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LAGOONAL LITTORINIDS: SHELL SHAPE AND SPECIATION

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ABSTRACT Variables related to shell shape have been measured in littorinids from brackish lagoons and coastal sites. After removal of size related effects, the data were analyzed using multivariate techniques. On Canonical Variate 1 there was good separation of the shells of the lagoonal animals from those of animals from the coast and a tidal lagoon. The former, for example, had lighter, and therefore thinner, shells for any given shell size and a smaller jugosity of the aperture lip. The lagoonal shells from Golam Head and the coastal animals from Robin Hood's Bay could each be separated clearly from the other samples. Although there are clear morphometric differences in the shells, it is not possible without appropriate breeding experiments to raise the lagoonal animals from *L. saxatilis* var. *lagunae* (*L. tenebrosa*) to species status. The importance of conserving lagoonal habitats is considered in terms of the preservation of biodiversity.

KEY WORDS: Littorinids, brackish lagoons, *Littorina saxatilis* var. lagunae

INTRODUCTION

There are three clearly recognizable taxa of rough periwinkles on European shores, the ovoviviparous *Littorina saxatilis* (Olivi), and the oviparous *L. arcana* Hannaford Ellis, and *L. compressa* Jeffreys. The last two taxa are non-contentious with regard to their species status. However, *L. saxatilis* has a wide range of habitats and includes populations of differing shell morphology some of which are found in discrete environments. This has led to various attempts to separate taxa from within this complex. Two of these are worthy of further investigation, i.e. *L. neglecta* Bean, which is found in the barnacle zone living sympatrically with "normal" *L. saxatilis* (*L. saxatilis* B) (Grahame et al. 1995, Hull et al. 1999), and *L. tenebrosa* (Montagu) which occurs in brackish lagoons (Barnes 1993). Two other forms within *L. saxatilis*, H and M, have also been recognized (Hull et al. 1996).

The subject of this paper is on littorinids that inhabit brackish lagoons. There are two problems. Firstly, the use of "*tenebrosa*" is confusing and is hence probably inappropriate (Barnes 1993). In the past it has been used in a very broad sense to include any littorinid with a high-spired shell occurring in sheltered locations including lagoons (Forbes & Hanley 1853). It has also been used in a much more restricted way to include only those littorinids which (a) have a small (usually <6 mm high), very fragile, smooth, plumply-whorled shell which is black or brown and often reticulated, and (b) live permanently submerged on macrophytes (such as *Chaetomorpha*) in brackish lagoons (Muus 1967).

Secondly, the situation is compounded by the presence in some lagoons of littorinids which fit the above description of *L. tenebrosa* whereas in others there are animals which live on the substrate, fit the wider definition of Forbes & Hanley (1853) and may be referred to as *L. saxatilis s.s.* (e.g. Barnes 1987). Furthermore, both forms have been reported as occurring in the same lagoon in some instances (Smith 1982), although Barnes (1993) was only able to find *L. saxatilis s.s.* in the Fleet, Dorset and Cemlyn Lagoon, Anglesey, at which sites *L. tenebrosa* had also previously been reported (Seaward 1980; Barnes 1987).

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Muus (1967) and Smith (1982) suggested that the "*tenebrosa*" animals are probably a distinct species, whereas Barnes (1993) concluded that, on the basis of shell variables, this is not the case and that, although the two forms appear to be reproductively isolated, they should be referred to as *L. saxatilis* var. *lagunae*. Reid (1996) also could find no case for species status for this form. In a preliminary study on five allozyme loci, Gosling et al. (1998) concluded that the two forms are genetically differentiated. However, in a more detailed investigation of 12 polymorphic enzyme loci, Wilson et al. (1999) found no allele unique to either form, and concluded that, although there is a barrier to gene flow between them, they are not distinct species.

Barnes' (1993) conclusions were based on five measurements of shell variables and two of operculum variables. The present study extends the number of measured variables and the number of populations in an attempt to clarify the situation, particularly since ecological barriers can result in populations diverging to species status in spite of close similarities at the molecular level (Morell 1999).

MATERIAL AND METHODS

Samples of lagoonal littorinids were obtained from both tidal and isolated habitats (Table 1, Fig. 1). In eastern England 28 lagoons at nine sites were visited. Littorinids were found in only five of these, representing four sites. They were also found in the Fleet in southern England and at Golam Head in the west of Ireland.

The Fleet is a tidal lagoon, open at its eastern end to the English Channel; the sample was taken from gravel on the seaward side of the lagoon near its eastern end (East Fleet). At Golam Head the sample was taken from the landward end of the lagoon near a small freshwater inlet and about 100 m from the seaward end. The animals were completely submerged on the alga *Chaetomorpha*. At its seaward end there are rocks which are continuous with those of the adjacent shore. At Alderton the lagoon is surrounded by shingle on its seaward side and is separated from the sea by a shingle dune about 6 m high. The animals occurred over a short stretch on this side; they were found on small stones near and at the water's edge and on the surface of the mud. Lagoonal littorinids were found in two lagoons at Cley which again were separated

TABLE 1.

Lagoonal and coastal sites.

Location	National Grid	N
Isolated lagoons		
Alderton, Suffolk, England	[1]TM 363419	53
Cley Eye, Norfolk, England	[1]TG 067447	50
Holkham Hole, Norfolk, England	[1]TF 886451	72
Snettisham (Shepherd's Port), Norfolk, England	[1]TF 649319	32
Lagoon with occasional incursions of sea water		
Golam Head, Galway, Ireland	[2]L 826214	54
Tidal lagoon		
The Fleet (east end), Dorset, England	[1]SY 664757	53
Open coast		
Golam Head, Galway, Ireland	[2]L 826214	38
Robin Hood's Bay, Yorkshire, England	[1]NZ 957058	50
Wells-next-the-sea, Norfolk, England	[1]TF 915456	50

[1] British National Grid; [2] Irish National Grid.

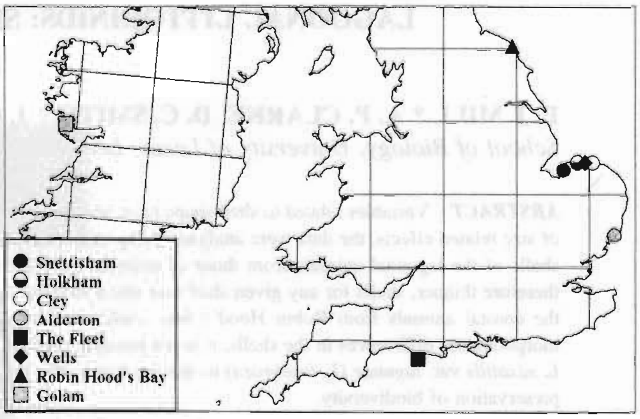

Figure 1. Location of sampling sites in Britain and Ireland.

from the sea by a gravel dune about 7 m in height. They occurred both on algal (*Chaetomorpha*) mats and on stones. At Holkham the lagoon was on the landward side of a very mature dune about 8 m in height covered in trees (Scot's Pine, Holm Oak, and Birch) and bushes. Similarly, the lagoon itself was largely surrounded by trees and bushes. The animals were found on the seaward side of the lagoon completely submerged on the alga *Chaetomorpha*, although some were also found on submerged wood. At Snettisham the lagoon was separated from the sea by a mature sand dune about 6 m in height. The animals were found completely submerged on stones well away from the edges of the lagoon.

At Golam Head tidal incursions occur on spring tides (Wilson et al. 1999). At Snettisham, sea water incursions are unlikely and would certainly be rare; at Alderton and, particularly, at Cley they would probably be even less likely, while at Holkham the only possible connection with sea water would be underground through the substrate. All of the lagoonal sites except Holkham were open and devoid of tall vegetation in their immediate vicinity. There were no coastal sites with littorinids in the close proximity of Alderton, Cley or Snettisham; the nearest coastal site to Holkham is 2.35 km away at Wells-next-the-sea. The condosity of the water was checked at Alderton and Cley and was in excess of 85% seawater.

Other samples were taken from rocky shores on the coast or, in the case of Golam Head, at the entrance to a lagoon (Table 1, Fig. 1). The sample from Golam Head was taken at the seaward end of the lagoon where the rocky shore merged with the edge of the lagoon, providing a very sheltered habitat. This was about 100 m from the site where the lagoonal sample was collected. The sample from Wells-next-the-sea was the closest site to the lagoon at Holkham (2.35 km). Robin Hood's Bay was chosen as representing a typical, somewhat sheltered, east coast boulder site.

The conventional measurements used in previous studies of

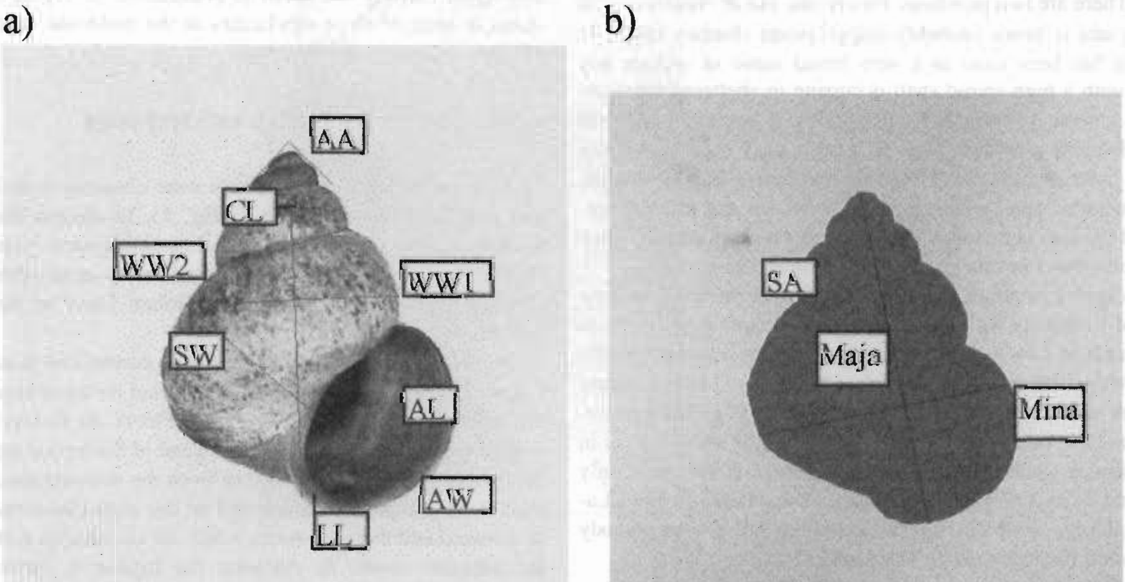

Figure 2. The measurements made on (a) the shell profile and (b) the shell silhouette. AA, apical angle, AL, aperture length (excluding the lip); AW, aperture width; CL, columella length; LL, lip length; Maja, major axis; Mina, minor axis; SA, surface area (in profile); SW, shell width (excluding aperture width); WW1, whorl width 1 (width of whorl at right angles to the columella axis); WW2, whorl width 2 (across the suture between the first and second whorls). In addition shell weight was measured.

littorinids were made, i.e. columella length, lip length, aperture length, width of first whorl at right angle to the columella axis, width of shell minus the aperture, aperture width, width of the suture between the first two whorls and the apical angle (Grahame et al. 1995). Three additional measurements were made, i.e. major axis (maximum linear dimension), minor axis (maximum linear dimension at right angles to the major axis) and shell profile area (Fig. 2). The shells were also weighed.

Canonical Discriminant Analysis, Principal Component Analysis, Factor Analysis and Discriminant Analysis were all carried out on the data after removing the effects of size by standardizing using the geometric mean (except of course for apical angle). Discriminant analysis was also carried out on the raw data.

RESULTS

The animals from Holkham and Golam Head were identified provisionally as *L. tenebrosa* on the basis that they lived permanently submerged on *Chaetomorpha*, their shells were less than 8 mm high and, particularly in the case of those from Holkham, were plumply whorled (Fig. 3a, b). Animals from East Fleet, Alderton and Cley possessed shells which were much more pointed (Fig. 3d-f); those from East Fleet reached 9 mm, while those from Alderton and Cley reached about 13 mm and 14 mm in height respectively; they were not permanently submerged. The shells of animals from Snettisham were somewhat intermediate (Fig. 3c); they were rather less bulbous than the Golam and Holkham animals, were found on stones not algae, but were permanently submerged and were less than 7 mm in height.

The shells of the coastal animals from Golam, Wells and Robin Hood's Bay were all fairly pointed and had a clear jugosity (relative aperture lip length) (Fig. 3g-i). Of the lagoonal animals, only the shells of those from East Fleet had anywhere near the same degree of jugosity. The Robin Hood's Bay shells reached a height of about 12 mm; those from Golam and Wells reached about 16 mm.

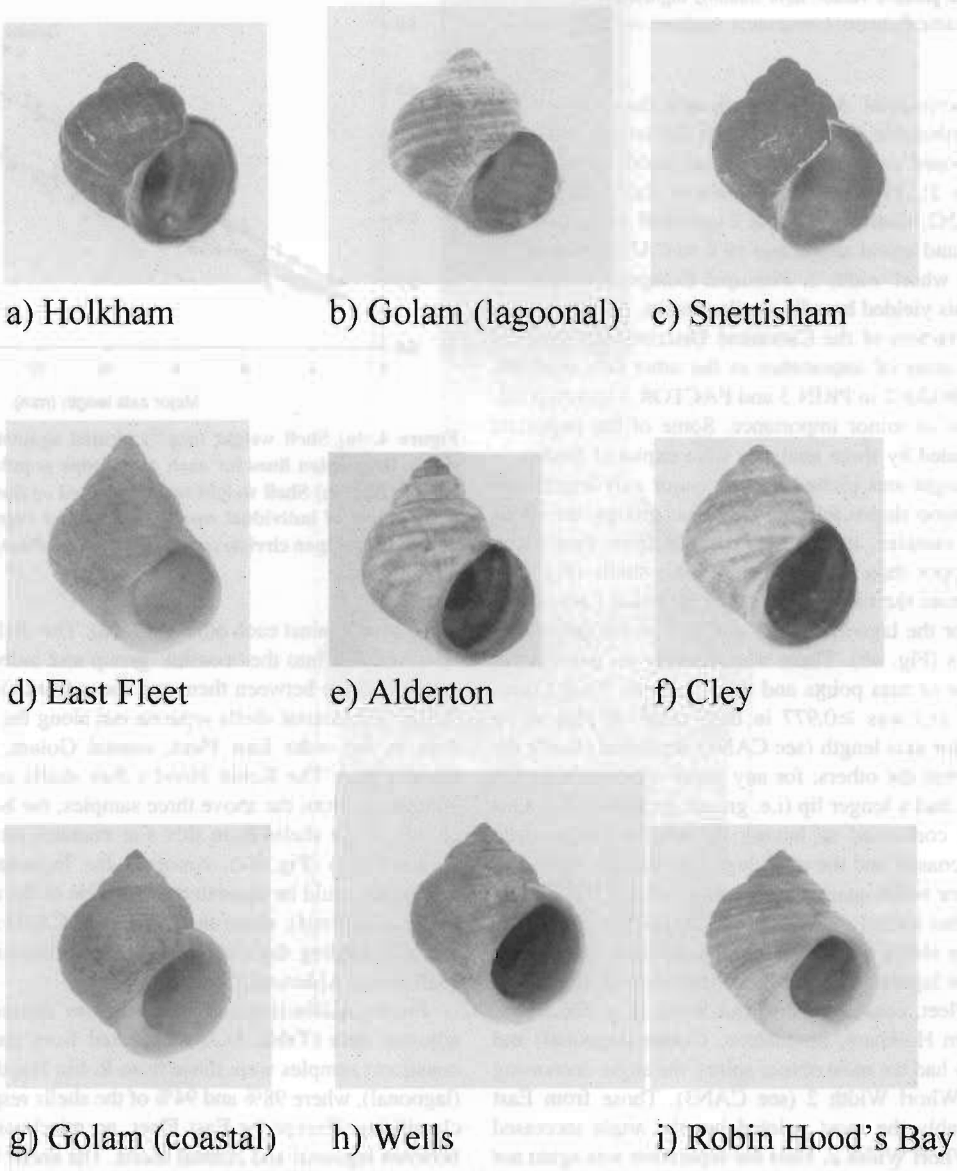

Figure 3. Profiles of shells from the localities stated. All shells are adjusted to the same overall width. Actual columella lengths are: (a) 5.1 mm, (b) 5.1 mm, (c) 5.5 mm, (d) 6.5 mm, (e) 7.2 mm, (f) 6.3 mm, (g) 11.5 mm, (h) 9.0 mm, (i) 9.2 mm.

TABLE 2.

Canonical Discriminant Analysis of the data after removing the effects of size by standardizing using the geometric mean.

	CAN1 51%	CAN2 23%	CAN3 18%
LL	0.9294	−0.2868	0.0811
WEIGHT	0.7228	0.1489	0.1981
SW	−0.0964	0.6524	0.4566
AA	−0.1989	−0.4188	0.6534
CL	−0.2341	0.4524	−0.2457
MINA	−0.4030	0.1948	0.4910
WW2	−0.4707	*0.6520	−0.4328
AL	−0.5671	0.1604	0.3739
AW	−0.6996	−0.4945	0.2274
MAJA	−0.7367	0.3475	0.0215
WW1	−0.7956	0.0083	−0.1916
SA	−0.8302	0.1815	0.4626

Dark shading, highest positive values; *light shading*, highest negative values.
* Not a high value using Principal Component Analysis and Factor Analysis.

Canonical Discriminant Analysis indicated that 51% of the variation was attributable to CAN 1, with lip length and shell weight being opposed to surface area, whorl width 1 and major axis length (Table 2). Twenty three percent of the variation was attributable to CAN2, with whorl width 2 and shell width opposed to aperture width and apical angle, and 18% to CAN3, with apical angle opposed to whorl width 2. Principal Component Analysis and Factor Analysis yielded broadly similar results, except that the second and third factors of the Canonical Discriminant Analysis were reversed in order of importance in the other two analyses. However, Whorl Width 2 in PRIN 3 and FACTOR 3 (corresponding to CAN2) was of minor importance. Some of the important relationships revealed by these analyses were explored further.

When shell weight was plotted against major axis length (see CAN1) the regression slopes fell into two clear groups, the shells from the coastal samples, together with those from East Fleet, having steeper slopes than the other (lagoonal) shells (Fig. 4a). Pooling the data from the two groups gave regression lines with a slope of 0.0448 for the lagoonal shells and 0.0604 for the coastal + East Fleet shells (Fig. 4b). There was virtually no overlap between the two sets of data points and the Spearman Rank Correlation Coefficient (r_s) was ≥0.977 in both cases. A plot of lip length against major axis length (see CAN1) separated clearly the coastal samples from the others; for any given major axis length the former always had a longer lip (i.e. greater jugosity). The East Fleet shells were confirmed as having lip lengths intermediate between those of coastal and the other lagoonal samples (Fig. 5a).

Plotting aperture width against shell width (see CAN2) did not separate the samples entirely along coastal versus lagoonal lines. Robin Hood's Bay shells were more similar to those of lagoonal animals, except for lagoonal Golam shells that were grouped with those from East Fleet, coastal Golam, and Wells (Fig. 5b).

The shells from Holkham, Snettisham, Golam (lagoonal) and Robin Hood's Bay had the most obtuse spires, the angle decreasing with increase in Whorl Width 2 (see CAN3). Those from East Fleet were noticeably the most pointed but the angle increased with increase in Whorl Width 2. Thus the separation was again not strictly coastal versus lagoonal (Fig. 5c).

CAN 1 expressed most clearly the separation of coastal and lagoonal animals and this is seen when the three canonical variates

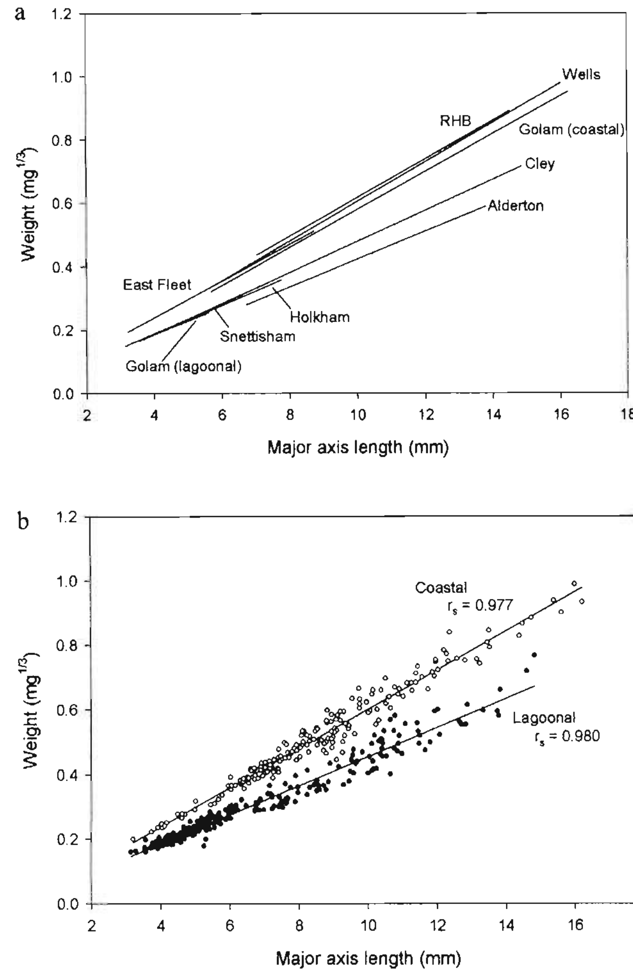

Figure 4. (a) Shell weight (mg$^{1/3}$) plotted against major axis length (mm). Regression lines for each of the nine populations. RHB, Robin Hood's Bay. (b) Shell weight (mg$^{1/3}$) plotted against major axis length (mm). Plots of individual measurements and regression lines for the pooled data. Open circles, coastal sites + East Fleet; filled circles, other lagoonal sites.

are plotted against each other (Fig. 6a). The shells from East Fleet, however, fall into the "coastal" group and indeed there is considerable overlap between them and those from Golam (coastal) and Wells. The coastal shells separate out along the CAN2 and CAN3 axes in the order East Fleet, coastal Golam, Wells and Robin Hood's Bay. The Robin Hood's Bay shells separate out almost completely from the above three samples; the box surrounding the cluster of 50 shells from this site contains only one other shell (from Wells) (Fig. 6a). Amongst the lagoonal shells, the only group that could be separated by rotation of the axes was that from Golam (lagoonal), along the CAN2 and CAN3 axes (Fig. 6b); the box surrounding the cluster of 54 shells contains only one other shell (from Alderton).

Finally a Discriminant Analysis was carried out on the size-adjusted data (Table 3). As expected from the above, the most consistent samples were those from Robin Hood's Bay and Golam (lagoonal), where 98% and 94% of the shells respectively were self classifying. Except for East Fleet, no misclassifications occurred between lagoonal and coastal shells. The shells from Cley showed the greatest degree of misclassification, with 20% classifying to Alderton, 12% to Holkham and 8% to Snettisham. Snettisham shells also had a high proportion of misclassifications, with 19%

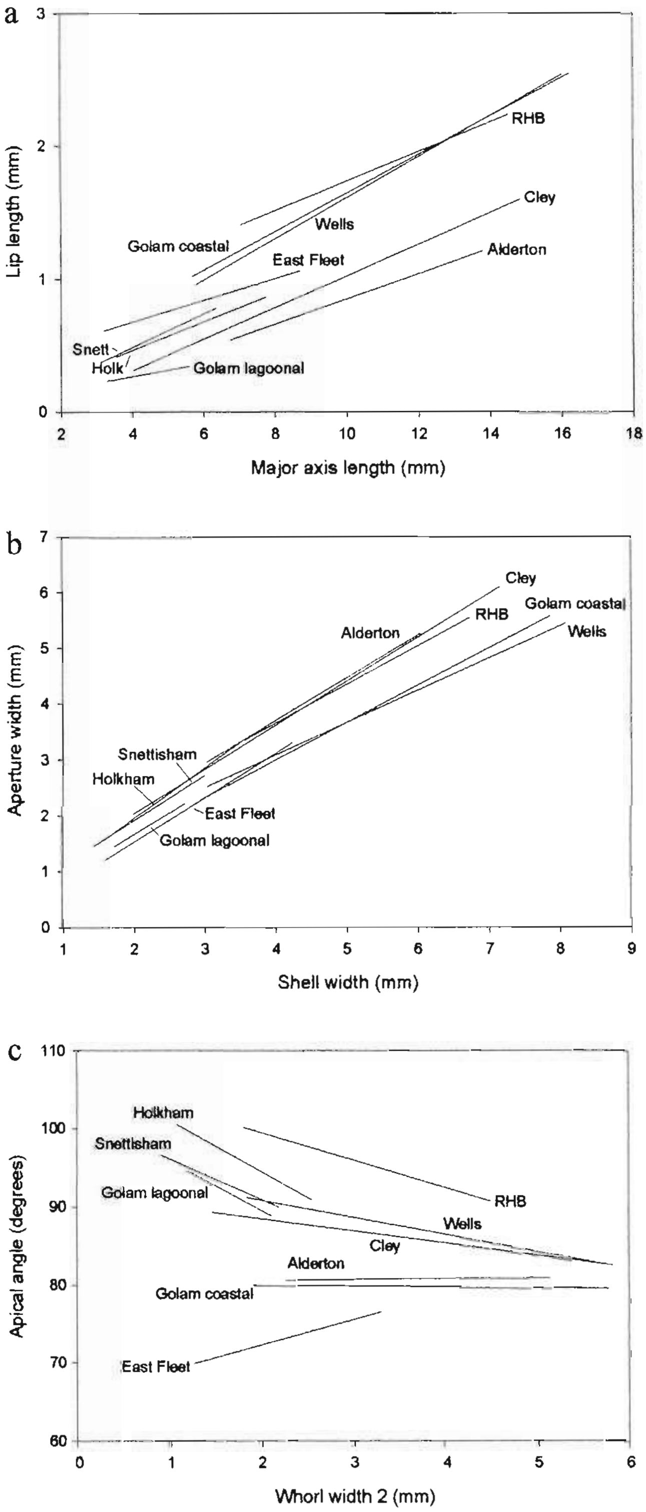

Figure 5. (a) Lip length (mm) plotted against major axis length (mm) with regression lines for each of the nine populations; Holk, Holkham; RHB, Robin Hood's Bay; Snett, Snettisham. (b) Aperture width (mm) plotted against shell width (mm) with regression lines for each of the nine populations; RHB, Robin Hood's Bay. (c) Apical angle (degrees) plotted against whorl width 2 (mm) with regression lines for each of the nine populations; RHB, Robin Hood=s Bay.

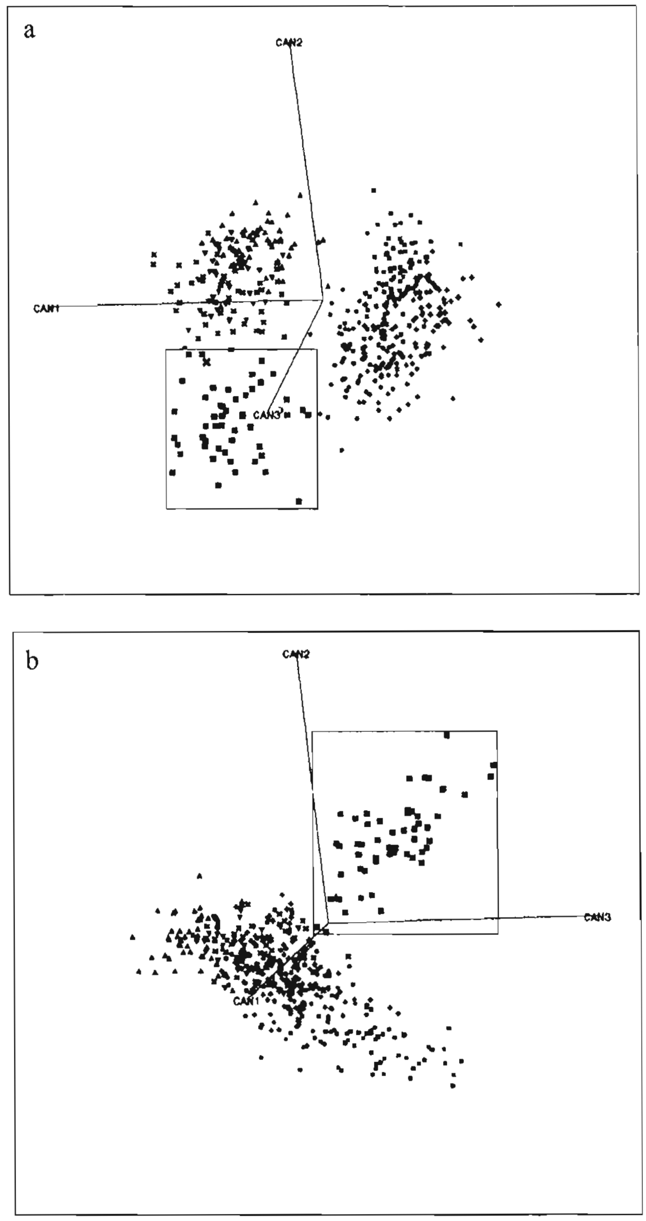

Figure 6. Plot of the data from the nine sites on Canonical Variables 1 (CAN1), 2 (CAN2) and 3 (CAN3). Canonical Variable axes at the mid point. +, Alderton; *, Cley; ▲, East Fleet; ▼, Golam coastal; ♦, Holkham; ■, Robin Hood's Bay and Golam lagoonal; ●, Snettisham; ✖, Wells. (a) The shells on the left are the coastal + East Fleet, and are separated from the lagoonal shells along the CAN1 axis. The square encloses the 50 Robin Hood's Bay shells plus one from Wells. (b) The square encloses the 54 lagoonal Golam shells plus one from Alderton.

classifying to Holkham, 9% to Cley and 3% to each of Alderton and East Fleet. Amongst the coastal samples, the highest proportion of misclassifications were from the coastal Golam shells, with 24% misclassifying to Wells and 11% to East Fleet. Twelve percent of Wells shells also misclassified to East Fleet. Of the East Fleet shells, 11% misclassified to coastal Golam and 2% to Cley. In most cases, using the raw data (i.e. not excluding size) reduced the proportion of misclassifications.

DISCUSSION

The *Littorina saxatilis* complex includes a wide variety of shell morphs and the taxon is thought to be undergoing differentiation

TABLE 3.

Discriminant analysis of the data after removing the effects of size by standardizing using the geometric mean.

From	Alderton	Cley	East Fleet	Golam (lagoon)	Golam (coastal)	Holkham	RHB	Snettisham	Wells	Error%	N
Alderton	89	9	0	0	0	0	0	2	0	11	53
Cley	20	60	0	0	0	12	0	8	0	40	50
East Fleet	0	2	87	0	11	0	0	0	0	13	53
Golam (lagoon)	0	4	0	94	0	2	0	0	0	6	54
Golam (coastal)	0	0	11	0	66	0	0	0	24	34	38
Holkham	0	7	0	0	0	83	0	10	0	17	72
RHB	0	0	0	0	0	0	98	0	2	2	50
Snettisham	3	9	3	0	0	19	0	66	0	34	32
Wells	0	0	8	0	12	0	0	0	80	20	50
Total											452

Values are percentages; shading indicates self classification.

(Fretter 1980, Ward & Warwick 1980) which might have arrived at, or in the future reach, species status for one or more of the morphs. This process is thought to be aided by direct development (Van Marion 1981; Janson 1982; Grahame & Mill 1989) and hence poor dispersal ability (Ward & Warwick 1980; Janson 1983; Janson & Ward 1984; Faller-Fritsch & Emson 1985). However, *L. saxatilis* is a rapid colonizer of offshore islands (Johannesson & Johannesson 1995). Furthermore, two other, closely related taxa, *L. arcana* and *L. compressa*, both show comparatively little variation in shell morphology and yet are direct developers. The main reproductive difference between *L. saxatilis* on the one hand and the other two species on the other, is that the former is ovoviviparous whereas the latter are oviparous.

It is clear from the data that the shells in this study can be separated into coastal + the Fleet and lagoonal, and CAN1 provides an axis for this separation. Lagoonal animals have a lighter shell than correspondingly sized coastal animals; also lagoonal shells lack the jugosity found in coastal populations. The position of the Fleet animals is not surprising as this lagoon is tidal and the sample was taken within a few hundred meters of the lagoon entrance. Of the other lagoonal animals, those from Golam (lagoonal) and Holkham satisfy the strict criteria of Muus (1967) for *Littorina tenebrosa*. However, although the shells from Golam (lagoonal) are clearly separable from those of other lagoonal samples, those from Holkham are not. This is somewhat surprising in view of the subjective impression of the shells (Fig. 3). However, this may be due to the (apparent) intermediate shape of the shells from Snettisham between those from Alderton and Cley on the one hand and those from Holkham on the other.

Barnes (1993) has concluded that there is currently insufficient evidence to accept a species status for *L. tenebrosa* (*sensu* Muus 1967) but that it is clearly distinguishable both in shell characteristics and habitat from *L. saxatilis s.s.* He suggested the varietal name *L. saxatilis* var. *lagunae* for the former.

Although, in the current study, the shells from Golam (lagoonal) separate out from those that came from other lagoons, there appears to be a gradation in shape, size and habitat in the lagoonal populations. It would not be surprising if we are witnessing different degrees of divergence in different lagoonal populations. This might be related to the age of the lagoon and hence to the period of their separation from coastal animals. Only those from Golam and Holkham fulfil all of the criteria for the status of *L. saxatilis* var. *lagunae* but others may have changed partially along this route, particularly those from Snettisham. At Snettisham the animals were found apparently permanently submerged but occurred on the substrate (rocks) rather than on macrophytes. At Cley they were on both the substrate and on floating mats of *Chaetomorpha*, and at Alderton were found on the substrate around the edge of the lagoon. Furthermore, in the last two sites, the animals were not permanently submerged and they reached a size similar to that achieved by the coastal animals. The gradation is reflected in the Canonical Discriminant Analysis, where the two taxa do not separate on the CAN2 axis; indeed the Holkham and Golam (lagoonal) shells fall into different groups when aperture width is plotted against shell width. Similarly, they do not separate on the CAN3 axis. Thus, when apical angle is plotted against whorl width 2, the Snettisham shells align with those from Holkham and Golam (lagoonal), but those from Cley and Alderton are more similar to the Wells and coastal Golam shells respectively.

The current view of speciation is generally that of Mayr (1942) in which a geographical barrier develops between populations, isolating them reproductively from each other. Following this, divergence occurs between the populations, even if the separated habitats are identical, and separate species ultimately evolve. However, there is another possible route for speciation, ecological speciation. Although the idea is not new, it has been brought into focus recently that ecological barriers rather than geographical ones may also be important for speciation (Morell 1999). Thus, ecological pressures could favor changes that eventually cause populations to become reproductively isolated in the absence of geographical barriers. It may be expected that populations that are ecologically separated but genetically similar to each other would be more likely to interbreed than comparable ones that have been separated geographically. However, this is not necessarily the case and size differences between genetically similar populations may be sufficient to produce reproductive isolation (Morell 1999).

It is entirely possible that this is the case with *L saxatilis* var. *lagunae* and *L. saxatilis s.s.*, since the former has clearly developed sexual maturity at a size much smaller than occurs in the latter. Indeed Barnes (1993) has suggested that *L. saxatilis* var.

lagunae may have a paedomorphic origin, as Raffaelli (1979) suggested for another taxon within the *L. saxatilis* complex, i.e. *L. neglecta*, and that, in the case of the former, small size is a requirement of living on submerged macrophytes such as *Chaetomorpha*. It follows from the above that *L. saxatilis s.s.* should interbreed with North American *L. saxatilis* but not with *L. saxatilis* var. *lagunae* occurring in the same lagoon. However, caution is required until the appropriate breeding experiments have been attempted. Furthermore, if, as seems to be the case, parallel evolution is occurring in two or more lagoons, and if the individuals in these populations can interbreed with each other but not with adjacent *L. saxatilis s.s.*, then it follows that any resulting "species" will have a polyphyletic origin.

In only one lagoon (Golam) were both *L. saxatilis* var. *lagunae* and *L. saxatilis s.s.* recorded and they were separated by some 100 m, the former occurring at the landward end of the lagoon, the latter adjacent to the rocky shore. It seems highly likely that the two populations are isolated reproductively (Wilson et al. 1999).

From a conservation point of view it is irrelevant as to whether or not we are dealing with two separate species or two morphs. It could be argued that, if priorities have to be decided, for example because of costs, it is more important to conserve at the species level. However, maximum biodiversity must be preserved so that evolutionary processes are allowed to continue. Brackish lagoons are a nationally rare habitat and have been accorded a "priority habitat type" under Annex 1 of the EU Habitats and Species Directive (Bamber 1998). It may be that many lagoons are very teneral, lasting only tens, or at best hundreds, of years (Bamber 1998), but others may be sufficiently permanent to allow complete separation of species to occur. Hence, it is vital that these lagoonal habitats be conserved.

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LITERATURE CITED

Bamber 1998. Conservation of brackish lagoonal faunas. *J. Conch. Special Publication* 2:265–270.

Barnes, R.S.K. 1987. A survey of the coastal lagoons of North Wales (R. Clwyd - Aberystwyth). September 1987. Nature Conservancy Council Report CSD 1123. Peterborough: Nature Conservancy Council.

Barnes, R. S. K. 1993. On the nature of the coastal lagoon winkles attributed to *Littorina tenebrosa* and *Littorina saxatilis*. *Cah. Biol. Mar* 34:477–495.

Faller-Fritsch, R. J. & R. H. Emson. 1985. Causes and patterns of mortality in *Littorina rudis* (Maton) in relation to intraspecific variation: a review. In: P.G. Moore & R. Seed, editors. The Ecology of Rocky Coasts. London: Hodder & Stoughton. pp. 157–177.

Fretter, V. 1980. Observations on the gross anatomy of the female genital duct of British *Littorina* spp. *J. Moll. Stud.* 46:148–153.

Gosling, E. M., I. F. Wilson & J. Andrews. 1998. A preliminary study on genetic differentiation in *Littorina saxatilis* from Galway Bay, Ireland: *Littorina tenebrosa* Montagu - a valid species or ecotype? *Hydrobiologia* 378:21–25.

Grahame, J. & P.J. Mill 1989. Shell shape variation in *Littorina saxatilis* and *L. arcana*: a case of character displacement? *J. Mar. Biol. Ass. U.K.* 69:837–855.

Grahame, J., P. J. Mill, S. Hull & K. J. Caley. 1995. *Littorina neglecta* Bean: ecotype or species? *J. Nat. Hist.* 29:887–899.

Hull, S., J. Grahame & P. J. Mill. 1996. Morphological divergence and evidence for reproductive isolation in *Littorina saxatilis* (Olivi) in Northeast England. *J. Moll. Stud.* 62:89–99.

Hull, S. L., J. Grahame & P. J. Mill. 1999. Heat stability and activity levels of aspartate aminotransferase and alanine aminotransferase in British Littorinidae. *J. Exp. Mar. Biol. Ecol.* 237:255–270.

Janson, K. 1982. Phenotypic differentiation in *Littorina saxatilis* Olivi (Mollusca, Prosobranchia) in a small area on the Swedish west coast. *J. Moll. Stud.* 48:167–173.

Janson, K. 1983. Selection and migration in two distinct phenotypes of *Littorina saxatilis* in Sweden. *Oecologia. Berl.* 59:58–61.

Janson, K. & R. D. Ward. 1984. Microgeographic variation in allozyme and shell characters in *Littorina saxatilis* Olivi (Prosobranchia: Littorinidae). *Biol. J. Linn. Soc. U.K.* 22:289–307.

Johannesson, K. & B. Johannesson. 1995. Dispersal and population expansion in a direct developing marine snail (*Littorina saxatilis*) following a severe population bottleneck. *Hydrobiologia.* 309:173–180.

Morell, V. 1999. Ecology returns to speciation studies. *Science* 284:2106–2108.

Muus, B.J. 1967. The fauna of Danish estuaries and lagoons. *Medd. Danm. Fisk.-og Havunders (Ny Ser.)* 5:1–316.

Raffaelli, D. 1979. The taxonomy of the *Littorina saxatilis* species-complex, with particular reference to the systematic status of *Littorina patula* Jeffreys. *Zool. J. Linn. Soc. U.K.* 65:219–232.

Reid, D. G. 1996. Systematics and Evolution of Littorina. Andover, England: The Ray Society. 463 pp.

Seaward, D. R. 1980. The marine molluscs of the Fleet, Dorset. *Proc. Dorset Nat. Hist. Archaeol. Soc.* 100:100–108.

Smith, S. M. 1982. A review of the genus *Littorina* in British and Atlantic waters (Gastropoda: Prosobranchia). *Malacologia* 22:535–539.

Van Marion, P. 1981. Intrapopulation variation of the shell of *Littorina rudis* (Maton) (Mollusca: Prosobranchia). *J. Moll. Stud.* 47:99–107.

Ward, R. D. & T. Warwick. 1980. Genetic differentiation in the molluscan species *Littorina rudis* and *Littorina arcana* (Prosobranchia: Littorinidae). *Biol. J. Linn. Soc. U.K.* 14:417–428.

Wilson, I.F., E.M. Gosling & W. Tapper 1999. The systematic status of the lagoon periwinkle, *Littorina tenebrosa*. *J. Mar. Biol. Ass. U.K.* 79:653–660.