
PHOTOGRAPHIC IDENTIFICATION OF *CNEMASPIS PSYCHEDELICA*: A USEFUL TOOL TO IMPROVE THE REGULATION OF INTERNATIONAL WILDLIFE TRADE

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Abstract.—Over-exploitation is considered as a major factor contributing to declines in wildlife populations. The recently described Psychedelic Rock Gecko (*Cnemaspis psychedelica*), which is endemic to two small islands in southern Vietnam, is an internationally protected species for which prescriptions of individual marking exist. *Cnemaspis psychedelica* is very small (5.82–7.57 cm snout-vent length) and may be too small for the use of transponders. No alternative permanent identification method has yet been found to be suitable for the species. In this study, we tested the intraspecific differences in the color pattern and the possibility of individual identification in *C. psychedelica* using the black and yellow reticulation pattern in the neck and occipital region of the body. We used photographic identification approaches by comparing images manually by eye and by applying two software applications. Our study showed that the life-color pattern of *C. psychedelica* is unique and stable for each adult individual for a period of at least 3 y. In addition to the manual photographic identification approach, the software application Wild ID was a useful tool for the image matching process and identification of individuals. This tool could assist authorities in wildlife conservation and should be commonly used in field research for a non-invasive individual identification.

Key Words.—CITES; conservation; lizards; non-invasive individual identification; Psychedelic Rock Gecko; Vietnam; Wild-ID

INTRODUCTION

The Psychedelic Rock Gecko (*Cnemaspis psychedelica*) was originally described from Hon Khoai Island, Ca Mau Province, in southern Vietnam (Grismer et al. 2010). Subsequently, Ngo et al. (2016) reported another population from Hon Tuong, a small isle in Rach Gia Bay, next to the type locality of the species. The population status of this species was first documented by Ngo et al. (2016) to be small overall but relatively stable and actively reproducing. Habitat destruction for infrastructure development on Hon Khoai Island and the introduced Long-tailed Macaques (*Macaca fascicularis*) were documented and could be increasing potential threats to the species (Ngo et al. 2016). As a result of recent research, *C. psychedelica* was assessed as Endangered (EN) in the International

Union for Conservation of Nature (IUCN) Red List of Threatened Species (Nguyen et al. 2016). Furthermore, the species is a desirable target in the international pet trade because of its extraordinary colorful appearance. Consequently, *C. psychedelica* specimens have been offered for sale online and at European reptile trade fairs for relatively high prices in recent years, shortly after the species description in 2010 (Auliya et al. 2016; Sandra Altherr et al., unpubl. report). The need to regulate the international trade in the species led to its inclusion in Appendix I of the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES) in 2016 and in Annex A of the European Union (EU) Council Regulation No. 388/97. In terms of national legislation, the species was listed as a protected species in Vietnam under governmental decrees No. 06/2019/ND-CP and No. 64/2019/ND-CP. Thus, domestic and

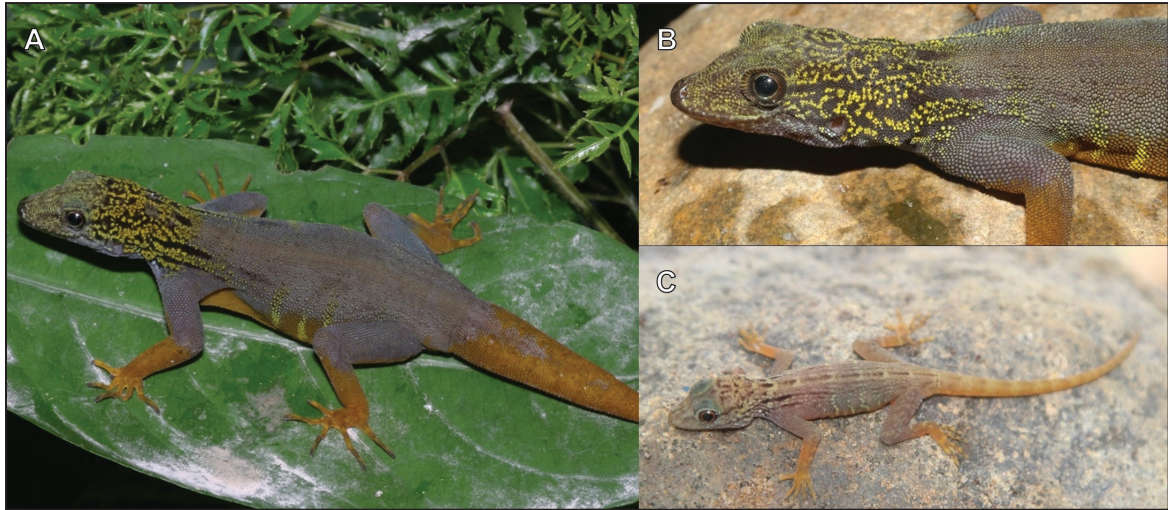


FIGURE 1. (A) General appearance of the Psychedelic Rock Gecko (*Cnemaspis psychedelica*) from Vietnam (Photographed by Truong Q. Nguyen). (B) Apart from the orange extremities, the black and yellow reticulation pattern in the neck and occipital region is the most remarkable characteristic of this gecko species (Photographed by Khoi V. Nguyen). (C) Shift in pattern and coloration in juveniles occurs at about six weeks of age (Photographed by Khoi V. Nguyen).

international trade in wild specimens of *C. psychedelica* for commercial purposes is prohibited. Recently, captive breeding in the species outside of its range has been reported, which can relieve the demand on wild animals for legal trade (Langner et al. 2020). The availability of geckos from captive breeding, however, may be insufficient to meet the demand for the international trade. Indeed, Ngo et al. (2018) documented the continued occurrence of wild captured specimens in trade at national and international markets, after the CITES listing had already taken effect. Therefore, the differentiation between legal and illegal specimens in trade poses a great challenge to responsible authorities.

To better control and trace specimens in trade and mitigate illegal trade in threatened species, such as *C. psychedelica*, a unique and permanent marking of the specimens in trade is useful. According to CITES Res. Conf. 12.10 (Rev. CoP15), specimens of Appendix I species bred in registered captive breeding operations have to be individually marked. With respect to EU law, the individual marking of all specimens of Annex A species is obligatory for the purpose of imports and commercial activities within the EU, based on Article 11 Paragraph 3 and Article 66 of the EU Commission Regulation (EC) No. 865/2006. While certain marking methods have been proposed for several taxa (such as closed rings for many birds, transponders for different vertebrates, photographic documentation for tortoises), no marking method has been officially recommended or identified to be suitable for *C. psychedelica*.

There are various techniques for identifying individual lizards such as toe-clipping, branding, tattooing, subcutaneous elastomer injections, and Passive Integrated Transponder (PIT)-tags (Sacchi

et al. 2010). Nevertheless, these techniques are not always practical because they are relatively expensive, ineffective, or have a potential negative impact on the welfare of the individual (Drechsler et al. 2015; Oscar et al. 2015; Matthé et al. 2017; Röhl 2018). Recently, identification approaches based on natural color and marking patterns of animals have been increasingly used to distinguish among individuals as a non-invasive technique to overcome the limitations of the other methods (e.g., Arzoumanian et al. 2005; van Tienhoven et al. 2007; Gamble et al. 2008). This method has been applied to several lizard species (e.g., Sacchi et al. 2010; Oscar et al. 2015; van Schingen and Böhmer 2018), including a gekkonid species (Röhl 2018). A requirement of photographic identification is that each individual possesses a unique body pattern, which is robust and stable over its entire lifespan without any substantial changes (Drechsler et al. 2015).

According to Grismer et al. (2010), *C. psychedelica* is distinguished from all other *Cnemaspis* species by having a unique color pattern; the detailed black and yellow reticulation pattern in the region of the neck and head (Fig. 1). We hypothesized that this colorful characteristic is stable throughout the entire lifespan (at least 6–7 y; unpubl. data) once individuals have reached maturity and that it is individually distinguishable. To test this hypothesis, we assessed captive specimens from a *Cnemaspis psychedelica* conservation breeding program that has been established on the grounds of the center of Wildlife at Risk (WAR) in southern Vietnam since 2015. This center was established to ensure the survival of the species also through *ex situ* measures (Ziegler et al. 2016). We predict that photographic identification can be used to identify individual lizards.

Here, we present the first study testing the suitability of the natural color pattern of *C. psychedelica* for photographic identification and also propose software that turned out useful for this purpose.

MATERIALS AND METHODS

Color pattern of *Cnemaspis psychedelica*.—The ground color of the dorsum of *C. psychedelica* is blue-grey to light purple and the venter is beige. Apart from the orange forelimbs, forelegs, hands, feet, and tail, the yellow reticulation in the neck and occipital region overlaid by black stripes is especially conspicuous. Additionally, the trunk bears four transverse yellow bars laterally (Grismer et al. 2010; Fig. 1). *Cnemaspis psychedelica* exhibits an ontogenetic pattern and color change: after hatching, juveniles show a pattern of dark longitudinal stripes and light transverse bars on a light purple to light grey body coloration. The tail is pale orange and transversely banded. The shift in pattern and coloration has been observed to start around five weeks after hatching and is far advanced in juveniles at 3 mo of age (Ziegler et al. 2016; Fig. 1). There are no differences in the color pattern between sexes (Grismer et al. 2010; Hai N. Ngo et al., pers. obs.). For intra-specific identification, we considered that the black and yellow reticulation pattern in the neck and occipital region would be best for identification because of the detailed pattern and the assumption that this pattern is unique in its characteristics for each specimen, despite the general similarity.

Recording and comparison of photographs.—We selected 12 adult individual *C. psychedelica*, kept in the WAR center in Binh Duong Province, southern Vietnam. Since the establishment of the breeding program in 2015, WAR center houses the largest known legal captive colony of the species, providing the opportunity to document the individual development over time. We took photographs of each individual in June and October 2017, May 2019, and July 2020. In 2019 and 2020 photographs were not available for all individuals that we initially included. To achieve the best possible shooting conditions and good photograph quality, we caught specimens by hand, photographed them outside of their enclosures, and released them immediately at their enclosure after photographing. We took images of the color pattern from above the head and laterally from both sides of the head (Fig. 1). We used a Canon Eos 10D mirror reflex camera (Canon, Ota City, Tokyo, Japan) and a Sony DSC-HX90 digital camera (Sony Corporation, Minato, Tokyo, Japan) to photograph lizards.

To determine the uniqueness and possibility of stability over time of the natural pattern of each

individual, we initially compared images visually. Because manual photographic identification is time-consuming and error-prone when dealing with large datasets, software has been developed and is commonly used to identify the stability of natural patterns of animals (e.g., Arzoumanian et al. 2005; Bolger et al. 2012; Oscar et al. 2015). Software algorithms were originally developed for one specific pattern of a certain species. Thus, it is recommended to compare different available algorithms to choose the most reliable version for the species of interest (Matthé et al. 2017). Therefore, we compared images with two freely available semi-automatic software applications after the manual visual analysis: APHIS (Oscar et al. 2015) and Wild ID (Bolger et al. 2012). APHIS was originally developed for a lizard and a salamander species, while Wild ID was developed for the pattern of giraffes. The preparation time for the use of these software applications is in both cases limited. For APHIS, images can be inserted without any pre-processing. For Wild ID, the user has to select the region of interest with the distinctive pattern and crop out other distracting elements.

In general, both software applications work in the same way. First, a database is created by gathering one image of each individual; in this case we used images from June 2017, which served as a reference for subsequent images. By running images against the database, a correlation score is calculated for each image, which is presented in a final ranking showing the most likely matching specimens. Finally, the match is manually checked by eye to confirm the result of the software identification. APHIS provides two procedures, and we used the pixel-based approach (Image Template Matching, ITM). Wild ID is fixed as a pixel-based algorithm as well. For detailed descriptions of the image matching code of the algorithm see Bolger et al. (2012) for Wild ID and Oscar et al. (2015) for APHIS. To assess the accuracy and suitability of a photographic algorithm, recognition rates should be calculated for known matching images of individuals (Matthé et al. 2017). The False Recognition Rate (FRR) is calculated as the ratio between the number of wrong matches and the number of total processed images. We considered a match as wrong if a true matching image was placed below a false matching image.

RESULTS

Our comparison of the images from June and October 2017 by eye revealed that all 12 individuals could be distinguished from each other on the basis of variation of the black and yellow reticulation pattern on the neck and occipital region. These differences were visible in pattern as viewed from the left and right side (lateral), and from above (dorsal), respectively (see Fig. 2, 3). Our

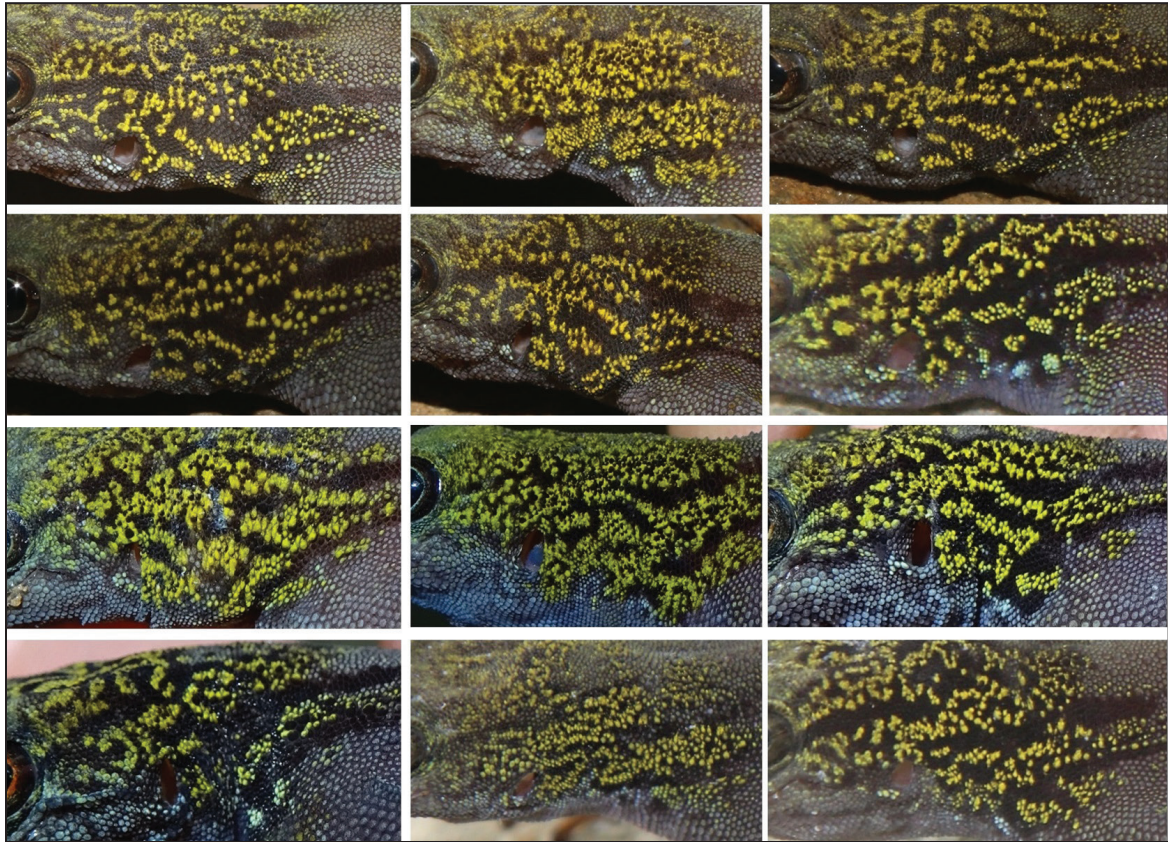


FIGURE 2. Variation in the lateral color pattern of the Psychedelic Rock Gecko (*Cnemaspis psychedelica*). (Photographed by Khoi V. Nguyen, Truong Q. Nguyen, and Thomas Ziegler).

manual comparison of the individual images from 2019 and 2020 with those of 2017 confirmed the stability of these individual patterns. Thus, we selected the pattern on the neck and occipital region of *C. psychedelica* to identify the temporal stability and the accuracy of the two software applications.

The software application Wild ID matched all lateral images correctly, except for one error after the addition of images from 2020 (FRR = 0–0.044; Table 1, exemplary matching process in Fig. 4). We found that most dorsal images were correctly matched as well, but a few errors occurred (FRR = 0–0.08; Table 1). In contrast, the application APHIS showed higher FRR values compared to Wild ID with increasing error rates especially after the addition of the images from 2019 and 2020. Fewer than every second dorsal image was

matched correctly (FRR = 0.58–0.72; Table 1). APHIS was less error-prone when comparing lateral images, but nevertheless error rates increased after including images from 2019 and 2020 (FRR = 0–0.40; Table 1).

DISCUSSION

Our study confirms that intra-specific difference of *C. psychedelica* based on the uniqueness of the black and yellow reticulation pattern on the neck and occipital region of each specimen, could be manually determined by eye and is considered suitable for individual identification using photographic methods. The stability of the pattern over the lifetime of an individual *C. psychedelica* has not yet been confirmed though, but we confirmed identification of individuals for 3 y and in

TABLE 1. Summary of calculated False Rejection Rates for the Psychedelic Rock Gecko (*Cnemaspis psychedelica*) using two software applications, APHIS and Wild ID.

Software/View Point	APHIS			Wild ID		
	2017	2019	2020	2017	2019	2020
Dorsal	0.583	0.700	0.722	0.083	0.050	0
Lateral (right side)	0	0.125	0.400	0	0	0
Lateral (left side)	0.083	0.111	0.130	0	0	0.044

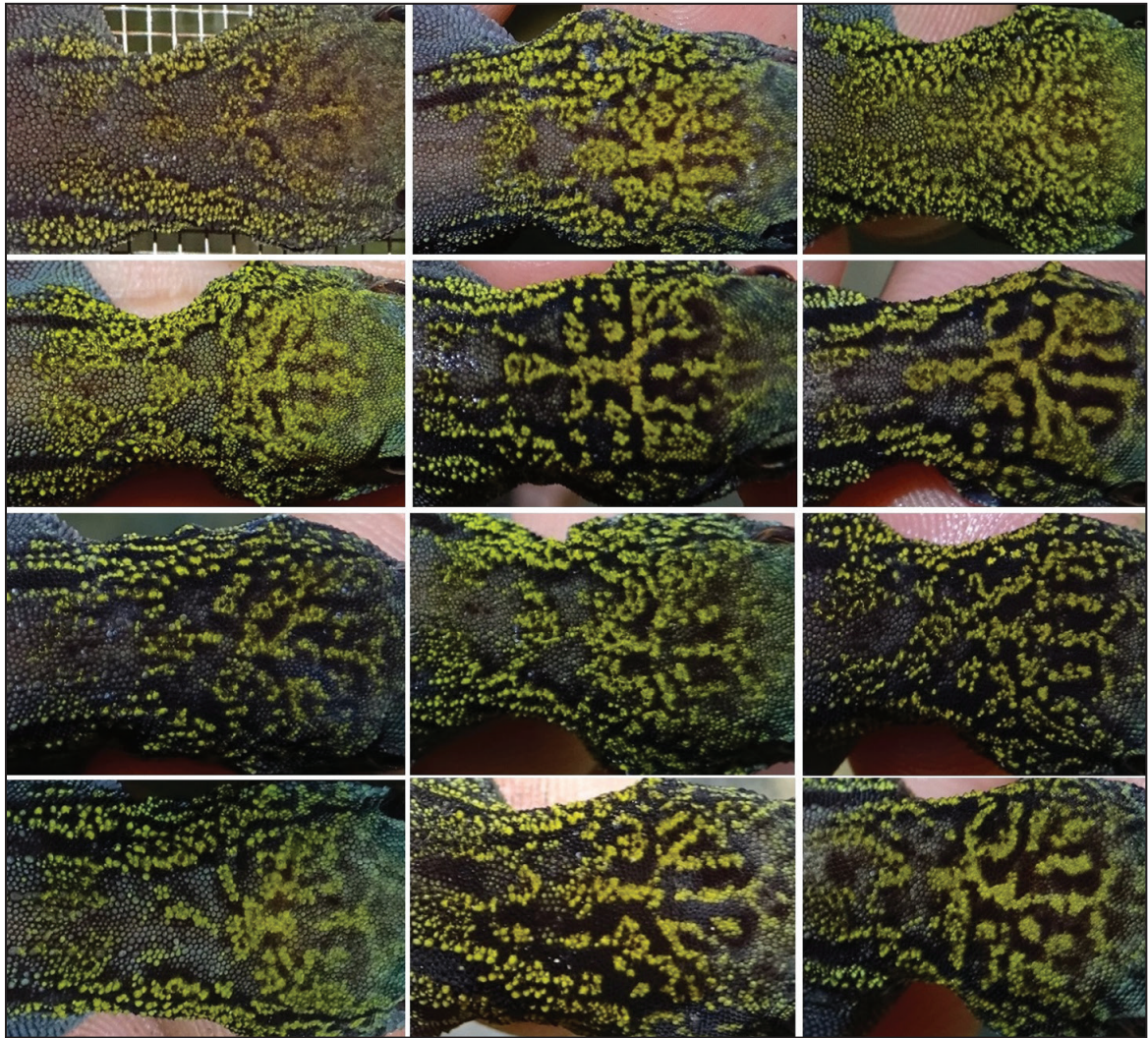


FIGURE 3. Variation in the dorsal color pattern of the Psychedelic Rock Gecko (*Cnemaspis psychedelica*). (Photographed by Khoi V. Nguyen, Truong Q. Nguyen, and Thomas Ziegler).

one case, almost 5 y (Fig. 5). Our data only shows the stability of the color pattern and individual identification for adults. We only documented the development of the pattern in juveniles in a few cases and we have not demonstrated the stability of color pattern. Therefore, further breeding-success in captivity would be required for a more detailed documentation of the ontogenetic color and pattern change process duration to determine minimum age and/or body size for the photographic identification method.

Our use of semi-automatic computer applications revealed that the Wild ID software is suitable to distinguish individual *C. psychedelica* both for lateral and dorsal patterns. The observed error rates were relatively low and comparable to other photographic identification studies using computer-assisted applications (mean FRR = 0.122; range, 0.007–0.250; Bolger et al. 2012). Large data sets are usually used to determine reliability of color patterns for individual identification and the

validity of our study thereby might be limited. Blurring and differences in perspective of our photographs likely led to some errors in identification. Problems in recognition due to differences in the quality of images are common, which is why standardized recording of high-quality photographs is advisable (Bendik et al. 2013). The application APHIS turned out to be rather unsuitable, which confirmed the observation that APHIS is sensitive to differences in exposure and perspective as well as body torsions (Oscar et al. 2015). Based on limited available data, the comparison of color patterns manually by eye without any software appears to be more reliable and time efficient. In particular, the human eye and mind is more sensitive in terms of differences in the quality, illumination, and perspective of images.

We recommend photographic documentation as a reliable marking method for *Cnemaspis psychedelica*, taking images from both lateral and dorsal perspectives of the neck. Based on this combination of photographs

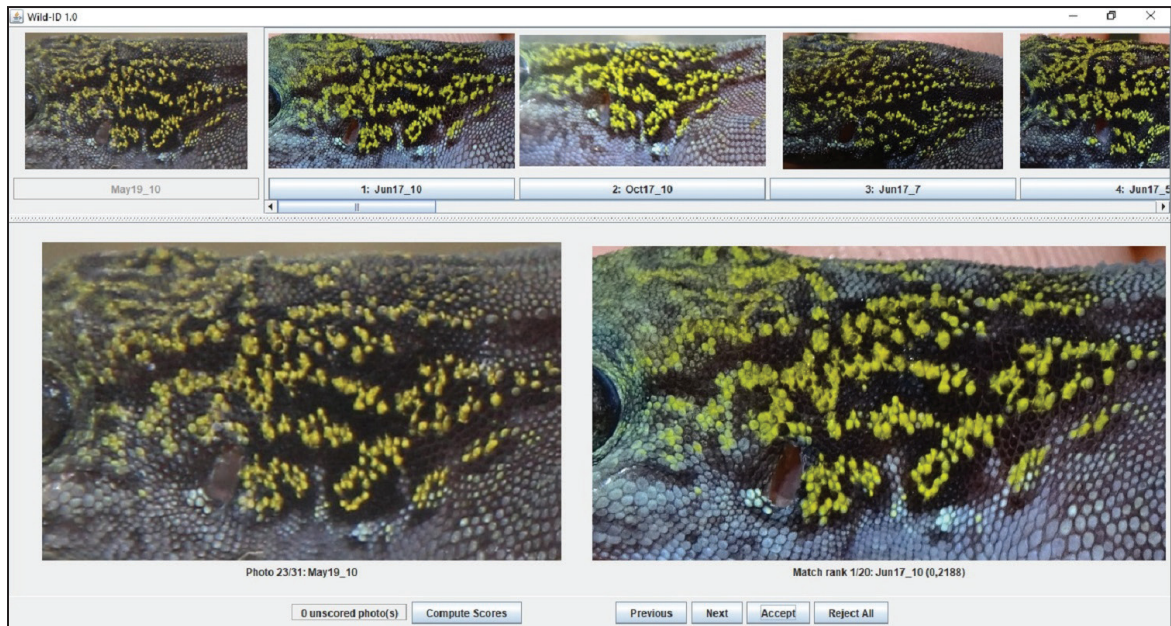


FIGURE 4. Example for a matching procedure of two images taken from a lateral view of the same individual of the Psychedelic Rock Gecko (*Cnemaspis psychedelica*) with the software application Wild ID.

using Wild ID software and visual inspection, an individual should be unambiguously recognizable on the basis of the black and yellow reticulation pattern. Photographic identification could be used as non invasive approach as well for mark-recapture studies to estimate population densities and sizes in the field instead of marking specimens. Ngo et al. (2016) evaluated the population status of *C. psychedelica* in its natural habitats by marking each captured individual with a permanent marking pen, but this marking technique does not last long and therefore is not suitable for long-term population assessments. Although PIT tags are often used in lizards for permanent identification, they

apparently cannot be used for *C. psychedelica* because individuals have low weights (0.70–13.6 g). According to German law (Verordnung zum Schutz wild lebender Tier- und Pflanzenarten, BArtSchV) 200 g is the minimum weight for reptiles (except turtles) to be marked with a transponder (Ngo et al. 2018; Röhl 2018; van Schingen and Böhmer 2019). Thus, photographic identification as a comprehensive mark-recapture approach can be applied for long-term monitoring activities to safeguard wild populations of *C. psychedelica* in its natural range. We recommend establishing a central database of images of wild and captive animals to serve as reference database for authorities in wildlife conservation.

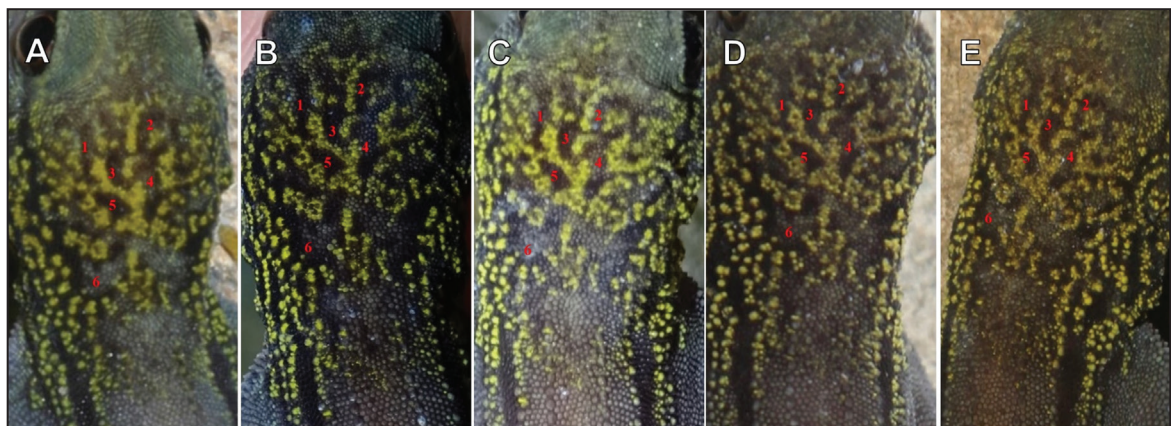


FIGURE 5. Series of the dorsal color pattern of the same individual of Psychedelic Rock Gecko (*Cnemaspis psychedelica*) over 5 y. The stability of the pattern is clearly visible despite slight divergences in illumination and recording angle of the images. Red numbers indicate noteworthy spots to simplify the identification. (A) November 2015 (Photographed by Thomas Ziegler). (B) June 2017 (Photographed by Thomas Ziegler). (C) October 2017 (Photographed by Thomas Ziegler). (D) May 2019 (Photographed by Truong Q. Nguyen). (E) July 2020 (Photographed by Khoi V. Nguyen).

Acknowledgments.—We thank the authorities of the Hon Khoai Forest Protection Unit, Forest Protection Department of Ca Mau Province, Vietnam. Collecting for the national *ex situ* program was based on the national collection permit issued by Hon Khoai Forest Protection Unit from 11 March 2015. We are grateful to Theo Pagel and Christopher Landsberg (Cologne Zoo), Sinh Van Nguyen (IEBR, Hanoi), for their support of conservation-based biodiversity research in Vietnam. This research was supported by the Vietnam Academy of Science and Technology (Project Code KHCBS.02/20–22) and partially funded by Cologne Zoo. Cologne Zoo is partner of the World Association of Zoos and Aquariums (WAZA): Conservation Project 07011, 07012 (Herpetodiversity Research, Amphibian and Reptilian Breeding and Rescue Stations). Research of Hai Ngoc Ngo in Germany is funded by the German Academic Exchange Service (DAAD).

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Herpetological Conservation and Biology



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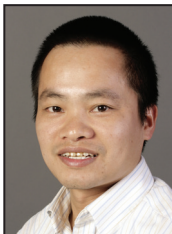
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