

Wanted! Dead or alive: the tale of the Brown's Grayling (*Pseudochazara amymone*)

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Abstract The Brown's Grayling (*Pseudochazara amymone*) is one of the most enigmatic and sought after species among European butterflies. Hiding its exact distribution for almost 40 years with the idea of protecting it, resulted in an increasing collector's interest, with market prices reaching up to 1,000 euro for a single female after its discovery in Albania. Aiming to demystify this butterfly and enable entomologists and conservationists to see the species in its natural environment, we provide detailed information on its distribution in south-eastern Albania. In addition, we modelled the potential species distribution to facilitate further surveys within its potential range. The modelled range of *P. amymone* is highly fragmented stretching from the central part of eastern Albania to northern Greece and is strongly bound to ophiolite

geological strata. The species was re-assessed as Endangered according to the IUCN criteria, with a predicted population decline due to construction of hydroelectric power plants in one of the locations. We argue that hiding valuable information regarding threatened insect species may have negative effects and we advocate publishing available distribution data so that conservation measures may be undertaken where and when necessary.

Keywords Lepidoptera · Nymphalidae · Satyrinae · Threatened species · Maxent · Species distribution model

Introduction

There is rarely any butterfly species that has captured so much attention among European butterflies as Brown's Grayling—*Pseudochazara amymone* (Brown 1976). Ever since its discovery in northern Greece (Brown 1976) it has become a 'holy grail' for butterfly collectors throughout Europe. The main reason for that is a rather vague definition of the type locality: 'mountains just north of Ioannina, Epirus, Greece, 650 m' (Brown 1976) which resulted in many unsuccessful surveys to locate this newly described species (Cuvelier 2010). The species was then noted by Arnscheid and Arnscheid (1981) from the Ioannina region, but without any further details or comments. It was also mentioned as *P. mamurra* (Herrich-Schäffer 1852) for south-eastern Albania (Fig. 1) by Misja and Kurrizi (1984); however, these records could not be verified. Further data on the species were not available until the publication of the Butterflies of Greece (Pamperis 1997). In this monograph, *P. amymone* was reported from four sites in Epirus and Greek Macedonia. However, the mystery around the species continued as no information on localities has been

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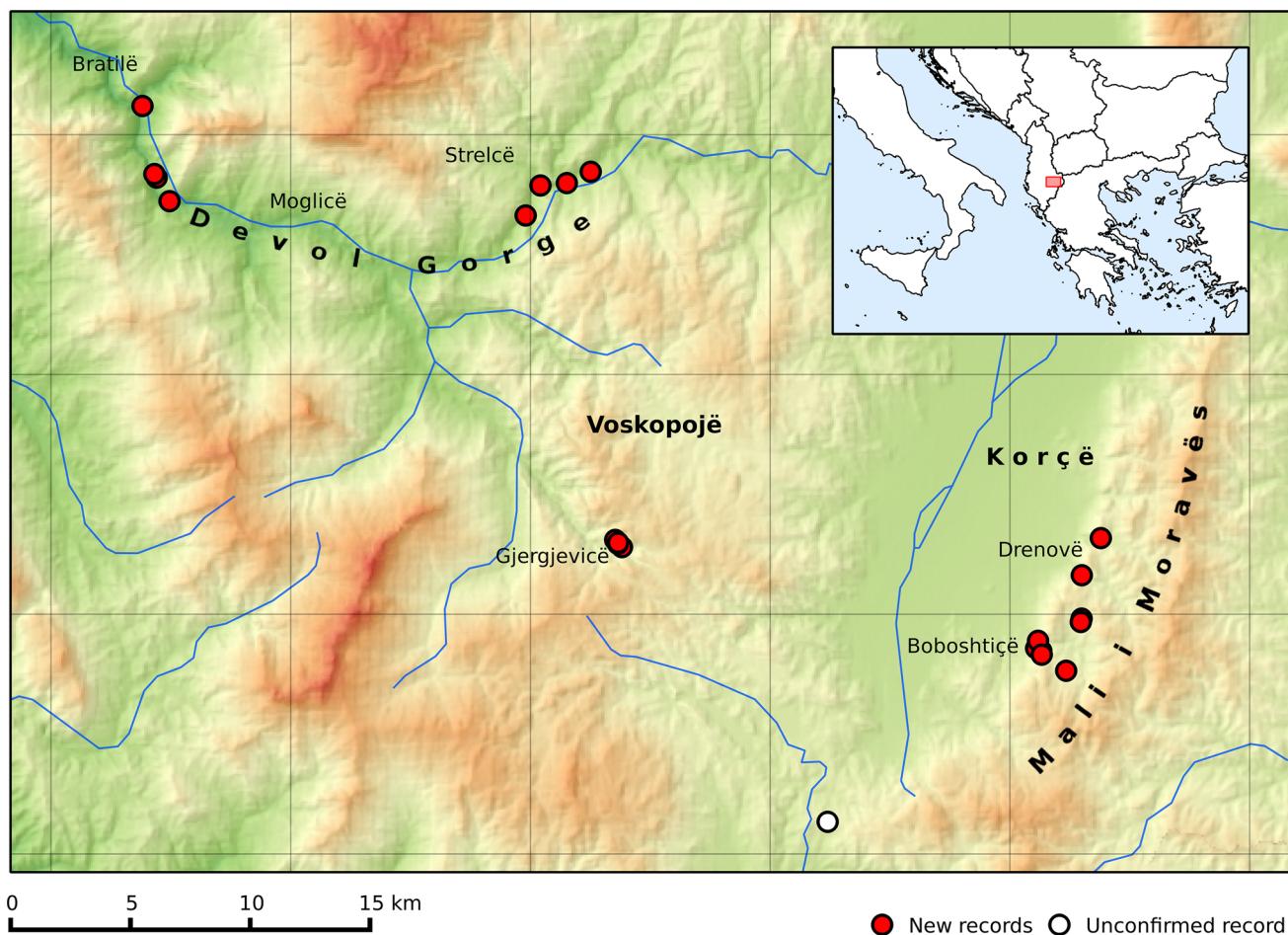


Fig. 1 Known distribution of the Brown's Grayling (*P. amymone*) in Albania shown on an UTM 10 × 10 km relief map with major rivers indicated. The locations are named in accordance with the distribution

section of this paper. New records from field surveys in SE Albania are shown along with a single unconfirmed record of the species from Misja and Kurzzi (1984)

revealed to protect the butterfly from collectors. In addition, a single photograph of a female leaves room for doubt of correct identification (Eckweiler 2012) and without available voucher specimens such records are hard to verify (Olivier 1999). In the second edition of the Butterflies of Greece (Pamperis 2009) the butterfly was recorded at 10 localities in the same region. Again all photographs depicted worn specimens with the female upper side photo giving the most reliable clue of the species presence in Greece (Eckweiler 2012). However, Cuvelier (2010) questions these photographs as pertaining to *P. amymone*. In the absence of additional information, the status of this taxon has been repeatedly questioned (Wakeham-Dawson and Dennis 2001; Kudrna et al. 2011). However, recent preliminary molecular studies show that *P. amymone* is closely related but genetically distinct from *P. mamurra* (Verovnik, unpublished data). In addition, the present European threat status of the species (i.e., Vulnerable) has been assessed solely on the basis of its presumed limited

distribution and potential habitat loss discerned from the absence of new records (van Swaay et al. 2011).

A breakthrough has been made by Eckweiler (2012) who found *P. amymone* in southern Albania. Again the information on exact locality was kept secret to protect this population and inspire other lepidopterists to find additional new localities. That, however, did not prevent commercial collectors from locating the species in 2013, possibly based on the habitat photo published by the author. During the summer of 2013, *P. amymone* was already on sale by several traders with high prices reaching up to 1,000 EUR for a single female (Insect Net 2013; Tarrier 2013; The Insect Collector 2013).

During our surveys in 2012 and 2013, we found *P. amymone* at several sites in south-eastern Albania. Here, we provide the first overview of its known range and assess its potential distribution using species distribution modelling techniques (Franklin 2010). We also discuss potential threats for the species and re-assess its threat status

according to the IUCN criteria (IUCN 2001). Through providing exact information on locations, we hope to demystify this poorly-known species and encourage entomologists to visit these sites and observe the species in its natural environment rather than trying to have the specimen in collections and thus supporting the insect dealers in earning profit on the species.

Materials and methods

Our first visit to Albania in 2012 formed part of the survey on the threat status of the Macedonian Grayling—*Pseudochazara cingovskii* (Gross 1973), as this species was listed for Albania without any further details (see Verovnik and Popović 2013; Verovnik et al. 2013 for details). During this visit, *P. amymone* was found at three different sites which triggered a more focused survey of its distribution in 2013. The survey was based mainly on the presence of potential suitable habitat identified using Google Earth satellite images searching for ophiolite rocks named after the brilliant green, snake-like serpentine minerals which are considered to be fragments of the oceanic lithosphere tectonically exposed on land by obduction during the geological history (Dilek and Newcomb 2003).

To predict the potential distribution of *P. amymone* in the southern Balkan Peninsula, we used Maxent species distribution models (Phillips et al. 2004, 2006) available from the dismo package in R. Maxent was chosen due to its capability of making powerful predictions with small sample sizes (Hernandez et al. 2006; Wisz et al. 2008). We based the model on *P. amymone* observations from south-eastern Albania, as no other precise data were available. Prior to analysis, duplicated points were removed from the data. As environmental parameters, a number of GIS layers covering potential butterfly distribution area were used: 1 km resolution WorldClim—BIOCLIM climatic parameters (Hijmans et al. 2005), 100 m resolution European Land Cover (EEA 2010), slope, slope aspect, and geology layers. The slope and slope aspect parameters were derived from 90 m Digital Elevation Data (Jarvis et al. 2008) in GRASS software, while aspect northness and eastness were derived from the aspect variable by simple calculation in R. The geological layer was produced in QGIS software, by digitizing 1:150,000 geological maps of Europe (Toloczyki et al. 1994). Possible multi-collinearity between numerical variables was tested using variance inflation factors (VIF) from the car package in R. The variables with the highest VIF score were removed in each cycle, leaving six out of 19 BIOCLIM parameters in the model (listed in Table 1). To check which environmental variables are valuable in explaining butterfly's distribution, we used percent contribution values and Maxent's jackknife analysis.

Table 1 Selected variables and their contributions to the Maxent distribution model to the of the Brown's Grayling (*P. amymone*)

Variable	Percent contribution
Geology	50.5
Land cover	18.6
Temperature seasonality (Bio4)	13.1
Mean temperature of wettest quarter (Bio8)	5.7
Precipitation of warmest quarter (Bio18)	5.2
Precipitation of wettest quarter (Bio16)	4.4
Aspect northness (AspectN)	1.2
Minimum temperature of coldest month (Bio6)	1
Isothermality (Bio3)	0.3
Slope	0.1
Aspect eastness (AspectE)	0

The names and derivation of the Bio3, Bio4, Bio6, Bio8, Bio16 and Bio18 variables can be found at <http://www.worldclim.org/bioclim>. For other variables please refer to “Materials and methods” section of this paper

Modelling is considered as an efficient tool for assessing species distributions in order to plan conservation actions for potentially threatened and less well studied species (Graham et al. 2004; Pearson 2007; Franklin 2010; Thomaes et al. 2008). However, in such cases it may be hard to obtain enough data and validate the model prediction. Recently, Pearson et al. (2007) proposed the jackknife validation of the model which is suitable for restricted occurrence data and provided simple software for calculating *P* values [supplemented in Pearson et al. (2007)]. The same authors also discussed using two threshold values, the lowest presence threshold (LPT) and fixed threshold of 0.1. This methodology is explicitly followed in constructing the *P. amymone* distribution model. Additionally, we evaluated the model using 53 data points of observed absence in geologically different substrate, which enabled us to calculate the area under the ROC curve (AUC) and true skill statistic (TSS) values (Phillips et al. 2006; Allouche et al. 2006).

Results

Distribution

During 2012, we found *P. amymone* at three sites in two separate areas: the Devoll Gorge and in two small valleys south of Boboshtiçë village near Korçë town (see Table 2, Fig. 1). At all three sites, only single specimens were observed along roads or watercourses in steep rocky gorges with sparse vegetation. In 2013, the western part of the Mali i Moravës mountain chain south of Korçë proved to

Table 2 List of the newly discovered Brown's Grayling (*P. amymone*) sites in south-eastern Albania

Site	Date	Latitude	Longitude	Altitude (m)
Korçë, Boboshticë, valley E of the village	11.7.2012	40°33"2'	20°46"40'	1,030
Korçë, Boboshticë, small gorge along the road SE of the village	11.7.2012	40°32"43'	20°46"46'	1,150
Korçë, Boboshticë, small gorge along the road SE of the village	20.7.2013	40°32"52'	20°46"36'	1,170
Korçë, Boboshticë, small gorge along the road SE of the village	16.7.2013	40°32"49'	20°46"45'	1,190
Korçë, Boboshticë, on the road to Dhardë	16.7.2013	40°32"22'	20°47"29'	1,220
Korçë, Drenovë, gorge SE of the village	21.7.2013	40°34"31'	20°47"57'	1,060
Korçë, Drenovë, gorge NE of the village	21.7.2013	40°35"21'	20°48"31'	1,100
Voskopojë, Gjergjivicë, NE of the village	18.7.2013	40°35"11'	20°34"13'	1,310
Voskopojë, Gjergjivicë, NE of the village	18.7.2013	40°35"7'	20°34"20'	1,220
Voskopojë, Gjergjivicë, NE of the village	18.7.2013	40°35"16'	20°34"9'	1,370
Voskopojë, Gjergjivicë, NE of the village	18.7.2013	40°35"13'	20°34"14'	1,330
Devoll Gorge, E of Strelcë village	10.7.2012	40°42"35'	20°31"27'	670
Devoll Gorge, E of Strelcë village	22.7.2013	40°43"15'	20°31"53'	700
Devoll Gorge, E of Strelcë village	22.7.2013	40°43"34'	20°33"22'	710
Devoll Gorge, E of Strelcë village	22.7.2013	40°43"19'	20°32"40'	700
Devoll Gorge, W of Moglicë village	22.7.2013	40°42"51'	20°20"53'	430
Devoll Gorge, W of Moglicë village	23.7.2013	40°43"22'	20°20"30'	450
Devoll Gorge, E of Bratilë village	23.7.2013	40°44"59'	20°20"2'	400

be one of the strongholds of the species with sightings in four different narrow valleys extending from the valley south-east of Drenovë village to the already known sites

near Boboshticë village. In some places, *P. amymone* was common, but still outnumbered by the much more abundant *P. mniszechii tisiphone* (Brown, 1980) with which it was observed syntopically. In Devoll Gorge, we found the species close to the site discovered in 2012 but also further down the valley, about 15 km to the west. Suitable habitat in the Devoll gorge is not contiguous and is separated by a long stretch of unsuitable habitat with different geology and plant communities. In the Devoll gorge, the only other species of the genus *Pseudochazara* was *P. anthelea* (Hübner, 1825), but only small numbers of specimens of both species were observed. Additionally, in 2013 *P. amymone* was found in a new region near Gjergjivicë village about 6 km south-west of Voskopojë town where it was again syntopic with *P. mniszechii tisiphone* and *P. anthelea*.

Altogether *P. amymone* has been found at 18 sites in three different locations covering a total of 13 2 × 2 km² (52 km²) squares (the grid cell size used by IUCN to calculate area of occupancy—AOO). Potential, but mainly inaccessible habitat is possibly much more extensive as our surveys were limited to the roads and more accessible slopes while major parts of the potential habitat were too steep to climb.

Distribution modelling indicated a larger potential range of the species stretching from the central part of eastern Albania to northern Greece where modelled suitable regions are much more fragmented (Fig. 2). It is interesting to note that no potential areas of distribution were detected near Ioannina—the putative type locality (Brown 1976). This may suggest that the type locality is further to the north than anticipated from Brown's inexplicit description of type locality or failure of the model to find potential habitat in that region. However, the unconfirmed record from Misja and Kurzzi (1984) was within the predicted distribution of the model. The jackknife model validation has shown high predictive ability. When a LPT threshold of 0.029 was used, the model success rate was 83 %, while a fixed threshold of 0.1 rendered a success rate of 72 % with high significance in both cases ($P < 0.0001$). The model reported very high AUC value of 0.989. The values of TSS score were 0.850 and 0.851 for LPT and a fixed threshold of 0.1 respectively. Among all environmental parameters, geology contributed 50 % of the resolution to the model, followed by Land Cover (19 %) and BIOCLIM parameters (30 %—Table 1). Maxent's jackknife analysis also showed that geology has the highest contribution to the model (Fig. 3).

Threats

Based on IUCN criteria (IUCN 2001), *P. amymone* should be considered Endangered under criteria B2ab(ii,iii,iv) as

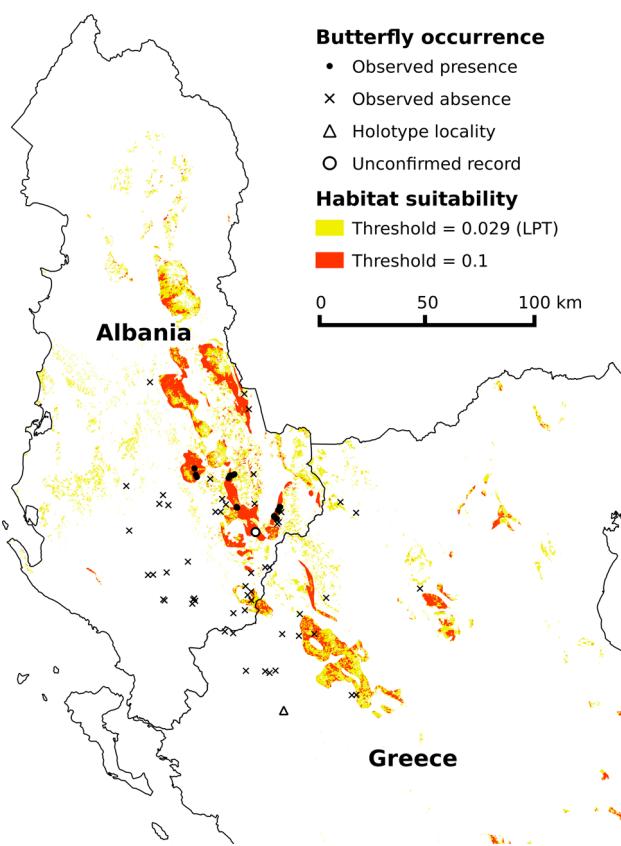
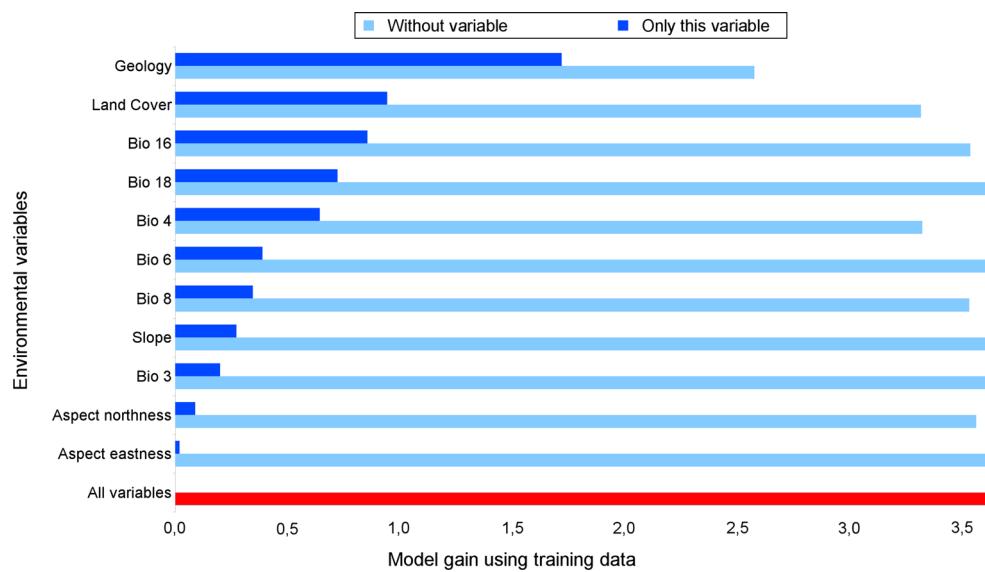


Fig. 2 Modelled distribution of the Brown's Grayling (*P. amymone*) in southern part of Albania and adjacent Greece. The observed presence and absence values are plotted on the map, along with single unconfirmed record from Misja and Kurzki (1984) and approximate location of the type locality

total area of occupancy does not exceed 500 km², it is known from no more than five locations, and there is a projected continuing decline of the species due to habitat loss. In Albania, the total area of occupancy is 52 km² while in Greece it is not known. Also, the number of confirmed locations is unknown in Greece, and due to the precautionary principle we could assume there is only one based on a single identifiable photo published (Pamperis 2009; Eckweiler 2012). Continuing decline is predicted due to habitat loss in the Devoll gorge region where a series of hydroelectric power plants will inundate large parts of the species habitat within the next decade (Statkraft 2013). This is at present the largest identified threat for the species as the initial work has already started and the project is mentioned among Stacrafts' company's major projects.

Commercial collecting is an additional general threat, although we consider it highly unlikely as a single cause of local extinction in any of the recently found *P. amymone* populations. That is mostly due to its inaccessible habitat on steep rocky terrain where adults are not easy to capture. Particularly the high-valued females are only rarely seen visiting flowers in areas accessible to collectors, while males commonly visit wet places along the roads and streams. However, the densities of *P. amymone* adults are lower than in other *Pseudochazara* species (e.g. Verovnik et al. 2013) which could make it more vulnerable for collecting in the long run. To prevent commercial collecting from damaging populations of *P. amymone*, collecting could be banned or regulated through its inclusion into the CITES appendix II.

Fig. 3 Jackknife test of variable importance for distribution model of the Brown's Grayling (*P. amymone*). The model gain is calculated for each variable alone, for each variable excluded and for all variables. A variable contains more useful information when it achieves high gain when used alone and when it lowers the gain when excluded. See Table 1 for explanation of the variable names



Discussion

Finding this enigmatic and emblematic species in three separate locations and 18 sites in south-eastern Albania gave us a unique opportunity to study its biology and potential distribution, but also a great responsibility as it could be considered as one of the most threatened endemic butterflies in Europe (van Swaay et al. 2011). Our surveys and the species distribution model suggested that the species occurrence is tightly linked to the geology of the area as *P. amymone* was always found on ophiolite geological strata in Albania. Geology is known to be an important factor limiting the distribution of several *Pseudochazara* species, in particular *P. cingovskii* and *P. orestes* De Prins and van der Poorten, 1981 which are present only on marble strata (Pamperis 2009; Verovnik et al. 2013). Owing to its chemical composition, ophiolites are characterised by a unique and scarce flora and fauna composition (Asenov and Pavlova 2009; Karataglis et al. 1982; Radford 1948). *P. amymone* is one of those species adapted to live in harsh environments, with its colour perfectly matching the colour of the brown-green rocks. It was not yet recorded far from this geological formation, thus, it would be interesting to test our hypothesis and provide additional data on the butterfly ecology.

The distribution model of *P. amymone* has shown great performance with only few available field observations, thus it can be considered a valuable tool for targeting further surveys (Pearson et al. 2007; Kumar and Stohlgren 2009). It should help to elucidate the situation in Greece which is highly important from a conservation point of view. Surveys north of the known *P. amymone* range in Albania could also provide additional records, and finding new distant locations may improve our model and enable more detailed model validation. Many variables used in the predictive model along with a small number of occurrence data may result in erroneous fit of the model, which is most likely shown as overprediction in this case (see Fig. 2). However, as Pearson et al. (2007) demonstrated, the Maxent approach can yield satisfactory model fits with as little as five observed occurrence data of a species. Predictions may be useful in the case of *P. amymone* helping us to identify potential areas where the species is present, target further field surveys for the species and for the planning of its protection.

Pseudochazara amymone is a prime example of how species should not be described and how damaging is to hide the exact position of the type locality. Although the latter has probably been done with good intentions it obviously had an opposite effect. Namely, the mystery around the species raised the interest of commercial collectors whose main goal is to collect and sell as many specimens as possible. Evidence of collecting, empty

envelopes, has been found in a small valley south of Boboshtiçë village during our surveys in 2013. It is also very likely that the type locality, known only to the describer, has already been destroyed in the past decades due to urban developments around Ioannina (Pamperis, comments to the authors). We will possibly never know how significant this loss is, as the overall situation in Greece remains unclear.

The reason to withhold information on the exact localities was to avoid collectors' pressure on this rare insect; however, there is not a single documented case of collectors entirely whipping out an insect species entirely by overkill (Thomas 1983). Making the information on species distributions unavailable for scientists and conservationists is therefore counterproductive and can have negative consequences, especially for species with limited geographic ranges. The main causes of extinctions worldwide are pollution, climate change, overexploitation of resources by the fast growing human population causing habitat loss and degradation, changes in agricultural practices, as well as synergistic effects of these threats (Purvis et al. 2000; Brook et al. 2008).

Collecting is an essential part not just of scientific studies, but also of faunistic surveys, as material can be checked subsequently if records are doubtful (Olivier 1999; Tarmann 2009). Even more importantly, correctly labelled material is indispensable for taxonomic or ecological studies and should be made available to the scientific community (GBIF, museums—Suarez and Tsutsui 2004). Here we come back to some collectors whose only interest is to possess a specimen even if unlabeled and commercial collectors earning money by collecting species of commercial interest. Such unlabeled material is useless for science while over-collecting is morally unacceptable and sheds bad light on entomologists in general. An additional side effect of commercial collecting is an inflation of named forms and subspecies which are sometimes not formally described and serve only to raise the value of the merchandise (Tennent 1999). The insect trade companies, which stand in between buyers and sellers, should therefore be more strictly controlled and should provide traceability of the material sold in order to prevent illegal collecting and loss of data.

Currently, *P. amymone* is not protected by any legislation and, therefore, formal steps to include it in national red lists and CITES appendix II need to be undertaken. With the information provided here, we hope to trigger further research on both the distribution and ecology of *P. amymone*. Additionally, we urge lepidopterists to publish their findings as every piece of information matters. This should enable more rigorous assessment of the threat status of *P. amymone* and hopefully provide enough evidence to start urgently needed conservation work for the species and its habitat.

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