

Recent invasion of European red foxes (*Vulpes vulpes*) on to Fraser Island (K'gari) and South Stradbroke Island

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Abstract Invasive predators are globally significant drivers of threatened fauna population decline and extinction, and the early detection of new incursions is critical to the chances of successful predator eradication and fauna conservation. Here, we provide evidence of the recent invasion of European red foxes (*Vulpes vulpes*) on to two large and internationally significant islands off the southeast coast of Queensland, Australia – Fraser Island (K'gari) and South Stradbroke Island. From camera trap footage collected on Fraser Island since 2009, foxes have now been observed on seven different occasions between 2012 and 2016. Two scats collected on South Stradbroke Island in 2013 and 2014 tested positive for fox DNA (and negative for *Canis* spp. DNA), with fox presence confirmed by subsequent camera trap footage in 2016. These data confirm the recent incursion of foxes on to these islands and suggest that small populations now exist there. Fraser Island and South Stradbroke Island represent key RAMSAR wetland areas of refuge for populations of multiple threatened fauna that have never been previously exposed to foxes. Fox impacts on these fauna can only be expected to increase without management intervention to eradicate them before they become widespread.

Key words: eradication, invasive species, island extinction, mesopredator, threatened fauna, water mouse.

INTRODUCTION

Invasive species are globally significant drivers of threatened fauna population decline and extinction (Genovesi *et al.* 2012; Bellard *et al.* 2016). These impacts are usually exacerbated on islands (Hanna & Cardillo 2014), which are often home to many endemic and migratory species. Mammalian predators are amongst the worst invasive species (Schoener *et al.* 2001; Doherty *et al.* 2016), and substantial effort goes into controlling the impacts of predators introduced to islands around the world (Jones *et al.* 2016).

The impacts of introduced mammalian predators have been particularly severe in Australia, where dingoes and other wild dogs (*Canis lupus dingo*), European foxes (*Vulpes vulpes*) and feral cats (*Felis catus*) have each been implicated in the declines and extinctions of multiple native fauna (Dickman 1996; Johnson 2006; Allen & Fleming 2012). Cats and foxes have been especially problematic, and they are both

seen as high priority pest species in need of active management and eradication where possible (Australian Government 2015). Cats naturalized quickly and have been present across the entire Australian continent for over a century (Abbott 2002). Foxes colonized Australia more slowly, are presently distributed across approximately 70% of the mainland, and continue to expand their range and impacts on threatened fauna (Saunders & McLeod 2007; Saunders *et al.* 2010). The early detection of new incursions of these invasive species is critical to the chances of successful eradication (Berry *et al.* 2007; Agriculture Victoria 2009; Gregory *et al.* 2014), especially on islands. The economic costs of invasion are far less if eradication is achieved early, and the potential negative impacts of invasive species can be averted following swift management action.

In this report, we describe the recent invasion of European red foxes to two internationally significant islands off the coast of southeast Queensland, Australia – World Heritage listed Fraser Island (K'gari) and South Stradbroke Island, both of which are within key RAMSAR wetland areas. Our aim is to outline the current state of evidence for fox

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occurrence at these sites and alert conservation practitioners to their presence.

EVIDENCE FOR FOXES ON FRASER ISLAND (K'GARI)

Camera traps have been used extensively on Fraser Island for several years to inform the management of dingoes and other extant wildlife and pest animal species. Over 600 000 camera trap images have been collected since 2009. From these images and one first-hand sighting, foxes have now been positively observed on seven different occasions over a four year period (Figs 1, 2) as follows:

- Photographed, 26th July 2012, intersection of Southern Rd and Yankee Jack Creek Rd;
- Photographed, 2nd July 2013, intersection of Southern Rd and Yankee Jack Creek Rd;
- Photographed, 19th July 2013, intersection of Southern Rd and Yankee Jack Creek Rd;
- Photographed, 19th July 2014, Buff Creek;

- Photographed, 23rd July 2014, intersection of Southern Rd and Yankee Jack Creek Rd;
- Photographed, (day unknown) August 2014, Buff Creek; and
- Sighted first-hand by G. O'Conner and S. O'Conner, 19th October 2015, Awinya fire break 10 km west of Dundubura.

EVIDENCE FOR FOXES ON SOUTH STRADBROKE ISLAND

State and local government officers have conducted annual invasive weed surveillance and control activities on South Stradbroke Island in early winter each year since the early 1990s, and an extensive pest animal survey of the island was also conducted in 2012 (L. Willsher, unpubl. data, 2012). No evidence of foxes was found during these surveys, or during any other activity on the island during this period. But during weed control activities in June 2013, fox footprints were observed in the sand at a single site (−27.860604, 153.425680), accompanied by a fresh



Fig. 1. European red foxes photographed on Fraser Island (K'gari) on (a) 26th July 2012, (b) 19th July 2013 and (c) 19th July 2014, and on South Stradbroke Island on (d and e) 15th November 2016 and (f) 30th November 2016. [Colour figure can be viewed at wileyonlinelibrary.com]

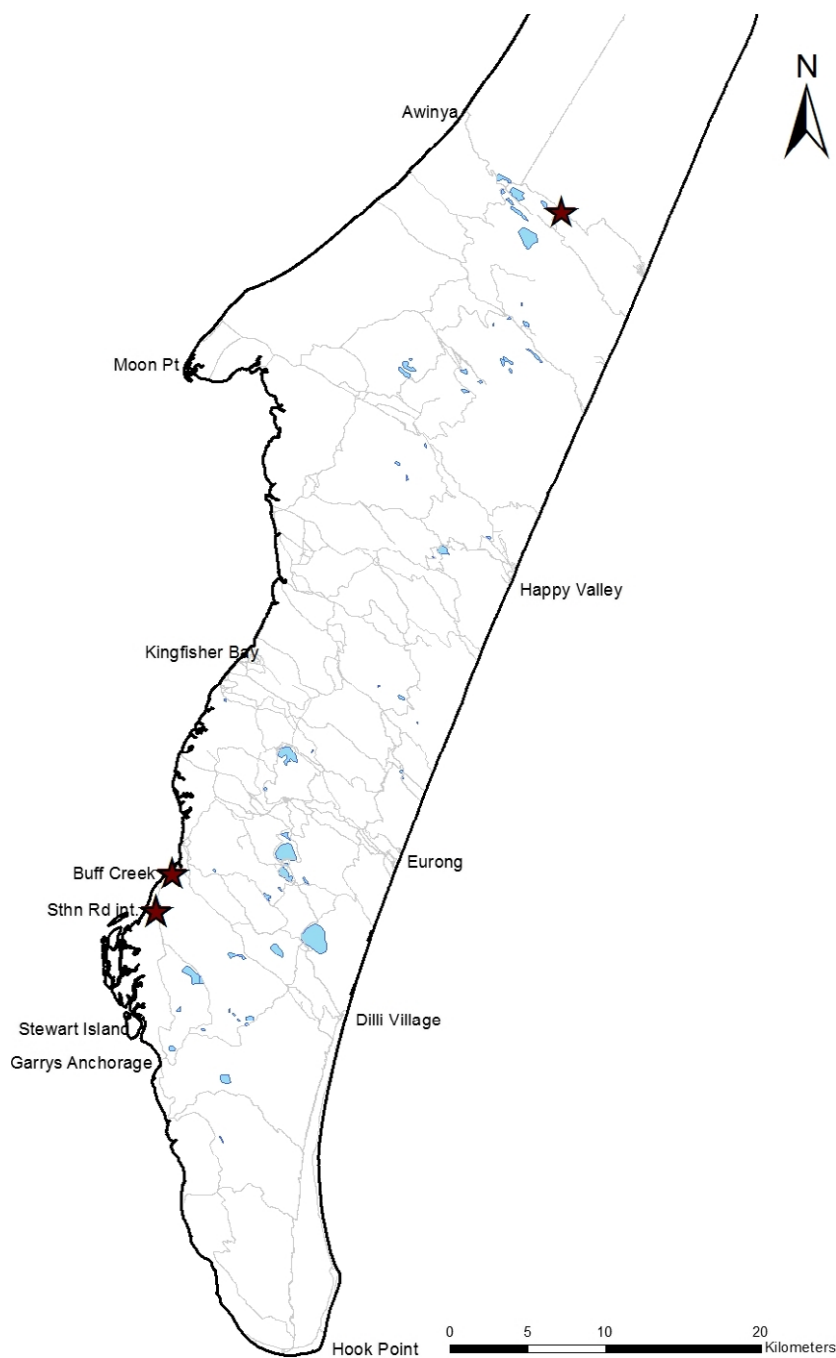


Fig. 2. Location of fox records (stars) on Fraser Island (K'gari), 26th July 2012 until 30th September 2016. [Colour figure can be viewed at wileyonlinelibrary.com]

fox scat. Fox footprints and another fresh scat were observed nearby ($-27.861867, 153.426278$) on the 15th May 2014. Fox footprints and scats can easily be confused with those of small domestic dogs (*Canis familiaris*), which frequently accompany people to the island. So, both scats were collected and analyzed for the presence of fox and/or dog DNA. No attempts were made to genotype individual foxes from the fox DNA in these scats (see below).

DNA was extracted from the 2013 fox scat within three weeks after collection from the field using a QIA-amp stool kit according to the manufacturer's instructions. During the extraction process, the scat was noted to contain a large amount of pale to dark brown hairs, some bone fragments, and an intact but unidentified claw $\sim 7\text{--}8$ mm long. The bones and claw were removed, so the scat subsamples used for DNA extraction included only scat matter

and some hair. We divided the scat into three, and undertook extractions from these three separate subsamples, that differed by proportion of hair. We used a skin fragment from a salted fox pelt (collected by B.A. a few months earlier from Noosa, on the Sunshine Coast) as a positive control, and blood from a domestic dog (ridgeback/cattle dog cross) as a negative control. DNA was extracted from the fox skin fragment and dog blood using a Qiagen DNeasy blood and tissue kit according to the manufacturer's instructions. All extracts were stored at -20°C . PCR for detection of fox DNA was based on the procedure described by Berry *et al.* (2007), with minor modifications to account for amplification chemistry. The reactions included a 12S intercal control primer pair to determine DNA viability. Briefly, reactions consisted of $1\times$ polymerase reaction buffer, $5\ \mu\text{g}$ BSA, $5\ \mu\text{mol}$ of each primer (*V. vulpes* cytochrome B specific pair and 12S DNA control pair), $2\ \text{U}$ DNA polymerase2, $2\ \mu\text{L}$ DNA extract, and a volume balance to $25\ \mu\text{L}$, with sterile nuclease-free water. Reactions were cycled at $1\times$ $94^{\circ}\text{C}/2\ \text{mins}$; $35\times$ ($94^{\circ}\text{C}/30\ \text{s}$, $58^{\circ}\text{C}/20\ \text{s}$, $72^{\circ}\text{C}/40\ \text{s}$); and $1\times$ $72^{\circ}\text{C}/2\ \text{mins}$. Amplification was resolved using 2.5% agarose gel electrophoresis and Gel-Red staining. We further tested for the presence of dog DNA in each sample. Reactions were performed using the same mastermix components as above, with a replacement canid-specific primer pair designed in-house for an unrelated project (J. Oakey, unpubl. data, 2016). Reactions were cycled at $1\times$ $94^{\circ}\text{C}/2\ \text{mins}$; $35\times$ ($94^{\circ}\text{C}/30\ \text{s}$, $52^{\circ}\text{C}/20\ \text{s}$, $72^{\circ}\text{C}/30\ \text{s}$); and $1\times$ $72^{\circ}\text{C}/2\ \text{mins}$. Amplification was also resolved using 2.5% agarose gel electrophoresis and Gel-Red staining.

This same process was followed for the 2014 scat. However, after removal of as much hair as possible from the scat, only one subsample was available for testing.

DNA viability tests confirmed all subsamples to contain intact DNA, except for Subsample 3 from the 2013 scat – the subsample that comprised almost entirely of hairs (Table 1). The fox skin fragment and all viable subsamples from both fox scats tested positive to the presence of fox DNA, and the dog blood tested negative for fox DNA. All samples tested negative to the presence of dog DNA, except for the dog blood.

We also undertook confirmation of these PCR results. The 12S viability amplicons and the fox-specific amplicons were excised and purified from the agarose gel using a commercial kit and the concentration was estimated by comparison to commercial standards. The nucleotide sequence was determined in two directions with dye termination sequencing using 1/8 strength BigDye terminator v3.1 according to the manufacturer's instructions,

purified through Sephadex G50 columns, and resolved using 3500xL genetic analyser (Life Technologies). Resulting chromatograms were proofread and aligned using Sequencher v4.8 (GeneCodes Corp., Ann Arbor, MI). Consensus sequences for each fragment were compared with each other and with the public-access database Genbank. The 183 bp 12S fragments from the fox scat subsamples indicated mixed samples, which was expected (i.e. DNA from the prey animals eaten by the fox was also present in the fox scat along with fox DNA). The 12S fragment from the fox skin fragment had 100% identity with 100% coverage ($E = 8-90$) with previously reported *V. vulpes* 12S sequences, up to 98% identity with other *Vulpes* spp., and up to 96% identity with other canids (J. Oakey, unpubl. data, 2016). The 12S fragment from the domestic dog blood had 100% identity with 100% coverage ($E = 2-90$) with previously reported *C. familiaris* 12S sequences. The 134 bp fox-specific fragment from the scat subsamples and the fox skin fragment were identical. This sequence showed 100% identity with 100% coverage ($E = 1-61$) with previously reported *V. vulpes* cytochrome B. There were no other significant identities suggested from Genbank. The 380 bp canid-specific fragment from the dog blood showed 100% match with equivalent fragments from a range of *C. familiaris* breeds and additional *C. lupus dingo* tissue samples tested within another unrelated project (J. Oakey, unpubl. data, 2016).

After confirming the presence of foxes on South Stradbroke Island using DNA analyses of these two scats, targeted camera trapping surveys were subsequently undertaken in the vicinity of the scats in 2016. A live fox was also observed first-hand on an access track east of the waste transfer station near Tipplers (-27.805433 , 153.427299) on the 11th

Table 1. PCR results from two fox scats collected on South Stradbroke Island in 2013 and 2014 (fox skin fragments were used as a positive control, and domestic dog blood was used a negative control)

| Sample year | Sample | 12S DNA viability | <i>Vulpes</i> presence | <i>Canis</i> presence |
|-------------|-----------------------------------|-------------------|------------------------|-----------------------|
| 2013 | Scat subsample 1 | + | + | - |
| | Scat subsample 2 | + | + | - |
| | Scat subsample 3 | - | - | - |
| | <i>V. vulpes</i> skin subsample 1 | + | + | - |
| | <i>V. vulpes</i> skin subsample 2 | + | + | - |
| | <i>C. familiaris</i> blood | + | - | + |
| 2014 | Scat subsample | + | + | - |
| | <i>V. vulpes</i> skin subsample 1 | + | + | - |
| | <i>C. familiaris</i> blood | + | - | + |
| | No template control | - | - | - |

August 2016 (S. Gava, pers. comm., 2017), and camera traps recorded foxes for the first time on the island on two occasions in November 2016 (Fig. 1).

DISCUSSION

The presence of foxes on Fraser Island was confirmed with photographic evidence (Fig. 1), and the presence of foxes on South Stradbroke Island was confirmed with DNA evidence from two separate fox scats (Table 1) and subsequent photographic evidence (Fig. 1). Verified first-hand sightings of foxes have also recently occurred on both islands.

That foxes have been observed on camera on multiple occasions over five successive years demonstrates that foxes have recently established on Fraser Island. Regular undergraduate biology courses involving analyses of hair and predator scat samples, spotlighting and driving along inland and beach tracks since 2005 have failed to return any evidence of foxes (S. Salisbury, pers. comm., 14 February, 2017). In the early 2000s, Baker (2004) conducted extensive footprint tracking studies across the island in addition to solicitation and analyses of predator hair (DNA) samples, which likewise failed to return any evidence of foxes. Foxes are often caught as bycatch in dingo trapping programs because both species are attracted to the lures used by trappers (Fleming *et al.* 2001), yet ongoing dingo trapping programs conducted on Fraser Island since 2001 (see Allen *et al.* 2015) have yielded no foxes. Park rangers have undertaken countless interviews with residents and island visitors since 2001 during 'campground briefings' (Allen *et al.* 2012a), which have also yielded no reports of foxes on the island. In addition to these efforts, extensive dingo diet studies conducted since the early 1990s have failed to yield any evidence of foxes in dingo scats (e.g. Twyford 1995; Behrendorff *et al.* 2016). Fauna surveys conducted during this time, in preparation for the nomination of Fraser Island as a World Heritage Area (Harmon-Price 1995), also failed to detect evidence of foxes. It therefore seems reasonable to conclude that foxes have not been present on Fraser Island until their recent detection on camera traps, as reported here. That fox records are still few (despite considerable surveillance efforts) suggests that this population is presently small, and perhaps more easily eradicable.

Historical weed surveillance efforts on South Stradbroke Island and an extensive pest animal survey of the island in 2012 yielded no evidence of foxes, suggesting that the fox incursion there was also recent. However, unconfirmed anecdotal reports of foxes from fisherman and visitors on South Stradbroke Island have occurred for over a decade, suggesting that the incursion of foxes there may have occurred

some time ago. In any case, our confirmation of fox presence via two DNA-positive scats collected on the island approximately 12 months apart and photographic evidence two years later provides unequivocal evidence of their presence on the island, and suggests that a small population of foxes may have also established there.

Foxes have been present in mainland areas adjacent to these two islands for many decades (Saunders & McLeod 2007), but have not been confirmed to occur on these islands until now. The source of the foxes and how they arrived on either island is unknown. However, it is likely that foxes arrived on both Fraser Island and South Stradbroke Island without human assistance given the close proximity (500–1500 m) of these large islands to mainland areas and adjacent islands where foxes are present and/or actively managed (e.g. North Stradbroke Island, Turkey Island; FCRC 2010; GCCC 2013), although we cannot discount translocation by humans. The timing of our records indicates that foxes have been present on Fraser Island since at least 2012, and on South Stradbroke Island since at least 2013.

In Australia, the only mammalian predator larger than foxes is dingoes. Fraser Island is home to a healthy and well-known population of dingoes, which occur across all areas of the island (Allen *et al.* 2015; Behrendorff *et al.* 2016). Dingoes are dominant over foxes, might have the capacity to limit fox abundance, and might therefore be considered an ally in preventing fox establishment (Letnic *et al.* 2012). However, dingoes did not prevent, have not limited, and are not limiting the continued invasion of foxes on mainland Australia, nor have dingoes prevented the many fox-caused and feral cat-caused declines and extinctions of threatened fauna on the mainland. Moreover, positive relationships between dingoes and foxes have been observed, and foxes may derive benefit from dingoes through kleptoparasitism (Allen *et al.* 2012b, 2013). There is evidence that high densities of foxes may also suppress dingoes (Allen *et al.* 2012b). Thus, although dingoes may interact negatively with foxes in some cases, Fraser Island's dingoes cannot be considered a reliable tool to mitigate the impacts of foxes and/or cats there. There are no dingoes on South Stradbroke Island, which now makes foxes the new terrestrial apex predator there.

Foxes have become one of the most destructive invasive predators in Australia, and represent a key threatening process for many species of native animals (DEWHA 2008). Their impacts are most severe on small and medium-sized mammals, turtles and shore birds, and substantial investment and effort goes into controlling fox populations and mitigating their impacts in many areas (Saunders *et al.* 2010). Both Fraser Island and South Stradbroke Island are located within key RAMSAR wetland sites, which are

home to internationally significant threatened and endemic fauna and flora (species lists available at: www.qld.gov.au/environment/plants-animals/species-list). Black-breasted button quail (*Turnix melanogaster*), beach stone-curlews (*Esacus magnirostris*), ground parrots (*Pezoporus wallicus wallicus*) and resting wader species, as well as marine turtles (e.g. green turtle *Chelonia mydas*, and loggerhead turtle *Caretta caretta*), long-nosed potoroos (*Potorous tridactylus*) and water mice (*Xeromys myoides*) are among those threatened fauna most likely to be placed at increased risk given the recent invasion of foxes to these areas. Indeed, local monitoring of water mice in the region over the last few years has revealed the new arrival foxes and their impacts on water mice nests, which includes the total and permanent destruction of water mice nests in some cases (Fig. 3; N. Kaluza, unpubl. data, 2017).

The recent invasion of foxes to these islands has important implications for managers and conservation practitioners. Both Fraser Island and South Stradbroke Island represent key areas of refuge for populations of multiple threatened fauna that have never been previously been exposed to foxes, and mitigating the impacts of introduced predators on islands is a key focus of the national Threatened

Species Strategy (Australian Government 2015). Failure to act and inadequate biosecurity response have contributed to the otherwise preventable extinctions of at least three native Australian vertebrates from offshore islands in the last 5 years (Woinarski *et al.* 2017). Fox impacts on threatened fauna (e.g. Fig. 3) can only be expected to increase without management intervention to eradicate foxes before they become widespread. Eradication of foxes from these islands is possible but will be challenging, less so for South Stradbroke Island (given its relatively small size) but especially for Fraser Island (where fox control activities need to carefully consider the risks of these activities to dingoes). Regardless of the challenges and risks, the ecological consequences of not eradicating foxes from these islands could be highly significant.

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Fig. 3. Fox damage to water mouse nests, showing (a) two foxes digging down a mound nest (foreground) at Maroochy River on the mainland on the 24th March 2016, (b) the levelled mound nest in the foreground and a single fox digging down another adjacent mound nest (in the background) on the 25th March 2016, (c) an intact mound nest at Wangoolba Creek on Fraser Island (K'gari) on the 2nd April 2016, and (d) suspected fox damage to the same mound on 7th September 2016. [Colour figure can be viewed at wileyonlinelibrary.com]

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