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HARMONIC DIRECTION FINDING: A NOVEL TOOL TO MONITOR THE DISPERSAL OF SMALL-SIZED ANURANS

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The suitability of harmonic direction finding for tracking of dispersing juvenile natterjack (*Bufo calamita*) and green toads (*B. viridis*) was evaluated in laboratory and field experiments. In a first step, dipole reflector tags were developed which combined low mass, small size and large detection range. The average mass was 114 mg, wire antenna length 42 mm and detection range usually varied between 2.5 m and 12.5 m – occasionally reaching 26 m – as assessed using a commercial portable scanning device RECCO 5000. In toads that had a snout-vent length of 22-24 mm, the mass of the reflector tag did not exceed 10% of the toad's body mass. Tags were externally attached by glueing to the dried dorsal skin of the toadlet. In a replicated laboratory experiment, almost all tags were shed 36 hr to 48hr after attachment. In 2001, 417 juveniles toads were equipped with reflector tags and their dispersal was studied in a natural habitat (Urmitz, Rhineland-Palatinate, Germany). The recovery rate of reflector tags was similar in *B. calamita* (35.9%, *n*=33) and in *B. viridis* (31.6%, *n*=103). The maximum distances between release and recovery site were 588 m in *B. calamita* and 665 m in *B. viridis*. Results obtained suggest that this new method is better suited for monitoring the migratory activity and habitat use of small terrestrial anurans than passive tagging systems presently in use, such as microtags and passive integrated transponders (PIT). Nevertheless, detection range is still too small to rival active monitoring systems such as radiotransmitters which remain unsuitable for small anurans.

Key words: Bufo calamita, Bufo viridis, postmetamorphic dispersal, passive tracking system

INTRODUCTION

Neighbouring populations are linked by dispersing individuals which do not only maintain gene flow but also counteract local extinction by recolonization of empty habitat patches (e.g. Hanski & Gilpin, 1997; Poethke *et al.,* 2003). Recent evidence even suggests that differences in dispersal ability among amphibians may play an important role in their sensitivity to the global decline phenomenon (Green, 2003). In many amphibian species, distances covered by dispersing juveniles by far exceed those of adults and thus, connectivity among populations mainly depends on the early terrestrial life stage (e.g. Dole, 1971; Breden, 1987; Sinsch 1991, 1997*a*). Nevertheless, our knowledge of the dispersal of juvenile amphibians is widely restricted to chance observations because of the absence of a suitable quantitative monitoring technique (e.g. Heyer *et al*., 1994, Cooke *et al.,* 2004). Active systems such as radio tags are still too large and heavy for most species and the detection range of passive systems such as passive integrated transponders or microtags rarely exceeds a 0.2 m (e.g. Sinsch, 1997*b*; Ott & Scott, 1999). Consequently, a new technique closing the gap between the currently used passive and active tracking systems in amphibian population ecology is urgently needed.

Harmonic radar (Riley *et al.*, 1996) and harmonic direction finding (Mascanzoni & Wallin, 1986) are techniques to locate diode tags which convert the frequency of an incoming radio signal to a harmonic frequency and reflect this harmonic as an outgoing signal. These techniques differ from active systems by the low mass and low price of tags and from passive ones by a larger detection range (Langkilde & Alford, 2002). Harmonic radar (radio detection and ranging) provides information on direction and distance of free-moving animals and has been successfully used to track bees (Riley *et al.*, 1996; Carreck *et al.*, 1999; Capaldi *et al.*, 2000), bumble bees (Osborne *et al*., 1999) and moths (Riley *et al.*, 1998). However, radar studies require heavy equipment which is usually stationary. Harmonic direction finding (HDF) provides only information on direction, while the exact location of the tracked animal has to be assessed by homing-in with a portable detector. This method has been successfully applied to ground-moving carabid beetles (Hockmann *et al.,* 1989), snails (Janßen & Plachter, 1998) and snakes (Webb & Shine, 1997; Engelstoft *et al.*, 1999). Surprisingly, harmonic direction finding has not yet been used to monitor dispersing amphibians in which size limitations prevent the use of radio tags as in most insects. In a pilot study, we adapted this technique to track free-ranging juvenile *Bufo calamita* and *B. viridis* toadlets by developing suitable reflector tags which were small enough to minimize potential effects on behaviour and still provided an acceptable detection range (Leskovar & Sinsch, 2002). As results obtained on a low number of individuals were promising, the suitability of HDF tracking of toadlets was tested in quantitative field experiments. In this paper, we present (1) a detailed description of optimized reflector tags weighing about

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114 mg; (2) the corresponding detection ranges under laboratory and field conditions; (3) a suitable attachment method for short-term studies on small-sized toads; and (4) the recovery rates of tags and migratory distances quantified in field trials with 92 *B. calamita* and 325 *B. viridis* toadlets.

MATERIAL AND METHODS

Dispersal of juvenile natterjack (*Bufo calamita*) and green toads (*B. viridis*) was studied using a harmonic direction finding system. In 2001, juveniles (17.5-44 mm SVL) were collected in the vicinity of the breeding pond within a sandy gravel pit area near Urmitz (Rhineland-Palatinate, Germany; details on habitat structure in Sinsch *et al.* 1999). During daytime, juveniles mostly burrow into the sand or hide below stones and wooden boards. Migratory activity is mainly restricted to the first half of the night. Snout-vent length (to the nearest mm) and body mass (to the nearest 10 mg) of toadlets of each species were recorded prior to experimental treatments.

Tracking Methods and Equipment

The HDF equipment used in this study consists of a commercial portable scanning device RECCO 5000 (size: 0.4 m × 0.1 m × 0.18 m, mass: 1.6 kg; supplier: RECCO AB, P.O. Box 4028, S-181 04 Lidingö, Sweden; price: ca. 5500 Euros) and self-built reflector tags (commercial silicium diode and antenna; price: ca. 0.50 Euro per unit). The hand-held RECCO 5000 includes a transmitter which emits a microwave (frequency: 917 MHz at 4-5 Watts), i.e. a directional signal, a detector for incoming microwaves which are transformed to an acoustic signal, and a headphone which allows an operator to evaluate amplitude and frequency of the acoustic signals. The self-built reflector tag used in this study is basically a dipole consisting of a diode connected with two antennas which reacts as the hollow antenna in commercial RECCO reflector tags (dipole design adapted from Janßen & Plachter, 1998). If the emitted microwave strikes the reflector diode, the incoming microwave is reflected with the doubled frequency (1834 Mhz), detected by the receiver of the RECCO 5000 and transformed to an acoustic signal of correspondingly increased frequency (Mascanzoni & Wallin, 1986). The rechargable batteries (NiMH, voltage: 6V) permit an operation period of about 3-4 hr.

Commercial reflectors provided by RECCO AB were too large and heavy to tag small animals. Therefore, in all tracking studies using HDF tags were specifically designed and assembled to meet size, shape and mass of the target organisms, some for implantation (Webb & Shine, 1997; Engelstoft *et al.* 1999), others for external attachment (Langkilde & Alford, 2002; this study). Here, we describe the design of the reflector tag developed for toadlets (Fig. 1). Tags consisted of a silicium diode (75V, 0.075A, device number 1N4148, size: 3.5 mm × 1.5 mm, mass: 44 mg), an isolated copper wire antenna and a copper foil antenna which were soldered to either pole of the diode. The length of the wire antenna was 42 mm (diameter: 0.2 mm), the surface area of the foil antenna 15-20 mm × 2.5-3 mm, depending on the size of the toadlet which was tagged. The total mass of a reflector tag averaged 114 mg (range: 101-130 mg; Fig. 1). The optimum length of the wire antenna corresponds to $\lambda/4 = 42$ mm with wave length λ [cm] = speed of light c [300,000 km/h] / frequency of transmitter f [917 MHz]. The wire antenna was flexible, but did not kink. Wire and foil antenna were fixed in a T-like position to each other because the detection range decreased to about 50%, if the angle between the antennas was less than 90°. A copper wire which was wound round the diode five and a half times linked the poles. This coil provides a return path to prevent the rectifying action of the diode producing a DC charge distribution that would tend to bias the diode into the non-conducting state, but is of high enough inductance to avoid shorting out the 917 MHz signal (Riley & Smith, 2002). We obtained the maximum response, if neighbouring windings did not have direct contact. The optimal position of the wire coil was fixed using nail varnish. Combinations of different colours permitted batch marks indicating species and release date. Size and shape of the foil antenna were optimized empirically as well as wire length and the number of windings between the poles.

Detection Range an Attachment Procedure

The maximum distance at which reflector tags were detectable under field conditions was assessed for 10 randomly selected tags. Four combinations of tag location and vertical distance between tag and detector were tested: (1) tag placed on ground surface, detector waved at 1.5 m above ground; (2) tag buried 0.05 m below ground surface, detector at 1.5 m above ground; (3) tag placed on ground surface, detector at 4.5 m above ground; (4) tag buried 0.05 m below ground surface, detector at 4.5 m above ground. If the detection range was less than 4 m, the detector was moved towards the tag until receiving the first signal. To explore the potential effect of tag contact with toad skin on detection range, we also tested the combination: (5) tag attached to a toad and placed at the ground surface, detector at 1.5 m above ground.

FIG 1. Reflector tag consisting of a silicium diode, a copper foil antenna and a copper wire antenna. The coin has a diameter of 16mm.

External adherence of tags to the moist skin of amphibians using glue is a difficult task. Relying on the long-term experience of attaching magnets to the skin of European *Bufo* for orientation experiments, we chose cyanoacrylate glue (Sekunden Alleskleber, Uhu) which evidently does not have adverse effects besides an acceleration of moulting frequency (e.g. Sinsch, 1987, 1992). However, unpublished observations on other species suggest that cyanoacrylate glue may cause severe wounds in the skin of European *Rana* spp. and *Hyla arborea* and that *Bufo* skin is the exception to the rule. Reflector tags were attached to the dried dorsal skin of a toadlet by bending the flexible foil antenna around the toad's dorsum and fixing it with a small drop of glue (Fig. 2).

Experiment 1: Endurance of Tagging Under Laboratory Conditions

Moulting is a limiting factor for the endurance of any tag attached to the skin of a toad. In *B. calamita* the period between two moults varies from 4.0 to 8.1 days (Sinsch *et al*., 1992). As the endurance of tagging can hardly be estimated in the field, we chose a laboratory approach to quantify the time between attaching and shedding the reflector tags. A total number of 87 juvenile *B. calamita* and 189 juvenile *B. viridis* were collected during July and August in the study area. They were randomly assigned to four groups, fitted with reflector tags and kept at room temperature and a natural light-dark cycle in plastic boxes (0.41 m × 0.61 m × 0.22 m) with moist sand (0.04 m deep) until all had lost the tags. Individual numbers and species composition of treatment replicates was: (1) 50 *B. viridis* and 33 *B. calamita*; (2) 60 *Bv* and 25 *Bc*; (3) 39 *Bv* and 14 *Bc*; (4) 40 *Bv* and 15 *Bc*. The number of toads with attached tags per replicate was counted every 12 hrs. At the end of the experiment toads were kept another 4-5 days in captivity to look for potential adverse effects of the treatment. After this period they were released again in their natural habitat.

Experiment 2: Dispersal of Tagged Juvenile *B. calamita* and *B. viridis* In Their Natural Habitat

Between 2 July and 21 October 2001, a total of 92 juvenile *B. calamita* and 325 juvenile *B. viridis* were collected during twelve 3-hr afternoon surveys in the study area. Collection and release date, group size and species composition were: 22 *B. calamita* and 26 *B. viridis* (2/07), 8 *Bc* and 18 *Bv* (10/07), 26 *Bv* (7/08), 24 *Bv* (17/08), 5 *Bc* and 40 *Bv* (24/08), 26 *Bv* (30/08), 5 *Bc* and 24 *Bv* (5/09), 2 *Bc* and 30 *Bv* (7/09), 24 *Bc* and 24 *Bv* (30/09), 5 *Bc* and 23 *Bv* (21/09), 5 *Bc* and 32 *Bv* (27/09), 16 *Bc* and 32 *Bv* (21/10). During and following the surveys toads were kept for 3-7 hr in plastic boxes (0.41 m × 0.61 m × 0.22 m before release at sunset. In order to reduce handling stress the tags were attached in the field immediately before releasing the toadlets. As a group, the tagged individuals were placed on moist ground (sand) below a wooden board close to the breeding pond (Fig. 3). As the release site was the same for all experimental groups, the colour of the wire antenna indicated the species and the colour of nail varnish the date of release. Thus, we could identify any tag detected with respect to the corresponding species and the release group.

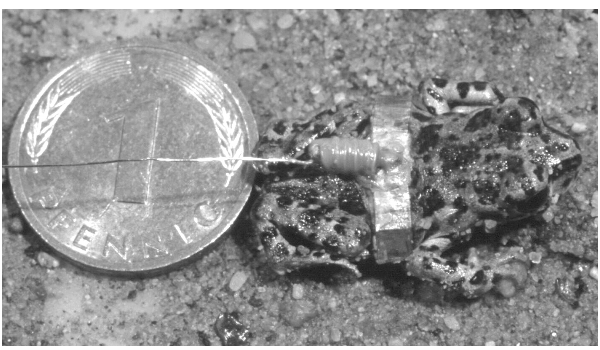

FIG 2. Juvenile *Bufo viridis* fitted with a reflector tag. The coin has a diameter of 16 mm.

We did not intend to follow the individual paths of toads during their dispersal to avoid disturbance of the resting or migrating individuals and therefore, surveys to detect tags began 4-6 days after release, i.e. when almost all toadlets had already shed the tags. Surveys were exclusively performed during daytime and limited by the maximum battery charge to 6-7 hr (two batteries were available). The first search of tags following a release began at the release site with the receiver switched to maximum intensity and the detector antenna was held horizontally (Engelstoft *et al*., 1999). In the close vicinity of the release site, the operator moved in a spiral by steadily increasing the distance to the release site by about 7-8 m per turn. However, the landscape did not permit an ideal spiral search at distances greater than about 120 m because large parts of the survey area were

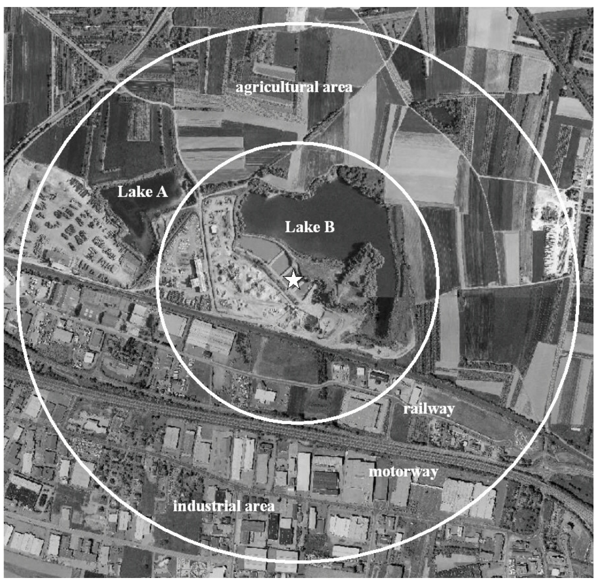

FIG 3. Aerial view on the study site. The star indicates the release site, the inner circle (diameter: 1 km) the most intensely surveyed area, the second circle (diameter: 2 km) the outer limit of the surveyed area.

covered by lakes, buildings, streets etc. (Fig. 3). Therefore, the accessible regions of the inner survey area (diameter: 1 km) were systematically searched in concentric circle segments until completely covered. A complete survey of the inner survey area lasted 18-20 hr. The survey within the outer search area (diameter: 2 km) was restricted to the area north of the railway because it soon became clear that toadlets did not cross the railway dam during the tagging period. As this area is considerably larger than the inner survey area, the outer region was scanned four times during 2001. A final complete survey to detect overlooked tags was performed two years after the last release, on 1 August 2003.

The most efficient method to detect the presence of reflector tags was waving the radar receiver at ca. 1.5 m above ground while moving through the habitat, because the signal intensity depends on the relative position of the linearly polarized receiver antenna to the tag (Janßen & Plachter, 1998). If the acoustic signal indicated the presence of a tag in the vicinity of the operator's position, its exact location was determined by a homing-in procedure, by progressively decreasing the sensitivity of the receiver until reaching the minimum. The location of buried tags often required time-consuming passing of soil through a sieve. Following tag identification its position was recorded as polar coordinates consisting of the direction to (Suunto compass) and distance from the release site (Bushnell laser range finder).

Statistical Analyses

All data sets were tested for normal distributions by determining standardized skewness and kurtosis. Distributions of maximum detection distance per treatment and frequency of toads with attached tags did not differ from a normal distribution and were consequently compared using ANOVA and a multiple range test following Bonferroni correction or *t*-statistics. Means are always given with corresponding standard error. In contrast, distribution of dispersal distances was significantly skewed and compared using the Mann-Whitney *U*-test and the Kolmogorov-Smirnov test. We fitted a double-logarithmic regression model to size-mass relationships and calculated the Pearson product moment correlation. Significance level was set at $\alpha=0.05$. All calculations were performed using STATGRAPHICS Plus for Windows, version 5.0.

RESULTS

Detection Range

The location of the tag relative to the scanning device influenced the detection range significantly (ANOVA, $F_{4,45}=36.03$, $P<<0.0001$; Fig. 4). Individual features of a tag, e.g. size and shape of antennas also influenced detection range. Even if exposed to identical environmental conditions, the maximum detection distance of the best tag was 1.92 times larger than that of the worst performing one (e.g. 12.5 m versus 6.5 m). Maximum detection distance (±SE) averaged 9.45±0.61 m, if tags were not attached to a toad and lay on the ground surface while the detector was moved at 1.5 m above ground. Any other test mode significantly reduced average detection range (Multiple range test, $P<0.05$; Fig. 4).

Nevertheless, maximum detection distance occasionally exceeded that of the standardized experiment. Preliminary tests following the assembly of tags within the University building yielded detection ranges of up to 30 m, e.g. if tags were placed on tables (75 cm above ground) and intervening structures between tag and detector were absent. In the field, maximum detection distance was 26 m in a reflector tag attached to a toad which hid within the moist sand of a slope, i.e. tag was at detector height. Nevertheless, the probability of detecting buried tags was lower than that of finding tags on the ground surface. Increasing the vertical distance between tag and detector did not increase detection range.

Biometric Features of Toads

In experiment 1 snout-vent length of experimental juveniles ranged between 18.5-34.5 mm in *B. calamita* (median: 26.5 mm, n=87) and 19.5-31.0 mm in *B. viridis* (median: 23 mm, n=189); in experiment 2 the corresponding values were 22.0-44.0 mm in *B. calamita* (median: 32.5 mm, n=92) and 17.5-32.0 mm in *B. viridis* (median: 25 mm, n=325). The overall size-mass relationship significantly differed between the juveniles of *B. calamita* (regression model: log10(SVL, mm) = -4.35 + 3.22 × log10(mass, g); n=189, R^2=98.3%) and *B. viridis* (regression model: log10(SVL, mm) = -4.53 + 3.32 × log10(mass, g); n=514, R^2=98.6%) with respect to slope (ANOVA, P=0.0072) and intercept (ANOVA,

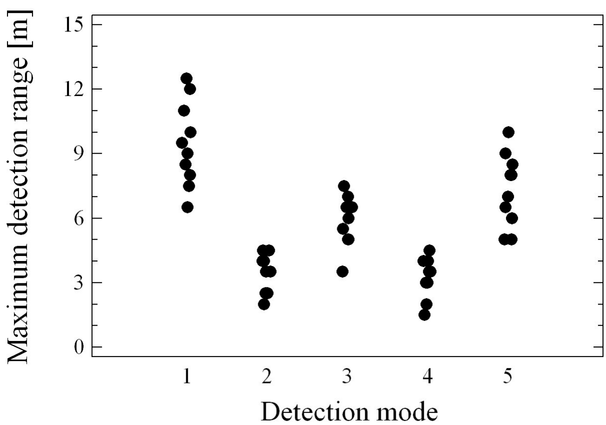

FIG 4. Maximum detection range of 10 reflector tags tested in five detection modes. (1) tag placed on ground surface, detector waved at 1.5 m above ground, detection range: 9.45±0.61 m (mean±SE); (2) tag buried 0.05 m below ground surface, detector at 1.5 m above ground, detection range: 3.30±0.31 m; (3) tag placed on ground surface, detector at 4.5 m above ground, detection range: 5.90±0.37 m; (4) tag buried 0.05 m below ground surface, detector at 4.5 m above ground, detection range: 3.25±0.29 m; (5) tag attached to a toad and placed at the ground surface, detector at 1.5 m above ground, detection range: 7.30±0.53 m. If detection range was less than 4.5 m (modes 3 and 4), the detector was moved towards the tag until receiving the first signal. Each dot represents an individual tag.

$P<0.0001$, Fig. 5). The tag load varied between 3.3% and 23.8% of individual body mass depending on toad size (Fig. 5). The snout-vent length at which the load amounted to ca. 10% of the individual body mass was 22-24.5 mm. The maximum individual tag load measured in experiment 1 was 19.4% in *B. calamita* (median: 7.0%) and 15.2% in *B. viridis* (median: 9.3%). The corresponding values for the field trials were 11.9% in *B. calamita* (median: 3.5%) and between 23.8% in *B. viridis* (median: 8.5%).

Experiment 1: Endurance of Tagging Under Laboratory Conditions

The number of tags still attached to the skin significantly varied in time in both species (3-factor ANOVA; $F_{6,45}$=1079.94, $P<<0.0001$; Fig. 6). In contrast, the time course of reflector loss did neither differ among the four replicate trials (3-factor ANOVA; $F_{3,45}$=1.02, $P>0.05$) nor between the two species (3-factor ANOVA; $F_{1,45}$=0.0, $P>0.05$). Single toads lost their tags during the first 12 hr following attachment. However, most toads shed the tags after 36 hr to 48 hr. Maximum duration of tag attachment was 72 hr. Attachment period of tags was unrelated to toadlet size. In a few individuals, skin which had been exposed to cyanoacrylate glue and/or the copper foil antenna was darker than normal. There was no mortality within a week after tagging.

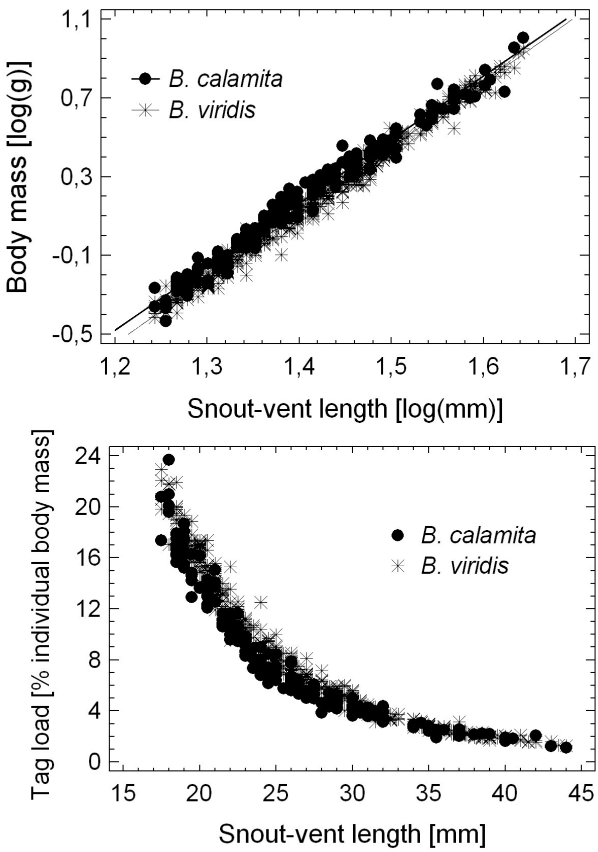

FIG. 5. Size-mass (top) and size-tag load (bottom) relationships in 179 juvenile *B. calamita* and 514 juvenile *B. viridis* studied in laboratory and field. For statistical details see text.

Experiment 2: Dispersal of Tagged Juvenile *B. calamita* and *B. viridis* in Their Natural Habitat

With a few exceptions reflector tags were located after the toads had lost them. Average recovery rates per release did not vary significantly between *B. calamita* (35.0±3.6%, n=9) and *B. viridis* (31.0±3.0%, n=12; t-test, t=0.86, $P>0.05$). The overall recovery rates of tags were 35.9% (n=33) in *B. calamita* and 31.6% (n=103) in *B. viridis*. Nine of the 136 recovered tags were detected during the final survey of the study area two years after the last release. In *B. calamita* five tags were found in cavities below stones which lay on the ground surface, six tags were buried up to 5 cm in sand, and 22 tags lay visible on the ground surface. In *B. viridis* one tag was located on a toadlet sitting in the pasture, five tags below stones, six buried in the sand, and 81 on the ground surface.

The maximum distance between release and recovery site was 588 m in *B. calamita* and 665 m in *B. viridis* (Fig. 7). Neither medians (68 m vs. 61 m; Mann-Whitney U-test, U=1782, $P>0.05$) nor shape of the species-specific distributions differed among each other (Kolmogorov-Smirnov test; two-sided large sample K-S statistic = 0.769, $P>0.05$). In both species, most of the

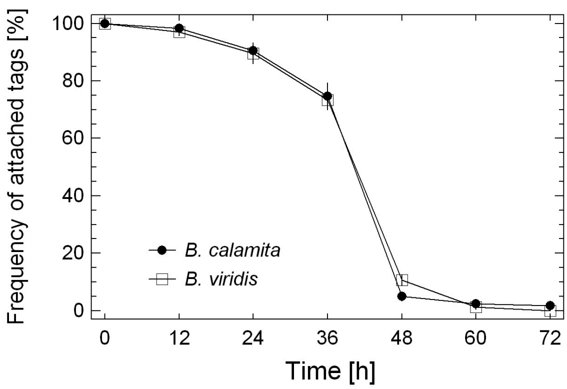

FIG. 6. Time course of reflector loss under laboratory conditions. Tags had been glued to the dorsal skin of 87 *B. calamita* and 189 *B. viridis*. Each symbol represents the average number (±SE, n=4 replicates) of tags still attached to a toad.

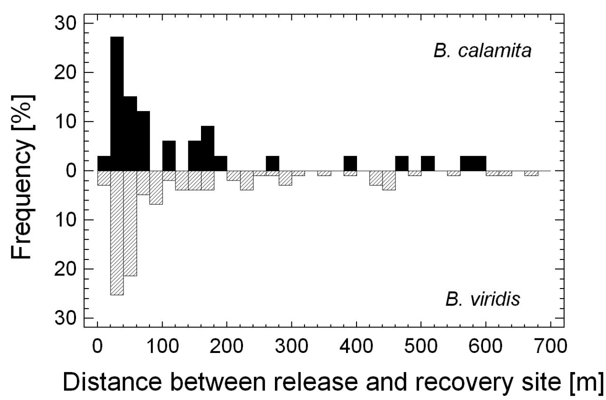

FIG. 7. Distances between release and recovery sites of attached reflectors (*B. calamita*: n=33; *B. viridis*: n=103). Data are presented as percent reflector tags within consecutive 20 m classes.

recovered reflectors (81.8% vs. 74.8%) were detected within a radius of 200 m around the release site. As the toadlets were not individually tagged, data on the potential effect of tag load on the dispersal distance were not available.

DISCUSSION

Harmonic direction finding has proved to be a suitable method to track non-climbing, free-ranging juvenile toads in their natural habitat, as demonstrated previously for carabid beetles (Hockmann *et al.*, 1989) and snakes (Webb & Shine 1997, Engelstoft *et al.* 1999). Compared to the few other available methods (e.g. microtags, passive integrated transponders), the small size and the low price of reflector tags combined with a detection range of about 7 m on average are intriguing advantages of HDF tracking (detailed discussion in Langkilde & Alford, 2002). Quantitative studies on the terrestrial dispersal of juvenile amphibians have come closer into the reach of population ecologists because now tagged toadlets can be located even if not visible to the observer. Before, it was necessary to locate the target organism by other means and to establish the presence of a tag in the already captured individual. Besides the obvious advantages of this method, there are several limitations which have to be considered, if planning a herpetological field study using HDF (Engelstoft *et al.*, 1999; Langkilde & Alford, 2002).

PREPARATION OF REFLECTOR TAGS

The tags which we describe here were optimized for HDF tracking of ground-moving juvenile toads and differ in size, mass and diode type from those used for other target organisms. When testing different types of diodes, we found that the detection range hardly depended on the diode type but mainly on antenna features. We used relatively large silicium diodes instead of Schottky diodes (e.g. Janßen & Plachter, 1998, Engelstoft *et al.* 1999) or germanium diodes (Langkilde & Alford, 2002) because they fitted best to our antenna design and smaller diodes would have required a more sophisticated electronic laboratory equipment. The dipole antenna design was chosen to facilitate attachment to the toad skin and to enhance signal reflection which is influenced by the alignment of tag and detector antennas (Janßen & Plachter, 1998). In contrast, studies on free-ranging snakes and captive hylids used a single antenna design with antenna lengths 70-130 mm (Webb & Shine, 1997, Engelstoft *et al.*, 1999, Langkilde & Alford, 2002). We refrained from using antenna of this length considering the small size of toadlets and the risk that the antenna becomes entangled with vegetation. As none of the recovered tags was caught within vegetation we assume that the external antenna length of 42 mm was an appropriate compromise between signal reflection capability and obstacle for movement within vegetation. Nevertheless, due to the external attachment tagged toadlets are more conspicuous to visually hunting predators than untagged ones and may suffer from an increased predation risk. This applies to those individuals which do not burrow during daytime as occasionally observed in juvenile green toads.

Due to diode size and antenna design, the resulting tag mass of about 114 mg was greater than that reported for vertebrates for snakes (4 mg; Engelstoft *et al.*, 1999) or snails (69 mg; Janßen & Plachter, 1998). According to the recommendations for radio tracking, the total mass of a tag should not significantly exceed 10% of a toad's body mass (Richards *et al.*, 1994). Applying this rule-of-thumb to HDF tracking by far most of the juveniles were below this limit, but several individuals with 22-24 mm SVL and all smaller than 22 mm passed the limit, reaching tag loads of more than 20% body mass. We were unable to study the potential effects of tag load on locomotory activity as recovered tags could not be assigned to individuals, but it is probable that dispersal velocity will be negatively affected with increasing relative tag mass. Consequently, more light-weighed tags are needed to track metamorphs of *B. calamita* (6-11 mm SVL) and *B. viridis* (11-18 mm SVL) as well as small juveniles.

ATTACHMENT OF TAGS

Owing to the small size and the shape of toadlets a surgical implantation of tags into the peritoneal cavity as commonly practiced with radio transmitters but also in HDF studies on snakes was not considered (Stouffer *et al.*, 1983; Webb & Shine, 1997; Engelstoft *et al.*, 1999). External attachment of reflector tags to the moist skin of amphibians is a greater challenge than glueing a tag permanently to a chitin exoskeleton or to a snail-shell. We are aware of only two other studies on amphibians in which HDF tags were used to track adult *Litoria lesueuri* (Langkilde & Alford, 2002) and adult *Hyla arborea* (J. Pellet unpubl.). In these studies tags were fixed with an elastic waistband. The period in which frogs remain tagged is potentially longer using waistbands than using glued tags, however, waistbands irritate frogs causing them to move more often (Langkilde & Alford, 2002). Moreover, experiences derived from fixing mechanical tracking devices with waistbands suggest that they may cause skin lesions, if they are worn more than a week (e.g. Sinsch, 1988; Heyer *et al.,* 1994). We preferred tag attachment with cyanoacrylate glue because this glue does not seem to damage skin in the European toad species *(B. bufo, B. calamita, B. viridis*; Sinsch 1987, 1992). However, cyanoacrylate glue considerably reduced the period between two moults from 4.0-8.1 days to less than 2 days in *B. calamita* (Sinsch *et al.*, 1992; this study). A consequence of accelerated moulting is the temporal limitation of the tracking period to mostly two nightly activity periods, if the rate of glue-induced tag shedding estimated in the laboratory is similar to that in the field. However, considering that about 2/3 of the tags were not recovered, the low endurance of the tag attachment guarantees that toadlets which are not recaptured will not suffer from potential long-term effects of the tag. The occasionally observed skin darkening was not limited to areas in direct contact with glue and more probably caused by the contact with the copper foil antenna.

DETECTION RANGE

The maximum distance at which HDF tags can be located using a RECCO detector is reported to depend on the antenna length and the tag's position over ground (Janßen & Plachter, 1998; Engelstoft *et al*., 1999; Langkilde & Alford, 2002). Tags placed on the ground surface are detectable from ca. 3 m at 20 mm antenna length and from ca. 20 m at 180 mm antenna length. If the tag's position is about 0.6-0.8 m above ground, maximum detection distances of ca. 60 m are feasible. Signal amplitude is greatly attenuated, if HDF tags are buried and they may even become undetectable (Engelstoft *et al*., 1999). Our data on ground-dispersing and burrowing toads corroborate these features into detail and suggest that under field conditions the average detection range is 7-8 m.

Further constraints influencing detection range are attenuation due to vegetation, to alignment of antenna and detector, and to the relative orientation of dipole antennas to each other (Janßen & Plachter, 1998; Engelstoft *et al*., 1999; Langkilde & Alford, 2002). While vegetation is usually sparse in our study area and did not interfere, the optimal T-like orientation during laboratory trials is unlikely to be maintained in the field because the flexible wire antenna surely changes its relative position when a toadlet crosses vegetation or burrows.

These complex influences on the reflected signal's amplitude make tag location by harmonic direction finding a time consuming tracking method which requires a lot of manpower to collect quantitative data. As large detection ranges are rather the exception than the rule, the distance between parallel scanning paths should be 4-8 m to reduce the number of overlooked tags.

What can be learned from the field trial with free-ranging juvenile *B. calamita* and *B. viridis*? Reflector tags do not hinder juvenile toads to disperse more than 600 m within a maximum of four days, but most probably during two consecutive nights, i.e. HDF-tracking is well-suited to study short-time dispersal in the natural habitat. Available data on the velocity of dispersing juveniles of comparable size are scarce, but a maximum of 800 m per night in *Rana pipiens* (Dole, 1971) and 600 m within three weeks in *B. calamita* (Sinsch, 1997*a*) do not suggest that tags substantially modify dispersal behaviour. We do not know whether the velocity reported is representative for undisturbed toads or not because prior to experimental estimates toads were handled and displaced from their capture site. At least experimental displacement is known to increase distances covered above the normal level during the first days following release (Sinsch, 1987). In adult *Litoria lesueuri* the presence of a tag and/or of a waistband was also observed to increase the number of movements and distance covered per hour (Langkilde & Alford, 2002). Thus, migratory velocity observed in this study – and probably in several others – may tend to overestimate that of undisturbed anurans.

The significance of a recovery rate of only about 1/3 of all tags in both species remains to be discussed. Several factors probably contributed to the failure to detect the missing 2/3 of tags: (1) some toadlets may have moved further than the documented 665 m. However, the number of wide-dispersers is not expected to be large because the recovery rate exponentially decreased with distance from the release site. (2) Some detectable tags have been probably overlooked during the surveys as in the natural habitat the scanning path may have deviated from the intended search pattern. As an additional nine tags have been detected two years following the final release, the proportion of overlooked tags may account for about 10-20% of the missing ones. (3) Other tags probably became undetectable due to either broken diode-antenna connection, unsuitable antenna position or simply for being lost in deep burrows. As these toads usually burrow during daytime, we assume that most of the missing tags are still within the inner study area but buried too deep to be detected with a RECCO device.

In conclusion, despite all of the shortcomings of HDF tracking of small anurans, we encourage population ecologists to use this method to obtain reliable field estimates on the direction and distance of postmetamorphic dispersal. It is surely an improvement on current methodologies – although the gap between the detection ranges of active and passive tracking systems has become smaller, it remains large.

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