POPULATION SIZE, STRUCTURE AND DISTRIBUTION OF AN UNEXPLOITED FRESHWATER PEARL MUSSEL *MARGARITIFERA MARGARITIFERA* (L.) POPULATION IN SCOTLAND

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Abstract *All published studies into Scottish populations of the globally threatened and endangered freshwater pearl mussel* Margaritifera margaritifera *have taken place on exploited (pearl fished) populations. In 2013, detailed studies took place on a large and unexploited freshwater pearl mussel population present in a small, remote Scottish river. This paper outlines the results of this study and compares the size, structure and distribution of this unexploited population with those from exploited Scottish populations.*

The unexploited population was estimated to contain approximately 0.6 million freshwater pearl mussels, holding the highest densities of mussels per km of river recorded in Scotland. Assessed against targets for assessing conservation sites, the population would be considered to be in favourable condition, holding high densities of freshwater pearl mussels (up to a mean of 84 mussels per m^2 over a 50 m×1 m transect area; highest density of 216 mussels in 1 m^2), a high proportion of juvenile mussels (23% of samples measured) and many juvenile mussels below 30 mm in size. Current population estimates for most exploited Scottish freshwater pearl mussel populations are far lower than former unexploited population estimates suggest.

This unexploited and undescribed population is considered to be the most important freshwater pearl mussel population in Scotland and the UK. Much conservation action is taking place on this species in the UK and this population is a suitable benchmark and reference site for comparisons on what a restored 'healthy' freshwater pearl mussel site should resemble. The undescribed population is threatened by a range of factors and these are discussed. The River X freshwater pearl mussel population is of global importance.

Key words Margaritifera, *pearl fishing, unexploited population, natural reference site*

INTRODUCTION

The natural range of the freshwater pearl mussel *Margaritifera margaritifera* stretches from northern Scandinavia, southwest through central Europe, the western European seaboard including the United Kingdom (UK), Ireland, France, Spain and Portugal (Young, Cosgrove & Hastie, 2001a). However, the freshwater pearl mussel is seriously threatened throughout its Holarctic range and is classified by the International Union for Conservation of Nature as Endangered (Mollusc Specialist Group, IUCN, 1996) in its world range. In Central Europe the population has declined by 95% (Degerman, Alexanderson, Bergengren, Henrikson, Johansson, Larsen & Söderberg, 2009) and it is classified as Critically Endangered in Europe (Moorkens, 2011). The largest remaining populations in Scotland, Ireland, Norway, Finland and Sweden are of international importance.

The species has a complex and fascinating lifecycle, which makes it vulnerable to a number of threats. Populations of freshwater pearl mussels have been studied across the UK, particularly in Scotland, where most of the extant populations occur. Approximately two thirds of Scottish rivers occupied by freshwater pearl mussels 100 years ago no longer do so or are close to losing their pearl mussels. In Scotland, the effects of pearl-fishing in particular have been implicated in this decline (although other factors are also involved). During a national survey of 160 historical freshwater pearl mussel rivers, destructive pearl-fishing was identified as a significant factor in the decline or disappearance of this species from 153 of 155 (99%) rivers (Cosgrove *et al*., 2000a).

As a consequence of its decline, detailed studies on most of the largest and important Scottish populations have been undertaken. However, all of those studies have taken place on exploited populations. The sizes, densities and age structures of twelve exploited *M. margaritifera* populations were investigated by Hastie *et al.* (2000a). Hastie (2006) subsequently investigated mortality patterns in exploited Scottish freshwater pearl mussel populations, with a special emphasis on the impact and effect of pearl fishing. Pearl

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fishing has taken place in the UK since pre-Roman times (Kunz & Stevenson, 1908), and as such practically all extant, accessible populations have a long history of destructive exploitation.

Despite being fully legally protected since 1998, large numbers of freshwater pearl mussels are killed every year in Scotland. For example, at least ten incidents of criminal damage, or suspected criminal damage, have been recorded by the Scottish police service annually since 2008. However, the police's National Wildlife Crime Unit believe that the annual number of such suspected crimes could be as high as thirty (Cosgrove, Hastie & Sime, 2012a).

In 2013, a full and detailed survey was conducted on one of only two known Scottish *M. margaritifera* populations considered likely to be unexploited. Because of the on-going illegal pearl-fishing threat, the watercourse name and location is treated as confidential and referred to as 'River X' in this paper. The River X is not designated i.e. it is not a Natura 2000 site, Site of Special Scientific Interest, or otherwise protected for conservation purposes.

There are a number of historical and logistical factors that led to River X remaining effectively unexploited. The river is remote and consequently has never been easy to access. According to a former pearl-fisher, in the past the managers of River X had a reputation of intolerance towards pearl-fishing and so very few pearl-fishers risked visiting the river (*pers. comm.*). Whilst the veracity of this claim cannot be independently corroborated, it may have helped create the impression of a no-go pearl-fishing area, especially when other easier to visit sites were available. Furthermore, the ex-pearl-fisher did not consider that the river had a good reputation for producing pearls. He fished the river twice (in 1976 and 1989) and reported that the river was 'untouched' from a pearl-fishing perspective (Cosgrove, 1997). It should be noted that the pearl-fisher only fished for crooked shells, reputed to be best for producing pearls, and carefully used non-destructive tongs, so that pearl mussels were returned alive to the river.

Given that much conservation effort is now being directed towards protecting and restoring *M. margaritifera* populations, it is important that a suitable benchmark or natural reference site for comparisons on what a restored 'healthy' freshwater pearl mussel population should look like is available. The objective of this study is to describe the population size, structure and distribution of a reference freshwater pearl mussel population and compare it with other Scottish populations, with an emphasis on exploitation by pearl fishing.

Freshwater pearl mussel habitat requirements

Freshwater pearl mussels are found in fast flowing unpolluted rivers, with detailed studies on Scottish freshwater pearl mussel populations suggesting that optimum water depths of 0.3–0.4 m and optimum current velocities of 0.25–0.75ms^{-1} at intermediate water levels are most suitable (Hastie *et al.* 2000b). River bed substratum characteristics appear to be the best physical parameters for describing freshwater pearl mussel habitat in Scotland. Freshwater pearl mussels prefer stable cobble/boulder dominated substrate with some fine substrate that allows the mussels to burrow (Cosgrove, Hastie & Young, 2000b). Adult and juvenile mussels tend to have similar habitat 'preferences', although adults are found over a wider range of physical conditions and juveniles appear to be more exacting in their requirements and sensitivity to environmental disturbance (Hastie *et al.* 2000b). Juvenile mussels require fine stable sediments, particularly clean sand and gravel. Of specific importance to freshwater pearl mussel survival are levels of silt, suspended solids, biochemical oxygen demand, calcium and chemical compounds generally associated with enrichment (eutrophication) i.e. nitrate and phosphate (Bauer, 1983).

Freshwater pearl mussel host requirements

Freshwater pearl mussels have a short parasitic larval phase on the gills of suitable native salmonid host fish. The larvae (glochidia) of *M. margaritifera* are very host-specific and can only complete their development on Atlantic salmon *Salmo salar* or brown/sea trout *Salmo trutta*. Usually juvenile fish (fry and parr) are utilised (Young & Williams, 1984). The presence of freshwater pearl mussels in any river therefore depends on salmonid host fish availability and there is some adaptive matching between local races of fish and mussels (Geist, Porkka & Kuehn, 2006). This complexity illustrates the many potential threats and pressures that operate on *Margaritifera* and its habitat.

METHODS

Survey methodology The following study was carried out by three teams of two experienced, specially trained and licensed freshwater pearl mussel surveyors in June 2013. The survey was directed towards establishing the relative abundance of freshwater pearl mussels and habitat suitability throughout all reaches of the main stem of River X. The river was entered and searched, where water depths allowed, using an adapted version of the standard Scottish shallow-water freshwater pearl mussel survey methodology (as described in Cosgrove *et al.* 2000a; Young, Cosgrove, Hastie & Henninger, 2001b; Young, Hastie & Cooksley, 2003) and recommended by Scottish Natural Heritage (SNH).

The main stem of the River X was subdivided into three survey reaches based on catchment characteristics and logistical considerations for the three survey teams:

- Lower reach, from the sea pool (downstream freshwater limit) upstream to the confluence of the first of two tributary watercourses (T1-T29), a distance of ca. 1,450 m.
- Middle reach, from the first tributary watercourse – to the second tributary watercourse (T30-T66), a distance of ca. 1,850 m.
- Upper reach, from the second tributary watercourse to a large freshwater loch (lake) (T67-T122), a distance of ca. 2,800 m.

Each fieldwork team concentrated survey effort in one of the three reaches. The survey reaches were sub-divided and numbered into consecutive 50 m transect sections, e.g. Transect 1 (T1) started at the further downstream freshwater reach and continued upstream for 50 m and then stopped (being recorded as 0–50 m in the results), T2 began at the end T1 and was also 50 m in length (being recorded as 50–100 m in the results) etc. This approach was carried out to the top of the main stem of the river at a large freshwater loch, with the last 50 m section being T122 (6,050–6,100 m upstream from the lowest freshwater reach at T1).

At the beginning of each 50 m transect section the river was entered by the two surveyors and a search conducted. Each surveyor recorded all visible mussels in a 50 m×1 m corridor. Transect surveys were conducted in parallel to avoid duplication. In practice this usually meant one transect count was conducted between 1 m out from the left bank to the middle of the river and the other between 1 m out from the right bank to the middle of the river. Occasionally where the water was too deep on one side of the river to survey, a single transect count was conducted. For each 50 m survey reach the two 50 m×1 m transects were undertaken and the number of mussels counted were added together, divided by two and multiplied by the mean permanently wetted river width for that 50 m survey section. It is recognised that mussel densities can and do vary within a river, but this sampling approach meant that close to a quarter of all substrate habitats in the river were surveyed. The mean width of the River X was 8.6 m (of which 2 m was always surveyed unless a 50 m transect was too deep to undertake). The substrate present in each 50 m transect section was recorded and classified using the standard Wentworth Scale (Wentworth, 1922).

The only practicable way of determining viability of a freshwater pearl mussel population is by estimating the relative abundance of juvenile mussels in overall population size profiles (Cosgrove *et al*. 2000a). This is carried out by conducting quadrat searches throughout reaches where mussels are present. It was not possible given the time and resources available to undertake quadrat searches in every 50 m survey reach. Instead, twenty five 1 m×1 m quadrats in total were laid out on the river bed spread over the lower, middle and upper survey reaches. All visible mussels in these quadrats were counted and then temporarily removed. Loose substrate was moved and displaced to reveal any hidden freshwater pearl mussels and, in particular, to search for any juvenile *M. margaritifera*. The substrate within each quadrat was carefully searched by sifting through all mobile/loose substrates for a minimum of 5 minutes. The depth these searches reached varied according to substrate composition, but in a typical quadrat searches tended to reach a depth of 5–10 cm below the surface. All mussels removed (both visible and hidden) were measured using dial callipers and then returned alive to their approximate original position in the river.

Mussel abundance categories Standard criteria were used for describing the abundance and status of the pearl mussel population, based on counts of visible mussels (Cosgrove *et al.* 2000a).

Table 1 Standard relative abundance terms and codes for 50 m×1 m transect counts

Visible mussels per 50 m×1 m transect	Visible mussel density per 1 m^2	Terminology	Abundance code
0	0	Absent	E
1–49	0.02–0.98	Rare	D
50–499	1–9.98	Scarce	C
500–999	10–19.98	Common	B
1000+	>20	Abundant	A

Any description of the conservation status of a mussel population must refer to the current ability of that population to recruit juveniles (Chesney & Oliver, 1998). The relative abundance and status terms used in this paper match those used in previous survey work (Table 1) are therefore based on the recommended terminology and, importantly, are directly comparable to those used for monitoring on Scottish pearl mussel designated sites which are used to report the status of designated sites to the European Commission (JNCC, 2005).

RESULTS

Population estimate Large numbers of freshwater pearl mussels were found in all three survey reaches and in all substrate habitats (Fig. 1). A summary of 50 m×1 m transect relative abundances is provided in Table 2 (which uses unadjusted visible count data). There were no 50 m×1 m transects where mussels were absent, with some stable and suitable substrate habitats present in all transects. The majority of 50 m×1 m transects (125) were assessed as having relative abundance 'Scarce' (C). Thirty six 50 m×1 m transects were assessed as either 'Common' (B) or 'Abundant' (A), with one 50 m×1 m transect (T82) holding 4,200 mussels (a mean visible relative abundance of 84 mussels per m^2 throughout the 50 m×1 m transect). A few 50 m×1 m transects (8%) were too deep to survey using standard shallow water survey methods.

For each 50 m survey reach two 50 m×1 m transects were undertaken. For example, in the lowest 50 m survey reach, the two 50 m×1 m transect counts recorded 50 and 20 visible mussels respectively. These metrics were added together and divided by two to provide a mean figure of 35 mussels per 50 m×1 m within this 50 m river section. This metric was then multiplied by the permanently wetted river width in this 50 m river section (not the mean width over the entire river length), which was 15 m in this part of the river, to give an estimated total of 525 mussels for this 50 m reach. This process was repeated throughout all 50 m river sections in the lower, middle and upper reaches.

Based on transect count data, an estimated total of 299,182 visible mussels were present in the lower (61,756), middle (75,066) and upper (162,360) survey reaches of the River X. A small number of pearl mussels (20 individuals) were present in the lowest 30 m of one of the two tributaries.

A total of 1,159 mussels were recorded and measured during twenty five 1 m×1 m quadrat searches (highest total density of 216 mussels in 1 m^2), comprising 627 visible and 532 hidden mussels. This gives a ratio of 627:532 visible to hidden mussels from the quadrat samples surveyed.

Of the 1,159 mussels measured, 262 were juveniles (23%). Hastie & Cosgrove (2002) demonstrated that searching for juvenile mussels in river bed sediments is inefficient, and that sampling bias towards larger, easier to see adults is considerable (with most detected). They calculated an average search efficiency of 50% for juvenile mussels, i.e. approximately half remained undetected during quadrat searches by experienced surveyors. Using Hastie & Cosgrove's (2002) work, it is possible to adjust the River X's population estimate to correct for this known sampling bias. As there were 262 juveniles recorded during quadrat searches, a 50% efficiency means that an additional 131 juvenile mussels were undetected during quadrat searches. As undetected these must be added to the proportion of hidden mussels, giving a new ratio of 627 visible: 663 hidden. Simple extrapolation of this ratio to the number of visible mussels from transect counts

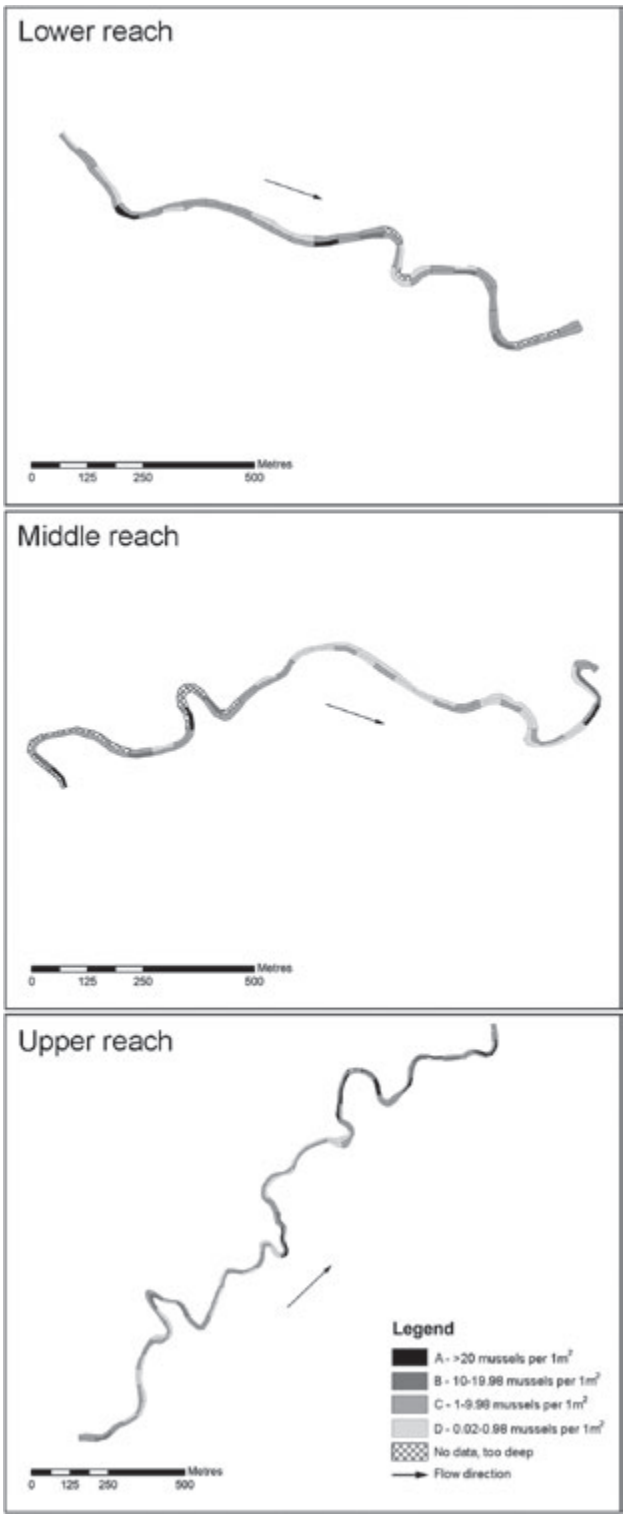

Figure 1 The relative abundance of freshwater pearl mussels in the River X, 2013

throughout the entire river (299,182 mussels) provides an additional 316,360 mussels.

Therefore, the total estimated population in River X is visible 299,182+hidden 316,360=615,542 freshwater pearl mussels.

Table 2 Relative abundance data for 50 m×1 m transect counts undertaken in River X

Visible mussels per 50 m×1 m transect	Abundance code	Number of 50 m×1 m transects
0	E	0
1–49	D	64
50–499	C	125
500–999	B	20
1000+	A	16
No data	Too deep	19

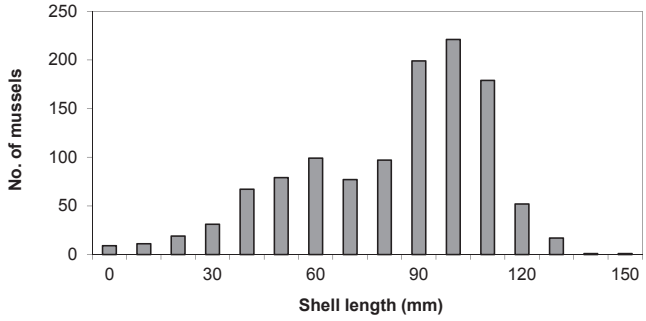

Figure 2 River X freshwater pearl mussel unadjusted shell length frequencies, 2013

Population profile Fig. 2 illustrates the unadjusted shell length profile of the freshwater pearl mussels recorded during twenty five quadrats in the lower, middle and upper reaches of River X.

To account for known sampling bias in the standard quadrat survey methodology, the number of mussels in each shell length category was 'corrected' using the mean size sampling efficiencies from Fig. 1 of Hastie & Cosgrove (2002), which plots sampling efficiency against mussel size as size class mid points using a fitted regression line and equation coefficient. For example, the sampling efficiency of the smallest juvenile mussels (age class 1–10 years) was 50% and for larger adult mussels (age class 61–100 years) was 99–100%. The adjusted shell length frequencies are presented for the River X in Fig. 3.

The unadjusted and adjusted population estimates and mean densities of pearl mussels per km of river for the three survey reaches are provided in Table 3.

Comparisons with other Scottish M. margaritifera populations The size, density and structure of twelve exploited Scottish *M. margaritifera* populations have previously been estimated by

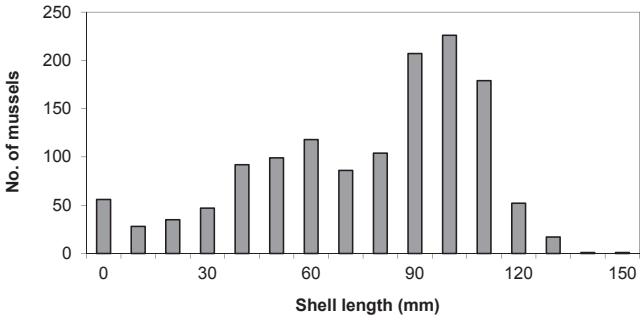

Figure 3 River X freshwater pearl mussel adjusted shell length frequencies, 2013

extrapolating from 50 m×1 m sample surveys (Hastie *et al.* 2000a) using similar methods to those presented for River X, but most estimates in Table 4 were based on smaller sample sizes (fewer transects). Total population estimates ranged from 2,000 to 10 million freshwater pearl mussels (Table 4). There were marked differences between the population size and profiles observed. Hastie *et al.* code-lettered their survey rivers A–L because of the threat posed by illegal pearl-fishing.

River X has the highest density of freshwater pearl mussels per km of any known river in Scotland (Table 4). In the rivers A–L (which are all designated sites), mussels are localised and only found within specific reaches, with large sections often devoid of freshwater pearl mussels. River X held pearl mussels throughout all reaches, with no sections devoid of freshwater pearl mussels (Fig. 1).

Although four rivers (D, F, J and K) are considered to have similar or greater overall total population estimates than River X, these rivers are much longer than the relatively short 6.1 km length of the River X (rounded to 6 km for comparative purposes with rivers A–L lengths in Tables 4 and 5).

Table 3 Population data for the three River X survey reaches

River X survey reach	Unadjusted population estimate	Final adjusted population estimate	Mean density of pearl mussels per km of river
Lower	61,756	127,058	87,626
Middle	75,066	154,442	83,482
Upper	162,360	334,042	119,301
Overall total	299,182	615,542	100,909

Table 4 Comparative size estimates of Scottish *M. margaritifera* populations

River	River length, to nearest km[a]	Transect based river population estimate[b]	Estimated number of mussels per km of river
A	8 (2)	N/A	N/A
B	**4 (2)**	**5,000**	**1,250**
C	**6 (1)**	**14,000**	**2,333**
D	140 (ca. 100)	1,300,000	9,286
E	7 (1.5)	35,000–70,000	5,000–10,000
F	18 (4)	600,000–1,200,000	33,333–66,666
G	14 (4)	187,000–375,000	13,357–26,786
H	21 (1.5)	N/A	N/A
I	6 (4)	<2,000	<333
J	80 (50)	900,000–3,700,000	11,250–46,250
K	170 (ca. 105)	10,000,000[c]	58,824[c]
L	**6 (0.5)**	**3,000**	**500**
X	6 (6)	615,542	100,909

[a] Lengths of rivers containing pearl mussels are in parenthesis.
[b] Estimates in bold based on complete visible mussel counts; otherwise based on extrapolated 50 m×1 m sample transect data.
[c] River K has very recently been resurveyed and the population has substantially declined (by at least 50%). Thus, the current population estimate is now likely to be <5 million mussels and so estimated mean number of mussels per km is likely to be <29,412.

Table 5 Unexploited river populations estimate of Scottish *M. margaritifera* populations

River	River length, to nearest km	Current river population estimate	Current estimate of mussels per km of river	Unexploited river population estimate
A	8 (2)	N/A	N/A	807,272 (201,818)
B	4 (2)	5,000	1,250	N/A[a]
C	6 (1)	14,000	2,333	N/A[a]
D	140 (ca. 100)	1,300,000	9,286	14,127,260 (10,090,900)
E	7 (1.5)	35,000–70,000	5,000–10,000	706,363 (151,364)
F	18 (4)	600,000–1,200,000	33,333–66,666	1,816,362 (403,636)
G	14 (4)	187,000–375,000	13,357–26,786	1,412,726 (403,636)
H	21 (1.5)	N/A[a]	N/A[a]	2,119,089 (151,364)
I	6 (4)	<2,000	<333	605,454 (403,636)
J	80 (50)	900,000–3,700,000	11,250–46,250	8,072,720 (5,045,450)
K	170 (ca. 105)	10,000,000[b]	58,824[b]	17,154,530 (10,595,445)
L	6 (0.5)	3,000	500	N/A[a]
X	6 (6)	615,542	100,909	605,454

[a] Estimate not provided because these watercourses are substantially narrower in width than River X.
[b] River K has very recently been resurveyed and sample surveys suggest the population has substantially declined (by at least 50%). Thus, the current population estimate is now likely to be <5 million mussels and so estimated mean number of mussels per km is likely to be <29,412.

The adjusted shell length profile in River X (Fig. 3) is one of a healthy and viable freshwater pearl mussel population. A large proportion of the River X population sampled (23%) were juvenile pearl mussels. A characteristic of most of the rivers surveyed by Hastie *et al.* (2000a) was the small proportion of juveniles recorded, suggesting a recent low level of recruitment in these populations. The three largest (longest) rivers surveyed (D, J and K) had by far the highest proportion of juvenile mussels in survey samples. Significant positive relationships between river size and levels of juvenile recruitment were observed by Hastie *et al.* (2000a) in these exploited populations.

Most survey work on Scottish populations, including rivers A–L, has taken place as part of routine monitoring and research of Scotland's 21 designated sites for freshwater pearl mussels. Common Standards Monitoring (JNCC, 2005) uses three mandatory biological criteria to assess status of the designated freshwater pearl mussel populations:

- there should be >5 mussels per m^2 in sample transects;
- at least 20% of the population should be juvenile (≤65 mm); and
- at least one mussel should be ≤30 mm (indicating recent successful recruitment).

Only one designated site fulfilled these criteria (River F), one was not assessed due to lack of survey information and the remaining designated sites were all classified as in unfavourable condition due to insufficient numbers of juvenile mussels and, at a number of sites, low population densities caused by a number of adverse pressures (Langan, Cooksley, Young, Stutter, Scougall, Dalziel, Feeney, Lilly & Dunn, 2007). Of the 21 Scottish designated sites, the most recent monitoring reported some evidence of criminal damage (exploitation) from 75% of these (Cosgrove *et al.* 2012a). Although not designated, the River X fulfils all three Common Standards Monitoring assessment criteria and there is no evidence of any criminal damage or exploitation from this river either.

DISCUSSION

The River X pearl mussel population profile is particularly interesting as it has: (i) a good spread of shell sizes – indicative of a healthy, functioning and unexploited population; (ii) a large proportion of juvenile mussels recorded in quadrats (23% measured); and (iii) although a tiny number of very large and old mussels were recorded (largest 155 mm in length), most natural mortality appears to occur once mussels reach

ca. 120 mm in size, with relatively few surviving beyond this size.

River X has a very large population of freshwater pearl mussels in a relatively short watercourse, with the estimated number of mussels per km considerably greater than any other known Scottish population (Table 4). This can be attributed to a number of key factors:

- mixed, stable and suitable substrate habitats present in all reaches;
- an absence of historic and current pearl-fishing mortalities;
- benign land-management practices in the catchment (maintaining suitable water quality and quantity through low intensity forestry, grazing and recreational management);
- healthy native host salmonid fish populations; and
- a lack of detrimental river engineering activities (the river uses its natural flood plain unconstrained).

The River X has exceptionally good pearl mussel habitat, with an absence of river engineering (one short 70 m section of bank stabilisation was recorded in the lower reach), excellent catchment management practices and a healthy host salmonid population further justifying it as a reference population.

River X is the only surveyed Scottish watercourse where pearl mussels are present throughout all river reaches. It was evident that all shallow, moderate and deeper areas in the catchment held pearl mussels, even where substrate habitats became unstable and apparently unsuitable (Fig. 4). Whilst some reaches of rivers A–L in Hastie *et al.* (2000a) held unsuitable habitats (and so would not be expected to hold pearl mussels), substantial reaches of suitable substrate habitats occurred, where mussels were absent. In all monitored Scottish populations, visible mussels in shallow water have been heavily exploited and in many areas partially or completely removed.

Rivers A–L have multiple additional pressures e.g. poor water quality/quantity, river engineering and declining host salmonid populations, which have undoubtedly adversely affected these populations (Langan *et al.* 2007).

Estimates of pearl-fishing mortality from other rivers have been made but are not considered accurate, due to a number of potential sources of error and crude assumptions associated with illegal pearl-fishing effort and sampling mussels (Hastie, 2006). Nevertheless, pearl-fishing has significantly impacted many pearl mussel populations; Cosgrove *et al.* (2000a) identified pearl-fishing as the main cause of decline in Scottish populations and found some evidence

Figure 4a Stable and suitable River X habitats held very large numbers of pearl mussels (Photo: D. Shields).

Figure 4b Partly stable River X habitats held large numbers of pearl mussels (Photo: D. Shields).

Figure 4c Unstable and typically unsuitable River X habitats also held moderate numbers of pearl mussels (Photo: D. Shields).

of pearl-fishing mortality in almost every population surveyed (the River X was not surveyed during this previous work).

Some populations have been more heavily exploited than others, with Cosgrove *et al.* (2000a) and Hastie (2006) attributing this to the perception that only certain rivers produce valuable pearls. In this regard some large populations have been relatively lightly exploited, whereas rivers with excellent reputations for producing pearls have been heavily exploited. Hastie (2006) found that whilst pearl-fishers generally targeted larger, older mussels, significant numbers of young mussels (incapable of producing pearls) were also regularly killed, highlighting the opportunistic and destructive nature of pearl-fishing.

If the density of freshwater pearl mussels present per km in River X is taken as representative of a natural reference population, then it can be used as a very crude measure of what might be missing in other, exploited, Scottish *M. margaritifera* populations if they were close to reference condition (Table 5). There are a number of assumptions with taking this approach (from assuming River X is typical of a reference population to assigning an area and length of river habitat likely to be formerly occupied by pearl mussels) and so the gross numbers should be treated with caution. River X is a relatively small (8.6 m mean width) river, comparable in width to rivers A, E, F, G and I; considerably wider than rivers B, C and L; and narrower than rivers D, H, J and K.

Two crude estimates of former population size for Rivers A–L are provided in Table 5. The first assumes that the entire river length held potentially suitable *M. margaritifera* habitats and this is known not to be the situation in most of these rivers. Therefore, this metric would likely be an over-estimate. The second metric (in parenthesis) only assumes that the length of river currently holding pearl mussels would have been capable of holding pearl mussels historically. This is known not to be the situation in most of these rivers, with large areas of suitable habitat currently devoid of pearl mussels due to historic exploitation, river engineering etc. and so the metrics in parenthesis are likely be an underestimate. Therefore, the former reference condition of these sites is likely to lie somewhere between these two estimates, excluding rivers B, C and L which are considerably narrower than River X and therefore estimates using River X data would be inappropriate.

What is apparent from Table 5 is that the current population estimates for most exploited *M. margaritifera* populations are far lower than former reference population estimates suggest. It is clear that most exploited rivers currently have population estimates that are less than a quarter of their likely former population size (the metrics vary depending upon the assumed length of formerly suitable habitat used for estimates). An exception to this is River F, where the current population estimate and predicted former population estimate are broadly similar. River F has been relatively lightly exploited, probably as a consequence of having a poor reputation for producing pearls, and is the only Scottish designated site currently assessed to be in favourable conservation status.

There are lots of anecdotal historical accounts of freshwater pearl mussels being formerly abundant in many Scottish rivers (e.g. Kunz & Stevenson, 1908; Goodwin, 1985; Woodward, 1994) before being pearl fished to extinction or virtual extinction, but none provide pre-exploitation density or population estimates. There are multiple accounts of intensive pearl fishing exploitation of rivers A–L (Cosgrove, 1997). Given this, these populations (with the possible exception of River F) are not likely to be suitable benchmark or natural reference sites because most mussels are probably missing.

M. margaritifera is a long-lived and slow growing species, with very low reproductive rates (Young & Williams, 1984) and it should therefore come as no surprise that historical and current illegal pearl-fishing exploitation has detrimentally affected almost every population in Scotland. It is evident from the results of this study, along with those of other monitored Scottish populations that the River X is unique and arguably the most important *M. margaritifera* population in Scotland and probably the UK. Indeed, in a recent review of Scottish freshwater pearl mussel populations (Hastie, 2011), the lack of any examples of completely undisturbed, unexploited and unstressed populations was highlighted as a major problem for comparative studies and conservation efforts.

Significant conservation efforts to protect and restore freshwater pearl mussel populations are

underway, for example, the current £3.5 million 'Pearls in Peril' LIFE +NATURE project co-funded by many organisations across Scotland, England and Wales aims to safeguard 21 important designated populations of freshwater pearl mussels (note this does not include River X, as the site is undesignated). With all but one of the Scottish designated sites in unfavourable condition, the importance of a suitable benchmark or natural reference site for comparisons on what a restored 'healthy' natural freshwater pearl mussel watercourse should resemble is clear.

The evidence of this site's international importance is overwhelming, River X is the only known UK freshwater pearl mussel population that is unexploited, unstressed and in a natural state. The densities of mussels throughout River X exceed all known sites and include a large proportion of juveniles. Recently, Cosgrove, Hastie, Watt, Sime & Boon (2012b) investigated the threats posed by climate change to Scottish *M. margaritifera* populations. They recognised that some pearl mussel populations may be lost regardless of effort and that scarce conservation resources should be targeted at catchments with native riparian woodland cover, upstream lakes and evidence of recent juvenile pearl mussel recruitment. River X has all three of these key characteristics which strongly suggests that, if managed properly, the pearl mussel population in this river would most likely persist in the long term.

There are reports of a second, potentially unexploited freshwater pearl mussel population from a remote area of Scotland. However, this population is apparently in deep water and has not been surveyed to date.

Threats to the River X pearl mussel population
There are four main potential threats to the River X freshwater pearl mussel population:

Pollution
There is an operational small hydro-electric scheme on one of the tributaries. In June 2013 there was an accidental hydraulic oil spill emanating from the hydro-electric scheme infrastructure posing a potential threat to the water quality of the lower reach of the River X, where ca. 20% of the pearl mussel population resides. There was no evidence encountered of direct mortality or damage to habitats caused by the pollution incident at the time of survey.

The response to the 'minor' pollution incident by the public bodies responsible also highlighted the risk from the lack of designation and associated information available. The response did not match the threat since the information available to the public bodies did not suggest any particularly special ecological interest in the River X catchment.

Pearl-fishing
Despite full legal protection, illegal pearl fishing still takes place annually in Scotland and so the River X is threatened by pearl fishing, although there was no evidence of any recent pearl fishing activity during the 2013 survey.

Decline in wild host fish populations
According to unpublished estate fish monitoring data, the host fish populations in the River X collapsed during the 1990s. The specific reasons why this occurred have not been established, but this happened across all monitored Scottish west coast pearl mussel river populations (Hastie & Cosgrove, 2001). It appears to have been halted and reversed in the River X by the stocking of sea trout *Salmo trutta*. However, during monitoring of fish in River X glochidia were only found on the gills of juvenile Atlantic salmon. Therefore, although the timing sea trout stocking appears to correlate with a recovery in juvenile pearl mussel recruitment, it may not be causative as there was no evidence of glochidia on the gills of sea trout.

An important and potentially manageable element is the mortality to wild host fish when they are at sea caused by two sea lice species *Lepeoptheirus salmonis* and *Caligus elongatus*, high densities of which are associated with marine fin-fish cages (e.g. Butler, 2002; Heauch, Bjørn, Finstad, Holst, Asplin, & Nilsen, 2005; Costello, 2006). Crucial to the long-term health of the only known unexploited and natural state UK freshwater pearl mussel population is the need to tackle the main threats posed to wild host fish on Scottish west coast rivers, including managing fin-fish cages in adjacent marine areas with wild host salmonid stock conservation in mind. This task is not assisted by the undesignated status of the River X's pearl mussel population and the fact that the site's importance and location is known by only a handful of people. The planning process with regard to protecting the river's pearl mussel population is fraught with

difficulties because of confidentiality and the fact that the site is undesignated and so is judged to be of low importance.

Insufficient protection measures

Whilst the freshwater pearl mussel is now legally protected under the Wildlife and Countryside Act 1981 (as amended) and is listed on Annexes II and V of the EC Habitats Directive and Appendix III of the Bern Convention, the River X pearl mussel population is threatened by insufficient site protection measures. Any activity that might disturb this species requires licencing from SNH. However, catchment land-use changes and the decline or destruction of wild host fish stocks could potentially take place, with little or no attention paid to effects on freshwater pearl mussels.

The absence of a site designation and the sensitive nature of the information about the mussel occurrence means that the knowledge required to ensure the mussels' continued survival rests with a few key individuals. While those individuals remain in position, this knowledge is available but once this changes, there is no guarantee that the necessary information will be passed on. Furthermore, even with key individuals in position, it is not always possible to gain suitable responses to threats, as evidenced by the response to the 2013 pollution incident. The most effective way to mitigate this risk would be to designate the site.

Site designation does, however, carry certain risks by making the location public knowledge. In this case though, it is considered that the threat of illegal pearl-fishing is outweighed by the threat of a decline in the host fish population or a detrimental change in catchment management. A series of benefits could accrue to the site with designation, largely from public engagement and targeted funding sources, such as agri-environment payments or initiatives such as the 'Pearls in Peril' Life +NATURE project. The lack of a designation excludes the full package of measures that otherwise might be available to underpin the protection and security of this river's mussel population.

There is a real risk of losing this, the most important freshwater pearl mussel population in the UK, due to threats posed by factors which are not addressed by the legal protection of the species. The lack of site designation is of concern and is a process that SNH should consider as a matter of urgency. It is important that this undescribed site, which is a suitable benchmark and reference site for comparisons on what a restored 'healthy' freshwater pearl mussel population should resemble, is properly protected as soon as possible. The River X freshwater pearl mussel population is of global importance.

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