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# ARAŞTIRMA MAKALESİ / RESEARCH ARTICLE

# THE ROLE OF INVERTED SUGAR SYRUP IN ENHANCING SEALED BROOD QUANTITY AND BEE COLONY PRODUCTIVITY IN WARRE HIVES

Warre Kovanlarda İnvert Şeker Şurubunun Kapalı Yavru Miktarını ve Arı Ailesinde Verimliliği Artırmada Rolü

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#### **ABSTRACT**

The primary aim of this study is to evaluate the impact of inverted sugar syrup supplementation on sealed brood quantity and colony strength in Warre hives, compared to traditional sucrose syrup feeding. The experiment consisted of 14 honey bee colonies, equally distributed between the control group (n = 7) and the experimental group (n = 7). The bee colonies were established at an apiary near Tran, Pernik region, Bulgaria. Our study shows that bee colonies fed inverted sugar syrup had significantly higher average values of sealed worker brood and colony strength compared to those in the control group from April to October in Warre hives. While these short-term results are promising and positive, long-term studies could reveal more about the lasting effects on bee health and productivity in this hive system. These results can help beekeepers choose better feeding strategies to maintain healthy and strong bee colonies in regions with insufficient pasture.

Keywords: Honey bees, Inverted sugar syrup, Bee colony strength, Sealed worker brood, Warre hives

## ÖZ

Bu çalışmanın temel amacı, invert şeker şurubu takviyesinin Warre kovanlardaki kapalı yavru miktarı ve koloni gücü üzerindeki etkisini, geleneksel sakkaroz şurubu ile beslemeyle karşılaştırarak değerlendirmektir. Deney, kontrol grubu (n=7) ve deney grubu (n=7) arasında eşit olarak dağıtılmış 14 bal arısı kolonisi ile gerçekleştirilmiştir. Arı kolonileri, Bulgaristan'ın Pernik bölgesi, Tran yakınlarındaki bir arılıkta kurulmuştur. Çalışmamız, Warre kovanlarda invert şeker şurubu ile beslenen arı kolonilerinin, Nisan'dan Ekim'e kadar olan dönemde, kontrol grubundakilere kıyasla kapalı işçi yavrusu ve koloni gücü ortalama değerlerinin anlamlı derecede daha yüksek olduğunu göstermektedir. Bu kısa vadeli sonuçlar umut verici ve olumlu olsa da, uzun vadeli çalışmalar bu kovan sistemindeki arı sağlığı ve verimliliği üzerindeki kalıcı etkiler hakkında daha fazla bilgi ortaya çıkarabilir. Bu sonuçlar, arıcılara yetersiz mera bulunan bölgelerde sağlıklı ve güçlü arı kolonilerini sürdürmek için daha iyi besleme stratejileri seçmelerine yardımcı olabilir.

Anahtar Kelimeler: Bal arısı, İnvert şeker şurubu, Arı ailesinin gücü, Kapalı işçi arı gözü, Warre kovan

## **GENIŞLETILMIŞ ÖZET**

Amaç: Bu çalışmanın temel amacı, invert şeker şurubu takviyesinin Warre kovanlardaki kapalı yavru miktarı ve koloni gücü üzerindeki etkisini, geleneksel sakkaroz şurubu ile beslemeyle karşılaştırarak değerlendirmektir.

Gereç ve Yöntem: Warre kovanlardaki koloni performansı üzerindeki invert şeker şurubu takviyesinin etkilerini değerlendirmek amacıyla, 2024'e kadar Nisan'dan Ekim kontrollü karşılaştırmalı bir çalışma yürütülmüştür. Deney, kontrol grubu (n=7) ve deney grubu (n=7) arasında eşit olarak dağıtılmış 14 bal arısı kolonisi içermiştir. Arı kolonileri, Bulgaristan'ın Pernik bölgesi, Tran vakınlarındaki (rakım 860 m) bir arılıkta konumlandırılmıştır. Kontrol grubu, (ağırlık/ağırlık) oranında sakkaroz çözeltisi (%50'si su içinde) ile beslenmiştir. Deney grubuna atanan arı kolonileri ise, sakkarozun kontrollü inversiyonu (dönüşümü) yoluyla hazırlanan şurubu almıştır. Kontrol ve deney grupları, sakkaroz çözeltisini ve invert şeker şurubunu zaman ve hacim açısından aynı şekilde uygulamışlardır. Besleme uygulaması iki dönemde yapılmıştır. Birinci dönem bahar teşviki ve ikinci dönem ise sonbahar beslemesiydi. İnvert hidroksimetil şuruptaki furfural (HMF) konsantrasyonu spektrofotometrik olarak belirlenmiştir. Deney ve kontrol grupları arasındaki kullanılarak karşılaştırmalar Student's t-testi yapılmıştır.

Bulgular: 15 Nisan'da (p<0.001), 27 Nisan'da (p<0.01), 9 Mayıs'ta (p<0.05) ve 21 Mayıs'ta (p<0.001) deney grubunun ortalama kapalı işçi yavrusu miktarı ile kontrol grubu arasında anlamlı farklılıklar bulunmuştur. Bahar dönemi için en yüksek kapalı işçi yavrusu alanı, deney grubunda 2 tarihinde (ortalama 24.157 kaydedilmiştir (Şekil 1). Bu tarihteki ortalama değerler istatistiksel olarak anlamlıdır (p<0.01). Sonbahar beslemesinde ise, 5 Ağustos'tan 4 Ekim'e kadar olan tüm tarihlerde deney grubundaki ortalama kapalı işçi yavrusu değerleri, kontrol grubuna kıyasla istatistiksel olarak anlamlı bir artış (p<0.001) göstermistir. İnvert seker surubu ile beslenen arı kolonileri, her iki test tarihinde de (Nisan ortası ve Ekim ortası) kontrol kolonilerine göre daha yüksek koloni gücü (gram cinsinden arı ağırlığı) sergilemiştir. Çalışmamız, invert şeker şurubu ile beslenen arı kolonilerinin, Warre kovanlarda Nisan'dan Ekim'e kadar kontrol grubundakilere kıyasla anlamlı derecede daha yüksek ortalama kapalı işçi yavrusu ve koloni gücü değerlerine sahip olduğunu göstermiştir.

Sonuç: Warre kovanları, doğal arı habitatını taklit etmek üzere tasarlanmıştır. Bu tasarım, arıların doğal olarak petek yapmasını tesvik ederek, yavru gelişimi için istikrarlı bir ortam sağlar. Sık sık yapılan kontrollerden kaçınılması, arı kolonisi üzerindeki stresi azaltır, ancak arıcıların müdahalesini sınırlar. Bu çalışmada, sonuçlar bal arılarının şeker şurubu ve invert şeker şurubunu yoğun olarak tükettiğini ortaya koymuştur. Şeker şurubu daha kolay sindirilebilir olduğu için, arıları daha fazla kaynağı üretimine ayırmaya teşvik düşünülmektedir. Bu kısa vadeli sonuçlar umut verici olsa da, Warre kovanlardaki bal arısı kolonilerinin genel durumu ve üretkenliği üzerindeki kalıcı etkileri belirlemek için uzun vadeli çalışmalara ihtiyaç vardır. Bu bulgular, arıcılara yetersiz mera bulunan bölgelerde sağlıklı ve güçlü arı kolonilerini sürdürmek için daha iyi besleme stratejileri seçmelerine yardımcı olabilir.

#### INTRODUCTION

It is evident that honeybees play a crucial role in preserving biodiversity in plants and maintaining the sustainability of ecosystems as pollinators (Hung et al. 2018). Many factors and stressors have been associated with bee colony losses in the last decades including pesticides, pathogens, parasites and nutritional limitations (Hsieh et al. 2024, Mayack et al. 2022). Honey bee nutrition is complex and depends on the environment. Honey bees rely on floral resources such as nectar and pollen and stored food (honey, bee bread) to meet their nutritional needs (Tsuruda et al. 2021, Wright et al. 2018). Nutrition in beekeeping is very important for maintaining honey bees' strong and healthy. Honey bees need a lot of different nutrients to grow and develop, such as carbohydrates, proteins, lipids, mineral elements, vitamins, and water (Frizzera et al. 2020). Carbohydrates serve as a critical energy source for sustaining bee colonies, rearing brood, and overall productivity (Ansaloni et al. 2025). Proper supplementation with appropriate sugar solutions can enhance bee health, improve foraging efficiency, and ultimately increase bee colony population density, supporting ecosystem pollination services (Mogren et al. 2018, Sammataro and Weiss 2013). The feeding of bee colonies is necessary when natural resources are not sufficient and do not meet the needs of the colony at a particular time or

area. Carbohydrates may even reduce the number of bees that die over winter (Cotter et al. 2011, Quinlan et al. 2023).

In the context of contemporary beekeeping practices, the provision of carbohydrates as a substitute for nectar has become essential. The effects of 65% sucrose and 65% invert sugar syrups on the survival and enzyme activity of caged honey bees were compared (Pavlovic et al. 2025).

Beekeepers often use sugar syrup to supplement their colonies' food supplies. Artificial sugar feeding is commonly used in beekeeping practices. Nowadays, different kinds of carbohydrate products are available on the market, such as: inverted sugar syrups, starch syrups (Szczęsna et al. 2021), and high fructose corn syrups (HFCSs) (Lazarov et al. 2025, Johnson et al. 2014, Ruiz-Matute et al. 2010, Wheeler and Robinson 2014) are commonly used in beekeeping practices.

When honey bees consume sucrose syrup, they produce an enzyme, invertase, which breaks down the sucrose into glucose and fructose. However, this process can be exhausting for the bees. Only monosaccharides, such as glucose, can enter the hemolymph (Crailsheim 1988). hypothesized that administering inverted syrups to winter bees may contribute to the prevention of exhaustion. This is based on the premise that the sucrose is converted into glucose and fructose during the production process. For this reason, inverted sugar syrup is a good alternative, as it reduces the energy bees expend on digestion (Ceksteryte and Racys 2006). By using inverted sugar syrup, beekeepers can support the health and productivity of bee colonies. This practice not only benefits individual hives but also plays a role in protecting the bees as pollinators.

Inverted sugar syrup made with honey (rather than commercial invertase) can be a better option for bees compared to artificial invertase-based syrups. Generally, there are no serious problems when feeding bees with syrup that has not been overheated (Matović et al., 2024).

The sealed worker brood is a critical metric for assessing the development of bee colonies and the impact of experimental beekeeping practices. Unlike other hive types, Warre hives emphasize natural comb construction and minimal intervention, which may influence brood development patterns.

The primary aim of this study is to evaluate the impact of inverted sugar syrup supplementation on sealed brood quantity and colony strength in Warre hives, compared to traditional sucrose syrup feeding. A more profound understanding of these nutritional dynamics could enable beekeepers to refine their hive management strategies, thereby fostering stronger and more productive colonies while maintaining Warre hives. The existing literature suggests that feeding experiments represent a component of the routine beekeeping practices. However, in Bulgaria, studies focusing on the Warre hives remain limited.

## **MATERIAL AND METHODS**

## Study design

A controlled comparative study was conducted from April to October 2024 to evaluate the effects of inverted sugar syrup supplementation on colony performance in Warre hives. The experiment comprised 14 honey bee colonies equally distributed between the control group (n=7) and the experimental group (n=7). The bee colonies were situated at an apiary near Tran, Pernik region, Bulgaria (altitude 860 m), characterized by a temperate climate and moderate productivity pastureland with periodic floral blooms (10-day flowering cycles of dominant meadow species).

#### **Experimental setup**

Colony standardization:

Initially, all bee colonies were equalized and balanced in strength for initial strength (6±1 frames of bees) and food reserves (2.5±0.5 kg honey stores). Furthermore, adjustments were made to equalize colony strength by redistributing frames of stored honey and brood. At the beginning of the experiment, the colonies have frames with wax combs. The top frame slat of Warre hives has dimensions of 315 mm in length and 20 mm in width.

#### **Treatment groups**

Control group supplementation

Control group fed 1:1 (w/w) sucrose solution (50% in water).

## **Experimental group supplementation**

Bee colonies assigned to the inverted sugar treatment received syrup prepared through controlled inversion of sucrose. The preparation

protocol was as follows: a supersaturated solution was prepared by dissolving 7.5 kg of sucrose in 2.5 L of water (3:1 w/v ratio). The solution was maintained at 38.0±0.5 °C using a water bath. Enzymatic inversion was achieved by adding 1.8 kg raw polyfloral honey (containing natural invertases). The mixture underwent intermittent stirring (15 min every 3 h) for 168±2 h (7 days) to ensure complete inversion. Inverted sugar syrup making machine (Civan supplies for inverted sugar syrup, Inter RR-VDU Digital Universal Timer) is used for the preparation of high-quality inverted sugar syrup. The inverted sugar syrup used in this study is a noncommercial product.

The control and experimental groups received sucrose solution and inverted sugar syrup, respectively, administered identically in terms of time and volume. The control and experimental treatments were administered via hive-top feeders at a quantity of 250 ml twice per week. Treatment was done during two periods. The first period was spring stimulation (from the initial inspection at 1st of April until the first major nectar flow on June 27 – 28th of June), and the second was autumn feeding (1st of August through 15th of October) in order to enhance the production of wintering bees and prepare honey stores needed for wintering.

## Hydroxymethyl furfural (HMF) determination

HMF concentration in inverted syrup was determined spectrophotometrically (White method; Bogdanov et al. 1997). Analyses were performed approximately 24±1 h post-preparation. Absorbance readings at 284 nm and 336 nm were obtained using a UV-Vis spectrophotometer (PG Instruments).

#### Sealed worker brood assessment

The sealed worker brood area was quantified using a standardized measurement frame divided into  $5 \times 5$  cm grid units (25 cm<sup>2</sup> each). Brood assessments were conducted at 12-day intervals throughout the experimental period (Delaplane et al. 2013).

### Colony strength quantification

The mass of the adult bee population was determined gravimetrically using the following

protocol. Bees were gently brushed from all frames into polyethylene collection bags. Samples were immediately weighed using a calibrated electronic balance, and the results were recorded as the weight of bees for each colony in grams. Bees were returned to their respective hives within 5 minutes of removal to minimize disturbance.

Colony strength assessments were performed at two critical stages:

Spring evaluation (15 April) – post-winter colony buildup

Autumn evaluation (15 October) – pre-winter population status

## Statistical analysis

The comparisons between the experimental and control groups were conducted using Student's t-test. The differences were considered significant at p < 0.05. The results are reported as mean  $\pm$  standard deviation (SD). All data were analyzed using SPSS Statistics 23.0 (IBM Corp.).

## **RESULTS**

In the present study, spectrophotometric analysis (White method) of inverted sugar syrup revealed HMF concentrations ranging from  $7.11 \pm 0.82$  mg/kg to  $10.93 \pm 1.04$  mg/kg (mean  $\pm$  SD).

The initial worker brood area in all selected colonies averaged approximately 7000 cells per colony across both groups. The results revealed that bee colonies fed by inverted sugar syrup initiated higher worker brood rearing. This tendency persisted until the beginning of June. Supplementation with inverted sugar syrup resulted in an increase in sealed worker brood area. Significant differences were found between the average sealed worker brood of the experimental group on 15 April (p < 0.001), 27 April (p < 0.01), 9 May (p < 0.05), and 21 May (p < 0.001) compared to the control group. The highest sealed worker brood area was noted on 2 June for the spring period in the experimental group (averaging 24157 cells) (Fig. 1). The average values on this date are statistically significant (p < 0.01).

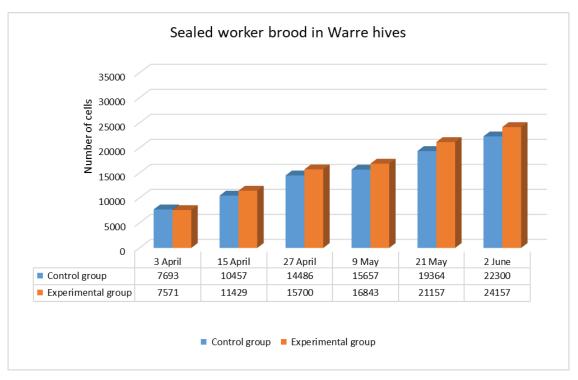


Figure 1. Sealed worker brood in Warre hives during spring feeding

In the autumn, the average values of sealed worker brood showed a statistically significant increase (p < 0.001) in the experimental group compared to the

control group for all dates from August 5 to October 4.



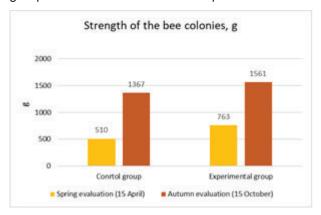
Figure 2. Sealed worker brood in Warre hives during autumn feeding

Figure 3 illustrates a Warre frame with a sealed worker brood area.



Figure 3. Frame covered with sealed worker brood in a Warre hive

Bee colonies supplied with inverted sugar syrup showed higher strength (weight of bees in grams) than control colonies in both test dates (15 April and 15 October) (Fig.4). Our results demonstrate statistically significant differences (p<0.001) in mean colony strength between experimental and control groups across both measurement periods.



**Figure 4.** Strength of the bee colonies (g) in the spring (15 April) and in the autumn (15 October)

## **DISCUSSION**

The hydroxymethylfurfural (HMF) content represents a critical quality parameter, and it is used as an indicator of thermal degradation or storage conditions of foods (Kus et al., 2005). The values received in this study fall well below the established

toxicity threshold for adult honey bees, which exceeds 50 mg/kg for chronic (LeBlanc et al. 2010).

The presence and development of sealed worker brood provide a direct measure of colony vitality, as they reflect successful brood rearing and overall hive productivity. Brood productivity is influenced by numerous factors, with nutrition playing a key role (Brodschneider and Crailsheim 2010). Supplemental feeding strategies have a direct impact on larval development and colony growth by providing essential carbohydrates for nurse bees.

Beekeepers often supplement honeybee colonies with sugar syrup or inverted sugar syrup to support brood rearing. In this respect, Rortais et al. (2005) found that a worker larva consumes 59.4 mg of carbohydrate during development.

In general, inverted sugar syrups are particularly useful for pre-winter feeding of honey bee colonies. However, their effects on colony productivity and brood development remain insufficiently studied for different types of hives. Pridal et al. (2023) reported that invert syrups are regarded as a form of sustenance with the capacity to improve the general status of the bee colonies before the winter season. This may be attributed to the fact that bees are not required to synthesize the enzymes necessary for sucrose breakdown when they consume monosaccharides such as glucose and fructose.

Several studies have investigated the effects of inverted sugar syrups on brood production, yielding varying results. Ceksteryte and Racys (2006) reported a significantly higher brood area in bee colonies fed yeast-inverted sugar syrup compared to standard sugar syrup during overwintering. Similarly, Eşanu et al. (2018) observed increased brood production in syrup-inverted feeding groups relative to controls. However, Pridal et al. (2023) found no statistically significant differences in brood development (p>0.05) or other growth parameters between colonies fed inverted versus sucrose-based syrups.

In the present study, results revealed that sugar syrup and inverted sugar syrup were highly consumed by the honeybees. It seems that because sugar syrup is more easily digestible, it stimulates the bees to distribute more resources to brood production.

Warre hives are designed to mimic a natural bee habitat. This design encourages bees to build comb naturally, fostering a stable environment for brood

development. The lack of frequent inspections reduces stress on the bee colony but limits beekeeper interventions.

#### Conclusion

Our study showed that bee colonies fed inverted sugar syrup had significantly higher average values of sealed worker brood and colony strength compared to those in the control group from April to October in Warre hives. While these short-term results are promising, long-term studies are necessary to determine the lasting effects on the general status and productivity of honeybee colonies in Warre hives. These insights can help beekeepers choose better feeding strategies to maintain healthy and strong bee colonies in regions with insufficient pasture.

**Author Contribution**: TT and RB conceived and planned the experiments. RB contributed to the analysis of the results and to the writing of the manuscript. The authors have read and agreed to the published version of the manuscript.

**Data Availability Statement**: All data are available upon request.

**Conflict of interest:** The authors declare no conflicts of interest.

**Ethical statement:** The ethical statement is not applicable to this study.

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