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published in
Ornis Fennica
2014

document version
Publisher's PDF, also known as Version of record

Ornis Fennica 91:244–255. 2014

Passive Integrated Transponders (PIT) on a small migratory passerine bird: absence of deleterious short and long-term effects

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Reviewed in Peerage of Science
Received 21 February 2014, accepted 18 July 2014

Passive Integrated Transponder (PIT) tags have been widely used for ornithological research. However, only few studies have reported their impacts on individuals. We assessed the efficacy of subcutaneous PIT tag implantation in the mantel area of adult Pied Flycatchers (*Ficedula hypoleuca*). In addition, we investigated the short-term effects of the tags on body mass changes during the breeding period, and potential long-term effects by examining local return rate and apparent survival over winter. We focused on the impacts of carrying PIT tags. We compared individuals with subcutaneously implanted PIT tags and individuals that had PIT tags implanted but lost their tags within days of implantation. Overall retention of subcutaneously implanted PIT tags was ~77% in both breeding males and females. The tag retention was influenced by implanter experience. Body mass changes of PIT tag implanted individuals during the breeding period did not differ from those individuals that lost the tag soon after implantation. The apparent survival of PIT tag implanted males and females did not differ from individuals without tags in previous years. Our results suggest that the retention of PIT tags on breeding adults may not adversely affect their body condition prior to migration and their apparent survival.

1. Introduction

Passive Integrated Transponder (PIT) tags are the key element in the application of Radio Frequency Identification (RFID) technology, primarily used for individual identification in many animal groups (Gibbons & Andrews 2004). A PIT tag is a small electromagnetic microchip encapsulated in a sealed glass tube which can be attached externally or implanted. These tags do not require a power source; a transceiver which generates an electromagnetic field is needed to activate the PIT tag after being attached or implanted. The unique alphanumeric code, emitted by each PIT tagged animal when passing close to a reader antenna, is detected and stored in a data logger. This technology enables gathering large quantities of accurate and reliable data, through a fully automated data collection system, while minimizing handling effects and observer effort. The long life of PIT tags and the relatively low cost of tags and readers (Bridge & Bonter 2011) minimize funding constraints in application to large-scale studies.

PIT tags have been widely used in ornithological research (see review: Bonter & Bridge 2011) including studies on provisioning and feeding rates (Kerry *et al.* 1993, Becker & Wendeln 1997, Boisvert & Sherry 2000, Freitag *et al.* 2001, Bley & Bessei 2008, Ringsby *et al.* 2009, Wilkin *et al.* 2009), incubation behavior (Cresswell *et al.* 2003, Zangmeister *et al.* 2009), pair formation (González-Solís *et al.* 1999), prospecting by breeding and non-breeding birds (Dittmann & Becker 2003, Dittmann *et al.* 2005, Thomson *et al.* 2013), survival (Becker *et al.* 2008a), temporal changes in body condition (Dittmann & Becker 2003, Limmer & Becker 2009), fledging (Johnson *et al.* 2013), post-fledgling movements (Michard *et al.* 1994, Nicolaus *et al.* 2008), dispersal (Becker *et al.* 2008b), homing behavior (Keiser *et al.* 2005) and identifying individual nests in a colony (Booms & McCaffery 2007). The use of PIT tags and applications for this technology are further likely to increase and it is therefore vital to ensure that any negative effects of these tags on individuals are negligible.

PIT tags can be either attached externally, to leg bands and feathers, or subcutaneously implanted. Both methods may have short-term and long-term impacts on the tagged birds. Externally attached PIT tags on leg bands are visible and no surgical procedures are required, however there is a high probability of tag loss (Bonter & Bridge 2011). Therefore, subcutaneous implants are generally preferred over external attachments due to better retention. On the downside, the procedure required for implantation is invasive and may cause harm to individuals. Furthermore, implanted PIT tags are invisible and often a portable reader is required to verify the presence of the tag. The location for subcutaneously implanted PIT tags varies between species (Bonter & Bridge 2011). For example, the back (Applegate *et al*. 2000, Jamison *et al*. 2000), the neck area (Clarke & Kerry 1998, Carver *et al*. 1999, Low *et al*. 2005) and pectoralis major in the belly area (Schroeder *et al*. 2011) have been identified as suitable locations to implant PIT tags. So far, no deleterious effects were found for large non-passerine birds when PIT tags were implanted subcutaneously (Clarke & Kerry 1998, Jamison *et al*. 2000, Gauthier-Clerc *et al*. 2004, Low *et al*. 2005). Yet, a few studies have reported or assessed the impacts of subcutaneously implanted tags on small passerines (Nicolaus *et al*. 2008, Schroeder *et al*. 2011).

In this study, we assessed the short and long-term impacts of subcutaneous PIT tags presence in Pied Flycatchers (*Ficedula hypoleuca*) during the breeding season. Short-term or long-term impacts of PIT tags on small migratory passerines during breeding have rarely been studied (Brewer *et al*. 2011). So far, studies that focused on small resident passerine species, PIT tags were either subcutaneously implanted or externally attached (Boisvert & Sherry 2000, Freitag *et al*. 2001, Wilkin *et al*. 2009) and no deleterious effects have been reported (Nicolaus *et al*. 2008, Brewer *et al.* 2011, Schroeder *et al*. 2011). Compared to resident species however, small migratory passerines go through rapid changes in their body condition prior to migration, and thus the effects of PIT tag insertion could be stronger. For example, Pied Flycatcher's lose 5–6% of their body mass during the breeding period (Slagsvold & Johansen 1998) and then accumulate the body mass during the migration (Schaub & Jenni 2000). Therefore, further studies on the short and long term-effects of PIT tags on migratory passerines are necessary to establish if PIT tags are free of harmful effects on small passerines.

Deleterious effects of PIT tags could be identified through two separate mechanisms. First, through the actual surgical implantation of the tags since this procedure may expose individuals to infectious diseases (Clarke & Kerry 1998). Second, through the movement (displacement) within the body of the subcutaneously implanted PIT tag (Clarke & Kerry 1998, Schroeder *et al*. 2011), that may impede locomotion and potentially damage tissues during their long distance flight. We focus on the second potential impact, as our study design does not permit controlling for the effect of the insertion procedure.

We firstly investigate the efficiency of the subcutaneous PIT tag implantation method described by Nicolaus *et al.* (2008) at mantel area. Secondly, we evaluate the specific effect of PIT tag retention on body mass change, as a measure of body condition and reproduction. Thirdly, we measured the long-term impact by examining apparent survival of PIT implanted individuals over winter.

2. Materials and methods

2.1. Species and study areas

The Pied Flycatcher is a small migratory passerine (12–13 g). European population of Pied Flycatchers winter in tropical West Africa (October–April) and are present in the breeding grounds from May to August (Lundberg & Alatalo 1992). The males arrive first and sing at natural cavities or available nest boxes. The females choose the nest sites or boxes, build the nests and lay 5 to 8 eggs. Only females incubate and the incubation period last for ~13 days. Both male and female perform parental care activities during the nestling period.

The data for this study were collected in two different study areas. Data testing short-term effects originates from a ~10,000 ha forest area in Kauhava, western Finland (63°10' N, 23°06' E), from May to July 2012 (hereafter, short-term impacts population).The forests consisted predominantly of Scots pine (*Pinus sylvestris*) interspersed with birch (*Betula spp*.) and spruce (*Picea abies*). Data for testing long-term effects originates from Ruissalo, south-west Finland (60°43' N, 22°16' E), during breeding seasons in 2005–2011 (hereafter long-term impacts population). The forests in Ruissalo are dominated by oak (*Quercus robur*) and Scots pine.

2.1.1. Short-term impacts population

A total of 140 nest boxes (11 × 11 × 17 cm; with 3.2 cm entrance hole) were placed in 35 different forest patches with minimum distance of ~600 m between two patches. Each forest patch contained four nest boxes placed at the corners of randomly selected 80 × 80 m square. The boxes were set up in early May 2012, before the arrival of Pied Flycatchers to the breeding sites (Lundberg & Alatalo 1992). We monitored all the nests from nest building to nestling phase approximately every 2–3 days. Females (N = 82) were captured at onset of incubation (mean ± SD: 14.9 ± 3.9 days from the first egg) while males (N = 50) were captured at the early nestling stage (26.4 ± 3.1 days from the first egg). All captured individuals were ringed with numbered metal tarsal rings, the body mass was measured using a digital scale (± 0.1g) and a PIT tag was subcutaneously inserted (see below). Tagged females were recaptured in the early chick-feeding phase (mean ± SD: 17.39 ± 3.46, range 5–23, median 18 days after insertion) and tagged males were recaptured in late chick-feeding stage (mean ± SD: 7.66 ± 2.15, range 5–19, median 7 days after insertion). Body mass was measured again and the presence of tags was checked thoroughly and later verified using PIT tag readers set up at the nest boxes.

2.1.2. Long-term impacts population

Approximately 230 nest boxes were permanently available to cavity nesting passerines. Each year (2005–2011), between 80 and 100 Pied Flycatcher pairs settled in these boxes for breeding. Nests were monitored every 4–5 days from the settlement period in early May, until chick fledging in July as part of ongoing long-term studies. In this population, females (N = 583) were captured during mid-incubation, while males (N = 551) were captured soon after arrival to the study site while prospecting nest boxes and also during the nestling phase. All captured individuals were ringed or their existing ring numbers noted. In 2010, PIT tags were subcutaneously implanted on all the birds captured. In all years the same procedure was used to capture and check the identity of individuals returning to the study population. In the year following PIT tag insertion (2011), all captured birds were also checked to detect PIT tag presence by using portable PIT tag reader.

2.2. PIT tag insertion

We used TROVAN ID® 100A Passive Integrated Transponders (2.12 × 11.5 mm, 0.1g) and an IID-102 implanter syringe to insert the tags. Tags were implanted subcutaneously following the method described in Nicolaus *et al*. (2008). Two people were involved in the process under field conditions: the handler held the bird firmly while the implantation procedure was carried out by the other person (hereafter implanter). Prior to insertion we wiped the feathers and skin close to the mantel area with cotton dipped in ethanol (80% ethyl alcohol). The skin was lifted with bulldog tissue forceps and then a small incision (2–3 mm) was made with the end of a sterilized needle containing the transponder attached to the implanter. The PIT tag was pressed gently underneath the skin through the incision and pushed forward using the blunt end of the bulldog tissue forceps. We sprayed two puffs of liquid plaster (Hansaplast Med®) to the wound opening and sealed the skin.

In the short-term impacts population, there were two field groups involved in PIT tag implantation, each group consisting of a naïve implanter and handler. In the long-term impacts population, all the implantations were done by an experienced implanter and handler. Implantation of PIT tags in both populations was done for the purpose of collecting provisioning and prospecting data of Pied Flycatchers.

In the short-term impacts population, we found that some birds had lost their PIT tags (see retention results). We used this loss of PIT tags as the control for our short-term investigation on the effects of tag-carrying birds. PIT tags were probably lost soon after the implantation (within 1–2 days), due to the movement of wings and dorsal muscles. In some birds, the inserted PIT tags partly protruded through the incision (callus was formed around the tag), but the wound had healed and the tag was secured on the bird ($N = 2$). These individuals were captured within 5 days of the tag implantation. This suggests that the healing of the small incision was rapid, probably because we used liquid-plaster which has faster wound healing properties ("moist wound healing technique"). Therefore, our assumption is that all PIT tag loss could only occur through the incision within 2 days of the initial subcutaneous implantation. From our data on PIT tag loss, this assumption appears valid. Therefore, in our test all individuals had PIT tags implanted, but those recaptured and found not to have retained PIT tags formed our control group of individuals (not carrying PIT tags), while individuals recaptured and found to still be carrying PIT tags were our treatment group of individuals (PIT tags retained).

This study was executed in accordance to Finnish Laws and regulations and under the approval of the animal experiment committee (permit number: ESAVI-2010-05480/Ym-23).

2.3. Statistical analyses

Firstly, we calculated the overall PIT tag retention for both males and females in the short-term impacts population. We used a generalized linear model with binomial error distribution with logit link to test whether implanter experience influenced the retention of PIT tags (coded as 1 or 0). In the full model, we included the following covariates: trapping session (a proxy of implanter experience which is expected to increase with the number of trapping sessions), sex of the bird, and naïve implanter's group (a factor with two levels "group A" and "group B"). Competing models were compared according to AIC values (Burnham & Anderson 2002) and results obtained from the best fitting model are reported.

Secondly, we investigated the effect of the presence of PIT tags (retained) on body mass change during the breeding period using generalized linear mixed model procedure with REML fit. We used body mass difference between initial capture (at the PIT injection) and recapture as our response variable. The males and females were generally captured and recaptured during the morning; therefore we did not include capture time in our analysis. Retention of PIT tags (i.e., a factor specifying whether individuals had retained their PIT tag or lost it soon after the insertion), individual sex, number of days between initial capture and recapture, clutch size and wing length (to control for body size) were used as covariates. The "site" was specified as random effect to account for potential pseudo-replication issues due to the nest boxes placed in the same forest patch. Again, all the competing models were ranked according to their AIC values and we report the results of the model that had the lowest value. All these analyses were performed using R version 3.0.2 for Windows (R Core Team 2013). The first analysis (implanter experience of PIT tag retention) was done by using the *glm* function and in the second

Table 1. Model summary of the generalized linear model with binomial error distribution which examined the effects of trapping session (implanter experience) on PIT tag retention of Pied Flycatchers. Parameter estimates were calculated using "group B" as a reference level and trapping session as predictor variable; two naïve implanter groups were named as "group A" and "group B".

Effect	Estimate ± SE	*z* value	*p*
Intercept	–0.60 ± 0.61	–0.985	n.s.
Trapping session	0.34 ± 0.16	2.125	< 0.05
Group (A)	1.62 ± 0.74	2.174	< 0.05
Trapping session × Group (A)	–0.26 ± 0.17	1.472	n.s.

analysis (PIT tag effect on body mass change) we used function *lmer* in the lme4 package (Bates *et al*. 2014).

Finally, we compared the effects of PIT tags on apparent survival of Pied Flycatchers in long-term impacts population. First, we used a two-sample test for equality of proportion with continuity correction to test whether there was a difference in local return rates (percentage of individuals recaptured over a winter) of the individuals with PIT tags in 2010 and individuals without PIT tags in previous years (2005–2009). This calculated local return rate was useful to compare our results with existing local return rates of Pied Flycatchers in similar latitude (Sanz 2001).

Second, we examined the effects of PIT tags on apparent survival (survival of individuals over a winter confounded by permanent emigration) for males and females using the Cormack-Jolly-Seber (CJS) model (Lebreton *et al*. 1992) in program MARK 7.2 (White & Burnham 2010). We assessed goodness-of-fit in U-CARE 2.3.2 for our global model ($\varphi_{(year)}\ p_{(year)}$, Choquet *et al*. 2009) where φ was apparent survival, and *p* was recapture probability. Heterogeneity due to "transience" or "trap-dependence" was negligible in our data set (Transience test 3.SR: standardized log odd-ratio $LOR_{male} = -0.219$, $P = 0.586$; $LOR_{female} = 2.302$, $P = 0.010$; Trap-dependence test 2.CT: $LOR_{male} = -0.281$, $P = 0.778$; $LOR_{female} = -0.627$, $P = 0.530$), however, there were overdispersion in both male and female data (median $\hat{c}$ values for males = 1.54, females = 2.80). We initially examined support for time-dependence in survival. We compared competing models nested within our global model using Akaike's Information Criteria with adjustment for small sample size (AIC_c) and overdispersion of data ($QAIC_c$) when necessary where $2 \geq \Delta AIC_c$ was considered to infer a difference in model support (Burnham & Anderson 2002). We also considered model selection uncertainty by model averaging the survival estimates. Then, we specifically test the effect of PIT tags; we applied parameter constraint using a dummy variable (1 = PIT, 0 = without PIT) to separate the PIT tag year (i.e., between 2010 and 2011) from years without PIT tags (2005–2010) in the design matrix (Cooch & White 2014). Here, we could not control for differing recapture rates between PIT tag classes because the PIT tag year was the last year in the data. Using time dependence or PIT tag classes would have resulted in problems with estimating the parameters for the last year (i.e., PIT tag year). Hence, constant recapture rates were used when examining

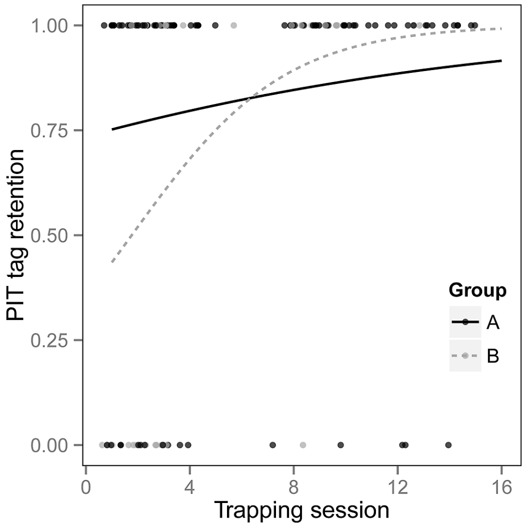

Fig. 1. PIT tag retention against the trapping session (implanter experience). PIT retention increased with the number of trapping session in both implanter groups (A and B).

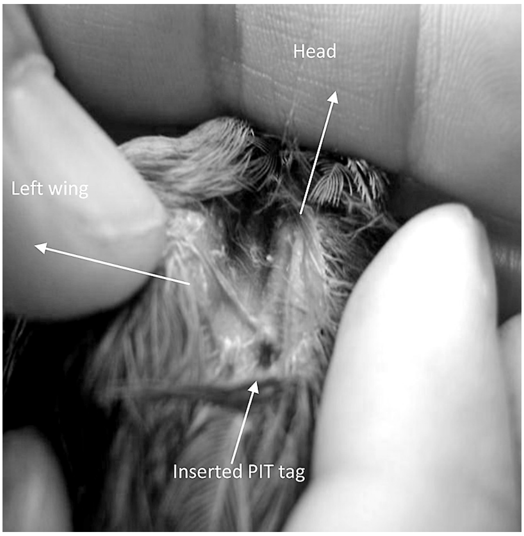

Fig. 2. Implanted PIT tag under the skin in the mantel area of Pied Flycatcher. The pattern of veins in mantel area may vary between individuals and thus researchers need to be cautious when inserting the implanter needle.

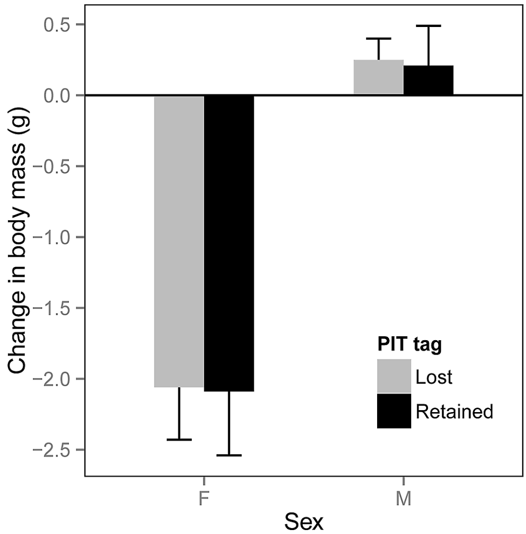

Fig. 3. Predicted body mass change of male and female pied flycatchers which retained PIT tags (dark bars) and lost PIT tags (light shaded bars) during the breeding period.

the effect of PIT tags on apparent survival, i.e., recapture rates were the same for both the PIT tag year and the non-PIT tag years.

3. Results

3.1. Tag retention

We captured and tagged a total of 132 individuals in the short-term impacts population, consisting of 50 males and 82 females. The overall PIT tag retention was 77.2% ($N = 102$); 12% ($N = 6$) of males and 29% ($N = 24$) females tags were lost during the study period. Overall, PIT tag retention increased with the trapping session (i.e. implanter experience). There was a difference in PIT tag retention between two naïve implanter groups (Table 1 and Fig. 1). We found that ~1% ($N = 1$) of PIT tags migrated to the side of the neck and ~2% ($N = 2$) tags came half way through the incision but the majority of the PIT tags (97%) did not move from the implanted mantel area (Fig. 2).

3.2. Body mass change

The body mass change of females with PIT tags (estimate ± SE: –2.09 ± 0.45 g, $N = 55$) did not dif-

Table 2. Summary of the linear mixed model analysis of the body mass change in male and female Pied Flycatchers implanted with PIT. Retention of PIT tags (factor specifying whether individuals lost or retained PIT tags), the sex, number of days between initial capture and recapture (recapture interval) were used as covariates. Forest patch (site) was specified as random effect for the intercept ("site" variance: 0.029; residual variance: 0.728). If the bootstrap confidence interval failed to include 0, then the *p*-value was deemed to less than or equal to 0.05.

Effect	Estimate ± SE	*t*	Lower	Upper
Intercept	–1.62 ± 0.19	–8.382	–1.99	–1.25
PIT tag (retained)	–0.04 ± 0.19	–0.200	–0.39	0.32
Sex (male)	1.15 ± 0.29	3.891	0.56	1.72
Recapture interval	–0.67 ± 0.14	–4.646	–0.95	–0.38

Table 3. Summary of model selection criteria for males and females used to determine competing models to estimate apparent survival of Pied Flycatchers in long-term impact population. The time-dependent survival with constant recapture probability model [$\varphi_{(year)}\ p_{(.)}$] was used to estimate apparent survival of birds with and without PIT tags. Akaike's Information Criterion adjusted for small sample sizes and overdispersion ($QAIC_c$), model deviance, number of parameters (N. Pr) arranged ascending by $QAIC_c$ value.

Model	$QAIC_c$	$\Delta QAIC_c$	Akaike weight	N.Pr	Model deviance
Males (median $\hat{c}$ = 1.54 corrected)					
$\varphi_{(year)}\ p_{(.)}$	473.22	0.00	0.84	7	31.15
$\varphi_{(.)}\ p_{(.)}$	478.54	5.32	0.06	2	46.65
$\varphi_{(PIT)}\ p_{(.)}$	479.35	6.13	0.04	3	45.45
$\varphi_{(year)}\ p_{(year)}$	479.80	6.58	0.03	11	29.46
$\varphi_{(.)}\ p_{(year)}$	480.43	7.21	0.02	7	38.36
Females (median $\hat{c}$ = 2.80 corrected)					
$\varphi_{(.)}\ p_{(.)}$	215.90	0.00	0.49	2	22.64
$\varphi_{(PIT)}\ p_{(.)}$	217.34	1.44	0.24	3	22.06
$\varphi_{(year)}\ p_{(.)}$	218.21	2.30	0.15	7	14.78
$\varphi_{(.)}\ p_{(year)}$	219.40	3.50	0.08	7	15.96
$\varphi_{(year)}\ p_{(year)}$	222.92	7.02	0.01	11	11.22

fer from females that lost PIT tags (estimate ± SE: –2.06 ± 0.37 g, N = 22; Fig. 3, Table 2). Males also did not show a difference in body mass change between the males carrying PIT tags (estimate ± SE: 0.21 ± 0.28 g, N = 41) and the males that lost tags (estimate ± SE: 0.25 ±0.15 g, N = 5; Fig. 3, Table 2). Overall, the body mass of females was decreased during the early incubation period to early nestling period whereas in males the body mass was increased early nestling period to late nestling period (Fig. 3).

3.3. Local return rate and apparent survival

In the long-term impacts population, a total of 483 males and 505 females were captured, marked and released over a five-year period (2005–2010). Of these, 124 males and 79 females were re-captured in the following years. In 2010, a total of 68 males and 78 females were implanted with PIT tags. Of these individuals, 15 (22%) males and 11 females (14%) returned to breed in 2011. The local return rates, 26.5% for males (two-sample proportion test, χ^2 = 0.126, P = 0.721) and 18.4% for females (two-sample proportion test; χ^2 = 0.018, P = 0.893), did not differ from the average local return rates over the five-year period (2005–2010).

We found good support for the time-dependent survival model with constant recapture probability for males, but not for females (Table 3). Model averaged apparent survival of males and females varied between 0.16 and 0.48; the PIT tag year being in the middle of the range (Fig 4A). Accordingly, in both sexes the additive model (with PIT tag) did not receive more support than the constant model; these models received roughly equal support (Likelihood Ratio test for males: χ^2 = 1.207, df = 1, P = 0.272; females: χ^2 = 0.579, df = 1, P = 0.446; Table 3). We used time-dependent survival with constant recapture probability models for both males and females to apply parameter constraint. The effect of the PIT tags in additive models were negative, but confidence intervals of the regression for both males and females exceeded zero (males: $\beta_{\text{PIT TAG}}$ = –0.44, 95% CI = –1.26–0.36; females: $\beta_{\text{PIT TAG}}$ = –0.51, 95% CI = –1.84–0.81). The model averaged apparent survival estimates of PIT tag carrying individuals were not different from the survival of non-PIT tagged individuals in both males (PIT tags (2010): estimate ± SE; 0.29 ± 0.04; no PIT tags: 0.33 ± 0.03; Fig.4B) or in females (PIT tags (2010): estimate ± SE; 0.27 ± 0.07; no PIT tags (2005–2010): 0.31 ± 0.05; Fig.4B). Hence, our data suggests no clear effect of PIT tags on apparent survival.

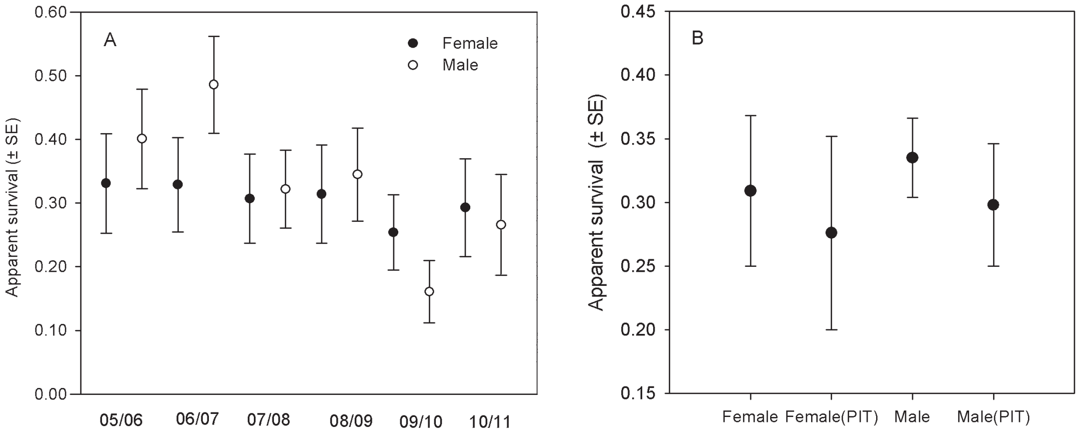

Fig. 4. Model averaged apparent survival estimates (± SE), A) for both Pied Flycatcher males and females in long-term impact population (survival interval indicated between years i.e., 05/06 for 2005–2006); B) model averaged apparent survival estimates of males and females with PIT tags did not differ from males and females without PIT tags (parameter constraint was applied to the model [$\varphi_{(year)}\ p_{(.)}$] by specifying a dummy variable: PIT tag year as 1, and non-PIT tag years as 0 in both cases).

4. Discussion

4.1. Short and long-term impacts of PIT tag implants

Our analyses revealed no measurable negative short-term or long-term effects on Pied Flycatcher individuals that retained subcutaneously implanted PIT tags. First, there was no effect on the body mass changes of males and females. Second, no substantial long-term effect was detected, as the estimated apparent survival of both males and females over five year period were not different from the estimated survival in the year after PIT tag insertion. In addition, our results also show that implanter experience has a significant impact on PIT tag retention.

Carrying subcutaneously implanted PIT tags did not significantly affect body mass change of females during the incubation and males during the nestling period. Our study therefore suggests that carrying a PIT tag does not significantly alter the overall condition of individuals since it does not cause critical changes in body mass. Observed body mass losses in our study were consistent with patterns well documented for this species during breeding (Freed 1981, Moreno 1989, Hillström 1995, Thomson *et al.* 2010). The body mass also varies during the day time in small passerines (Baldwin & Kendeigh 1938); hence analyses based only on capture or recapture body masses and need to correct for diurnal variation. In this study, however, we used body mass changes, between minimum five days to maximum 23 days period, which requires no correction for capture and recapture time. Nevertheless, small impacts of PIT tag presence during the breeding season, or during the refuelling period prior to migration, may become detectable in longer-term survival and return rates.

We found that the return rates of flycatchers carrying PIT tags were not different from the annual local return rate of individuals without the tag in the studied population. The return rate of females (14%) with PIT tags was similar to annual female local return rates found at the same latitude in a previous study (13.6%, see comparative data: Sanz 2001). There may be a substantial number of birds that disperse from the site in subsequent breeding seasons, and are thus undetected (Marshall *et al.* 2004).

We assume that dispersal propensity and detection in the study site are the same for individuals with and without PIT tags. However, there may be several other factors that impact local return rates of Pied Flycatchers, for example local immigration (Chernetsov *et al.* 2009), environmental pollution (Eeva *et al.* 2006) and weather conditions in wintering ground (Laaksonen *et al.* 2006). The main caveat of this study is absence of the same year control birds without PIT tags; we need to be cautious about the inference from our study. From our results, it seems that year 2011 could be a normal year despite the tags presence so the possible deleterious effects on the apparent survival are not extreme. Given the large variation of apparent survival between years we also cannot rule out that the individuals from year 2011 show high apparent survival even without the PIT tags.

Our data was sourced via field work not specifically designed to test the impacts of PIT tags. Therefore while our study provides evidence that there were no substantial negative impacts of carrying PIT tags in a small migratory passerine, we cannot completely discount the possibility that PIT tags may harm birds (see studies on geolocators: Arlt *et al*. 2013; Gómez *et al*. 2014). However, our study has a novel aspect in focusing on the effects of carrying the PIT tag during breeding and subsequent migration while controlling for the invasive procedure to subcutaneously implant the tags. Our controls are unique; these individuals experienced the surgical implantation process, but lost tags shortly afterwards and were without the potential burden of carrying PIT tags. An experimental design simulating this control would have ethical considerations, and our detailed approach of following the fate of PIT tags during our study facilitated using such a control group.

4.2. Implantation process and tag location

Successful PIT tag retention depends on the experience and attachment method. In our study, once the implanter's experience improved, retention increased from 69% (in the first five trapping sessions) to 88% (the last five trapping sessions). The percentage retention in our study birds (~77%) was higher than in previous studies in non-passerines species: the 30% for Adélie Penguins (*Pygoscelis adeliae*); (Clarke & Kerry 1998), and the 59% for Common Terns (*Sterna hirundo*); (Becker & Wendeln 1997). In these species, tag losses were reduced using adhesive glue and changing location of insertion (Bonter & Bridge 2011), whereas in our study we used liquid-plaster for better healing and tag losses were reduced through improved implanter experience. Increased retention with increasing implanter experience was expected. Our study, however, provides an estimate of the time and the number of individuals required to increase the retention that would be useful when this technique is applied to other similar passerine species.

The PIT tag implantation technique here used, originally described in Nicolaus *et al*. 2008, appears to be suitable for small migratory passerines like the Pied Flycatcher (see: Fig. 2). Indeed, the mantel area, close to the neck, seems a good place to implant PIT tags in small passerines as retention was ~77% and low PIT tag migration within the body (~3%) in our study. In previous studies on small passerines, PIT tags were implanted in the abdominal area close to pectoralis major; neck and lower back (see Introduction). For example in House Sparrow (*Passer domesticus*) PITs were generally implanted to both the abdominal (pectoralis major) and the neck area, but it has been suggested the neck area is more suitable than pectoralis major due to lower risk of PIT losses (Schroeder *et al*. 2011). Intramuscular implantation is also recommended, alternative to pectoralis major, to decrease the chances of PIT tag loss (Elbin & Burger 1994). In Common Tern (*Sterna hirundo*) PITs implanted at the back were lost more than the PITs implanted at breast area due to the extensive preening behavior (Becker & Wendeln 1997). Therefore, choice of the location and the individual behavior are equally important to attain better retention.

The size of the incision is also important because the inserted PIT tags occasionally move through the opening, though it is sealed with liquid-plaster, because of the rapid movement of the wings and the associated muscles. Therefore, the needle size (e.g., N125 with MK10 syringe in Biomark® products) should be carefully selected as to make a small incision via which small PIT tags can be transferred under the skin. The opening and the skin should be closed properly and either glued with topical adhesive or liquid plaster soon after implanting the PIT tag. Furthermore, we observed individual differences in vein patterns of Pied Flycatchers in the skin in mantel area which can cause difficulties when implanting PIT tags in some individuals.

Researchers should test the potential impacts of the marking methods they are using and PIT tags are no exception. However, despite being a commonly used technique, only few studies have tested such impacts. Sharing information on the implantation process is useful to ensure this methodology improves and minimizes problems for groups planning future studies using PIT tags. Our results are therefore a useful addition to the existing literature testing the impacts of PIT tags in avian studies. We also encourage more studies that investigate the impacts of PIT tags even within the broader scope of other research questions.

Acknowledgements. We are particularly thankful to the following researchers for their work in Ruissalo, and for providing us their unpublished data: Toni Laaksonen, William Velmala, Sara Calhim, Wiebke Schuett and Päivi Sirkiä. We thank Ryan Miller, Jorma Nurmi, Elina Ode, Stefan Siivonen, MarjoTuronen, Rauno Varjonen and Ville Vasko for their help in the field in Kauhava. Prithiviraj Fernando, Jeniffer Pasteroni, Lise Ruffino and Veli-Matti Pakanen provided the technical information and valuable comments on the manuscript. Funding for this project was provided by the Academy of Finland project # 138049 to RLT. The Center for Conservation and Research-CCR in Sri Lanka provided logistic support while preparing the manuscript. Funding was provided to CPR by Erasmus Mundus EXPERT programme.

Passiivisten RFID-sirujen hyödyntäminen muuttavilla varpuslinnuilla: ei haitallisia lyhyt- tai pitkäaikaisvaikutuksia

Passiivisia RFID-siruja on viime aikoina käytetty paljon lintutieteellisessä tutkimuksessa. Kuitenkin vain harvat tutkimukset ovat raportoineet sirujen vaikutuksia yksilöihin. Tutkimme kirjosiepolla (*Ficedula hypoleuca*) niskan alueelle sijoitetun ihonalaisen mikrosiruimplantin tehokkuutta menetelmänä. Lisäksi tutkimme mikrosirujen lyhytaikaisia vaikutuksia lintujen pesimäajan painonmuutoksiin ja mahdollisia pitkäaikaisia vaikutuksia paluun ja hengissä selviämisen todennäköisyyksiin.

Vertasimme lintuyksilöitä, jotka säilyttivät mikrosirut ja joilta sirut putosivat pian toimenpiteen jälkeen. Sekä koiralla että naarailla 77 % siruista säilyi paikallaan ja säilyvyyteen vaikutti toimenpiteen suorittajan kokemus. Ryhmällä, jossa siru säilyi paikallaan, pesimäajan painonmuutokset olivat samanlaisia kuin ryhmällä, jotka menettivät sirut. Sirutettujen koiraiden ja naaraiden hengissä selviämisen todennäköisyys oli samanlainen kuin edellisinä vuosina yksilöillä, joilla ei ollut siruja. Tuloksemme viittaavat siihen ettei mikrosiru vaikuta haitallisesti pesivien lintujen kuntoon tai hengissä selviämiseen.

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