A pattern-based tool for long-term, large-sample capture-mark-recapture studies of fire salamanders *Salamandra* species (Amphibia: Urodela: Salamandridae)

Jeroen Speybroeck[1,*], Koen Steenhoudt[2,$]

[1] *Research Institute for Nature and Forest, Kliniekstraat 27, 1070 Brussels, Belgium. *Corresponding author. E-mail:* jeroenspeybroeck@inbo.be, jeroen.speybroeck@hotmail.com
[2] *Waterschaapstraat 30, 1570 Galmaarden, Belgium*
[$] Programming and database development was solely done by K.S.

Abstract. Solid population studies depend on reliable capture-mark-recapture (CMR) methodology. The available methods for such studies on amphibians are often invasive, unsuitable for long-term studies, time-consuming and/or expensive. We present a new software tool, connected to a MS Access database, ManderMatcher, for CMR study of fire salamanders (*Salamandra salamandra* and related species) by means of a robust matching algorithm using 44 pattern characteristics. Metadata related to standardised counts (e.g., weather and lunar variables) as well as a myriad of individual sighting variables can be entered and saved as well. Application of the presented method to a database of 9,397 sighting records gathered over a period of eight years, as well as a random sample validation, demonstrate the accuracy of the applied matching algorithm. Differences with other methods are discussed. The program is made freely available for download and widespread application is encouraged, especially given the contemporary context of a fungal disease threatening survival of fire salamander populations.

Keywords. *Salamandra salamandra*, pattern recognition, capture-mark-recapture, software, methodology, free download

INTRODUCTION

Long-term demographic studies depend on reliable capture-mark-recapture (CMR) methodology to model population size and abundance, survival and detection rate, as well as to provide data on a myriad of life history features such as individual growth, reproductive cycles, longevity, dispersal and migration (e.g. Ferner, 1979; Schmidt et al., 2002; Schmidt, 2004; Amstrup et al., 2005). A number of methods are available for practical execution of such studies in amphibians. Toe clipping as well as the use of tattoos or passive or active transponders, however, all require a certain degree of (at best tem-

porary) damage to the subject's body, and are thus commonly labelled as invasive and, at least potentially, damaging (e.g. Davis and Ovaska, 2001; Le Galliard et al., 2011). Furthermore, given the regenerative capacity of certain taxa, namely salamanders, toe clipping becomes unsuitable for long-term studies (e.g., Ferner, 1979; Davis and Ovaska, 2001) and may in certain cases even reduce recapture and survival rates (McCarthy et al., 2009). Use of transponders sets additional limitations such as being costly and fairly time-consuming in the field (Ott and Scott, 1999), as well as in some cases having low retention rates in smaller individuals and species (Ryan et al., 2014), while detection and recapture rates may differ

according to the size of the used tags (Ousterhout and Semlitsch, 2014).

Species with an individually varying pattern of colour spots offer the possibility of recognising animals without the need to apply invasive techniques. A number of (more or less) automated image analysis tools has been developed, such as WildID (Bolger et al., 2012), AmphIdent (Drechsler et al., 2015), I³S (Van Tienhoven et al., 2007, with newer, improved versions having been successfully applied to the newt *Triturus carnifex* by Sannolo et al., 2016, as well as to reptiles, as reviewed by Sacchi et al., 2016), APHIS (Moya et al., 2015) etc. While these claim to offer more robust recognition procedures and automatic pattern recognition, they usually require labour-intensive image processing steps in order to select a certain 'area of interest'. Thus, they do not necessarily guarantee faster processing than alternatives based on interpretative pattern coding. One (AmphIdent) is even fairly expensive, especially for non-institutional users. Additionally, while pattern shape becomes of greater importance, spot shapes may alter drastically given the position of the animal within the picture. As such, higher demands are set on the quality of the pictures, resulting in more time spent properly positioning live and fairly hard to immobilise animals while collecting data.

The use of non-invasive recognition techniques to study fire salamander *Salamandra salamandra* evolved throughout the last decades, making use of the typical pattern of yellow spots on a black background in this species. Earlier authors used time-consuming, direct comparison of photographs to recognise individuals (e.g., Feldmann, 1971; Klewen, 1985; Seifert, 1991). Kopp-Hamberger (1998) coded different *S. salamandra* pattern types into an alphanumerical code, while not ruling out occurrence of type doubles, thus causing obvious problems in larger samples. Carafa and Biondi (2004) elaborated on this by combining a colour typology with a software application storing data into an MS Access database for the Italian subspecies *S. s. gigliolii*, using a sample of 233 individual animals. Given the limited description of the applied characteristics in their paper and the fact that the software was not made freely available, use of and detailed insight into this methodology remained unavailable to a wider audience. A different approach was presented by Šukalo et al. (2013), using the number of glandular pores located within the boundaries of the yellow spots. In many subspecies of *S. salamandra*, however, the number, size and shape of these spots is known to change throughout an individual's lifetime (Eiselt, 1958; Klewen, 1991; Mutz, 1992; Bogaerts, 2002). Becoming stable at adulthood only (Klewen, 1991), pattern changes may be common, limiting indeed the use of colour patterns for

individual recognition at earlier life stages. While the degree of colour pattern alteration during early terrestrial life may vary considerably between *S. salamandra* subspecies (Bogaerts, 2002), a relatively stable pattern is said to be reached after at least two or three years (Eiselt, 1958). This leads to changes in pore count within the span of a single year in some individuals, even in animals larger than 12 cm (Speybroeck, unpublished data). Thus, the method of Šukalo et al. (2013) seems of limited value for studies spanning more than a couple of months at most, and a methodology combining a multitude of characteristics seems preferable.

As such, use of an individual-specific code which can be applied to easily collected and processed images, which is sufficiently stable over time and can be queried by an automated matching algorithm applied to a database, is of merit. We present a new software tool, connected to a MS Access database, ManderMatcher, for CMR study of *Salamandra salamandra* and other spotted species of the genus by means of a robust matching algorithm using 44 pattern characteristics. We demonstrate how this tool can serve study of large sample sizes involving a high number of different individuals.

MATERIAL AND METHODS

Salamandra salamandra and related spotted species (*S. algira, S. corsica* and *S. infraimmaculata*) are characterised by a black background colour covered with a variable amount of most commonly yellow (but in certain taxa rather orange or red) spots or stripes (Thiesmeier, 2004; Beukema et al., 2016b; Speybroeck et al., 2016). The top of the head is usually covered with yellow on the eyes and parotoid glands, while additional spots may be present on the snout tip. The tail may be crossed by spots as well. Toes may be black or yellow. This pattern layout offers potential for coding each animal's pattern into a series of numbers. These numbers can be supplied to an algorithm that matches each pattern with pattern codes from previously collected data.

Pattern code

The spot pattern is coded into 44 characteristics. Each of those indicates the number, presence or mutual relation between the yellow spots within well-defined areas of the salamander body: head (11 characteristics), back and tail (19) and toes (14). In case of doubt, characteristics can be left blank and will not be taken into account by the matching algorithm. As patterns may vary over time, reliable use is advised for animals with a total length of more than 14 cm only. Matching may work on smaller animals, depending on the stability of that animal's pattern as well as the portion of characteristics that are left blank by the user. Thus, while no reliable assessment of subadult and juvenile population size can be made with this

(or any) pattern-based method, it may allow certain other studies which do not require tracing every single individual (e.g. growth analysis).

In contrast to criteria used in some of the other cited methods, each of the 44 characteristics has a clear definition, allowing a limited array of entered values only and leaving little room for variation caused by interpretation. All are discussed and illustrated in the manual of the freely available program, serving as supplementary material to this paper (available for download, along with the program itself, at http://www.hylawerkgroep.be/jeroen/index.php?id=85). For demonstration to new users, a version containing already some data is also provided. The ManderMatcher website also offers data exploration tools for data stored in the program's database).

Program

A software tool, ManderMatcher, was developed to allow CMR recognition and storage of other visit and sighting related data. The program was written in the VB.NET (Visual Basic) language, using the freely available basic version of Visual Studio Express. Practical use is discussed at length in the manual (see above link). The program basically consists of three main windows.

The first is the 'Visits' window, available after launch as the left tab (Fig. 1). It lists the visits and allows to enter metadata related to that visit. Visits are time-restricted count events, covering e.g. a standardised transect or habitat surface.

The second window is the 'Sightings' window (Fig. 2), available as the second tab, next to the Visits window. The 'pivot animal', shown at the upper left, will be matched with the entire database of available sightings at the right on the basis of the pattern code that was entered for it. At the lower left, criteria can be specified to filter and restrict the database entries which are returned at the lower right. The as such selected entries are sorted by their resemblance to the pivot animal, with the most similar entry at the top.

The third window is accessed through the buttons for editing one of the two sightings shown at either the upper left or

Fig. 1. 'Visits' window for edit and creation of visits, and entry of environmental data in the program ManderMatcher.
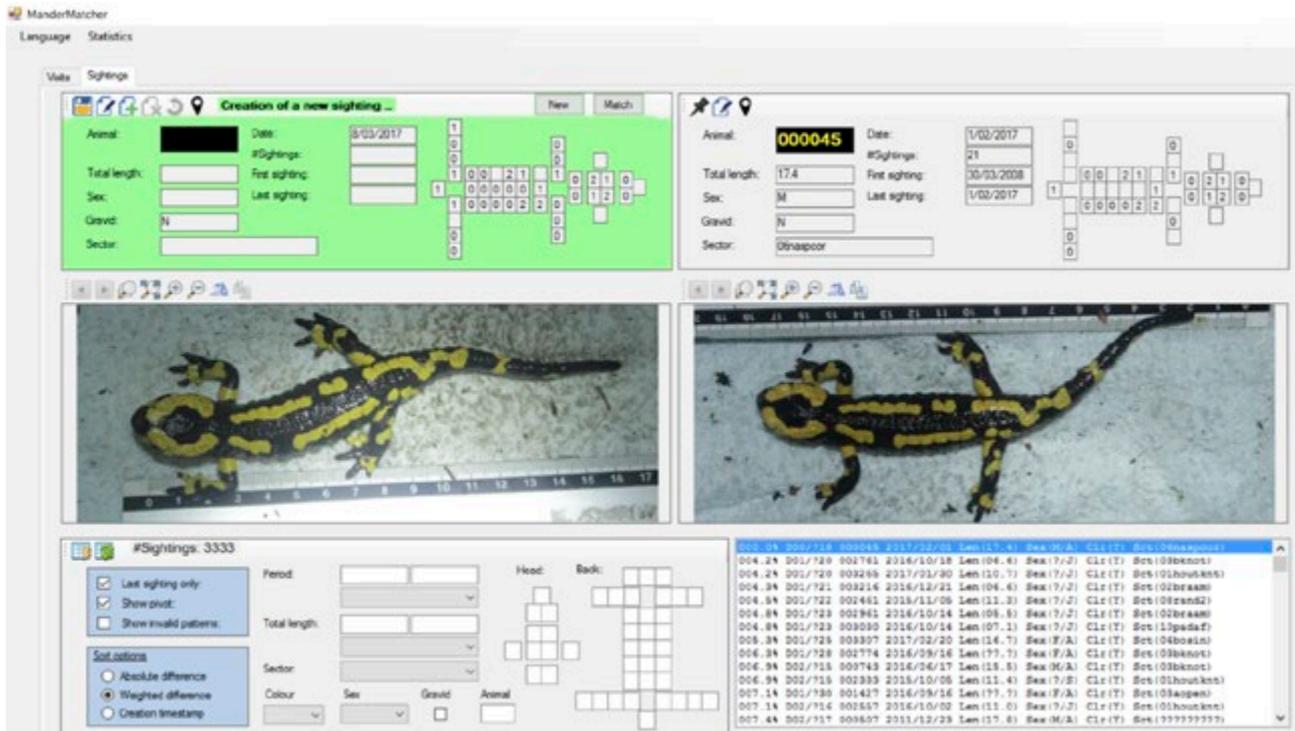
Fig. 2. 'Sightings' window for recapture recognition and database querying in the program ManderMatcher.
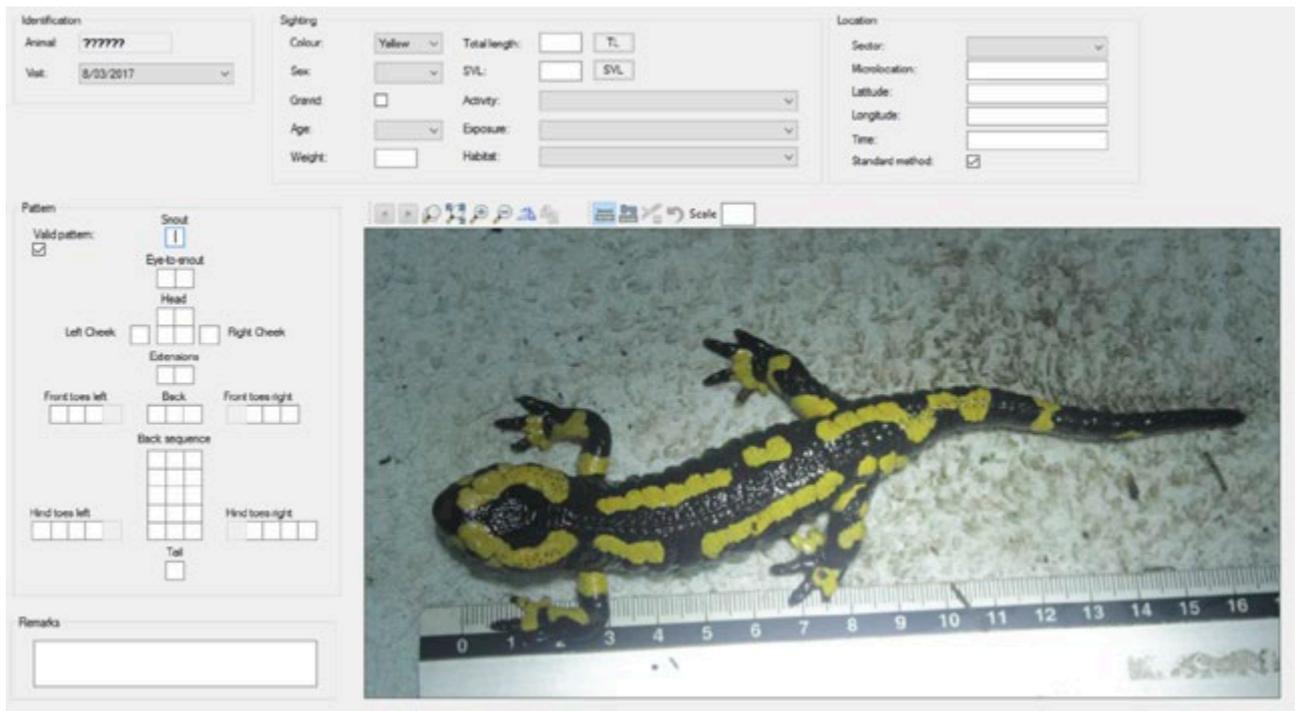

Fig. 3. Window for entry of individual sighting records in the program ManderMatcher.

the upper right. This window is used for entry of new sightings, as well as for editing of existing sightings. At the left, the CMR pattern code is entered, while the upper fields allow to store several other variables. Hitting the 'OK' button at the right after data entry/edit leads back to the 'Sightings' window and initiates the matching process.

In the cited available methods (see Introduction), a list of observations ordered by decreasing match likelihood is the endpoint of the matching process. Unlike any other, however, our method offers filter criteria to reduce this list (by entering features which are considered to be stable) as an additional tool to detect recaptures. This is particularly useful for matching younger specimens, which are likely to have undergone certain pattern changes.

Additional variables

Besides CMR matching, the program offers several tools which are relevant for ecological study of salamanders. A first set involves data related to the individual sighting, based on characteristics of the animal. We refer for a full list to the provided download link and the manual of the program, and only provide a general outline here. Biometric fields are available – weight and length measurements (total length and snout-vent length). The latter can be measured within the program itself if a calibration object is properly inserted into the picture. Ecological aspects can be documented in the shape of discrete variables to discern patterns in e.g. activity and habitat use throughout salamander life history. Several fields allow further specification of details concerning the encounter location. All individual-related data is entered through the individual sighting entry window (Fig. 3).

A second set of variables, which are entered through the 'Visits' window (Fig. 1), consists of metadata related to the count event (visit), including an array of environmental variables which can be registered both at start and end of the count event. All are defined and discussed in the program's manual.

Database

All data is saved into a MS Access database, which offers ample opportunity to query the entered data and export for analysis.

Application

The program was applied to *S. s. terrestris* data collected in a forest in the town of Merelbeke, Flanders, Belgium. Sampling was performed along a standardised transect of 1.3km. From March 2008 to January 2017, 169 sampling events were executed by night during activity-provoking weather conditions. All encountered post-metamorphic fire salamanders were captured, photographed and released.

Validation

To validate the applied search algorithm, a random sample of 100 sightings of adult salamanders was drawn from the database. One-by-one visual photograph comparison was conducted.

RESULTS

Application

A total of 9,397 sighting records were collected during standardised transect sampling events. Of these,
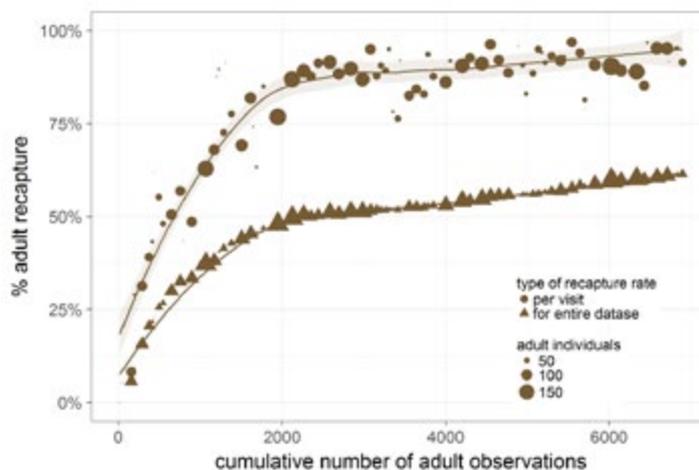

Fig. 4. Recapture rate by cumulative number of sighting records of adult animals, by visit (circles) and for entire accumulating dataset (triangles) with non-parametric, locally weighted regression smoother (loess) and 95% confidence interval. Point size relates to absolute number of encountered adults within each visit (encounter event) for both point types (circles and triangles), presented as a continuous variable.

6,899 are sightings of animals larger than 14 cm, here treated as adults. These adult sightings consist of (re) captures of 1,477 different individual adult salamanders. The overall percentage of animals captured at least twice, as obvious from the growing, cumulative database is shown as a function of the growing total of adult individual observations by the triangles in Figure 4, each data point representing a capture event. The cumulated dataset recapture rate amounts to 62.4% in the entire database at the final instance of data collection. The circles in the graph, however, not showing the percentage of recaptures in the cumulated database but the percentage within the data of a single visit, reach far higher values and, for instance, correspond to 91% of the adult individuals captured during the last visit (at the extreme right in the graph).

Validation

The random sample comprised 89 adult individuals, of which 80 were encountered only once within the sample, 7 were encountered twice, and 2 individuals were encountered three times. The exact same result was obtained by application of the program, even though matching of these 100 randomly selected sightings was done against up to 9,397 records, thus making successful match finding more complicated than if 100 new records were to be entered into an empty database. While matching through individual picture comparison took about 4 hours, matching 100 new records one by one with an unstructured collection of 9,397 photographs would take an impractical amount of time. While the speed of using MandermMatcher differs depending on the size of the database and recapture rate within the data, using the matching algorithm and finding a match or not (excluding entry of other data) takes a user which is sufficiently acquainted with the program an average of 2 minutes or less per record.

The uniqueness of certain code combinations can be calculated theoretically and from our data on *S. s. terrestris*. As an example, we consider the toe characteristics. Fourteen toe characteristics (with potential values 0 and 1) corresponds to 2^{14} or 16,384 theoretical combinations. Using our data to assess the number of actual combinations, we used a subset of 1,597 observations for which all toe characteristics were determined. These observations relate to 737 individual animals. Within these 737 individuals, 488 different toe colour combinations occur, of which 52.2% are found in a single individual only. The most commonly encountered combination (i.e., all black toes) is shared by 5.4% of the individuals.

DISCUSSION

While the actual recapture numbers shown for the example population greatly depend on the applied count methodology and, possibly, the nature of the investigated population, they serve as illustration that with sufficient effort high recapture success can be achieved. Manual validation showed no mismatches in comparison with application of the algorithm. After the program's initial release, as substantiated by this paper, tools for exploratory analysis are under development (Speybroeck and Steenhoudt, in prep.). These include analysis of population dynamics through space and time and mapping features, but also analysis of pattern variability, allowing the recognition variables to serve a second purpose, apart from CMR matching.

Several non-invasive methods exist for CMR recognition of individuals. Some may require a lot of time in the field and/or behind the desk. We have demonstrated how MandermMatcher allows effective matching of flexible-bodied fire salamanders at a swift rate, requiring limited handling time per record in the field, no photograph processing at the desk and easy and fast record entry. During a high number of recapture events (169), the method was successfully applied to a dataset of 9,397 records and 1,477 adult individuals, which is clearly larger than any to which other methods were applied (e.g., 1,606 records of 1,321 individuals in Drechsler et al., 2015; 852 photos of 324 individuals in Sannolo et al., 2016). More importantly, the dataset spans a period of eight years. This is particularly relevant to assess how changes of pattern over time may affect recognising individual animals. Using data from a single season does not offer assurance that the quality of the matching process remains sufficiently high over time. For example, Sannolo et al. (2016) used data collected over a four month period of a single year. While these authors claim the ventral spot pattern of *Triturus carnifex* to be stable over time (citing Arntzen and Wallis, 1999, who however do not provide proof for this statement), this seems unlikely, as Drechsler et al. (2015) showed this to be incorrect for the closely related *T. cristatus*. Only by using data from a period of time which is long enough to span a relevant portion of adult life and growth of the considered taxon (e.g., three years in Drechsler et al. 2015, eight years in our study), the impact of pattern changes (whether they exist and/ or are relevant or not) on the use of the recognition algorithm can be assessed. While the dorsal pattern of adult fire salamanders is traditionally considered to remain stable over time (e.g., Eiselt, 1958; Klewen, 1991), changes may occur (Speybroeck, unpublished data). Application of MandermMatcher to data collected over eight years has

shown that the algorithm is sufficiently robust to prevent such changes from causing matching problems, which is likely to be an advantage of using numeric coding of the pattern, instead of image analysis tools basing matching on the extent of pigmentation. ManderMatcher does not require photographs to be taken in a standard way (e.g., with the animal perfectly stretched), which is particularly time-consuming when dealing with salamanders, such as the species we studied, and newts, including the subject species of Drechsler et al. (2015) and Sannolo et al. (2016). Finally, in contrast to all other tools, our program is a comprehensive research tool, including tools for length measurement and a myriad of variables related to each individual and each record.

While the pattern coding was so far only applied to *S. s. terrestris*, the character definitions offer sufficient precision and resolution for use with most spotted taxa within the *Salamandra* genus (*S. s. salamandra, S. s. almanzoris, S. s. terrestris, S. s. longirostris, S. s. bejarae, S. s. morenica* and most *S. s. bernardezi*) as well as *S. algira, S. corsica* and *S. infraimmaculata*). Two exceptions, however, may exist. Populations with a high portion of animals with poorly defined spots (e.g., populations of *S. s. gallaica, S. s. crespoi,* and part of *S. s. bernardezi* (i.e., populations formerly assigned to *S. s. alfredschmidti* but placed in synonymy by Beukema et al., 2016a)) may offer coding difficulties, whereas taxa with a high portion of animals with low numbers of spots (e.g., *S. s. fastuosa, S. s. gigliolii*) will have identical values for a number characteristics across many individual animals. However, considering the frequency distribution of actual toe colour combinations in our data, we have demonstrated how powerful the algorithm may be. Only 5.4% of the individuals share the most common combination, allowing to drastically reduce the number of individuals to be compared by using the toe characteristics alone. Further use will proof applicability to other taxa/populations, and it seems likely that for some of the taxa with less well-defined dorsal spots (e.g., *S. s. gallaica*) toe colour may provide sufficient discerning power.

The program is made freely available for use by any researcher, while the authors applaud getting in touch with them for potential collaboration, applying the same methodology across fire salamander taxa. While this method is presented for a single taxon (as was the case for the initial publication of most other methods, e.g., Van Tienhoven et al., 2007; Drechsler et al., 2015), the presented approach has potential to be used to develop similar tools for use with other species, applying a modified code with relevant characteristic definitions. It is particularly useful for species which have a pattern that can be coded to a sufficient number of characteristics.

How high this number has to be in order to allow precise matching, depends on the sample size and the frequency distribution of possible values of each characteristic. As such, using a smaller number of characteristics may be sufficient for smaller datasets. For the species at hand, population studies may serve as a crucial baseline at this point in time, given the imminent threat presented by lethal fungal infection caused by *Batrachochytrium salamandrivorans* (Martel et al., 2013, 2014). After decimating the Dutch population (Spitzen-van der Sluijs et al., 2013), severe fungus-caused mortality has occurred at several sites in Belgium and the fungus has also been detected in Germany (Spitzen-van der Sluijs et al., 2016). Thus, the availability of an easy-to-use, inexpensive CMR methodology for study of both infected and non-infected populations is of great value.

REFERENCES

Amstrup, S.C., McDonald, T.L., Manly, B.F.J. (2005): Handbook of Capture-Recapture Analysis. Princeton University Press, Princeton.

Arntzen, J.W., Wallis, G.P. (1999). Geographic variation and taxonomy of crested newts (*Triturus cristatus* superspecies): morphological and mitochondrial DNA data. Contrib. Zool. **68**: 181-203.

Beukema, W., Nicieza, A., Lourenço, A., Velo-Antón, G. (2016a). Colour polymorphism in *Salamandra salamandra* (Amphibia: Urodela), revealed by a lack of genetic and environmental differentiation between distinct phenotypes. J. Zool. Syst. Evol. Res. **54**: 127-136.

Beukema, W., Speybroeck, J., Velo-Antón, G. (2016b). Quick Guide *Salamandra*. Curr. Biol. **26**: R689-R700.

Bogaerts, S. (2002): Farbkleidentwicklung bei einigen Feuersalamanderarten und –unterarten. Amphibia **1**: 4-10.

Bolger, D.T., Morrison, T.A., Vance, B., Lee, D., Farid, H. (2012): A computer-assisted system for photographic mark–recapture analysis. Methods Ecol. Evol. **3**: 813-822.

Carafa, M., Biondi, M. (2004): Application of a method for individual photographic identification during a

study on *Salamandra salamandra gigliolii* in central Italy. Ital. J. Zool. Supp. **2**: 181-184.

Davis, T.M., Ovaska, K. (2001): Individual recognition of amphibians: effects of toe clipping and fluorescent tagging on the salamander *Plethodon vehiculum*. J. Herpetol. **35**: 217-225.

Drechsler, A., Helling, T., Steinfartz, S. (2015): Genetic fingerprinting proves cross-correlated automatic photo-identification of individuals as highly efficient in large capture-mark-recapture studies. Ecol. Evol. **5**: 141-151.

Eiselt, J. (1958): Der Feuersalamander *Salamandra salamandra* (L.), Beiträge zu einer taxonomischen Synthese. Abh. Ber. Naturk. Vorgesch. Magdeburg **10**: 77-154.

Feldmann, R. (1971): Felduntersuchungen an westfälischen Populationen des Feuersalamanders, *Salamandra salamandra terrestris* LACÉPÈDE, 1788. Dortmunder Beitr. Landeskunde, Naturw. Mitt. **5**: 37-44.

Ferner, J.W. (1979): A review of marking techniques for amphibians and reptiles. Soc. for the Study of Amphibians and Reptiles Herp. Circ. **9**: 1-41.

Klewen, R. (1985): Untersuchungen zur Ökologie und Populationsbiologie des Feuersalamanders (*Salamandra salamandra terrestris* LACÉPÈDE 1788) an einer isolierten Population im Kreise Paderborn. Abh. Westf. Mus. f. Naturkunde **47**: 1-51.

Klewen, R. (1991): Die Landsalamander Europas. Ziemsen Verlag, Stuttgard.

Kopp-Hamberger, M. (1998): Eine Methode zur individuellen Erkennung von Feuersalamandern (*Salamandra salamandra terrestris*) abhand des Zeichnungsmusters. Salamandra **3**: 239-244.

Le Galliard, J.-F., Paquet, M., Pantelic, Z., Perret, S. (2011): Effects of miniature transponders on physiological stress, locomotor activity, growth and survival in small lizards. Amphibia-Reptilia **32**: 177-183.

Martel, A., Spitzen-van der Sluijs, A., Blooi, M., Bert, W., Ducatelle, R., Fisher, M.C., Woeltjes, A., Bosman, W., Chiers, K., Bossuyt, F., Pasmans, F. (2013): *Batrachochytrium salamandrivorans* sp. nov. causes lethal chytridiomycosis in amphibians. Proc. Natl. Acad. Sci. USA. **110**: 15325-15329.

Martel, A., Blooi, M., Adriaensen, C., Van Rooij, P., Beukema, W.M., Fisher, M.C., Farrer, R.A., Schmidt, B.R., Tobler, U., Goka, K., Lips, K.R., Muletz, C., Zamudio, K.R., Bosch, J., Lötters, S., Wombwell, E., Garner, T.W.J., Cunningham, A.A., Spitzen-van der Sluijs, A., Salvidio, S., Ducatelle, R., Nishikawa, K., Nguyen, T.T., Kolby, J.E., Van Bocxlaer, I., Bossuyt, F., Pasmans, F. (2014): Recent introduction of a chytrid fungus endangers Western Palearctic salamanders. Science **346**: 630-631.

McCarthy, M.A., Weller, W.F., Parris, K.M. (2009): Effects of toe clipping on survival, recapture and return rates of Jefferson salamanders (*Ambystoma jeffersonianum*) in Ontario, Canada. J. Herpetol. **43**: 394-401.

Moya, Ó., Pep-Luis, M., Sergio, M., José-Manuel, I., Andreu, R., Antonio, R., Giacomo, T. (2015): APHIS: A new software for photo-matching in ecological studies. Ecol. Inform. **27**: 64-70.

Mutz, T. (1992): Vergleich der Farbkleidentwicklung bei Unterarten des Feuersalamanders. Urodela Info **3**: 4-5.

Ott, J.A., Scott, D.E. (1999): Effects of toe-clipping and PIT-tagging on growth and survival in metamorphic *Ambystoma opacum*. J. Herpetol. **33**: 344-348.

Ousterhout, B.H., Semlitsch, R.D. (2014): Measuring terrestrial movement behavior using passive integrated transponder (PIT) tags: effects of tag size on detection, movement, survival, and growth. Behav. Ecol. Sociobiol. **68**: 343-350.

Ryan, K.J., Zydlewski, J.D., Calhoun, A.J.K. (2014): Using passive integrated transponder (PIT) systems for terrestrial detection of blue-spotted salamanders (*Ambystoma laterale*) *in situ*. Herpetol. Conserv. Biol. **9**: 97-105.

Sacchi, R., Scali, S., Mangiacotti, M., Sannolo, M., Zuffi, M.A.L. (2016): Digital identification and analysis. In: Reptile ecology and conservation: a handbook of techniques. Dodd, K.C., Ed, Oxford University Press, Oxford.

Sannolo, M., Gatti, F., Mangiacotti, M., Scali, S., Sacchi, R. (2016): Photo-identification in amphibian studies: a test of I³S Pattern. Acta Herpetol. **11**: 63-38.

Schmidt, B.R., Schaub, M., Anholt, B.R. (2002): Why you should use capture-recapture methods when estimating survival and breeding probabilities: on bias, temporary emigration, overdispersion, and common toads. Amphibia-Reptilia **23**: 375-388.

Schmidt, B.R. (2004): Declining amphibian populations: the pitfalls of count data in the study of diversity, distributions, dynamics, and demography. Herpetol. J. **14**: 167-174.

Seifert, D. (1991): Untersuchungen an einer ostthüringischen Population des Feuersalamanders (*Salamandra salamandra*). Artenschutzreport Heft **1**: 1-16.

Speybroeck, J., Beukema, W., Bok, B., Van Der Voort, J. (2016): Field guide to the amphibians and reptiles of Britain and Europe. Bloomsbury Publishing, London.

Spitzen-van der Sluijs, A., Martel, A., Asselberghs, J., Bales, E.K., Beukema, W., Bletz, M.C., Dalbeck, L., Goverse, E., Kerres, A., Kinet, T., Kirst, K., Laudelout, A., Marin da Fonte, L.F., Nöllert, A., Ohlhoff, D., Sabino-Pinto, J., Schmidt, B.R., Speybroeck, J., Spikmans, F., Steinfartz, S., Veith, M., Vences, M., Wagner, N.,

Pasmans, F., Lötters, S. (2016): Expanding distribution of lethal amphibian fungus *Batrachochytrium salamandrivorans* in Europe. Emerg. Infect. Dis. **22**: 1286-1288.

Spitzen-van der Sluijs, A., Spikmans, F., Bosman, W., De Zeeuw, M., Van der Meij, T., Goverse, E., Kik, M., Pasmans, F., Martel, A. (2013): Rapid enigmatic decline drives the fire salamander (*Salamandra salamandra*) to the edge of extinction in the Netherlands. Amphibia-Reptilia **34**: 233-239.

Šukalo, G., Dorđević, S., Golub, D., Dmitrović, D., Tomović, L. (2013): Novel, non-invasive method for distinguishing the individuals of the fire salamander (*Salamandra salamandra*) in capture-mark-recapture studies. Acta Herpetol. **8**: 41-45.

Thiesmeier, B. (2004): Der Feuersalamander. Laurenti Verlag, Bielefeld.

Van Tienhoven, A.M., Den Hartog, J.E., Reijns, R.A., Peddemors, V.M., (2007): A computer-aided program for pattern-matching of natural marks on the spotted raggedtooth shark *Carcharias taurus*. J. Appl. Ecol. **44**: 273-280.