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LONG TIME SERIES DATA SHOWING RECENT RECOVERY OF GASTROPOD POPULATIONS FROM EFFECTS OF TRI-BUTYL TIN AT THE SHETLAND OIL TERMINAL

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Abstract

Surveys of gastropod (dogwhelk, *Nucella lapillus*) populations at the Shetland Oil Terminal in Sullom Voe for the effects of tri-butyl tin (TBT) from shipping have been commissioned by the terminal's Environmental Advisory Group (SOTEAG) and now represent one of the longest time series of biological effects data available (1987 – 2009). These data from 20 shoreline sites around the oil terminal in the voe and less affected waters of Yell Sound, show a substantial recovery of TBT-induced imposex as measured by vas deferens sequence index (VDSI), relative penis size index (RPSI) and female sterility associated with decreasing shipping traffic, reduced usage of free association antifoulants and now the complete ban imposed by the IMO on TBT usage on large vessels in 2008. Data are presented that show in 2009, for the first time and following the IMO ban, no incidence of imposex-induced female sterility at any of the surveyed sites. At some of the most impacted sites, some metrics of imposex are now at or close to zero (sterility, RPSI) and the rate of recovery of VDSI is greater than previously recorded. Data are assessed using OSPAR methodologies and assessment criteria and presented alongside data showing associated changes in dogwhelk population structure in the area.

Keywords: gastropod, imposex, TBT, dogwhelk, biological effect

Introduction

Tributyltin (TBT) compounds have been recognised as some of the most toxic substances released into the aquatic environment. Extensive reviews have been published outlining the toxicity of TBT to aquatic organisms (e.g. Hall and Pinkney, 1985; Laughlin and Linden, 1987; Muller *et al.*, 1989; IMO, 1989; IPCS, 1990).

Sullom Voe is a large fjordic inlet on the mainland of Shetland (Fig. 1). The mouth of the Voe is approximately 5 km wide, and the Voe extends approximately 13 km southwards (Dooley, 1981). A large oil terminal situated on the promontory of Calback Ness was opened in November 1978. There was a peak in the tonnage and number of crude and gas tankers visiting the terminal in 1984 and the tonnage and numbers have since fallen (Fig. 2). TBT contamination arises from visiting tankers (Bailey and Davies, 1988) and, up until 1986, from TBT antifoulants used on towing vessels, navigational buoys and harbour craft (Shetland Islands Ports Authority and

Shetland Islands Council, pers comm, 1991). Reducing shipping traffic, a historical change from free association to co-polymer TBT based antifoulants and recent IMO restrictions on the use of TBT on large vessels (no new applications in 2003 and no exposed TBT paints on vessels in 2008) means that TBT inputs to the area should be reducing.

Extensive laboratory and field investigations have been undertaken demonstrating the occurrence of male sexual characteristics in female dogwhelks (a condition termed imposex; Blaber, 1970) resulting from exposure to TBT. The sensitivity and usefulness of using the dogwhelk as an indicator of TBT contamination is well established (e.g. Gibbs *et al.*, 1987; Bailey and Davies, 1989) and has been included in international monitoring programmes under the Oslo and Paris Commission (Davies *et al.*, 1997; Gubbins *et al*., 2004).

The following paper details results of surveys that used the common dogwhelk (*Nucella lapillus* L.) as an indicator of TBT contamination arising from oil terminal operations in Sullom Voe from 1987 - 2009. The aim of these surveys has been to generate time series data on the effects of TBT contamination in and around Sullom Voe and, in recent years, assess if the dogwhelk populations have shown any further recovery since the ban on TBT on large vessels in 2008.

Figure 1. Sullom Voe and yell Sound showing the position of the Oil Terminal and sampling sites. Sites labelled with a "J" are where juvenile dogwhelks have been sampled to derive information on population reproductive state and recent impacts from TBT.

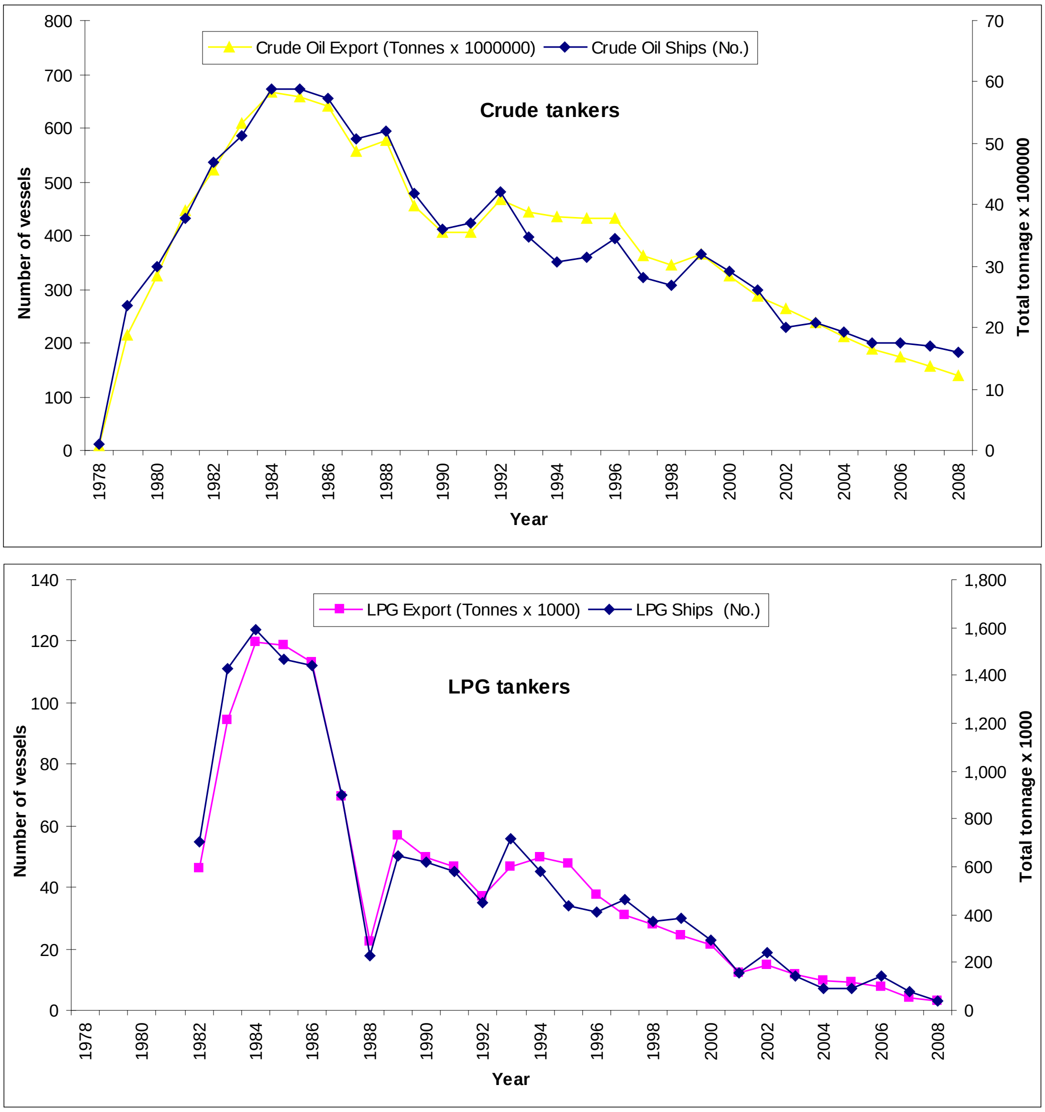

Figure 2. Number and total tonnage of tankers visiting Sullom Voe, 1978 – 2008.

Methods

In summers of 1987, 1990, 1991, 1993, 1995, 1997, 1999, 2001, 2004, 2007 and 2009, samples of approximately 40 adult dogwhelks (identified by thickened shell rim and the presence of "teeth"; Crothers, 1985) were collected from 20-21 sites around Sullom Voe and the waters of Yell Sound (Fig. 1, Table 2).

The shell length of each animal was measured, and individuals (at sites 1, 3, 5, 9, 12) were classified by their shell length according to observations by Moore (1936), i.e. juveniles (10-15 mm shell length), sub-adults (15-21 mm), and un-toothed adults (21-26 mm and 26-35 mm). At each of the juvenile and sub-adult survey sites, an attempt was made to obtain 20 individuals from each of the above size classes (and 40 toothed adults) The degree of imposex, as measured by Relative Penis Size Index (RPSI) and Vas Deferens Sequence Index (VDSI), was determined using international standard techniques (OSPAR, 2002).

The development of imposex in dogwhelks may be divided into seven stages, depending upon the developmental state of both the penis and vas deferens in the female (Gibbs *et al.*, 1987). Stage 0 is identified where no signs of imposex can be seen. Stage 1 can be identified when the vas deferens begins at the site of the vulva with Stage 2 also showing a small penis behind the right eye tentacle. As imposex progresses, the vas deferens starts to develop from the penis (Stage 3) and will become continuous (Stage 4). Eventually, vas deferens tissue may proliferate over the opening of the vulva (Stage 5), rendering the female incapable of breeding since she can no longer release egg capsules. The trapped egg capsules form a solid mass within the capsule gland. In this final Stage (Stage 6), the capsule gland may eventually rupture, causing premature death of the female. Each of the seven Stages of imposex is known as a Vas Deferens Sequence (VDS) stage and calculation of the mean VDS for a group of females provides the Vas Deferens Sequence Index (VDSI) which may be used to compare the reproductive competency of different populations.

The VDS was determined for each female and the mean VDS calculated to provide an estimate of the VDSI of the population.

The Relative Penis Size Index (or RPSI, Gibbs *et al.*, 1987) was calculated from penis length measurements of the dogwhelks as follows:

$$\frac{(\text{mean female penis length})^3}{(\text{mean male penis length})^3} \text{X} 100 = \%\text{RPSI}$$

The greater the penis growth in females, the higher the RPSI value. An RPSI of 12.5 %, for example, indicates that the mean female penis length is half that of the male.

The proportion of sterile females observed in a population was calculated as the number of stage 5 and 6 individuals divided by the total number of females in the sample (x100).

During all surveys estimates of dowhelk abundance were recorded during collection by measuring collection rates and expressing abundance as individuals collected per minute. This provides a fairly crude but effective measure of relative population abundance.

The level of imposex is assessed against Oslo and Paris Commission (OSPAR) assessment criteria to provide meaningful picture of the chemical and biological significance of the imposex levels observed. The criteria for each assessment level and the significance is given in Table 1 below (OSPAR, 2004).

Table 1. Oslo and Paris Commission biological effects assessment criteria for imposex in *Nucella lapillus*, based on VDSI (OSPAR, 2004).

Assessment class	***Nucella* VDSI**	**Effects and impacts**
A	VDSI = <0.3	The level of imposex in the more sensitive gastropod species is close to zero (0 - ~30% of females have imposex) indicating exposure to TBT concentrations close to zero, which is the objective in the OSPAR strategy of hazardous substances.
B	VDSI = 0.3 - <2.0	The level of imposex in the more sensitive gastropod species (~30 - ~100 % of the females have imposex) indicates exposure to TBT concentrations below the EAC derived for TBT. E.g. adverse effects in the more sensitive taxa of the ecosystem caused by long-term exposure to TBT are predicted to be unlikely to occur.
C	VDSI = 2.0 - <4.0	The level of imposex in the more sensitive gastropod species indicates exposure to TBT concentrations higher than the EAC derived for TBT. E.g. there is a risk of adverse effects, such as reduced growth and recruitment, in the more sensitive taxa of the ecosystem caused by long-term exposure to TBT.
D	VDSI = 4.0 - 5.0	The reproductive capacity in the populations of the more sensitive gastropod species, such as *Nucella lapillus*, is affected as a result of the presence of sterile females, but some reproductively capable females remain. E.g. there is evidence of adverse effects, which can be directly associated with the exposure to TBT.
E	VDSI = > 5.0	Populations of the more sensitive gastropod species, such as *Nucella lapillus*, are unable to reproduce. The majority, if not all females within the population have been sterilized.
F	VDSI = -	The populations of the more sensitive gastropod species, such as *Nucella lapillus* and *Ocinebrina aciculata*, are absent/expired.

Results and Discussion

TBT-induced imposex in gastropod populations is most evident at sites within Sullom Voe, close to the location of the Oil Terminal (Table 2, Figure 3). Historically, populations at these locations have been heavily affected, with significant incidence of female sterility at some sites, and absence of juvenile dogwhelks from some sites as a result. Further away from the Terminal, in Yell Sound, the impacts from TBT have always been significantly less with minimal levels of RPSI and VDSI recorded and little or no incidence of female sterility (Gubbins 2009, Gubbins et al., 2005, 2008).

Table 2 Identification of sampling sites and current (2009) imposex status (incidence of imposex occurrence in females, RPSI, VDSI, and % sterile females) of populations at these locations.

Site No	Site Name	Size	Incidence of Occurrence %	% RPSI	VDSI	% sterile females
1	Easterwick	10-15	17	0.00	0.167	
		15-21	27	0.00	0.273	
		21-26	22	0.00	0.222	
		26-35	11	0.00	0.111	
		Adults	45	0.00	0.455	0
2	Burgo Taing	Adults	31	<0.01	0.577	0
3	Billia Skerry	10-15	43	22.38	1.571	
		15-21	78	0.11	1.667	
		21-26	83	0.13	1.667	
		26-35	71	0.02	1.643	
		Adults	65	0.13	1.500	0
4	Scarf Stane	Adults	70	<0.01	0.950	0
5	East of Ollaberry	10-15	18	0.00	0.182	
		15-21	27	0.00	0.273	
		21-26	13	0.00	0.125	
		26-35	63	<0.01	0.175	
		Adults	33	<0.01	0.381	0
6	Grunn Taing	Adults	59	0.00	0.588	0
7	Tivaka Taing	Adults	100	0.54	2.636	0
8	Noust of Burraland	Adults	100	0.41	3.526	0
9	Mavis Grind	10-15	83	0.04	1.167	
		15-21	100	10.63	2.358	
		21-26	91	0.17	2.273	
		26-35	100	1.08	3.000	
		Adults	94	1.08	3.667	0
10	Voxter Ness	Adults	83	0.01	1.300	0
11	Northward	Adults	95	0.31	3.143	0
12	Kames	10-15	90	1.06	2.300	
		15-21	83	<0.01	1.000	
		21-26	75	1.42	1.273	
		26-35	85	0.06	1.308	
		Adults	93	0.26	3.000	0
13	Skaw Taing	Adults	100	0.26	2.765	0
14	Grunna Taing	Adults	100	0.67	3.235	0
15	Orfassary	Adults	27	<0.01	0.400	0
16	Samphrey/The Helliach	Adults	28	<0.01	0.333	0
17	Uynarey	Adults	29	<0.01	0.381	0
18	Little Roe	Adults	79	0.03	1.947	0
19	The Brough	Adults	41	0.01	0.375	0
20	Norther Geo	Adults	20	0.00	0.200	0

The current (2009) status is shown in Figure 3, with sites assessed according to their OSPAR classification (A-F). The majority of sites within Sullom Voe close to the terminal fall into class C indicating that individuals are exposed to TBT concentrations above the EAC, but are not experiencing female sterility as a result of imposex. Populations further from the Terminal, in the waters of Yell Sound show either class A or B status indicating either that imposex status is as close to background as can be determined by the sampling methodology or that exposure is occurring but not to a level that would cause adverse effects (i.e. TBT concentrations are not above the EAC).

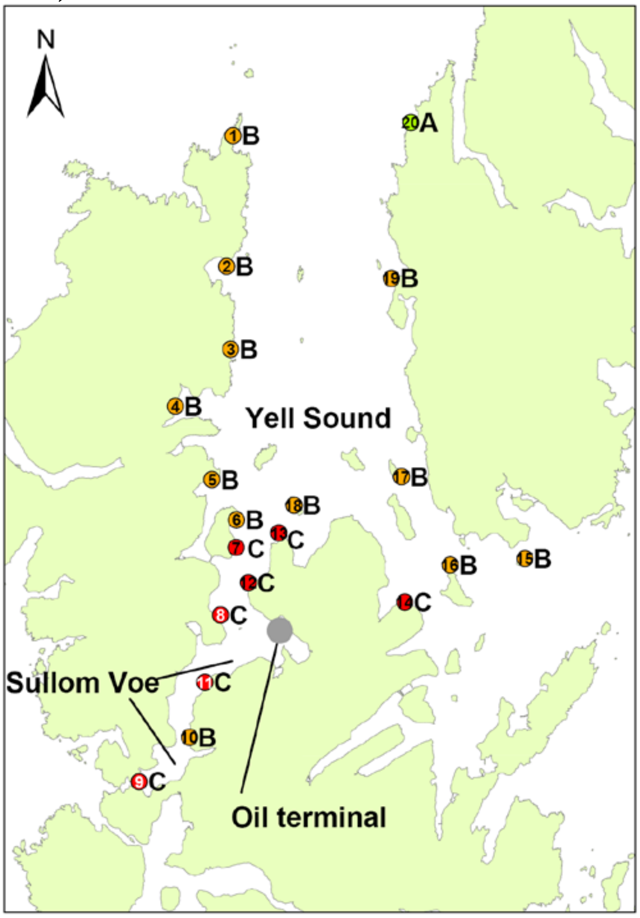

Figure 3. 2009 OSPAR classification status of sites based on VDSI measurements.

Over the time series of data for these sites, it is apparent that imposex has declined markedly since the early to mid-1990s when imposex was at its peak. VDSI was high within the Voe (>4 at many sites) for much of the period 1987 – 1999, while at sites outside the Voe, recovery of VDSI is evident in the late 1990s (Figure 4). Similarly RPSI, a less sensitive metric, shows recovery from early 1990 levels through to 2009 at highly impacted sites within the Voe (Figure 5). These results suggest that even prior to the complete ban on TBT, imposex levels were recovering as a result of reduced TBT inputs from changes in paint formulation and the reduction in shipping traffic (Figure 2).

A decrease in the incidence of female sterility in the most heavily affected populations near the terminal is also apparent. This recovery was noted by the late 1990s and, potentially as a result of the TBT ban on large vessels, 2009 is the first year that zero female sterility has been recorded at all sites in the area (Figure 6). The most significant reductions in VDSI at heavily affected sites have also been recorded between 2007 and 2009 following the ban on TBT on large vessels (Figure 4).

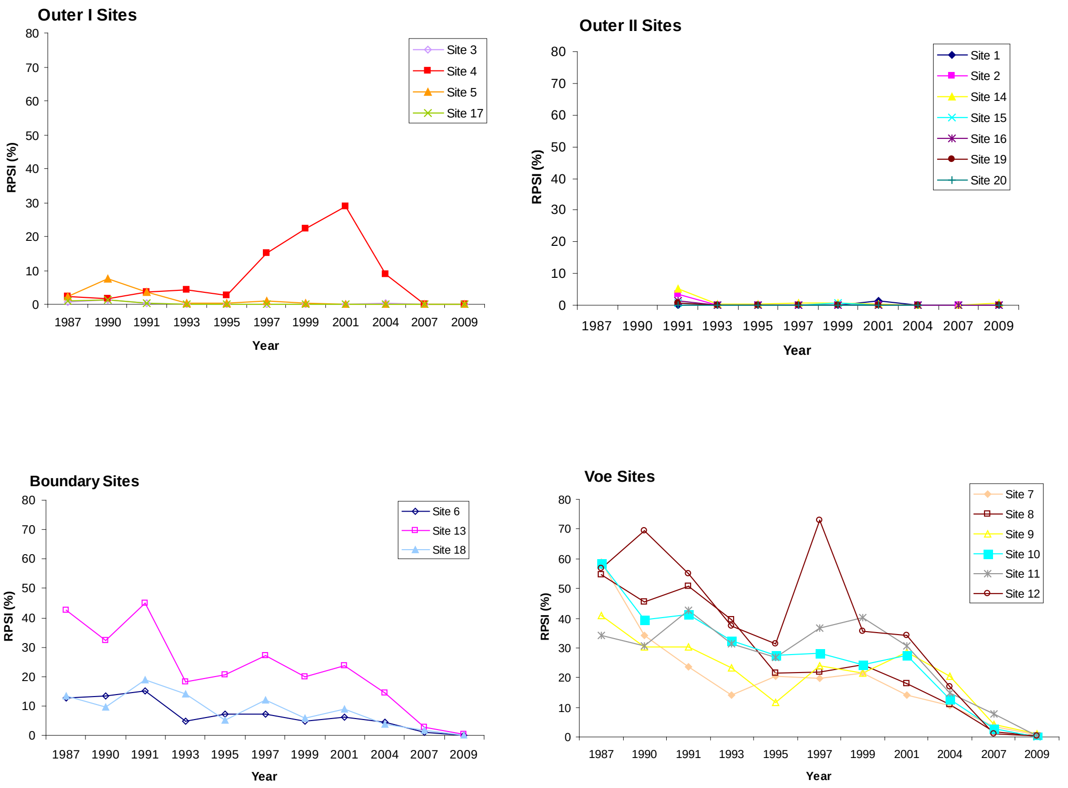

Figure 4. RPSI values for all adults in populations of sites separated by geographical grouping (1987 – 2009)

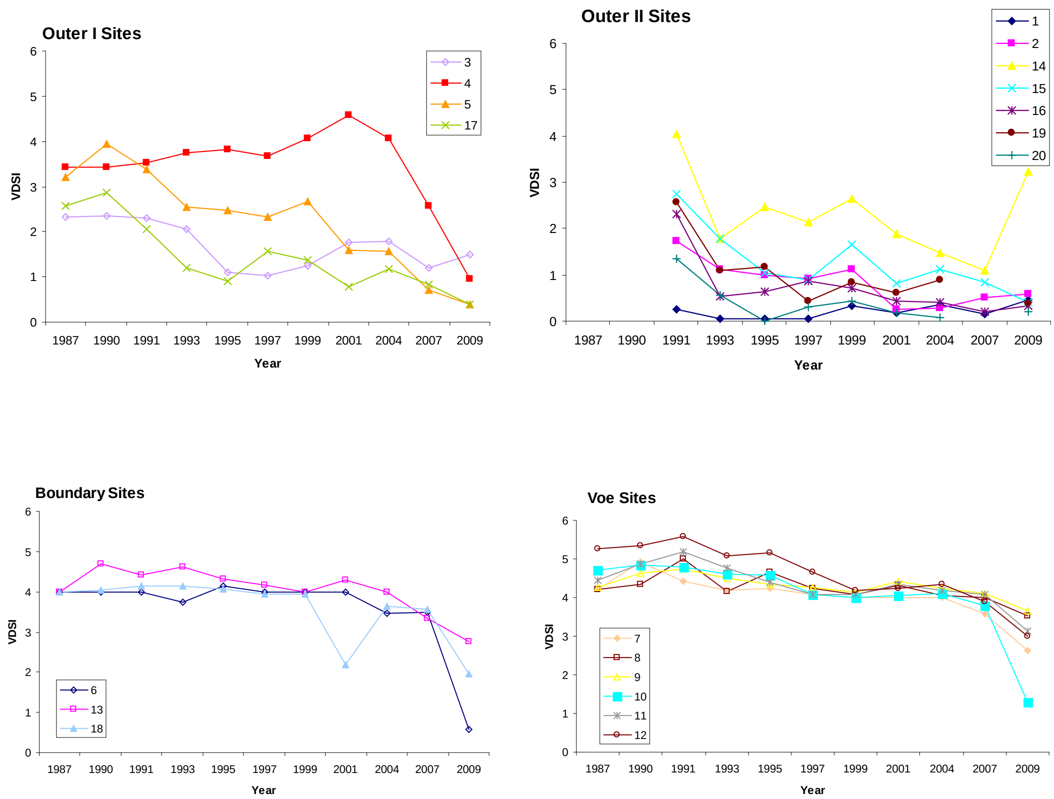

Figure 5. VDSI values for all adults in populations of sites separated by geographical grouping (1987 – 2009)

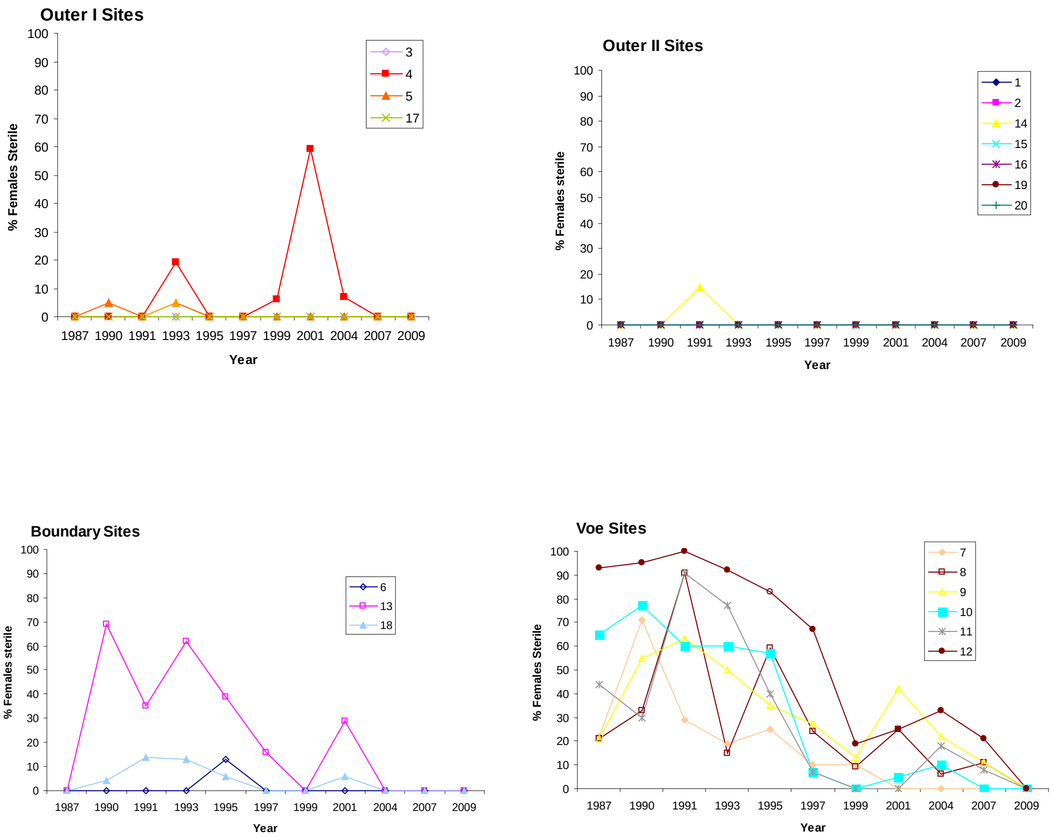

Figure 6. Percentage of adult females in each population recorded as sterile. Data separated by geographical grouping (1987 – 2009).

The continued presence of imposex in juveniles (Table 1) indicates that TBT exposure is still occurring, affecting young dogwhelks to a lower extent than previously recorded. This suggests that availability of TBT, perhaps from historical inputs to the sediments, is a residual issue now that direct inputs to the Voe have ceased.

There is also evidence from population abundance estimates that reproductive capability is returning to previously heavily affected populations. The Kames (site 12) is of particular interest, because of its proximity to the terminal, because it was the site where high levels of imposex first became apparent and because juvenile production was apparently non-existent there in 1991. The population in The Kames study area has increased considerably since then, particularly since 2004 (see Figure 7) (Moore & Gubbins, 2009).

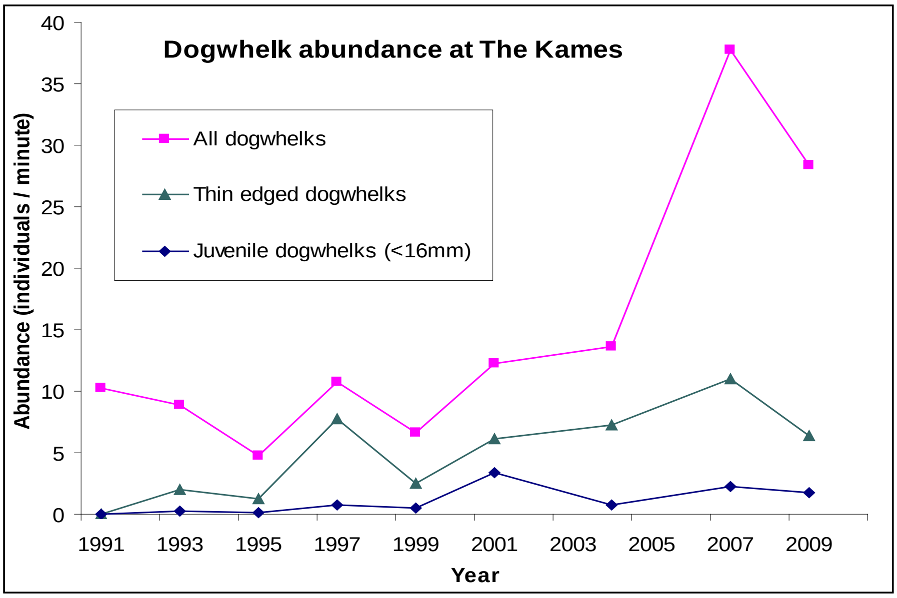

Figure 7. Increase in abundance of one of the most heavily affected populations in Sullom Voe at The Kames (1991 – 2009)

Conclusions

The study of dogwhelk populations at Sullom Voe Oil Terminal has generated one of the longest term biological effects monitoring data sets available. The results have shown how TBT inputs to a particular location have affected the most sensitive gastropod species, *Nucella lapillus*, over 20 years at the whole-organism and population level. The spatial extent and temporal trends of impact have been described and related to changes in TBT inputs. Most significantly this is the first published study that the authors are aware of to show significant recovery of imposex affected populations following the 2008 IMO ban on TBT on large vessels, with data from 2009 suggesting that for the first time in over 20 years, sterile females are absent from all previously affected populations.

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