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Research Article

Morphological identification of four stored grain pests in Misan Province Southern- Iraq

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Abstract

The present study aimed to morphological identification of four species of pests that infect stored grains in Misan province in south Iraq during the period from January 2021 to December 2022. Samples of infecting grains included chickpeas, white and red beans, Indian and amber and American rice, bulgur, sesame, rice flour, flour, dried and canned dates, as well as samples of corn were collected from different regions, including the homes and local and central markets and general company for grain trade Misan branch. The results show that four species belonging to the order Coleoptera return to three families under the four genera. These species are *Tribolium castanum* Herbst,1797,*Latheticcus oryzae* Waterhouse, 1880, *Trogoderma granarium* Everts ,1898, *Oryzaephilus mercator* Fauvel, 1889.

Keywords: Coleoptera, Grain Stored Pests, Identification, Morphological, Misan province Store grain, pests, Stored grains.

Introduction

Stored grains collected after harvest, include products and foods processed from them (Solà Cassi, 2018). About 10% of the grains stored after harvest are damaged by insects, rodents, microorganisms and improper storage (Banga et al., 2020). Insects are among the main and important pests of stored grains (Rajashekar et al., 2010). Insects do not cause economic losses as a result of consuming grain alone, but they are also one of the main causes of the spread of pollutants. Among the 600 species of insects present in stored grain, 100 species cause economic losses to grain and its products (Neethirajan et al., 2007). And more than 60 species of these pests

belong to the order Coleoptera and Lepidoptera, and species return to the order Coleoptera constitute about 75% of the pests that infecting stored grains (Abbasi et al., 2021; Jian, 2019).

Pests are divided depending to their feeding method into primary and secondary pests .That first bite uninfected grains with their chewing mouthparts. The larvae and adults are harmful and dangerous because of their ability to attack grain embryos, which leads to a reduction in germination and protein content. An example of these pests is *Callsobruchus maculatus.* Secondary pests are pests that infect grains infected with primary pests, as they are unable to infect uninfected grains, an example of which is *Tribolium castaneum* (Dowell et al., 1999; Sabeat, 2017; Stejskal et al., 2005).

The morphological identification depends on the external characteristics of each species and for all stages of life, starting from the special features of the egg, larva, and pupa and adult . The most important of these morphological characteristics is the presence or disappearance of spots on the body and seta, and their distribution over different areas of the body, and the presence or absence of seta on the hind legs and appendages distributed over the whole body (Liu et al., 2011). This is the first study in Misan province –south Iraq .There are many morphological studies in Basrah province, including the study (Grzywacz et al., 2017), they conducted a diagnosis study on four species of the muscidae family in Basrah province. In the same year (Abed & Kareem, 2022) used a scanning electron microscope to diagnosis the fourth larval instar of mosquitoes and due to the lack of studies on these pests from the taxonomic aspect, the study was designed to survey and isolate insects found in grains and collected from different areas in Misan province.

Material and methods

Pest-infected grain samples Wheat, flour and rice were collected over the whole year, from January 2021 until December 2022, and from different regions of Misan province, which included the central province, local and central markets, and districts. And from the General Company for Grain Trade in Misan with its three branches, and it was located within the city center, as the first site is located on the Basra-Amara Road, and the second site is located on Al-Tayeb Road, nearby to the plastic factory. The third site is on Batira – Amara Road within the Batira Military Airport Road.

The three warehouses differed in their ways of storing wheat, as in the first site the wheat was stored in Iron Elevators, while in the second site the storage was in the form of stacks, while in the third the storage was in the form of sheds.

Samples of animal feed were also brought from the General Company for Grain Trade in the first site, and the samples that were brought from the central and local markets included chickpeas, white and red beans, Indian and amber and American rice, bulgur, sesame, rice flour, flour, dried and canned dates, as well as samples of corn.

Colonies of pests that infect stored grain

Colonies of pests that infect stored grain were reared on media free of infection depending on the grains infect them as these media were previously treated at a temperature of 50˚ c for 10 minutes in a oven to get rid of any infection in the grains (Ahmady et al., 2016) 250 grams of infection – free grains are placed in plastic bottles with a capacity of 300 ml each with four replications for each of the study areas. and a pair of insects male and female are inside each bottle using a aspirator in case the insect is flying and then ozzle of bottle is covered with a piece of cloth and the colonies renewed after each generation by taking the insects emerging from the eggs to make new colonies.

Preparation of permanent slides for insects

Insects to be studied are placed on a piece of cork. This piece of cork is placed in a glass flask with a capacity of 100 ml. Half of it contains water. It is heated for 20 minutes at a temperature of 50-60 °C without the insects touching the water. The flask is covered by a Petri dish to prevent the leakage of water vapor which works on relent and thus the ease of separating the body parts and muscles and not breaking them. Then those parts are placed in a glass flask with a capacity of 100 ml. One-third of it contains potassium hydroxide (KOH) of concentration 10 % and left it to boil for 10 minutes on a hot plate to clarify the sutures and plates and facilitate the removal of the muscles attached to some plates so that they appear clear during the examination and then washed three times with distilled water It is left in water for five minutes each time, after which it is placed in ethyl alcohol at concentrations of 30%, 50%, 70 %and 100% each concentration for a period of five minutes. It is transferred to xylene for five minutes and placed on a glass slide and install with DBX glue. A drop of glycerin is placed and surrounded by a plastic ring in a way that suits the height of the sample. The top and bottom were painted with nail dye to adhere the cover. If bubbles appear, the slide is placed in an electric oven at 60°C degrees for 24 hours to get rid of the bubbles.

Results

The results of the morphological identification study showed that four species of pests infecting stored grains returns to the order Coleoptera were diagnosed as a study for the first time in Misan province. Species belong to three families of four genera:

Order: Coleoptera

Sub order: Polyphaga

1-Family: Tenebrionidae, Latreille, 1802

Subfamily: Tenebrioninae, Latreille

Genus:*Tribolium* Macleay, 1825

Tribolium castaneum, Herbst,1797

Genus:*Latheticus* Waterhouse, 1880

Latheticus oryzae Waterhouse, 1880

2-Family:Silvanidae

Subfamily:silvaninae Kirby, 1837

Genus: *Oryzaephilus* Ganglbauer, 1899

Oryzaephilus mercator Fauvel, 1889

Family: Dermestidae3-

Subfamily:Megatominae Leach,1815

Genus: *Trogoderma* Dejean,182

Trogoderma granarium Everts,1898

Morphological description of Adults

***Tribolium castaneum* (Herbst, 1797**)

The general shape of body: Elongated cylindrical, its color is light brown to reddish brown at times, dark brown to black in the last stages of the adults, flat from the ventral side and slightly convex from the dorsal side, the body length is 3.5-4 mm and the width is 1-1.5 mm (Fig. 1 A and B).

Head: Small in size compared to the remnant of the body part, reddish-brown to dark brown, semi square in shape, 1 mm long and 0.5 mm wide, irregularly distributed small punctures spread over the surface of the head .(Fig1 A) **Antennae:** 0.5 mm long, consisting of 11 segments, longer than the length of the head, the scape is rectangular and elongated, the pedicel is shorter in length and smaller than the scape, the three peripheral segments appear to have suddenly enlarged, the last segment is circular and the remaining segments are square in shape, the sides of the three peripheral segments have soft setae , the middle six segments appear semi triangular in shape(Fig1 A) **Compound eyes:** dark black in color, small, renal in shape, the distance between them on the dorsal side is greater than on the ventral side (Fig. 2 A).

Thorax: light brown to dark brown, the anterior margin of the pronotum is wide and less wide than the posterior margin, which is semi round, and is larger than the mesothorax and metathorax ,convex from the dorsal side, more wide than the width of the head, the front margin is parallel to the sides and the lateral margin appear to be curved inward, there are small irregular punctures with clear distances covering the surface ,The scutellum is triangular in shape, small in size from the dorsal side, on the ventral side Two triangular openings near the anterior margin of the pronotum is the place where the coxa of the front legs join. The coxa of the middle legs are located on the posterior margin of the mesothorax. The hind legs coxa embed into the sternal plate of the thir d abdominal segment and do not divide it. (Fig. 2 B and C)

Thoracic appendages

The fore wings: light to dark brown strong sheath, meet in a straight line in the middle of the dorsal line and cover the two hind wings,their anterior end is rectangular, and the posterior end is tapering .It has longitudinal lines extending from front to back, but some of these lines do not reach their end.(Fig. 2 D).

Front legs:1 mm long, coxasemi-rounded, trochanter triangular in shape and dark in color, femur wide but less wide than the femur of the hind legs, the base of the femur has a dark spot and on surface have seta, the tibia is triangular in shape longer than the femur and contains long spurs at in the internal side have seta .The tarsus consists of five segments, and the first four tarsus segments are small, of semi equal length, with soft seat, and the last segment is elongated, triangular in shape, ending with two pairs of long simple curved claws (Fig. 2 E)

Middle Legs: similar to the front legs in length and the shape of the remnant of the parts except for the coxa, which are closer to the circular shape, and the first tarsus segment is longer than the second tarsus segment (Fig. 2 F)

Hind legs: larger than the coxa of the front and middle legs, and their coxa are embedded in the abdominal plate of the third abdominal segment. They are oval and elongated. The trochanter is dark brown and the coxa is based on it. The femur is wider than the femur of the front and middle legs and its close end to the tibia is dark, the tibia is elongated, and its inner margin has two long spurs. The tarsus is made up of four segments, and the last segment is elongated and ends with two pairs of claws (Fig. 2 G)

Abdomen: light brown to dark brown, consisting of five segments. Segments are rectangular in shape. The first four segments have a transversal anterior and posterior margin. The fifth segment

has a rounded top. The sternal abdominal plates are gradient in size, as the first and second sternal abdominal plates are equal in size. The third is smaller than the first and second, and the fourth is shorter than the third (Fig. 2 H).

Figure 1. *Tribolium castaneum;* A) dorsal side; B) ventral side

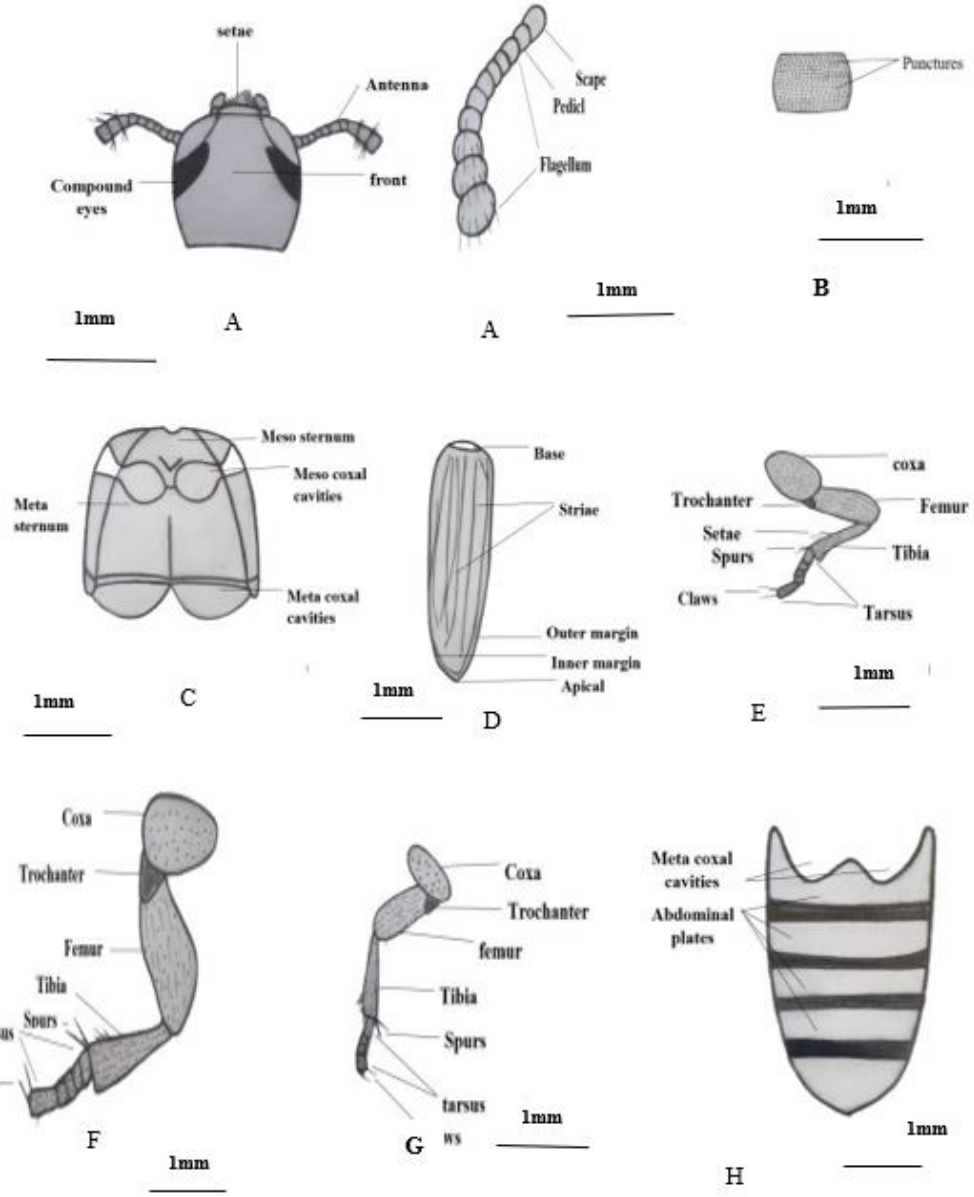

Figure 2. A) Head; B) Pronotum; C) Meso and Metathorax D) Elytron; E) Foreleg; F) Middle leg; G) Hind leg; H) Abdomen.

***Latheticus oryzae*Waterhouse,1880**

The general shape of body: Yellowish brown, elongate cylindrical, dorsally flattened, parallel sides, body length 3-3.5 mm (Fig 3 A and B).

Head: Wide, looks more like a square shape, elongated, 0.5 mm, longer than the head of *Tribolium castaneum*, and for this reason it is called the flour beetle with a long head, compressed from the dorsal and ventral sides. It is located inside the furrow of the pronotum (Fig.4 A).

Antennae:0.5 mm long, 11segments, clavate, the scape is small in size, irregular in shape, the pedicle is small in size, semi-circular, the flagellum consists of nine segments, the first four segments are small in size, from the fifth to the tenth segments, they vary in size and appear to be overlapping with each other, the ninth terminal segment is smaller, elongated cylindrical (Fig. 4 A).

Compound eyes small, dark black in color, renal in shape, diverging from the dorsal side and close together from the ventral side, surrounded by a clear suture, the divisions of the facets are clear (Fig. 4 A).

Thorax: yellowish to light brown, semi-rectangular, of greater length compared to the width. Small punctures spread on the dorsal surface with clear distances. The anterior margin of the pronotum is wider than the posterior margin close to the base of the elytron. The pleuron are parallel, the scutellum is triangular in shape, small in size, the mesothorax is rectangular in shape, small, the anterior end of the metathorax is narrower than the posterior end, the mesothorax and metathorax are longer than the mesothorax and metathorax in the species *Tribolium* castaneum, the coxa of the hind legs are embedded within the sternal plate of the third abdominal segment and this plate does not divide, a line extends from the margin of the posterior side of the metathorax to the middle and does not reach the front this line called meso groove (Fig. 4 B and C).

Thoracic appendages

Elytron: Sheaths are strong, light brown to bright yellow, elongated, with hexagonal punctures spread over their surface arranged in the form of longitudinal lines. Some of these lines reach the tapering end, while others do not (Fig. 4D).

Front legs: 0.5 mm long, light brown, the coxa have a rounded base and a conical apex, the trochanter triangular shape is small in size, the femur is elongated and wide in the middle and narrows towards the apex and is rounded at the base near the area of its connection with the tibia, tibia is elongated, triangular in shape, more broadly at the base. It carries a long spur at one edge. The tarsus is equal in length, consisting of 5 segments, the first four are equal in size, except for the last, elongated, triangular shape that ends with two pairs of claws (Fig. 4 E).

Middle legs: like the front legs, except for the coxa which are circular in shape, and the femur is shorter and less wide in the than the front legs (Fig. 4 F)

Hind legs: the coxa are oval in shape wide with a light brown color, the trochanter triangular shape is dark brown, the femur is wide and light brown in color, the front end is narrow, elongated and

wide at the posterior end, the tibia is triangular in shape, narrow at the top and wide at the base, there are two spurs at the posterior end of the tibia in the area where the tibia connects with the tarsus, the tarsus consists of four segments, the first is triangular in shape, elongated, the second and third segments are triangular in shape, while the fourth is longer than the remnant of the tarsus segments, ending with two pairs of claws (Fig. 4 G.).

Abdomen: dark brown, five segments, the sternal abdominal plates are convex, the first plate is broad and large, the second and third are equal in length and size, the fourth is broader but shorter than the second and third ventral sternal plates, the fifth is narrow and short tapering (Fig. 4 H).

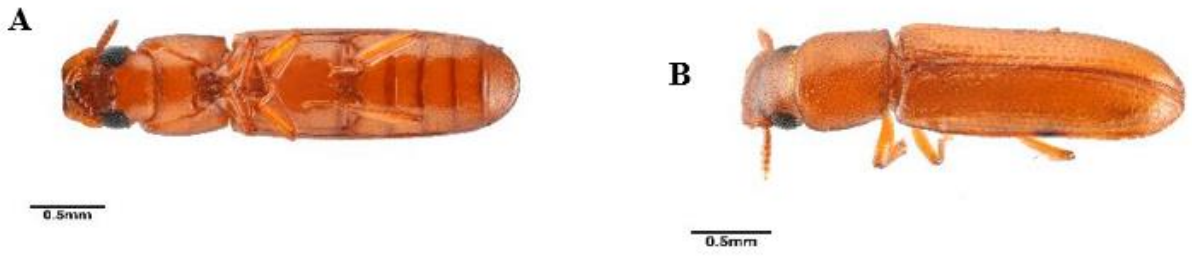

Figure 3. *Latheticus oryzae:* A) Ventral side; B) dorsal side

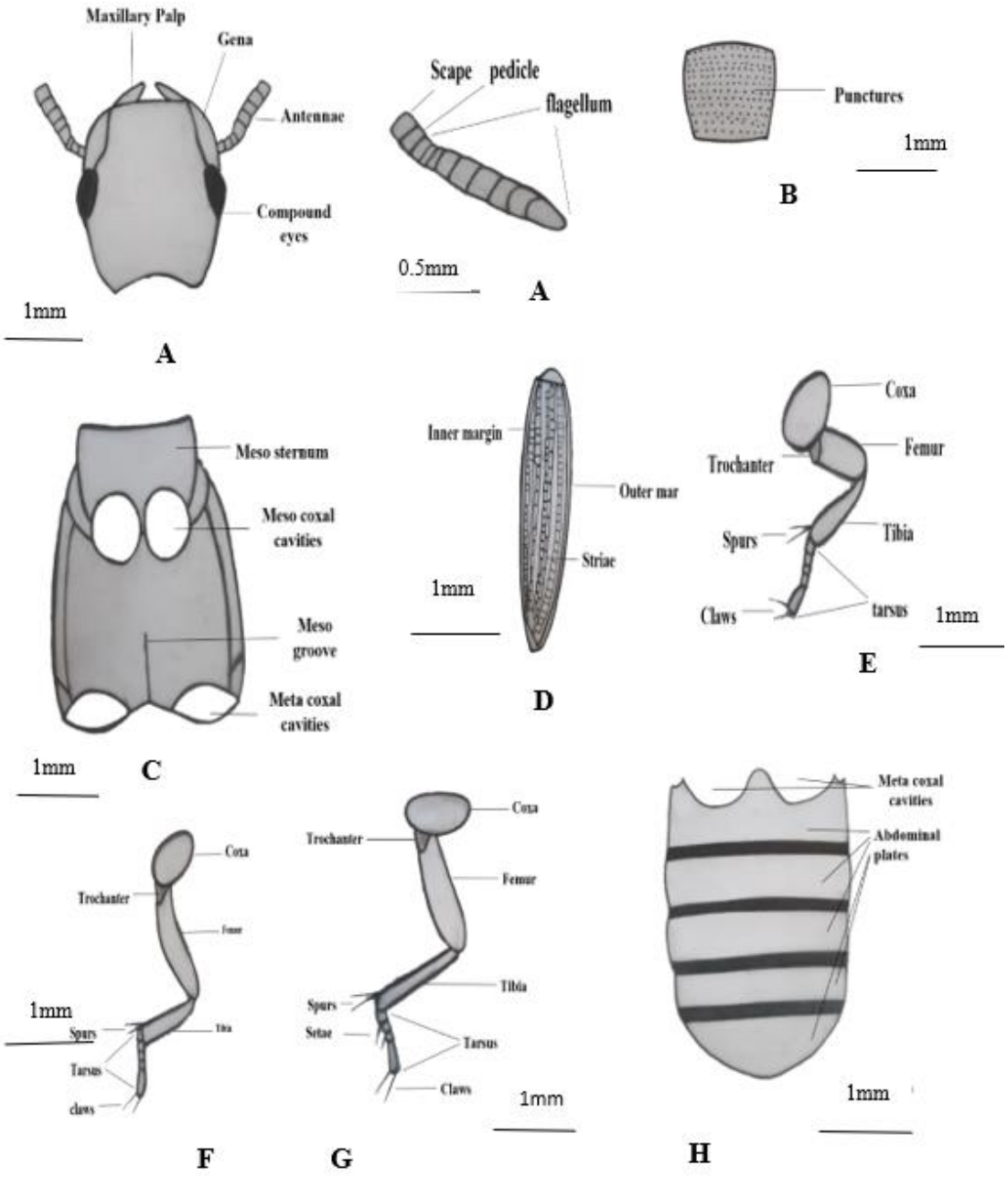

Figure 4. A) Head and appendage; A) Antannae; B) Pronotum; C) Meso and Metathorax; D) Elytron; E) Foreleg; F) Middle leg; G) Hind leg; H) Abdomen

***Oryzaephilus mercator* Fauvel,188**

The general shape of body:reddish-brown to dark brown, 2-2.5 mm long, thin, elongated,there are circular punctures and light-colored scattered on the surface of the body. The body is covered from the ventral side with dense, shiny seta (Fig.5 A and B).

Head: semi-triangular, 0.5 mm long, less wide than the remnant of the body, covered with fine and dense punctures and seta , the posterior end is wider than the front end, behind the compound eyes there is a triangular temple, which is a distinctive characteristic of this species (Fig. 6 A) **Antennae:** long, brown, capitate, consisting of 11 segments, longer than the length of the head, the scape is cup-shaped, the pedicle is smaller in size and elongated in shape, the third segment is shorter than the second, segments of 4-8 equal in size are semi-spherical, while the three terminal segments of segment 9-11 are cup-shaped, segment 10 is wider than segment 9, the last segment is round in shape, the sides of the terminal five segments have soft and fine seta (Fig. 6A) **Compound eyes:**Closer to the thoracic region, black in color, clear in a semi-circular shape, protruding outside the head, the facets are very clear, the temple is a triangular structure with an acute angle behind the eye and this makes it distinguished from the species*O. surinamensis*, the distance between the compound eyes from the dorsal and ventral sides is large (Fig. 6 A).

Thorax:Semi-square, elongated, parallel sides, dark brown, dense punctures spread on its surface, there are six teeth on the two lateral margin that give a saw-like shape, the distance between these teeth is uneven, and these teeth diverge in the anterior angle between prominent, distinct and non-protruding and undistinguished teeth, the scutellumis triangular in shape, with a dense and deep punctures ,the mesothorax merges with the pronotum by elongated neck. The hind legs are embedded in the margin of the third sternal abdominal plates and do not divide them (Fig. 6 B and C).

Thoracic appendages

Elytron: Reddish-brown to dark brown, elongated, the anterior end is more rectangular than the posterior end tapering, covers all five abdominal segments, punctures of different sizes distributed in the form of nine longitudinal lines, the color of the punctures ranges from reddish brown to dark brown, contains short brown seta (Fig. 6 D).

Front legs: dark brown in color, small coxa, trochanter oval elongated, smaller than the coxa, looks like a support for the coxa , the femur is wide, cylindrical and darker in color than the remnant of the parts, enlarged in the middle, the tibia is semi-elongated triangle, the front of the tibia is narrow compared to the broad end, the tarsus consists of five segments, the first is longer than the second, and the second and third are equal in size, the fourth is small and the fifth is triangular in shape, elongated, ending with two pairs of semi-arched claws (Fig. 6 E).

Middle legs: similar to the front legs, except for the coxa, which are rounded and larger in size than those of the front legs (Fig. 6F).

Hind legs: the coxa are oval in shape, elongated, the trochanter is semi-rectangular, the femur is wide in the middle, narrow at the end close to the trochanter and circular at the end close to the tibia, the tibia is elongated, semi-triangular, the inner margin has short teeth and the outer margin has short spines, in the femur of the hind legs of males there is a spine that does not exist in females (Fig. 6G).

Abdomen: dark brown, elongated, numbering five segments or plates. The sternal ventral plates appear convex. Their surface is covered with soft, delicate, and shiny seta. The abdominal plates are gradient in size. The first stern abdominal plate is larger than the second, third, and the fifth is oval, tapering at the end (Fig.6 H).

Figure 2. *Oryzaephilus Mercator:* A) Ventral side; B) Dorsal side

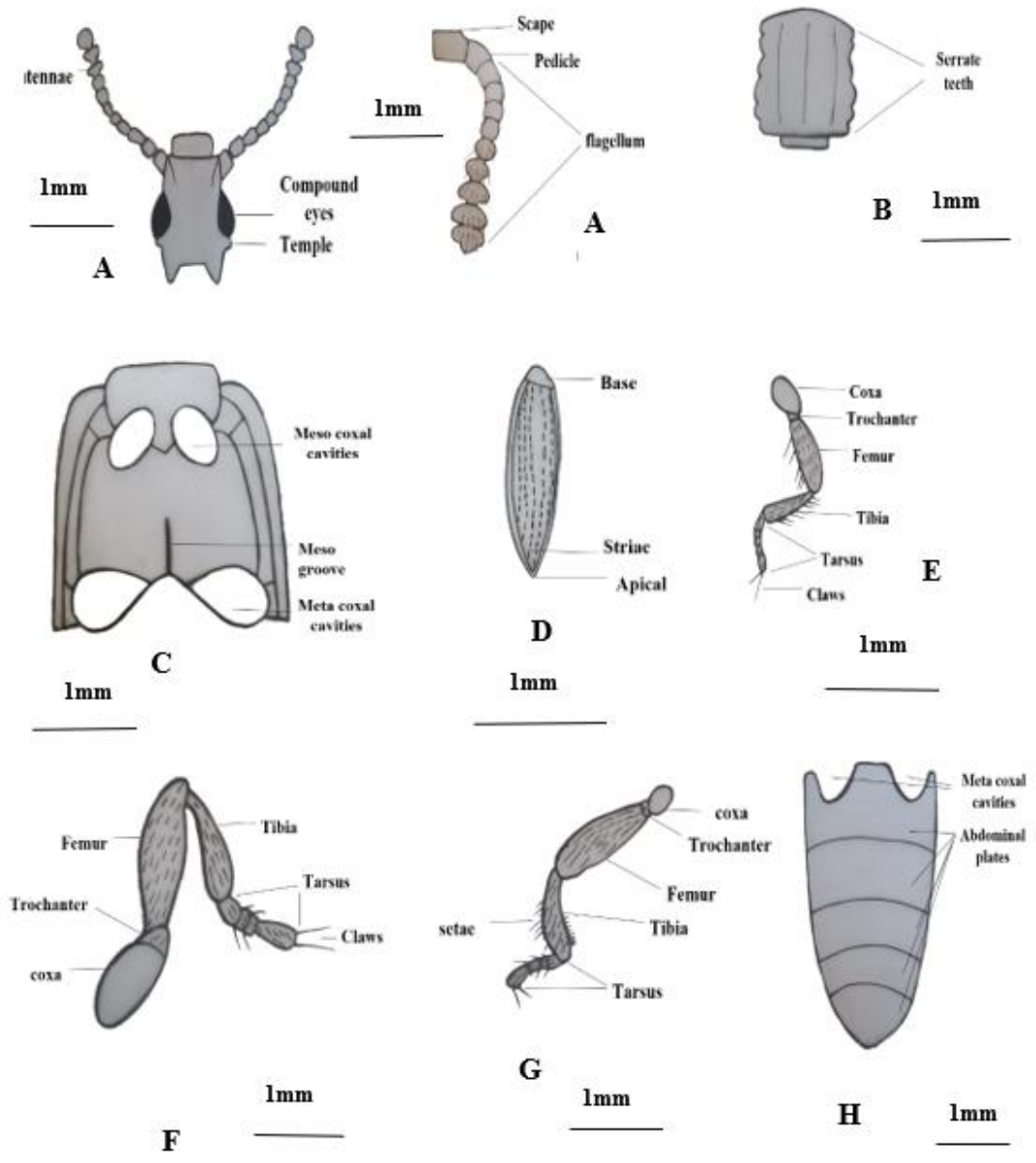

Figure 6. A) Head and appendage; B) pronotum; C) Meso and Metathorax; D) Elytron; E) Foreleg; F) Middle leg; G) Hind leg; H) Abdomen

***Trogoderma granarium* Everts,1898**

The general shape of body: Body length from 1.5-2.5 mm, radius 0.5-1.5 mm, oval in shape , the head and thorax and abdomen are dark brown in color ,the color of antenna and legs range from red to reddish brown, the body is covered on the dorsal and ventral sides by very fine, soft seta (Fig. 7 A and B.).

Head: black, small, curved under the pronotum, in the front a simple medial eye Ocellus, one between the compound eyes, spread on the surface of the head numerous dense small puncturs in size and cover the head with fine and short soft seta at varying distances, the maxillary palpus are yellow in color, elongated with a pointed head. It has very soft and fine seta (Fig. 8 A)

Antennae: 11 segments, located inside a grave in the sides of the dorsal of pronotum, brown to yellowish-brown in color, the scape is cup-shaped, the pedicel is semi-circular, similar in size to the scape, the five medial segments are equal in size, the segments of 8-10 take the form of a club-Shape and the last segment is a tapered end (Fig. 8A).

Compound eyes: brown to black in color, the margin surrounded by them is zigzag, on both sides of the head in a semi-circle, facets are not well clear (Fig. 8A).

Thorax: dark brown to black, the front margin is semi-concave, the posterior margin is V-shaped, the lateral margin is curved towards the inside, the surface of the pronotum is provided with dense, soft, bright yellow seta, the scutellum is small in size and triangular in shape (Fig. 8 B and C).

Thoracic appendages

Elytron: range from light brown and reddish brown to dark brown, the anterior end is rectangular in shape and the posterior end is semi-oval, black seta spread sparsely on the surface of the elytron in different areas and at different distances, and there are dense punctures on their surface at different distances as well, spots of different shapes distrusted on the edge and mid elytron (Fig. 8D)

Front legs: 0.7 mm long, brown to yellow in color, short, semi-oval coxa, visible with a darker brown color than the remnant of the parts, the trochanter is irregular in shape, the femur is huge and cylindrical with a yellowish-brown color, the tibia is light brown, elongated, cylindrical, at the external margin is a row of short spines, the top of which contains two short spurs. the tarsus consists of five segments ending in two pairs of semi-curved claws (Fig. 8 E).

Middle legs: similar to the front legs, except for the coxa, which are circular in shape. The remaining parts of the legs are similar to the front legs (Fig. 8F)

Hind legs: the coxa are more broad and elongated, a wide base and a narrow apex, the trochanter is semi-oval, the femur is wide at the place of its connection with the trochanter and narrow in the area of its connection with the tibia, the tibia is elongated, on the outer margin five soft seta, the tarsus consists of five graduated, semi-overlapping segments that end with two pairs of claws (Fig. 8G).

Abdomen: wide, light brown to dark brown, the sternal ventral plates are convex, graduated in size, the fourth and six abdominal plates are equal in length, while the third is wider than the remnant of the sternal plates (Fig. 8H)

Fig. 7. *Trogoderma granarium:* A) Ventral side; B) Dorsal side

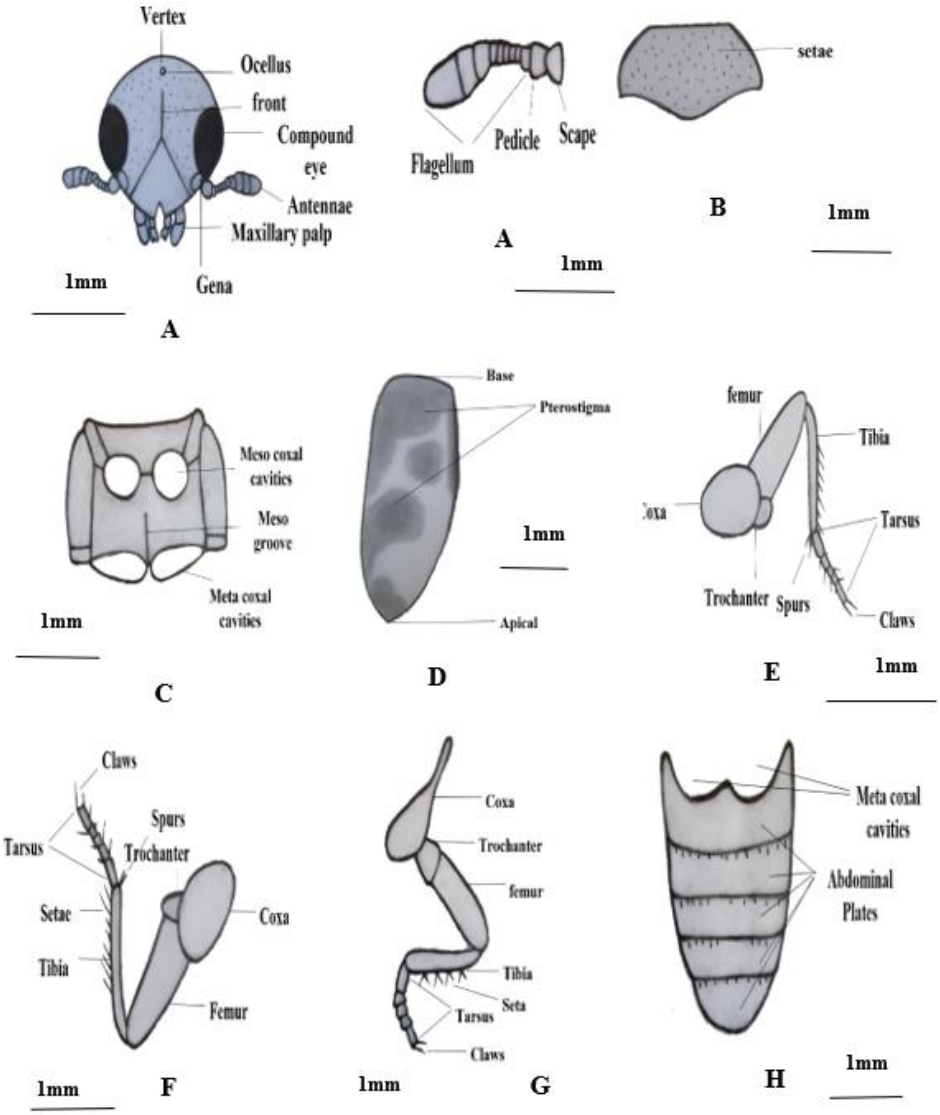

Figure 4. A) Head and appendage; B) pronotum; C) Meso and Metathorax, D) Elytron; E) Foreleg; F) Middle leg; G) Hind leg; H) Abdomen

Discussion

The four species of diagnosed pests have been recorded in many countries of the world. They have been recorded in Africa, Asia, North and South America, Europe and the Middle East (Hagstrum, 2016; Rodríguez-Cabo et al., 2021; Szito, 2006) indicated that *Trogoderma granarium* is not found in Australia and New Zealand .The species *Tribolium castaneum* was recorded in many Arab countries, including Egypt, Libya, Palestine, Sudan and Lebanon (Al Ali, 1977). It was also recorded Iraq for the first time in 1918 on stored rice, flour and wheat (Buxton & Mellanby, 1934) and recorded in Basrah province after an infection of stored dates ,called the red rusty flour beetle or the red flour beetle (Ahmad et al., 2021).

As for the species *Latheticus oryzae*, it was recorded by (Lobl et al., 2008) in Iraq for the first time, and the species *Oryzaephilus mercator* was recorded for the first time in Iraq by (Shalaby et al., 1970), and the last species, *Trogoderma granarium* , was recorded by (Rassoul, 1976).

The description of the local species, *Tribolium castaneum*, is identical to the species described by (Salman et al., 2018) in the characteristics of the antennae of this species . The antennae is connected to the head in a hole called the antennal socket , and it consists of the scape with few yellow and short seta located in the scape, but the flagellum consists of nine segments , the terminal three are capitate , long, shiny, yellowseta ,pedicle has fewer spines than the spines in the scape. The species *Latheticus oryzae* is called long-headed flour beetle. The description of the sample studied in Misan identical to the description of (Rees, 2004) in adults are flat, thin, the general color of the body is yellowish-brown, parallel sides, the segments of the four antennae are equal in size and the last segments are smaller than the other four segments this species is present in a lower rate compared to the previous species, in Egypt recorded by Andre in 1927 (Badawi, 1972). The species *Oryzaephilus mercator* prefers oilseeds, nuts, oats and dried fruits (Barnes, 2002; Syarifah Zulaikha et al., 2018). The characteristics of the studied local species are identical with the characteristics mentioned in the key, which was worked by (Thomas & Woodruff, n.d.), in that the anterior pronotal angle gradually develops, and behind the compound eyes, the structure of the temple is triangular in shape, and the head is triangular – elongated . The local species, *Trogoderma granarium* its description identical with was mentioned by (Singh et al., 2017), that the adults are oval in shape, the hairs are distributed over the entire body, the spots on the sheath are light and the segments of the three terminal antennae are like a club.

Identification Key of four stored grain pests in present study

1- The temple is triangular in shape and the teeth serration on both of the pronotumare blunt …….. *Oryzaephilus mercator.*

- There is no temple and it does not contain shape serration teeth on the sides of the pronotum …….. 2.

2- The distance between the compound eyes on the side is small and the terminal tertiary segment of the antennae are suddenly enlarged……. *Tribolium castaneum* .

- The distance between the compound eyes on the side is large and the terminal tertiary segment of the antennae are not suddenly enlarged……3.

3- The head is long , the antennae are gradual in size and the labial palps are triangular in shape …….*Latheticus oryzae* .

- The head is circular , the antennae are capitate and the labial palps are prominent ……..*Trogoderma granarium.*

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