# On the taxonomy of the erythrina moths *Agathodes* and *Terastia* (Crambidae: Spilomelinae): Two different patterns of haplotype divergence and a new species of *Terastia*

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**Abstract**: Members of two species complexes in the family Crambidae feed on ornamental and medicinal plants of the genus *Erythrina* (Fabaceae), commonly known as coral beans and/or coral trees, in tropical and subtropical regions worldwide. Erythrina leaf-rollers (genus *Agathodes*) and erythrina borers (genus *Terastia*), although different ecologically and phenotypically, are sister genera. Each is represented by several very similar species. The erythrina leaf-rollers comprise a species complex that, in the New World, consists of *Agathodes monstralis* Guenée, 1854 **rev. stat.** and *A. designalis* Guenée, 1854. We analyzed morphology and DNA barcoding data of populations throughout the geographic range and concluded that North American *A. monstralis*, which was previously demoted to the status of a subspecies of *A. designalis*, is a valid species. The two are sympatric in Costa Rica. In contrast, the erythrina borer, *Terastia meticulosalis* Guenée, 1854, which has a similar geographic distribution to *A. designalis*, shows no divergence in DNA barcodes and morphology in the New World. However, DNA barcode analyses and detailed morphological examinations of the Asian and African populations of *Terastia meticulosalis* species group show that these represent several species. Thus, a new species, *Terastia africana* Sourakov, **sp. nov.,** is described and the status of *T. subjectalis* Lederer, 1863 as a valid species is confirmed.

**Key words**: DNA barcoding, morphology, cryptic species, biogeography, allopatric speciation, clinal variation, co-evolution, Pyraloidea

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## INTRODUCTION

Worldwide, erythrina leaf-rollers (*Agathodes* Guenée (Crambidae)) and erythrina borers (*Terastia* Guenée (Crambidae)) are economically significant species, feeding on a group of popular ornamental and medicinal plants in the genus *Erythrina* L. (Fabaceae). *Erythrina* encompasses over 100 species of shrubs and trees (Bruneau, 1996) and is distributed throughout subtropical and tropical regions worldwide. These plants have numerous uses, depending on the geographic region, species, and local traditions (e.g., Powell & Westley, 1993; Brown, 2011), and they are frequently cultivated both inside and outside their native ranges. Such cultivation efforts are frequently impacted by the caterpillars of these moths (e.g., Raven, 1974). For instance, the erythrina borer *Terastia meticulosalis* Guenée has recently become a problem for California *Erythrina* growers (pers. comm. between Buzz Uber, Crop Inspection Service, Valley Center, CA and AS). Understanding the taxonomy of pest species can be important for integrated pest management because groups of related species often have unique pheromones, host plant and habitat preferences, distributional ranges, and parasite/parasitoid associations.

The erythrina leaf-rollers belong to the genus *Agathodes*, which includes 15 species that are phenotypically very similar to one another and that are distributed throughout the world's tropics. The Old-World *Agathodes* species have host associations with different species of *Erythrina*; for example, in Africa, *Agathodes musivalis* Guenée, 1854 caterpillars feed on Tiger Claw, *Erythrina variegata* L., and African Coral Tree, *E. caffra* Thunb., while in Asia, *Agathodes ostentalis* (Geyer, 1837), feeds on *E. variegata* and Bat's Wing Coral Tree, *E. vespertilio* Benth. There are three New-World *Agathodes* species, two of which (*Agathodes designalis* Guenée, 1854 and *Agathodes monstralis* Guenée, 1854 (Fig. 1)), are very similar and form a complex that is distributed from Georgia to Arizona in the USA and range as far south as Argentina, feeding on a variety of *Erythrina* species (Sourakov, 2011, and references therein). The third, relatively dissimilar species, *Agathodes transiens* Munroe, 1960, was described from Bolivia. Munroe (1960 p. 172) called *A. transiens* "the most primitive of its group… [showing] remarkable resemblance to *Liopasia*". While there is no life history information for *A. transiens*, the other two species, as caterpillars, share host plant species with *Terastia*, with the former eating leaves and the latter boring into stems (e.g., Sourakov, 2011, 2012). Munroe (1995, unpublished) considered the two to be sister genera based not only on their food choice but also on their morphology. Erythrina borers and erythrina leaf-rollers are found sharing host plant species throughout the tropics. As a consequence, and because of their similar morphology, the two genera have been treated together throughout their taxonomic history (Guenée, 1854; Lederer, 1863; Hulst, 1886; Munroe, 1995; Munroe, unpublished) and they are frequently addressed together by other biologists (e.g., Wakamura & Arakaki, 2004). While there are two distinct species groups in *Terastia*, represented by *T. meticulosalis* and by *T. egialealis* (Walker, 1859), *T. meticulosalis* is always listed by taxonomists as a Neotropical species. Yet, in many taxonomic lists, such as those for Africa, India, or Hawaii, erythrina borers resembling the Neotropical *Terastia meticulosalis* are also listed under this name (e.g., Zimmerman, 1958; Mathew, 2006; Goff, 2011). Part of the confusion came from the revision of the group by Hampson (1899), who considered the Asian *T. subjectalis* Lederer, 1863 synonymous with *T. meticulosalis*. Munroe, in his 1995 checklist, validated both species, but, understandably, due to the format of the publication, had no opportunity to provide his reasons for this decision. Hence, the distribution and taxonomy of *T. meticulosalis* group has remained unclear, and the present publication is an attempt to clarify it using conventional and modern taxonomic tools.

Guenée (1854: 209) described *A. monstralis* and *A. designalis* as distinct species: *Agathodes monstralis* from "Amérique septentrionale?" and *Agathodes designalis* from "Brésil y Amérique septentrionale?". At the end of this work, he provided a single generalized illustration for all *Agathodes* species, and his descriptions of *A. monstralis* and *A. designalis* mostly refer to size and forewing color, without any description of genitalia. Although it is hard to deduce diagnostic characters from his descriptions, it is clear that Guenée observed differences that he believed merited separate species status. Dyar *et al.* (1902: 378) and Holland (1903: 393) treated *A. designalis* as a synonym of *A. monstralis*, probably because the latter name appears first in Guenée (1854), or perhaps because they were more familiar with North American Lepidoptera. However, Munroe (1995) treated *monstralis* as a subspecies of *A. designalis*, and since then the name *A. designalis* has generally been used for the North American populations of erythrina leaf-rollers (e.g., Powell & Opler, 2009).

In the present paper, we analyze morphology and DNA barcodes of a number of populations of *Agathodes* from North, Central and South America and conclude that two species should be recognized, as was originally proposed by Guenée (1854). We contrast this analysis with similar analyses of *Terastia meticulosalis*, in which we found that the haplotypes of the latter species in the New World are extremely uniform. We also redescribe New World *T. meticulosalis* and Asian *T. subjectalis*, and conclude that the African population represents a well-diverged indigenous species, which is described below.

## MATERIALS AND METHODS

**Morphology.** Adult specimens were studied in the collection of the McGuire Center for Lepidoptera and Biodiversity, Florida Museum of Natural History (MGCL) and were borrowed from Bohart Museum, UC Davis, CA. The collection of the Smithsonian Institution (USNM) was also examined, as were DNA-barcoded specimens from the ongoing biodiversity inventory of the Área de Conservación Guanacaste (ACG) in northwestern Costa Rica (Janzen *et al.*, 2009), now deposited in the USNM; specimens from this inventory are uniquely coded with yy-SRNP-xxxxxx and all their data are available at http://janzen.sas.upenn.edu/caterpillars/database.lasso. Dissections of genitalia were performed using the standard procedure, slightly modified from Robinson (1976), of soaking the abdomens in a solution of 10% KOH for 24 hours, removing and descaling the genitalia, staining them with chlorazol black for 30 seconds, and mounting them on a glass slide with euparal under dissecting microscope. Forewing length (from the wing base to the apex) was measured using electronic calipers. Photographs of specimens were taken with a Canon EOS camera using flash, with the exception of Fig. 2C-F, which were taken without flash. For genitalia photographs, the camera was connected to a motorized microptic camera system and controlled using a StackShot macro photography rail and the software EOS Utility 2.6. This allowed for multiple images to be photographed and stitched together using Helicon Focus stacking software. The voucher codes in figures correspond to those on specimen labels. Detailed information on each specimen is provided in the Appendix.

**DNA sequencing and analysis.** We obtained sequences already available from GenBank and BOLD (http://www.boldsystems.org) and new sequences obtained at the MGCL's molecular lab/University of Florida sequencing facility (ICBR). The latter sequences were obtained from leg tissue of *Terastia* and *Agathodes* specimens recently reared by the first author in Florida (see Sourakov, 2011, for details on rearing procedures), and wild-collected dry specimens at MGCL (for specimen data, see Appendix). We amplified a 655-bp segment of mitochondrial cytochrome oxidase subunit I known as the COI barcode region. All genomic extractions, polymerase chain reactions and DNA sequencing were carried out following standard DNA barcoding procedures for Lepidoptera. Total DNA was extracted from butterfly legs using a QIAGEN DNeasy Blood & Tissue kit and eluted in 50-100 µl of Buffer AE. A 687-bp fragment of the COI DNA barcode was amplified using the LepF1 and LepR1 primers (Hajibabaei *et al.* 2006; deWaard *et al.* 2008). Each PCR contained 2 µl DNA templates, 1× PCR buffer, 2 mM $MgCl_2$, 0.1 mM dNTPs, 0.2 µM of each primer, and 0.2 µl Platinum Taq DNA polymerase (Invitrogen) in a total volume of 20 µl. PCR conditions were: 94°C for 1 min; 5 cycles of 94°C for 30 s, 45°C for 40 s, and 72°C for 1 min; 35 cycles of 94°C for 30 s, 51°C for 40 s, and 72°C for 1 min; and 72°C for 5 min. PCR amplicons were analyzed by gel electrophoresis followed by Sanger sequencing. Alignment and editing of the COI barcode sequences were performed in Geneious Pro 5.5.3. Sequences were aligned using BioEdit version 7.0.5.3 (Hall, 1999) and the alignment was manually edited and checked by eye. Data matrices were analyzed using Bayesian inference (BI), as implemented in MrBayes 3.1.2 (Huelsenbeck & Ronquist, 2001; Ronquist & Huelsenbeck, 2003). A GTR substitution model with gamma-distributed rate variation across sites and a proportion of invariable sites was specified before running the program for 10,000,000 generations with default settings of the MrBayes program. The process was monitored by inspection of the Standard Deviation of Bipartition Frequencies (the average standard deviation of split frequencies was 0.007980, and the majority of bipartitions had the value 0.0000) and the Potential Scale Reduction Factor (PSRF had value 1.0000 for the great majority bipartitions, and some bipartitions had a value from 1.001 to 1.006). The first 2500 trees (out of 10000) were discarded prior to computing a consensus phylogeny and posterior probabilities. Dendroscope (Huson *et al.*, 2007) was used to edit trees for publication. New sequences were uploaded to GenBank (see Appendix).

## RESULTS

Analysis of morphological and molecular characters from the *Agathodes designalis* and *Terastia meticulosalis* species groups led us to conclude that each of these groups comprise several allopatric or parapatric species. In *Agathodes*, these results require the elevation of *monstralis* to the species level; in *Terastia*, they confirm *subjectalis* as a valid species, and necessitate the description of an additional species from Africa.

### Redescription of *Agathodes monstralis* Guenée, 1854, Rev. Stat.

(Fig. 1A,B; 2A,C,E; 3D; 4B-D; 5; 6A,C,E; 8C; 9)
*Agathodes designalis monstralis*; (Munroe, 1995)
syn. *Stenurges floridalis* Hulst, 1886, p. 156 (type locality: USA, Florida, Indian River)

**Type:** The Guenée type(s) of *A. monstralis* has not been located in the Museum National d'Histoire Naturelle, Paris (MNHN), where many other pyraloid types of Guenée are otherwise located, and it was very likely lost prior to its arrival there (pers. comm. between staff of that museum and AS). The original description indicates the type locality as "Amérique septentrionale?".

**Adult:** For this study, specimens from Gainesville, Florida, Arizona, Mexico, Dominican Republic, and Costa Rica were used. The genetic data (Fig. 7) and the genitalic morphology (Fig. 4) show potentially taxonomically significant differences between individuals of this species from Florida and Costa Rica, but much more study of individuals from intervening areas is required to resolve this taxonomic issue. Dorsal FW 12-15 mm, basally dirty olive, wine-pink in discal area, marbled wine-olive-rust color postdiscally. Dorsal HW

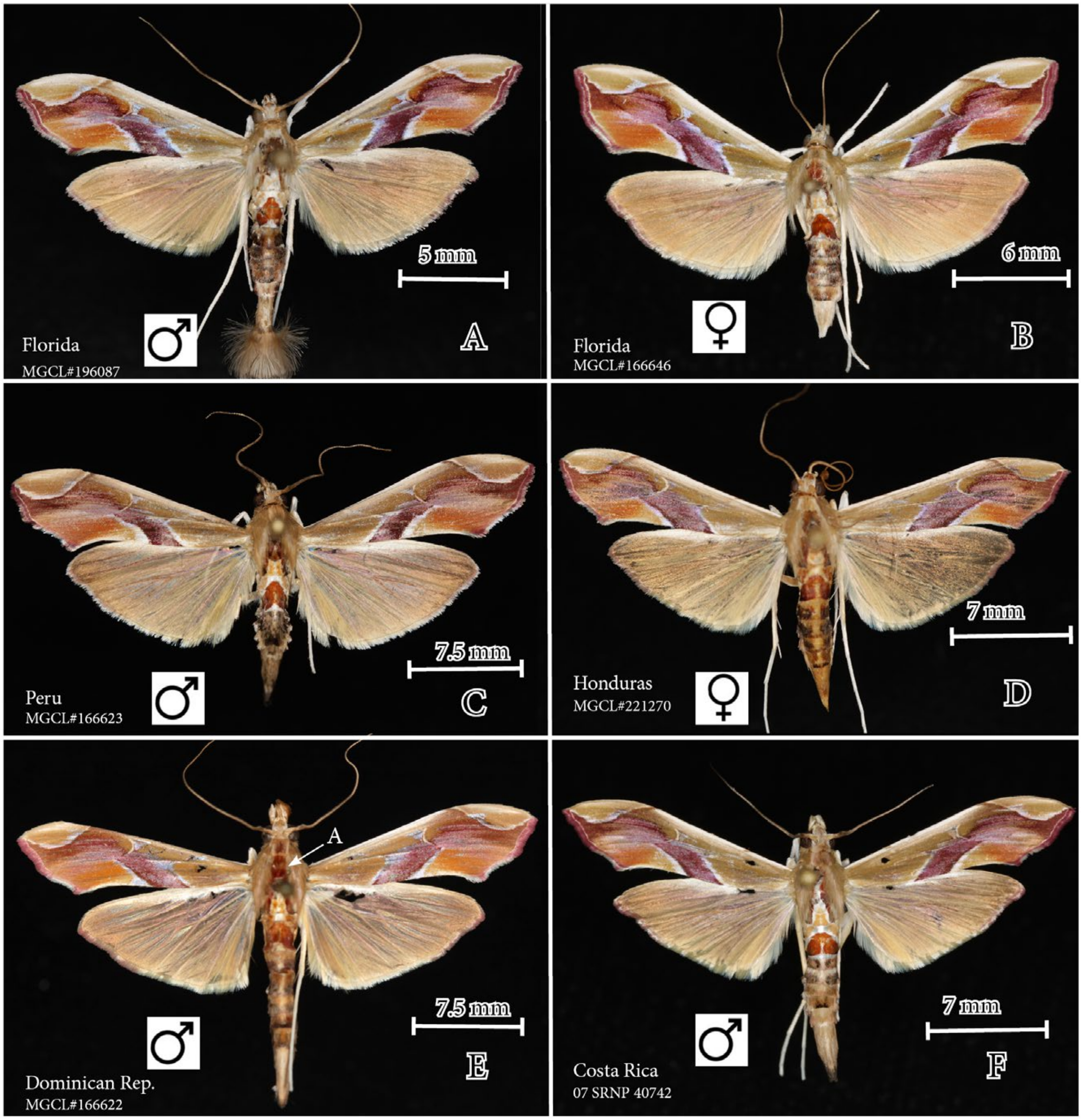


**Fig. 1.** *Agathodes monstralis* and *Agathodes designalis* species complex. See text and Fig. 2, 4 for diagnostic characters.

uniformly pale brown, slightly pinkish towards apex, more reflective in males (Fig. 1A,B; 2A,C,E; 8C), and more opaque in females (Fig. 1B); thorax olive-colored with anteriorly pointing wine-colored 'V'-shaped marking; ventral side of both wings are uniformly beige-colored (Fig. 8C); thorax and abdomen with burgundy, olive and rusty bands bordered with white lines dorsally (Fig. 9A), and much paler ventrally (Fig. 8C); eyes, filiform antennae, palpi and proboscis are olive-colored; legs white (Fig. 9). In males specimens, hair-pencils may protrude as in Fig. 1A, 9C, although usually they are hidden; antennae long, thin, filiform. In live specimens, abdomen is held in an upright position (Fig. 9).

**Male genitalia:** Valvae rounded to more pointed with short sporadic hairs (Fig. 4B). Uncus located dorsally to anal tube, long and narrow, ending in a club-like formation with setae. Vinculum with pair of elongate hair-pencils that extend slightly past uncus (Fig. 4C). Aedeagus simple, straight, with slight trifurcation at apex and about as long as valvae (Fig. 4D).

**Female genitalia:** Simple in structure, short, densely setose lobes, ductus bursae about three times longer than corpus bursae; bursa and ductus bursae membranous, except for lightly sclerotized subbasal collar; bulla seminalis connects at about one-third from proximal end of corpus bursae; sclerotization of antrum limited (Fig. 5).

**Immature stages:** Detailed life history information and illustrations can be found in Sourakov (2011, 2012). The eggs are mostly found in pairs or in triplets. They are flat, pancake-shaped, white and translucent to the extent that the larva, when formed inside, becomes clearly visible. The micropyle is located not mid-dorsally, as it is in most Lepidoptera eggs, but laterally, at the narrow ridge that joins the dorsal and the ventral egg surfaces. The 1st-2nd instars are cryptic and feed on the leaf underside by consuming only the lower cuticle and the mesophyll. In the Floridian spring, larvae live in silk shelters that they build among flowers and flower buds of *Erythrina herbacea*; older larvae of the Floridian summer and fall generations feed on the tender upper leaves, and, as they mature, make nests inside older and lower leaves. These larvae are aposematically colored, with orange head and white subdorsal stripes and dark spots (Fig. 6A,C,E). The pupa is light brown and the larva makes a double-layered cocoon before pupation. The diapause stage, at least in north Florida, is a prepupa sheltered inside the cocoon.

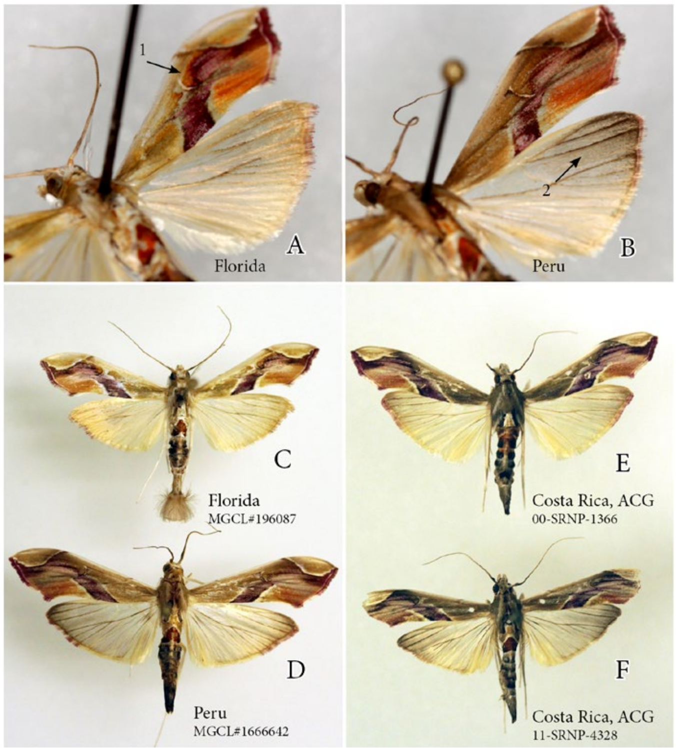


**Fig. 2.** (A, C, E) *Agathodes monstralis* and (B, D, F) *Agathodes designalis* showing slight differences in wing coloration.

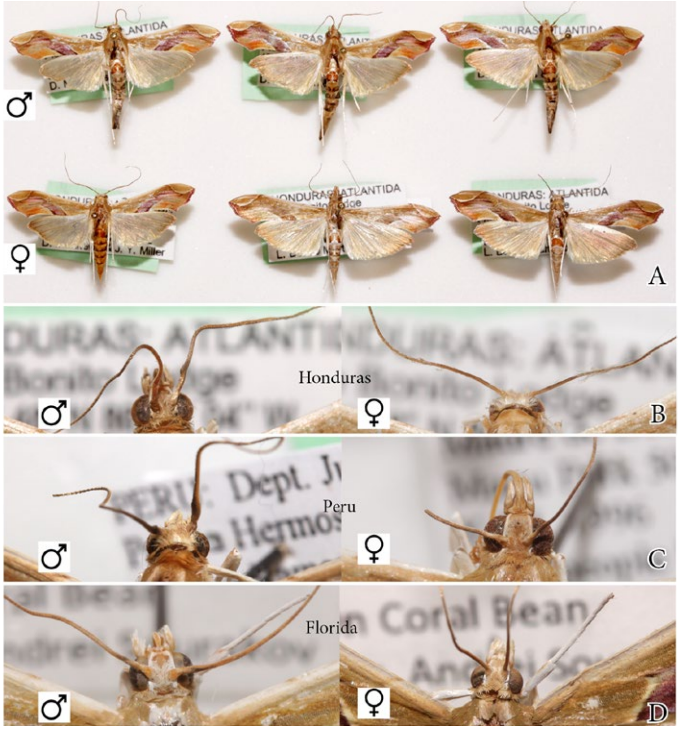


**Fig. 3.** Sexual dimorphism in *Agathodes designalis* and *A. monstralis*: (A) A series of *Agathodes designalis* from Honduras, males with longer abdomens. (B-D) Bulbous organ at the base of male antennae in *A. designalis* (B,C) and lack of thereof in *A. monstralis* (D).

## Redescription and diagnosis of *Agathodes designalis* Guenée, 1854

(Fig. 1C,D; 2B,D,F; 3A-C; 4A)

**Type:** The type(s) of *A. designalis* has also not been located in the MNHN and was very likely lost in a similar manner to the type(s) of *A. monstralis* as discussed above. The original description refers to “Brésil y Amérique septentrionale?” as the type locality.

**Adult:** Specimens from Argentina, Peru, Ecuador, Honduras and Costa Rica were studied. FW 14-18 mm; dorsal FW similar to that of *A. monstralis* as described above; dorsal HW darker towards its apex, grayish-translucent (Fig. 2B,D,F). Sexual dimorphism, as in *A. monstralis*, is very subtle. Specifically, males can be recognized by longer abdomens (due to longer tufts of hairs at tip; Fig. 3A). Also, males have bulbs at base of antennae that are not present in females (Fig. 3B,C); these are conspicuously absent from *A. monstralis*.

**Male genitalia:** Valvae tapered, with longer and denser hair tufts toward apex than in *A. monstralis* (Fig. 4A).

**Female genitalia:** Similar to *A. monstralis*.

**Immature stages:** The fourth instar larvae reared in ACG, northwestern Costa Rica, appear to be different from those of *A. monstralis*, having greatly reduced black markings (Fig. 6B,F) in the few specimens that were photographed.

**Diagnosis:** *Agathodes monstralis* is very similar to *A. designalis*, but that can also be said about its resemblance to all *Agathodes*, such as *A. modicalis* Guenée, 1854, illustrated here from Sumatra, the widespread Asian species *A. ostentalis*, or the African *A. musivalis* (Fig. 8). The entire hindwing of *A. monstralis* is uniformly pale brown with a pink margin (Fig. 2A,C,E), while it is more translucent in *A. designalis* and shows a color gradient shifting from cream-gray at the base to gray-black towards the apex (Fig. 2B,D,F). This can be better observed in photographs taken without flash as in Fig. 2, since the reflection from flash obscures the difference (Fig. 1). If it were not for DNA barcode data from ACG, it would have been hard to identify phenotypic differences between specimens of the two species (Fig. 2E,F), while the differences are more obvious if populations of *A. designalis* from South America are compared with those of *A. monstralis* from Florida (Fig. 2C,D). It seems likely that there is some clinal variation in wing pattern from Central America northwards for *A. monstralis*, and southwards for *A. designalis*. This is also supported by apparently clinal variation in the shape of valvae (Fig. 4F). Guenée, in his original descriptions of the two species, noted that the antennae of male *A. designalis* are swollen at the base, while he made no mention of this character in the original description of *A. monstralis*. While *A. designalis*, for which we have large samples from Peru and Honduras, indeed exhibits this character in males, it is absent in females (Fig. 3A-C). In males from Florida (Fig. 3D (left)) this character is greatly subdued, and it is easy to see why Guenée might have thought it absent. In the male genitalia, the tips of the valvae in *A. designalis* are tapered and equipped with considerably longer setae, which are also more numerous than those of *A. monstralis* (Fig. 4). The valvae maintain more or less the same tapered shape in specimens from Central and South America, while in Florida they are noticeably more oval (Fig. 4B,F,G). The club-like tip of the uncus in *Agathodes monstralis* from Florida appears to be slightly narrower than that of *Agathodes designalis*. Guenée also noticed differences in the size of the moths, namely a 30 mm wing span for *A. monstralis* and 33 mm for *A. designalis*; our statistical analyses of forewing (FW) length suggest that size is a reliable character to separate most populations in Latin America from the population in the southeastern United States. For example, wild-caught Peruvian moths were significantly larger ($P<0.0001$, Student T-test) than wild caught moths from Arizona (FW length of $16.1\pm0.9$ ($N=10$) vs. $14.2\pm0.9$mm ($N=16$)). The Florida reared specimens were even smaller ($12.8\pm0.6$ ($N=19$)), but this could be due to rearing conditions. It would not be surprising if this difference in size was an expression of phenotypic plasticity related to caterpillar diet, as is often the case in moths (e.g. Sourakov, 2015). A comparison of larvae from Florida, Costa Rica and Argentina showed that there are also differences in the immature stages between *A. monstralis* and *A. designalis*: in Florida (*A. monstralis*), 5th instar larvae have on every segment a complete set of prominent black sclerotized tubercles that form paired subdorsal spots, and singular supraspiracular, subspiracular and subventral spots (Fig. 6A). In the same instar of the Costa Rican wet-forest population (*A. designalis*), these spots are reduced to barely noticeable dots in all but the first two thoracic segments (Fig. 6B). The subdorsal white line, on the other hand, is more prominent in the Costa Rican population, and there are black markings on the orange head that are absent in 5th instar larvae of the Florida population. The Costa Rican 6th (final) instar larva has subdorsal spots on every segment similar to those of the mature larva from Florida (Fig. 6C,D), but it lacks the prominent supraspiracular, subspiracular and subventral spots on all but the first two thoracic segments. A mature larva figured by Bourquin (1945) from northern Argentina has a pattern similar to that of the Costa Rican larva figured here, which may mean that the absence of lateral black spots is widespread in *A. designalis*.

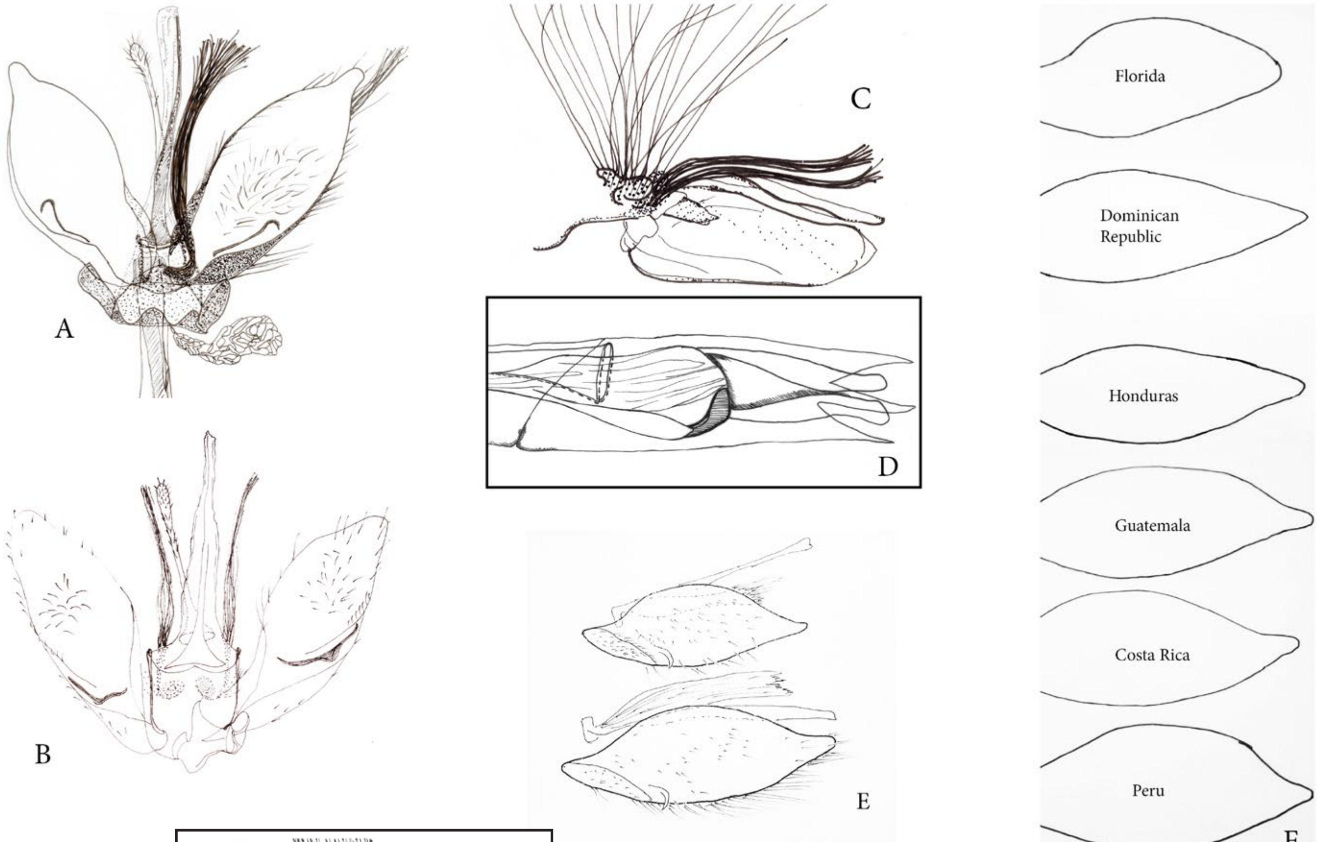


**Fig. 4.** Male genitalia of *Agathodes designalis* from Peru (A) and *Agathodes monstralis* from Florida (B-D): (A, B) Ventral view, open, hair-pencils removed; (C) lateral view; (D) tip of aedeagus. (E) Comparison of valvae of sympatric populations of Costa Rican (ACG) *Agathodes* with different haplotypes: top: 10-SRNP-7374 - *Agathodes designalis* JQ533043 in Fig.7; bottom 05-SRNP-66393 - *Agathodes monstralis* JQ539644 in Fig. 7.
(F) Valvae shape comparison across the distribution range of the species complex indicates possible clinal variation or presence of several species.
Voucher numbers (A)196083; (B)196080; (F) Florida - 196083; Dominican Republic - 196082; Honduras - 221265; Guatemala - 196079; Costa Rica 07-SRNP-40742; Peru - 196083.

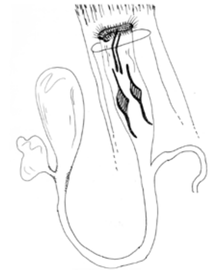


**Fig. 5.** Female genitalia of *Agathodes monstralis* (Florida).

**Table 1.** Genetic distance between *Agathodes* populations. The sister genus representative, *Terastia meticulosalis*, is used for comparison.

| Species/ locality | *Agathodes nr. musivalis* (Gabon) | *Agathodes monstralis* (ACG, Costa Rica) | *Agathodes designalis* (ACG, Costa Rica) | *Agathodes designalis* (Honduras) | *Agathodes monstralis* (Florida) | *Agathodes designalis* (Peru) | *Terastia meticulosalis* (Florida) |
|---|---|---|---|---|---|---|---|
| *Agathodes nr. musivalis* (Gabon) | - | 5.2% | 5.6% | 5.6% | 5.9% | 5.9% | 10.1% |
| *Agathodes monstralis* (ACG, Costa Rica) | 5.2% | - | 1.6% | 1.3% | 1.6% | 1.6% | 10.6% |
| *Agathodes designalis* (Costa Rica) | 5.6% | 1.6% | - | 0.3% | 1.6% | 0.3% | 10.7% |
| *Agathodes designalis* (Honduras) | 5.6% | 1.3% | 0.3% | - | 1.6% | 0.3% | 10.6% |
| *Agathodes monstralis* (Florida) | 5.9% | 1.6% | 1.6% | 1.6% | - | 2.0% | 10.7% |
| *Agathodes designalis* (Peru) | 5.9% | 1.6% | 0.3% | 0.3% | 2.0% | - | 10.8% |
| *Terastia meticulosalis* (Florida) | 10.1% | 10.6% | 10.7% | 10.6% | 10.7% | 10.8% | - |

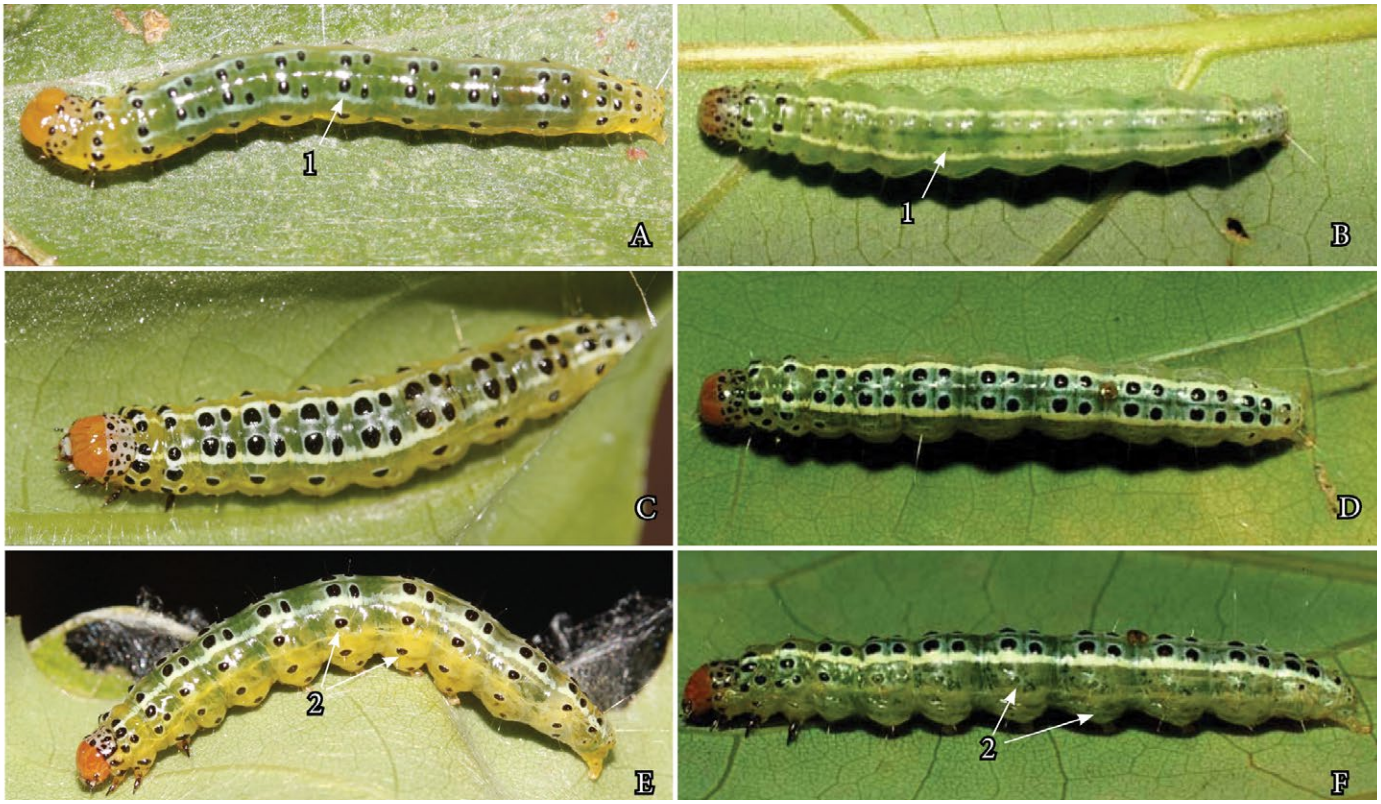


**Fig. 6.** Larvae of *Agathodes monstralis* from Florida (A, C, E) and *Agathodes designalis* from Costa Rica (B, D, F). (A-B) Penultimate instar; (C-F) Last instar.

**Molecular Analysis:** The DNA barcoding analysis (Fig. 7) grouped all Florida individuals with a subset of the ACG specimens, but these populations formed two separate clusters. These clusters represent, in our view, *A. monstralis*, that likely is distributed throughout southern United States and southward to Central America. Remaining ACG specimens clustered with specimens from the tropical wet forests of Honduras and Peru, and represent *Agathodes designalis* (Fig. 7, yellow cluster). The green cluster, representing specimens from ACG, are closer in genitalic characteristics to *A. designalis* but have haplotype and hindwings more similar to those of *A. monstralis*. Much more work, including morphological and molecular studies from extensive areas of the New World and the Caribbean from where the moths are known to occur but as yet unstudied, is necessary to better delineate species boundaries and hybridization zones. However, it seems that at ACG there is a contact point of two species, that in close proximity show a high level of genetic divergence (1.6%) which is similar to that in comparison with the population in Florida, thousands of miles away (Table 1). This genetic distance between two almost sympatric populations is five times greater than that found among the wet forest populations from as far as Costa Rica, Honduras and Peru, which all share haplotypes with differences around 0.3%. Therefore, it seems that there is a high degree of genetic isolation between ACG's cryptic species. By elevating the status of *monstralis* to species level, we bring attention to the fact that there are at least two species present within the *A. designalis* complex in the New World.

## Redescription and diagnosis of *Terastia meticulosalis* Guenée, 1854

(Fig. 10A-C; 11; 12B,E; 13; 14B; 15A,F)

syn. *Megastes coeligenalis* Hulst, 1886, p. 156 (type locality: USA, Florida, Indian River)

**Type:** As with the *Agathodes* species that he described, Guenée's (1854) type(s) of *Terastia meticulosalis* has not been located in the MNHN and was very likely lost prior to his collection arriving at that museum. The original description gives the type locality as "Haiti" and, therefore, if a neotype ever needs to be designated, it should be from the island of Hispaniola, from which the species was originally described.

**Adult:** Here, we figure specimens from Hispaniola as well as from Florida (Fig.10A-C). The Hispaniolan specimens are from the MGCL collection and are labeled as follows: Dominican Rep.: Prov. Barahona, nr. Filipinas Larimar Mine, 6-11 VII 1993, R. E. Woodruff, mercury vap. light. MGCL-195942. The approximate coordinates for this locality are 18°9'11.8044" N, 71°7'8.7384" W. Twelve additional specimens with the same data are also in the MGCL collection. The Florida specimens were reared by the first author from larvae, which were collected on *E. herbacea* behind the Florida Museum of Natural History, Gainesville (29°38'9.5" N, 82°22'10.9" W). Forewing length ranges from 14 mm (some reared specimens from summer stem-boring generation in Florida) to 20 mm (wild caught specimens from Peru), with the average for Hispaniolan series approximately 17 mm. Males and females are quite similar, with female FW markings tending to be more beige-brown (Fig. 10A,B; 11); thorax, abdomen and forewing marbled; hindwing white with dark margin.

In live specimens, the abdomen is held in an upright position and the scales covering it form concave pockets that produce an impression of an insect (praying mantis) head when viewed from most angles (Fig. 11). The fake eyes deflate if the moth is disturbed or killed, and hence in the collection specimens do not appear out of the ordinary. It is likely that these organs function as a false head to deflect or deter attacks by predators, as the moth also makes slight movements with the tip of its abdomen while keeping its true head still (Sourakov, pers. obs.).

**Male Genitalia:** Valvae very square and broad, with slight broad projection at apex of costal region (Fig. 12B, 13A). Antemedial region of

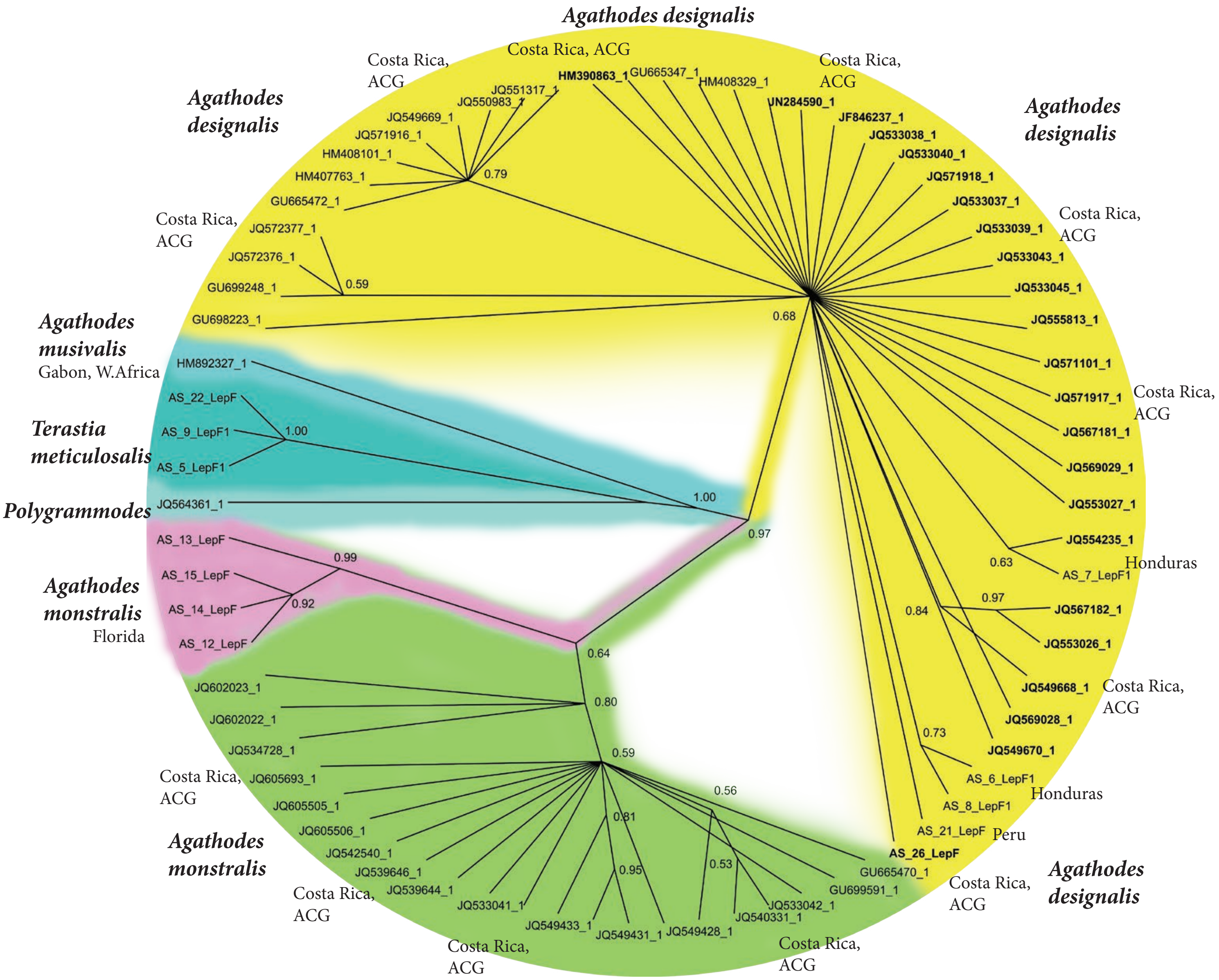


**Fig. 7.** Bayesian inference tree based on 655bp of COI for 69 specimens of *Agathodes*, representing at least two species: *Agathodes monstralis* (highlighted pink) and *Agathodes designalis* (yellow). Specimens forming the clade highlighted green occur sympatrically with *A. designalis* in Area Conservacion Guanacaste, in northern Costa Rica, but are genetically distinct, perhaps part of a cline of *A. monstralis* that extends from Florida to Central America. The numbers at the nodes indicate posterior probability. For details corresponding to specimen numbers in this figure, see Appendix.

each valva with single spinose projection, roughly perpendicular to tegumen; projection with relatively narrow base and slightly hooked tip. Uncus as long as valvae and tapering at tip. Pair of hair-pencils with ovate, membranous base attached to vinculum. Saccus tapered. Aedeagus simple, straight, membranous.

**Female genitalia:** Simple in structure, with ductus bursae 4 times longer than corpus bursae (Fig. 14B).

**Immature stages:** Detailed life history information and illustrations were recently published in Sourakov (2011, 2012). Eggs are usually laid singly at the tip of the branch at the point of petiole attachment to the stem, or among flower buds or bean pods (depending on the season). The eggs are white, shapeless and very soft, but sculptured. The young larvae burrow into the stem, flower, pod, or petiole and are brownish white with a black head and sclerotized prothoracic shield; larvae that feed on seeds turn pink before pupation, while the ones that feed inside the stem do not. The pupa is light brown and enclosed in a double-layered cocoon.

**Diagnosis:** Phenotypically, *Terastia meticulosalis*, the new species from Africa described below, and the Asian *T. subjectalis* are hard to distinguish, while two other species of *Terastia* (*T. egialealis* (Walker, 1859) from Africa and *T. margaritis* (Felder & Rogenhofer, 1875) from India) are quite distinct (Fig. 15). *Terastia meticulosalis* has a deeper notch at the forewing apex, a more tapered hindwing apex, and straighter hindwing margin. *Terastia subjectalis*, described below, have broader dark hindwing margins. The valvae (Fig. 13) of *T. subjectalis* are narrower and more rounded than are those of *T. meticulosalis*. Also, the valvae of *Terastia meticulosalis* each have a slight, broad projection at the apex, differing in shape from *Terastia subjectalis* (Fig. 13B). The two species diverged not only in their DNA barcode but also in their genitalic morphology more than in their wing pattern, a situation commonly encountered in Lepidoptera.

Dissections of *Terastia* specimens from New Guinea revealed a slight morphological divergence from Asian mainland specimens, with the diagnostic character being the shape of the valvae (Fig. 13). Dissections and DNA barcodes of specimens from Malawi suggest that African populations deserve

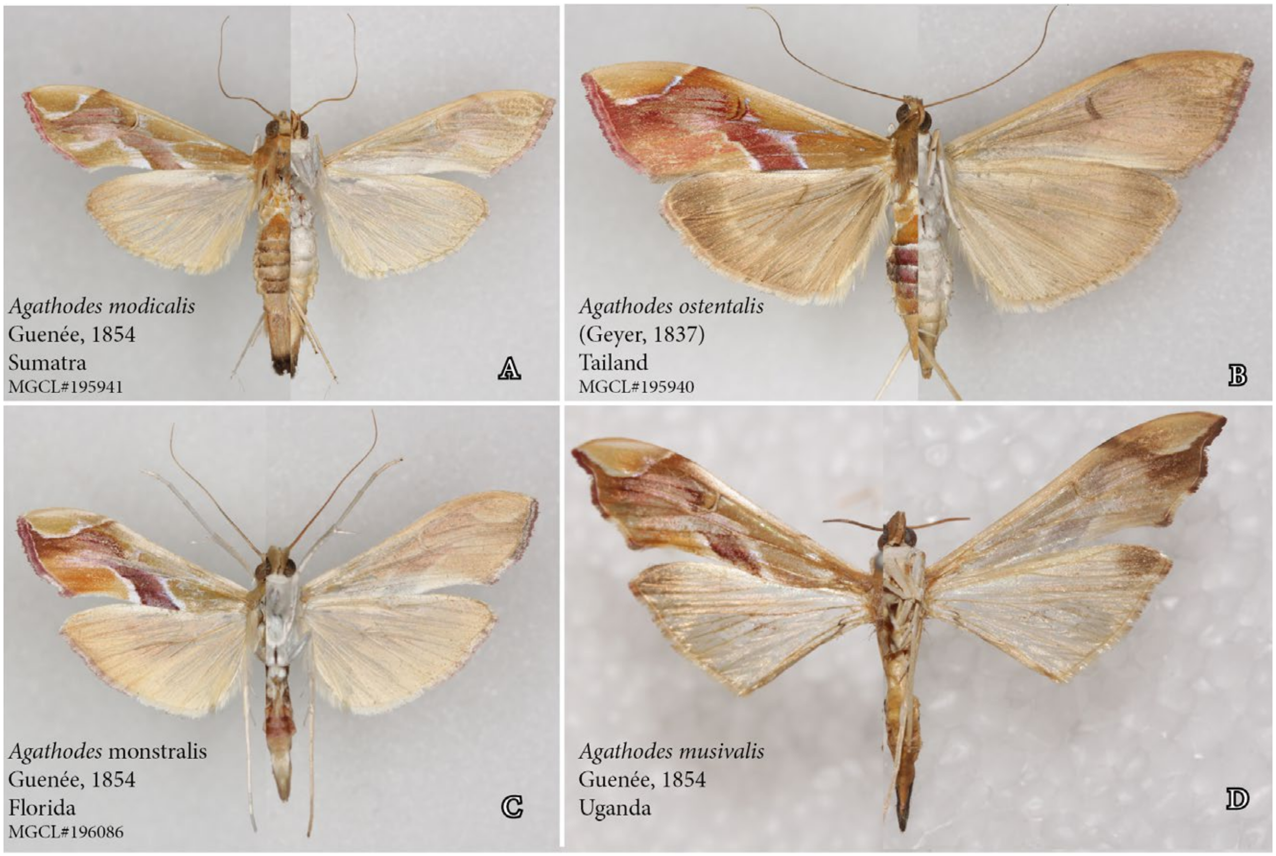


**Fig. 8.** Representatives of the genus *Agathodes* exhibit similar wing patterns worldwide.

the status of separate species, as described below. The wing pattern of the latter species is almost indistinguishable from *T. meticulosalis*, although all specimens examined are lighter in color. Male genitalia of the two new species from Malawi and New Guinea exhibit slight variation in the apical shape of the valvae, as mentioned above; their valvae also each have a single spine whose base is wider than that of the spine on each valva of *T. meticulosalis* (Fig. 12). Finally, the saccus appears to be tapered in *T. meticulosalis*, whereas it appears rounded in the new *Terastia*. However, the saccus is often obscured by the large membranous bases of the hair-pencils during preparation of the dissections, so this observed variation may be artifactual.

All of the *Terastia* species treated here are superficially very similar (Fig. 10), and have been usually referred to as *T. meticulosalis* in various regional checklists and identified as such in major collections, such as, for example, the USNM.

**Molecular Analysis:** Unlike *Agathodes*, in the New World *Terastia meticulosalis* showed a remarkable uniformity in the DNA barcode region. Specimens from as far apart as Florida, Cuba, Honduras, Costa Rica, Ecuador and Peru showed little differences in their haplotypes, with a maximum divergence of 0.2% (Table 2, Fig. 16). At the same time, the Asian relative, *Terastia subjectalis*, which has been frequently considered conspecific with *T. meticulosalis* (e.g., Hampson, 1899; Dyar, 1902; Bhattcharya & Mandal, 1976; Hutson, 1920, etc.), showed a divergence of 6.9-7.1% from its New World congener. The divergence of the African specimens (described as a new species below) is equally high in comparison with both Asian and the New World specimens. This analysis, together with phenotypic differences, suggests that Asian species are sister to the New World/African clades.

## Redescription and diagnosis of *Terastia subjectalis* Lederer, 1863

(Fig. 10D-F; 12C,F; 13; 14C; 15B,G)
syn. *Megaphysa quadratalis* Walker, (1866), p. 1527-1528 (type locality Indonesia: Java)

**Type**: Lederer's type series is stated to be from East Indies and the Philippines, and hence locating and examining these specimens will be crucial for pinpointing the type locality and establishing the identity of this name.

**Adult:** The specimens used in this study are from the collection of the Bohart Museum, University of California, Davis, CA, USA and from the MGCL collection. The Bohart specimens are labeled as follows: PAPUA NEW GUINEA, GULF, Ivimka Res. Sta. – Lakemaku Basin, 120m, 7°44' S 146°30' E, 16-25. XI 1999 S. L. Heydon, N. Schiff , T. A. Sears. 14-26 April 2000 and 1-15 March 2000, T. Sears collector. The single Papuan female available for this study (Fig. 10F) is labeled: PAPUA NEW GUINEA, Morobe Prov – Tekadu, 10 January 2000, T. Sears & Binatung Brigade. "FLMNH-MGCL" labels have been added to the Papuan specimens with numbers 166630 (male) and 166631 (female), which numbers correspond to those of the genitalic dissections (slides). A specimen collected in Vietnam by the first author on April 30, 2013 in Dakrong (Dakrong province) was also studied and is labeled MGCL-166644, which number corresponds to that of the genitalic dissections (slides). Specimens from Java and Sumatra (Indonesia), New Caledonia and the Philippines were examined at the USNM. FW length 18-19 mm in male (N=5), 19 mm in female (N=1). Female (Fig. 10 F) has a darker forewing and broader, more pronounced hindwing margin.

**Male Genitalia:** Valvae very broad, rounded (Fig. 12C, 13). Antemedial region of each valve with single spinose projection, roughly perpendicular to tegumen; projection with relatively wide base and slightly hooked tip. Uncus as long as valvae and tapering at tip. Pair of long, dense hair-pencils with ovate, membranous bases attached to vinculum. Saccus appears rounded. Aedeagus simple, lightly sclerotized at base (Fig. 12F).

**Fig. 9.** *Agathodes monstralis*, Florida, (A-male, B-female) natural position. (C) Pinned male specimen with hair-pencils everted.

**Female genitalia:** Simple in structure (Fig. 14C). Lobes small, strongly setose. Ductus bursae long, with ductus bursae 6 times longer than corpus bursae, narrow, membranous, with a small sclerotized collar at ostial end.

**Distribution and phenology:** The species is likely to occur throughout Southeast Asia, and Australia wherever *Erythrina* host plants are found. The many isolated populations from the Mainland Eurasia, Australia and many islands throughout the Pacific may prove to be, upon closer examination, a complex of several allopatric species.

**Diagnosis:** This species is very similar to *T. meticulosalis* (Fig. 10, 12, 15), but the broader dark border along the hindwing margin may be a reliable character for separating *T. subjectalis* from its New World and African relatives. Specimens from Java and Sumatra (Indonesia) and New Caledonia in the USNM collection share this character with the *T. subjectalis* specimens illustrated. Additional diagnostic characters are found in the male genitalic structures, particularly the shape of the valvae, the width of the base of the single spine on the antemedial valve, and possibly the shape of the saccus. These genitalic characters are most useful for distinguishing *T. subjectalis* from *T. meticulosalis*.

## ***Terastia africana*** Sourakov, **new species**

(Fig. 10G,H; 12A,D; 13; 14A, 15C,H)

**Type:** Holotype: male, MGCL-166636, MALAWI: Mulanje Dist. Mulanje Mtn., Ruo Cyn. 20-23 Feb 2009, 900m, R. Murphy, montane woods. Approximate coordinates: 15°54'2.3508" S, 35°29'24.8778" E. Allotype: female, MGCL-166637, locality same as holotype; MGCL numbers correspond to those of the genitalic dissections (slides).

**Adult:** Male (Fig. 10G) smaller (FW=20 mm) than female (Fig. 10H) (FW=23mm). Otherwise, sexes quite similar to one another and to *T. meticulosalis*, as described above.

**Male Genitalia:** Valvae very broad and rounded (Fig. 12A, 13). Antemedial region of each valve with single spinose projection, roughly perpendicular to tegumen; projection with relatively narrow base and slightly hooked tip. Uncus as long as valvae and tapering at tip. Long, dense hair-pencils with ovate, membranous bases attached to vinculum. Aedeagus simple, lightly sclerotized (Fig. 12D).

**Female genitalia:** Simple in structure (Fig. 14A). Lobes small, strongly setose. Ductus bursae long, about 4-5 times longer than corpus bursae, narrow, membranous, with a small sclerotized collar at ostial end.

**Etymology:** *Terastia africana* **n. sp.** is named after the continent where it was collected.

**Distribution and phenology:** The species is likely to be widespread in Africa wherever *Erythrina* host plants are found. More material from Africa is necessary in order to assess the distribution of this species. Additional African specimens from this complex examined at the USNM collection from Kenya, Malawi, and South Africa were similar in wing pattern to the types illustrated here.

**Diagnosis:** This new species is very similar to *T. meticulosalis* and *T. subjectalis* (Fig. 10, 15), but because they are allopatric, knowing the locality from which the specimen came will assist in identification even without dissection or DNA barcoding. Some male genitalic characters distinguish *T. africana* from *T. meticulosalis*, including the shape of the valvae, the width of the base of the single spine on the antemedial valve, and possibly the shape of the saccus (Fig. 12-13). *Terastia africana* can be distinguished from *T. subjectalis* by the lack of a broader dark margin on the hindwing, and from sympatric *T. margaritis*, by its wing pattern (Fig. 15).

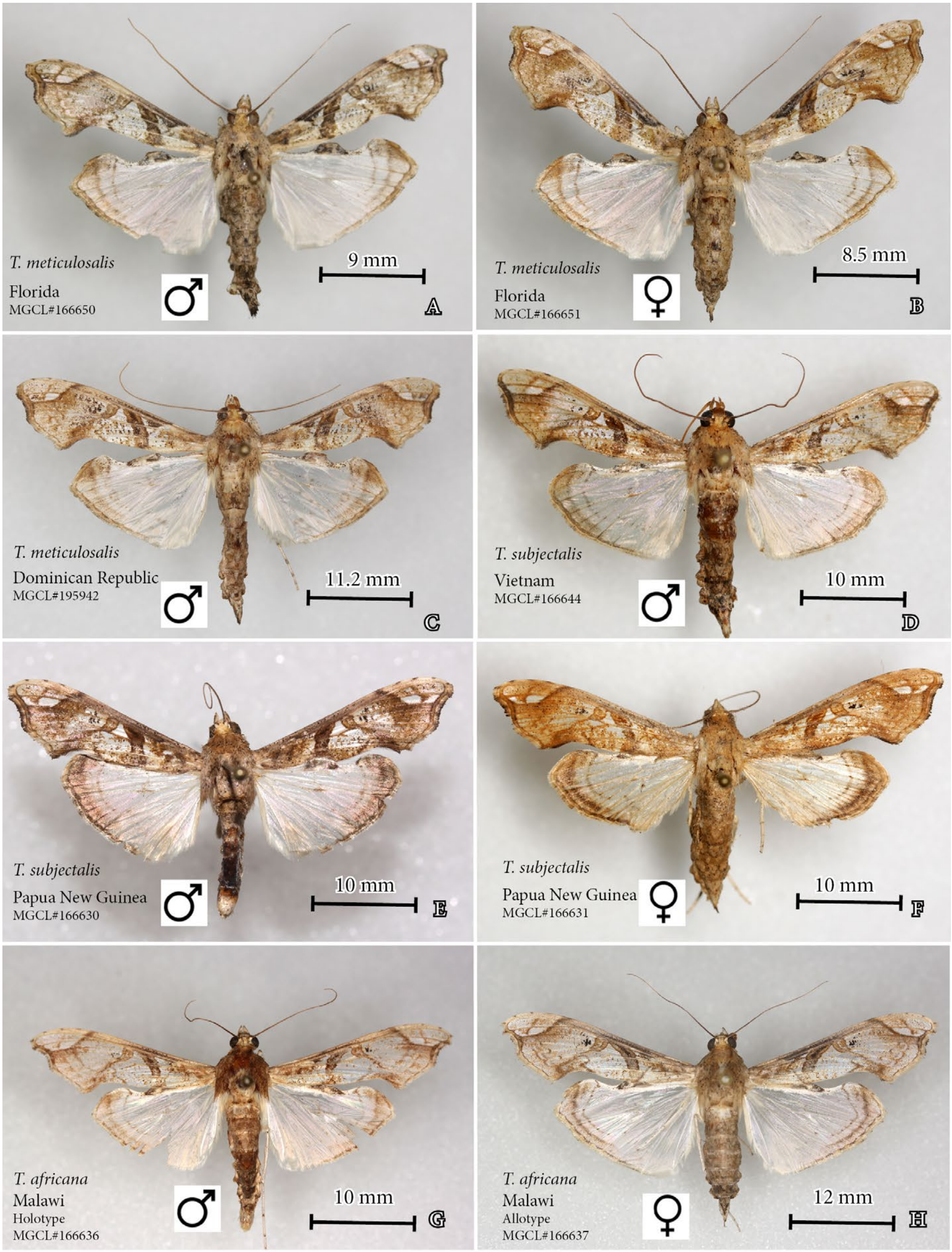


**Fig. 10**. *Terastia meticulosalis* species group, including (A-C) *T. meticulosalis*, (D-F) *T. subjectalis*, and (G-H) *T. africana* **sp. n**.

**Fig. 11**. *Terastia meticulosalis*, Florida, natural posture (A,D - female). (B,C,E,F - male). Both sexes show characteristic "false head" formed by specialized scales. These scales create hollow bulbous structures resembling insect eyes. This feature is pronounced only in live specimens in resting position, and the "false eyes" deflate when the moth is disturbed, and are almost unnoticeable in dead specimens.

**Table. 2.** Genetic distance (based on DNA-barcode region) between analyzed populations of *Terastia.*

| ***Terastia* population** | *T. meticulosalis* Honduras | *T. meticulosalis* Florida | *T. subjectalis* Vietnam | *T. africana* Malawi |
|---|---|---|---|---|
| *T. meticulosalis* (Honduras) | - | 0.2% | 7.2% | 6.9% |
| *T. meticulosalis (*Florida) | 0.2% | - | 7.1% | 6.7% |
| *T. subjectalis* (Vietnam) | 7.2% | 7.1% | - | 5.3% |
| *T. africana*, **sp. n.** (Malawi) | 6.9% | 6.7% | 5.3% | - |

**Molecular Analysis:** The DNA barcode obtained from the type specimen shows a 6.7-6.9% distance from *T. meticulosalis* and 5.3% distance from Asian *T. subjectalis* (Table 2, Fig. 16). This distance is 30 times greater than occurs among Neotropical specimens and is equivalent to that separating *T. meticulosalis* and *T. subjectalis*, strongly supporting the separate species status of the African population.

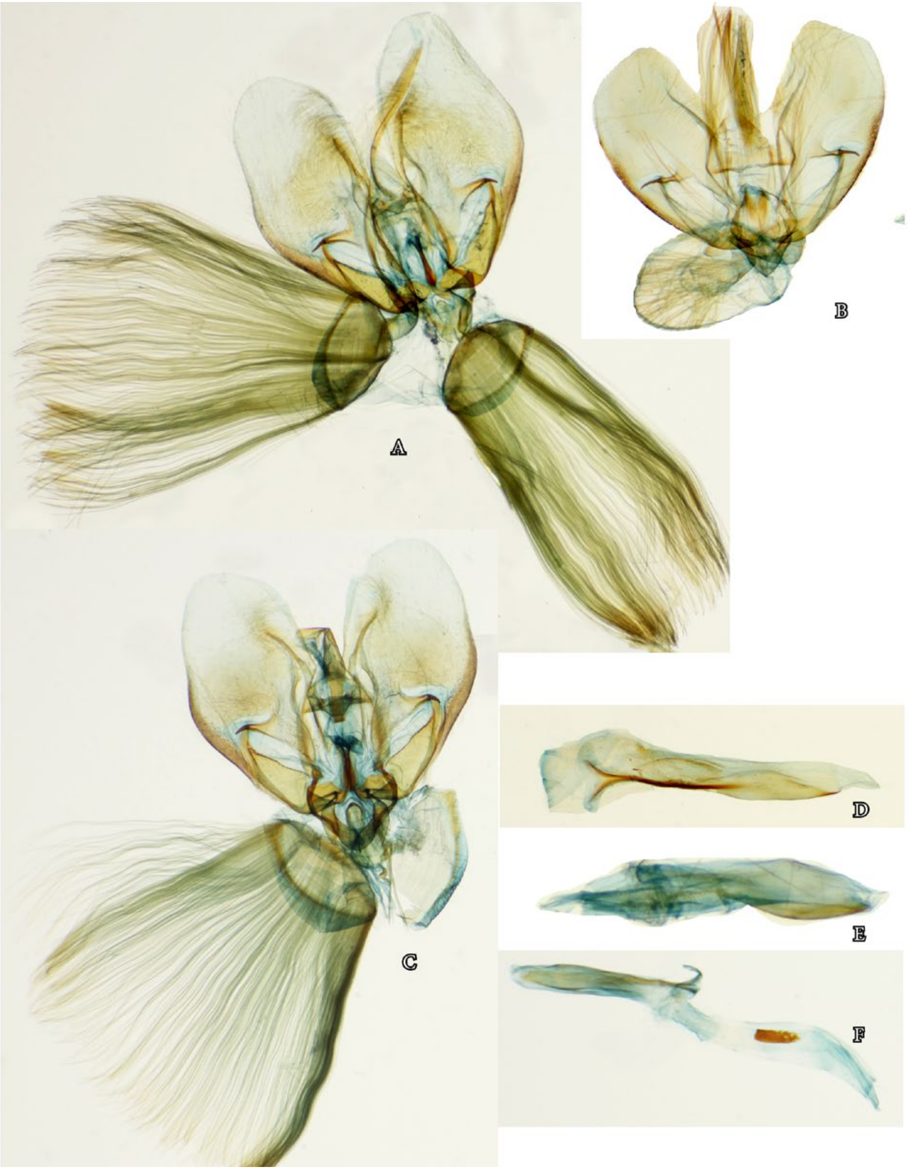


**Fig. 12.** Male genitalia (A-C), with aedeagi (D-F) separated: (A, D) *Terastia africana*, **sp.n.**, Malawi; (B,E) *T. meticulosalis*, Florida; (C, F) *T. subjectalis*, Papua New Guinea. Voucher numbers (A,D) 166636, (B,E) 165868, (C,F) 166630.

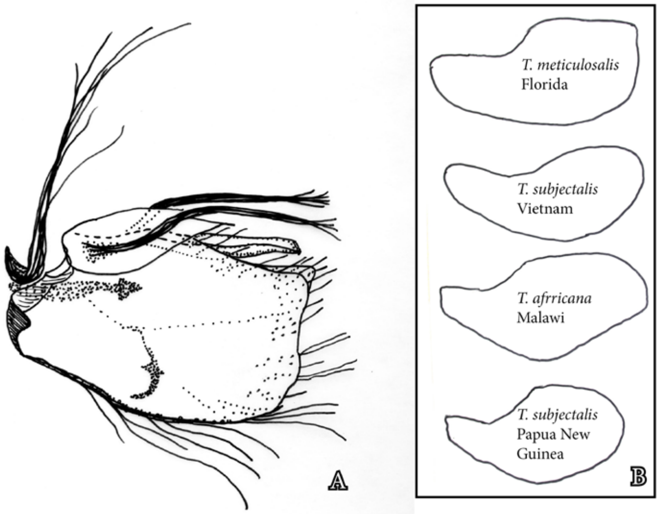
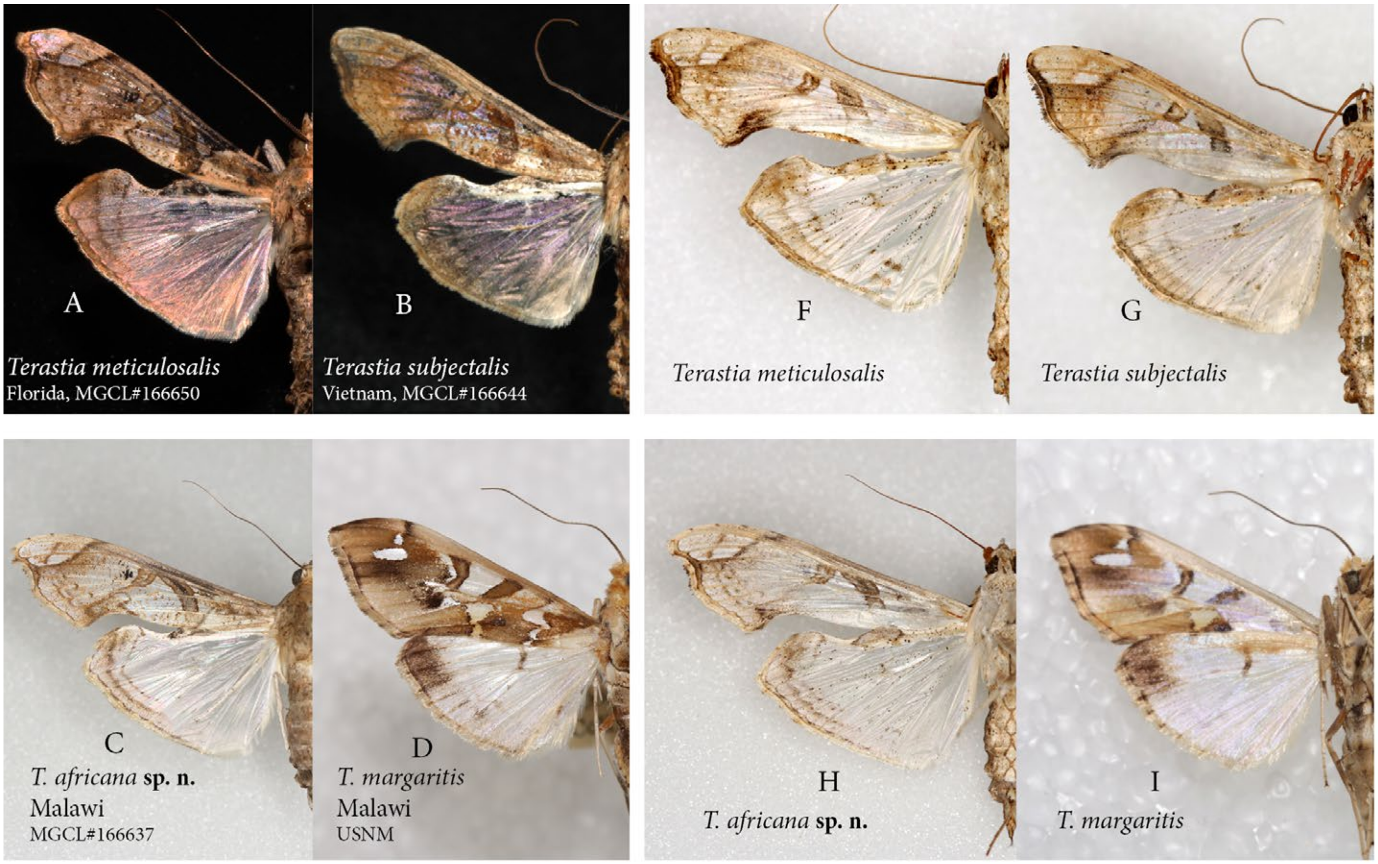


**Fig. 13**. (A) Male genitalia of *Terastia meticulosalis*, Florida, lateral view; (B) Valvae shape differences in three species of *Terastia*. Voucher numbers as in Fig. 12, *T. subjectalis* (Vietnam) – 166644.

## DISCUSSION

This present study emphasizes the importance of routine DNA barcoding as a tool for answering and posing questions; the results of our molecular analysis led us to examine other characteristics of *Agathodes* and *Terastia* specimens. Considering the proposed co-evolution of the two genera through niche partitioning (Sourakov, 2011), the observed lack of divergence in *Terastia meticulosalis* in the New World as compared to *Agathodes* probably lies in its presumed superior dispersal ability. Sourakov (2012) observed that *Terastia meticulosalis* has a strong hovering flight similar to that of sphinx moths (Sphingidae). *Terastia meticulosalis* can live in captivity for at least three weeks, developing eggs rather slowly and ovipositing solitarily. All of these features may facilitate continuing gene exchange among populations.

However, populations of the *T. meticulosalis* species group found on different continents are clearly non-conspecific, and



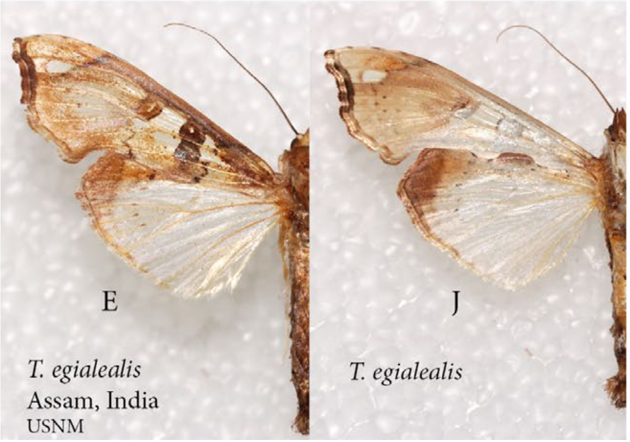


**Fig. 15.** Members of the genus *Terastia.* (A-E) Dorsum; (F-J) Ventrum.

**Fig. 14.** Female genitalia: (A) *Terastia africana*, **sp.n.**, Malawi; (B) *T. meticulosalis*, Florida; (C) *T. subjectalis*, Papua New Guinea. Voucher numbers (A) 166637, (B) 165869, (C) 166631.

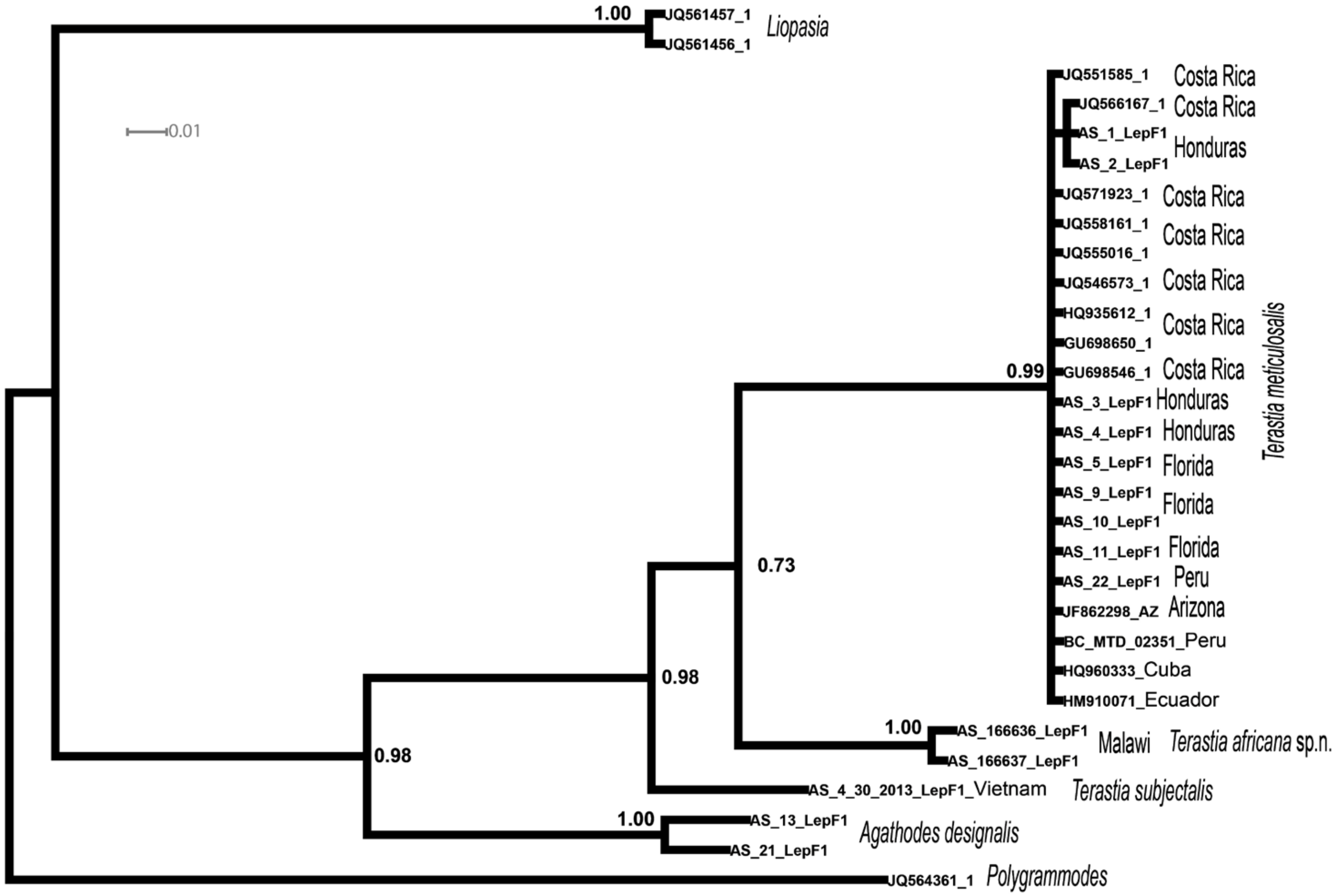


**Fig. 16.** Bayesian inference tree based on 655bp of COI for erythrina borers. *Terastia meticulosalis* is represented by 22 specimens from throughout the New World that show no divergence. Other lineages represent almost identical *Terastia subjectalis* from Asia, *T. africana* **sp. n.**, two closely related genera, *Agathodes* and *Liopasia*, and an outgroup genus, *Polygrammodes*. See Appendix for details corresponding to specimen numbers in this figure.

while their phenotypic divergence is modest, their genetic divergence is substantial. The genus *Liopasia* Möschler is also frequently confused with *Terastia* and *Agathodes* due to wing pattern similarities, and they have also recently been found to utilize *Erythrina* seeds in their larval diet (Pereira *et al.,* 2014). The inclusion of *Liopasia* and *Agathodes* in the analysis of *Terastia* DNA barcodes helped to demonstrate that the two genera are well diverged.

In contrast with almost complete absence of genetic divergence in Terastia meticulosalis throughout its distribution range in the New World, DNA barcodes of *Agathodes* formed at least three separate clusters in the same area. One can hypothesize that the observed differences in haplotype divergence are due to differences in the biology of the two moths. In the lab, *Agathodes* live a shorter time and are more fragile (e.g., die easily from dehydration) than are *Terastia* (Sourakov, pers. obs.), and they may not venture as far in search of food plants. However, *Agathodes* has been as successful as *Terastia* in colonizing remote tropical islands across the world, with 15 species recognized. It is possible that a closer look at additional *Terastia* populations will also yield additional species, just as has happened in *Agathodes*. We view this paper as a preliminary investigation of the taxonomy of erythrina moths.

## ACKNOWLEDGMENTS

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## **Appendix:** Specimen information used in the analysis

| **Specimen Figs** | **Voucher specimen #** | **Species name** | **Locality - Country** | **Locality-Additional** | **Sequencing facility** | **GenBank number** |
|---|---|---|---|---|---|---|
| JQ554235.1 | DHJanzen:09-SRNP-100033 | *Agathodes designalis* | Costa Rica | ACG, Sector Rincon Rain Forest, Rio Negro, E. Cuatro Bocas (habitat: Rainforest) | BOLD | JQ554235.1 |
| JQ533038.1 | DHJanzen:10-SRNP-7355 | *Agathodes designalis* | Costa Rica | ACG, Sector San Cristobal, Tajo Angeles, (habitat: dry-rainforest) | BOLD | JQ533038.1 |
| JQ533040.1 | DHJanzen:10-SRNP-7357 | *Agathodes designalis* | Costa Rica | ACG, Sector San Cristobal, Tajo Angeles, (habitat: dry-rainforest) | BOLD | JQ533040.1 |
| JQ533037.1 | DHJanzen:11-SRNP-40809 | *Agathodes designalis* | Costa Rica | ACG, Sector Rincon Rain Forest, Sendero Venado (habitat: rainforest) | BOLD | JQ533037.1 |
| JQ533039.1 | DHJanzen:10-SRNP-7356 | *Agathodes designalis* | Costa Rica | ACG, Sector San Cristobal, Tajo Angeles, (habitat: dry-rainforest) | BOLD | JQ533039.1 |
| JQ533043.1 | DHJanzen:10-SRNP-7374 | *Agathodes designalis* | Costa Rica | ACG, Sector San Cristobal, Tajo Angeles, (habitat: dry-rainforest) | BOLD | JQ533043.1 |
| JQ533045.1 | DHJanzen:10-SRNP-7378 | *Agathodes designalis* | Costa Rica | ACG, Sector San Cristobal, Tajo Angeles, (habitat: dry-rainforest) | BOLD | JQ533045.1 |
| AS-26-LepF1 | DHJanzen:07SNRP-40744 and MGCL-166643 | *Agathodes designalis* | Costa Rica | ACG, Sector Rincon Rain Forest, Rio Francia Arriba, 400m (habitat: rainforest) | MGCL | KU252856 |
| AS-6_LepF1 | MGCL-221268 | *Agathodes designalis* | Honduras | Atlantida: Pico Bonito (habitat: rainforest) | MGCL | KU252846 |
| AS-7_LepF1 | MGCL-221269 | *Agathodes designalis* | Honduras | Atlantida: Pico Bonito (habitat: rainforest) | MGCL | KU252847 |
| AS-8_LepF1 | MGCL-221270 | *Agathodes designalis* | Honduras | Atlantida: Pico Bonito (habitat: rainforest) | MGCL | KU252848 |
| JQ551317.1 | DHJanzen:08-SRNP-105957 | *Agathodes designalis* | Costa Rica | ACG, Rincon Rain Forest, Estacion Caribe (habitat: rainforest) | BOLD | JQ551317.1 |
| GU698223.1 | DHJanzen:09-SRNP-104311 | *Agathodes designalis* | Costa Rica | ACG, Sector Santa Elena, Potrero Grande (habitat: dry forest) | BOLD | GU698223.1 |
| JQ572376.1 | DHJanzen:06-SRNP-107498 | *Agathodes designalis* | Costa Rica | ACG, E. Rincon de la Vieja, Hotel Borinquen, Canopy Tours (habitat: dry-rainforest) | BOLD | JQ572376.1 |
| JQ572377.1 | DHJanzen:06-SRNP-107499 | *Agathodes designalis* | Costa Rica | ACG, E. Rincon de la Vieja, Hotel Borinquen, Canopy Tours (habitat: dry-rainforest) | BOLD | JQ572377.1 |
| JQ549669.1 | DHJanzen:08-SRNP-104052 | *Agathodes designalis* | Costa Rica | ACG, Sector Cacao, Bosque aguirre, E. Vol. Orosi (habitat: dry-rainforest) | BOLD | JQ549669.1 |
| JQ549668.1 | DHJanzen:08-SRNP-104051 | *Agathodes designalis* | Costa Rica | ACG, Sector Cacao, Bosque aguirre, E. Vol. Orosi (habitat: dry-rainforest) | BOLD | JQ549668.1 |
| JQ549670.1 | DHJanzen:08-SRNP-104053 | *Agathodes designalis* | Costa Rica | ACG, Sector Cacao, Bosque aguirre, E. Vol. Orosi (habitat: dry-rainforest) | BOLD | JQ549670.1 |
| JQ550983.1 | DHJanzen:08-SRNP-105496 | *Agathodes designalis* | Costa Rica | ACG, Javalina, Manta Pizote RAIN | BOLD | JQ550983.1 |
| JQ569029.1 | DHJanzen:07-SRNP-106297 | *Agathodes designalis* | Costa Rica | ACG, Rincon Rainforest, Estacion Caribe (habitat: rainforest) | BOLD | JQ569029.1 |
| JQ553027.1 | DHJanzen:08-SRNP-107833 | *Agathodes designalis* | Costa Rica | ACG, Estacion Botarrama, Manta Loma (habitat: rainforest) | BOLD | JQ553027.1 |
| JQ553026.1 | DHJanzen:08-SRNP-107832 | *Agathodes designalis* | Costa Rica | ACG, Estacion Botarrama, Manta Loma (habitat: rainforest) | BOLD | JQ553026.1 |
| GU698517.1 | DHJanzen:09-SRNP-104912 | *Agathodes designalis* | Costa Rica | ACG, Sector Santa Elena, La Angostura, (habitat: dry forest) | BOLD | GU698517.1 |
| HM408101.1 | DHJanzen:09-SRNP-108586 | *Agathodes designalis* | Costa Rica | ACG, Estacion Botarrama, Manta Porton (habitat: rainforest) | BOLD | HM408101.1 |
| HM408329.1 | DHJanzen:09-SRNP-108800 | *Agathodes designalis* | Costa Rica | ACG, Estacion Botarrama, Manta Porton (habitat: rainforest) | BOLD | HM408329.1 |
| AS-21_LepF1 | MGCL-166642 | *Agathodes designalis* | Peru | Dept. Junin, Pampa Hermosa Lodge, nr. San Ramon, 1220 m. (habitat: rainforest) | MCGL | KU252854 |
| GU665347.1 | DHJanzen:09-SRNP-55086 | *Agathodes designalis* | Costa Rica | ACG, Sector Santa Maria, Sendero Termales (habitat: dry-rainforest) | BOLD | GU665347.1 |
| JQ549428.1 | DHJanzen:98-SRNP-3455 | *Agathodes designalis* | Costa Rica | ACG, Sector Cacao, Estacion Cacao (habitat: rainforest) | BOLD | JQ549428.1 |
| GU665471.1 | DHJanzen:08-SRNP-40595 | *Agathodes designalis* | Costa Rica | ACG, Rt 917, E. Cuatro Bocas (habitat: rainforest) | BOLD | GU665471.1 |
| GU665472.1 | DHJanzen:08-SRNP-40596 | *Agathodes designalis* | Costa Rica | ACG, Rt 917, E. Cuatro Bocas (habitat: rainforest) | BOLD | GU665472.1 |
| HM390863.1 | DHJanzen:09-SRNP-1139 | *Agathodes designalis* | Costa Rica | ACG, Rt 917, E. Cuatro Bocas (habitat: rainforest) | BOLD | HM390863.1 |
| JQ571101.1 | DHJanzen:06-SRNP-105540 | *Agathodes designalis* | Costa Rica | ACG, Sector Santa Elena, La Angostura (habitat: dry forest) | BOLD | JQ571101.1 |
| HQ934753.1 | DHJanzen:10-SRNP-106182 | *Agathodes designalis* | Costa Rica | ACG, Rt 917, E. Cuatro Bocas, N. Rincon de la Vieja (habitat: rainforest) | BOLD | HQ934753.1 |

| **Specimen Figs** | **Voucher specimen #** | **Species name** | **Locality - Country** | **Locality-Additional** | **Sequencing facility** | **GenBank number** |
|---|---|---|---|---|---|---|
| HM407763.1 | DHJanzen:09-SRNP-104431 | *Agathodes designalis* | Costa Rica | ACG, Sector Santa Elena, Potrero Grande (habitat: dry forest) | BOLD | HM407763.1 |
| GU699248.1 | DHJanzen:09-SRNP-106464 | *Agathodes designalis* | Costa Rica | ACG, Sector Cacao, Estacion Cacao, (habitat: dry-rainforest) | BOLD | GU699248.1 |
| JQ555813.1 | DHJanzen:07-SRNP-113791 | *Agathodes designalis* | Costa Rica | ACG, Rincon Rainforest, Estacion Caribe (habitat: rainforest) | BOLD | JQ555813.1 |
| JQ571916.1 | DHJanzen:06-SRNP-106829 | *Agathodes designalis* | Costa Rica | ACG, E. Rincon de la Vieja, Hotel Borinquen, Canopy Tours (habitat: dry-rainforest) | BOLD | JQ571916.1 |
| JQ571918.1 | DHJanzen:06-SRNP-106831 | *Agathodes designalis* | Costa Rica | ACG, E. Rincon de la Vieja, Hotel Borinquen, Canopy Tours (habitat: dry-rainforest) | BOLD | JQ571918.1 |
| JQ571917.1 | DHJanzen:06-SRNP-106830 | *Agathodes designalis* | Costa Rica | ACG, E. Rincon de la Vieja, Hotel Borinquen, Canopy Tours (habitat: dry-rainforest) | BOLD | JQ571917.1 |
| JN284590.1 | DHJanzen:10-SRNP-112710 | *Agathodes designalis* | Costa Rica | ACG Estacion Cacao, Toma de Agua (habitat: rainforest) | BOLD | JN284590.1 |
| JQ567182.1 | DHJanzen:07-SRNP-104093 | *Agathodes designalis* | Costa Rica | ACG, Sector Santa Rosa, Luces (habitat: dry forest) | BOLD | JQ567182.1 |
| JQ567181.1 | DHJanzen:07-SRNP-104092 | *Agathodes designalis* | Costa Rica | ACG, Sector Santa Rosa, Luces, (habitat: dry forest) | BOLD | JQ567181.1 |
| JF846237.1 | DHJanzen:10-SRNP-112492 | *Agathodes designalis* | Costa Rica | ACG, Sector Cacao, Toma de Agua (habitat: dry-rainforest) | BOLD | JF846237.1 |
| JQ533042.1 | DHJanzen:10-SRNP-7372 | *Agathodes monstralis* | Costa Rica | ACG, Sector San Cristobal, Tajo Angeles (habitat: dry-rainforest) | BOLD | JQ533042.1 |
| JQ533041.1 | DHJanzen:10-SRNP-7373 | *Agathodes monstralis* | Costa Rica | ACG Sector San Cristobal, Tajo Angeles (habitat: dry-rainforest) | BOLD | JQ533041.1 |
| AS-12_LepF1 | MGCL-166648 | *Agathodes monstralis* | USA | Florida: Gainesville (habitat: upland pine) | MGCL | KU252850 |
| AS-13_LepF1 | MGCL-166647 | *Agathodes monstralis* | USA | Florida: Gainesville (habitat: upland pine) | MGCL | KU252851 |
| AS-14_LepF1 | MGCL-166646 | *Agathodes monstralis* | USA | Florida: Gainesville (habitat: upland pine) | MGCL | KU252852 |
| AS-15_LepF1 | MGCL-166645 | *Agathodes monstralis* | USA | Florida: Wocahoota rd, Micanopy (habitat: hardwood forest) | MGCL | KU252853 |
| GU665470.1 | DHJanzen:08-SRNP-1044 | *Agathodes monstralis* | Costa Rica | ACG, Sector San Cristobal, Sendero Huerta (habitat: rainforest) | BOLD | GU665470.1 |
| JQ569818.1 | DHJanzen:06-SRNP-103791 | *Agathodes monstralis* | Costa Rica | ACG, Sector San Cristobal, Rio Blanco Abajo, S. Cuatro Bocas (habitat: rainforest) | BOLD | JQ569818.1 |
| JQ569028.1 | DHJanzen:07-SRNP-106296 | *Agathodes monstralis* | Costa Rica | Rincon Rainforest, Estacion Caribe (habitat: rainforest) | BOLD | JQ569028.1 |
| JQ549430.1 | DHJanzen:98-SRNP-2446 | *Agathodes monstralis* | Costa Rica | ACG, Sector Cacao, Sendero Nayo (habitat: dry-rainforest) | BOLD | JQ549430.1 |
| JQ602023.1 | DHJanzen:06-SRNP-9179 | *Agathodes monstralis* | Costa Rica | ACG, Sector San Cristobal, Rio Blanco Abajo (habitat: rainforest) | BOLD | JQ602023.1 |
| JQ534728.1 | DHJanzen:06-SRNP-9180 | *Agathodes monstralis* | Costa Rica | ACG, Sector San Cristobal, Rio Blanco Abajo, E. Cuatro Bocas (habitat: rainforest) | BOLD | JQ534728.1 |
| JQ602022.1 | DHJanzen:06-SRNP-9182 | *Agathodes monstralis* | Costa Rica | ACG, Sector San Cristobal, Rio Blanco Abajo, E. Cuatro Bocas (habitat: rainforest) | BOLD | JQ602022.1 |
| JQ605506.1 | DHJanzen:03-SRNP-13116 | *Agathodes monstralis* | Costa Rica | ACG, Sector Santa Rosa, Laguna Escondida (habitat: dry forest) | BOLD | JQ605506.1 |
| JQ549429.1 | DHJanzen:98-SRNP-10886 | *Agathodes monstralis* | Costa Rica | ACG, Santa Rosa NP, (habitat: dry forest) | BOLD | JQ549429.1 |
| JQ549433.1 | DHJanzen:98-SRNP-10885 | *Agathodes monstralis* | Costa Rica | ACG, Santa Rosa NP, (habitat: dry forest) | BOLD | JQ549433.1 |
| JQ539644.1 | DHJanzen:05-SRNP-66393 | *Agathodes monstralis* | Costa Rica | ACG, Sector Mundo Nuevo, Porton Rivas, W. Rincon de la Vieja (habitat: dry-rainforest) | BOLD | JQ539644.1 |
| JQ539646.1 | DHJanzen:05-SRNP-66395 | *Agathodes monstralis* | Costa Rica | ACG, Sector Mundo Nuevo, Porton Rivas, W. Rincon de la Vieja (habitat: dry-rainforest) | BOLD | JQ539646.1 |
| JQ542540.1 | DHJanzen:06-SRNP-56090 | *Agathodes monstralis* | Costa Rica | ACG, Sector Mundo Nuevo, Porton Rivas, W. Rincon de la Vieja (habitat: dry-rainforest) | BOLD | JQ542540.1 |
| JQ540331.1 | DHJanzen:06-SRNP-56795 | *Agathodes monstralis* | Costa Rica | ACG, Sector Mundo Nuevo, Mamones, N. Castilla (habitat: dry forest) | BOLD | JQ540331.1 |
| JQ605505.1 | DHJanzen:07-SRNP-56487 | *Agathodes monstralis* | Costa Rica | ACG, Sector Mundo Nuevo, Quebrada Tibio Perla (habitat: dry forest) | BOLD | JQ605505.1 |
| JQ605693.1 | DHJanzen:02-SRNP-5492 | *Agathodes monstralis* | Costa Rica | ACG, Sector El Hacha, Finca Araya (habitat: dry forest) | BOLD | JQ605693.1 |

| **Specimen Figs** | **Voucher specimen #** | **Species name** | **Locality - Country** | **Locality-Additional** | **Sequencing facility** | **GenBank number** |
|---|---|---|---|---|---|---|
| JQ549432.1 | DHJanzen:00-SRNP-319 | *Agathodes monstralis* | Costa Rica | ACG, Sector San Cristobal, Cementerio Viejo (habitat: rainforest) | BOLD | JQ549432.1 |
| JQ549431.1 | DHJanzen:00-SRNP-320 | *Agathodes monstralis* | Costa Rica | ACG, Sector San Cristobal, Cementerio Viejo, S. Cuatro Bocas (habitat: rainforest) | BOLD | JQ549431.1 |
| GU699591.1 | DHJanzen:09-SRNP-105565 | *Agathodes monstralis* | Costa Rica | ACG, Sector Mundo Nuevo, Manta Cañon (habitat: dry forest) | BOLD | GU699591.1 |
| HM892327.1 | TDGAB-0797 | *Agathodes* sp. | Gabon, West Africa |  | BOLD | HM892327.1 |
| JQ564361.1\| | DHJanzen:07-SRNP-112267 | *Polygrammodes* nr. *multifenes* | Costa Rica | ACG, Sector Mundo Nuevo, La Perla (Tanque) (habitat: dry forest) | BOLD | JQ564361.1\| |
| AS-5_LepF1 | Na (destructive sampling) | *Terastia meticulosalis* | USA | Florida: Gainesville (habitat: upland pine) | MGCL | KU252845 |
| AS-22_LepF1 | MGCL-166652 | *Terastia meticulosalis* | Peru | Manu Park, Dept. Madre de Dios | MGCL | KU252855 |
| AS-1_LepF1 | MGCL-221335 | *Terastia meticulosalis* | Honduras | Atlantida: Pico Bonito (habitat: rainforest) | MGCL | KU252857 |
| AS-2_LepF1 | MGCL-221337 | *Terastia meticulosalis* | Honduras | Atlantida: Pico Bonito (habitat: rainforest) | MGCL | KU252858 |
| AS-3_LepF1 | MGCL-221338 | *Terastia meticulosalis* | Honduras | Atlantida: Pico Bonito (habitat: rainforest) | MGCL | KU252859 |
| AS-4_LepF1 | MGCL-221339 | *Terastia meticulosalis* | Honduras | Atlantida: Pico Bonito (habitat: rainforest) | MGCL | KU252860 |
| AS-9_LepF1 | MGCL-166649 | *Terastia meticulosalis* | USA | Florida: Gainesville (habitat: upland pine) | MGCL | KU252849 |
| AS-10_LepF1 | MGCL-166650 | *Terastia meticulosalis* | USA | Florida: Gainesville (habitat: upland pine) | MGCL | KU252861 |
| AS-11_LepF1 | MGCL-166651 | *Terastia meticulosalis* | USA | Florida: Gainesville(habitat: upland pine) | MGCL | KU252862 |
| JQ551585.11 | DHJanzen:98-SRNP-2446 | *Terastia meticulosalis* | Costa Rica | ACG, Estacion Botarrama, elev147m (habitat: rainforest) | BOLD | JQ551585.1 |
| JQ555016.1 | DHJanzen:07-SRNP-112964 | *Terastia meticulosalis* | Costa Rica | ACG, Sector Santa Rosa, elev. 251m (habitat: dry forest) | BOLD | JQ555016.1 |
| JQ546573.1 | DHJanzen:11-SRNP-103142 | *Terastia meticulosalis* | Costa Rica | ACG, Sector San Cristobal, elev. 575m (habitat: rainforest) | BOLD | JQ546573.1 |
| JQ571923.1 | DHJanzen:11-SRNP-103142 | *Terastia meticulosalis* | Costa Rica | ACG, Sector: Hotel Borinquen Canopy tours, 700m (habitat: dry-rainforest) | BOLD | JQ571923.1 |
| HQ935612.1 | DHJanzen:10-SRNP-108876 | *Terastia meticulosalis* | Costa Rica | ACG, Sector Pailas, elev. 1055m (habitat: dry-rainforest) | BOLD | HQ546573.1 |
| GU698546.1 | DHJanzen:09-SRNP-104632 | *Terastia meticulosalis* | Costa Rica | ACG, Sector Santa Elena, elev. 300m (habitat: dry forest) | BOLD | GU698546.1 |
| JQ558161.1 | DHJanzen:08-SRNP-102402 | *Terastia meticulosalis* | Costa Rica | ACG, Sector Rincon Rain Forest, Javalina elev. 288m (habitat: rainforest) | BOLD | JQ558161.1 |
| GU698650.1 | DHJanzen:09-SRNP-104521 | *Terastia meticulosalis* | Costa Rica | ACG, Sector Santa Elena, elev. 20m (habitat: dry forest) | BOLD | GU698650.1 |
| JQ566167.1 | DHJanzen:07-SRNP-102878 | *Terastia meticulosalis* | Costa Rica | ACG, Sector Trocha ICE, 850m (habitat: dry-rainforest) | BOLD | JQ566167.1 |
| HM903511 | BC ZSM Lep 19920 | *Terastia meticulosalis* | Ecuador | Pichincha prov., elev 2,310m | BOLD | HM903511 |
| BC-MTD-02351_Peru | BC MTD 02351 | *Terastia meticulosalis* | Peru | Huanuco, Lat.-9.614; long. -74.945 | BOLD | na |
| JF862298_AZ | CNCLEP00074996 | *Terastia meticulosalis* | USA | Arizona, Lat. 31.4468; long. -110.307 | BOLD | JF862298 |
| HQ960333_Cuba | BC MTD Lep 447 | *Terastia meticulosalis* | Cuba | Holguin, elev. 700m | BOLD | HQ960333 |
| AS-4-30-2013_LepF1_Vietnam | MGCL-166644 | *Terastia subjectalis* | Vietnam | Dakrong prov.: Dakrong | MGCL | KU252863 |
| JQ564361.1 | DHJanzen:07-SRNP-112267 | *Polygrammodes multifenestrata* | Costa Rica | ACG, Sector Mundo Nuevo, (habitat: dry forest) | BOLD | JQ564361.1 |
| JQ561457.1 | DHJanzen:07-SRNP-109185 | *Liopasia sp.* | Costa Rica | ACG, Alajuela, Rincon Rainforest (habitat: rainforest) | BOLD | JQ561457.1 |
| JQ561456.1 | DHJanzen:07-SRNP-109184 | *Liopasia sp.* | Costa Rica | ACG, Alajuela, Rincon Rainforest, (habitat: rainforest) | BOLD | JQ561456.1 |
| AS_166636_LepF1 | MGCL-166636 | *Terastia africana* **sp. n.** | Malawi | Mulanje Dist., Mulanje Mnts. Ruo Cyn.. elev. 900m | MGCL | KU252864 |
| AS_166637_LepF1 | MGCL-166637 | *Terastia africana* **sp. n.** | Malawi | Mulanje Dist., Mulanje Mnts. Ruo Cyn.. elev. 900m | MGCL | KU252865 |