

Yonca Hortumlu Böceği, *Hypera postica* (Gyllenhal 1813) (Coleoptera: Curculionidae)'nın Zararı ve Larva, Pupa ve Ergin Gelişim Morfolojisi

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ÖZET

Bu çalışmada, yonca bitkisinde zararlı bir tür olan Hypera postica (Gyllenhal 1813) (Coleoptera: Curculionidae)'nın zararı, larva, pupa ve ergin gelişim morfolojisi, stereomikroskop ve taramalı elektron mikroskobu kullanılarak incelenmiştir. H. postica yumurtaları sarımsı renkte ve nispeten küçüktür. H. postica larvalarının başı ovaldır. Larvalar sarımsı-yeşilimsi renktedir. Larvaların sırtında uzunlamasına beyaz bir şerit vardır. Larvaların vücudu segmentlidir ve üzerinde setalar vardır. Larvalar, larva aşamasından prepupal aşamaya kadar sarımsı-yeşilimsi renkten kahverengiye dönüşür. Bu aşamada C şeklinde bir görünüme sahiptir. Baş ve vücudun diğer kısımlarında çok sayıda seta ayırt edilir. *H. postica* pupaları sarı renklidir. Kokon yapısı şeffaf ve ağsıdır ve farklı kalınlıklarda iç içe geçmiş delikli bantlar şeklinde görünür. *H. postica* erginlerinin vücut rengi parlak ve kontrastlı görünür. Ergin *H. postica*'nın vücut yapısı setalarla kaplı olup iki farklı seta yapısı bulunmaktadır. H. postica dünyanın birçok bölgesinde yoncanın verimini, kalitesini düşürmekte olup ekonomik kayıplara neden olmaktadır. Bu calışma ile vonca zararlışı olan ve ekonomik öneme sahip bu böceğin gelişim morfolojisinin belirlenmesi ve bu zararlıya karşı daha etkin mücadele yöntemlerinin geliştirilmesine katkı sağlanması amaclanmıştır.

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Adult development morphology and pupae, larval and damage of Alfalfa Weevil *Hypera postica* (Gyllenhal 1813) (Coleoptera: Curculionidae)

ABSTRACT

In this study, adult developmental morphology and the pupal, and larval of *Hypera postica* (Gyllenhal 1813) (Coleoptera: Curculionidae), which is a harmful species in alfalfa, and its damage in alfalfa were investigated using stereomicroscope and scanning electron microscope. *H. postica* eggs are yellowish in color and relatively small. The head of *H. postica* larvae is oval. Larvae are yellowish-greenish in color. The larvae have a longitudinal white stripe on the back. The body of the larvae is segmented and has setae on it. Larvae change from vellowish-greenish to brown in the larval to the prepupal stage. At this stage, it has a C-shaped appearance. Numerous setae are distinguished on the head and other parts of the body. *H. postica* pupae are yellow. The cocoon structure is transparent and reticulate, appearing as intertwined perforated bands of different thicknesses. The body color of H. postica adults appears bright and contrasting. The body structure of adult *H. postica* is covered with scales, and two different scales were found. *H. postica* reduces the yield and quality of alfalfa in many parts of the world and causes economic losses. This study aimed to determine the developmental morphology of this economically important alfalfa pest and contribute to the development of more effective control methods against this pest.

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INTRODUCTION

Curculionidae family is a group of insects, commonly known as the chambrel bugs, and are widespread throughout the world. The family is one of the largest families of insects, with approximately 83,000 described species (Oberprieler et al., 2007). In addition to causing structural and nutritional damage to various plants, some species of Curculionidae also play a role in spreading plant diseases (Parker et al., 2016). The alfalfa weevil, Hypera postica (Gyllenhal, 1813) (Curculionidae), is a destructive pest of alfalfa (Medicago sativa L.), the world's most important cultivated plant (Hoffmann, 1963). Damage caused by larvae consists of defoliation, which reduces the yield and quality of forages and causes economic losses in many regions of the world (Goosey, 2012; Saeidi & Moharramipour 2017; Soroka et al., 2020). H. postica is an oligophagous insect that feeds exclusively on leguminous plants of the genus *Medicago*. However, it can feed on several species of related genera, such as Melilotus, Trifolium, and Trigonella, in most alfalfagrowing regions worldwide. Heavily infested fields may appear silver or white as most leaves are destroyed or completely consumed. Severe pest pressure can destroy crops (Fick, 1976). The alfalfa weevil, *H. postica* (Coleoptera, Curculionidae), was studied to determine some biological characteristics in nature. It was found that females lay eggs on alfalfa stems in November, spend the winter in both egg and adult stages, and from mid-February, when the average temperature is above 9 °C, the adults migrate back into the field, mate, and start laying eggs. At the same time, it was determined that larvae emerged in early March from the eggs laid in the fall (Gözüaçık & Irec, 2019).

The family Curculionidae has a legless (apod) larva type. Larvae are generally seen in a "C" shape. The free pupal type is seen in the Curculionidae family. There are distinct wing marks on the pupa. The pupal period is spent in the soil or on the host plant (Lodos, 1960).

Studies on the morphology of larvae, pupae, and adults of Curculionidae are as follows. The morphology of immature larvae, pupae, and adults of Hypera temperei (Curculionidae) and Phloeosinus tacubayae (Curculionidae) were studied (Germann, 2021;Cervantes-Espinoza et al., 2023). The larvae and pupae were examined in Phelypera supply (Boheman, 1834)(Curculionidae) *Gymnetron* species (Curculionidae), Smicronyx smreczynskii Solari, 1952 (Curculionidae), (Vanin et al., 2012; Skuhrovec et al., 2022; Sprick, & Gosik, 2023). Skuhrovec (2006), H. *postica* explained its periods using chaetotaxy. Skuhrovec (2004) also described mature larvae of the

subgenus Hypera.

Studies on this subject are quite limited and no study has been conducted on H. postica, an important species. Therefore, in this study, larval, pupal, and adult developmental morphology of H. postica and the damage caused by it on alfalfa plants were investigated.

MATERIAL and METOD

Stereomicroscopy (SM)

In this study, *H. postica* was collected from alfalfa fields (*Medicago sativa* L.) from Bingöl, Turkey, in May 2022. Adult, pupa, and larvae of *H. postica* were photographed under an Olympus SZX7 SM.

Scanning electron microscopy (SEM)

For scanning electron microscopy (SEM), adult, larva, and pupae specimens were fixed in 2.5%glutaraldehyde (pH 7.2, phosphate buffered), rinsed three times with phosphate buffer, and separately dehydrated 15 min by using an ethanol progressive series (70%, 80%, 90%, and 100%). The tissues were transferred to hexamethyldisilazane, and then samples were dried with air. They were mounted on SEM stubs and coated with a gold sputter coater (Polaron SC 502). The samples were then photographed SEM (JEOL JSM 6060 LV).

RESULTS and DISCUSSION

H. postica adult females lay their eggs in groups on the stem parts of the clover plant. The eggs are yellowish and quite small (Figure 1a). Figure 1b shows newly hatched greenish and C-shaped larvae. The newly hatched larvae move to the terminal leaves and feed, causing small holes as they feed. Larvae and adults cause the plant to dry out (Figures 1c-h).

Larvae

Larvae when they first hatch, their heads are dark black and their bodies are off-white. In the second period of the larvae, the body color is light yellowgreenish. Larvae (L); 1st (L1) and 2nd (L2) stages are similar to each other. 3rd (L3) period. In larvae, the body is greenish. The 4th (L4) instar is also light green. From the third instar onwards, the dorsal and lateral parts of the larvae are white lines (Anonim, 2008). The fourth instar (mature) larva is 8 mm long the body is greenish, with a distinct white line running along the dorsal area present. Larvae go through 4 stages (Gözüaçık, 2022).



Şekil 1. (a).Yonca zararlısının yumurta kümesi. (b). Yonca zararlısının ikinci dönem larvaları. (c-f). Larvaların Medicago sativa üzerindeki zararı. (g, h). Yonca zararlısı ergin bireyleri ve Medicago sativa üzerindeki zararı.
Figure 1. (a). Egg mass of Alfalfa weevil. (b). Second instars larvae of Alfalfa weevil. (c-f). Damage of Alfalfa weevil larvae on Medicago sativa. (g, h). The adult individual and damage of Alfalfa weevil on Medicago sativa.

The head of *H. postica* larvae is oval. 4th instar larvae of *H. postica*, yellowish-greenish in color (Figure 2a). The larvae have a longitudinal white stripe on their back (Figure 2a). The larvae body is segmented (Figures 2b-d) and has setae on it (Figures 2c, d). The body surface is soft and flexible. Larvae move wormlike by contracting and relaxing. The head, thorax, and abdomen gradually become prominent in the developing larvae (Figure 2b). *H. postica*, spiraculum on abdominal segments I–VIII positioned above the dorsopleural lobe (Skuhrovec, 2004). In the transition from larva to prepupa, the larvae change color from yellowish-greenish to brown (Figure 3a). After this stage, they molt and begin to form the outline of the adult form. At this stage, it has a C-shaped appearance (Figure 3b). Two different setae, long and short, were found on the head of *H. postica* larvae. When SEM photographs are examined, we see the posterolateral and distal setae (Figure 3c). Numerous setae are distinguished on the head and other parts of the body (Figures 3b-f). Mouthparts on the head are distinguishable (Figure 3e).



Şekil 2. (a). Sırtta beyaz şeritli (St) ve sarımsı-yeşilimsi renli 4. dönem larva *H. postica*'nın genel görünümü (b). 4. İnstar olgun larvanın lateral görünümünün SEM görüntüleri. T-toraks, A1-A8-abdominal segmentler (c). *H.postica* larvasının baş kısmının SEM görüntüsü. (d) Larvaların son abdominal segmentleri (SEM). *Figure 2. (a). General view of H. postica 4th instar larva yellowish-greenish in color with a white stripe on the back* (St) (b). SEM images of lateral view of mature larvae at 4th instar. T-thorax, A1-A8-abdominal segments

(c). SEM images of the head of H.postica larvae. (d) The last abdominal segments of larvae (SEM).

Larvae of another species, *Lignyodes bischoffi* Blatchley, 1916 (Curculionidae) and *L. enucleator* (Curculionidae), unlike planned *H. postica* larvae, have white to whitish-yellow thorax and abdominal segments. The setae on the body are thin, orange, and distinct (Gosik et al., 2017).

Metadonus vuillefroyanus (Curculionidae), as for the larvae, the head changes from dark brown to black. All thorax and abdominal segments are greenish with longitudinal white stripes on both parts of the body. However, it has a thick long yellow stripe on its back and pink and purple stripes with small black stripes inside. All hairs are thorn-like and are located on distinct black protrusions in skinny white transverse stripes (Skuhrovec & Bogusch, 2016).

While the caudal spines in the larvae of *H. postica* show growth in the first stage towards later stages, there is a decrease in *P. tacubayae* (Curculionidae) (Cervantes-Espinoza, et al., 2023). *Adosomus roridus* (Curculionidae) has a brown or dark brown head when mature larvae are examined. All thoracic and abdominal segments are white, with a long light brown stripe only on the dorsal part of the protonum (Trnka et al., 2015).



Şekil 3. (a-f). Larvadan prepupaya geçiş. (a) Larvadan prepupaya geçişin genel görünümü (SM). (b-f) *H. postica* larvasının abdominal segmentlerin SEM görüntüleri (ds: distal setae, ps: postero-lateral setae).

Figure 3.(a-f). Transition from larva to prepupa. (a) General view of transition from larva to prepupa (SM). (b-f) SEM images of the abdominal segments of H. postica larva (ds: distal setae, ps: postero-lateral setae).

We observed that the abdominal segments in *H. postica* consist of eight segments. Similarly, *H. arator* (Linné, 1758), *H. denominanda* (Capiomont, 1868), *H. jucunda* (Capiomont, 1868), *H. nigrirostris* (Fabricius, 1775), *H. plantaginis* (De Geer, 1775), In *H. suspiciosa* (Herbst, 1795), *H. venusta* (Fabricius, 1781) and *H. viciae* (Gyllenhal, 1813), the abdominal segments were reported to consist of eight segments (Skuhrovec, 2004).

Skuhrovec (2006), stated that *H. postica* larvae have

one seta in the first instar, 3 in the second instar, and 3 setae in the abdominal regions in the third and fourth instars.

Pupae

Figures 4a-d show the skin change of *H. postica* as it transitions from the prepupa stage to the pupa stage. Pupae is the period known as dormancy. *H. postica* pupae are yellow (Figures 4e, f). Pupae of another species, *A. roridus* (Curculionidae), have a different color from whitish to yellow (Trnka et al., 2015). *P.*

tacubayae (Curculionidae) pupae (Cervantes-Espinoza et al., 2023) are similar to *H. postica* pupae. The color of the pupae of *M. vuillefroyanus* (Curculionidae) is yellow on the body and greenish on the abdomen (Skuhrovec, & Bogusch, 2016).



Şekil 4. (a-f). *H. postica*'da prepupadan pupaya geçişin SM fotoğrafları. *Figure 4. (a-f). SM photographs of the transition from prepupa to pupa in H. postica.*

Adult

The cocoon structure is a transparent and reticulated layer and appears as intertwined perforated bands of different thicknesses (Figures 5a-h). The pupal cocoons in *P. schuppeli* (Curculionidae) are spherical, cage-like, and translucent white (Vanin et al., 2012). The stage of the last larval instar of *H. temperei* (Curculionidae) built a net cocoon, whose colour reached from transparent-whitish to slightly brownish (Germann, 2021). As the prepupa inside the cocoon matures, its color changes from yellow to brown (Figures 5a, b). SEM photographs show the structure of the cocoon fibers woven by the last-stage larvae (Figures 5c-h).



Şekil 5. (a,b). *H. postica*'nın kokon içindeki prepupası (SM). (c). Kokonun genel görünümü (SEM). (d-h). Son dönem larvası tarafından örülen kokon liflerinin mikro yapısı (SEM).

Figure 5. (a,b). Prepupa of H. postica in a cocoon (SM). (c). Genel view of cocoon (SEM). (d-h). A microstructure of the cocoon fibers spun by a last instar larva (SEM).

In Figure 6a, a pupa is seen inside the cocoon. The free pupa is seen in *H. postica* (Figure 6b). Figure 6c shows the transition from prepupa to adult. At this stage, white setae are distinguished on the elytra (Figure 6c). The body of *H. postica* adults ultimately acquires a bright brownish color. The instrestium in the dorsal part of the elytra also consists of a series of points (Figure 6d). In SEM photographs of adults of H. postica, the lateral and ventral parts can be seen, and their body lines are clear. Adults have a rostrum on their heads that starts from the eyes (Figures 6e, f). H. postica rostrum is "short and robust", but H. temperei (Curculionidae) rostrum is "long and slender" (Germann, 2021). The body structure of H. postica is covered with scales, and two different scales were found (Figures 6g, h). The first one shows a thick sword-like structure at the base and becomes thinner towards the tip. The second one resembles a tulip and is forked. Both flake types have parallel lines on their surfaces (Figure 6h).

The shape and patterning of the scales on the elytra varies from species to species. In *Cionus hortulanus* (Curculionidae), the scales are elongated and parallelsided, the apical part is serrated, the lateral side is smooth, and has large holes on the surface (Erbey, & Candan, 2015). In the *Cionus opens* (Curculionidae), the scales are long and elliptical; the apical part is sharpened; the surface of the scales has two or three longitudinal protrusions; the serrated part of the interstrain on the lateral side near the apical part has a series of clearly erect scales (Erbey, & Candan, 2015).





- Şekil 6. (a). Pupanın kokon içindeki genel görünümü. (b). Serbest pupanın genel görünümü (SM). (c). Prepupadan ergine geçiş (SM). Elytra üzerinde beyaz setalar. (d). Gelişimini tamamlamış ergin (SM). (e, f) Yetişkinlerin SEM görüntüleri yanal ve ventral wiev. (g, h). Elytrasındaki pulların SEM görüntüleri (*:lale benzeri pullar, ▶:kılıç benzeri pullar).
- Figure 6. (a). General view of the pupa inside the cocoon. (b). General view of the free pupa (SM). (c). Transition from prepupa to adult (SM). White setae on elytra. (d). Fully developed adult (SM). (e, f) SEM images lateral and ventral view of adults. (g, h). SEM images of scales in the elytra (*: tulip-like scales, swordlike scales).

The larva, pupa, and adult forms of this alfalfa pest, *H. postica,* have been described in detail, and the similarities and differences with other species are explained and contribute to taxonomic methods. It also forms the basis for studies on biological control against this species. Chemical residues in the plant, serious damage to non-target organisms, and the risks posed to the environment and human and animal health have very serious consequences. As a result, the pest biology, economic damage threshold natural enemies existence, alternative methods of struggle and all these should be handled within the framework of 'Integrated Struggle'is considered useful to be taken (Gözüaçık, 2022).

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Contribution Rate Statement Summary of Researchers

The authors declare that they have contributed equally to the article.

Conflict of Interest

The authors have declared no conflict of interest.

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