

# **METAMORPHOSIS**

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# Development and early stages of *Mylothris chloris chloris* (Fabricius, 1775) (Lepidoptera: Pieridae)

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- **Abstract:** In this paper, the full life-history of *Mylothris chloris chloris* is discussed with morphological descriptions of its early stages and multiple photo illustrations, documented in Liberia, West Africa. The early stages of *M. chloris* are compared with those of *M. agathina*.
- Key words: Lepidoptera, Pieridae, Mylothris chloris, early stages, pre-imaginal stages, life-history, Loranthaceae.
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### **INTRODUCTION**

*Mylothris chloris* (Fabricius, 1775) is a common pierid butterfly of wide ecological tolerance, distributed in the Guineo-Congolian forest zone, the forest-savannah transition and the fringing humid savannah areas from Senegal and The Gambia to Uganda, Western Kenya and Western Tanzania. Currently two subspecies are recognised, *M. chloris chloris* (Fabricius, 1775) and *M. chloris clarissa* Butler, 1888, with the latter being restricted to outlier rainforest areas in Uganda and Western Kenya (e.g. Kakamega Forest) (Williams, 2020).

The species is replaced in Eastern (including the Horn of Africa) and Southern Africa by the closely related *M. agathina* (Larsen, 1991; Williams, 2020), which was for a long time treated as a subspecies of *M. chloris*, possibly based on similarities of pre-imaginal stages (Larsen 2005). Interestingly, the author did not find published descriptions of the development and early stages of *M. chloris* except for a single drawing illustration of presumably a fully grown larva and pupa in Carcasson (1981).

# MATERIAL AND METHODS

Captive rearing was performed using 3L air-tight tupperware containers, kept indoor at room temperature (24–25°C). Fresh mistletoe leaf shoots were supplied every second day or sometimes earlier, when the amount of food was critically low. The containers were cleaned of frass and leaf-litter every day to avoid the appearance of mould or infections. After eclosion, freshly emerged specimens were placed on a bamboo stand in open air for drying.

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# THE LIFE-HISTORY OF M. CHLORIS CHLORIS

On the 6<sup>th</sup> July 2020, the author spotted a cluster of 116 glossy-yellow eggs laid on the underside of a leaf of a mistletoe (Loranthaceae) species which infested a grapefruit (*Citrus x paradisi*) tree in a garden (Fig. 1) in Yekepa town, northern Liberia at the foothills of Nimba Mountains. As *M. chloris chloris* individuals were previously observed fluttering around the various mistletoe-infested trees, it was suspected that the eggs would probably belong to *M. chloris chloris*, although another, yet unidentified species of *Mylothris* was also seen at the same area. The eggs remained on the plant for another two days before they were removed on the 8<sup>th</sup> July for rearing.



Figure 1 – The grapefruit tree and mistletoe in the residential area Yekepa, Liberia from which the *M. chloris chloris* eggs were collected.

From the eggs, 88 larvae (caterpillars) hatched on the 12<sup>th</sup> July. They consumed the eggshells and probably all other remaining unhatched eggs too, because visibly no remnants remained on the leaf. The tiny first instar larvae remained together, they quickly skeletonised the already browning but still soft leaf, then moved onto a freshly picked one. During movement, they spun a silk line,

creating a small, invisible web on the surface of the leaf, also on the tupperware. They most probably had their first moult after just one day as they appeared almost twice their original size already on the 14<sup>th</sup> July. They also changed their colour from bright yellow to darker bicoloured glossy green-yellow (see detailed morphological description for each larval stage below).

They grew fast but remained bi-coloured for another three days. They divided into smaller groups but remained gregarious, and each group moved to different leaves, possibly as a general survival strategy to avoid food shortage caused by the complete defoliation of a single shoot of the mistletoe plant. Division to smaller clusters could be more efficient also against the various predators and parasitoids. Before feeding, they always moved in a single line (Fig. 2), similarly to various species of Processionary Moth (Thaumetopoeidae), as described in Williams (2020) quoting Trimen & Bowker (1889), describing the feeding habits of *M. agathina* larvae. During their entire larval stage they continued to spin the above mentioned silk line, which they also used to climb back on to the food plant, as in case of disturbance when the larvae tend to jump off with a sudden, twisting movement. On the 21st July, they were already in third instar, still similar in colour but with a new pattern, as a white lateral line appeared on the larvae and the thick setae that cover the entire body of the larvae became apparent (Fig. 3). The moult happened unnoticed between 14th and 21st July, and the larvae ate the moulted skin. In the fourth instar, on the 23<sup>rd</sup> July, they reached their final colour and pattern. They started the next moult already on the 25th July, thus the fourth instar lasted only three days (Fig. 4).



**Figure 2** – *Mylothris chloris chloris* second instar larvae moving to the feeding site in a line.



Figure 3 – Mylothris chloris chloris third instar larvae.

On the 28<sup>th</sup> July the fully grown larvae moved around the storage boxes to find a potentially vertical or sub-vertical position for pupation. Many clustered together on the box or on mistletoe leaves, prepared for pupation. The first larvae pupated on the 30<sup>th</sup> July and all specimens completed their pupation by the 1<sup>st</sup> August.



**Figure 4** – Freshly moulted fifth instar larvae of *M. chloris chloris* and one still in fourth instar (third one from lower left with the smaller black head capsule).

The pupal stage lasted eight days and the first few butterflies eclosed during the night of 7<sup>th</sup> August (Figs 5–7). The majority of specimens eclosed on the 8<sup>th</sup> August, while another 6–8 hatched in the morning hours of the 9<sup>th</sup> August. Altogether, just over 80 specimens eclosed from pupae, resulting in ~70% rearing success. In the next few days, numerous butterflies stayed in the garden, flying around all mistletoe-infested trees. On the 9<sup>th</sup> August a pair in copula, the female fresh and the male worn, was found in the garden (Fig. 8). On the 14<sup>th</sup> August two egg-clusters were spotted on the same mistletoe individual, where the first cluster of eggs was collected.

# **DESCRIPTION OF EARLY STAGES**

**Eggs:** Height approximately 1 mm, width 0.6–0.7 mm, shiny bright yellow, barrel-shaped with longitudinal ribs (best visible with magnifying glass) and eighth warts of lighter yellow colour, forming a circle on the edge of the plate of the micropyle (Fig. 9). They were laid on the mistletoe leaves (either surfaces), evenly spaced in clusters of approximately 100 eggs (leaving usually the height of the egg between each egg). Both the morphology of the eggs and the oviposition pattern are very similar to those of *M. agathina* as described and illustrated in Williams (2020) citing Clark (1949).

#### Larvae:

<u>1<sup>st</sup> instar:</u> Body length 1.3 mm, growing to 2 mm, width 0.3 mm, body shape evenly cylindrical. Colour is bright glossy yellow, covered with short white setae, the head capsule black, slightly broader than the body (Fig. 10). <u>2<sup>nd</sup> instar:</u> Body length 3 mm, growing to 10 mm, width 0.5–1 mm, bi-coloured, darker glossy green and yellow anteriorly, yellow posteriorly with light brown anal plate. This could be a result of a still semi-transparent body filled with green parechymal tissue of the food plant in roughly two-thirds of the body, being digested and converted into frass towards the posterior section of the



Figure 5 – Mylothris chloris chloris male eclosion.



**Figure 6** – *Mylothris chloris chloris* freshly hatched male pumping its wings and excreting waste (meconium) accumulated in the butterfly's body during the pupal stage.



**Figure 7** – *Mylothris chloris chloris* freshly hatched female drying itself.



Figure 8 - Mylothris chloris chloris pair in copula, found in the garden two days after eclosion of the majority of the butterflies.

tract; the fresh frass is also light brown as seen on Fig. 11. The head capsule remains black, its width is equal to its body's (Figs 2,11).

 $3^{rd}$  instar: Body length 15 mm, growing to 20 mm, width 2.5 mm, still bi-coloured but its glossiness is less conspicuous, as it is now densely covered with white hairs and white dots appear all over the body at the base of the setae. Another new feature is a prominent, white longitudinal line laterally that appears also behind the black head capsule, which is slightly narrower than the width of the body, also sparsely covered with white hairs (Fig. 3).

 $4^{\text{th}}$  instar: Body length 22 mm, growing to 25–26 mm, width 4–5 mm, colour more mottled, green-yellow and light brown in colour with a blackish dorsal dashed line, densely scattered with white dots all over the body at the base of the numerous 1–2 mm long white hair-like setae. Head capsule is black, approximately 3 mm in diameter, with white hair-like setae (Fig. 12).



Figure 9 – The evenly distributed eggs of *M. chloris chloris*.



Figure 10 – Mylothris chloris chloris first instar larvae.



Figure 11 – Mylothris chloris chloris second instar larvae.

<u>5<sup>th</sup> instar</u>: Body length 25–26 mm, growing to 33 mm, width 5–6 mm, colour has not changed much, however, the fully grown body is slightly darker and its glossiness has disappeared. The skin is now stretched to its maximum length and width, and the rings between the segments are well visible as they lack the white hair-like setae and the dots at their base. The size of the head capsule is only slightly larger than in 4<sup>th</sup> instar and is visibly narrower than the width of the body (Fig. 13). The fully grown larvae turn glossy ochreous-yellow powdered with white specks before pupation (Fig. 14).



**Figure 12** – *Mylothris chloris chloris* fourth instar larvae, already showing the final pattern and colouration.



**Figure 13** – *Mylothris chloris chloris* fully-grown fifth instar larvae.



**Figure 14** – *Mylothris chloris chloris* pupation (left) and fresh pupa (right, dorsal view).

Pupae: Length approximately 20–23 mm, width 5–6 mm excluding a 2-2.5 mm cephalic projection/horn. Generally, the pupae of M. chloris chloris are identical to the different colour forms of the pupae of *M. agathina*: "The chrysalides both of Agathina and Poppea [= rueppellii] very much resemble bird-droppings with mistletoe seeds intermixed." as re-quoted from Williams (2020) quoting Trimen & Bowker (1889). Also, on the thorax the "small anterior acute black tubercle on each side, and on median ridge a series of three white, blackedged, broad, blunt, tubercular processes, slanting forward", and on the abdomen "along median ridge a series of seven small white black-edged tubercles, of which the second, third, and fourth are blunter and larger than the rest; both the sixth and seventh segments bearing on each side a large, broad, acute, slightly forward-curved, tooth-like white projection". The colour of the fresh pupa is glossy greenish yellow with white specks, later the ground colour turns glossy white or creamy yellowishwhite with black and brown pattern of variable extent. There are pupae with generally darker colouration, where the entire wing-case and the eyes are black and/or brown, but there are also almost completely whitish ones with only small black and/or brown spots on the tip of some of the tubercles (Figs 14-16).



**Figure 15** – Dorsal view and colour patterns of *M. chloris chloris* pupae.



**Figure 16** – Mylothris chloris chloris pupa (lateral view) just before eclosion, showing the forewing's upper side pattern.

Numerous further illustrations of each stages are also available online following the link: <u>https://abdb-</u> africa.org/species/mylothris\_chloris\_chloris

# DISCUSSION

The full life-cycle of *M. chloris chloris* from eggs to the first flight of the newly eclosed imagos lasted 32 days, roughly eight days shorter than in *M. agathina* as described in Williams (2020) quoting Clark (1949, 1994). Nevertheless, the captive rearing setup provided constantly favourable conditions of  $24-26^{\circ}$ C, not affected by the variable weather of the high rainy season. This probably shortened the development of the butterflies and had possibly also had a positive effect also on the success of their rearing, which under natural conditions highly depends on weather, predation, the abundance of parasitoids and even on fungal or viral infection.

All stages of *M. chloris chloris* were virtually identical to those of *M. agathina* – eggs, fully-grown larvae and pupa illustrated in Williams (2020). It is therefore not surprising that Larsen (2005) still treats the latter as a subspecies, based on the morphology of the early stages.

Slight differences in the size of the larvae appear between the development of *M. chloris chloris* and *M. agathina* according to literature records. While the author recorded 10 mm of body length already in the second instar and up to 26 mm in the fourth instar of *M. chloris chloris*, Williams (2020) citing Clark (1949) observed the body length to be from 4–6 mm in second instar and 13–18 mm in fourth instar larvae of *M. agathina*. Although the body size of larvae highly depends on environmental conditions, particularly weather and food quality and availability (e.g. Gupta *et al.*, 2005; Lytan & Firake, 2012), a misinterpretation of instars could have occurred during the rearing of *M. agathina*, as during the rearing of a second batch of *M. chloris chloris*, very similar values of body size in both instars were measured.

The food plant of *M. chloris chloris* in Liberia could not identified to genus level because during the rearing there were no flowers on the plant to aid identification.

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