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ORIGINAL PAPER

Reproductive strategies and energy sources fuelling reproductive growth in a protracted spawner

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Abstract Most marine invertebrates experience variable environments and for broadcast spawners, fertilisation success increases with greater synchronisation of spawning, so a capital breeding strategy is predicted. However, this prediction should be tested for species with protracted breeding seasons, since it is not clear how reproduction is fuelled over several consecutive months of spawning. The simultaneous hermaphrodite scallop *Pecten fumatus* was used to test the hypothesis that protracted spawning is supported by both capital and income strategies, depending on the state of energy reserves and food availability at the time of oocyte maturation. The study was carried out in Great Bay, Tasmania, Australia (147.335W, 43.220S) in 2010/2011. The use of glycogen, protein and lipid in the muscle, gonad, and digestive gland was examined, along with the role of atretic eggs as an alternative energy source for oogenesis and maturation. The reproductive stage of an individual was determined using only the ovaries. *P. fumatus* uses a capital breeding strategy early in the reproductive cycle during winter and spring (August–October) with muscle glycogen and protein and digestive gland lipid providing energy for oogenesis. Given there was no evidence of energy stores being used later in the reproductive cycle in late spring and summer (November–March), when less food was available for direct fuelling of reproduction, it appears that metabolites produced from oocyte lysis may have fuelled oogenesis. Recycling of energy from oocyte resorption must be considered as part of the strategy of energy use to fuel reproduction in marine invertebrates.

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Introduction

Strategies for energy acquisition and allocation for reproduction lie along a continuum between income breeding, with energy for reproduction sourced directly from food available in the environment, and capital breeding, with energy collected and stored in advance until needed for reproduction (Drent and Daan 1980; Tuomi et al. 1983; Stephens et al. 2009). For ectotherms, the energetic and demographic costs associated with storage, maintenance, and use of body reserves are less than for endotherms (Bonnet et al. 1998), suggesting that ectotherms will be more likely to use capital energy sources to fuel reproduction. While this may hold true for amphibians and reptiles (Bonnet et al. 1998), this is not the case for fishes (review by McBride et al. 2013). The strategy for energy acquisition and allocation in fish varies according to the pattern of oocyte recruitment; synchronous and group synchronous oocyte development is generally associated with capital breeding, while asynchronous oocyte recruitment is associated with income breeding, but oogenesis is augmented using a capital strategy (McBride et al. 2013).

Several characteristics of sessile or semi-sessile broadcast spawning marine invertebrates suggest use of a capital breeding strategy over an income breeding strategy. Given availability of food is seasonally variable, particularly for filter-feeders in temperate regions, storing energy for

reproduction is predicted. Moreover, fertilisation success in broadcast spawners requires synchronisation of spawning among individuals in close proximity (i.e. cm) to one another (Babcock et al. 1994; see Levitan 1995 and references therein). Therefore, storing energy for use in oogenesis allows synchronised maturation of oocytes among individuals in the spawning population independent of food availability. Additionally, for endotherms the increased body mass associated with storing energy reserves can affect an individual's vulnerability to predation (Hedenstrom 1992; Gosler et al. 1995), or compromise acquisition of food (reviewed by Witter and Cuthill 1993). This does not seem applicable to semi-sessile marine invertebrates, since stored energy can assist in recovery from swimming escape responses (Brokordt et al. 2000a, b).

Scallops (family Pectinidae) are useful semi-sessile marine invertebrates to explore energy allocation associated with reproduction because the gonad and main energy storage sites, the adductor muscle and digestive gland, are easily isolated. Three main energy sources supporting oocyte growth and maturation have been identified (reviewed by Barber and Blake 2006); lipid stored in the digestive gland, carbohydrate stored mainly in the adductor muscle, and protein also stored in the adductor muscle (Barber and Blake 2006). Temporal changes in stored energy reserves, e.g. glycogen, lipids and protein in the muscle and digestive gland, are associated with gametogenesis (Comely 1974; Barber and Blake 1981; Sundet and Vahl 1981; Epp et al. 1988; Brokordt et al. 2000a; Brokordt and Guderley 2004), suggesting that most scallops that are either semelparous or have a single spawning event per year are likely to adopt a capital breeding strategy to meet energetic requirements for gametogenesis. In contrast, scallops with more than one pronounced spawning season (e.g. *Chlamys varia* in Bay of Brest, France and *Pecten maximus* in Galicia, Spain) will change strategies depending on food supply at the time of maturation; in periods of limited food availability a capital breeding strategy is used, while when excess food is available, an income breeding strategy is adopted (Shafee 1981; Pazos et al. 1997).

Many marine invertebrates, including scallops, have a protracted spawning season and individuals in the population produce and release a number of batches of eggs (Cantillanez et al. 2005; Geiger et al. 2010). However, it is not clear what energy source is used to support this spawning pattern. Identifying energy use and allocation to reproduction based on temporal patterns for species with protracted spawning and asynchronous oocyte development is challenging because several reproductive stages may be present on a single sampling occasion (Domínguez-Petit et al. 2010; Alonso-Fernández and Saborido-Rey 2012). Therefore, the dynamics of energy storage and reproduction should be analysed using both temporal patterns and by reproductive stage. Additionally, in scallops, breakdown of mature oocytes may provide a fourth source of energy for protracted spawning species to support the next batch of growing oocytes (Le Pennec et al. 1991). Given that species with protracted spawning frequently have atretic (necrotic) eggs present it is possible that this source of energy is part of their reproductive strategy (Román 2002).

Pecten fumatus is a simultaneous hermaphrodite with a protracted spawning season involving several partial spawning events (Sause et al. 1987; Fuentes 1994; Young et al. 1999; Mendo et al. 2014). As spawning lasts 5–6 months during spring and summer in Tasmania (Mendo et al. 2014), the aim of this study was to determine whether spawning activity was supported by a capital or income strategy, or combination of both. We examined the use of glycogen, protein, and lipid for oogenesis, and the role of atretic eggs as an alternative energy source for egg growth and maturation. We hypothesised that oogenesis early in the spawning season is fuelled by stored energy reserves, while maturation of later batches of oocytes is supported by higher food concentrations in the spring.

Methods

Environmental variables

From August 2010 to March 2011 monthly mean sea surface temperature (SST) and chlorophyll-*a* [Chl-*a*] for the study area were obtained from MODISA satellite imagery (NASA Ocean Biology 2013) at a 4-km scale and processed using MATLAB v. 7.2 (MATLAB 2006). Sea surface temperature data were retrieved from the information pixel closest to the study area, while [Chl-*a*] (mg m^{-3}) data were retrieved from an average of the five pixels closest to the study area. These data were used as proxies for temperature (SST) and food availability [Chl-*a*] in the area during the study period.

Reproductive cycle

A total of 16 samples of approximately 25 adult scallops [shell length (SL) >100 mm] were collected by divers every 15–20 days from August 2010 to late March 2011, from Great Bay in the D'Entrecasteaux Channel, southern Tasmania (147.33590W and 43.22028S, 12 m depth). Scallops were kept alive in seawater filled plastic containers until processing in the laboratory. Each scallop was measured for SL (to the nearest 1 mm), total weight, gonad weight, adductor muscle weight, shell weight, and digestive gland weight (to the nearest 0.1 g). The gonad was divided into halves longitudinally so that both the ovary and testis were

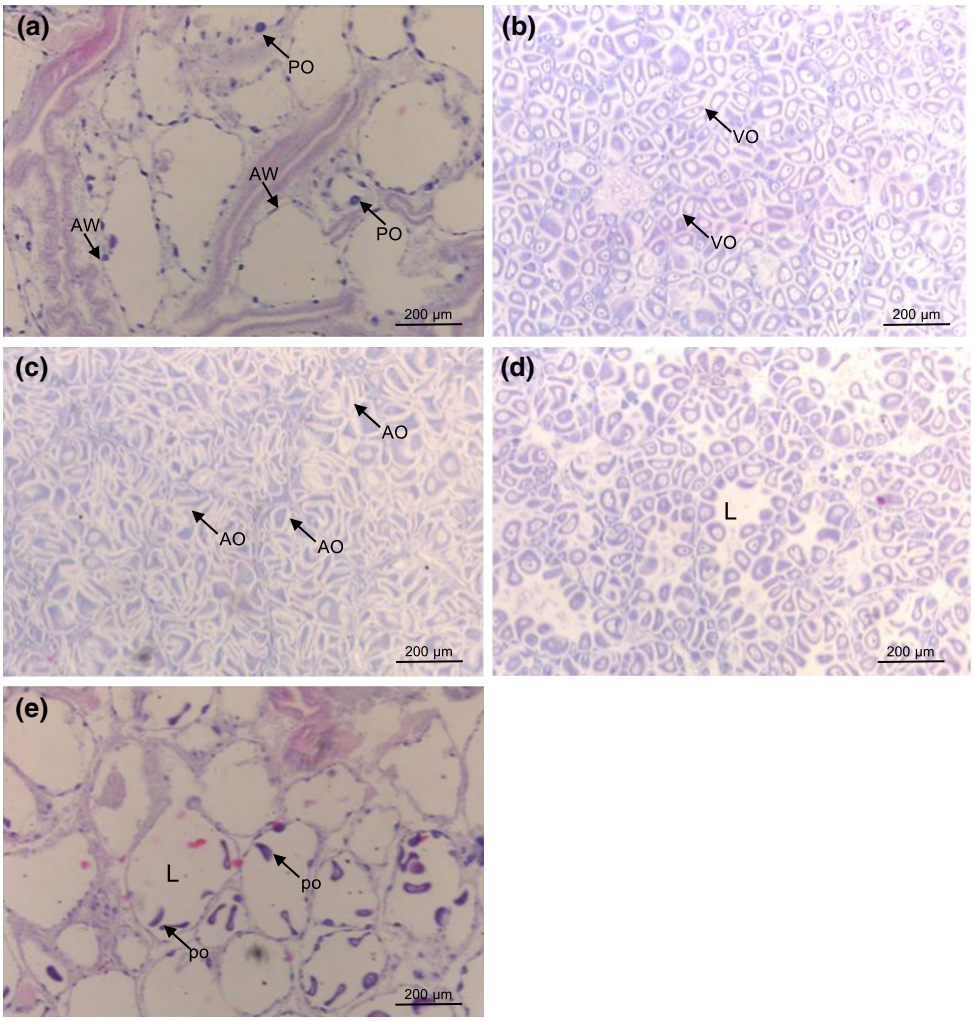

Fig. 1 Histological sections of *Pecten fumatus* ovaries showing different stages of oogenesis: **a** developing, **b** mature, **c** atresia, **d** partial spawning and **e** fully spawned. *AW* acinus wall, *PO* previtellogenic oocyte, *VO* vitellogenic oocyte, *AO* atretic oocyte, *L* lumen, *po* pedunculated oocyte. See Table 1 for the description of the reproductive stages

present in both halves. One half was fixed in FAACC (formalin, acetic acid and calcium chloride) for gonad histology (Winsor 1994) and the other frozen at −40 °C for proximal analysis.

To estimate spawning time and reproductive effort, the gonadal mass index was estimated for each specimen after Bonardelli and Himmelman (1995). First, the slope *b* was obtained from the log-linear regression of scallop SL and gonad mass of *P. fumatus* for each collection date. This slope *b* was then used to calculate the gonad mass Y' for a standard scallop measuring 105 mm (the mean SL of scallops in this study):

$$\text{gonad mass for a standard scallop}_{ij} = \left(Y_{ij}\right)/\left(L_{ij}^{b}\right) * \left(105^{b}\right)$$

where *i* represents the *i*th scallop, *j* is the collection date, *Y* is the gonad mass in g, L is the SL in mm, b is the slope from the regression of log10 gonad mass on log10 SL (Bonardelli and Himmelman 1995). Muscle and digestive gland mass indices were calculated following the same procedure as described above.

Histological examination of gonads was used to identify factors contributing to changes in gonad mass that could be associated with oogenesis and spawning. A decrease in the gonad mass index may be due to either spawning or resorption of gametes, and this can only be determined histologically. Fixed gonad tissue was transferred to 70 % ethanol and stored for at least 48 h, before being embedded in paraffin and sectioned to 6 µm. Sections were stained with haematoxylin and eosin and mounted in a mixture of distyrene, tricresyl phosphate and xylene (DPX synthetic resin mountant) (Kiernan 2008).

To determine the reproductive stage of individuals, a frequency distribution of the oocytes was generated for 14–18 ovaries on 13 of the 16 sampling dates using 50 random points distributed in the ovary with Coral Point Count with Excel extensions (CPCe) version 4.1 (Kohler and Gill 2006). The ovary contains a large number of acini, whose walls are composed of connective tissue and primary germ cells. The lumens of the acini are more or less filled with gametes in varying stages of oogenesis, depending on the reproductive stage (Fig. 1). Reproductive

Table 1 Descriptions of oogenesis stages in *Pecten fumatus*

Oogenesis stage	Description
Developing	Previtellogenic oocytes of various sizes adhering to acinus walls, formation of oocytes in acini; inter-acinal tissue still present (Fig. 1a)
Mature	Large ovarian acini filling space, predominance of fully-developed vitellogenic oocytes (Fig. 1b)
Atresia	Oocytes deformed (jig-saw puzzle appearance); staining affinities change (Fig. 1c)
Partial spawning	Initiation of oocyte release, decrease in number of vitellogenic oocytes in lumen (Fig. 1d)
Fully spawned	Very few free oocytes in lumen; remaining oocytes pedunculated (Fig. 1e)

Stages modified from Mason (1958) and Sause et al. (1987)

stages were identified for ovaries following a scale modified from Mason (1958) and Sause et al. (1987) (Table 1; Fig. 1). When the acinus structure was clearly evident (i.e. Figure 1a) under the random point, the reproductive stage was classified using the appearance of the acini and the oocytes (Table 1; stages developing, partial spawning, or fully spawned). When the acinus structure had broken down or its wall was difficult to observe, the appearance of the oocyte under the random point was assigned a reproductive stage (stages mature or atresia). For each individual the stage in oogenesis was assigned as the most frequently observed stage of the acini and oocytes, excluding atretic acini, because this provided an assessment of the stage of oogenesis. Oocyte lysis was analysed separately. The percentage of atretic oocytes in each scallop was recorded from the random point assessment, and these data were incorporated in the MANOVA analysis detailed below to determine whether any limitation in energy sources in the muscle, digestive gland and gonad could explain the occurrence or extent of atretic oocytes.

Proximal composition

Every month from August 2010 to March 2011, the proximal composition of muscle, gonad (female and male sections combined) and digestive gland tissue was determined for 4–6 randomly selected individuals. Tissues were initially frozen, then freeze-dried, weighed, and ground with a mortar and pestle. Subsamples of each tissue were used to determine concentrations of glycogen, protein, and lipid, which were multiplied by the total dry weight of each tissue to estimate total content per tissue. In gonads, as only half of the tissue was available for proximal analysis, a linear regression between dry and wet weight was determined and used to convert total wet mass to total dry weight. Glycogen concentration was quantified by breakdown of glycogen into glucose units using amyloglucosidase (from *Aspergillus niger*) (Burton et al. 1997). Glucose concentration was measured before and after the use of amyloglucosidae using an Amplex Red Glucose/Glucose Oxidase Assay kit (Life Technologies) and the difference calculated. Glycogen standards from oyster (Type II, Sigma Cat. N G8751) were prepared concomitantly with tissue samples to generate a standard curve for glycogen concentration (Simon and Jeffs 2011). Total nitrogen was determined using a Thermo Finnigan EA 1112 Series Flash Elemental Analyser. Samples were combusted using tungstic oxide on alumina as an oxidising agent followed by reduced copper wires as a reducing agent. The results were calibrated using a certified sulphanilamide standard, and total protein was calculated from total nitrogen using the factor 6.25 (Giese 1967). The concentration of total lipids was determined by the method of Bligh and Dyer (1959) using chloroform/methanol (2:2) and estimated gravimetrically. Energy conversion factors to estimate the total energy content g^{-1} of dry tissue were 17.14 kJ g^{-1} for glycogen (Brody 1945), 17.97 kJ g^{-1} for proteins (Beukema and De Bruin 1979), and 35.20 kJ g^{-1} for lipids (Beukema and De Bruin 1979).

Data analysis

To assess whether the frequency of scallops in each reproductive stage was the same on each of the 16 sampling dates, a Chi-square test of independence was used; if the analysis was significant standardised residuals were used to determine where differences between observed and expected frequencies were large (>2). Differences in the mean standard mass and total energy content of different tissues were compared among the 8 months using a one-way ANOVA. Normality of residuals was assessed visually by plotting the residuals. Homogeneity of variances was assessed using the Bartlett's test (Bartlett 1937). An adjusted Welch's test was used to test equality of means when variances were unequal (Welch 1951) in which the degrees of freedom were adjusted, often producing fractional values. Post hoc pairwise t-tests with a Bonferroni correction were used to determine which sampling dates differed (Wright 1992).

To identify which sources of energy were associated with each reproductive stage the differences in average energy content from glycogen, protein and lipids in the gonad, digestive gland and adductor muscle and the

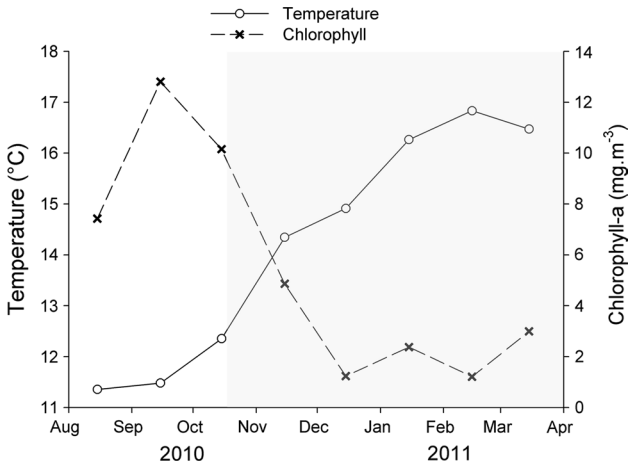

Fig. 2 Monthly mean sea surface (°C) and [Chl-*a*] (mg m^{-3}) in Great Bay, D'Entrecasteaux Channel, August 2010–March 2011. *Grey shading* indicates protracted spawning season

average per cent atresia of 44 scallops in different stages of oogenesis were compared using a one-way MANOVA. Since the fully spawned stage precedes the developing stage and the acinal structures of the gonads are very similar (Fig. 1), the two stages were combined for this analysis. There were strong correlations among some of the energy sources in muscle, digestive gland, and gonad; therefore, only six variables (glycogen in muscle, protein in muscle, protein in gonad, lipid in gonad, lipid in digestive gland and per cent atresia) were used. A sequential Bonferroni (Holm's method) was used to adjust the p values from the pairwise contrasts among the reproductive stages (Quinn and Keough 2002). The Shapiro–Wilk multivariate normality test was used to assess multivariate normality and the Box's M test was used to test homogeneity of covariance matrices using a p value <0.005 to reject the null hypothesis (Huberty and Petoskey 2000). A canonical discriminant analysis followed the MANOVA to identify the variables that explained the differences in the centroid means among the oogenesis stages. Statistical analysis was conducted using the R software package (R Development Core R Development Core Team 2010).

Results

Environmental variables

Sea surface temperature was lowest in August and September, started to warm in October, was highest in February and cooled thereafter (Fig. 2). [Chl-*a*] concentration increased from August to a peak in September, after which a gradual decrease was observed until December and the lowest [Chl-*a*] was recorded thereafter (Fig. 2). Spawning started when temperatures were rising and [Chl-*a*] values were decreasing and continued throughout the warmer months (Fig. 2).

Temporal patterns of gonad mass and oogenesis stages

Average muscle, digestive gland, and gonad mass standardised for SL (105 mm scallop) varied significantly for samples collected August 2010–late March 2011 ($F = 52.44$, df 15, 146.54, $p < 0.001$; $F = 39.08$, df 15, 146.55, $p < 0.001$; $F = 6.77$, df 15, 146.64, $p < 0.001$, respectively). Mean muscle mass was least between August and October 2010 (about 9 g) after which it increased steadily until January 2011, remaining at around 16 g until late March 2011 (Fig. 3a). Digestive gland mass was also lowest August–September (~2 g) but increased rapidly in October to remain ~4 g November–January before declining to an intermediate level in February and March (Fig. 3b). Gonad mass was most variable early in the study period, especially from August to November (Fig. 3c). Mean gonad mass was greatest at the beginning of September (about 5 g) and declined to low levels at the beginning of October (about 3 g). Throughout October and November gonad mass showed a fortnightly pattern of increasing and decreasing mass. These variations in gonad mass were not related to spawning events, as only minor spawning events were observed starting mid-October. There was a small increase in gonad mass in January, after which time mean gonad mass declined (Fig. 3c).

The proportion of different reproductive stages in ovaries changed through time ($\chi^2 = 183.15$, df 36, $p < 0.001$); a greater percentage of individuals in the developing stage (88 %) was observed at the beginning of October and in mid-December 2010 (44 %) (Fig. 3d). Mature individuals occurred in more or less the same proportions (averaging 73 %) throughout the study period, apart from at the beginning of October (12.5 %) and mid-December (25 %) when a greater proportion of developing individuals were present, and in February (22.5 %) when most individuals had fully spawned. From November to late March, a greater percentage (47 % on average) of individuals with partially spawned and fully spawned stages were observed (Fig. 2d).

Based on the frequency of reproductive stages and gonad mass index, the reproductive cycle could be divided into an initial gonad maturation (Aug–Sep) phase with small values of muscle and digestive gland indexes, followed by a protracted spawning phase (Oct–Mar). Adductor muscle and digestive gland mass were least during the maturation phase (Fig. 3a, b). Spawning was detected in October and was associated with an increase in the muscle and digestive mass index. The lowest gonad mass was observed January–late March 2011 when 30–63 % of individuals had fully spawned ovaries (Fig. 3c, d).

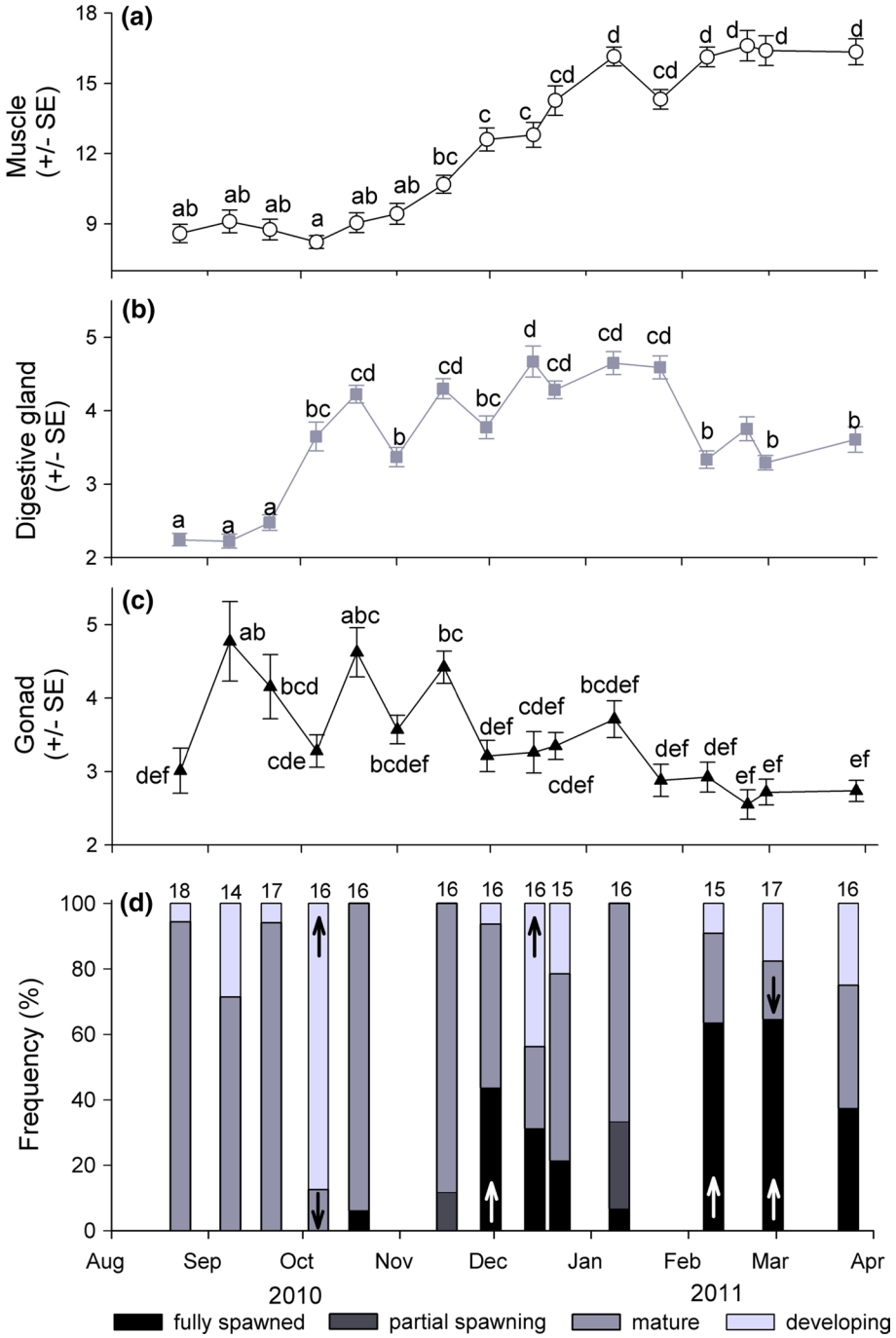

Fig. 3 Changes in mean (± SE) **a** muscle, **b** digestive gland, and **c** gonad mass for a standardised 105-mm SL scallop, August 2010–March 2011. *Different letters* in **a**, **b**, and **c** indicate significant differences among sampling dates. **d** Per cent frequency of females in each oogenic stage August 2010–March 2011. *Arrows* indicate where frequencies were > or < expected in each stage. *Numbers above bars* indicate number of scallops examined histologically on each date

Atresia during the reproductive period

The mean per cent frequency of atretic eggs in the ovary varied from 22.3 to 76.5 % over the 8 months of sampling, with this proportion varying significantly between samples ($F = 4.64$, df 12,195, $p < 0.001$). Other than a more than threefold decline between February and late March, there was no evidence of a strong temporal trend in the changes in the mean per cent frequency of atretic eggs (Fig. 4).

Energy content

The adductor muscle had the greatest mean energy content among the three tissue types ($F = 134.60$, df 2, 82.73, $p < 0.001$) in the form of protein and glycogen with a very small contribution from lipids (Fig. 5a). Average energy content in the muscle derived from glycogen varied significantly over time ($F = 68.82$, df 8,13.03, $p < 0.001$); very little glycogen was present in the muscle tissue from August to October, after which time a 50-fold increase

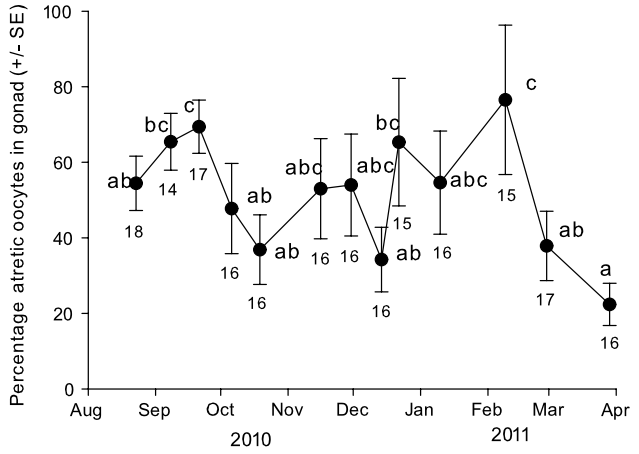

Fig. 4 Mean per cent frequency (±SE) of atretic oocytes in ovary August 2010–March 2011. Means with *different letters* were significantly different from one another. *Numbers* below *error bars* indicate number of scallops sampled on each date

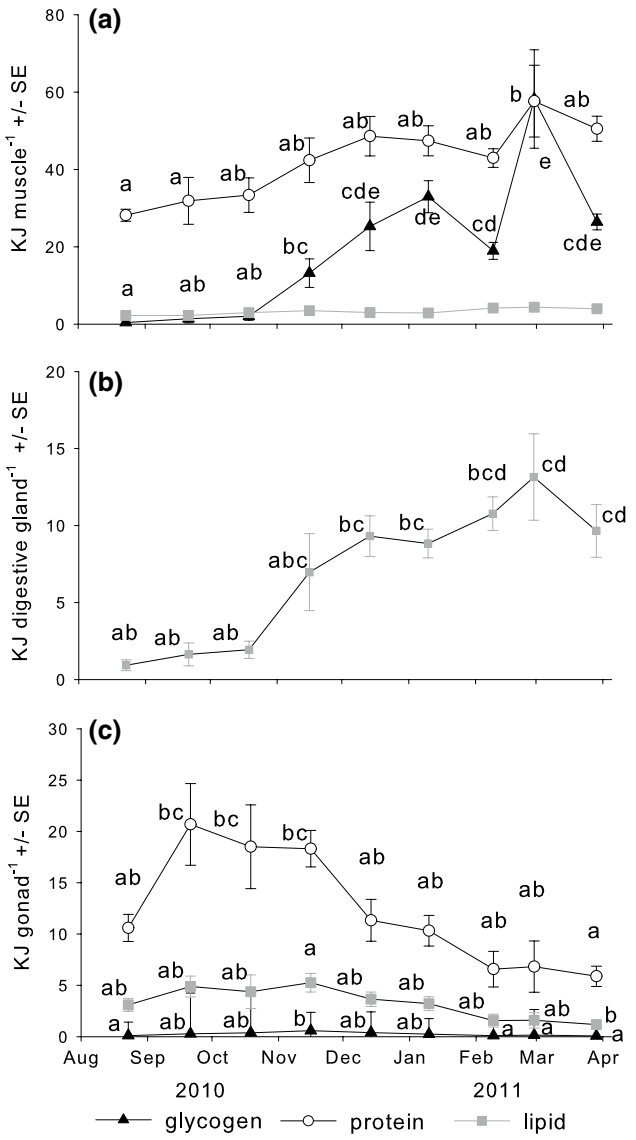

Fig. 5 Changing mean (±SE) energy content as glycogen, protein and lipid (kJ tissue^{-1}) in **a** muscle, **b** digestive gland, and **c** gonad. Note *y*-axis scales differ among panes, $n = 4$–6 sample^{-1}. *Different letters* denote significant differences for each energy source over time

occurred from October to February, followed by a sharp decrease in March (Fig. 5a). Average muscle energy derived from protein also varied temporally ($F = 3.38$, *df* 8, 35, $p = 0.005$), increasing from the least amount in August to its greatest in February (Fig. 5a). Energy from the lipid in muscle showed no evidence of a temporal change over the 8 months ($F = 1.95$, *df* 8, 35, $p = 0.084$). Energy content from lipids in the digestive gland varied significantly over time ($F = 11.29$, *df* 8, 35, $p < 0.001$) with a similar pattern of change as the glycogen content in muscle (Fig. 5b).

Less total energy was present in the gonad compared to the muscle and was mainly protein and lipid (Fig. 5c). Average energy content in the gonad from protein, lipids and glycogen changed significantly during the study period ($F = 5.26$, *df* 8,35, $p < 0.001$; $F = 2.90$, *df* 8, 35, $p = 0.013$; $F = 4.11$, *df* 8,35, $p = 0.001$, respectively). Average protein values were greatest during maturation in September and then declined steadily during spawning to approximately a quarter of what was present initially. While average lipid values were significantly different between the beginning and end of the spawning phase, lipid content was relatively low and the change was not great (Fig. 5c). Average glycogen levels were very low in the gonad, and the changes through time were very small (Fig. 5c).

Dynamics of energy use and storage by oogenesis stage

Energy storage showed a clear pattern of change that could be related to reproduction. Differences in proximal composition were evident among the reproductive stages ($F = 3.982$, *df* 2,12, $p < 0.001$), largely between mature and fully spawned stages ($F = 10.429$, *df* 6,30; $p < 0.001$), with partial spawning individuals similar to both mature and fully spawned individuals. Differences between mature and fully spawned stages were driven by the percentage of atresia, glycogen and protein content in the muscle, and lipid in the digestive gland (Fig. 6). The percentage of atresia was greatest in scallops with mature gonads but lowest in fully spawned individuals. Mature scallops were associated with greater values of lipid and protein in the gonads. Partial spawning and fully spawned individuals were associated with higher values of glycogen and protein in muscle and lipid in the digestive gland (Fig. 6). The percentage of atresia was unrelated to any of the energy sources (Fig. 6).

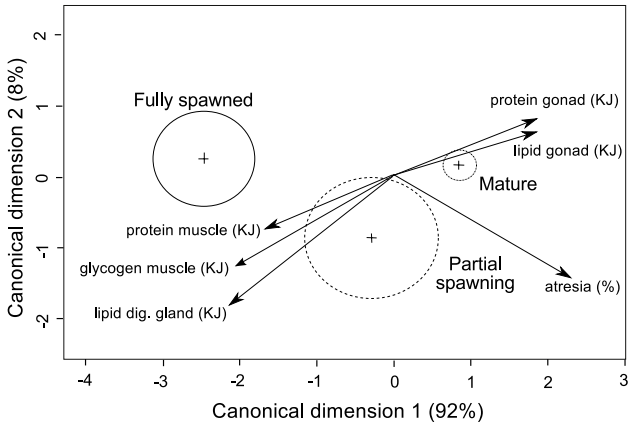

Fig. 6 Centroid means with 95 % confidence ellipses, for three oogenesis stages on first two canonical discriminant dimensions. Direction and length of vectors show strength and nature of correlation with each variable and canonical discriminant axes. Per cent variability among three centroid means explained by each axis

Discussion

Pecten fumatus spawned over a protracted period, with evidence that energy sources for gamete production varied through time. The initial peak of oogenesis in winter–spring (August–October) was most probably fuelled by stored energy substrates, while the production of eggs for spawning over summer (November–March) appeared to be fuelled by recycling of mature oocytes that were not released during the first spawning event. Examining seasonal changes in energy reserves by reproductive stage showed that mature scallops had less muscle glycogen and protein and digestive gland lipid content compared to fully spawned scallops. This pattern of energy use to fuel gametogenesis is typical of pectinids (Barber and Blake 1981; Strohmeier et al. 2000; Arellano-Martinez et al. 2004; Brokordt and Guderley 2004). However, oocyte maturation later in the spawning season occurred when energy substrates in the muscle and digestive gland were increasing, despite [Chl-*a*] (a proxy of food availability) decreasing. As this suggests there was sufficient food to allow energy storage, this does not exclude the possibility that ingested energy was also contributing to egg maturation, i.e. an income strategy. However, the presence of substantial numbers of atretic oocytes throughout the spawning period suggests that metabolites produced by the recycling of gonad products via atresia (oocyte lysis) were definitely being used to support oogenesis for the latter part of the spawning cycle. A similar mechanism of energy recycling for oocyte maturation occurs in *Pecten maximus*, where energy for successive cohorts of developing oocytes was supplied through resorption of atretic material (Dorange and Le Pennec 1989; Le Pennec et al. 1991). Therefore, it appears that individuals in scallop populations with a protracted spawning season either use a capital breeding strategy, or they use energy for an extended spawning period by resorbing oocytes in the earlier batch of oocytes (present study), or a combination of capital and income strategies (Román et al. 2002). Teleost and pectinid species that are batch spawners with asynchronous oocyte development use this combination of energy strategies to fuel reproduction (review by McBride et al. 2013).

The reproductive cycle in *P. fumatus* was characterised by the presence of atretic oocytes during the whole study period, which is not uncommon for pectinids that undergo protracted spawning, e.g. *P. fumatus* (Young et al. 1999), *Pecten maximus* (Tang 1941; Motavkine and Varaksine 1989; Duinker and Nylund 2002), *Placopecten magellanicus* (Barber et al. 1988), *Argopecten irradians irradians* (Epp et al. 1988) and *Argopecten purpuratus* (Cantillanez et al. 2005). Mature oocytes in scallops have a relatively short residence time in the gonad and if not released undergo lysis, initiated by putative lysosomes present in mature oocytes (Dorange and Le Pennec 1989). Oosorption in the polychaete *Nephtys hombergi* converts energy in mature oocytes back into stored energy and somatic growth which can be used to fuel reproduction in the following year (Olive et al. 1981). The large numbers of atretic oocytes in the gonads of *P. fumatus* may be a similar energy conservation process to support protracted spawning independent of external energy, but in this case fuelling oocyte development in the current spawning season. As such, oosorption may be an important process supporting the semi-annual or protracted spawning reproductive strategy of pectinid species (Román 2002).

There was no evidence that either temperature or stores of glycogen, protein or lipids were associated with the degree of oosorption throughout the spawning season of *P. fumatus*. While temperature and food availability are factors known to influence gamete resorption in marine invertebrates (Barber et al. 1988; Paulet et al. 1988; Soudant et al. 1996; Olive et al. 1997), these associations are not consistently present among populations, e.g. *Pecten maximus* (Paulet et al. 1988; Strand and Nylund 1991; Pazos et al. 1996). Several other possibilities should be investigated to identify the processes driving oocyte resorption in scallops. It is possible that, as in polychaetes, a critical level of energy is needed to support survival which would be reduced if energy was lost as gametes; hence, the need to recycle energy (Olive et al. 1981, 1997). Alternatively, if mature oocytes cannot be retained for long in the gonad, then the energy from the oocytes can be recycled and used to fuel the next batch of maturing oocytes if external spawning cues are not received to trigger release

of oocytes (Babcock and Keesing 1999; Styan and Butler 2003; Mendo et al. 2014).

If oosorption is related to the semi-annual or protracted spawning reproductive strategy of pectinid species (Román 2002), then quantifying spatial and temporal extent of atresia in individuals and populations adopting different reproductive and energy strategies will inform the adaptive significance of atresia and its role in supplementing energy demands for oogenesis. The resorption of mature oocytes presents difficulties in interpreting temporal patterns of change of the gonadosomatic index (GSI), as declines in GSI may be a function of either spawning or atresia. Similarly, counts of vitellogenic eggs may overestimate fecundity if atresia is a regular part of a process of energy recycling. Obtaining accurate estimates of fecundity and the timing of spawning events will require knowledge of the temporal patterns of atresia and the factors influencing atresia. Given the almost twofold difference in mean gonad mass observed for *P. fumatus* on the scale of weeks, an intense sampling program is necessary to describe the reproductive dynamics in scallops (Román 2002). In the longer term, identifying the factors triggering atresia will not only allow correct interpretation of temporal changes in GSI, but also identification of spawning failure in a population.

In conclusion, exploring seasonal changes in energy reserves as a function of oogenesis stage has shown that muscle glycogen and protein and digestive gland lipid content in mature *P. fumatus* provide energy to support an initial peak in oocyte production. Given that there was no evidence of energy stores being used later in the season at a time when food availability was low, it appears that metabolites produced from oocyte lysis may have fuelled subsequent oogenesis. This study supports a model that scallops with protracted spawning initially use a capital breeding strategy, with energy either from carbohydrate or lipid stores, and then oocyte lysis-derived energy to sustain maturation. However, we cannot eliminate the possibility that an income strategy fuelled or partly fuelled oocyte maturation later in the spawning season. The reasons that atresia occurs so extensively in *P. fumatus* are unclear, especially given that the degree of atresia was not associated with the current nutritional condition or water temperature, and further exploration of the process is required. This will also allow predictions of when temporal changes in GSI are a function of spawning events or when substantial atresia is occurring. Recycling of energy from oocyte resorption represents a strategy of energy use to fuel reproduction that may be important in other marine invertebrates.

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