

Translocation of the little whirlpool ramshorn snail:
Monitoring Update
November 2018

Highways England

ii

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1 Introduction

The little whirlpool ramshorn snail *Anisus vorticulus* is a small, aquatic snail with a dorsoventrally flattened shell approximately 5 mm in diameter. It is a UK Biodiversity Action Plan Priority Species and the only British non-marine gastropod which is a European Protected Species. It is also listed in Annex II of the EU Habitats and Species Directive and therefore requires the designation of Special Areas for Conservation (Annex II). In the UK, populations of *Anisus vorticulus* have been declining since the 1960s and although the precise cause is not clear, it is thought that drainage, over frequent dredging, and eutrophication are all likely to be contributing factors (JNCC, 2007; Van Damme, 2012).

Anisus vorticulus is also challenged by extreme dispersal limitation, which may prevent it expanding its range into suitable habitat even if conditions improve (Niggebrugge et al. 2007). Using translocation to assist with expanding the range of the species may therefore be appropriate, and to this end a Pilot Translocation of Anisus vorticulus was conducted in the summer of 2016 to test both the feasibility of such action and the response of the species to it (AECOM/Abrehart Ecology 2016a). The Pilot Translocation involved the movement of 800 Anisus vorticulus from (Figure 1), following an in-depth analysis of the habitat preferences of Anisus vorticulus to identify suitable donor sites (AECOM/Abrehart Ecology 2016b).

An initial monitoring survey of the newly translocated populations, from the Pilot Translocation, conducted in late October 2016, gave promising results regarding the survivorship of *Anisus vorticulus* at the receptor sites in (AECOM/Abrehart Ecology 2017b).

Following the Pilot Translocation in 2016 (AECOM/Abrehart Ecology 2016b) and the initial promising monitoring results (AECOM/Abrehart Ecology 2016c), the translocation exercise was extended in line with the original suggestions of the feasibility report (AECOM/Abrehart Ecology 2016b), whereby the translocation could be expanded based on the results of initial translocations, and the methods employed refined according to results. The second translocation involved moving additional *Anisus vorticulus* from the donor ditches at a new donor site at the working title of Translocation 2017, to differentiate the second phase from the Pilot Translocation. Due to a small number of *Anisus vorticulus* available for Translocation 2017 in June 2017, Translocation 2017 was also conducted in October/November 2017. Translocation 2017 is described in detail under separate cover (AECOM/Abrehart Ecology 2018b). This current report should therefore not be read in isolation and reference should be made to the Translocation 2017 translocation report.

As per the terms of the translocation licence, monitoring surveys will continue every six months over a period of five years post translocation. The findings presented in the current report are from:

- The fourth monitoring visit to the Pilot Translocation sites (July 2018)
- The fifth monitoring visit to the Pilot Translocation sites (November 2018)
- The second monitoring visit to the Translocation 2017 [June] sites (July 2018))
- The second monitoring visit to the Translocation 2017 [June] sites (November 2018)

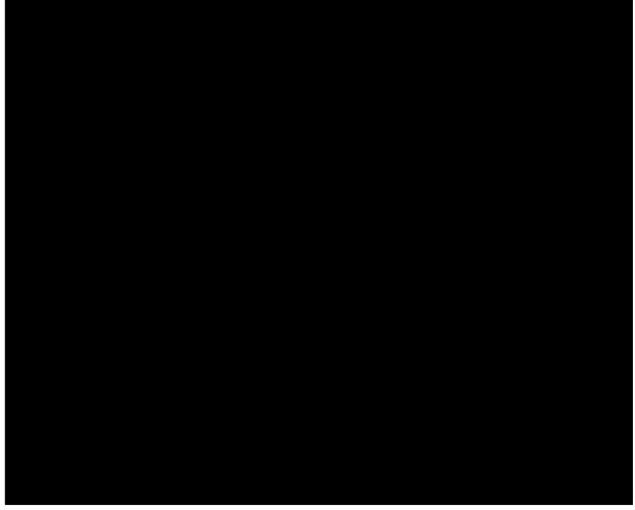


Figure 1. Location of

2 Methods

2.1 Licence requirements

2.1 Licence requirements
Natural England licences were required for each aspect of this project, including surveying (disturbing <i>Anisu vorticulus</i>), collecting full aquatic invertebrate samples for laboratory analysis (killing <i>Anisus vorticulus</i>), and for the translocating from the translocation of <i>Anisus vorticulus</i> , to the
Translocations of <i>Anisus vorticulus</i> were carried out in accordance with Translocation Licence 23292. Subsequent surveys and sample collection were conducted in accordance with Survey Licence 25961.
- -
Full Licence details are provided in Appendix A.

2.2 Monitoring Method

The Pilot Translocation of 800 *Anisus vorticulus* from took place in May 2016, with Translocation 2017 taking place in June and October/November 2017 (AECOM/Abrehart Ecology 2016b). During Translocation 2017, an initial 1,000 *Anisus vorticulus* were moved in June 2017, with a further 2,000 animals moved in October/November 2017.

The monitoring surveys presented here were conducted in July 2018 and November 2018, following an initial monitoring survey of Pilot Translocation in late October 2016 as reported in AECOM/Abrehart (AECOM/Abrehart Ecology 2016b). In 2017 a resurvey and additional translocations were conducted in June and October/November.

Data and sample collection was conducted by a pair of surveyors, including an experienced on-site mollusc surveyor period of the pair of surveyors, including an experienced on-site mollusc surveyor period of the period of the

2.2.1 Post 2016 and 2017 Translocation sampling

As part of the monitoring schedule for the pilot and	2017 translocations, post translocation	on sampling surveys were
conducted at all donor and receptor sites across	(Donor 1 and 2) and	(Donor 3 and Receptor
1) (Figure 1), and (receptor),	(receptor) and	(donor)
(location shown in Figure 1). The survey focused cl	hiefly on the presence/absence of liv	e <i>Anisus vorticulus</i> at the
receptor sites, and the continuing persistence and ne	ecessary abundance of Anisus vorticul	us at the donor sites.

The donor ditch monitoring was carried out in July and November 2018; whereby all ditches that had been identified as being suitable donor ditches for *Anisus vorticulus* were revisited. The ditches surveyed included both those part of the Pilot Translocation and the additional ditches included in Translocation 2017. Monitoring at the donor ditches aimed to ensure that the translocation has caused no long-term negative effects on *Anisus vorticulus* populations. In addition to checking the abundance of *Anisus vorticulus*, the wider mollusc community was assessed to ensure that the disturbance has not caused any long-term shifts in species composition and / or abundance.

Samples were collected using the sweep net method described in Section 2.2.2. Samples were taken at either end of the original donor ditch sections. As at the receptor sites, all mollusc species were identified in the field, and abundance of each was quantified. Any *Anisus vorticulus* present were individually counted and photographed for later age class assessment (methods described in Section 2.3).

2.2.2 Receptor ditch monitoring

In July and November 2018, all ditches that had received a population of *Anisus vorticulus* were revisited for the purposes of monitoring. The ditches surveyed included both those part of the Pilot Translocation and the additional ditches included in Translocation 2017 (and the purposes of monitoring repeatability, samples were collected using the same sweep netting method as the Pilot Translocation monitoring report (AECOM/Abrehart Ecology 2016c, first described in AECOM/Abrehart Ecology 2016a). This method was developed to minimise disturbance at the receptor sites, by taking smaller samples than using a typical sweep net protocol (as was used in the detailed surveys described in AECOM/Abrehart Ecology 2015, 2016a, and 2017b) (see photo number xxx). It is acknowledged that the simplification of the sweep technique may result in a slightly lower detection rate for *Anisus vorticulus* than the method described in AECOM/Abrehart Ecology 2016a; however, minimising disruption of translocated populations during monitoring was considered a priority.

Beginning directly next to the canes marking each original translocation placement point, a 0.5mm mesh net was drawn towards the bank in a single sweep, covering $0.25m^2$ to a depth of 25cm. The vegetation was gently agitated during the sweep, but any surrounding vegetation was disturbed as little as possible. Samples were also taken from 1m either side of the original placement points to assess whether the translocated individuals had begun to disperse within the ditch. During the October/November 2017 and 2018 re-surveys, additional samples were taken at midpoints between these main receptor sites, to further assess the potential spread of introduced animals along the ditch.

The material collected during the sweep was placed in a white gridded tray filled with water from the same ditch area. Molluscs were released from the collected vegetation by agitating the contents of the tray, after which excess vegetation was then removed. The floating contents of the tray (chiefly vegetation and larger invertebrate species) were poured back into the ditch, with molluscs retained in the bottom of the tray; it is accepted that a small proportion of molluscs may be lost at this stage, attached to some of the floating vegetation, but previous tests of this method have shown such losses to be negligible personant, pers. obs.). The material remaining was then evenly distributed across the tray for assessment. Any *Anisus vorticulus* present were individually counted and photographed for subsequent estimation of age (see Section 2.3). After identification, the sample was carefully replaced at the point of collection.

Figure 2. Locations of donor and receptor sites at



Figure 3. Location of donor sites at for Phase 2 of Translocation 2017.



Figure 4. Locations of receptor sites at the second of the



Figure 5. Locations of receptor sites at for phase 2 of Anisus vorticulus translocation in 2017.



2.3 Life stage classification

All Anisus vorticulus found at the receptor sites, and collected from the donor sites, were photographed to allow the subsequent assessment of the age profile of the population. The animals at each receptor site were grouped together on a laminated sheet of gridded paper, which was appropriately labelled. Using the photographs, the shell diameter of each individual was measured using ImageJ software (v1.50i; Rasband 1997-2016). The scale for measurements was set using the grid squares of the paper in each photograph. Shells were measured from the edge of the shell aperture, through the central point of the whorl of the shell, to the opposite outer edge of the shell (Figure 6). All measurements were taken in mm.

The Anisus vorticulus were classed by age according to their size following the guidelines devised by Glöer & Groh (2007). Those with a diameter <2.5mm were classed as juveniles; small adults were classed as between 2.5-3mm (this was the size at which copulation was first observed; Glöer & Groh 2007); large adults were classed as having a diameter >3mm.

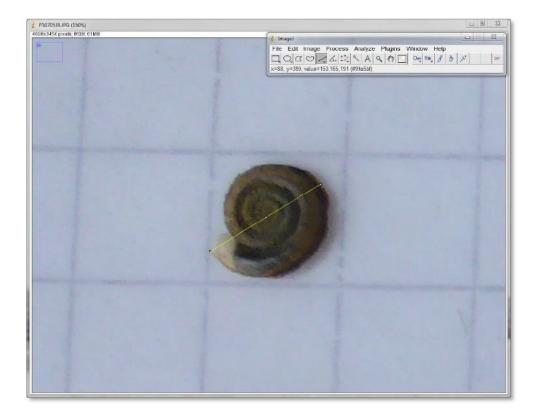


Figure 6. Measurement of shell diameter using Image J software. The full diameter of the shell was measured in each case, from the outer edge of the aperture, passing through the central whorl, to the opposite edge.

3 Results

3.1 Survivorship of translocated Anisus vorticulus

3.1.1 Overview of results

Table 1. Numbers of *Anisus vorticulus* placed during Pilot Translocation to provide the placed during placed during Pilot Translocation to provide the placed during six, twelve, eighteen and 26-month surveys post-translocation.

		Number of Anisus ve	orticulus			
Receptor site number	Number Translocated	Six-month check October 2016	Twelve-month check June 2017	Eighteen Month Check November 2017	26 Month Check July 2018	30 Month Check November 2018
1	100	10	2	25	13	15
2	100	6	2	17	8	1
3	100	39	0	9	0	1
4	100	29	0	29	35	11
5	100	10	3	2	1	3
6	100	18	0	6	1	7
7	100	2	0	0	0	0
8	100	11	0	10	8	1
Totals	800	125	7	98	64	39

3.1.2 July 2018

In July 2018, the forth monitoring visit to (26 month check) was undertaken. Live *Anisus vorticulus* were found at six of the eight receptor sites used as part of the Pilot Translocation. Where live specimens were found, the numbers encountered varied from very low (1) to significant (35). When compared to the June 2017 survey, this represented a significant increase in animals found - showing an upward trend in numbers of *Anisus vorticulus* at all receptor sites, excluding locations 3 and 7. No *Anisus vorticulus* have been found at Receptor Site 3 for the last two summer visits and no *Anisus vorticulus* have been found at Receptor Site 7 since the first 6 month check.

All but two specimens found in July 2018 were small juveniles (Figure 8). This contrasted with the 12-month check conducted in June 2017, where only large adult snails were found - though only seven individuals were found. This indicated that breeding had occurred in the past year. The numbers found during this re-survey (64) were significantly higher than in the previous early summer re-survey of 2017, when only 7 were found across all sites (Figure 9).

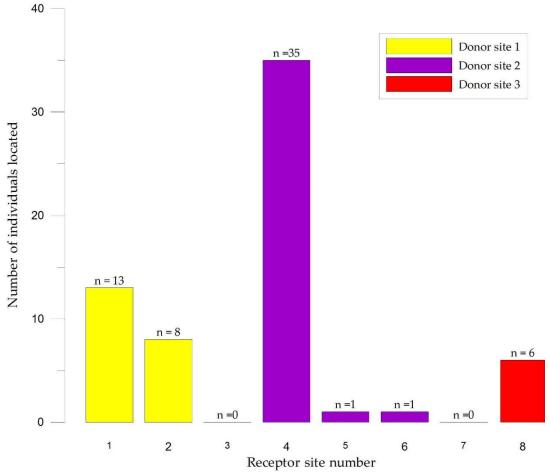


Figure 7. Frequency of living Anisus vorticulus observed at each receptor site at a living anisus vorticulus observed at each receptor site at living anisus vorticulus observed at each receptor site at living anisus vorticulus observed at each receptor site at living anisus vorticulus observed at each receptor site at living anisus vorticulus observed at each receptor site at living anisus vorticulus observed at each receptor site at living anisus vorticulus observed at each receptor site at living anisus vorticulus observed at each receptor site at living anisus vorticulus observed at each receptor site at living anisus vorticulus observed at each receptor site at living anisus vorticulus observed at each receptor site at living anisus vorticulus observed at each receptor site at living anisus vorticulus observed at each receptor site at living anisus vorticulus observed at each receptor site at living anisus vorticulus observed at living anison vorticulus o

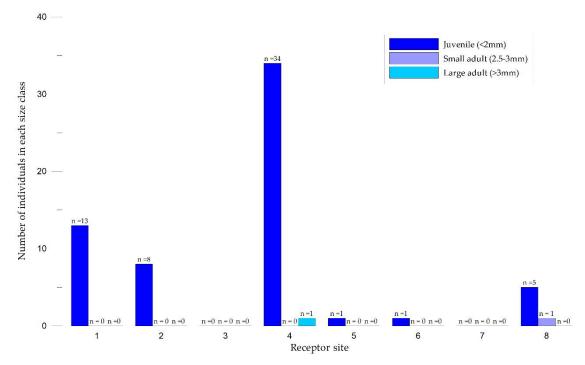


Figure 8. Age class of living *Anisus vorticulus* observed at each receptor site at a second in July 2018.

3.1.3 November 2018

At the end of November 2018, the fifth re-survey visit of the receptor was undertaken (30-month check).

It was found that the ditch suffered a disturbance incident in the late autumn of 2018. A cow had fallen into the ditch and spent at least a day trying to get out. During this time, it had walked along the bottom of the ditch.

As a result, vegetation and sediment

along much of the ditch was significantly disturbed – an area covering all but two of the receptor sites.

Despite this disturbance, *Anisus vorticulus* was found at seven of the eight sample sites (Figure 9). Numbers ranged from 0 to 15 animals found in a pair of sub-sample points.

A large proportion (99%) of the *Anisus vorticulus* found across the receptor sites were juveniles (Figure 10), these proportions are the reverse to those seen in the June 2017 survey, where only adults were found.

Of the 38 juveniles, 25 were above 2mm showing that they were approaching young adult size. Only 13 were smaller juveniles. The only young adult found was 2.6mm wide, showing it was only just into this size class. No full adults were located at this time.

At Receptor Site 7, no specimens of Anisus vorticulus were located for the fifth time.

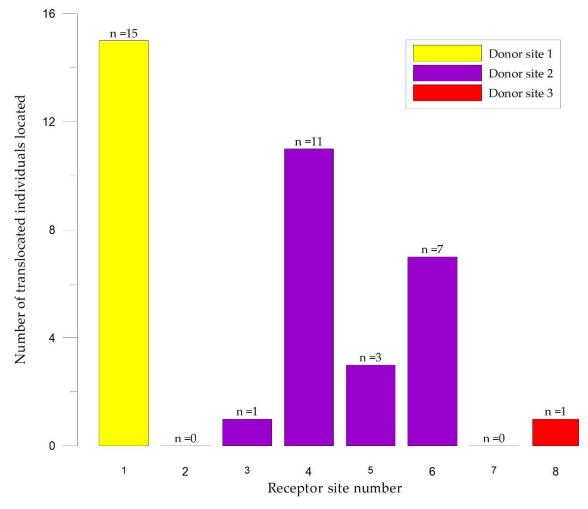


Figure 9. Frequency of living *Anisus vorticulus* observed at each receptor site at a living an in November 2018

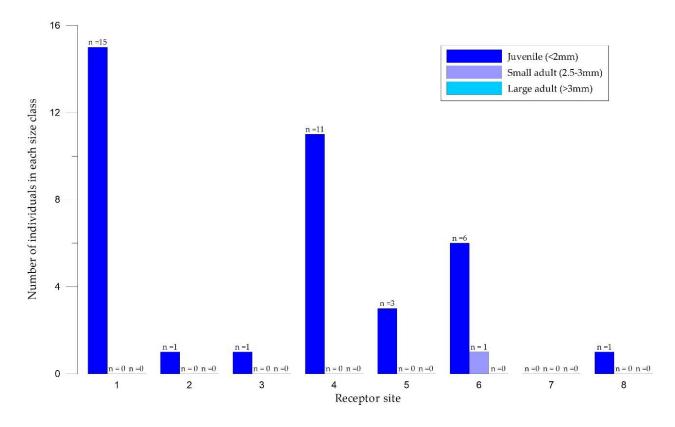


Figure 10. Age class of living *Anisus vorticulus* observed at each receptor site at a living an in November 2018.

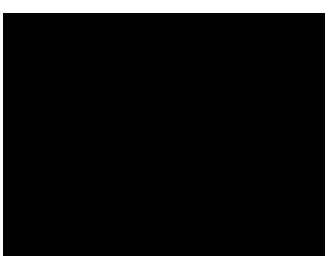
3.1.4 Damgate receptor sites - cow damage.

During the late autumn of 2018, Abrehart Ecology went to re-survey the receptor ditches at and found that both ditches had been heavily disturbed. The first two receptor locations were unaffected; however, all others had signs of considerable habitat movement. At this time of year there is a change in the vegetation visible within the ditches, with the water soldier (*Stratiotes aloides*) sinking to the bottom of the ditch and the frogbit (*Hydrocharis morsus-ranae*) disintegrating and forming turions for next year's growth phase. This type of ditch is uniform in appearance with occasional emergents still standing visibly in the ditch margins (Figure 13).



Figure 11. Normal looking ditch in July 2018 Figure 12. Cow disturbed ditch November

The ditch appearance on arrival in November 2018 was that of very disturbed ditch vegetation (Figures 11 - 13), which differed considerably from a ditch that is mechanically cleared - where the vegetation is removed from the ditch and placed as a spoil on the top and rear edge of the marsh. Here, the vegetation was disturbed and remained floating horizontally on the surface of the water. There was also evidence of the ditch base sediment through the water column and particularly visible on the surface of any *Stratiotes aloides* leaves. The water was opaque and the



sediment when disturbed had a very strong anoxic smell. It is assumed that the dying scattered vegetation was rotting and causing an anoxic layer to form. It was promising that there were *Anisus vorticulus* found in each of the sample locations, excluding Receptor Site 7. Site 7 has consistently had very low numbers from the beginning of the translocation, with only 2 animals recorded at the first 6-month survey and then none since. It is interesting that the donor population that came from the same marsh system are showing the least growth of all the donor populations.

Figure 13. Cow disturbed ditch | November 2018

3.2 Survivorship of translocated *Anisus vorticulus* to in July 2018

In July 2018, a monitoring survey of the Translocation 2017 receptor sites at was carried out.

Samples were taken 1m either side of the original placement points, as well as at the five release sites. A single simple sweep was taken at each location, as described in section 2.2.2. This reduced size sweep had shown to collect *Anisus vorticulus* at sites in Norfolk and Suffolk, but it may not always pick up very low numbers of *Anisus vorticulus*. This reduced sweep effort was important so as not to overly disturb the translocated population which had been moved within six months. In addition, four samples were taken from near the middle of the ditch, to assess if the population was spreading out from the original release sites at the ditch edge.

Results here showed that there was a continued presence of *Anisus vorticulus* within the ditch; however, numbers recorded were better than was expected for a summer survey. 11 *Anisus vorticulus* were found at site 1a, 22 at 2a, and 1 at 2b. In total, 40 *Anisus vorticulus* were found across the three receptor areas – comprising 21 sample points. These animals were mainly large adults, with an average size of 3.4 mm. The average of those translocated in June 2017 (Sample Site 1a) was 3.1mm. The low numbers of juveniles indicate that there was breeding on the site, although limited at this time.

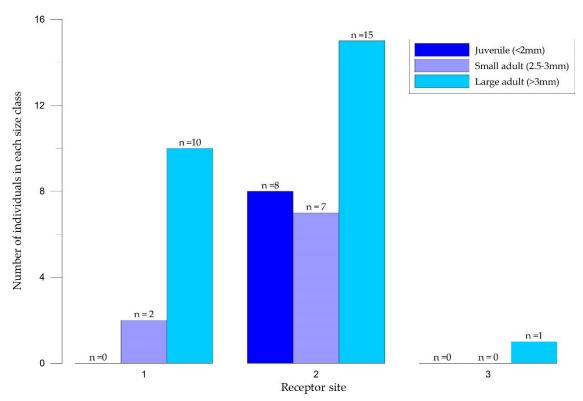


Figure 14. Age class of living *Anisus vorticulus* observed at each receptor site at site at July 2018. Shading indicates the number of individuals in different age classes.

3.3 Survivorship of translocated *Anisus vorticulus* to in November 2018

In November 2018, a monitoring survey of the Translocation 2017 receptor sites at was carried out

Again, samples were taken 1m either side of the original placement points, as well as at the five release sites. Sampling methodology was as those described in Section 3.2.

Results here showed that there was a continued presence of *Anisus vorticulus* within the ditch. The numbers of *Anisus vorticulus* at the receptor areas ranged from moderate (31 in 2b) to high (284 in 2a). *Anisus vorticulus* was found at each of the 15 receptor sub-sample sites, these numbers ranged from one to 122 in the south facing samples in area 2a. The majority of the *Anisus vorticulus* collected were juveniles, indicating a significant breeding event over the summer period. The presence of such a large number of juveniles indicated that there was significant breeding on the site.

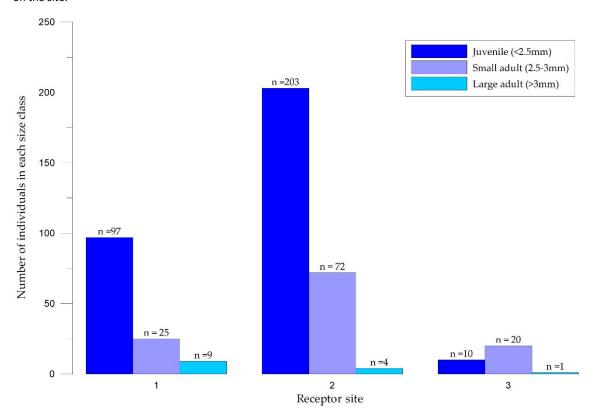


Figure 15. Age class of living *Anisus vorticulus* observed at each of the three receptor site at November 2018. Shading indicates the number of individuals in different age classes.

3.4 Survivorship of translocated *Anisus vorticulus* to in July and November 2018

In July and November 2018 re-survey surveys were undertaken at the three receptor sites at No Anisus vorticulus were found during the 2018 monitoring visits.

As no Anisus vorticulus were found during the re-survey the site, the reasons for this are unknown and a future study will attempt to study the reasons why.

3.5 Re-survey of *Anisus vorticulus* donor sites in July and November 2018

A re-survey of the four donor sites (four samples from two ditches at two samples from one ditch at at any and four samples from two ditches at two samples from two ditches at two samples from one ditch at age class and population size. This survey showed that there was still a high density of *Anisus vorticulus* within each of the donor ditches. Over the two survey periods the numbers of *Anisus vorticulus* collected increased at all sample points. Up to 550 *Anisus vorticulus* were found in individual samples within Donor ditch 4 in the November survey. This showed that there has been no discernable effect on the populations following the removal of *Anisus vorticulus* for the translocations. All other samples taken indicate that there were still strong populations at each site.

However, during July surveys only low numbers (123 *Anisus vorticulus*) were recorded across all the sample sites, with particularly low numbers at (5 *Anisus vorticulus*) and at

In the October /November donor site re-surveys 1079 *Anisus vorticulus* were collected and returned.

Site	Date	Sample Number	Number	
	July 2018	1a	35	
	July 2018	1b	8	
	July 2018	2a	12	
	July 2018	2b	22	
	July 2018	3a	1	
	July 2018	3b	4	
	July 2018	4a	29	
	July 2018	4b	12	
Site	Date	Sample Number	Number	
	November 2018	1a	47	
	November 2018	1b	17	
	November 2018	2a	19	
	November 2018	2b	22	
	November 2018	3a	21	
	November 2018	3b	11	
	November 2018	4a	392	
	November 2018	4b	550	

Table 2. Numbers of Anisus vorticulus found at each sample point at the donor sites.

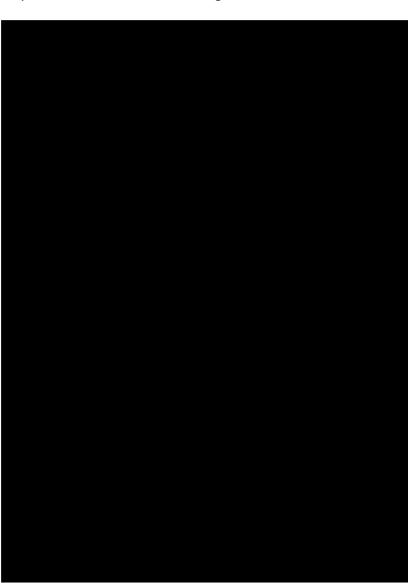
3.6 Colonisation of wider ditch areas

As part of the monitoring survey of all three receptor sites samples were taken one metre either side of the original translocation release points and at points between the receptor areas. This was to see if *Anisus vorticulus* was dispersing from the initial translocation sites.

3.6.1 2018

A number of samples were taken and *Anisus vorticulus* were found at several of these locations over the two visits (July and November 2018). Evidence for dispersal was found six months post-translocation within the receptor sites and at each period since. *Anisus vorticulus* were found in the 1m² areas immediately adjacent to the original translocation sites (AECOM/Abrehart Ecology 2018c).

At the mid channel samples were taken in the dense *Utricularia vulgaris* (Figure 16) covering the ditch, the samples were taken at full reach of the net going 120cm into the ditch from the edge. The samples were only taken from the surface 30cm of the vegetation.



Receptor site 1a (June 2017 translocation): the two positive samples (taken from sample site 1a -2 and 1a -4) had 5 adults and 3 adults respectively (Figure 17). This indicates the population has dispersed out into the ditch, possibly finding more suitable habitat than the margin. None were found around receptor site 2b.

Anisus vorticulus was found to be dispersing from the initial sites with animals being found to the east and west of samples areas 1a and 2a, with little colonisation in 2b at the western end of the ditch.

It was not possible to sample central ditch areas in November, as all vegetation had sunk at this time of year.

Figure 16. receptor site in July 2018

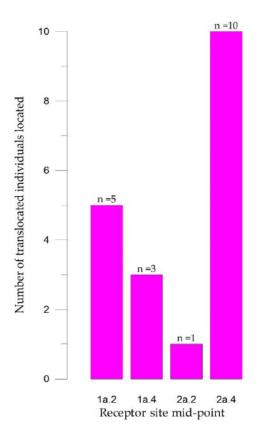


Figure 17. Frequency of living Anisus vorticulus observed at each mid-channel point between each receptor site Beccles Marshes (July 2018).

3.6.2

Samples were taken either side of the original translocation points, at 5m either side of these points, and mid-channel if possible (only possible if floating vegetation was present).

Only in the July re-survey period were any mid sample *Anisus vorticulus* located. In the north south receptor ditch *Anisus vorticulus* were only found between sites 2–4, and here only one animal was recorded. No animals were found in the two other sample points.

In the east west receptor ditch *Anisus vorticulus* were found in good numbers between sites 4-6, with a total of 20 found. Between points 6–8 three animals were found.

These numbers roughly correlate with the numbers found at each of the sample points, with site 4 consistently supporting the highest numbers.

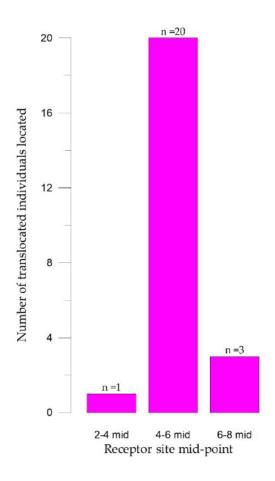


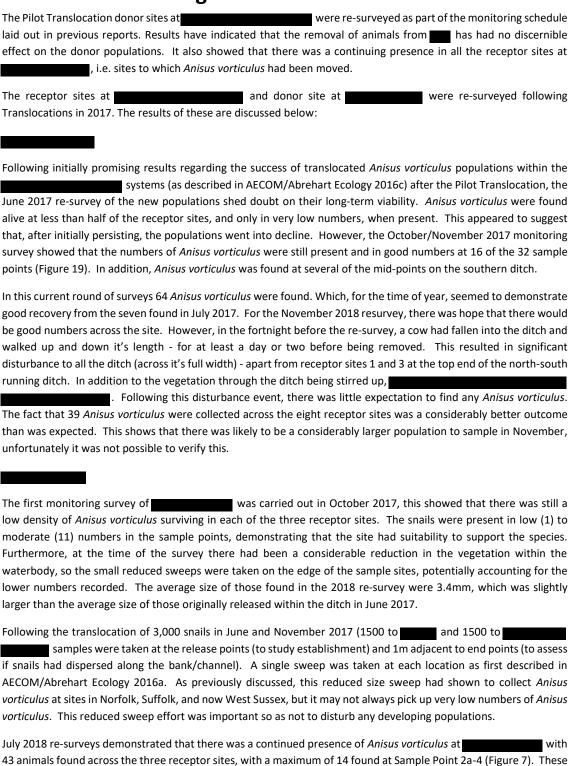
Figure 18. Frequency of living Anisus vorticulus observed at each mid-point between each receptor site

3.6.3

Within the sample sites no *Anisus vorticulus* were found at any sample site, either within the receptor sites or the mid points.

4 Discussion

4.1 2018 Monitoring results



were mainly large adults with an average size of 3.4mm; the average size of those translocated in the June 2017 was

3.1mm. Nine juvenile *Anisus vorticulus* were identified within the samples, indicating that there has been breeding on the site for the second year - though still limited at this time, this is unlikely to be a stress event reaction as over a year had passed and these records were for the second breeding period. At the time of the monitoring survey in November 2018 there had been a considerable reduction in the vegetation within the channel, so the reduced sweeps were taken on the edge of the sample sites. Previously this produced a lower number of *Anisus vorticulus* recovered. In November's survey the story was considerably different, with 441 *Anisus vorticulus* collected across the 3 receptor areas. The large majority of these (310) were juveniles at an average of 2mm, with the majority of these coming from receptor area 2, this was located on the northern bank of the ditch.

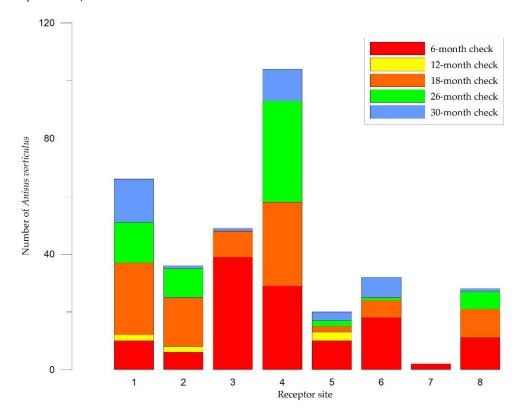


Figure 19. Combined numbers of *Anisus vorticulus* found at each pair of translocation site at at each survey period.

The second monitoring survey of ______ was undertaken in July 2018, no Anisus vorticulus were found. The habitat had not changed significantly (from surveys conducted in 2017) and as such the site is still considered suitable to support Anisus vorticulus. In July 2018 the water level was at least 30cm lower than in the November 2017 survey.

In addition to this the winter of 2017/18 was wet, with extensive flooding across the valley for a considerable period in March 2018 - ______ Though this may not affect Anisus vorticulus, the fact that there was a large flood event ______ means there may have been a considerable amount of pollutants from ______ introduced into the ditches, which may have influenced mollusc survival. Despite this, there are abundant populations of Segmentina nitida within this portion of the valley.

Monitoring conducted in June 2017 found limited numbers of *Anisus vorticulus* within the proposed donor ditch, though the November 2017 re-survey and translocation collection found high numbers. In the winter of 2017/18 these two ditches were cleared under a class licence (CLS002636) This clearance was of 50m sections on alternative sides of the ditches, leaving large areas uncleared. Though the central channel along the length had been removed, there were still wide (1-2m) ronds of flooded poached ditch margin remaining. In the cleared sections, the vegetation had been removed leaving a shallow profile margin that was clear of vegetation and consisted of exposed soils. In March 2018 a brief look at these areas still revealed a low density of *Anisus vorticulus* within the cleared areas. The July 2018 survey found high numbers within the sample area and a complete cover of frogbit (*Hydrocharis morsus-ranae*) and ivy-leaved duckweed (*Lemna trisulca*) and the November 2018 survey found very high numbers of *Anisus vorticulus* with 615 and 849 found in two simple samples taken. This indicates that the clearance of the ditch has created an environment that was suitable for a considerable breeding event. This density of population was scattered along this ditch, as assessed during the monthly monitoring aspect of this project.

This large number of *Anisus vorticulus* found in the recently cleared ditch is something that has been noted across the over the summer of 2018, whether it is to do with the hot summer or better detectability this year is hard to say, though investigations into this are being looked at.

4.1.1 Anisus vorticulus population and habitat changes

Seasonality

The abundance of *Anisus vorticulus* and the age profile in populations can vary considerably from month to month within the same ditch. As part of this project, a monthly monitoring study has been started with the aim of determining how *Anisus vorticulus* responds over a single 12 month period. Seven ditches were chosen, four as the donor sites already studied and three new sites as found during separate survey for the Broads Authority. These new monitoring sites are all within the sample site, two points have been chosen at a 20m interval along a ditch. At each of these points a single simple sample is taken – as described earlier in this report. This is rinsed in the field and the *Anisus vorticulus* are picked out and placed onto a gridded sheet of paper for measuring to enable a size class assessment to be made.

The 2018 fieldwork was started later than in other years due to contractual arrangements. So, the sampling has shifted from May in 2016 to July in 2018. The winter sampling is now in November. In the previous report we discussed the potential difficulty in finding small juveniles in a sample and that they may be washed out of the sample. Our studies to date have shown this not to be the case. If juvenile *Anisus* vorticulus are in the sample, our sampling methods are likely to locate them. A 500micron net was used for all the sampling and over 40 juveniles under 500 micron have been collected and measured to date. In some of the samples there were still large numbers of these small juveniles, though they were not all picked out due to time restraints. In the first four months of sampling nearly 13,000 *Anisus vorticulus* have been collected and picked out. As many live animals are returned to the sample areas as possible following this procedure.

Inter-annual variability

There appears to be co	onsiderable variation in the populations at each donor site from year to year. $ $	īhis year (2018)
several surveys were ca	arried out across the UK, with a large number of samples taken along	and
at	. There were large numbers of Anisus vorticulus at some of these s	ites ,with up to
2000 Anisus vorticulus	s collected in a single sample point in some ditches	
). The	long hot summer may have contributed to this being a 'good year' for Anisus v	orticulus.

There is little data available regarding the inter-annual fluctuation of absolute numbers of *Anisus vorticulus* within sites, and such information would be useful to know when planning conservation translocations. This project is collecting this inter annual data from the monitoring work being carried out at each donor and receptor site for five years post translocation and the two years post translocation. For example, knowing the extent to which population levels can vary could help to determine what level of buffering is needed in terms of numbers to allow a population to persist in the longer term.

Insufficient population size

This recent round of sampling has shown that the population had in general increased over the twelve-month period since the Pilot Translocation and Phase 2 translocations were conducted. At there were relatively high numbers recorded in July and, although the November numbers were low, the cattle disturbance event will have influenced later samples. At present, it appears that the sample numbers placed at each location (50 per sub sample at was sufficient to produce a viable population - following the latest re-survey results.

At 100 animals were placed at each of the 15 sample points, in July there were low numbers re-found, though this was very different in the November surveys. Here too it appears that there were sufficient numbers moved.

Future re-surveys will show continued trends across the sample sites.

Abiotic variables

It has been noted that *Anisus vorticulus* builds up numbers very quickly under optimal conditions but does not thrive under sub-optimal conditions (Glöer & Groh 2007). It may therefore be the case that the environment in parts of the receptor ditches are not fully appropriate to support a long-term population of *Anisus vorticulus* (Recent work at has shown considerable variability along the length of a ditch. Where the population varies considerably along the length of different ditches (Abrehart Ecology 2018). As part of the monthly monitoring aspect of this project, samples were taken at 10 intervals along each donor ditch and has shown limited variation along the ditch length.

For the initial Pilot Translocation, receptor sites were chosen based on a multivariate analysis of the vegetation and mollusc communities present (see AECOM/Abrehart Ecology 2016a, 2016b, 2016c), which indicated a strong association of *Anisus vorticulus* with certain groups of plants and other mollusc species.

Some preliminary tests of abiotic factors were conducted during the initial analysis, including neighbouring land use and ditch profile, but water chemistry was not initially considered as it was believed that variations in plant communities would reflect the underlying abiotic variables. However, samples were collected for water chemistry analysis during the summer of 2016. Subsequent analysis of the data indicates that water chemistry is an important factor to consider in relation to *Anisus vorticulus* going forward. For example, a strong inverse correlation was found between the presence/absence and abundance of *Anisus vorticulus* and calcium concentrations (one-way ANOVA: $F_{1,41}$ =10.16, p<0.005), in agreement with the findings of Watson & Ormerod (2004). There is also a suggestion of a strong correlation of the presence/absence of *Anisus vorticulus* with levels of total organic carbon (one-way ANOVA: $F_{1,41}$ =10.57, p<0.005). While a relationship with calcium concentration is most likely to be a product of underlying geology and hydrology, the latter may relate to factors such as ditch clearance frequency and adjacent land use. It was also noted that the receptor locations appear to be located in areas of sub-optimal levels for these two variables.

Studies to look at variability in water chemistry along the length of a ditch is being looked at as part of other aspects of this project and will be reported on separately.

vegetation communities.

5 Future Work

The relatively poor understanding of the requirements of *Anisus vorticulus* as a species make it difficult to determine the precise reason (or reasons) for the declines and increases observed, and more research is needed to shed light on the problem. A concurrent project is currently looking at the monthly distribution of *Anisus vorticulus* at seven sites across Suffolk and Norfolk – including those used as donor sites for the three phases of the translocation. This study is looking at the monthly variation in numbers and age of *Anisus vorticulus* collected. This work will enable us to analyse the changes in population age structure at seven distinct sites supporting *Anisus vorticulus* across a single year.

vorticulus collected. This work will enable us to analyse the changes in population age structure at seven distinct sites supporting Anisus vorticulus across a single year.
A final translocation of <i>Anisus vorticulus</i> was carried out in October/November 2018 to
This Highways England work is being carried in conjunction with other projects with this species across the Broads NP (Broads Authority) and Pulborough Brooks (RSPB) in West Sussex. The Broads Authority studies have managed to locate two new marsh systems that now support <i>Anisus vorticulus</i> that in previous surveys (in 1997) were not identified. In addition this 2018 survey also found three marsh systems where it was no longer found. The number of samples taken may simply have missed any low-density levels of <i>Anisus vorticulus</i> possibly present. The survey is looking at effects of ditch maintenance and management regimes on the populations of <i>Anisus vorticulus</i> across a single marsh system.
Monitoring at six-monthly intervals will continue as per terms of translocation licence, but more regular (monthly) monitoring of a subset of ditches (at and another reference site) will give a clearer indication of the annual population dynamics of the study populations.
Further insights into the role of water chemistry on the distribution of <i>Anisus vorticulus</i> will need to be looked. In addition to the strong relationships observed between calcium and total organic carbon and the presence/absence of <i>Anisus vorticulus</i> at studies on other populations in the UK have indicated that water chemistry plays an important role in site suitability (e.g. Watson & Ormerod 2004). Assessment of the levels of these and other chemical components may therefore provide vital information as to the potential suitability of sites as receptor locations for future translocation, as closer matches could be made between environmenta conditions. By taking water and sediment samples at all sites surveyed to date, levels of different chemical components (such as calcium, nitrates, and phosphates) could be assessed in relation to the presence/absence of <i>Anisus vorticulus</i> identified during detailed surveys in 2016-2017. This would give a better indication as to whether relationships with certain chemicals are consistent across as in the conditions.
conditions. By taking water and sediment samples at all sites surveyed to date, levels of different chemical components (such as calcium, nitrates, and phosphates) could be assessed in relation to the presence/absence of Anisus vorticulus identified during detailed

process for choosing future receptor sites. Sampling water from multiple ditches may also indicate certain portions of different sites that are more or less suitable. For example, when mapped out the data from showed variability at a fine scale within the site, resulting in some ditches being occupied by *Anisus vorticulus* while other remained vacant, despite apparently suitable

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Appendix A - Licences