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Breaking down the mussel (*Mytilus edulis*) shell: which layers affect oystercatchers' (*Haematopus ostralegus*) prey selection?

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1   **ABSTRACT**

2   Predators are able to identify fine characteristic features of prey and use them to

3   maximize the profitability of foraging. Oystercatchers *Haematopus ostralegus* select thin-

4   shelled mussels *Mytilus edulis* to hammer through because they are easier to crack than

5   thick-shelled mussels. But mussel shells are composite structures, so we need to ask what

6   it is about these thin-shelled mussels that make them vulnerable. Here we show that the

7   mussels damaged by Oystercatchers were mainly distinguished by having a significantly

8   thinner prismatic layer than undamaged mussels. Regression analysis indicated that the

9   Oystercatchers' shell selection was independently influenced by the thickness of the

10   prismatic and nacreous layers, but the coefficient for the thickness of the prismatic layer

11   was almost one and half times that for the nacreous layer. Thus the thickness of the

12   prismatic layer largely determines the vulnerability of the mussel shells. Oystercatchers

13   were more likely to attack mussels by the right valve than the left, and this tendency was

14   accentuated in larger mussels and those with a thicker nacreous layer.

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16   *Key words*: Anti-predation; Foraging behaviour; *Haematopus ostralegus*; Mussel shell

17   layer; Oystercatcher; Prey selection

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1. Introduction

20   Rate maximising foraging theory states that predators maximise the profitability

21   of foraging by making numerous decisions such as where and when to search for prey

22   and which prey to select (Sih 1993, Sutherland *et al*. 1996). Predators are able to select

23   the more vulnerable and most profitable prey on the basis of their morphological and

1 behavioural characteristics. Hence predators' selective behaviour effectively ranks prey

2 accordingly to their profitability. On the other hand, prey develop many behavioural and

3 morphological adaptations that tend to resist predation pressure.

4   Oystercatchers are extreme specialists in their foraging behaviour, preying on

5 hard-shelled molluscan organisms (Goss-Custard 1996; Nagarajan et al. 2002a,b,c).

6 They use different techniques to prey on various species of molluscs, but within a species

7 always select relatively thin shelled prey, within length classes as well as overall

8 (Hulscher 1996; Sutherland and Ens 1987; Sutherland et al. 1996; Nagarajan et al. 2002b;

9 Zwarts et al. 1996). Mussel feeding, ventral hammering Oystercatchers selectively attack

10 ventrally thin shells, whereas dorsally hammering Oystercatchers selectively attack

11 dorsally thin shells (Durell and Goss-Custard 1984). They also attack the weaker valve

12 of a given mussel (Nagarajan et al. 2002c). This selectivity presumably occurs because

13 thinner shells require less effort to break, as has been confirmed by laboratory

14 experiments with model Oystercatcher bills (Meire 1996; Nagarajan et al. 2006).

15   However, this previous work has ignored the fact that mollusc shells are

16 composite structures. In order to understand the selection pressures that the risk of bird

17 attack imposes on mussels, we need to know whether there is one component of their

18 shells whose strength is particularly important in resisting such attack. Mussel shells are

19 made up of three layers, the outer organic periostracum, a calcified prismatic layer, and

20 an inner calcareous nacreous layer (Chen et al. 2004). The layers have different

21 microstructures (Zuschin et al. 2003). The strength and toughness of the shell are

22 determined by the thickness of the individual layers and their microstructure (Currey

23 1988). The periostracum is made of sclerotized proteins (Watabe 1988). The prismatic

1   layer is composed of calcite prisms surrounded by a conchiolin matrix, with the prisms

2   oriented at an angle to the surface. The inner, nacreous, layer consists of flattened tablets

3   of aragonite enveloped in conchiolin, placed horizontally to the surface (Harris 1990):

4   this is the oldest and mechanically strongest layer (Taylor and Layman 1972; Currey and

5   Taylor 1974; Currey 1977, 1988; Evans et al. 2001; Chen et al. 2004).

6   To further elucidate the ecological and behavioural significance of thin-shelled

7   mussel selection by Oystercatchers, therefore, we investigated the difference in the

8   thickness of the different shell layers between undamaged mussels and mussels that had

9   been damaged in the way that is characteristic of Oystercatcher attack. In common with

10   much recent research (e.g. Nagarajan et al., 2002a, 2002b), the present paper focuses on

11   ventrally damaged mussels. The aim was to determine the relative importance of the

12   different layers in defending the mussels from bird attack.

13

2. Materials and methods

15   *2.1. Study area*

16   Mussels were collected during November and December 2004, and February 2005, from

17   a site on the western side of the Exe estuary, southwest England, near Cockwood Harbour

18   (mussel bed number 3 of McGrorty and Goss-Custard 1993, 50°37'N, 03°27'W).

19   During winter this bed has a substantial population of Oystercatchers, which feed on the

20   mussels.

21   *2.2. Mussel collection and measurements*

22   Mussels that had recently been damaged by Oystercatchers were identified by their

23   broken shells and traces of adductor muscle inside the shell. Only ventrally damaged

1   shells were collected. For every freshly damaged shell, one or two undamaged mussels

2   of similar length were collected from a location as near as possible (Nagarajan et al.

3   2002b), if any could be found. In total 107 ventrally damaged and 115 undamaged

4   mussels were collected during November and December 2004 and 135 ventrally damaged

5   and 121 undamaged mussels were collected during February 2005. Before measurement,

6   the shells were scrubbed to remove mud and epi-biotic growth, and the undamaged

7   mussels were placed in warm water for 5 minutes to sterilize the sample and kill the

8   animals in the shells. The flesh was then removed by gentle scraping, and the shells were

9   left to dry at room temperature before the valves were individually marked. The length

10   of the mussel was measured using vernier callipers that were accurate to 0.05 mm. The

11   left and right valves of the mussel were identified using the methods of Nagarajan et al.

12   (2002c).

13   The valves were glued in rows to fibreboard sheets using a hot-glue gun. They

14   were arranged so that the long dimension of the valve was perpendicular to one edge of

15   the sheet, with the midpoints of the long dimension of all mussels lying along a single

16   line. A band saw with a metal-cutting blade was used to cut along this line. Another,

17   parallel, cut was made to one side of this line, to produce strips of board bearing shell

18   cross-sections approximately 5 mm wide, for convenient viewing in a microscope. To

19   prepare the cross-sections for viewing under the microscope, a thin coat of aerosol

20   lacquer was applied and an identifying mark was made with permanent marker at the

21   ventral margin of each section.

22   The cross-sections were top-illuminated and observed under a stereoscopic

23   dissection microscope at 10x magnification (Fig. 1). The constituent shell layers were

1 distinguished by colour. For each valve, a single measurement of layer thickness was

2 taken at the midpoint ventral location using the microscope graticule, calibrated by

3 measuring a slide showing 0.01 mm divisions. For the damaged mussels, the thickness

4 measurements were collected on the undamaged valves only, since in many cases the

5 damage affected the regions that were to be measured. Measurements were taken at the

6 midpoint of the long dimension. Layer thicknesses were recorded to the nearest 0.01

7 mm.

8 *2.3. Statistical methods*

9 The distribution of damage between valves was examined using a 1-sample χ^2 test.

10 Statistical analyses of the thickness measurements were performed using Minitab and

11 SPSS statistical packages. Mussels were included in the analysis if they had no signs of

12 attack, and the ventral thickness of both valves could be measured (undamaged mussels:

13 261 cases), or if they had been ventrally damaged on one and only one valve and the flesh

14 had been removed (damaged mussels: 230 cases). Correlations between the thicknesses

15 of layers were examined using Pearson coefficients, so that the effects of length class

16 (with which thickness would be expected to be correlated) could be partialled out.

17 The sources of differences in layer thicknesses were investigated using three

18 separate factorial repeated-measures analyses of variance, using General Linear Model

19 (GLM) procedures. In all models, mussels were taken as the unit of analysis. The first

20 analysis of variance involved all mussels, and tested the effects of Layer (periostracum,

21 prismatic and nacreous) as a within-mussel factor, and Collection date

22 (November/December vs. February), Length class (40-50 mm, 50-60 mm or 60-70 mm)

23 and Damage condition of the mussel (undamaged vs. damaged) as between-mussel

1 factors. For this analysis, thicknesses of layers for the undamaged mussels were taken as

2 the mean of values from left and right valves. Since this involved a difference in

3 treatment between the two levels of the Damage condition factor, supplementary GLM

4 analyses were carried out comparing the intact left valves of right-damaged mussels with

5 the left valves of undamaged mussels, and the intact right valves of left-damaged mussels

6 with the right valves of undamaged mussels, to check whether any distortion had been

7 introduced. The second analysis of variance involved damaged mussels only, using as

8 factors Layer, Collection date and Length class as before; in addition, Side of damage

9 (left or right) was used as a between-mussel factor. The third analysis of variance

10 involved undamaged mussels only, using as factors Layer, Collection date and Length

11 class as before, and in addition Valve (left or right) as a second within-mussel factor.

12 Since Mauchly's test indicated non-sphericity in some cases, lower-bound levels were

13 used for testing the significance of all within-mussel effects. Where post hoc

14 comparisons between multiple factor levels were made, Bonferroni's correction to

15 significance levels was used.

16

17   Binary logistic regression was used to investigate the sources of Oystercatchers

18 selection of mussels to attack, and their selection of valves to attack within a mussel.

19 These analyses also allowed us to see whether the three shell layer thicknesses

20 independently affected Oystercatchers' mussel and valve selection, and to predict the

21 threshold prismatic layer thickness of vulnerable mussels. The ventral thicknesses of

22 periostracum, prismatic and nacreous layers were used as predictor variables in the

23 regression models. Interaction terms (mean-referenced) between Length class and each

1   of the three layer thicknesses were also included. Full models were computed, followed

2   by a forward stepwise procedure to identify a smaller set of independent variables that

3   would predict the dependent variables efficiently. The appropriateness of assumptions

4   such as linearity, homoscedasticity and the absence of multicollinearity were strictly

5   checked when developing the models (Nagarajan *et al*. 2002). Two separate regression

6   models were developed. The first used data from all mussels, and aimed to predict

7   whether they would be damaged or undamaged; in the case of undamaged mussels,

8   means from the two valves were taken to avoid spurious inflation of the degrees of

9   freedom in the regression, while in the case of damaged mussels, measurements from the

10   undamaged sides were used. The second used only damaged mussels, and aimed to

11   predict whether they would be damaged on the left or the right valve; in this case,

12   thickness measurements from the undamaged valve were used.

3. Results

3.1. Distribution of damage between valves

In the majority of the damaged mussels, the damage affected the right valve (79% of

those with unilateral damage), significantly more than would be expected by chance;

$\chi^2_1=60.40$, *P*<0.001).

3.2. Descriptive statistics for thickness of the layers

In the undamaged mussels, the periostracum was the thinnest layer (mean in the

undamaged mussels 0.038mm, s.d. 0.020), and the prismatic the thickest (mean 0.75mm,

s.d. 0.29); the mean thickness of the nacreous layer was 0.37mm, s.d. 0.22. Fig. 2 shows

that these differences existed also in damaged mussels, and in all length classes, although

the prismatic and nacreous layers were both thicker in larger mussels than smaller ones.

1 It also shows that among the undamaged class, larger mussels were thicker than smaller

2 mussels, in the prismatic and nacreous layers but not in the periostracum, and that

3 damaged mussels tended to be thinner than undamaged ones, again in the two inner layers

4 only. The nacreous layer was absent at the ventral measurement point in one or other

5 valve in 11% of undamaged mussels; these cases were included (with a value of zero) in

6 the computations that follow since there seemed to be no grounds for excluding them and

7 such mussels would presumably be relatively vulnerable to attack. The thicknesses of the

8 different layers of a given valve were correlated, but most of the correlations were

9 negative. With length class partialled out, in the undamaged class the thickness of the

10 inner (nacreous) layer was significantly negatively correlated with the thickness of the

11 outer (periostracum) layers (Pearson coefficient -0.20, P<0.01) and the middle (prismatic)

12 layer (-0.16, P<0.05). In the damaged class, the corresponding correlations were -0.44

13 and -0.21 (P<0.01 in both cases); however the middle and outer layer thicknesses were

14 had a positive correlation of 0.20 (P<0.01).

15 *3.3. Significance of differences in thickness between the layers*

16   In the GLM analysis of all mussels using Layer, Collection date, Length class and

17 Damage condition as factors, the main effects of Layer, Damage condition and Length

18 class were significant ($F_{1,463} = 686.39$, P<0.001; $F_{1,463} = 50.47$, P<0.001; $F_{2,463} = 4.01$,

19 P<0.05, respectively). The interaction of Layer with Damage condition was significant

20 ($F_{1,463} = 18.51$, P<0.001); the mean difference between damaged and undamaged mussels

21 was 0.00mm in the periostracum, 0.20mm in the prismatic layer and 0.04mm in the

22 nacreous layer. Post hoc tests showed that the difference was significant only in the

23 prismatic layer (t_{463} values of 0.37, 6.37, and 1.83, P =.71, P<.001 and P=.07 for the

1 periostracum, prismatic and nacreous layers respectively). The interaction of Length class

2 with Damage condition was also significant ($F_{1,463} = 4.97$, $P<0.01$). The mean total

3 thickness of undamaged mussels varied from 1.06mm in the 40-50mm length class to

4 1.28mm in the 60-70mm class, with the thickness of the 60-70mm class being

5 significantly greater than that of either of the other two ($t_{463} = 3.22$ for 40-50mm vs. 60-

6 70mm, $P<0.01$ and $t_{463} = 4.15$, $P<0.001$ for 50-60mm vs. 60-70mm classes). In the

7 damaged mussels, however, mean thickness was approximately 0.91mm in all size

8 classes, with no significant differences. There were no significant interactions involving

9 Collection date, nor was its main effect significant. As noted above, for this analysis, the

10 values used for layer thicknesses for undamaged mussels were the means from the two

11 valves, whereas those for damaged mussels were from the single undamaged valves; the

12 supplementary analyses in which single valves from undamaged mussels were used in the

13 comparison gave qualitatively similar results.

14 In the GLM analysis of damaged mussels only, using Layer, Collection date,

15 Length class and Side of damage as factors, the only significant effect was that of Layer

16 ($F_{1,202} = 155.04$, $P<0.001$). In the GLM analysis of undamaged mussels only, using

17 Layer, Length class, and Collection date as factors, the main effects of Layer and Length

18 class were significant ($F_{1,255}= 599.98$, $P<0.001$; $F_{2,255} = 11.06$, $P<0.001$); the main effect

19 of Collection date was not. There were also significant interactions between Layer and

20 Length class ($F_{2,255} = 3.16$, $P<0.05$), Layer and Valve ($F_{1,255} = 11.20$, $P=0.001$), and

21 Layer, Valve and Collection date ($F_{1,255} = 5.00$, $P<0.05$). The Layer and Length class

22 main effects were qualitatively the same as in previous analyses. The form of their

23 interaction can be seen in Fig. 2. The periostracum was much the same thickness in all

1 sizes of mussel. The prismatic layer was thicker in the largest size class, and the

2 difference between 50-60mm and 60-70mm size classes was significant on a post hoc test

3 (t_{255}=3.11, P<0.01). The nacreous layer was thicker in each successive size class, and the

4 difference between both the 40-50mm and 50-60mm size classes and the 60-70mm size

5 classes was significant on a post hoc test (t_{255} values of 3.13 and 3.22, P<0.01 in each

6 case).   The interaction of Layer and Valve took the form of left valves having thinner

7 periostracal and prismatic layers, but thicker nacreous layers, than right valves; these

8 differences were all significant on post hoc tests (t_{255} values of 3.92, 3.12, and 3.09,

9 P<.001, .01 and .01). This effect was substantially more marked in the February sample

10 than in the November/December sample, as reflected in the significant three-way Layer

11 by Valve by Collection date interaction. These effects can also be seen in Fig. 2.

12 *3.4 Contributions of the layers to mussel selection*

13   Table 1 provides the results of the logistic regression analyses. The analysis of

14 mussel selection (for which the dependent variable was Damage condition) showed

15 significant overall prediction by the independent variables when they were all included.

16 Damaged mussels had significantly thinner prismatic and nacreous layers than

17 undamaged mussels, and the regression coefficient for the thickness of the prismatic layer

18 was almost one and half times greater than that for the nacreous layer. The same was true

19 if the interaction terms were excluded. Neither length nor season had significant effects.

20 The interaction between the thickness of the inner layer and length class had a significant

21 effect, in the direction of the inner layer thickness having more effect on mussel selection

22 in the larger mussels. The results were not substantially changed when the stepwise

23 procedure was used to eliminate non-significant regressors from the model.

1   The analysis of Side of damage did not show significant prediction overall if all

2   variables were included, and the only individually significant effect found was due to

3   Length class, with the tendency for right rather than left valves to be damaged

4   accentuated in larger mussels. However, when the stepwise entry procedure was used to

5   reduce the model, the only effect identified as significant was that a thicker nacreous

6   layer accentuated the tendency for mussels to be damaged on the right valve.

7

8   4. Discussion

9   *4.1. Why do Oystercatchers select mussels with a thin prismatic layer?*

10   The present data show that Oystercatchers are independently influenced in their selection

11   of thin-shelled mussels by the thickness of the prismatic and nacreous layers of the shell,

12   but most strongly by the thickness of the prismatic layer (see Fig 2 and Table 1). It thus

13   gives a more detailed understanding of the well-established tendency for the mussel

14   shells damaged by Oystercatchers to be thinner than control, undamaged shells (e.g.

15   Sutherland et al. 1996, Nagarajan et al. 2002a,b,c). The difference between damaged and

16   undamaged mussels was greatest in the prismatic layer. The difference in nacreous layers

17   fell short of significance in the analysis of variance, but was found be significant with the

18   slightly different approach of the logistic regression analysis. However it is clearly a

19   lesser effect: in the logistic regression analysis of mussel selection, the coefficient or the

20   prismatic layer's thickness was almost one and half times that for the nacreous layer

21   (Table 1).

22   Presumably the Oystercatchers' selection reflects the resistance of the shells to

23   fracture, which is an adaptation to physical stress (Zuschin et al. 2003). Resistance to

1 fracture is a function of both strength (maximum compressive force required to break an

2 object) and toughness (resistance to propagation of cracks). These properties are in turn

3 determined by the organic contents, microstructure, patterns of arrangement, and

4 adhesive matrix of the shell (Currey 1988; Kamat et al. 2000; Zuschin et al. 2003; Chen

5 et al. 2004). The prismatic and nacreous layers both comprise lamellar structures of

6 calcified materials supported by a glycoprotein matrix, which together provide the

7 strength and toughness to the shell (Watabe 1988).

8   As noted above, the nacreous is the oldest and mechanically strongest layer. The

9 propagation of cracks from the prismatic layer into the nacreous layer is resisted because

10 of its brick wall structure and associated nanometric-scale arrangement (Currey 1988).

11 The relatively important role of the prismatic layer may result from its specialized micro-

12 structural properties. It has a crossed lamellar structure which has the highest nominal

13 fracture toughness (Feng et al. 2000; Chen et al. 2004) and requires additional energy for

14 crack propagation (Kamat et al. 2000). Such a structure retards and diverts cracks

15 (Currey 1988): they travel immediately to the junction of the middle and inner layers but

16 invade the inner layer with difficulty. Furthermore when there is a difficulty in the travel

17 path of a crack, it tends to split into two roughly orthogonal directions, reducing its

18 energy (Currey 1988). Therefore, the thicker prismatic layer allows non-catastrophic

19 failures of a shell by acting as a crack tolerant sacrificial layer when a load is placed on

20 the shell. To crack a mussel successfully the nacreous layers of the shell must be

21 damaged, and to allow such damage the prismatic layer must be thin enough to allow the

22 impact of the bird's blows to reach the nacreous layer. We argue that this is why

23 Oystercatchers select mussels with a thin prismatic layer.

1   *4.2. Effect of thin shell selection by Oystercatchers on mussels*

2   Selective predation by Oystercatchers may well affect the thicknesses of the

3   various layers of the shells of undamaged mussel shells remaining available later in the

4   season. Oystercatchers arrive on the Exe estuary in large numbers in early autumn and

5   remain there throughout the winter. In the same study area, Nagarajan (2000) found that

6   Oystercatchers selectively attacked mussels between 40 and 55 mm in length. They

7   selectively consume thinner shelled mussels (Durell and Goss-Custard 1984; Cayford and

8   Goss-Custard 1990; Meire 1996; Nagarajan et al. 2002b); even within thin mussels they

9   are capable of discriminating thinner valves down to a threshold level of 0.036 mm, and

10   thus attack the weaker valve (Nagarajan et al. 2002c). During the winter, mussels neither

11   grow nor reproduce (McGrorty 1997; Nagarajan 2000), and we might expect, therefore,

12   that selective predation on thin-shelled mussels would increase the average thickness of

13   the remaining mussels by February. However, there may be some changes in the mussels

14   that are not due to predation. Nagarajan et al. (2006, 2008) found that the thickness and

15   strength of mussels changes across the winter in relation to the fluctuations in the salinity

16   and temperature, and it may be that these water quality effects are layer-specific, and

17   offset the effects of predator selection to different extents for different layers.

18   The seasonal change that we observed in the undamaged mussel population took

19   the form of a three-way interaction, between Layer, Valve and Collection date. The

20   asymmetry between left and right valves (with left valves having thinner outer and

21   middle layers and thicker inner layers than right valves) was substantially more marked in

22   the February collection than in November/December (see Fig. 2). This effect can

23   plausibly be attributed to a selective effect of the Oystercatchers' foraging behaviour.

1   Nagarajan et al. (2002c) showed that Oystercatchers on the Exe estuary tend to attack

2   mussels by the right valve, and this tendency is higher than can be explained by the

3   proportion of mussels whose right valves are thinner than their left valves. This tendency

4   was strongly confirmed in the present data, and the results reported in Table 1 suggest

5   that it is somewhat accentuated in mussels that are generally stronger, because they are

6   larger or because of a thicker nacreous layer. Because the birds' mussel selection is most

7   influenced by the thickness of the prismatic layer, they will selectively deplete mussels

8   with thin prismatic layers in their right valves. The combined effect of these two

9   tendencies would account for precisely the differences we observed.

10   In the longer term, predation pressure will favour shell repair mechanisms.

11   Trussell and Smith (2000) found that exposure of flat periwinkles *Littorina obtusata* to

12   shore crab *Carcinus maenas* effluent can induce an 8-47% increase in shell thickness in

13   just 90 days. Lowell et al. (1994) found that the strength of *L. obtusata* shells increased

14   faster than could be accounted for either overall shell mass or thickness at the point of

15   force application, and they suggested that the strengthening could be due to other factors

16   such as changes in the shell microstructure or shape. Although neither of these studies

17   looked at the relation between the shell layers and predation pressure, we suspect that

18   mussels could give priority to the growth and repair of prismatic layer over other layers,

19   both because the prismatic layer is cheaper and quicker to produce than the other layers,

20   and it plays a dominant role in resisting cracks and damages.

21   *4.3 How do Oystercatchers detect thin shells?*

22   Previous studies have shown that, in addition to selecting for shell thickness,

23   ventral hammering Oystercatchers attack mussels that have fewer barnacles, are brown in

1   colour, ventrally flat, and are less well attached to the substrate (Nagarajan et al. 2002b;

2   Sitters 2000). However shell thickness remains the prime factor (Durell and Goss-

3   Custard 1984; Cayford and Goss-Custard 1990; Meire 1996; Nagarajan et al. 2002a,

4   2002b). To date, no study has attempted to show whether the selection is active or

5   passive, that is, whether Oystercatchers are able to identify a thin-shelled mussel before

6   they attack it, or simply abandon thicker shelled ones after they have tried,

7   unsuccessfully, to attack them. In the present study site, Oystercatchers select 85% of

8   their mussels from under weed (Nagarajan et al. 2002b), which suggests that they are not

9   using visual cues to select thin shelled mussels. The current paper shows that the

10   Oystercatchers selectively attack mussels that have thinner prismatic and, to a lesser

11   extent, nacreous layers. Since these layers are hidden under the outermost periostracum,

12   the present results make active visual selection of thin mussels even less likely. However

13   the present data make it possible to address this question in a more precise way, by

14   investigating what proportion of the mussels that Oystercatchers start to hammer they

15   subsequently discard, and comparing this with the proportion that are suitable for

16   opening. In a previous, unpublished study on our present study site, we found that about

17   38% of mussels are abandoned after hammering has started. The best-fitting logistic

18   regression equation for Damage condition correctly classified 70% of undamaged

19   mussels on the basis of shell thickness. This means that 70% of mussels had shell

20   characteristics that made them more likely to be found undamaged than damaged, and

21   these mussels can be regarded as relatively unsuitable for attack by Oystercatchers.

22   Because the data come from different studies, no statistical test of the difference of these

23   two proportions (38% and 70%) is possible, but the difference is substantial enough to

suggest that the Oystercatchers may have been able to recognise some unsuitable mussels before trying to open them. However, Oystercatchers do reject some mussels after a few test blows, presumably because they detect their thickness by sound or feeling. According to unpublished data of the present authors, when Oystercatchers successfully hammered their way into a mussel, it took them 83.8 ± 1.5 seconds to do so (74.9 ± 2.7 s for the 40-50mm size class, 87.2 ± 2.5 s for the 50-60mm class, and 88.2 ± 2.7 s for the 60-70mm class; all data means $\pm$ SE). However, when they started hammering a mussel and then abandoned it (without obvious disturbance by another bird) they did so after 16.7 ± 0.8 seconds (15.3 ± 1.4 s, 16.4 ± 1.2 s, 14.6 ± 1.4 s for the three length classes in order). These figures are somewhat lower than those of Sitters (2000), established on the same basis from mussel bed 20 of Exe estuary, who found that the average time spent on unsuccessful mussels was 24.4 seconds. However, taken together with our present results, they lead to the same conclusion: the selection of thin-shelled mussels by Oystercatchers is in part an active process, but most rejections occur during rather than before hammering.

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2 Fig. 1. A section of a mussel shell (*Mytulis edulis*) as viewed through a microscope. The

3 cross-sections were top-illuminated and observed under a stereoscopic dissection

4 microscope at 10x magnification. The layers within the shell are identifiable by their

5 shading and location: (a) the outer periostracum, (b) the middle, prismatic layer, and (c)

6 the nacreous layer.
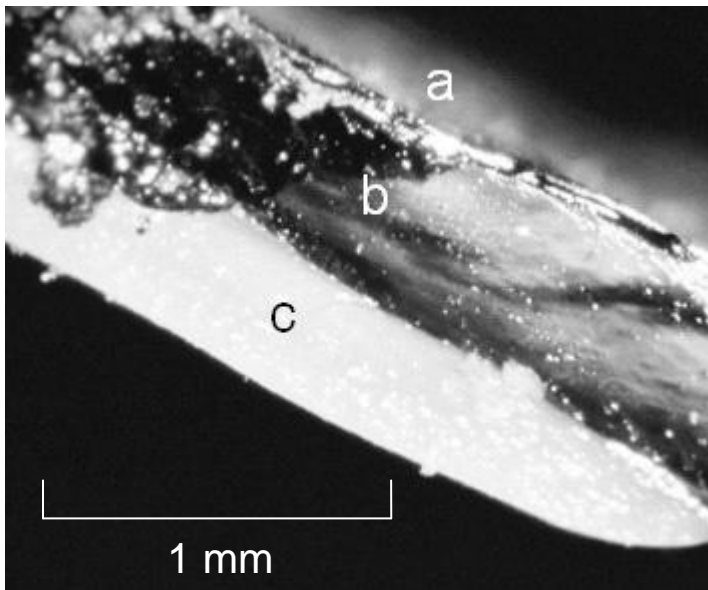
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Fig. 2. Variations in the mean thickness of different layers of mussel shells (*Mytilus edulis*), for shells of different length classes in each of two collection periods. Data for damaged shells are available from the undamaged valve only (so Damaged – Left valve data come from mussels damaged on the right valve), so separate sample sizes apply to each valve. Data for undamaged shells are available from both valves so a single sample size applies to both valves. The horizontal axis shows the length class (mm) and the vertical axis shows mean thickness of shell layers (mm) of mussels, ± standard error; note the different scales used for each layer.
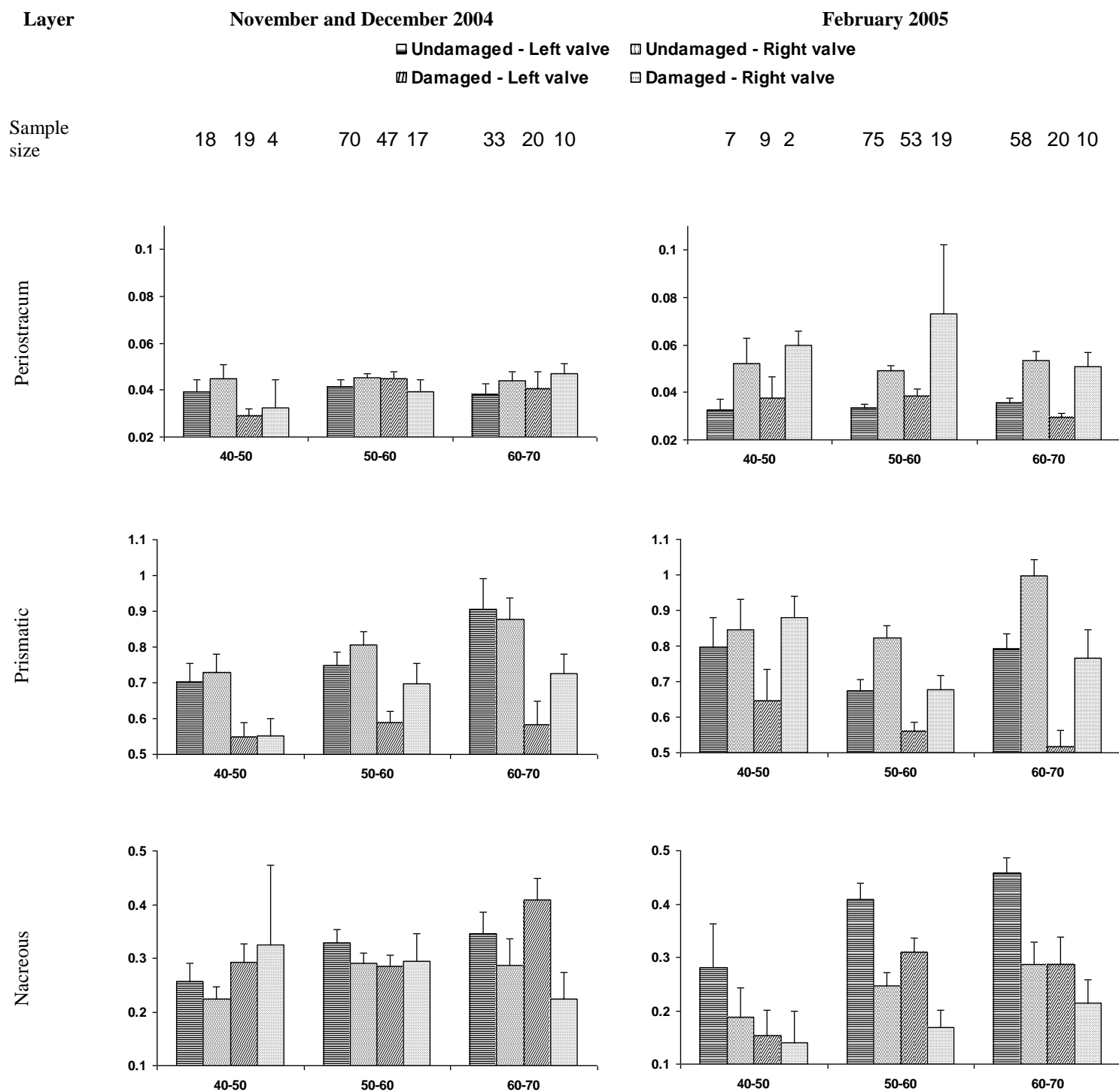
1 Table 1. Binary logistic regression equation models to explore the independent effect of mussel (*Mytilus edulis*)

2 shell layers in predicting damage by Oystercatchers (*Haematopus ostralegus*). Entries are regression coefficients ±

3 standard errors, with Wald statistics in parentheses.

Regressors and model test statistics	Dependent variable			
	Damage condition (Damaged=1, *N*=214; Undamaged=0, *N*=261)		Side of damage in damaged mussels (Left=2, *N*=66; Right=1, *N*=168)	
	Complete model	Stepwise model	Complete model	Stepwise model
Constant	3.33± 0.66 (25.29***)	3.05±0.42 (53.30***)	3.32±1.13 (8.61**)	1.92±0.37 (26.72***)
Periostracum (mm)	-3.56±5.65 (.40)		10.08±11.08 (.83)	
Prismatic layer (mm)	-3.56±0.50 (50.72***)	-3.70±0.48 (58.95***)	-0.33±0.96 (.12)	
Nacreous layer (mm)	-2.39±0.59 (16.59***)	-2.39±0.57 (17.27***)	-1.90±1.10 (2.99)	-1.90±0.96 (3.92*)
Collection Date (Nov=0, Feb=1)	-0.01±0.23 (.003)		-0.14±0.35 (.17)	
Length class	-0.11±0.17 (.36)		-0.64±0.32 (3.98*)	
Periostracum x Length class	9.46±8.28 (1.30)		-12.80±16.20 (.63)	
Prismatic layer x Length class	-1.08±0.81 (1.80)		-1.47±1.52 (.94)	
Nacreous layer x Length class	-2.91±1.07 (7.39**)	-2.59±0.94 (7.61**)	-0.86±1.72 (.25)	
χ^2	102.47***	98.36***	9.27	4.03*
Degrees of freedom	8	3	8	1
Log-likelihood	551.38	555.48	213.48	218.75
Nagelkerke Pseudo-R^2	0.26	0.25	0.07	0.03

4 *$P<0.05$; **$P<0.01$; ***$P<0.001$

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