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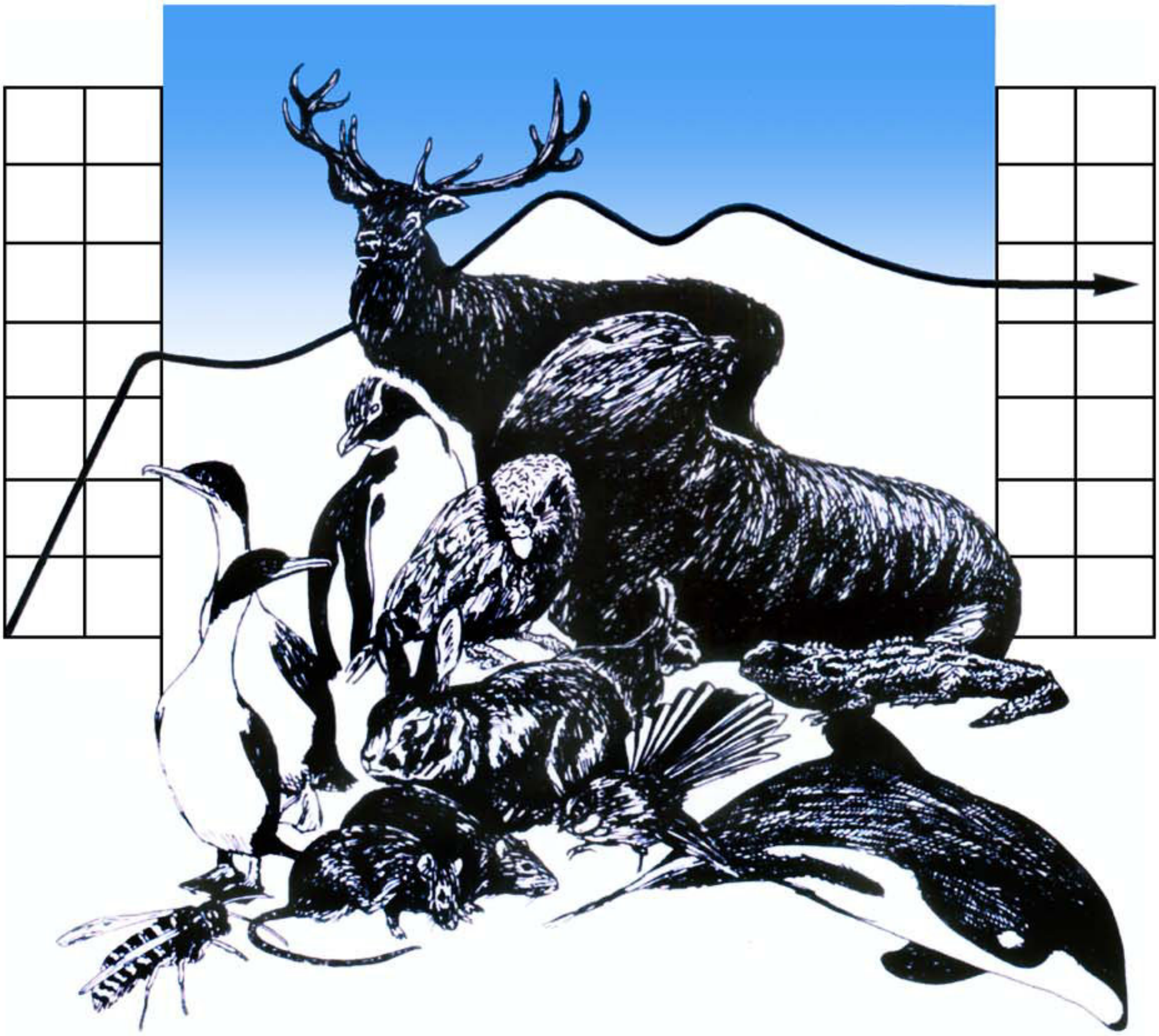

WILDLIFE MANAGEMENT

Are transponders a reliable primary mark for yellow-eyed penguins?

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Are transponders a reliable primary mark for yellow-eyed penguins?

A report on the effectiveness of current transponder use in yellow-eyed penguins, for the Department of Conservation and associated agencies

April 2014

Wildlife Management Report

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Executive summary

Yellow-eyed penguins (*Megadyptes antipodes*) or *hōiho* were monitored and surveyed on the Otago coastline over a two month period during the austral spring of 2013. The purpose of this was to clarify and determine whether there were issues with the current monitoring set up of yellow-eyed penguins, across the dual marked population (marked with both passive integrated transponders (PIT) and flipper bands), as well as to determine whether PIT tag transponders are reliable enough to be the primary marking technique used on the species.

Twenty nine dual marked yellow-eyed penguins, across 66 nests had their PIT tags identified by two models of Allflex transponder readers (RS200, RS320) for a total of 117 reading events. They were scanned multiple times across two different anatomical planes (parallel / perpendicular or coronal / transverse) to detect whether or not there was a significant difference between the method used and successful identification of the PIT tags.

It was discovered our PIT tag detection results compared to numerous studies and privately contacted authors were markedly different to what was expected. Although it was found that the correct identification of the PIT tags being identified was not an issue (i.e. all 29 dual marked birds were identified by their PIT tag at least once during the study); the rate of which these PIT tags being successfully detected was. Our results only showed a quarter (~25%) of the ability to detect PIT tags per scan than expected elsewhere (95-100% / scan).

On top of this, c. 5% of PIT tags were not detected when they were in fact present and active inside the individual. Combined these are serious issues as it may take up to four scans to determine the identity of a bird, meaning that increased time and effort would need be spent per nest visit, further increasing the anthropogenic stress caused to the individual during an already stressful time. If PIT tags are not detected when they are actually present this could also lead to inaccurate or incorrect information being recorded affecting proper identification as well as population modelling parameters.

To determine whether PIT tags are the best and most cost-efficient method for monitoring yellow-eyed penguins, it will be important to track the successfulness of the new transponder reader model (RS420), while also researching other potential marking methods and techniques that could be used along side or instead of PIT tags (and flipper bands).

Also within this document:

- A detailed account of the current set up and how individual yellow-eyed penguins are identified using PIT transponders.

- A pros and cons list of using this technology, while stating and comparing the current issues found with our system with other studies from around the world using similar equipment.

1. Introduction

Marking animals either externally or internally is integral to the research and management of wildlife populations (Beausoleil *et al.* 2004). Without the ability to identify individuals, scientists and conservation managers would struggle to actively study and monitor many species and wild populations due to their phenotypic similarities. However, all animal marking techniques have costs, whether they are financial, logistical or ethical (Petersen *et al.* 2006), and there is a need to balance out the potential information and insights gained, over the negative impacts caused by marking, handling and disturbing individuals (Dugger *et al*. 2006). In other terms, it has to be determined whether the information obtained from marking is worth the risk and associated consequences placed on the marked individual (Beausoleil *et al.* 2004).

Crucial information such as lifetime reproductive success, survival, foraging ecology, diet, dispersal and migration can only be studied and obtained if animals can be individually identified (Parker & Rankin 2003; Gibbons & Andrews 2004; Maho *et al.* 2011).Throughout the decades there have been considerable advancements in marking technology, and numerous refinements in procedures to best and most efficiently mark animal species (Beausoleil *et al.* 2004). However, factors such as body shape, size and mobility, all influence the decision on how to mark a species or population, and thus marking technique has to be carefully chosen on a case by case basis (Beausoleil *et al.* 2004).

1.1. Passive integrated transponders as a primary mark for wildlife management

Since the mid-1980s passive integrated transponders (PIT) have been used in hundreds of studies of vertebrates, from mammals and reptiles to amphibians and fish, and are now also compact enough for use on numerous large invertebrate species (Buhlmann & Tuberville 1998; Gibbons & Andrews 2004; Hill *et al.* 2006). PIT tags are commonly referred to as transponders, microchips and radio frequency identification tags (RFID). PIT tags are small electronic units containing an antenna coil, capacitor and circuit board encased in a small, biologically inert glass or resin capsule (Jansen *et al.* 1999; Roussel *et al.* 2000). Most PIT tags have a diameter of ~2mm but can vary in length from <11mm to over 32mm depending on their capabilities and various other factors (Beausoleil *et al.* 2004).

PIT tags are most commonly inserted subcutaneously or intra-abdominally with a hollow needle, taking less than a minute to complete the procedure (Klindtworth 1998; DOC 2012). PIT tags can also be attached to an animal externally (Hill *et al.* 2006), although recent studies have shown that this can lead to higher frequencies of tag loss (Schroeder *et al.* 2010), as well as harmful changes in behaviour and fitness in numerous bird species (Jamieson *et al.* 2000; Gauthier-Clerc *et al.* 2004).

PIT tags do not contain their own power source or memory, and only transmit a unique identification number or code when held in an electromagnetic field produced by an appropriate transceiver (Prentice *et al.* 1990; Gibbons & Andrews 2004; Fielder 2011). Due to their size and obscurity once properly inserted they are ideal marks for use in long term studies where other forms of tagging or marking are inappropriate; this is especially noted in long lived, marine birds where an external mark may reduce swimming or diving efficiency (Culik 1994; Buhlmann & Tuberville 1998; Gibbons & Andrews 2004). The first published study of PIT tag usage in penguins occurred in 1992 in king penguins (*Aptenodytes patagonicus*) (Gendner *et al.* 1992), and are still currently being used for many penguin studies today, including the yellow-eyed penguin (*Megadyptes antipodes*).

1.2 Yellow-eyed penguins and marking of individuals

Since the landmark population study undertaken by Richdale (1951), yellow-eyed penguins have been marked in numerous ways for individual identification (McKinlay 2001). Metal leg bands were first applied as an identification mark by Richdale. However later studies in the 1970s to the present day use aluminium or stainless steel flipper bands due to the leg and lower body injuries that the metal leg bands caused (McKinlay 2001). Webtags have been used to mark first-born chicks at study sites on the Otago Peninsula and in the Catlins, however these were removed before fledging because of the risk of tag loss and ripping of foot webbing (J.T. Darby, pers. comm. 2013). Yellow-eyed penguins and their chicks are still flipper banded at sites where lifetime reproductive success information is collected, and when individuals are released from rehabilitation centres (12 to 14 breeding sites in Otago; M.J. Young pers. comm. 2014).

Over the past decade PIT tags have been introduced as a primary mark for yellow-eyed penguins on Codfish Island, Enderby Island, Stewart Island and specific areas on the Otago coast. As of 2012, over 800 PIT tags have been implanted in yellow-eyed penguins by the Yellow-eyed Penguin Trust (YEPT), the Katiki Point Charitable Penguin Trust (KPPT) and the Department of Conservation (DOC) (DOC 2012). A small proportion of these PIT-tagged yellow-eyed penguins are dual marked with a flipper band. At last count, 94 of the 346 “rebanded” birds (i.e. individuals that have had their identification mark changed) were both flipper banded and PIT tagged; with the vast majority (87/94) being from the Otago coast (DOC, unpublished data). Data from these dual marked individuals may be of use to conservation managers to determine the degree of tag loss for either marking technique.

Although the techniques for transponder insertion have been refined in terms of best practice, there have been reoccurring issues noted by users of PIT tags, which include: failure to read some tags (i.e. “faulty” tags), failure of the readers to detect implanted PIT tags, increased disturbance of yellow-eyed penguins that do not have an external mark, and reliance on manual detection of PIT tags. At present no studies of tag loss or failure have been undertaken that are specific to the PIT tag hardware being used on yellow-eyed penguins. It appears that the majority of problems relate to the manual detection of transponders using a range of handheld readers, rather than the transponders falling out of the animals post-insertion. The agencies involved seem to be no closer in using these tags effectively to save time, effort and cost compared to the information they were already obtaining from flipper banded birds.

1.3 Identifying the context for marking and studying yellow-eyed penguins

Yellow-eyed penguins are one of the rarest species of penguin on the planet (Ellenberg & Mattern 2012). They are classified as "Endangered (EN) B2b(iii)+c(iv)" by the World Conservation Union (IUCN) (Seddon *et al.* 2012, unpublished); and are listed as threatened (Nationally Vulnerable) by the New Zealand Threat Classification System (Miskelly *et al.* 2008). This, along with the recent and unexplained die-off of at least 58 adult birds, five juveniles and four chicks on the Otago Peninsula alone, as well as other declines throughout their range (Ellenberg & Mattern 2012, DOC, unpublished data); demonstrate that it is now more vital than ever that yellow-eyed penguins are studied and appropriately monitored to ensure their continued survival and that the correct conservation actions are taken for their benefit.

Not only is the decision to monitor yellow-eyed penguins beneficial for this species alone, but potentially for the local marine ecosystem as a whole. Penguins are excellent bio-indicators of change in aquatic environments, as seen in the CCAMLR Ecosystem Monitoring Programme (Gendner *et al.* 2005; Petersen *et al.* 2006; Boersma 2008). Hence their monitoring could be the best and one of the more cost efficient way of maintaining one of New Zealand's best assets: our shoreline and coastal waters.

1.4 Known problems with the use of transponders as a primary identification mark

Although known to occur, few published wildlife management studies comment on failures or fault with transponder technology. The most recent and best example to date, is a study by Dann *et al.* (2013) who commented that transponder failure was c. 5% in the first year of implantation in free-living little penguins (*Eudyptula minor*), with gradual reductions as time went on. Hence due to lack of study whether or not this result is common in penguins or other species is therefore up for debate. Becaus eof this information to compare and contrast to our yellow-eyed penguin transponder results had to be collected by privately contacting authors from published literature. Rates of failure noted by researchers from several published and unpublished studies of transponder use are displayed on the next page in Table 1.

Table 1: Noted results and anecdotal accounts of transponder failure rates (from around the world).

Species	**Transponder type**	**Reader type**	**Failure rate % (# failed/# total)**	**Source**
Adelie penguins (Pygoscelis adeliae)	???	Automated weigh bridge	c. 11% (was up to 27%)	K. Newbery pers. comm. 2014
Kakapo (Strigops habroptilus)	???	Allflex handheld reader	No issues over past 14 years	J. Ledington pers. comm. 2014
Little penguins (Eudyptula minor)	Trovan	Automated	c. 5% (during first year, and ~1% after first year)	Dann et al. 2013
Little penguins (Eudyptula minor)	Allflex	A static DOC automated reader Handheld Allflex reader	Small number of failures noted, but number not recorded	M. Rumble pers. comm. 2014
New Zealand sea lions (Phocarctos hookeri)	Trovan	Handheld reader	c. 11% (transponders fell out). Issues in only 2 adults.	L. Chilvers pers. comm. 2014
Numerous animal species	Mostly used Allflex gear	Mostly used Allflex gear	No records kept, noted always certain individuals which are difficult to detect	J. Clarke pers. comm. 2014
Southern rockhopper penguins (Eudyptes chrysocome)	TRIS - Allflex (23mm)	Automated gateway system	No PIT tags could be read by system (had to change tags)	N. Dehnhard pers. comm. 2014
Southern rockhopper penguins (Eudyptes chrysocome)	RFID - Texas Instruments (23mm)	Automated gateway system Allflex RS320	c.0.6% (1/166) (transponder loss)	N. Dehnhard pers. comm. 2014; P. Maud pers. comm. 2014
Taikos (Pterodroma magenta)	TRIS - Allflex (11mm)	Automated gateway system	c. 2.5% (4/160) (different issues with tags)	G. Taylor pers. comm. 2014
Yellow-eyed penguins (Megadyptes antipodes)	TIRIS - Allflex (23mm)	Allflex RS200 reader	c. 5% (4/80) (failed immediately after insertion)	M.J. Young pers. comm. 2014

Although PIT tag and transceiver technology has improved over the years, they still have issues which might make other forms of animal identification more appropriate or reliable

(Buhlmann & Tuberville 1998; Beausoleil *et al.* 2004). The pros and cons of PIT tags are mentioned below in Table 2.

Table 2: Lists of pros and cons of transponders as individual marks for wildlife management.

PROS	**CONS**
Reliability of tag detection is considered high	PIT tags and hardware are an expensive and ongoing cost
Reliability of reading accuracy is considered high	Without an external mark, other forms of reporting of marked individuals become obsolete
May reduce human error if tag numbers are stored and transferred electronically	Manual detection of all animals requires more disturbance to determine presence/absence of a PIT tag
Internal and permanent mark, protected by the body	PIT tags are known to fail post-insertion, migrate to other parts of the body, or may fall out of the body
Internal mark does not negatively affect survival or breeding performance	The supporting reader hardware can fail, either due to power or antenna loss
Tags are tolerant of a wide temperature range	Transcription errors may occur if tag detection systems do not store tag numbers
Automated tag detection may reduce capture events and yield larger numbers of detection events	PIT tags may change their orientation for reduced performance

Advantages of using PIT tags

Reliability of tag detection is considered high

One of the main advantages of using PIT tags is their high reliability of tag detection and reading accuracy. Although there are exceptions, Gibbons & Andrews (2004) found that most studies had a tag detection rate of 95-100% every scan, and a reading accuracy rate of ~100%. Since this process is electronic it almost entirely eliminates the possibility of human error when properly recording data.

Internal and permanent mark, protected by the body

PIT tags are internal and permanent. If they are implanted correctly they should last as long as animal itself (Buhlmann & Tuberville 1998; Gibbons & Andrews 2004). The procedure of implanting a PIT tag itself is relatively quick, and the birds do not seem to be too affected by the treatment (M.J. Young pers. comm. 2013; Dann *et al.* 2013). The surface of glass PIT tags are roughened to prevent them slipping out during the healing process, and in some cases, tissue glue is used to close the hole left by the needle to prevent early loss of the transponder.

Due to the protection provided by the animals' body, and the construction of PIT tags themselves, PIT tags rarely break. Conill *et al.* (2000) found that out of 686 PIT tags only 0.4% of broke or stopped working throughout the study period. One PIT tag found in a dead yellow-eyed penguin broke after collection, but the data was able to be recovered (YEPT 2011).

Internal mark does not negatively affect survival or breeding performance

If the animals are of an adequate size and ecological activities are not inhibited by the tagging and monitoring process, there is virtually no direct evidence of PIT tag induced impacts occurring in marked individuals (Gibbons & Andrews 2004). Unlike some external marks, studies have shown that PIT tags show little to no impact influencing growth rates (Low *et al.* 2005), mating performance (Watanuki *et al.* 1994; Gibbons & Andrews 2004), predator susceptibility (Culik *et al.* 1993, Petersen *et al.* 2006) or swimming speed and energy expenditure (Culik & Wilson 1991; Culik *et al.* 1993; Culik *et al.* 1994; Banasch *et al.* 1994), although these results have mostly been detected in captive birds. A study by Schroeder *et al.* (2010) found no evidence of adverse effects occurring from PIT tags on smaller bird species.

Tags are tolerant of a wide temperature range

PIT tags function equally as well under extreme hot and cold conditions, compared to other marking types (Gibbons & Andrews 2004). Though this is not especially important for yellow-eyed penguins and our local, temperate climate.

Automated tag detection may reduce capture events and yield larger numbers of detection events

PIT tags can be used with automatic scanning stations, weighbridges or pathways. Automated stations can not only continuously detect PIT tags of animals that move close enough to the antenna, but can also gather other important such as bird movements, time spent away from nest, as well as recording weights (Gendner *et al.* 2005; Fiedler 2011). Although these automatic scanning stations vary greatly in specifications and cost, they can reduce the disturbance and stress posed by manual detection of transponders by humans on the tagged animals, as there is no recapture and they offer a high detectability rate (Beausoleil *et al.* 2004; Gibbons & Andrews 2004; Gendner *et al.* 2005).

Disadvantages of using PIT tags:

PIT tags and hardware are an expensive and ongoing cost

As it is with a lot of practical applications of technology, whether or not PIT tags systems are a viable option may be due to budget constraints. Establishing a long-term identification system using PIT tags is significantly more expensive than most other marking methods, such as metal bands, tags or branding equipment, for example (Beausoleil *et al.* 2004; Fiedler 2011). In addition to the bulk amount of PIT tags that need to be purchased, multiple and relatively expensive reading apparatus need to be purchased for cost-effective and efficient nest monitoring (Beausoleil *et al.* 2004; M.J. Young pers. comm. 2013). In addition, data collection incurs a human cost in the form of increase time and wages, unless automated systems have been established.

Without an external mark, other forms of reporting of marked individuals become obsolete

PIT tags also result in fewer recoveries of birds made by the public in contrast to a visible, readable external mark (Dann *et al.* 2013). A study of yellow-eyed penguins caught in set nets indicated that birds with an external mark were more likely to be reported as dead than unmarked birds (Darby & Dawson 2000). If a transponder fails or falls out of an animal and it has no other external mark, there is a chance that it will be re-marked, thereby skewing population estimates.

Manual detection of all animals requires more disturbance to determine presence/absence of a PIT tag

Unlike external marks, which can be identified from a distance, internal marks such as PIT tags cannot. Hence if the PIT tags have to be detected manually, a person has to get close enough to the animal to scan and record its identity (usually <30cm away, but as close as 5cm depending on the type of scanner (Beausoleil *et al.* 2004; Maho *et al.* 2011). As stated earlier, these periodic recaptures can negatively affect tagged animals, especially around breeding time, and such negative impacts caused by human-induced stress have been observed in yellow-eyed penguins (Ellenberg *et al*. 2007; Ellenberg *et al*. 2009; P.J. Seddon and M.J Young pers. comm. 2013). External marks such as bands can be read unobtrusively from a distance using binoculars or by photograph, meaning, not all animals will need to be disturbed to determine their identity. Automatic scanners largely reduce the anthropogenic impact placed on tagged individuals; however PIT tags still have to be within range of an antenna for a successful PIT tag read (c. 1m; Fiedler *et al.* 2011).

PIT tags are known to fail post-insertion, migrate to other parts of the body, or may fall out of the body

Although not often recorded in literature, PIT tag failure can and does occur (Buhlmann & Tuberville 1998). Tag failure often occurs in two distinct ways: as mentioned previously equipment failure (breakages), and through tag loss. Due to PIT tags and readers being completely electronic, the devices are liable to faults and may eventually stop working, or not work at all from the start. Since PIT tags cannot be scanned pre-insertion, due to the metal needle short-circuiting the electric field or the thickness of the needle sheath preventing a read (M.J. Young pers. comm. 2013; K. Newbery pers. comm. 2014). It is only immediately after implantation that tags can be checked to see whether they are active or not. While no studies published were found to have looked directly at the equipment failure, PIT tag read failure or how and why it occurs; it is thought that it is during the implantation process that the majority of damage seems to occur (Conill *et al*. 1994; Sutterluery 1996).

However the loss of PIT tags from the body has been better documented than equipment failure rates in published literature; and is also thought to be primarily caused by inadequate implantation practices (Prentice *et al.* 1990; Freeland & Fry 1995). Loss rates can dramatically vary between bird species and implantation procedures, results from 1 to 4% (Hindell *et al*. 1996; Clarke & Kerry 1998; Gauthier-Clerc *et al*. 2004; Dann *et al*. 2010), up to 30% in penguins have been observed (Clarke & Kerry 1998), and can trend all the way up to 41% in other bird species (Becker & Wendeln 1997).Tag loss rates of ~30% have also been found in the New Zealand sea lion (B.L. Chilvers pers. comm. 2104).

Although it is hard to compare PIT tag loss rates across different species due to such factors as lack of data, and differences between implantation practices, it is generally assumed that PIT tag loss is dependent on three factors: depth of PIT insertion; the location and site of insertion, and; post implantation management of the insertion hole (Low *et al*. 2005). If properly managed, PIT tag loss can be reduced to negligible levels if the puncture hole is covered with some form of topical adhesive (Lebl & Ruf 2010; Saraux *et al*. 2011). For example during Clarke & Kerry's (1998) two-year study of Adélie penguins (*Pygoscelis adeliae*); they found that after changing their PIT tag implantation procedure to one which required the wound to be covered immediately after insertion, tag loss decreased dramatically from 30% (n = 65/217) to 1% (n = 1/90) (Boersma & Rebstock 2009). It is important to note that even if covered sometimes tags themselves can be rejected by the body of the animal and pushed out through the skin, although this is a rare occurrence and has rarely been reported (Gibbons & Andrews 2004).

The migration of tags through the individuals' body has also been noted in numerous studies and in numerous animal species (Jackson & Bunger 1993; Buhlmann & Tuberville 1998; Camper & Dixon 1999; van Dam & Diez 1999). When PIT tags move internally through an animals' body it not only complicates identification and scanning (Gibbons & Andrews 2004;

Beausoleil *et al.* 2004), but can also be dangerous. Studies have shown that inserting an adequate-sized PIT tag, subcutaneously into the back of a birds neck (as with yellow-eyed penguins; DOC 2012) have the highest retention rate (Jackson & Bunger 1993), and lowest tag migration rates (Rao & Edmondson 1990; Dann *et al.* 2013).

Clarke & Kerry's (1998) findings showed that migration distances of PIT tags of up to 5 cm was common in Adélie penguins (n = 13/20). With most cases the tag had moved slightly to one side of the neck or the other. However in two Adélie penguin chicks PIT tags were found alongside the trachea and oesophagus – this movement could lead to the possibility of obstructing blood flow or damaging vital nerves in the region as well as organ damage (Lambooy & Merks 1989; Clarke & Kerry 1998). Of the 20 individuals examined by Clarke & Kerry (1998), five showed no tag movement and in two individuals the PIT tags no longer existed and were ejected from the body.

There was also a case described where a PIT tag was found to be covered by a slimy biofilm - potentially harbouring pathogenic organisms that were incorporated at the time of implantation (Clarke & Kelly 1998). A recent post mortem report carried out on a yellow-eyed penguin also suggested that there was also an area of infection around the PIT tag implantation site at the back of the individuals' neck (M.J. Young pers. comm. 2014). Although these two incidents appear to be separate, and no other cases have been published; if this contamination is more common there could be a potential risk for the long term survival of PIT tagged birds, especially with the combination of PIT tag migration mentioned above. Proper cleaning of equipment and the animal itself is necessary to avoid contamination of the wound site.

The supporting reader hardware can fail, either due to power or antenna loss

Using PIT tags as a primary mark with no secondary external mark has the disadvantage of, if for some reason, the equipment or reader fails to read in the field (i.e. due to a flat battery), birds can not be identified at all (Beausoleil *et al.* 2004). It is also very difficult or near impossible to find and identify a non functional PIT tag in an individual (Fiedler 2011).

Transcription errors may occur if tag detection systems do not store tag numbers

Handheld readers have little or no storage of large (15-digit) PIT tag numbers, therefore manual detection often relies on transcription into a field notebook, therefore increasing the risk of transcription errors. Some handheld readers have Bluetooth capability, where PIT tag numbers can be transferred to a PDA or Smartphone, with data transfer and management systems.

Other factors that are known to hinder or affect PIT tag scanning results

The orientation of the PIT tag alone has been known to affect whether or not a successful read will transpire (S. Parker pers. comm. 2014; N. Ratcliffe pers. comm. 2014). Since these tags are not anchored to the individual a 90 degree rotation change may make the tag unreadable, especially in an automatic reading system (K. Newbery pers. comm. 2014). The Allflex user manual state that for successful PIT tag reads at the maximum distance, the axes of the transponder scanner and the antenna coils must be perpendicular to one another.

The quality and performance of the equipment can also vary between brands and conditions in which they are used. It has been found that generally the best scanning results come from using the same brand equipment (due to slight differences in production) and this has been supported by Ryan *et al.* (2010).

Metal objects have also been identified to change and distort the magnetic field generated during the reading of the PIT tag. Hence nearby metal objects during scans can reduce reading distance and potentially accuracy. The same is also found in electronic products such as computer screens and cell phones.

The last factor that needs to be accounted for is charge or battery level. The Allflex user manual also states that low battery can affect and reduce reading distances compared to fully charged batteries or power sources.

It is essential to fully understand PIT tag loss and failure rates for yellow-eyed penguins; as significant underestimates for survival parameters could occur due to inaccurate information provided by missing identities, thus making models less realistic and inaccurate, conversely affecting conservation management plans and programmes (Dann *et al.* 2013).

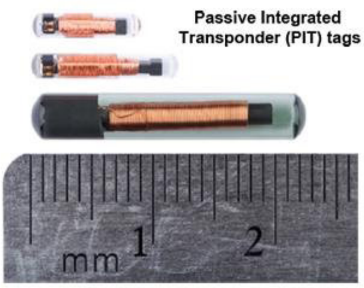
1.5 Types of hardware currently being used for manually detecting transponders in yellow-eyed penguins

The Department of Conservation uses Allflex brand PIT tagging equipment (both PIT tags and readers) on numerous species of animals including yellow-eyed penguins (DOC 2012; Graeme Taylor pers. comm. 2014). Due to the non-colonial behaviour and the vast range they occupy (McKinlay 2001; Ellenberg & Mattern 2012), yellow-eyed penguins are currently manually identified as automatic transponder set ups (i.e. stationary field-based readers) are not cost-effective at this point in time, as they would need to be installed across at least 52 known breeding sites in Otago alone).

As mentioned previously, the two main pieces of equipment used for PIT tag identification are the implanted PIT tag and the reader used to detect and identify the tag itself. The Department of Conservation's best practice manual states that half-duplex ISO encoded 23mm TIRIS PIT tags must be used, which are inserted into the back of bird's neck with a single-use hollow needle (DOC 2012).

To read the 15-digit code produced by these PIT tags, DOC, KPPT and the YEPT use several different models of Allflex readers and these are: the handheld RS200 compact reader; the RS320 yellow stick reader, and; more recently the RS420 green stick reader. These models not only vary in dimensions, specifications, read distances and practicality, but also price; with each model having its own strengths and weaknesses.

Note: all information below comes from Allflex user manuals.



Examples of different size PIT tags, the bottom tag is the size used in Yellow-eyed penguins and costs $12 each

The RS200 compact reader (NZ$300 + GST):

The RS200 compact reader

This model is the smallest out of the three (125.5mm (L) x 70 mm (W) x 24 mm (H)) and is rectangular in shape, compared to the “wand” / “sword” shape of the others. The RS200 takes a 9V battery alkaline battery, from which it can perform over 2,500 PIT tag reads before the batteries need to be replaced. It has an inaccuracy rate (incorrectly identifying a PIT tag identification number) of less than 1 in 10^6, and is fully compatible with numerous varieties of PIT tag.

Due to its size, shape, variability in reading other tags and price it is the most practical transmitter for identifying yellow-eyed penguins, and can easily be placed into a pocket or bum-bag. However, the RS200’s lack of memory and reading distance required to successfully identify a PIT severely impact its overall usefulness and efficiency.

The RS200 can only store one PIT tag identification number at a time (the last PIT tag code encountered). While this may not be a problem if the user follows the proper data collection methods, and records detailed notes while out in the field; the inability to re-check previous scanned birds for misreads or mistakes can be a data integrity issue. However the RS200’s major weakness is its small reading distance, this model only has a reading distance of 5-10cm as stated in the user manual. This reading proximity is further reduced as the device gets older and degrades. Some RS200 readers have to be touching, or virtually touching the back of the bird’s neck before a PIT tag number is detected. This not only puts the user in danger from being bitten by a penguin, but also places further unnecessary stress on the bird itself.

The RS320 yellow stick reader (NZ $1800 + GST):

The RS320 yellow stick reader

This model is the complete opposite in terms of physical dimensions to the RS200 compact reader. The RS320 is “wand”/“sword” shape and is 800mm in length (including handle) and weighs c. 660 grams. This model was originally made for farmers with PIT tagged cattle and sheep, hence is a naturally larger and more robust piece of equipment. Although it is six times more expensive than the compact unit, the RS320 has a read zone of 20 to 35cm (and has been known to locate PIT tags half a metre away; DOC 2012), thus reducing the chance of bird biting the operator as well as stress level of the bird itself, as the user does not have to get as close for a successful read. This ability also makes it easier to scan birds that are in hard to reach nests or are at awkward angles from the user. Although these stick readers are not as practical to carry as the RS200 model, they can be attached to backpacks and are light enough to be carried for field work.

The RS320 is battery powered but this model uses a Ni-CAD rechargeable battery pack that is recommended to be charged for 16 hours overnight for a day’s worth of use. The user manual states that this model can store up 3099 PIT tag numbers (and other important data), however the ones used by the Dunedin branch of DOC and Otago University only seem to only be able to store and review 15 PIT tag identities – although still an upgrade over the compact reader.

PIT tag numbers can be uploaded to a nearby computer or PDA device via Bluetooth or cable, for secure storage and to avoid transcription errors, making it possible to amend field notes at a later date if required.

The RS420 green stick reader (NZ $2000 + GST):

The RS420 green stick reader

The RS420 is the new and improved model of the RS320, and has not yet been used to extensively scan yellow-eyed penguins. This model although shorter than its predecessor at 670mm, weighs slightly more at 830g.

Along with the usual upgrades of being more reliable, user friendly and offering a larger memory to store tag numbers, this model also boasts a Li-ion battery that produces a longer battery life of around eight to 16 hours depending on scanning function, faster recharge times of only three hours and a greater reading distance of up to 42cm from the tip of the device.

The RS420 vibrates the handle instead of emitting a beep when a tag has been successfully read and recorded, thus further reducing the stress impact placed on yellow-eyed penguins who can become startled by loud sounds during the nest check process (M.J. Young pers. comm. 2014).

2. Rationale for research

The Department of Conservation, the Yellow-eyed Penguin Trust and the Katiki Point Charitable Penguin Trust are currently entirely reliant on manual detection of PIT tags in yellow-eyed penguins using low-power handheld readers. PIT tag and hardware failure rates are not readily reported in published literature, with the majority of published research using high-power automated systems for collection of PIT tag data, which are not comparable with the situation at hand.

This study will address the rate of accurate detection of transponders in yellow-eyed penguins using the current marking and manual detection methods, as well as determining a rate of fault or failure with individual PIT tags.

Due to the snapshot nature of this research, this study does not address the detection of transponders or their apparent degradation over time.

3. Methods

During the austrial spring of 2013, three trained observers scanned yellow-eyed penguin PIT tags during regular nest checks across two areas of the Otago coast. Two Allflex RS320 yellow stick readers and one Allflex RS200 handheld reader were sourced from the Department of Conservation and the University of Otago. Each day the yellow stick readers had two out of four fully charged batteries randomly allocated between them to ensure proper functionality. While the handheld reader had a new Energizer^{TM} non-rechargeable battery replaced as required. Before each day of use and after each change of battery, each unit was tested using a test unit transponder, and all three units detected this test unit on their first scan in perpendicular and parallel panning motions.

During the nest visits (from the start of October – to the end of November 2013), adult yellow-eyed penguins sitting prone on the nest (or standing around the nest if a pair was present) were scanned using either a handheld or a yellow stick reader, four times in parallel to the beak (coronal plane; the assumed orientation of the implanted transponder) and four times in perpendicular (transverse plane, see Figure 1 below). The 'read' button was pressed on the scanner unit which was waved over the neck area of the attending bird(s) for the full read time per scan. In total 203 nest approaches were undertaken on 66 different nests; 132 individuals were encountered during the survey, of which 29 were found to be dual marked.

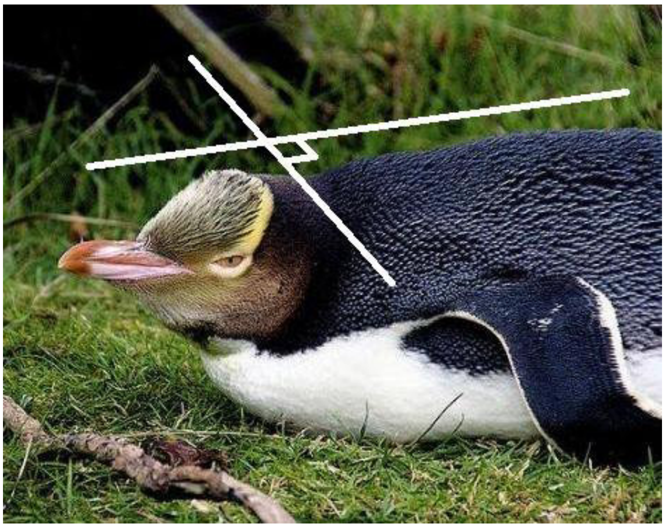


Figure 1: A yellow-eyed penguin prone on a nest, with a diagram of the direction of transceiver scanning motions (Parallel – beak to tail, Perpendicular – flipper to flipper)

The frequency of first detection was recorded separately for the parallel and perpendicular scans for each bird, and for each reader type. The data was transferred from notebooks into

Microsoft Excel and the frequencies of detection were analysed for both scanner types and scanning directions.

Note: It was not always possible to scan prone yellow-eyed penguins on the nest in both directions, as there were many obstructions present, including tree stumps and rocks. In addition, some of the attending birds protested at the level of disturbance caused by waving the reader above them, either attacking the transponder reader and the operator, or moving their position on the nest to a less-favourable reading position. In these instances, the operator collected as much information as possible without causing further stress to the bird attending the nest.

4. Results

(a) Summary data

Analysis of encountered individuals at the 66 nests checked showed that 75 of the 132 (56.8%) that were scanned were found to not have a transponder; and were instead identified by their flipper band.

Twenty nine of the 132 (21.9%) of the breeding adults were identified using both their flipper bands and their transponder (dual marked). Twenty seven of these birds were found to have been marked earlier in 2013, one in May 2012, and one bird in January 2010.

Twenty two of the 132 (16.7%) adults were not checked for a transponder, either because they left the nest during the nest check, or the transponder reader battery failed during the nest round; these birds were not detected or found in subsequent nest checks.

(b) Data gathered from dual marked yellow-eyed penguin nest visits

For the 29 adults that had both a transponder and a flipper band, a total of 117 scanning events were recorded in either a parallel or perpendicular scan. Most dual marked birds were scanned more than once. The majority of these scans were undertaken using the yellow stick reader (109 of 117, 93.2%), with only 8 scans using the handheld reader (6.8%). The handheld reader was not favoured by any of the three observers for various reasons during nest visits. However it was felt that it was appropriate to include the data to increase the sample size, albeit marginally.

In total, 111 of 117 (94.9%) scans in either parallel or perpendicular resulted in a transponder being detected on one of four passes with the stick reader or the handheld reader (Table 3). Six of 117 (5.1%) scans in either parallel or perpendicular resulted in a transponder not being found at all, four of the six were when the RS200 compact reader was used, while the remaining two were from the RS320 yellow stick readers (Tables 3, 5, 6). Removing the handheld reader data from the sample decreased the number of undetected transponders to 1.8% (Table 4), however this did not improve the distribution of the data from one to four scans remarkably (Tables 3 and 4).

Table 3: Manual detection of 23mm TIRIS transponders in yellow-eyed penguins/hōiho (Megadyptes antipodes) using Allflex handheld and Allflex RS320 yellow stick readers for 117 reading events for 29 individual penguins, reading either in parallel or perpendicular. The data here represent the first time the transponder was detected on one of four scans only. Cumulative totals indicate the percentage of transponders detected as the number of scans increases.

Reading event, all reader types	N	Percentage	Cumulative
Transponder detected on first scan	28	23.9%	23.9%
Transponder detected on second scan	23	19.7%	43.6%
Transponder detected on third scan	28	23.9%	67.5%
Transponder detected on fourth scan	32	27.4%	94.9%
Transponder not detected after four scans	6	5.1%	100.0%
TOTAL NUMBER OF SCANS	**117**		

Table 4: Manual detection of 23mm TIRIS transponders in yellow-eyed penguins/hōiho (M. antipodes) using the Allflex RS320 yellow stick readers for 109 reading events for 29 individual penguins, reading either in parallel or perpendicular. The data here represent the first time the transponder was detected on one of four scans only. Cumulative totals indicate the percentage of transponders detected as the number of scans increases.

Reading event, RS320 yellow stick reader	N	Percentage	Cumulative
Transponder detected on first scan	28	25.7%	25.7%
Transponder detected on second scan	20	18.3%	44.0%
Transponder detected on third scan	28	25.7%	69.7%
Transponder detected on fourth scan	31	28.4%	98.2%
Transponder not detected after four scans	2	1.8%	100.0%
TOTAL NUMBER OF SCANS	**109**		

Our data also found that neither the parallel, nor the perpendicular scanning technique were significantly better at detecting PIT tags on the first pass of the transponder unit (p-value = 0.397, α = 0.05; Tables 5 and 6). Although the parallel scanning method shows that twice the amount of PIT tags were not detected after four attempts (n=4, compared to n=2 for perpendicular method) this was also found to not be significant.

Table 5: Manual detection of 23mm TIRIS transponders in yellow-eyed penguins/hōiho (M. antipodes) using the Allflex handheld and the Allflex RS320 yellow stick readers for 63 reading events for 28 individual penguins, reading in parallel only. The data here represent the first time the transponder was detected on one of four scans only.

Reading event, all reader types, parallel only	N	Percentage	Cumulative
Transponder detected on first scan	16	25.4%	25.4%
Transponder detected on second scan	13	20.6%	46.0%
Transponder detected on third scan	13	20.6%	66.7%
Transponder detected on fourth scan	17	27.0%	93.7%
Transponder not detected after four scans	4	6.3%	100.0%
TOTAL NUMBER OF SCANS	**63**		

Table 6: Manual detection of 23mm TIRIS transponders in yellow-eyed penguins/hōiho (M. antipodes) using the Allflex handheld and the Allflex RS320 yellow stick readers for 54 reading events for 29 individual penguins, reading in perpendicular only. The data here represent the first time the transponder was detected on one of four scans only.

Reading event, all reader types, perpendicular only	N	Percentage	Cumulative
Transponder detected on first scan	12	22.2%	22.2%
Transponder detected on second scan	10	18.5%	40.7%
Transponder detected on third scan	15	27.8%	68.5%
Transponder detected on fourth scan	15	27.8%	96.3%
Transponder not detected after four scans	2	3.7%	100.0%
TOTAL NUMBER OF SCANS	**54**		

To account for pseudoreplication in the dataset, the raw data was pooled and averaged across each individual penguin and the amount of scans it took to detect its transponder. From here a two-factor ANOVA was manually applied to detect whether or not a difference between the scanning directions was evident, in addition to whether or not there was an interaction-effect between the amount of scans and method used. The results found that there were no significant differences found between the scanning direction (method) used ($F_{\text{-value } (1, 87)}$ = 0.02633; α <0.05) and no interaction-effect was detected ($F_{\text{-value } (24, 87)}$ = 1.27; α <0.05).

Our results show that compared to numerous other studies and anecdotal evidence, the current yellow-eyed penguin transponder set up is an issue for conservation management of this species. Although the transponder readers do not seem to have any trouble identifying the correct PIT tag identification code, there is a significant issue with the rate at which these are being successfully detected. It was assumed that the results found would follow a Poisson distribution, where the data would be strongly skewed to the left (i.e. the majority of counts would be detected on the first scan, and very few on the second, third or fourth) however, this is not the case (see Tables 3-6). It was also concluded that the direction or orientation scanning method did not impact or change this outcome, even when pseudo-replication was taken into account.

5. Discussion

As stated previously Gibbons & Andrews (2004) found that most studies had a transponder detection rate of 95-100% per scan. The current yellow-eyed penguin PIT tag findings showed a detection rate of around a quarter of what should be expected. This is of significant concern as it may take up to four scanning attempts to detect and identify an individual; if this penguin is stressed or decides to flee - identification might not be possible for an extended period of time. This result is also magnified due to the Department of Conservation and other agencies presently struggling to detect and identify more than 80% of all breeders with manual detection (with flipper bands and transponders), with most sites being more in the range of 30-50% breeder identification rates per year (M.J. Young pers. comm. 2013).

It is also important to note that the all of the transponders that were not detected after four scans (in either direction) were actually still intact and active inside of the animal. This was proven as their PIT tag code was detected and recorded at a later date, but for unknown reasons were not detected at the time.

For best results in scanning PIT tags Allflex claims that the axes of the transponder and the reader must be perpendicular to one another. However we did not see such a trend in our results even when the data was controlled for pseudoreplication. Even if the tags have randomly changed orientation over time post-implantation, a favoured direction of detection was still not picked up when individual birds were scanned on multiple occasions.

The exact reason these results occurred are not fully understood. Whether or not it was due to equipment failure or other issues (PIT tags and/or transponder reader), human error or another factor (i.e. tag migration) was not determined in this study. Although an RS320 battery did fail during the early dates of nest checks, and was discarded after failing to detect the test transponder after another over night charge. This point is thought to be a crucial inclusion in this document to state that we did have an initial problem with the equipment used. Whether or not this is a more common occurrence and is ignored and not reported in published literature or is an isolated event is unknown. But it is these types of incidences that can and do alter transponder identification rates, and so might affect other similar studies using comparable devices.

Ryan *et al.* (2010) found that the Allflex PIT tags, which are used in yellow-eyed penguins, showed relatively high read distance variance between transponders. Whether or not this finding has an influence upon successful read rates was not resolved in the conclusion of this study, but it should not be ruled out altogether. Although, it is alarming that the vast majority of studies and anecdotal evidence from people using similar hardware have not found the results and issues we readily came across (Table 1).

In stating this comparisons between other published studies and the results we obtained have to be taken *cum grano salis*, and on a case by case basis. This is due to the majority of studies using transponders in wildlife use automated systems, where the power draw is significantly higher (sometimes from mains power) than that of a handheld reader used for manual detection. High-power systems for transponder detection can dramatically change and improve results of detection rates and read distances.

The recent addition of the Allflex RS420 green stick reader to the coastal Otago yellow-eyed penguin identification programme looks to be a much needed upgrade to the previous RS320 readers, if manual detection of transponders is still to be a part of the monitoring programme. Although not monitored or analysed in this study, early reports have come off favourable and have indicated a 100% detection rate on the first attempt on newly tagged chicks. Whether or not the model will work equally as well on older PIT tags and individuals is still unclear; however this could be the solution to the current problems and may ensure the practical use of transponders being used on yellow-eyed penguins in the near future.

5.1 Problems with transponders and readers to be resolved by conservation managers

The results indicate that none of the yellow-eyed penguins lost their PIT tags, as each bird encountered was read at least once using the RS320 yellow stick reader. However, the readers themselves, or their rechargabable batteries may be faulty. It is recommended that a "scanning protocol" be established, to ensure that all birds read for a PIT tag are detected.
I recommend the following:

- The results of the current study, four passes with an RS320 resulted in c. 95% of PIT tags being detected, therefore four scans should be sufficient to detect an implanted PIT.

- The RS420 green stick reader detected 100% of freshly implanted tags, and the scanning protocol needs to be tested using a larger sample size, as for the RS320 yellow stick reader, to determine a "scanning protocol" for the use of this advanced model.

- The RS200 handheld should not be used for scanning birds that are incubating or mobile birds (moving birds) – just for birds that are "in the hand".

The risks associated with not scanning a bird an appropriate number of times are high, based on the data presented. This could result in a much lower detection rate for marked birds, which would be to the detriment of monitoring and management. Most importantly, a failure to appropriately determine the identity of a marked individual may result in a bird being marked twice, without being checked appropriately for an RFID, without this identity being linked to its original identity.

DOC and other agencies need to collaboratively establish a time frame for implementing the roll out of this technology as it becomes available. For example, the RS420 reader is now available if some agencies wish to upgrade, which may yield significantly more recoveries of yellow-eyed penguins than an RS320, depending on the number of times a bird has been scanned. Establishing a timeframe in association with Allflex may allow some agencies to determine “trial periods” for using the new technology; as well as considering how frequently parts and models need to be upgraded.

The yellow-eyed penguin monitoring programme in Otago and elsewhere collect data on individuals and breeding productivity by site, which indicates dispersal, longevity, lifetime reproductive success, pair-bond duration, and individual statistics, including season to season breeding success, time spent in temporary captivity, recoveries outside of the breeding season, and eventually death. Manual detection of transponders is a labour-intensive means of obtaining this data, particularly when it is also delicately balanced with technology that may or may not be working to collect the necessary information. Much of the data (particularly dispersal, longevity and recoveries) can be collected using an automated detection system.

Reliance on manual detection of PIT tags should only be considered for breeding birds laying prone at the nest, rather than approaching non-breeding or mobile birds, which are likely to take fright when approached. Information from the latter is extremely important to determine survival information, and it is strongly recommended that automated detection techniques be established for determining the identities of these non-breeding birds.

5.2 Secondary external marks as a measure to prevent double-marking

It is recommended that the Department of Conservation and other agencies research, gain ethics approval and trial an appropriate secondary mark for yellow-eyed penguins marked with a transponder. A secondary mark may prevent birds from going undetected, or being marked twice, or more.

Such marks include:

Flipper bands – The current method applied to the majority of marked yellow-eyed penguins; however they have fallen out of favour for multiple reasons. This is partially due to the numerous negative impacts that they have been noted to cause on numerous penguin species (see Jackson & Wilson 2002; Beausoleil *et al.* 2004; Petersen *et al.* 2006; Maho *et al.* 2011); including evidence of harm (such as feather wear and feather loss) caused to local yellow-eyed penguins. Flipper bands also need to be maintained and fixed with a tool kit if they become twisted or sprung open, thus causing more work for rangers and other personnel responsible for penguin welfare which could be spent elsewhere. Moreover there is also a global push by such organisations as the Scientific Committee on Antarctic Research (SCAR) to stop or heavily reduce the implementation of flipper bands on penguins worldwide, in favour of other marking types (Petersen *et al.* 2005).

Foot tattoos –Although little information seems to have been published on their usage in penguins, they have been used in other sea bird monitoring programmes such as in boobies and albatross. Tattooing is considered the most permanent method for marking wildlife, and hence can be used to determine loss rates of less permanent marks (Beausoleil *et al*. 2004; Mellor *et al*. 2004). In New Zealand tattooing on animals has been used on reptiles, amphibians and pinniped species, although has only ever been used alongside other marking techniques and not as a primary mark (Beausoleil *et al*. 2004). Tattoos generally cause few problems (cause no energetic cost to animal, minimal effects on behaviour or physiology, do not need to be maintained), and can be applied with numerous portable tattooing devices (Mellor *et al*. 2004). However they also have the disadvantage of requiring recapture or a person to get close enough to manually identify the individual. As a secondary marking technique however, handling would only be necessary if the primary mark had failed.

Rubber leg O-rings – No studies I have come across have explicitly used this as a marking method for penguins. However Technical Advisor Bruce McKinlay believes these might be worth looking into. A study on African penguins (*Spheniscus demersus*) found that using experimental rubber flipper bands caused less drag in the water in comparison to steel banded flipper bands; there use did also not affect breeding success compared to non-banded individuals (Barham *et al.*2004). Whether or not the leg injuries will once again occur is unknown at this point until trials are implemented.

Web tags – Are the last viable option for tagging yellow-eyed penguins, and have so far been unsuccessfully trialled by John Darby. I would personally not advise this as a viable marking alternative due to the importance and thermo-regulatory properties of penguins' feet (Frost *et al.* 1975; McCafferty *et al.* 2013). Fatal damage could be caused if the web tags were ripped out or damaged the penguins feet in one way or another (foot injuries are common for yellow-eyed penguins, due to bites from barracouta (*Thyrsites atun*)). Most web tags are also very small and cannot be read from a distance (may also require a magnifying glass to read), making them less practical than other methods (Boersma & Rebstock 2009).

5.3 Establishing automated detection systems with higher power draw for detecting transponders

As suggested previously, developing a roll out for an automated detection system (including a trial of different models for different situations, high power vs. low power and locations) is an important next step for yellow-eyed penguin detection. Tailoring a system to the birds and their breeding areas, rather than the other way around (e.g. bringing something off the shelf for cattle) is an important consideration for future monitoring, especially in hard to reach localities. Systems like these could not only increase the identification rates and information gathered of yellow-eyed penguins across the coast, but also heavily reduce the nesting birds stress levels. These set ups could potentially also help detect non-breeding birds which DOC has struggled to currently achieve in the last decade.

5.4 Areas for future research

This study is only the beginning of solving and implementing a long lasting, cost-effective management programme for yellow-eyed penguins. From here the first thing that needs to be looked into is determining the detection rates for Allflex RS320's and RS420's transponders over time, i.e. do the transponders degrade depending on how long they are inside the animal? The results we have presented here form a baseline for determining not only rates of detection for different readers, but the rates of detection for individuals over time.

A protocol for how to most effectively monitor yellow-eyed penguins (using both the RS420 and RS320) needs to be established. Stating not only the methods of how best to scan PIT tag devices, but also the best ways to record and analyse data to create uniformity from all regions yellow-eyed penguins is required. This protocol needs to cover how to best deal with scanning and identification issues that might occur, so they are known and properly understood so they can be recorded, fixed or altered if need be.

A secondary mark may be required if the PIT tag issues identified are not soon amended. Alternative marks would most likely need to be external and different from the current option of flipper banding. A study on other appropriate marking methods such as foot tattoos should be initiated to see whether they are a more practical alternative if PIT tags alone are not satisfactory.

Trials of automated detection systems should also be established to see how feasible these can be for yellow-eyed penguins. Initially small systems (similar the ones used in the Chatham Island taiko; Taylor *et al*. 2012) could be set up at or around previously identified nests to establish their viability. From there decisions can be made upon whether they are the best expenditure of resources and should be continued on a greater scale.

6. Conclusions and recommendations for stakeholders

This report should only be the very beginning on what is the best, and most cost efficient way to mark yellow-eyed penguins for the foreseeable future. Results have indicated that at the current time there are issues with current PIT tag monitoring, and these need to be rectified. In the short term if the Allflex RS420 transponder reader proves to be the capable and reliable PIT tag scanner that it has so far appears to be, then the issue should soon be resolved with the current RS320's being replaced by the newer addition. However if the detectability issues are still occurring on a regular basis, a new form of marking or identification may need to be introduced. Whether this is via purpose-built, automated PIT identification systems, or applying another type of external mark so far remains to be seen. A full review of current yellow-eyed penguin marking techniques including the use of flipper bands and other possible alternatives; as well as the small-scale introduction of automated yellow-eyed penguin detection systems need to be considered the next step in yellow-eyed penguin monitoring.

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