Biosecurity and Vector Behaviour: Evaluating the Potential Threat Posed by Anglers and Canoeists as Pathways for the Spread of Invasive Non-Native Species and Pathogens

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Abstract

Invasive non-native species (INNS) endanger native biodiversity and are a major economic problem. The management of pathways to prevent their introduction and establishment is a key target in the Convention on Biological Diversity's Aichi biodiversity targets for 2020. Freshwater environments are particularly susceptible to invasions as they are exposed to multiple introduction pathways, including non-native fish stocking and the release of boat ballast water. Since many freshwater INNS and aquatic pathogens can survive for several days in damp environments, there is potential for transport between water catchments on the equipment used by recreational anglers and canoeists. To quantify this biosecurity risk, we conducted an online questionnaire with 960 anglers and 599 canoeists to investigate their locations of activity, equipment used, and how frequently equipment was cleaned and/or dried after use. Anglers were also asked about their use and disposal of live bait. Our results indicate that 64% of anglers and 78.5% of canoeists use their equipment/boat in more than one catchment within a fortnight, the survival time of many of the INNS and pathogens considered in this study and that 12% of anglers and 50% of canoeists do so without either cleaning or drying their kit between uses. Furthermore, 8% of anglers and 28% of canoeists had used their equipment overseas without cleaning or drying it after each use which could facilitate both the introduction and secondary spread of INNS in the UK. Our results provide a baseline against which to evaluate the effectiveness of future biosecurity awareness campaigns, and identify groups to target with biosecurity awareness information. Our results also indicate that the biosecurity practices of these groups must improve to reduce the likelihood of inadvertently spreading INNS and pathogens through these activities.

Introduction

Invasive non-native species (INNS) are a primary driver of biodiversity loss and a major economic problem, with management and mitigation costing an estimated US$120 billion in the USA [1], US$6.3 billion in Australia [2] and US$2.6 billion in the UK each year [3]. Their ecological impacts range from habitat degradation, to competition with native species, to the introduction of pathogens and disease[4–6]. As the eradication of an established INNS is rarely possible [7,8], preventative management is an important and cost effective control strategy [9]. To this end, the management and prevention of INNS introductions is recognised as a global priority for biodiversity conservation and is listed as one of the Convention on Biological Diversity's (CBD) Aichi biodiversity key targets for 2020 [10].

Freshwater ecosystems are particularly vulnerable to INNS [11]. They are exposed to a wide range of transmission pathways including fish stocking, the redirection of water supplies, release of boat ballast and bilge water, release of exotic and ornamental plant and animal species, and the transfer of recreational angling and boating gear between sites [11–16]. Recent research indicates that fishing, boating and leisure activities are collectively responsible for almost 40% of aquatic species introductions into Europe [17]. The management of vectors such as these is considered to be one of the most effective strategies to prevent introduction and spread of invaders since numerous INNS threats can be controlled simultaneously [18,19].

In the UK, freshwater ecosystems contain seven of the UK Environment Agency's 10 'most wanted' invasive species [20] and are thought to be threatened by a further 11 [17]. Despite the estimated 4 million recreational anglers [21] and 404,000 canoe owners [22] in the UK, these groups have received little attention with regard their potential role in the introduction and secondary spread of aquatic invasive species and pathogens.

Since many INNS can survive in damp environments for a number of days or even weeks (Table 1), the potential exists for their introduction and spread between catchments on wet equipment used by anglers and boaters [12,23]. Prominent

examples include the zebra mussel (*Dreissena polymorpha*) introduced to Ireland on the hulls of boats [26]; the pathogen *Aphanomyces astaci* (causative agent of crayfish plague) vectored in mud and on damp angling gear [23,24]; and the killer shrimp (*Dikerogammarus villosus*) which is able to survive for up to 15 days on damp angling equipment [25].

The aim of this study is to investigate the biosecurity risk posed by recreational anglers and canoeists in the UK. Specifically, we aim to i) identify potential pathways of INNS introduction and spread, ii) identify 'higher risk' groups who should be targeted with biosecurity guidance and iii) provide a baseline against which to evaluate the effectiveness of future biosecurity advice.

Materials and Methods

Ethics Statement

The questionnaire satisfied the University of Leeds' guidelines on ethical conduct (Ethics reference BIOSCI 12-043). All data was collected, stored and analysed anonymously. Respondents were asked for two items of demographic data (age group and sex) but no data was collected that would enable any respondent to be identified.

An online questionnaire survey was conducted using Bristol Online Surveys software [26]. The secretaries of 316 angling clubs and 241 canoeing clubs in England were contacted from listings in the UK Environment Agency's 'Where to Fish?' guides, online angling club databases and the British Canoe Union's list of canoe clubs, and asked to circulate the questionnaire to their members.

Anglers and canoeists were asked about the type and frequency of angling/canoeing carried out. In order to gain a representative overview of how far each respondent typically travels to take part in their sport, they were asked to list the three sites that they fish/canoe at most recently and the three sites that they fish/canoe at most frequently. The six sites were geocoded into latitude and longitude coordinates with Python's *Geopy* toolbox, using the Google Maps API. The catchment that each site fell into was identified using the *Extraction* tool in the ArcGIS Spatial Analyst extension within the ArcGIS 10.1 [27] Geographic Information System software, according to the European Union Water Framework Directive catchment areas.

Respondents were asked about the equipment used during each trip, and how frequently it was i) dried and ii) cleaned after use; iii) which cleaning products were used, if any; and iv) whether equipment had been used overseas, and in which countries. Anglers were also asked about their use of live bait (invertebrates and fish). Canoeists were asked about the factors that influenced whether they cleaned and dried their equipment at the end of each trip. To do this, they were asked to score each of 6 factors on a likert scale from 1 (not important) to 5 (very important) depending on how these factors influenced their decision to clean equipment after use. The factors were: i) availability of a hose, ii) cost of cleaning products, iii) time taken to clean boat, iv) availability of information about how to clean boat and v) how dirty their boat appeared to be. Canoeists were also asked if they were aware of the 'Check, Clean, Dry' biosecurity campaign launched by the UK Department for Environment, Food and Rural Affairs (Defra)

Table 1. Approximate survival times of notifiable freshwater pathogens listed by the World Organisation for Animal Health (OIE) and freshwater INNS listed in the Environment Agency's 10 'most wanted' invasive species or as one of the potential invaders threatening Great Britain and Ireland [17].

Species	Survival time outside host (pathogens) or in damp conditions (INNS)		Reference
PATHOGENS			
Gyrodactylus salaris	2–5 days	R	[53]
Koi herpes virus	3 days	P	[54]
White spot syndrome virus	3–4 days	R	[55]
Aphanomyces astaci	16 days	P	[56]
Batrachochytrium dendrobatidis	7 days	P	[57]
Amphibian ranaviruses	1 month	P	[58]
Infectious haematopoietic necrosis	1 month	R	[58]
Spring viraemia of carp	5 weeks	R	[59]
Viral haemorrhagic septicaemia	49 days	R	[60]
INVASIVE NON-NATIVE SPECIES			
Topmouth gudgeon (*Pseudorasbora parva*)	Minutes (fish)	P	N/A
Zebra mussel (*Dreissena polymorpha*)	3–5 days	P	[61]
Signal crayfish (*Pacifastacus leniusculus*)	3–7 days	P	[62]
Killer shrimp (*Dikerogammarus villosus*)	15 days	P	[25]
Floating pennywort (*Hydrocotyle ranunculoides*)	No data available	P	
Parrots Feather (*Myriophyllum aquaticum*)	No data available	P	
Chinese mitten crab (*Eriocheir sinensis*)	No data available	P	
Ponto-caspian shrimp (*Echinogammarus trichiatus*)	6 days	R	[63]
Quagga mussel (*Dreissena rortriformis bugensis*)	3–5 days	R	[61]
Mean survival time (where known)	15 days		

P = species or pathogen is already present, R = this species poses a significant threat to UK freshwaters.
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in 2010 to see whether there were differences in the biosecurity practices of those who had and had not heard of the campaign. Respondents were asked about their awareness of the biosecurity campaign on the last page of the online questionnaire having already answered questions about their actions in order to minimise potential bias.

The answer options for closed-format questions were determined through consultation with Environment Agency Fisheries Officers, UK Rivers Trusts and biosecurity experts from the Centre for Environment, Fisheries and Aquaculture Science (Cefas). A prototype version of the questionnaire was also piloted with 15 anglers and 12 canoeists to prevent any misunderstandings, to check that the online questionnaire worked effectively and to ensure that sufficient answer options had been provided in the closed questions. The full questionnaire is available as supplementary material (Questionnaire S1).

Hazard Scores

In order to explore the relative biosecurity hazard posed by different groups of anglers and canoeists, respondents were scored against a set of criteria from 1 (low hazard) to 5 (high hazard) by interrogating them against a set of criteria (Table 2). The criteria were: the number of catchments visited (N), the frequency of the activity (F), the frequency of equipment cleaning (C) and the frequency of equipment drying (D). **Individual hazard score = N*F*C*D.**

By multiplying hazards together, interactions between factors were incorporated into the overall hazard score. As there is

Table 2. Scoring scheme for the criteria against which each individual was assessed in the hazard analysis.

Risk Factor	Description	Hazard Score
Frequency of angling	Once a month or less frequently	1
	Once every three weeks	2
	One a fortnight	3
	Once a week	4
	More than once a week	5
Number of catchments visited	1 catchment	0
	2 catchments	2
	3 catchments	3
	4 catchments	4
	5 or 6 catchments	5
Cleaning of equipment	After every trip	1
	Every 2–5 trips	2
	Every 6–10 trips	3
	Every 11+ trips	4
	Never	5
Drying of equipment	After every trip	1
	Every 2–5 trips	2
	Every 6–10 trips	3
	Every 11+ trips	4
	Never	5

Scores from 1–5 correspond to a hazard gradient from 1 (very low) to 5 (very high). As we were considering secondary spread between river catchments, respondents scored 0 if they said that they only visited one catchment.
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insufficient data available with which to inform the relative importance of the different risk criteria, we gave them equal weighting when calculating the hazard scores. As we were primarily investigating the potential role of anglers and canoeists in the secondary spread of INNS between UK catchments, anglers and canoeists who only visited one catchment scored zero. Regardless of how frequently they cleaned or dried their equipment, or how frequently they travelled, their total score would remain zero as they posed no likelihood of moving invasive species or aquatic pathogens to another catchment.

Results

Fifty two angling clubs and 70 canoeing clubs circulated the online questionnaire to their members. In total, 599 canoeists and 960 anglers completed the questionnaire (response rates 17% and 25%, respectively).

Following best practice advice of White et al. [28], our sample was verified using demographic information to ensure that respondents were representative of the angling/canoeing communities. Of the 960 angler respondents, 98% were men with the highest proportion of respondents aged between 55 and 64 (31%) followed by 45 and 54 (22%) and 65+ (17%). To ascertain whether our sample was representative of the UK angling population, a Kolmogorov–Smirnov test was performed in 'R' [29] to compare the age distribution of angler respondents to the age distribution of holders of Environment Agency rod licences in 2011- a requirement of all UK freshwater anglers. No significant difference was detected between the ages of the two groups (K-S Test, $D = 0.24$, $p > 0.05$), nor was there a significant difference between the sex ratios of the two groups, with 2% of angler respondents and 5% of rod license holders being female (K-S Test, $D = 0.3$, $p < 0.05$).

Seventy percent of the 599 canoeist respondents were men, a sex ratio which was not significantly different from British Canoe Union figures on the sex ratio of UK recreational boat users (British Canoe Union 2011) ($D = 0.7$, $p > 0.05$). Respondents were from a broad range of age groups, with 35–44 year old and 45–54 year old groups with the greatest number of respondents (16.4% and 26%, respectively). Unfortunately, data on the age profiles of UK canoeists were unavailable.

Respondents were broadly spread across different angling and canoeing categories. Forty four percent of anglers were coarse anglers (typically pleasure anglers using rods from the bank and catching any freshwater fish other than game fish), 25% were game anglers targeting trout, 13% were match anglers (typically angling in heavily stocked commercial fisheries and frequently travelling to different sites to attend competitions), 7% were barbel anglers, 6% were game anglers targeting salmon and 5% were pike anglers. Almost half of canoeist respondents (47%) were recreational canoeists using rivers while 21% canoed on lakes, 19% were competitive canoeists, 11.9% were sea kayakers who also took part in freshwater canoeing and 1.5% were long distance touring canoeists.

Potential for Secondary Spread

Anglers visited a mean of 2.25 different UK catchments (Figure 1). There was a significant difference in the number of catchments visited by categories of angler (ANOVA $F_{5,954} = 9.56$, $p < 0.001$). Posthoc (LSD) tests revealed that salmon anglers – who visited a mean of 2.79 catchments - travelled significantly further than any other group of angler ($p < 0.05$). Canoeists visited a mean of 2.84 different catchments (Figure 1). There was a significant difference between the distances travelled by different

categories of canoeist (ANOVA, $F_{4,594} = 6.17$, $p < 0.001$), however, posthoc (LSD) tests revealed that touring canoeists - who travelled to the highest number of catchments (mean 3.33) - did not visit significantly more catchments than other groups ($p > 0.05$), while competitive canoeists did visit significantly fewer catchments than the other groups (mean of 2.53, posthoc tests $p < 0.05$ when compared to each of the other groups).

Sixty four percent of anglers and 79% of canoeists used their equipment in more than one catchment within a fortnight (Table 3), the mean duration that pathogens/INNS listed in Table 1 survived in damp conditions. Forty nine percent of canoeists and 12% of anglers visited more than one catchment within a fortnight and neither cleaned nor dried their kit between uses (Table 3). The geographic movements of these higher risk anglers and canoeists are displayed in Figure 2.

Of the 614 anglers and 470 canoeists who used their equipment in more than one catchment within a fortnight, 22% of anglers and 10% of canoeists cleaned their kit after every use, 80% of anglers and 33% of canoeists dried their kit after every use and 21% of anglers and 6% of canoeists both cleaned and dried their kit after every use, the biosecurity advice recommended by Defra (Figure 3). Of the anglers who cleaned their kit after each use, 49% used tap water, 31% used disinfectant and 30% used detergent. Of the canoeists who cleaned their kit each time, 81% used tap water, 15% used detergent and 4% used disinfectant.

A large proportion of the anglers who travelled to more than one catchment within a fortnight without cleaning equipment after each trip used equipment associated with INNS/pathogen accumulation: rubber or felt soled waders (used by 36% and 4%, respectively) and keep nets (used by 25%).

Hazard scores

Overall, anglers had lower hazard scores than canoeists, due to the higher proportion drying equipment, and the lower number of catchments visited (Table 3).

When different types of angler were compared, the median hazard scores of different groups were significantly different (Kruskal Wallis H = 29.80; 5 df; $p < 0.001$). Salmon anglers had the highest average hazard score, followed by trout anglers while pike and barbel anglers had the lowest (Table 4). In contrast, there was no significant difference between the hazard scores of different categories of canoeist (Kruskal Wallis H = 2.086; 4df; $p > 0.05$).

However, competitive canoeists had the highest mean hazard score and touring canoeists had the lowest (Table 3).

Canoeists were asked about the factors affecting whether they cleaned equipment after use. The availability of a hose was the most important factor (mean score 3.1 out of 5) followed by time availability (mean score 2.86 out of 5). The least important factors were the cost of cleaning products and the availability of information about how to clean equipment (both scored a mean of 1.9 out of 5). The 22 percent (130) of canoeist respondents who had heard of the 'Check Clean Dry' campaign exhibited biosecurity hazard scores that were 40% lower than those who had not (Kruskal-Wallis: H = 10.99; df 1; $p < 0.001$).

Potential for Introduction to the UK

A large proportion of anglers (53%) and canoeists (46%) had used their equipment overseas, trips of between 260 km and 9500 km from their last site in the UK (excluding Ireland). The majority visited other countries within Europe (84% of anglers and 96% of canoeists) although 20% of respondents had used their equipment in North America and 7% in Australasia.

Within Europe, France and Ireland were the most popular angling destinations (visited by 17% and 16% of anglers, respectively). France and Austria were the most popular canoeing destinations (visited by 69% and 20% of travelling canoeists, respectively). Three percent of anglers had used their equipment in Norway where the salmon louse *Gyrodactylus salaris* poses a particular biosecurity threat to the UK [30].

Ninety nine percent of the 446 anglers who used their equipment overseas and went angling at least once a fortnight, failed to clean their equipment after every use, and 18% neither cleaned nor dried their equipment after every use. Moreover, 29% of the anglers who travelled overseas, fished at least once a fortnight and neither cleaned nor dried their equipment between uses used rubber waders, 5% used felt-soled waders and 19% used keep nets. Of the 241 canoeists who used their equipment overseas and at least once a fortnight and used their equipment overseas, 94% failed to clean it after every use, 71% failed to dry it after very use and 69% neither cleaned nor dried it after use.

Use of Live Bait

Three hundred and seventy five of 960 angler respondents (39%) used live bait. Of those, 34% indicated using maggots, 34%
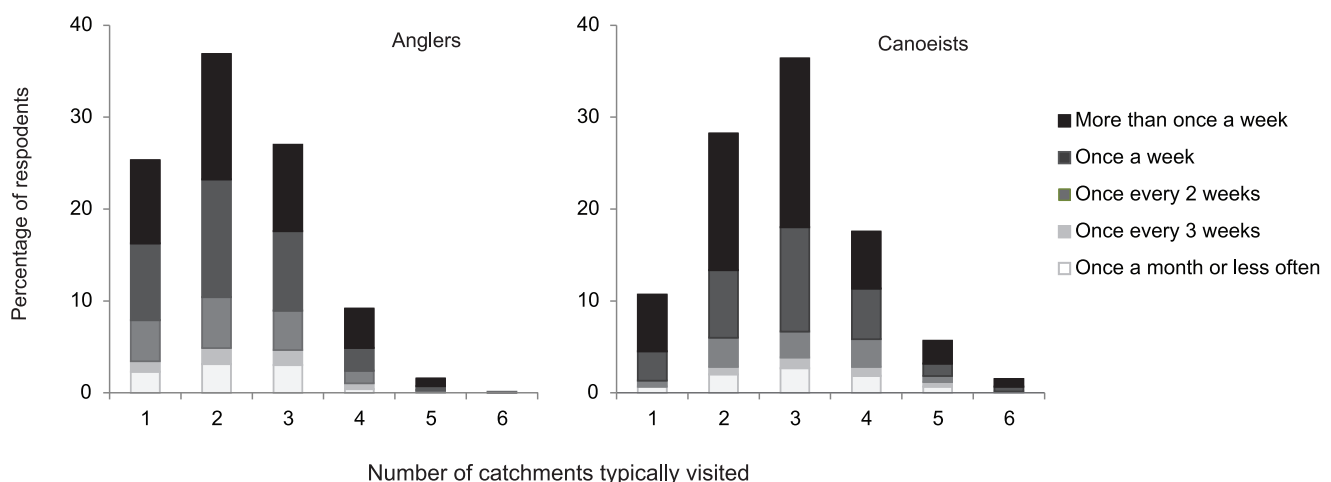

Figure 1. Typical number of UK catchments visited by canoeists and anglers. Shading shows the frequency with which respondents travelled between the catchments that they visited.
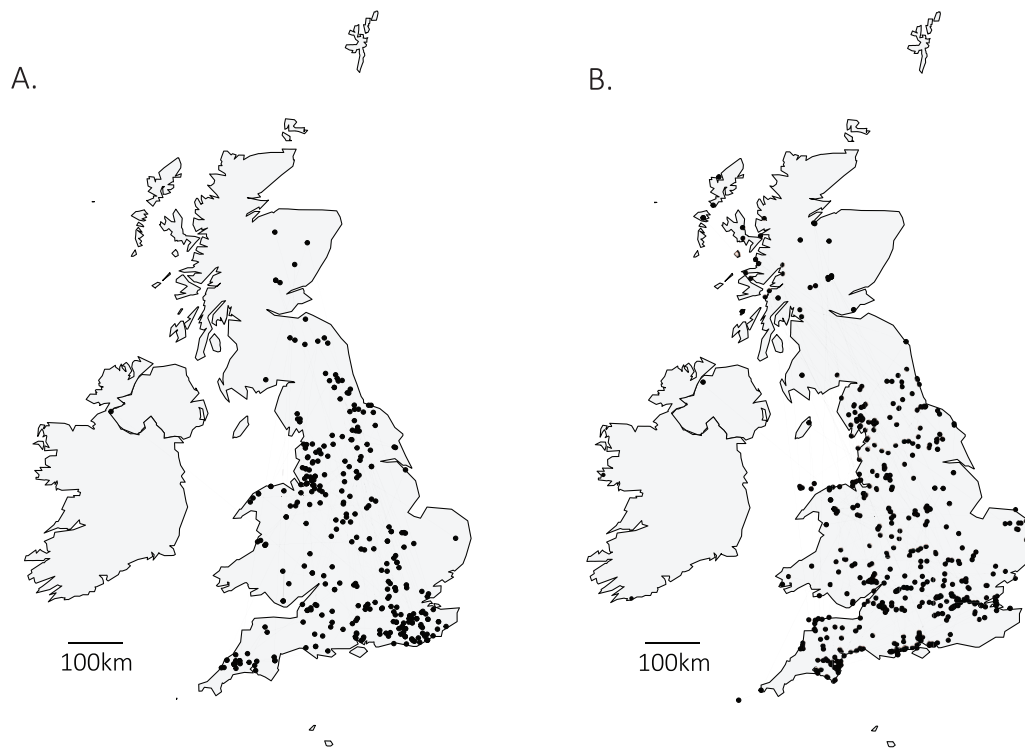doi:10.1371/journal.pone.0092788.g001

Figure 2. Maps showing the last three UK sites visited and by A) anglers and B) canoeists who visited more than one catchment within a fortnight and failed to clean or dry their kit between uses. Points show the sites and lines connect sites visited by an individual within a fortnight.
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indicated using bait fish, 23% indicated using earthworms and 18% indicated using bloodworms. The most commonly used bait fish were roach (*Rutilus rutilus*), rudd (*Scardinius erythrophthalmus*), perch (*Perca fluviatilis*), minnows (*Phoxinus phoxinus*) and gudgeon (*Gobio gobio*). The use of live bait was highest amongst pike anglers (47%) and barbel anglers (44%). Live bait use was lowest among trout anglers (36%).

Although the source varied between bait types (Table 4), the majority of bait users sourced bait from angling shops. Catching or collecting bait was the second most popular source of live bait. Of the 140 anglers who caught their own bait, fish and earthworms were most commonly sourced (77% and 17%, respectively) (Table 4). Baitfish was most commonly caught at the same site that the angler intended to use it (84%). However, 16% of anglers collected their bait at a different site from where it was to be used and 7% released unused bait which had been from a different site into the river/lake after use (Figure 4).

One hundred and forty bait users (37%) released unused bait into the water body at the end of their angling trip including bloodworms (9%) and baitfish (63%). Although the majority of anglers who released unused baitfish into the water had caught their fish at the same site, posing no biosecurity hazard, three had caught their baitfish at another site and one angler released bait fish sourced from a bait dealer. In addition, 9% of bait users released unused bait onto the land. This included 18 anglers who released unused earthworms on the river bank after use, all of which had been sourced from a different site to where they were released.

Discussion

This is the first study exploring the potential biosecurity risk posed by anglers and canoeists in the UK. Our results highlight the fact that a high proportion of anglers and canoeists use their equipment frequently, and in multiple UK catchments, within the time that a range of INNS and aquatic pathogens can survive in damp conditions. This coupled with the low frequency with which anglers and canoeists clean and dry their equipment suggests that these groups may have the potential to act as vectors for their spread. The results are in accord with studies in North America which showed that anglers and boaters were travelling hundreds of kilometres between sites [31,32] and were frequently transporting muddy waders [31], and the remnants of invasive aquatic plants on their boats [33,34]. Within this research, we investigated the movement patterns of anglers and canoeists to evaluate the biosecurity risk that they would pose should they become contaminated with INNS or pathogens. We acknowledge that not all anglers and canoeists will use their equipment in a water body which has INNS present and of those who do, a low proportion of boats or equipment will become contaminated [35]. However, the large number of anglers (4 million) and canoe owners (404,000) in the UK and the frequency with which these groups appear to be using their equipment at different sites suggest that these groups pose an important pathway for the spread INNS should their equipment become contaminated.

Table 3. Relative hazard scores for different categories of angler and canoeist.

Category	Median hazard score (0–625). Brackets indicate inter-quartile range.	% travelling to 2+ catchments	% travelling to 2+ catchments AND doing activity ≥ once per fortnight	% travelling to 2+ catchments AND doing activity ≥ once per fortnight AND not cleaning their equipment after every use	% travelling to 2+ catchments AND doing activity ≥ once per fortnight AND not drying their equipment after every use	% travelling to 2+ catchments AND doing activity ≥ once per fortnight AND neither cleaning nor drying their equipment after every use	% doing activity ≥ once per fortnight AND neither cleaning nor drying their equipment after every use AND using their equipment overseas
Game anglers (salmon)	40 (IQR 60)	88.5	80.3	63.9	19.7	19.7	8.2
Game anglers (trout)	30 (IQR 60)	82.6	68.6	56.8	14.0	13.6	8.1
Match anglers	20 (IQR 64)	74.2	72.6	52.4	15.3	12.9	8.9
Coarse anglers	16 (IQR 50)	69.6	56.8	45.1	11.9	11.2	9.0
Pike anglers	16 (IQR 52.5)	75.0	66.7	47.9	6.3	6.3	4.2
Barbel anglers	13.5 (IQR 60)	67.1	60.0	40.0	4.3	4.3	5.7
All anglers (n = 960)	20 (IQR = 50)	74.7	64.0	49.9	12.5	11.9	8.23
Competitive canoeists	100 (IQR 210)	85.6	83.8	55.0	79.3	52.3	36.0
River canoeists	96 (IQR 195)	89.4	74.6	54.1	68.6	50.9	31.1
Lake canoeists	80 (IQR 226)	87.2	77.6	50.4	68.6	48.8	19.2
Sea kayakers	75 (IQR 197.5)	97.2	85.9	47.9	67.6	40.8	18.3
Touring canoeists	60 (IQR 110)	100	88.9	44.4	77.8	44.4	11.1
All canoeists (n = 599)	80 (IQR = 220)	89.3	78.5	52.6	70.6	49.5	27.7

IQR = Interquartile range. Percentages show the co-occurrence of biosecurity hazards associated with potential transmission.
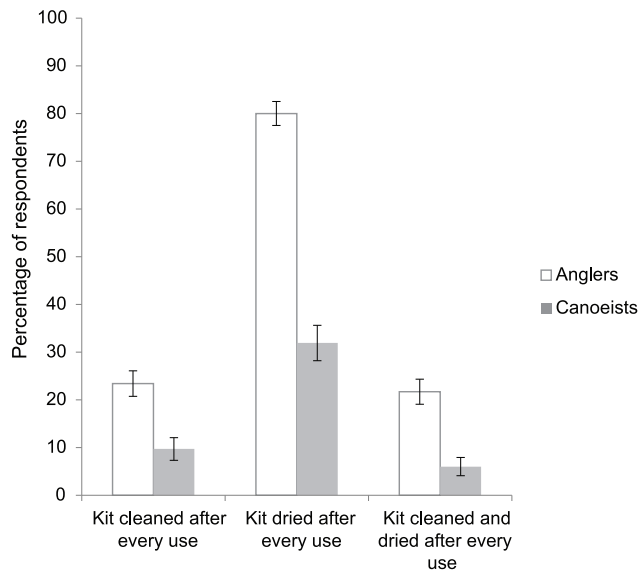doi:10.1371/journal.pone.0092788.t003

Figure 3. Percentage of anglers and canoeists who visited more than two catchments within a fortnight and who either cleaned, dried or cleaned and dried their equipment after every use. Error bars show 95% Confidence Interval.
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Potential for Secondary Spread

Fewer canoeists cleaned or dried equipment than anglers and only a small proportion of both groups used disinfectant. The low proportion of canoeists cleaning their equipment reflected the behaviour of boat users at Lake Simcoe, Canada where 19% rinsed their boats after use and only 3.2% allowed them to dry out for at least 5 days between uses [35]. Despite the seemingly high proportion of anglers drying equipment after use, complete drying is required in order to kill INNS and pathogens through desiccation [36,37]. Considering the frequency with which anglers used their equipment it seems improbable that complete desiccation would have been achieved between trips.

The high frequency with which respondents took part in their activity may be an artefact of distributing the questionnaire to angling and canoeing clubs; themselves hubs of particular enthusiasts. Questionnaire surveys distributed in an 'opt in' manner can lend themselves to self-reporting bias [28], however
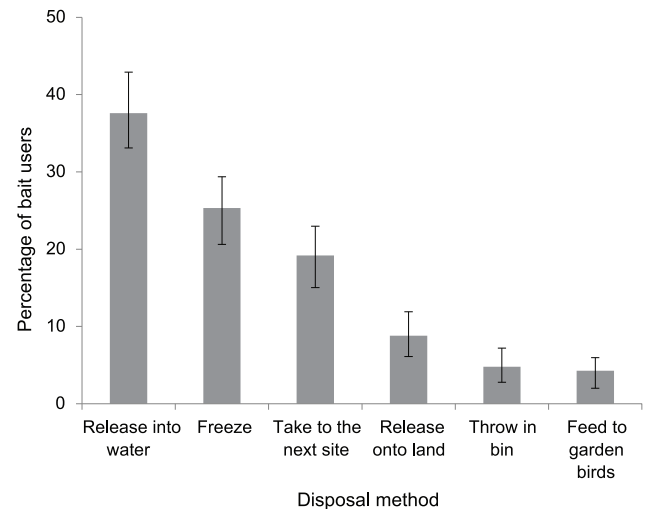

Figure 4. Disposal methods for live bait (fish and invertebrates) used by anglers. Error bars show 95% Confidence Interval.
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due to data protection restrictions preventing us from contacting individuals in a more structured manner, this was an effective way of obtaining a large sample size. While care was taken to design an un-biased survey, we recognise that when people are asked about their individual actions in an environmental context, some 'good behaviour' bias may exist.

Potential for Introduction to the UK

A high proportion of anglers and canoeists used their equipment overseas, primarily elsewhere in Europe. Moreover, a low proportion of both groups cleaned their equipment between uses – actions which may risk the inadvertent introduction of new aquatic invaders and pathogens from overseas. A number of the species mentioned in Table 1 such as the salmon ecto-parasite *Gyrodactylus salaris* and invasive amphipod *Dikerogammarus villosus* pose a hazard to native species in the UK, yet exist in regions of Europe where anglers and canoeists had used their own equipment and failed to clean it after each use (Table 3) [30,38].

Countries such as New Zealand communicate a strong biosecurity message to water users at the international border, insisting that anglers and boaters check and disinfect their

Table 4. The source and method of disposal of live bait by anglers.

		Bloodworms	Maggots	Earthworms	Fish
Source of bait	Bait dealer	22.1	4.8	7.1	2.4
	Catch own	8.8	10.3	29.4	90.5
	Purchased at fishery	4.4	0.8	0.0	2.4
	Tackle shop	64.7	84.1	63.5	4.8
Disposal method	Release into water	17.6	29.4	22.4	64.5
	Release onto land	7.4	7.9	21.2	0.8
	Freeze	27.9	18.3	18.8	27.4
	Throw in bin	7.4	4.0	5.9	3.2
	Take to next site	33.8	32.5	29.4	4.0
	Feed to garden birds	5.9	7.9	2.4	0.0

Figures show percentages for the source and disposal of each type of bait.
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equipment before entering the country, as well as banning the use of hazardous equipment such as felt soled waders [39]. Considering the high proportion of UK water users who appear to use equipment overseas, there is likely to be a benefit from communicating a similar biosecurity message to these groups at UK ports and airports.

Use of Live Bait

Release of bait fish from another site or from a bait dealer, the release of unused aquatic bloodworms from an unknown source and the disposal of earthworms on banksides after use pose clear biosecurity hazards. In the USA, the release of live bait has been the third largest source of non-native fish introductions [40] as well as being a major vector for the spread of invasive earthworms which have been associated with changing soil composition and local extinction of native plants [41].

The number of anglers who released bait fish having caught them at another site or from a bait dealer was much lower in our study than a comparable study in Maryland, USA, where 65% of anglers released unused bait which was frequently (and illegally) non-native crayfish [42]. Nonetheless, the release of unused bait fish by UK anglers is a recognised practice [43], and previous introductions of bait fish in the UK have been negatively correlated with the abundance of native fish species [43]. The release of bait is still therefore a potential route by which invasive species could be moved between UK catchments and one which needs controlling. Although more than a decade old, a study of anglers' attitudes to conservation in 2001 showed that only 19% of anglers saw the release of non-native bait fish as a conservation concern [44], despite the fact that failure to prevent the introduction of an invasive species risks prosecution under the Wildlife & Countryside Act, 1981.

The control of bait movement is also important to control the indirect spread of associated INNS including *A.astaci* zoospores, zebra mussels (*Dreissena polymorpha*) and Asian clams (*Corbicula fluminea*) which can be transported via the gastrointestinal tract of fish [45,46] and may be moved between sites by anglers using live bait fish [47].

Biosecurity

Our results indicate that in addition to the management of INNS vectors such as boat ballast water, and fish stocking, effective biosecurity practices are required to reduce the likelihood of INNS spread by recreational water users such as canoeists and anglers. In Europe, a large proportion (36%) of non-native species introductions are thought to have been due to fishing, boating and leisure activities [48]. Our results suggest that angler and canoeist activities pose a potential pathway for the spread of INNS or pathogens. As evidence indicates that pre-emptive management is effective at reducing the likelihood of aquatic INNS invasion via anthropogenic pathways [49], improving the biosecurity practices of these groups is important.

The lower proportion of canoeists cleaning/drying equipment after use as well as the higher proportion visiting three or more catchments was reflected in canoeists having higher biosecurity hazard scores than anglers. When canoeists were asked about factors affecting their cleaning/drying behaviours, the most important factor was the availability of a hose or cleaning station. The provision of more cleaning stations in 'hot spot' locations where boat and angling traffic is highest could therefore be fundamental to improving biosecurity practices.

Amongst anglers, we found game anglers, fishing salmonids, and match anglers to have the highest biosecurity hazard scores. Within the canoeing community, competitive canoeists and river

canoeists had the highest hazard scores These groups were characterised by frequent canoeing/angling trips, often several times a week, visits to multiple catchments, and a low proportion of individuals cleaning and/or drying their equipment after every use. Biosecurity information should be targeted towards these groups of anglers, but we recognise that there was a lot of variation in biosecurity hazard scores within groups of canoeists so focusing biosecurity information towards specific types of canoeists may be less effective.

The UK Department for Food, Environment and Rural Affairs (Defra) launched a 'Check, Clean, Dry' biosecurity awareness campaign in 2010 in response to the first reports of the killer shrimp *Dikerogammarus villosus* in the UK [50]. The campaign was based on a biosecurity campaign in New Zealand designed to prevent the secondary spread of didymo (*Didymosphenia geminata*). In New Zealand, 80% of recreational water users were aware of the national 'Clean, Check, Dry' campaign and the spread of didymo appears to have slowed since the campaign was launched [39]. Our results indicate that the UK 'Check, Clean, Dry' biosecurity campaign has only reached a small proportion of water users to date. Nonetheless, canoeists in our study who reported awareness of the campaign also exhibited lower biosecurity hazard scores.

Public engagement is vital to effectively manage INNS [51]. It is therefore important to engage with recreational water users to raise awareness and regularly evaluate the effectiveness of biosecurity campaigns, not only to ensure that they are having the desired effect, but to provide evidence to the public that their actions make a difference [52]. Our results highlight the need to increase biosecurity awareness among recreational water users; however it is important to engage with these groups so that they continue to enjoy their sport whilst taking biosecurity into account. We have provided an important baseline against which to monitor the effectiveness of future biosecurity awareness campaigns. Our data also identify groups who pose a higher biosecurity hazard, and should therefore be targeted as a priority.

Finally, our data on equipment use can inform experiments that evaluate the effectiveness of different decontamination measures to prevent the survival of INNS and pathogens on angling and canoeing equipment and our spatial data can used to parameterise network models to identify hotspot locations to target with biosecurity control measures.

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Supporting Information

Questionnaire S1 This table outlines the questions which appeared in the online questionnaire distributed to canoeists and anglers using Bristol Online Surveys software.
(DOCX)

Author Contributions

Conceived and designed the experiments: LGA PCLW PDS AMD. Performed the experiments: LGA. Analyzed the data: LGA. Wrote the paper: LGA PCLW PDS GDS AMD.

References

1. Pimentel D, Zuniga R, Morrison D (2005) Update on the environmental and economic costs associated with alien-invasive species in the United States. Ecological Economics 52: 273–288. doi:10.1016/j.ecolecon.2004.10.002

2. CSIRO (2009) Biosecurity and invasive species. Available: http://www.csiro.au/~/media/CSIROau/Divisions/CSIRO%20Entomology/EntomologyBiosecurityOverview_Ento_PDF%20Standard.pdf. Accessed: 10 Mar 2013.

3. Williams F, Eschen R, Harris A, Djeddour D, Pratt C, et al. (2010) The economic cost of invasive non-native species on Great Britain. CABI Project No VM10066 CABI Europe-UK. Available: http://prerel.sgk.cabiorg.cab.semcs.net/uploads/projectsdb/documents/6534/Economic%20Costs%20of%20INNS%20to%20the%20British%20Economy%20-%20Final%20Report%20v3.pdf. Accessed: 24 Jul 2012.

4. Prenter J, MacNeil C, Dick JT., Dunn AM (2004) Roles of parasites in animal invasions. Trends in Ecology & Evolution 19: 385–390. doi:10.1016/j.tree.2004.05.002

5. Okamura B, Feist SW (2011) Emerging diseases in freshwater systems. Freshwater Biology 56: 627–637. doi:10.1111/j.1365-2427.2011.02578.x

6. Hatcher MJ, Dunn AM (2011) Parasites in ecological communities: from interactions to ecosystems. Cambridge, UK; New York: Cambridge University Press.

7. Mack RN (2000) Biotic invasions: causes, epidemiology, global consequences, and control. Ecological Applications 10: 689–710.

8. Kolar CS, Lodge DM (2001) Progress in invasion biology: predicting invaders. Trends in Ecology & Evolution 16: 199–204.

9. Caplat P, Coutts SR (2011) Integrating Ecological Knowledge, Public Perception and Urgency of Action into Invasive Species Management. Environmental Management 48: 878–881. doi:10.1007/s00267-011-9747-8

10. Secretariat of the Convention on Biological Diversity (2011) Strategic Plan for Biodiversity 2011–2020 and the Aichi Targets "Living in Harmony with Nature." Secretariat of the Convention on Biological Diversity.

11. Strayer DL, Findlay SEG (2010) Ecology of freshwater shore zones. Aquatic Sciences 72: 127–163.

12. Rahel FJ (2007) Biogeographic barriers, connectivity and homogenization of freshwater faunas: it's a small world after all. Freshwater Biology 52: 696–710. doi:10.1111/j.1365-2427.2006.01708.x

13. Keller RP, Zu Ermgassen PSE, Aldridge DC (2009) Vectors and Timing of Freshwater Invasions in Great Britain. Conservation Biology 23: 1526–1534. doi:10.1111/j.1523-1739.2009.01249.x

14. Havel JE, Shurin JB (2004) Mechanisms, effects, and scales of dispersal in freshwater zooplankton. Limnology and Oceanography 49: 1229–1238.

15. Oidtmann BC, Thrush MA, Denham KL, Peeler EJ (2011) International and national biosecurity strategies in aquatic animal health. Aquaculture 320: 22–33. doi:10.1016/j.aquaculture.2011.07.032

16. Jacobs MJ, MacIsaac HJ (2007) Fouling of fishing line by the waterflea Cercopagis pengoi: a mechanism of human-mediated dispersal of zooplankton? Hydrobiologia 583: 119–126.

17. Gallardo B, Aldridge DC (2013) The "dirty dozen": socio-economic factors amplify the invasion potential of 12 high-risk aquatic invasive species in Great Britain and Ireland. Journal of Applied Ecology. doi: 10.1111/1365-2664.12079

18. Briski E, Wiley CJ, Bailey SA (2012) Role of domestic shipping in the introduction or secondary spread of nonindigenous species: biological invasions within the Laurentian Great Lakes. Journal of Applied Ecology: n/a–n/a. doi:10.1111/j.1365-2664.2012.02186.x

19. Chan FT, Bailey SA, Wiley CJ, MacIsaac HJ (2012) Relative risk assessment for ballast-mediated invasions at Canadian Arctic ports. Biological Invasions. Available: http://www.springerlink.com/index/10.1007/s10530-012-0284-z. Accessed: 23 Jul 2012.

20. Environment Agency (2011) Biodiversity's Most Wanted. Available: http://www.environment-agency.gov.uk/news/132112.aspx.

21. Environment Agency (2004) Our nations' fisheries: The migratory and freshwater fisheries of England and Wales – a snapshot.

22. British Canoe Union (2011) Watersports and Leisure Participation Survey.

23. Taugbøl T, Skurdal J, H\aastein T (1993) Crayfish plague and management strategies in Norway. Biological Conservation 63: 75–82.

24. Reynolds JD (1988) Crayfish extinctions and crayfish plague in central Ireland. Biological Conservation 45: 279–285.

25. Fielding N (2011) Dikerogammarus villosus: preliminary trials on resistance to control measures. Freshwater Biological Association Newsletter 54.

26. University of Bristol (2013) Bristol Online Surveys.

27. Environmental Systems Research Institute (ESRI) (2012) ArcGIS. CaliforniaR-Redlands

28. White PCL, Jennings NV, Renwick AR, Barker NHL (2005) Questionnaires in ecology: a review of past use and recommendations for best practice. Journal of Applied Ecology 42: 421–430. doi:10.1111/j.1365-2664.2005.01032.x

29. R Development Core Team (2012) R: A Language and Environment for Statistical Computing. Vienna, Austria: R Foundation for Statistical Computing. Available: www.R-project.org.

30. Peeler EJ, Gardiner R, Thrush MA (2004) Qualitative risk assessment of routes of transmission of the exotic fish parasite Gyrodactylus salaris between river catchments in England and Wales. Preventive Veterinary Medicine 64: 175–189. doi:10.1016/j.prevetmed.2004.05.005

31. Gates KK, Guy CS, Zale AV, Horton TB (2009) Angler awareness of aquatic nuisance species and potential transport mechanisms. Fisheries Management and Ecology 16: 448–456. doi:10.1111/j.1365-2400.2009.00694.x

32. Drake DAR, Mandrak NE (2010) Least-cost transportation networks predict spatial interaction of invasion vectors. Ecological Applications 20: 2286–2299.

33. Johnson LE, Ricciardi A, Carlton JT (2001) Overland dispersal of aquatic invasive species: a risk assessment of transient recreational boating. Ecological Applications 11: 1789–1799.

34. Stasko AD, Patenaude AL, Strecker AL, Arnott SE (2012) Portage connectivity does not predict establishment success of canoe-mediated dispersal for crustacean zooplankton. Aquatic Ecology 49: 9–24.

35. Kelly NE, Wantola K, Weisz E, Yan ND (2012) Recreational boats as a vector of secondary spread for aquatic invasive species and native crustacean zooplankton. Biological Invasions 15: 509–519. doi:10.1007/s10530-012-0303-0

36. Jerde C, Barnes M, DeBuysser E, Noveroske A, Chadderton L, et al. (2012) Eurasian watermilfoil fitness loss and invasion potential following desiccation during simulated overland transport. Aquatic Invasions 7: 135–142. doi:10.3391/ai.2012.7.1.015

37. Poznańska M, Kakareko T, Krzyżyński M, Kobak J (2012) Effect of substratum drying on the survival and migrations of Ponto-Caspian and native gammarids (Crustacea: Amphipoda). Hydrobiologia 700: 47–59. doi:10.1007/s10750-012-1218-6

38. MacNeil C, Platvoet D, Dick J, Fielding N, Constable A, et al. (2010) The Ponto-Caspian "killer shrimp", Dikerogammarus villosus (Sowinsky, 1894), invades the British Isles. Aquatic Invasions 5: 441–445. doi:10.3391/ai.2010.5.4.15

39. Ministry for Primary Industries (2008) Check Clean Dry. Available: http://www.biosecurity.govt.nz/biosec/camp-acts/check-clean-dry.

40. Padilla DK, Williams SL (2004) Beyond ballast water: aquarium and ornamental trades as sources of invasive species in aquatic ecosystems. Frontiers in Ecology and the Environment 2: 131–138.

41. Keller RP, Cox AN, Van Loon C, Lodge DM, Herborg LM, et al. (2007) From bait shops to the forest floor: earthworm use and disposal by anglers. The American Midland Naturalist 158: 321–328.

42. Kilian JV, Klauda RJ, Widman S, Kashiwagi M, Bourquin R, et al. (2012) An assessment of a bait industry and angler behavior as a vector of invasive species. Biological Invasions. Available: http://www.springerlink.com/index/10.1007/s10530-012-0173-5. Accessed: 24 Jul 2012.

43. Winfield IJ, Fletcher JM, James JB (2011) Invasive fish species in the largest lakes of Scotland, Northern Ireland, Wales and England: the collective UK experience. Hydrobiologia 660: 93–103.

44. Williams AE, Moss B (2001) Angling and conservation at Sites of Special Scientific Interest in England: economics, attitudes and impacts. Aquatic Conservation: Marine and Freshwater Ecosystems 11: 357–372. doi:10.1002/aqc.466

45. Oidtmann B, Heitz E, Rogers D, Hoffmann RW (2002) Transmission of crayfish plague. Diseases of aquatic organisms 52: 159–167.

46. Gatlin MR, Shoup DE, Long JM (2012) Invasive zebra mussels (Driessena polymorpha) and Asian clams (Corbicula fluminea) survive gut passage of migratory fish species: implications for dispersal. Biological Invasions. Available: http://www.springerlink.com/index/10.1007/s10530-012-0372-0. Accessed: 5 Dec 2012.

47. Peeler EJ, Feist SW (2011) Human intervention in freshwater ecosystems drives disease emergence. Freshwater Biology 56: 705–716. doi:10.1111/j.1365-2427.2011.02572.x

48. Gallardo B, Errea MP, Aldridge DC (2011) Application of bioclimatic models coupled with network analysis for risk assessment of the killer shrimp, Dikerogammarus villosus, in Great Britain. Biological Invasions 14: 1265–1278. doi:10.1007/s10530-011-0154-0

49. Dresser C, Swanson B (2012) Preemptive legislation inhibits the anthropogenic spread of an aquatic invasive species, the rusty crayfish (Orconectes rusticus). Biological Invasions 15: 1049–1056. doi:10.1007/s10530-012-0349-z

50. Madgwick G, Aldridge DC (2011) Killer Shrimps in Britain: hype or horror? British Wildlife. Available: http://www.arrt.org.uk/media/BWKillerShrimp.pdf. Accessed: 1 Feb 2013.

51. Gozlan RE, Burnard D, Andreou D, Britton JR (2013) Understanding the Threats Posed by Non-Native Species: Public vs. Conservation Managers. PLoS ONE 8: e53200. doi:10.1371/journal.pone.0053200

52. Baruch-Mordo S, Breck SW, Wilson KR, Broderick J (2011) The Carrot or the Stick? Evaluation of Education and Enforcement as Management Tools for Human-Wildlife Conflicts. PLoS ONE 6: e15681. doi:10.1371/journal.pone.0015681

53. Olstad K, Cable J, Robertson G, Bakke T (2006) Unpredicted transmission strategy of Gyrodactylus salaris (Monogenea: Gyrodactylidae): survival and infectivity of parasites on dead hosts. Parasitology 133: 33–41.

54. Shimizu T, Yoshida N, Kasai H, Yoshimizu M (2006) Survival of koi herpesvirus (KHV) in environmental water. ÕÚ̃ÓÕ̃Õ̃Ñ = Fish Pathology 41: 153–157.

55. Nakano H, Hiraoka M, Sameshima M, Kimura T, Momoyama K (1998) Inactivation of penaeid rod-shaped DNA virus (PRDV), the causative agent of penaeid acute viraemia (PAV), by chemical and physical treatments. Fish Pathology 33: 65–71.

56. Oidtmann B (2000) Disease in freshwater crayfish Environment Agency. 9–18.

57. Johnson ML, Speare R (2003) Survival of *Batrachochytrium dendrobatidis* in water: quarantine and disease control implications. Emerging Infectious Diseases 9: 922.

58. Nazir J, Spengler M, Marschang R (2012) Environmental persistence of amphibian and reptilian ranaviruses. Diseases of Aquatic Organisms 98: 177–184. doi:10.3354/dao02443

59. Ahne W (1976) Untersuchungen über die Stabilität des karpfenpathogenen Virusstammes. Fisch und Umwelt 2: 121–127.

60. Ahne W (1982) Vergleichende Untersuchung über die Stabilität von vier fischpathogenen Viren (VHSV, PFR, SVCV, IPNV). Zentralblattfür Veterinarmedizin 29: 457–476.

61. Ricciardi A, Serrouya R, Whoriskey FG (1995) Aerial exposure tolerance off zebra and quagga mussels (Bivalvia: Dreissenidae): implications for overland dispersal. Canadian Journal of Fisheries and Aquatic Sciences 52: 470–477.

62. GB Non Native Species Secretariat (2011) GB non-native organism risk assessment scheme: Pacifastacus leniusculus - Signal crayfish.

63. Martens A, Grabow K (2008) Risk of spreading of nonindigenous Amphipoda due to overland transport of recreation boats. Lauterbornia 62: 41–44.