle tarım alanlarında balarısı kolonilerinin taşınabilme özelliği bu böceğe olan talebi arttırmaktadır. Arıların ekonomiye olan katkıları ABD'de yapılan bir araştırmaya göre 14.6 milyar dolar civarındadır. Ancak ABD'de 2006 yılından başlayarak günümüze kadar balarısı kolonilerinde azalma gözlenmektedir. Sebepleri tam olarak anlaşılamasa da bu durum şu şekilde ifade edilmektedir: 1) yetişkin arı sayısında ani düşme, 2) koloni etrafında ölü arıların olmayışı ve 3) diğer parazitlerin bu kovanlara geçişinin uzun sürmesidir. Bu durum Koloni Çökme Bozukluğu (Colony Collapse Disorder) olarak adlandırılmıştır.

ABD ve tüm dünya medyasında geniş yer alan bu konu, Avrupa'da COLOSS grubunun oluşmasına neden olmuş ve tüm Avrupa ülkeleri birleşerek bu ölümleri araştırmaya başlamıştır. 2007-2009 arasında ABD'deki kadar olmasa da Arap dünyasında özellikle de Orta Doğu'da arı ölümleri yaşanmıştır. Bir çok neden GMO'lardan tutun, mikrodalgalara, cep telefonu baz istasyonlarına, nanoteknolojiye ve iklim değişikliklerine kadar herşey bu ölümlerden dolayı suçlanmıştır.

Birçok bilimsel çalışmaya göre Koloni Çökme Bozukluğu ile birçok neden arasında ilişki bulunmuştur, bunlar; nosema, varroa, ilaç olarak kullanılan kimyasallar ve diğerleri olarak sıralanmaktadır. Bir çalışmaya göre İsrail Akut Paraliz virüsü ile KÇB arasında yüksek ilişki bulunmaktadır. Ancak kesin sebebi bu denilememiş ve birçok diğer neden daha ortaya konmasına rağmen bu durumu değiştirmemiş ve arı ölümleri devam etmiştir. Ölümler Orta Doğu'da 1985-1987 yılları arasında Varroa'nın görülmesinden bu yana ilk defa bu kadar yüksek seviyelere çıkmıştır. O zamanlar Varroa ile mücadele bilinmediğinden ölümler %50'ye varmıştır. Orta Doğu'da şimdiki ölümlere bakacak olursak güney Lübnan'da %90, Suriye'de %50 ve Irak'ta %90 ile en fazla ölümlerin yaşandığı ülke olmuştur.

Tüm elde edilen bulgular değerlendirildiğinde Orta Doğu'da birçok neden ön plana çıkmaktadır. Bunlar;

1-Viral Hastalıklar: 18 viral etmen olmasına rağmen 2007-2008 yıllarında ölen kovanlarda en çok Deforme kanat Virüsü (DWV), Yavru Kese Virüsü (SBV), Akut Arı Paraliz Virüsü (ABPV) ve İsrail Akut Paraliz Virüsü (IAPV) bulunmuştur. Fakat sadece bunlar ölümlere neden olmaktadır demek yanlıştır

çünkü ölen ya da ölmekte olan kolonilerde bu virüslerin kombinasyonları yer almaktadır.

2-Varroa: Orta Doğu'da varroa ilk 1980'da İsrail'de görülmesinden sonra 1987 yılında tüm Orta Doğu'da sorun haline gelmiştir. Sonraki yıllarda birçok kimyasal bu parazitin mücadelesinde kullanılmıştır. Koloni Çökme Bozukluğu ile direk bir ilişki kurulamamasına rağmen koloni sağlığını yakından etkileyen etmenlerden birisidir.

3-Nosema: Diğer etmenlerde olduğu gibi direk ilişki kurulamamış olmasına rağmen başlangıçta ilk nedenlerden biri olarak gösterilmiştir. Sonraki çalışmalar ise bunun doğru olmadığını göstermiştir. Ancak 2 farklı sporun varlığı tespit edilmiştir.

4-Koloni yönetimi: Arıcılar arasındaki genel düşünce "balarılarının en temel düşmanı arıcının kendisidir" çünkü kötü koloni yönetimi arılığa ve kovanlara tüm olası etmenleri hastalıkları, parazitleri, virüsleri ve tüm kötü durumları getirmektedir. Ürdün'de koloni yönetimine etki yapanlar arasında a) dışarıdan gelen arılar, b) polen yerine kullanılan malzemeler, c) ana arı değişimi, d) eski çerçevelerin kullanımı üzerinde durulmaktadır. Tüm bu sayılanlar direk olmasa da koloni sağlığını etkilemekte ve Koloni Çökme Bozukluğuna neden olduğu düşünülmektedir.

Sonuç: İnternette Koloni Çökme Bozukluğunu araştırdığımızda bir milyondan fazla doküman bulunduğu görülmektedir; bunlardan sadece bin tanesine ulaşılabilmektedir. Bu da bize medya hakkında bilgi vermekte ve medya tarafından koloni çökme bozukluğunun abartıldığı ortaya çıkmaktadır ancak bu durum arı ölümlerinin olmadığı anlamı taşımamaktadır. Fakat bu abartıdan herkes payını almıştır, çevre, arıcı, bilim adamları, araştırmacılar hatta arılar dahil. Farklı dünya ülkelerinde büyük paralar bu durumun araştırılmasına ayrılmıştır. Çok açıktır ki arıcılar son bir kaç yıldır kayıpları yaşayanlardır ve maalesef günümüze kadar bu ölümler hakkında bilimsel olarak kesin bir neden tanımlanamamıştır. Bununla beraber yapılan araştırmalar arı araştırmacılarına balarısı kolonisinin sağlığını nelerin etkilediğini anlamalarına neden olmuştur. Dolayısı ile bu durumun daha detaylı araştırılabilmesi için ülkeler hatta kıtalar arası ortak araştırmaya ihtiyaç vardır.