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## THE LIFE CYCLE OF *MESOSEMIA MEVANIA* (HEWITSON 1857) (RIODINIDAE) IN A LOWER MONTANE HUMID FOREST IN COLOMBIA

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**Abstract.** The life cycle and behavior of *Mesosemia mevania* from eggs to adults in a lower montane humid forest in the Parque Ecológico Piedras Blancas (Antioquia, Colombia) are described and illustrated. For each stage, morphological characteristics are described as well as average size and development time. The average duration of the life cycle was 77.3 days. The host plant was *Notopleura macrophylla* (Rubiaceae) (Ruiz & Pav.) C. M. Taylor.

**Additional key words:** immature stages, *Notopleura macrophyll*

Immature stages of riodinid butterflies are not well known, with host plants and first stadia morphology having been described for only 13–15 % of species (DeVries *et al.* 1992; DeVries 1997; Hall *et al.* 2004). Detailed life cycles have only been published for a handful of species: *Anatole rossi*, by Ross (1964); *Juditha molpe*, by Callaghan (1982); *Stalachtis susana*, by Callaghan (1985); *Synargis brennus*, by (Callaghan 1986); *Nymphidium lisimon attenuatum*, *Phaenochitona sagaris satnius* and *Metacharis ptolomaeus*, by (Callaghan 1988); *Metacharis ptolomaeus* and *Napaea nepos orpheus*, by Callaghan (1991); *Theope guillaumei cecropia*, by DeVries & Hall (1996); *Dodona egeon*, *Dodona eugenes*, *Dodona dipoea*, by Callaghan (1997); *Juditha caucana*, by Hall & Harvey (2001) and *Calydna sturnula*, by Hall *et al.* (2004).

The genus *Mesosemia* (Hübner 1819) contains approximately 130 Neotropical species (Callaghan & Lamas 2004) distributed from Mexico to South America, but especially diverse in lowland forest habitat in the Amazon (DeVries 1997). Although it is one of the most frequently observed genera among the riodinids (DeVries 1997), its biology and immature stadia are not well known (DeVries *et al.* 1992; DeVries 1997; Hall *et al.* 2004).

*Mesosemia mevania* (Hewitson 1857) is commonly found in forest understory in the Andean region in Colombia, Ecuador, Peru and Venezuela. In Colombia it is found in all three Andean Cordilleras, on the Pacific

coast and in the Magdalena and Cauca valleys, at altitudes ranging from 800 to 2400m (García-Robledo *et al.* 2002). Host plants include species of Rubiaceae: *Psychotria poeppigina*, *P. macrophylla* and *Palicourea angustifolia* (Valencia *et al.* 2005).

This article describes and illustrates immature stages of *Mesosemia mevania*, representing the second detailed description of the life cycle of a species belonging to this genus after Stichel (1924).

### MATERIALS AND METHODS

The study was carried out between April and September 2005 in the Parque Ecológico Piedras Blancas nature reserve (Antioquia, Colombia) (6° 8' 20" N - 75° 30' 20" W), at an altitude of 2300 to 2400m, which has an average temperature of 15°C and average annual rainfall of 1965mm (IDEAM 2003). Immature individuals (eggs and larvae of different instars) were collected in the field and reared in laboratory until adult. Some of the eggs were collected immediately after oviposition events by females. Behavioral observations of immature individuals were made in the field and in the laboratory.

**Rearing in the laboratory.** Laboratory rearing was carried out at an average temperature of 16.7°C (max: 23.5 and min: 10.3) and a relative humidity of 76.7% (max: 100 and min: 51.1). Eggs were held in plastic containers until eclosion, after which they were separated at first instar and monitored individually

throughout their life cycle. Larvae obtained in the field were reared as a group, conserving the same number of individuals found on the host plant. Some of these were reared on host plants grown in plant pots in order to observe group behavior.

To determine the development time for the eggs, only those collected immediately after observing the oviposition event were taken into account (for which the date and time of oviposition was known). Development time for larvae, from first instar until chrysalis, was established from all the eggs collected, whether or not the oviposition event was observed.

For each individual for each of the five larval instars we recorded development time, initial and final larval size, width of head capsule and pupal size. Lengths were measured with millimeter lined paper under a stereo microscope. These data were used to calculate minimum, maximum, average and standard deviation values for each instar.

Larvae from each instar were killed in hot water and initially fixed in Kahles solution and finally transferred to 80% alcohol (Borror *et al.* 1989; Holloway *et al.* 1992). These specimens, as well as the adults, were deposited in the Piedras Blancas Entomological Museum (MEPB- National Collections Register No. 147) and the Entomological Collection of the Universidad de Antioquia (CEUA National Collections Register No. 036). The host plant was identified and deposited at the Herbarium of the Universidad de Antioquia (HUA # 147845).

## RESULTS

**Description of immature stages.** *Eggs* (Fig. 1a). Echinoid-shaped eggs, 0.6mm long and 0.8mm in diameter ( $n = 132$ ). Viewed under a stereomicroscope, the chorion has diminutive round cells, surrounded by smooth crests with hairlike structures. The surface surrounding the micropyle is smooth and slightly concave. Eggs are yellow in color for the first few days after oviposition and gradually become clearer until eclosion. Eggs take an average of 17 days to hatch (16–18 days,  $n = 17$ ).

*First instar* (Fig. 1b). Average length at the beginning of the instar: 2.2mm ( $sd = 0.4$ ;  $n = 103$ ) and 3.6mm at the end ( $sd = 0.6$ ;  $n = 64$ ). The body is trapeze-shaped, with small lateral, rounded lobes at the base of segments T2 to A8, covered with dorsal chalazae with long feathery setae. T1 is higher than other segments, with a fleshy lobe or verruca on each side with seven divergent chalazae, most of which are dark; some point cephalad, covering the head. From T2 to A8, two dorsal chalazae are present per segment. In T2–A1 and A8, one lies close to the midline, the other below. From A2 to A7, one is anterior, the other posterior, further dorsad. The closest chalazae to the midline on T2 and A1 are white, the others are dark; on T3 both are dark; on A8 both are white. On A2 to A7, the anterior chalaza is white, the posterior is dark. On A9 and A10, there are three dorsal setae, the two anterior are white and the posterior is dark. The posterior edge of A10 has three dark setae which point caudad. On each side of A8, dorsally posterior to the spiracle, there is an extrusible tentacle, similar to the nectary organs present on myrmecophilous riodinid larvae described by DeVries (1997). T1 has two lateral setae per side, there are three on T2 and T3, one of which is shorter; on A1 to A7, there is a group of four infraspicular setae, one of which is shorter and points dorsad.

On A8, there are six or eight infraspicular setae, one is shorter and points dorsad. On T1 to T3, there are two setae on each leg and two on each proleg of A10. Initially, larvae are transparent and become green after feeding. Spiracles are circular and white, located more ventrally and posterior on the prothorax than on the abdomen. The head is visible at rest; it is round, transparent yellow with transparent long and short setae. The head capsule measures 0.5mm in width on average ( $n = 26$ ), with black ocelli and light brown mandibles. Average duration of the stage is 7.3 days (5–12 days,  $n = 64$ ).

*Second instar* (Fig. 1c). Initially 4.9mm in length ( $sd = 1.5$ ;  $n = 62$ ), final length 6.7mm ( $sd = 1.5$ ;  $n = 59$ ). The body is green with a shape similar to the previous instar, covered with small spinules and chalazae. These are greenish-aquamarine at their base with feathery black setae. Each verruca on the prothorax has numerous divergent chalazae as well as white setae on the anterior edge of the segment extending cephalad and covering the head. On each side of the dorsal part of T2, T3 and A1, there are three or four chalazae, joined at the base, two anterior and one or two posterior. On A2 to A7, there are two dorsal chalazae, joined at the base; one is close to the midline, the other below; there are also one or two posterior chalazae, joined at the base. A8 has two chalazae joined at the base, and like the previous stadium, there is an extrusible tentacle posterior (dorsal) to the spiracle and surrounded by long, thick setae. A9–A10 has numerous black setae dorsally and white setae on the posterior edge. A sclerotized or well-differentiated anal shield was not observed. There is a short seta on and anterior to the spiracle from A1 to A7, as well as a longer one, posterior (dorsal) to the spiracle. These are white and elliptic in shape. There are more abundant lateral lobes, barbed setae and chalazae present on T2 to T8 than in the previous stadium. Over the first pair of legs, there is a verruca with a tuft of setae. The head capsule is on average 0.8mm in width ( $n = 27$ ). Average duration of the stage is 7.2 days (4–10 days,  $n = 59$ ).

*Third instar* (Fig. 1d). Initial length 8.9mm ( $sd = 2.0$ ;  $n = 59$ ), final length 11.2mm ( $sd = 1.5$ ;  $n = 58$ ). The body has a shape similar to the previous instar with a green aquamarine mid-dorsal stripe, extending from T2 to A8, bordered by a thick line interrupted at the middle of each segment. The chalazae are the same color as in the previous stadium. The head, lateral lobes of each segment and ventral region are lime green. The position of the dorsal chalazae on T1 to A1 is the same as the second instar. There are two anterior chalazae on A2 to A7, joined at the base, one closer to the midline, the other below. There also two posterior chalazae, joined at the base, one anterior and above the other. A8, like in the previous instar, has an extrusible tentacle (Fig. 1e). A9 to A10 have numerous dorsal chalazae and setae, as well as on the edge of the segment. There are lateral setae as in the previous stadium. In addition to the dorsal chalazae, there are also smaller, thick, black setae on each segment. The head capsule has an average width of 1.1mm ( $n = 45$ ). Average duration of the stage is 5.8 days (3–9 days,  $n = 59$ ).

*Fourth instar* (Fig. 1f). Initial length 13.2mm ( $sd = 2.0$ ;  $n = 58$ ), final length 16.5mm ( $sd = 2.3$ ;  $n = 58$ ) (Table 2). Body shape, position of setae and chalazae same as previous instar. The color varies slightly, on the dorsal, central region, from T2 to A8 there are three thin aquamarine longitudinal lines. Viewed dorsally, the head is hidden under the prothorax. There are verrucae on top of the last pair of prolegs, with setae hidden by the anal shield. A8 is the same as in previous instar, with an extrusible tentacle. The head capsule averages 1.4mm in width ( $n = 9$ ) (Table 2). Average duration of the stage is 4.2 days (3–7 days,  $n = 58$ ).

*Fifth instar* (Fig. 1g). Initial length 19.2mm ( $sd = 2.5$ ;  $n = 58$ ), final length 20.7mm ( $sd = 1.6$ ;  $n = 58$ ). Body is similar to previous instar, but with numerous clear blue spots on all segments. The width of the prothorax is reduced to almost the width of the head capsule, which is an average of 3mm ( $n = 58$ ). A8 is the same as in the previous instar, with an extrusible tentacle. Average development time until prepupal stage is 4 days (2–7 days,  $n = 58$ ). After the fourth day, the larva begins the prepupal stage (Fig. 1h), becoming dark brown with fine yellow lines bordering the segments and a yellow mid-dorsal line with lateral emerald green spots parallel to this. The prepupa is suspended with the head pointing down, and is adhered to the substrate by the

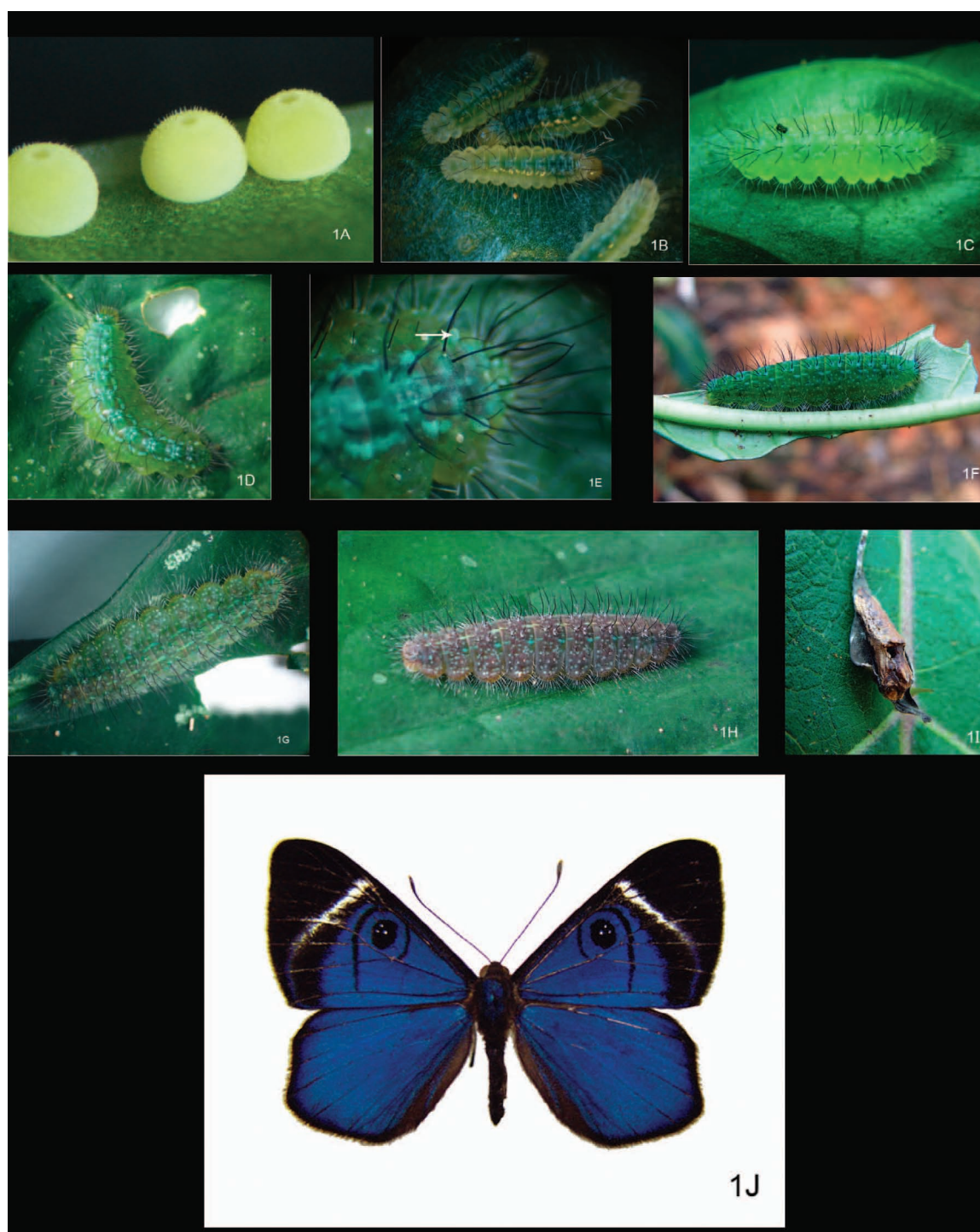


FIG 1. Immature stages and imago: **a)** Egg, **b)** First instar, **c)** Second instar, **d)** Third instar, **e)** Third instar, arrows show extrusible tentacle on A8, **f)** Fourth instar, **g)** Fifth instar, **h)** Prepupa, **i)** Pupa, **j)** Dorsal view, adult male.



cremaster which is surrounded by a silk pad. Average length 19.5mm (sd = 1.3; n = 58). The prepupa takes an average of 3.5 days to transform into the pupa (1–6 days, n = 58). Total development time is 7.5 days (n=58).

**Pupa** (Fig. 1i). Length 19mm, width 5.9mm, on average (n = 58). Pupa is suspended with the head pointing downwards and attached to the substrate by means of a large and flattened cremaster as well as a very thin silk thread, which passes dorsally over the second abdominal segment. Shape and coloration of the body is very cryptic, resembling a dry leaf or the face of an animal. Laterally, the body appears ventrally flattened; the first abdominal segment is depressed with a pair of dark marks. A2 is greater in height and has two crests on either side of the midline, with a line of hairs which look like eyebrows; two black marks join those on A1, giving the appearance of eyes. The mesothorax resembles a nose with two small protuberances with tufts of hairs on either side of the dorsal midline. On abdominal segments A3 to A6, there is an interrupted dorsal midline and lateral black marks. The body is covered with small, white setae, denser nearer the head. The A3 spiracle is hidden. The imago (Fig. 1j) emerges after 28.4 days on average (25–32 day; n = 56).

**Biological observations.** All eggs and larvae of *M. mevania* were observed on *Notopleura macrophylla* (Ruiz & Pav.) C.M. Taylor (Fig. 2), generally on plants between 20 and 60cm in height. A total of 132 eggs were collected on 40 plants, 17 immediately after observing oviposition events by three females. The remaining 115 were collected without recording the time of oviposition. Eggs were distributed in clusters of two to eleven eggs per leaf (one group per plant, 40 groups in total), and only three eggs were found by themselves. Larvae were only present on four of the 40 plants where eggs were found. Generally, each group of eggs was found on the edge of the underside of the leaf, organized in a line, separated by a few millimeters between each egg or in contact with one another.

The complete life cycle of *M. mevania* requires an average of 77.4 days (n = 58) from the egg to the emergence of the adult. The instar requiring most development time is the pupa, with 28.4 days on average, followed by eggs with 17 days. When the larvae hatch, they make a circular hole at the apex of the egg and do not feed on the chorion. In the field, observations showed that larvae of different instars fed on very young plants with succulent stalks and leaves. During the first three stages, larvae are semi-gregarious; two to five individuals were found on the underside of a leaf on each plant. In general they are active both by day and by night, feeding at the same time as each other on the leaves, from the central vein outwards on the underside. In the fourth and fifth instars only one or two larvae were found per leaf, with a maximum of three larvae per plant (field observations). They are active mainly at night, a common trait in many species of riodinids (DeVries 1997). Larvae prefer the upper side of the leaf and feed from the edges inwards, until the whole leaf has been eaten.

In the laboratory, the prepupae generally moved away

from the food plant to fix themselves to the walls of the container and form a chrysalis. In the cases where larvae were reared together on the host plant in a plant pot, they often pupated on the edges of the pot.

Adults were frequently observed in the understory at less than 2m in height, or in clearings, up to 5m above the ground. They flew from one plant to another, where they perched on the leaves with their wings half open. The adults were certainly found where there were immature larvae.

**Parasitoids.** Only 2.3% (n=3) of the 132 eggs collected in the field had been parasitized by *Telenomus* sp (Hymenoptera: Scelionidae). Parasites were not observed in any other stadia.

#### DISCUSSION

**Habitat and food plant.** The study area is principally made up of large pine plantations surrounding small fragments of native forest, dominated by *Quercus humboldtii*. This area corresponds to the lower montane moist forest (Im-MF) life zone (Holdridge 1987).



Figure 2. *Notopleura macrophylla*, host plant of *Mesosemia mevania*.

The host plant, *Notopleura macrophylla* (Ruiz & Pav.). C.M. Taylor (Rubiaceae), previously cited as *Psychotria macrophylla* Ruiz & Pav, is a shrub reaching a maximum height of 2.20m. Plants are distributed in small groups, along streams or in humid, shady areas and exclusively in native forest.

#### Oviposition behavior and behavior in larvae.

The host plant of *M. mevania* in Parque Ecológico Piedras Blancas belongs to the Rubiaceae family, the same host family as the majority of species of *Mesosemia* (Beccaloni *et al.* 2008). Females oviposit in clusters, on young plants and generally where oviposition has not taken place previously. Although an oviposition event was only observed in three females, it is likely that each of the 40 groups of eggs collected came from a single female. Observations showed that eggs within each group generally hatched on the same day and had been placed very close together on the leaf, in a similar fashion to those observed in oviposition events.

According to DeVries *et al.* (1992), DeVries (1997) and Stamp (1980), the majority of riodinids lay isolated eggs. Semi-gregarious behavior of the larvae or oviposition in small clusters has not previously been reported for any species of Mesosemiini. Among the riodinids, semi-gregarious larvae have only been reported for Eurybiini, Riodinini, Helicopiini, Emesiini, Lemoniini and Nymphidiini (DeVries *et al.* 1992), Stalachtis (Callaghan 1985), whereas females of *Ancyluris*, *Emesis*, *Thisbe*, *Theope* and *Nymphidium* oviposit in small clusters or individually, depending on circumstances (DeVries *et al.* 1994). Stamp (1980) states that oviposition behavior depends on the structural and ecological characteristics of the host plant and proposes that a grouped distribution of the host plant, as is the case in *N. macrophila* in the study zone, favors clustered oviposition because it reduces time spent searching for host plants by the female. It is probable that a female of *M. mevania* oviposits on several nearby host plants, given that several plants within a patch are often found to contain eggs. However, this could be a disadvantage to *M. mevania* with regard to predators and parasitoids, although results show that rates of parasitism were very low.

The cryptic appearance may be a factor in avoiding parasitism as suggested by Eisner & Meinwald (1965) and Damman (1986). The yellow color of the eggs resembles that of the leaf underside and the slightly flattened larvae are a very similar green to that of the host plant. The pupae resemble dry leaves. Additionally, IV and V stadia larvae have another apparent method of defence in the form of a green secretion produced when they are disturbed, possibly to repel predators.

Courtney (1984) suggests that oviposition tactics

depend principally on fecundity in species laying groups of eggs, fecundity is high, even though some females may increase the number of eggs depending on the density of the host plant. In the case of *M. mevania*, the limiting factor on number of eggs laid per plant may be the size of the host plant. In the laboratory, observations of the groups of larvae in the containers showed that when they reach instar IV or V, two individuals are completely capable of consuming a whole plant before pupating. It is probably for this reason that larvae go from being semi-gregarious in the first instar to solitary in the last instar. Despite larvae being able to eat a whole plant before pupating, cannibalism was never observed.

In the field, when larvae enter the prepupal stage, it is probable that they move away from the host plant to form a chrysalis close to the ground. This was observed in the laboratory with the larvae reared on plants grown in plant pots.

**Morphological notes.** With regard to larval morphology, the appearance and coloration of the larvae is similar to that of *M. rhodia* (Stichel 1924). The pupa is similar to *Leucochimona vestalis*, illustrated by DeVries (1997). The imago emerges after 28.4 days on average, a shorter time than *Mesosemia rhodia*, which has a 45 day duration (Stichel 1924).

One of the most interesting observations was that of structures similar to tentacle nectary organs located at the same place as in *Synargis brennus* (Callaghan 1986), (Ross 1966) and other myrmecophilous riodinids (DeVries 1997). However, unlike in these species, the body of *M. mevania* is covered by numerous setae. There are bearded setae and chalazae on the lateral lobes, and the tentacles themselves are surrounded by setae, which would make them rather inaccessible to ants. In the field we observed no ants associated with the larvae, no secretions were detected from these organs, and the larvae did not have a reflex of rolling around when disturbed, a behavior to which myrmecophilous larvae are prone (DeVries 1997).

This is the second report on the presence of these structures in *Mesosemia* and Mesosemiini. Stichel (1924) described similar structures for *M. rhodia* on A8. Since Mesosemiini are not known to be myrmecophilous (DeVries 1997), the presence of these organs in *M. mevania* and *M. rhodia* suggests that more detailed field observations on larval behavior and more detailed studies on other species of *Mesosemia* and Mesosemiini would be very interesting.

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