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Do non-native invasive fish support elevated lamprey populations?

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Summary

1. Managing populations of predators and their prey to achieve conservation or resource management goals is usually technically challenging and frequently socially controversial. This is true even in the simplest ecosystems but can be made much worse when predator–prey relationships are influenced by complex interactions, such as biological invasions, population trends or animal movements.

2. Lough Neagh in Northern Ireland is a European stronghold for pollan *Coregonus autumnalis*, a coregonine fish and for river lamprey *Lampetra fluviatilis*, which feeds parasitically as an adult. Both species are of high conservation importance. Lampreys are known to consume pollan but detailed knowledge of their interactions is scant. While pollan is well known to be a landlocked species in Ireland, the life cycle of normally anadromous river lamprey in Lough Neagh has been unclear. The Lough is also a highly perturbed ecosystem, supporting several invasive, non-native fish species that have the potential to influence lamprey–pollan interactions.

3. We applied stable isotope techniques to resolve both the movement patterns of lamprey and trophic interactions in this complex community. Recognizing that stable isotope studies are often hampered by high-levels of variability and uncertainty in the systems of interest, we employed novel Bayesian mixing models, which incorporate variability and uncertainty.

4. Stable isotope analyses identified trout *Salmo trutta* and non-native bream *Abramis brama* as the main items in lamprey diet. Pollan only represented a major food source for lamprey between May and July.

5. Stable isotope ratios of carbon in tissues from 71 adult lamprey showed no evidence of marine carbon sources, strongly suggesting that Lough Neagh is host to a highly unusual, nonanadromous freshwater population. This finding marks out the Lough's lamprey population as of particular scientific interest and enhances the conservation significance of this feature of the Lough.

6. *Synthesis and applications*. Our Bayesian isotopic mixing models illustrate an unusual pattern of animal movement, enhancing conservation interest in an already threatened population. We have also revealed a complex relationship between lamprey and their food species that is suggestive of hyperpredation, whereby non-native species may sustain high lamprey populations that may in turn be detrimental to native pollan. Long-term conservation of lamprey and pollan in this system is likely to require management intervention, but in light of this exceptional complexity, no simple management options are currently supported. Conservation plans will require better characterization of

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population-level interactions and simulation modelling of interventions. More generally, our study demonstrates the importance of considering a full range of possible trophic interactions, particularly in complex ecosystems, and highlights Bayesian isotopic mixing models as powerful tools in resolving trophic relationships.

Key-words: Bayesian, conservation dilemma, *Coregonus autumnalis*, hyperpredation, *Lampetra fluviatilis*, pollan, potamodromous, River lamprey, stable isotope analysis in R, stable isotope

Introduction

The conservation of threatened species is rarely straightforward and can be greatly complicated when predators and their prey are both threatened in the same location. In such instances the abundances and fortunes of both species are likely to be closely coupled, and so identifying conservation priorities often presents managers with an ongoing dilemma (Lalas *et al*. 2007). Although such conflicts between the interests of predators and prey are relatively commonplace, management can be complicated further by the presence of invasive, non-native species. It is widely acknowledged that invasive species represent one of the major threats to global biodiversity and ecosystem function (Sala *et al*. 2000), and are particularly problematic in freshwater systems (Marchetti, Moyle & Levine 2004; Clavero & García-Berthou 2006), where invasive species have contributed to the decline or loss of endemics (Lever 1996; Williamson 1996). One mechanism arising from the abundance of invasive, non-native species is that of hyperpredation, whereby predators are sustained or increase in number via the energy inputs provided by invasive prey, which in turn leads to increased impacts on native prey species (Smith & Quin 1996; Courchamp, Langlais & Sugihara 1999).

Lough Neagh is the largest area of freshwater in Great Britain and Ireland, and while it has been colonized by a number of non-native fish (Harrod *et al*. 2001), it remains home to species that are important for conservation and commercial fishery interests. *Coregonus autumnalis* Pallas is common in Lough Neagh and is the only Irish vertebrate that is not found elsewhere in Europe (Whilde 1993). This coregonine fish, known locally as pollan, is currently found in only five lakes in Ireland and in Lough Neagh is the focus of a small commercial fishery. The Irish population has been isolated from other *C. autumnalis* populations for long periods of time and is unusual in many ways. Elsewhere in their distribution, pollan are anadromous, migrating to the sea through large rivers in Arctic Russia and North America, while in Ireland the species is entirely lacustrine. Apart from the loss of anadromy, the Irish populations display distinctive life-history characteristics (e.g. fast growth and small body size) (Harrod *et al*. 2001). As a result of their long isolation and the ecological differences between Irish and Arctic populations of *C. autumnalis*, some have called for Irish pollan to be considered as a separate, endemic species *C. pollan* (Kottelat & Freyhof 2007). Pollan populations are in decline and are a priority species for conservation in Ireland and Northern Ireland (Harrod *et al*. 2001).

Lough Neagh also contains a population of river lamprey *Lampetra fluviatilis* Linnaeus. River lamprey are listed on Annex II of the EC Habitats Directive as a Species of Community Interest, which places the species among the highest priorities for conservation protection at a European level. Adult lampreys feed on fish by attaching externally and consuming the host's tissue and fluids. Lamprey can alternately be considered parasites or perhaps as predators where their attacks result in the death of the host. For clarity of terminology, here we consider lamprey as predators and construe their relationship with food species as a predator–prey interaction. Some lamprey species have been known to have substantial impacts on prey species in freshwater bodies (Harvey, Ebener & White 2008). Previous work suggested that pollan are a major prey species for lamprey (Kennedy & Vickers 1993; Harrod *et al*. 2001; Goodwin *et al*. 2006). However, these earlier studies are compromised for two reasons. First, they rely on counting fish bearing the characteristic scars of past lamprey attacks and so do not account for pollan that are killed by lamprey. Secondly, they have tended not to consider all the fish species within the Lough as potential prey items for lamprey.

If pollan are a major prey item for lamprey, conservation of the two species presents something of a dilemma for managers. It would be facilitative to identify the nature of the interactions between lamprey and pollan in order that effective management practices can be developed for both species. Moreover Lough Neagh also holds large populations of the native brown trout *Salmo trutta* Linnaeus, and introduced, non-native roach, *Rutilus rutilus* Linnaeus, and bream, *Abramis brama* Linnaeus, all of which are candidate prey for lamprey (Kennedy & Vickers 1993).

Generally, river lamprey adopt an anadromous life cycle, where juveniles migrate to the sea or to estuarine areas where they mature before returning to freshwater to spawn and die. Rare potamodromous populations have been found in Lakes Onega and Ladoga in Russia (Berg 1948) and in Loch Lomond in Scotland (Maitland, Morris & East 1994; Adams *et al*. 2008). Recent observations have suggested that at least some of the Lough Neagh river lamprey spend their entire life in freshwater (Goodwin *et al*. 2006), which, if verified, would greatly increase the conservation importance of this population.

Here, we combine stable isotope analyses and direct observations of lamprey foraging scars on prey fish to examine lamprey predation. Because stable isotope ratios of prey items are incorporated into the tissues of consumers in a predictable manner (Hobson & Clark 1992; Bearhop *et al*. 2002), stable isotope mixing models have increasingly been used to quantify the proportion of prey items in the diets of consumers (Inger *et al*. 2006a, b; Inger *et al*. 2008) and to determine trophic position as isotope ratios of nitrogen increase with trophic level. Recent advances have provided powerful Bayesian frameworks (Jackson *et al*. 2008; Moore & Semmens 2008), allowing incorporation of variability in sources, trophic enrichment factors (TEF), and additional unquantifiable variation into these models, thus providing robust estimates of the proportion of different prey items in consumer diets. Using these approaches we may quantify lamprey diet within Lough Neagh, identify sources of variation in diet and determine whether any fish species are preferentially eaten. In addition, isotopic ratios of consumer tissues reflect those of the habitat in which the tissues were synthesized (Hobson & Clark 1992; Inger *et al*. 2006a; Inger & Bearhop 2008), and one of the strongest isotopic gradients is found in stable carbon signatures of marine and freshwater environments (Inger *et al*. 2006b; Adams *et al*. 2008). Therefore, we also used our analyses to determine whether Lough Neagh lamprey had been foraging in a marine environment, and hence whether or not they adopt an anadromous life cycle.

Materials and methods

SAMPLING PREDATOR AND PREY POPULATIONS

Lough Neagh was sampled for both lamprey and putative prey species under license and with permission of the fishery owner. Fish were sampled using seine (draft) nets (length = 82 m, height 4·5 m, mesh = 8 mm knot-to-knot). Samples were taken between 0700 and 1300 at eight different, randomly selected positions (selected by GIS), on each of 3–4 days in five sampling periods from May to October 2006 (total samples $n = 122$). Fish were identified to species, measured (total length ± 1 mm) and returned to the Lough, with the exception of scarred fishes, which were retained for analysis.

To give an initial indication of which species of fish were attacked by lamprey, we recorded the number of individuals of each species with lamprey scars. A subsample of these scarred fish were randomly selected and muscle samples were taken for stable isotope analysis. We also randomly sampled pollan from a commercial (Quibros, Cookstown, UK) gill-net fishery on 12 occasions from 16 may 2006 to 26 October 2006.

Lamprey were also collected whilst sampling the Lough for prey species, and from the commercial gill-net pollan fishery, and the Toomebridge Eel Fishery. Total length (± 1 mm) and body mass (wet weight) (±1 g) were recorded for each lamprey. Blood and muscle samples were collected for stable isotope analysis, and sex was determined by dissection.

STABLE ISOTOPE ANALYSIS

Stable isotope ratios of consumer tissues reflect that of their diet and habitat usage in predictable ways. As different metabolically active tissues turnover at different rates, the rates of incorporation of new material from the environment are also different (Hobson & Clark 1992; Carleton & del Rio 2005). The isotopic ratios of different tissues therefore reflect diet and habitat usage over different temporal periods. In this study, we utilized blood plasma, blood cells and muscle tissue, representing increasing turnover times and so reflecting diet and habitat usage over longer periods.

Lamprey blood samples were separated into plasma and red blood cells (RBC) by centrifugation at 600 ***g*** for 10 min. All tissues were dried at 80 °C for 48 h. Lipids are isotopically light in carbon, and so by removing lipids, carbon isotope ratios better reflect those of their diet. Lipids contain little nitrogen and so have minimal effects on δ^{15}N (Ingram *et al*. 2007; Mintenbeck *et al*. 2008). Lipids were extracted from dried muscle tissue using a Soxhlet apparatus with a 1 : 1 ratio of chloroform and methanol. All samples were then ground into a homogenous powder, before being weighed into tin cups for analysis.

Analysis was carried out at the Natural Environment Research Council Life Sciences Mass Spectrometry Facility. Stable carbon and nitrogen isotopes measurements were carried out using continuous flow isotope ratio mass spectrometry (CF-IRMS), using a Costech (Milan, Italy) ECS 4010 elemental analyser interfaced with a Thermo Electron (Bremen, Germany) Delta Plus XP mass spectrometer. Stable isotope ratios are reported as δ values and expressed in ‰, according to the following: $\delta X = [(R_{\text{sample}}/R_{\text{standard}}) - 1] \times 1000$, where X is ^{13}C or ^{15}N and R is the corresponding ratio ^{13}C/^{12}C or ^{15}N/^{14}N, and R_{standard} is the ratio of the international references PeeDee Belemnite for carbon and AIR for nitrogen. Replicate analyses of internal lab standards during measurements reported in this study yielded standard deviations better than 0·24‰ for δ^{15}N and 0·04‰ for δ^{13}C.

ISOTOPIC MIXING MODELS

Stable isotope mixing models are routinely used to estimate the proportion of food sources in the diet of consumers, although their utility is often constrained in underdetermined systems, or when the sources are highly variable (Phillips & Gregg 2001; Moore & Semmens 2008). Recent advances in statistical techniques utilising a Bayesian approach to the analysis of stable isotopic data overcomes a number of these limitations by incorporating uncertainty into the mixing models (Jackson *et al*. 2008). We used the SIAR package (Stable Isotope Analysis in R; Parnell *et al*. 2008) part of the open source statistical language R (R Development Core Team 2007) that allows each of the sources and the TEF to be assigned as a normal distribution, rather than a single datum. SIAR also allows the user to consider variation within and between consumers. This is achieved by incorporating into a single analysis multiple isotopic measurements (from different tissues) from each individual, and by sampling multiple individuals for each group, in this case sample month. Critically, SIAR also includes a residual error term to account for any other, unquantified sources of variation.

The SIAR requires data on the TEF, dietary sources and consumers. As no lamprey-specific TEFs were available these were calculated based on results from multiple fish species. A meta analysis by McCutchan *et al*. (2003) found a mean of 0·5 ± 0·13‰ for Δ^{13}C across several tissue types (mostly muscle and whole organism material). Subsequent studies of sea bass *Dicentrarchus labrax* Linnaeus, fed a diet of sandeel *Ammodytes marinus* Raitt however found much higher fractionation values and recommended a figure of 2‰ be used (Barnes *et al*. 2007). We therefore used a figure of 1 ± 1‰ for Δ^{13}C to incorporate the range of both studies. In case of Δ^{15}N, meta analysis found a value of 3·3 ± 0·26‰ (McCutchan *et al*. 2003), although again subsequent studies of fish fed a high protein fish diet have recorded higher fractionation values of up to 3·98‰ (Sweeting *et al.* 2007). We included a large standard deviation of 1‰ to the value of 3·3‰ to account for variation found in other studies. Therefore, our input parameters can theoretically take values covering the full range of TEFs from 0‰ to 2‰ for Δ^{13}C and 2·3–4·3‰ for Δ^{15}N thus incorporating all the variation indentified in a recent review of TEFs (Caut, Angulo & Courchamp 2009). To ensure that our results were not sensitive to the TEF we carried out a sensitivity analysis, by varying the mean TEFs by ± 1 SD from the values in McCutchan *et al.* (2003). We then examined the mean proportional values for pollan and bream for our first sampling period (May). Altering $\Delta\delta^{13}$C shifted the proportion of bream by 0·005 and pollan by 0·006 for bream. $\Delta\delta^{15}$N had a more significant effect and the proportions changed by 0·11 for bream and 0·04 for pollan. We also increase the mean $\Delta\delta^{13}$C to 3·13‰ utilizing the equations of Caut *et al.* (2009), and found no changes in our general conclusions.

For all the fishes potentially consumed by lamprey, neither $\Delta\delta^{13}$C nor $\Delta\delta^{15}$N varied significantly over the course of the study. We therefore used the mean and standard deviation of each source in the models; hence we consider the full range of variability in dietary isotope signatures within the models.

An estimate of the amount of variation associated with an individual consumer allows SIAR to make more robust estimates of the said individual's diet, hence we incorporated data from all tissues sampled (muscle, RBC and plasma) into a single analysis. To account for intra-population variability we included data from all individuals sampled within a particular month within the analysis to determine (a) the importance of each prey item in the diet, (b) how, if at all, prey choice varied with time and (c) how the results from the isotopic analysis compare with scarring rates.

STATISTICAL ANALYSIS

All tests were carried out in SPSS Version 12.0 (SPSS Inc., Chicago, IL, USA). Data were tested for normality using Kolmogorov-Smirnov tests, and homoscedasticity using *F*-tests or Levene's tests. Non-normal data were transformed. Proportional/percentage data were arcsine square-root transformed prior to analysis. Stable isotope ratios were analysed using general linear models (GLM) (type III SS) with the isotopic ratios as the dependent variables. For lamprey prey items, species was included as a fixed factor and Julian day as a covariate. For lamprey sample tissue (muscle, blood plasma and RBC), capture method (seine nets, pollan gill-net fishery and eel fishery), were included as factors, and Julian date and body mass were included as covariates. The minimal adequate models were achieved by backwards elimination of nonsignificant terms from the saturated model.

Results

PREDATOR AND PREY POPULATIONS

A total of 5211 fish were collected in seine net samples from Lough Neagh. We assumed that our sample procedure was not selective for any particular species and that our samples represent actual abundances within the lough (see below). Roach and pollan were by far the most abundant species followed by eel *Anguilla anguilla* Linnaeus, perch *Perca fluviatilis* Linnaeus, bream, brown trout, 3-spined stickleback *Gasterosteus aculeatus* Linnaeus, gudgeon *Gobio gobio* Linnaeus and finally roach/bream hybrids (Fig. 1a,b).

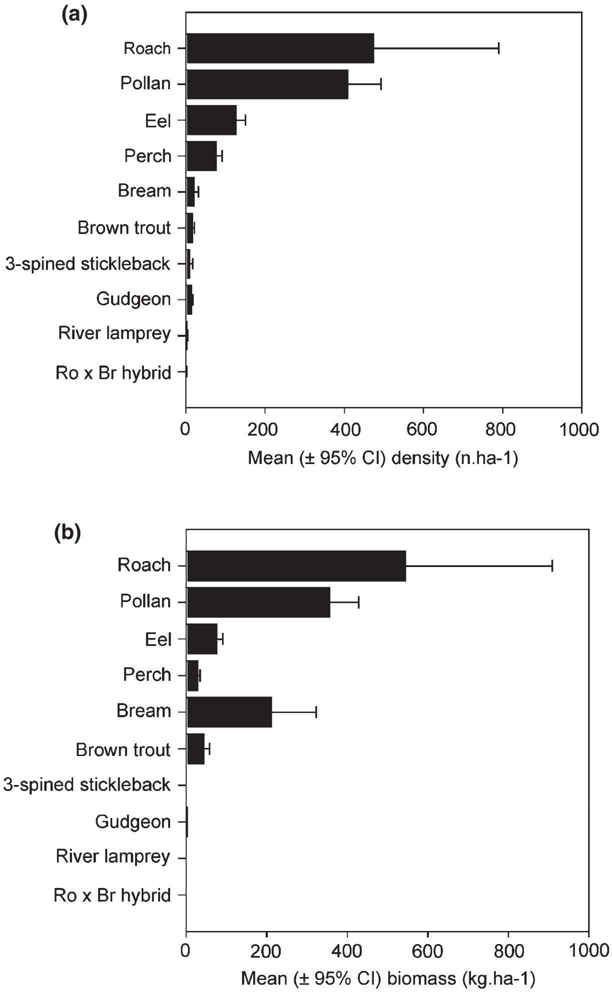

Fig. 1. Estimates of mean fish density (n ha^{-1}) (a) and fish biomass (kg ha^{-1}) (b) of each species within the Lough Neagh fish community from seine net sampling.

A total of 148 lampreys were captured, 25 from Lough Neagh seine net sampling, 12 from the Lough Neagh pollan gill-net fishery and 111 from the Toomebridge eel fishery. No significant differences in length or body mass were found between the sexes, or capture sites. Log length and log body mass of lamprey were significantly related ($F_{[1,144]} = 1631$, $P < 0{\cdot}001$, $r^2 = 0{\cdot}82$), as were Julian date and length ($F_{[1,144]} = 102$, $P < 0{\cdot}001$, $r^2 = 0{\cdot}42$), and Julian date and body mass ($F_{[1,144]} = 76$, $P < 0{\cdot}001$, $r^2 = 0{\cdot}35$), suggesting growth of a single cohort.

SCARRING RATES

A total of 4089 lampreys and fish (of eight species and one roach × bream hybrid) from the seine net sampling were examined for scarring, of which 193 individuals (4·7%) of five species were carrying lamprey scars (Table 1). There were significant differences in the proportions of each species carrying lamprey scars ($F_{[4,63]} = 7{\cdot}92$, $P < 0{\cdot}001$). The highest proportion of scarred individuals was among trout ($\bar{x} = 32\%$, $n = 33/102$), followed by bream ($\bar{x} = 7\%$, $n = 14/134$),

Table 1. Number of fish of each species sampled (n), number with lamprey scars (n_{scar}), the percentage (%) of sampled fish with Lamprey scarring present and (L) mean fish length in mm.

	Pollan				Trout				Bream				Roach				Perch				Gudgeon			
Sample	n	n_{scar}	%	L	n	n_{scar}	%	L	n	n_{scar}	%	L	n	n_{scar}	%	L	n	n_{scar}	%	L	n	n_{scar}	%	L
31 May to 7 June	291	15	5	183	20	6	30	279	20	2	10	183	17	0	0	178	59	0	0	114	7	0	0	153
23–28 June	634	50	8	280	31	10	32	243	20	0	0	280	164	9	5	183	45	1	2	120	15	0	0	134
24–26 July	395	33	8	220	20	8	40	285	4	0	0	220	40	0	0	130	11	0	0	123	4	0	0	129
23–25 August	309	22	7	286	16	4	25	269	29	4	14	286	145	4	3	113	129	0	0	133	21	0	0	135
16–18 October	336	6	2	327	15	5	33	305	61	8	13	327	248	5	2	202	96	1	1	144	28	0	0	130
Mean			6	259			32	276			7	259			2	161			1	127			0	136
Total	1965	126			102	33			134	14			614	18			340	2			75	0		

pollan ($\bar{x} = 6\%$, $n = 126/1965$) roach ($\bar{x} = 2\%$, $n = 18/614$) and perch ($\bar{x} = 1\%$, $n = 2/340$) (Table 1). No scars were found on gudgeon, eel, stickleback or on other lamprey. We found significant differences in scarring rates between sampling periods ($F_{[4,61]} = 6{\cdot}7$, $P < 0{\cdot}001$) and species ($F_{[4,61]} = 13{\cdot}4$, $P < 0{\cdot}001$).

A total of 4808 pollan from the gill-net fishery were checked for lamprey scarring on 12 sampling occasions (Fig. 2). Over all sampling events, 490 (10·2%) scarred pollan were found. The proportion of scarred pollan peaked in May and June, before falling to below 1% for the remainder of the sampling period (Fig. 2).

STABLE ISOTOPE ANALYSIS

A total of 64 individuals of seven potential prey species were sampled for stable isotope analysis (Fig. 3). There were significant differences among species for δ^{13}C ($F_{[6,56]} = 5{\cdot}98$, $P < 0{\cdot}001$) and δ^{15}N ($F_{[6,56]} = 11{\cdot}92$, $P < 0{\cdot}001$). No significant differences were found with sample date (Julian day) for either δ^{13}C ($F_{[6,56]} = 0{\cdot}01$, $P = 0{\cdot}977$) or δ^{15}N ($F_{[6,56]} = 1{\cdot}72$, $P = 0{\cdot}195$).

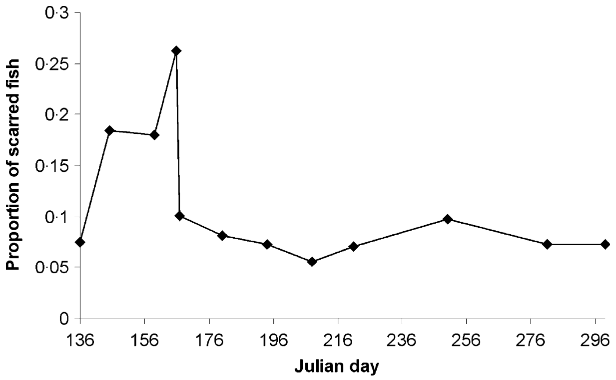

Fig. 2. Proportion of pollan found with lamprey scars from the commercial gill-net fishery, over 12 sampling events between 16/05 and 26/10. Mean sample size = 398, SD = 96.

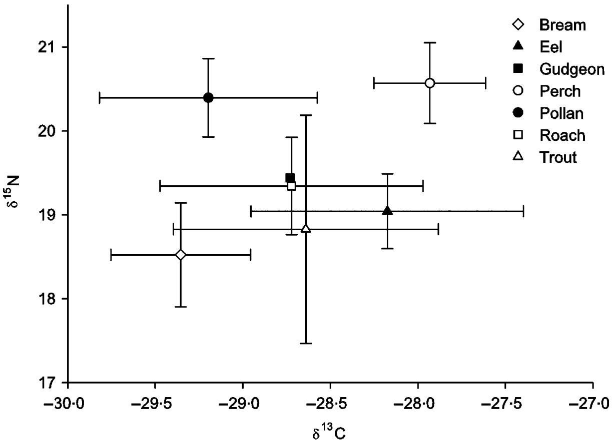

Fig. 3. Biplot of mean (±SD) stable isotopic ratios of carbon and nitrogen for putative lamprey prey species. Bream $n = 13$, eel, $n = 4$, gudgeon $n = 1$, perch $n = 9$, pollan $n = 12$, roach $n = 15$, trout $n = 10$.

A total of 71 lampreys (25 from seine nets, 12 from the gill-net fishery and 34 randomly selected from the eel fishery sample) were collected between May and November 2006. Stable isotope ratios were measured from muscle, RBC and blood plasma (Fig. 4) from each individual. All tissues from all individuals had very low δ^{13}C values, indicative of having been synthesized in a freshwater habitat. For comparison, mean δ^{13}C estimated from for 11 marine fishes was −17·8‰ ± 0·3 (SE) (Etheridge *et al.* 2008) (Fig. 5).

The GLM ($r^2 = 0{\cdot}57$) identified significant differences in δ^{13}C of lamprey between capture methods ($F_{[2,189]} = 12{\cdot}10$, $P < 0{\cdot}001$, seine net $\bar{x} = -28{\cdot}1$, SD = 0·98; pollan fishery $\bar{x} = -27{\cdot}6$, SD = 0·59; eel fishery $\bar{x} = -28{\cdot}1$, SD = 0·72), and a significant interaction between tissue type and capture method ($F_{[2,189]} = 37{\cdot}09$, $P < 0{\cdot}001$). All samples showed isotopic values consistent with feeding in a freshwater habitat (Fig. 5).

The GLM ($r^2 = 0{\cdot}77$) for δ^{15}N found significant differences between capture method ($F_{[1,186]} = 7{\cdot}785$, $P = 0{\cdot}001$, seine net $\bar{x} = 20{\cdot}45$‰, SD = 1·88; pollan fishery $\bar{x} = 20{\cdot}22$‰, SD = 1·70; eel fishery $\bar{x} = 21{\cdot}8$‰, SD = 0·54), tissue type ($F_{[1,186]} = 3{\cdot}76$, $P = 0{\cdot}025$) and significant differences with time (Julian date) ($F_{[1,186]} = 162{\cdot}55$, $P < 0{\cdot}001$) (Fig. 6), and body mass ($F_{[1,186]} = 121{\cdot}34$, $P < 0{\cdot}001$). The model also identified significant interactions between tissue type and body

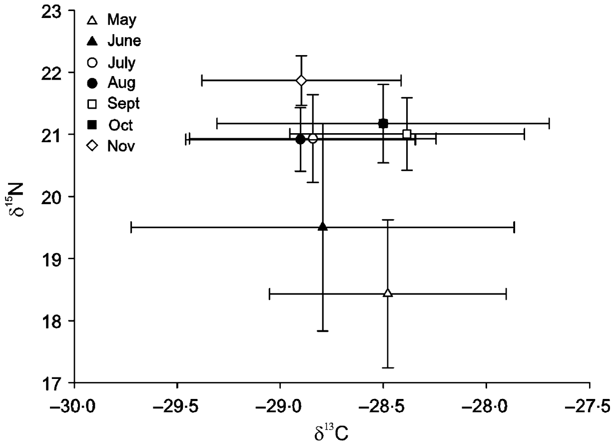

Fig. 4. Biplot of mean $\delta^{13}C$ and $\delta^{15}N$ (± SD) blood plasma from lamprey by month. May $n = 6$, June $n = 5$, July $n = 4$, August $n = 10$, September $n = 12$, October $n = 9$, November $n = 22$.

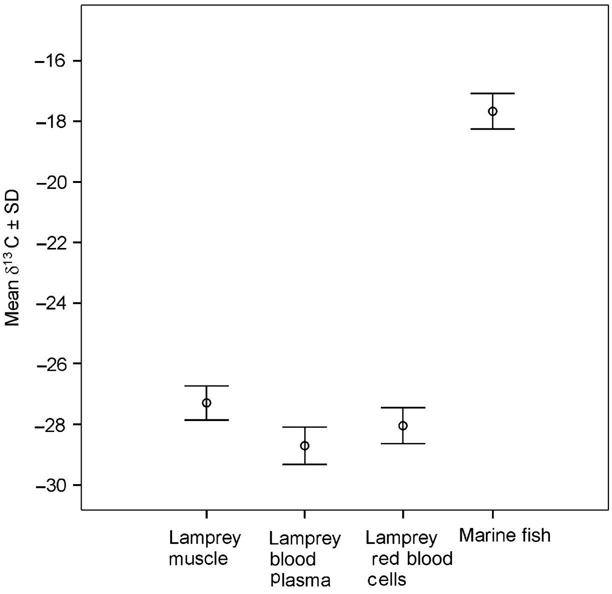

Fig. 5. $\delta^{13}C$ values of lamprey muscle, blood plasma and red blood cells and muscle samples from marine fish, taken from Etheridge *et al.* 2008. The lower values for the lamprey tissue are indicative of tissue synthesized in a freshwater habitat.

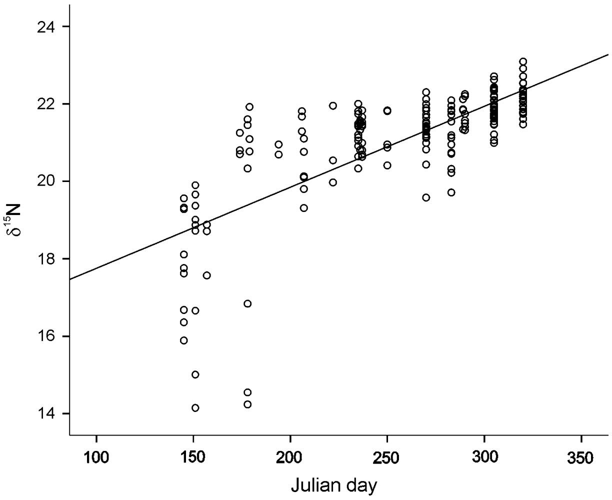

Fig. 6. Linear regression of $\delta^{15}N$ from all lamprey tissue samples with Julian date, $R^2 = 0{\cdot}53$.

mass ($F_{[1,186]} = 121{\cdot}34$, $P < 0{\cdot}001$), site and Julian date ($F_{[1,186]} = 8{\cdot}72$, $P < 0{\cdot}001$), and body mass and Julian date ($F_{[1,186]} = 89{\cdot}51$, $P < 0{\cdot}001$). The relationship between $\delta^{15}N$ and time is particularly strong ($r^2 = 0{\cdot}538$, $P < 0{\cdot}001$) with $\delta^{15}N$ increasing with time (Fig. 6).

ISOTOPIC MIXING MODELS

Isotopic values from all lamprey muscle, blood plasma and blood cell samples for each month (May $n = 18$, June $n = 15$, July, $n = 12$, August $n = 30$, September $n = 36$, October $n = 33$, November $n = 69$) were used in the SIAR mixing model. The use of multiple tissues (with different turnover rates) allows us to make robust estimates of diet, incorporating recent and longer-term foraging habits. The model utilizes all the available data and calculates the most likely range of proportions for each food source. Perch, eel, gudgeon, stickleback and lamprey were not included as potential prey items, because of very low levels of scarring (0 or < 1%). The results clearly demonstrate that the most important food sources for lamprey in Lough Neagh are trout and bream, with roach and pollan constituting a much smaller proportion of the diet (Fig. 7). SIAR produced a range of feasible solutions to the mixing problem to which we can then assign credibility intervals (CI), analogous to confidence intervals used in frequentist statistics. For example we can predict with 95% credibility that, in June, pollan represent between 0% and 40% of the diet (Fig. 7). Pollan contributes the least of the principal prey species to the lamprey's diet throughout the year, accounting for between 0% and 40% of the diet (95% CI) between May and July, and subsequently becoming a very minor prey item between August and November, representing between 0% and 12% of the lamprey's diet (95% CI). The proportion of roach drops between May (0–49%) and October (0–34%) and then increases again in November (4–36%) (95% CI). Trout represented the most important food source in May (3–60%), and continued to be a major source until October (2–48%), but were less important in November (0–24%) (95% CI). The proportion of bream in the diet increased throughout the year, becoming the largest single prey item in the diet from June until November, at which point it represents between 50% and 77% of the diet (95% CI).

Discussion

Our work makes several important contributions: first, we show that a native predator, the river lamprey, is relying on both native prey of conservation importance (pollan) and non-native prey that have been introduced to the water body. The reliance on non-native prey increases later in the year, suggest-

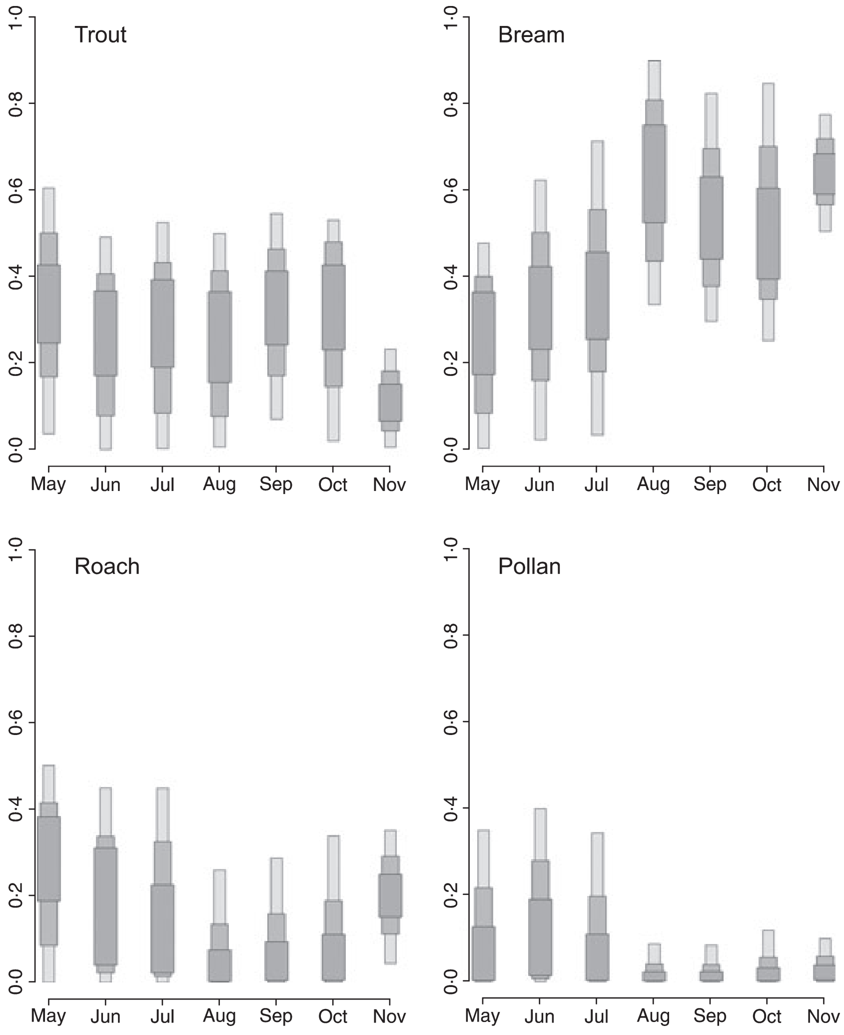

Fig. 7. Proportions of each food source in the lamprey diet predicted from stable isotope analysis mixing models. Each plot shows proportions for each prey species by month from May to November. Boxes indicate 50% 75% and 95% Bayesian credibility intervals.

ing that the predator population may be sustained at higher numbers than would otherwise be the case and thus there is potential for hyperpredation on the native pollan. Secondly, we have identified a rare potamodromous population of river lamprey in Lough Neagh which is of significant conservation interest in itself.

With respect to potamodromy, stable isotope analysis of three different tissues (with different temporal turnover rates) from 71 lampreys, sampled from May until November, provide compelling evidence that none of this sample had foraged in a marine environment. It remains possible that an unsampled proportion of the population does migrate to the sea, though our sampling of lamprey at all locations of the lough and over a protracted season would suggest otherwise.

River lamprey in Lough Neagh depend to a large extent on native trout and non-native bream and roach and to a much lesser extent on native pollan. It is clear that lampreys are selective feeders as our analyses suggest rates of predation that are markedly out of proportion to the relative abundance of species as sampled by seine netting. This relies on the assumption that our sampling protocol is unbiased, and we have no reason to believe that this is not the case. This finding supports recent work demonstrating active prey selection by invasive sea lamprey *Petromyzon marinus* (Harvey, Ebener & White 2008) but is in contrast to previous studies that suggest lamprey are opportunistic feeders (Swink 1991), selecting prey items based on size only.

Although pollan were the second most abundant fish species in the Lough, they formed only a small proportion of lamprey diet. Previous work has identified pollan as lamprey prey in Lough Neagh (Kennedy & Vickers 1993; Harrod 2001; Goodwin *et al.* 2006). Goodwin *et al.* (2006) found 0–13% of sampled pollan were scarred whilst Harrod *et al.* (2001) found 1·2% scarred. These previous studies relied exclusively on fish scarring data as a proxy for lamprey predation and such data are less likely to reflect accurately the diet of lamprey, as they only represent individuals that have survived a lamprey attack. Nonetheless, scarring data from the gill-net fishery in both this and previous work (Goodwin *et al.* 2006) showed striking similarities with the results of our isotopic mixing model, both methods indicating that lamprey consume most pollan in May–July.

Our mixing model indicates that the proportion of roach in the diet is consistently higher than that of pollan, although scarring levels are lower, possibility because of higher mortality rates in the former. By contrast, brown trout are much less abundant than either roach or pollan and yet they formed a higher proportion of the lamprey's diet. Their larger size and tendency to swim near the surface may make them easier targets for lamprey. High rates of trout consumption are also supported by the high rates of scarring among trout in this study (mean = 32·1%, range = 25–40%) and previous reports (43%; Kennedy & Vickers 1993).

Perhaps the most surprising result is the importance of bream in the diet of lamprey. Both scarring and stable isotope data suggest that bream are important for lamprey, and both indicate the increased contribution of bream to the diet later in the year. This pattern is consistent with the increase in lamprey $\delta^{15}N$ values from May to November, and the significant correlation with lamprey size. As lamprey grow it appears they undergo ontogenetic shifts in their diet and forage on increasingly large species of fish, that are themselves feeding at a higher trophic level. We suspect that the lower $\delta^{15}N$ values (lying below the regression line) earlier in the season (Fig. 6) represent remnants of the immature filter-feeding stage of the lamprey life cycle, which feed at lower trophic levels. Previous studies (Goodwin *et al*. 2006) have suggested that the absence of scars on pollan later in the year may be as a result of seasonal, vertical feeding migrations of lamprey (Berg 1948; Applegate 1950). This suggestion is consistent with our observation of seasonal variation in consumption of pollan, furthermore our data indicate that later in the year lamprey switch from pollan to bream, which are generally benthic foragers (Persson & Brönmark 2002).

Overall it is clear that a large proportion of lamprey diet consists of invasive, non-native species (roach and bream). This pattern of trophic interactions has two potential interpretations for the threatened native pollan population. First, the presence of abundant non-native species may act to decrease lamprey predation pressure on pollan. Alternatively, the abundance of non-native species may sustain artificially high lamprey numbers resulting in hyperpredation of pollan.

Predation of invasive, non-native species by native predators is often considered as beneficial as predation may limit the abundance of invaders (Levine, Adler & Yelenik 2004; Noonburg & Byers 2005). Invasive prey, however, can also act as a supplementary resource for predators, which may facilitate an increase in predator numbers, subsequently impacting native prey populations. This process, termed hyperpredation (Smith & Quin 1996; Courchamp *et al*. 1999), can have severe detrimental consequences for native species (Roemer *et al*. 2001; Roemer, Donlan & Courchamp 2002). In this specific situation, interspecific competition with invasive, non-native species has previously been identified as a threat to the native pollan population (Whilde 1993; Rosell 1997; Harrod *et al*. 2001), though it has not previously been suggested that roach and bream interact with pollan numbers via predation by lamprey.

It is also possible that the abundance of non-native prey species has enabled Lough Neagh lamprey to achieve a larger body size (potentially resulting in a greater level of fatal attacks). Other freshwater-feeding lamprey populations tend to be smaller than sympatric anadromous lamprey (Berg 1948; Abou-Seedo & Potter 1979; Maitland *et al*. 1994; Adams *et al*. 2008). Stable isotope analysis of the Loch Lomond population demonstrated that a larger morph is anadromous, whereas the smaller morph feeds only in freshwater (Adams *et al*. 2008). It has been suggested that the smaller morph is the result of slow growth caused by feeding in freshwater (Potter & Beamish 1977). The freshwater lampreys found within Lough Neagh are of typical size for the species, and we found no evidence for a smaller body size morph. Whether this is a result of the fish community of Lough Neagh containing larger species on which larger lamprey can feed remains a matter of conjecture, but presents a clear path for future research.

This study has two distinct implications for managers. The potamodromous river lamprey of Lough Neagh is one of only a few such populations in the world. This should be taken into account in evaluating the contribution lamprey make to biodiversity of Lough Neagh and of Ireland more generally.

Understanding the trophic relations between lamprey and pollan is made much harder with the knowledge acquired here of the significance of invasive, non-native prey to lamprey. At face value the short-term, and low frequency of lamprey consumption of pollan suggests that pollan are not a particularly important food source for lamprey. Thus the local commercial pollan fishery appears unlikely to impact directly upon lamprey populations. Conversely, it is much harder to gauge the impact of lamprey predation on pollan. Although pollan are consumed less than roach or bream, if these species are sustaining higher lamprey populations native pollan and trout populations may experience higher rates of predation than would be the case if roach or bream were absent. Hypothetically, targeted removal of non-native species from the Lough would, in due course, reduce lamprey populations. However, in the period when roach or bream were becoming less abundant and lamprey were short of food, predation of pollan and trout is likely to increase. The consequences of management interventions therefore remain very hard to predict. Basic modelling of the population dynamics of these populations may reveal a range of management options though these will require better knowledge of both fish and lamprey populations.

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