Marine Pollution Bulletin 127 (2018) 342–351

Contents lists available at ScienceDirect

Marine Pollution Bulletin

journal homepage: www.elsevier.com/locate/marpolbul

Volcanic ash in the water column: Physiological impact on the suspension-feeding bivalve *Mytilus chilensis*

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ARTICLE INFO

Keywords:
Mytilus chilensis
Volcanic ash
Physiological responses
Weight loss
Suspension-feeding
Mussels
Clearance rate
Oxygen consumption rate

ABSTRACT

Ashes settling into the sea from volcanic explosions expose suspension-feeding species to reduced seston quality. Adults and juveniles of the mussel *Mytilus chilensis* were exposed for 15 days to the phytoplankton *Isochrysis galbana* together with various concentrations of ashes. We then quantified impact on survival and physiology. Although no individuals died during the experiment, by the end of the study clearance rates and oxygen consumption rates had decreased substantially, and tissue weight of mussels exposed to the highest ash concentrations declined substantially. Gills showed no physical damage, but did show abundant mucus secretion in response to ash particles. Moreover, as the relative proportions of microalgae to ash in the diet decreased, individuals showed increasing preferential ingestion of microalgal particles. Increased ash content in the diet altered physiological rates and activated distinct particle selection with a high production of pseudofeces and high energy costs, with potential long-term consequences.

1. Introduction

Periodic volcanic eruptions are natural phenomena that can produce global catastrophic effects (Self, 2006), due to the emission of large quantities of gas and ash particles into the atmosphere. Depending on the magnitude of the eruption, the volcanic ashes can remain for several weeks in the troposphere and for months in the stratosphere (Klűser et al., 2013), with high dispersion around the world (Durant et al., 2012; Vernier et al., 2013). The physical and chemical properties of the elements expelled in these events, can lead to negative effects on the physiology of plants (Hirano et al., 1995), arthropods (Buteler et al., 2011; Wolinsky et al., 2013), and fishes (Newcomb and Flagg, 1983), as well as on human respiratory health (Horwell and Baxter, 2006).

When ashes rain down along the coasts, they can cause displacement and massive mortality of benthic organisms (Jewett et al., 2010). Suspended in the water column, ashes become a particularly serious problem for many organisms, through abrasion (e.g. physical damage) or by altering various physiological processes, such as respiration (Newcomb and Flagg, 1983) or feeding (Shirakawa et al., 1984). With juvenile salmon, for example, ashes adhere to the gills, damaging the gill epithelium and making it difficult for the animals to obtain oxygen (Newcomb and Flagg, 1983). For freshwater zooplankton, ingestion of ashes has been shown to damage the digestive tract, again through abrasion (Wolinsky et al., 2013). To our knowledge, similar studies have not been previously reported for marine suspension-feeding invertebrates, even though the volcanic ash can obviously enter the water column and become part of the available particulate material in suspension (seston). Suspension-feeding organisms– especially bivalve molluscs—are therefore likely to be strongly affected, since they are forced to interact directly with the abrasive ash particles as they circulate seawater in their mantle cavity during the process of feeding and/or respiration. In these animals, changes in the concentration and quality of seston can strongly modify physiological processes, including clearance rates, which are typically reduced at high seston concentrations (Newell et al., 2001). Moreover, high particle concentrations–particularly of inorganic particles (Bayne et al., 1993)–can cause particles to be rejected as pseudofeces (Navarro and Velasco, 2002), a phenomenon with clear energetic costs: in addition to the normal costs of suspension-feeding there are now added costs generated by the selection and removal of particles in the form of mucous aggregations (Beninger and St-Jean, 1997a, 1997b; Beninger et al., 1999; Beninger and Dufour, 1996; Kiørboe et al., 1980).

Alterations in diet particle composition may also affect rates of oxygen consumption (Hutchinson and Hawkins, 1992; Bayne and

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https://doi.org/10.1016/j.marpolbul.2017.12.024
Received 25 August 2017; Received in revised form 7 December 2017; Accepted 8 December 2017
Available online 21 December 2017

Thompson, 1970), which, together with changes in other physiological responses, may affect both growth (Bayne and Widdows, 1978; Bayne and Newell, 1983) and survival (Irlandi et al., 1997; Akberali and Trueman, 1985). Pre-ingestion particle selection has been previously demonstrated in some suspension-feeding molluscs (Beninger et al., 2007; Levinton et al., 2002; Bougrier et al., 1997; Chaparro et al., 2013), although which particles are ingested and which are rejected as pseudofeces is in many cases still not clearly established (Rosa et al., 2013). However, a high concentration of inorganic particles can activate pseudofeces production, thereby improving the quality of the particles that are actually ingested in comparison to what is available (Jørgensen, 1996). In this way, suspension-feeding organisms can regulate ingestion rates and improve the nutritional quality of ingested material relative to what is available (Navarro and Widdows, 1997; Ward and Shumway, 2004). Thus, faced with the persistent presence of suspended volcanic particles, suspension-feeders could possibly distinguish between nutritive particles and volcanic ash particles and expend extra energy in eliminating the latter as pseudofeces.

The Southern-Austral part of Chile is widely exploited for obtaining mussel spat (*Mytilus chilensis*) and doing massive culture of this sessile, suspension-feeding mussel (231,659 tons of these mussels were harvested in 2014; SERNAPESCA, 2015). In addition to the economic importance of this species, it also plays an important ecological role in structuring macrofaunal communities (Acuña et al., 2012). The recent volcanic eruptions in the area (especially that of the recent Calbuco volcano eruption, April 2015, Delgado et al., 2017, Vidal et al., 2017) have led to serious questions about the impact of ash particles on the survival and physiological functioning of *M. chilensis*. Suspension-feeding mytilids direct incoming particles towards the pallial cavity, where they are retained by the gill filaments and then moved to the ventral food grooves where they become embedded in a mucous cord. Particles are then displaced along the food groove by cilia, and moved towards the anterior region of the animal (Garrido et al., 2012; Navarro et al., 2011). Once they reach the labial palps, the mucous cords are disaggregated and particles are then either directed towards the mouth for ingestion or rejected as pseudofeces (Beninger et al., 1999; Beninger and St-Jean, 1997a). In this process, the labial palps play a major role in determining which particles get ingested and which get rejected (Garrido et al., 2012). Thus it is possible that by actively participating in collecting abrasive volcanic material from the seston, the gills could suffer some level of physical damage, which may in turn explain any variations seen among individuals in clearance rates, changes in physical condition (e.g. tissue content), or survival.

In the present research we studied the impact of diets varying in volcanic ash content on adult and juvenile survival of *Mytilus chilensis* over a 2 week period of exposure. We also monitored individual variation in oxygen consumption and clearance rates along with changes in individual biomass for animals fed the different diets. We also quantified the capacity of these mussels to select between microalgae and volcanic ash particles for ingestion or rejection, to assess their ability to thereby improve the quality of the ingested food in comparison to what is available in the water column. Electron microscope studies also allowed us to identify possible physical damage to the gills, caused by the abrasiveness of the volcanic material retained during the clearance process.

2. Material and methods

2.1. Obtaining the experimental material

Adults and juveniles of *Mytilus chilensis* were obtained by divers in the Caleta Amargo sector, Los Ríos Region, southern Chile (39° 52′ 30″ S, 73° 25′ 34″ W) during the southern hemisphere summer of 2016, and taken to the laboratory, where they were kept for a maximum period of 1 week before being used in the experiments. The specimens were maintained in a 200 L aquarium with circulating seawater, taken from the natural environment.

Volcanic ash obtained from the Calfuco volcano explosion of April 2015 was collected by hand in July 2015 near Puente Chico, in the Lakes Region of southern Chile (41° 27′ S, 72° 49′ W) and taken to the laboratory. The ashes were forced through an 80 μm sieve, and particles retained by the sieve were then discarded. The smaller particles that passed through the sieve were then subdivided into different size classes. For this, we suspended the particles in distilled water and by using different times of decantation the particles were subdivided into a number of size classes. The size fractions thus obtained were then dried for 48 h in an oven at 60 °C. In this way we obtained a fraction with ash particles 3–20 μm in diameter; this fraction was used in our experiments (see supplementary material 1).

2.2. Survival and physiology of M. chilensis exposed to diets containing volcanic ash

2.2.1. Survival of M. chilensis

An experiment was conducted to identify potentially lethal impacts of volcanic ash particles on *M. chilensis*. Adults (n = 180 individuals, 5.2 ± 0.3 cm shell length) and juveniles (n = 180 individuals, 2.1 ± 0.3 cm shell length) were both tested. Both received as food a mixture of phytoplankton (*Isochrysis galbana*), at a standard concentration of 50,000 cells mL^{-1} (equivalent to 1.5 mg dry weight; Navarro and Chaparro, 2002) and one of several different concentrations of volcanic ash: 0 mg ashes L^{-1} (control), 30 mg ashes L^{-1}, 60 mg L^{-1}, 90 mg L^{-1}, 120 mg L^{-1} and 150 mg L^{-1}. The maximum ash concentration tested in this study corresponded to the mass that was found in a freshwater lake (Pire Lake) close to the Puyehue-Cordón Caulle volcano following the 2012 eruption (Balseiro et al., 2014). For our experiments we used 18 forty-liter aquaria for adults and 18 twenty-liter aquaria for juveniles, filled with seawater taken directly from the natural environment (14 ± 1 °C and 32 ± 1 PSU) and filtered as described below. The seawater was forced through filters of 20 μm, 5 μm and 1 μm and then sterilized with UV light before being added to the aquaria. The seawater was changed in all aquaria each morning. Food was added once each morning and again in the afternoon; any settled feces and detritus was removed during the next morning before the new food was added.

Control adults and juveniles were distributed among 3 aquaria and fed only a unialgal diet (*I. galbana*) for the full 15 day period. For all treatments, particles were kept in suspension within the aquaria by using a submersible pump and constant aeration.

2.2.2. Effects on oxygen consumption rates (OCR) and clearance rates (CR)

OCR and CR were determined at the end of the 15-day survival study, to document physiological changes caused by exposure to volcanic ash.

2.2.2.1. Oxygen consumption rate (OCR). At the end of the 15-day survival assessment study, OCR was determined for 12 adults and 12 juveniles subsampled from each treatment (72 adults and 72 juveniles examined in total). Specimens were individually installed in hermetic glass chambers filled with filtered seawater (14 ± 1 °C and PSU 32 ± 1) that had previously been saturated with oxygen by bubbling. The volume of seawater in each chamber varied between 150 mL and 1000 mL, depending on the size of the individuals being tested. The aerated seawater was allowed to sit undisturbed for 15 min before animals were added, to facilitate the escape of any remaining air bubbles. The animals were then individually placed inside the respiration chambers while the chambers were submerged under water, to prevent the entry of air; the chambers were then sealed while still underwater, for the same reason. Oxygen concentrations were determined non-invasively every h for up to 3 h, using a PSt3 optimal oxygen sensor (PreSens Precision Sensing GmbH). Oxygen concentrations were also monitored in 2 control aquaria with similar conditions as the experimental aquaria, but without animals inside.

OCR was calculated as follows:

$$OCR = (((C_0 - C_f) - PC) \times Vol)/Time$$

Where:

OCR = oxygen consumption rate,

C_0 = initial oxygen concentration in the chamber,

C_f = final oxygen concentration in the chamber,

PC = mean value of oxygen within the control chambers,

Vol = volume (L) of fluid in the chambers (discounting the mussel volume), and

Time = hours between measurements.

2.2.2.2. Clearance rate (CR). At the end of the 15-day feeding period, we collected 12 juveniles and 12 adults from each treatment ($n = 72$ individuals in total for each of the two groups). Each individual was placed in either a 2 L or 4 L aquarium, depending on the size of the specimens being tested (juveniles or adults). The aquaria were filled with seawater (14 ± 1 °C and 32 ± 1 PSU) that had been previously filtered to 1 μm. Before introducing animals into the aquaria, enough *Isochrysis galbana* suspension was added to achieve an initial concentration of 50,000 cells mL^{-1}. The water in each aquarium was aerated constantly, which also kept the phytoplankton in suspension. For each quantification of CR, we also sampled the water from two control aquaria, which were identical with the others except without animals inside.

Algal concentrations were determined using 50 mL subsamples from each aquarium, both at time zero (start) and at 1 h intervals, up to a maximum of 3 h or until the concentrations dropped by > 40% of the initial concentrations. The number of particles per mL was quantified twice in 0.5 mL subsamples using a Beckman Model Z2 Coulter Counter equipped with an aperture of 100 μm. After each quantification, the remaining volume of the sample was immediately returned to the aquarium of origin to minimize volume changes over time.

CR was expressed as L filtered h^{-1} g^{-1} dry tissue weight for adults, and as L filtered h^{-1} 0.1 g^{-1} dry tissue weight for juveniles and calculated according to Coughlan (1969).

2.3. Electron microscopy of gill lamella

To identify any physical damage to the gills caused by the volcanic ashes suspended in the water column, pieces of gill lamellae were taken on day 15 from 3 control individuals and from 3 individuals that had been exposed to the highest ash concentration (150 mg L^{-1}) for the full 15 d, and prepared for scanning electron microscopy (SEM). Gill samples were dehydrated for 10 min at each of the following alcohol concentrations: 50%, 60%, 70%, 80%, 90%, and 95%, and were later washed twice in 100% ethyl alcohol and then critical point dried using a Hitachi model HPC-2 desiccator, with pressurized CO_2 as a transitional fluid. Samples were placed on aluminum studs (porta samples) and plated with gold. Samples were then examined using a model LEO-420 scanning electron microscope attached to a PC.

2.4. Dry tissue weight

Dry tissue weights were quantified for all experimental and control animals on d 15, at the completion of the experimental exposure period. Shell lengths were recorded to the nearest 0.1 mm as the distance from the anterior end of the shell to the posterior end, using digital calipers, and the tissue was then carefully removed from each individual and placed in pre-weighed, numbered aluminum foil containers. Samples were then dried for 48 h at 60 °C and placed in a desiccator; dry weights were later determined to the nearest 0.1 mg using an A & D Model GR-202 Semi MicroBalance.

2.5. Preferential selection of particles from the seston: index of electivity

These experiments were designed to determine whether the mussels could select against ash particles, and the extent to which the relative concentration of ash and phytoplankton particles influenced this "index of electivity". Thirty adults of *M. chilensis* (50 ± 3 mm long shell) were cleaned of epibionts and maintained in the laboratory in flowing seawater for up to a week before being used in the experiments.

For these experiments, we used 6 one-L aquaria per treatment (total $n = 36$), each filled with 1 μm filtered seawater. The phytoplankton *Isochrysis galbana* was added to each aquarium to attain an initial concentration of 50,000 cells mL^{-1} (equivalent to 1.5 mg dry weight; Navarro and Chaparro, 2002). Animals were held at one of the following concentrations of volcanic ash: 0 mg ashes L^{-1}, 30 mg ashes L^{-1}, 60 mg L^{-1}, 90 mg L^{-1}, 120 mg L^{-1} and 150 mg L^{-1}. The seawater in each aquarium received constant aeration. One adult mussel was then placed into each aquarium. Five minutes after the animal began processing seawater through its mantle cavity we took 3 samples of seston near the inhalant area of each individual. Samples of approx. 0.5 mL of water were deposited on microscope slides and then photographed using a camera mounted onto a fluorescence microscope. For each sample, we took one photograph using epifluorescence illumination and another with normal light. After 2 h, the experiment was stopped; samples of the pseudofeces were then extracted from the bottom of each tank and deposited into 5 mL Eppendorf tubes. The contents were then diluted with 2 mL of filtered seawater and stirred vigorously to disperse the particles. Then, 3 sub-samples were taken from each tube, put on glass microscope slides, and photographed with either epifluorescence or normal light. Photographs from the inhalant area and from pseudofeces samples were taken with an Olympus BX41 microscope equipped with a Q-imaging camera, and the epifluorescence images were taken using a TRICT filter. Fluorescence microscopy allowed us to determine the amount of microalgae present in the pseudofeces, while the normal lighting allowed the identification of ashes. Q IMAGING 6.0 software was used to capture the images and the image processor GIMPPORTABLE was used to identify and count the microalgae.

Both ash particles and microalgae were counted in the same area of each sample. The resulting data were used to estimate the electivity index (EI) (Jacobs, 1974, modified by Baker and Levinton, 2003), which identifies the degree of acceptance or rejection of organic and inorganic particles by suspension-feeders. The EI was calculated as follows:

$$EI = -\{(S - P)/[(S + P) - (2 \times P \times S)]\}$$

where:

P = proportion of particles of interest in pseudofeces (specific number of particular particles/total particles),

S = proportion of particles of interest in suspension (specific number of particular particles/total particles).

The EI can range from − 1.0 to 1.0. A positive value for EI indicates that type of particle was preferably selected for ingestion (i.e., that particle type was underrepresented in the pseudofeces compared with the numbers found in suspension), while a negative value of EI indicates rejection (i.e., that type of particle was enriched in the pseudofeces compared with what was found in suspension). Values close to zero indicate the absence of selection. In either direction.

2.6. Clearance rate in diets containing different proportions of microalgae and ash particles

A total of 150 adults of *M. chilensis* were cleaned of epibionts, measured to the nearest 0.1 mm in shell length, and maintained for one week in aquaria with circulating seawater taken directly from the natural environment. The individuals used in these experiments had shell lengths of 50 ± 3 mm ($n = 150$).

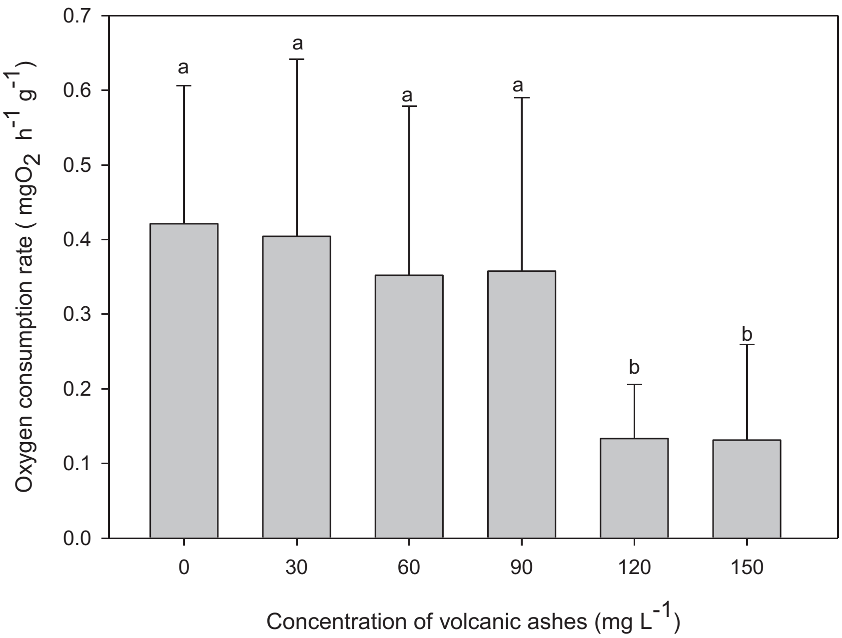

Fig. 1. *Mytilus chilensis*: The impact of volcanic ash on oxygen consumption rates in adults, after exposure for 15 days to diets containing a constant concentration of 50,000 cells *I. galbana* mL^{-1} and different amounts of volcanic ash. Each bar represents the mean ± standard deviation per treatment (total $n = 12$ individuals per treatment). The different letters on the bars indicate significant differences between means ($p < 0.05$).

2.6.1. Preparation of diets and experimental aquaria

We prepared five diets with different ratios of volcanic ash particles to micro-algae as follows: 100:0, 75:25, 50:50, 25:75 and 0:100. Ash particles that were used in preparing the diets for this experiment had a spherical diameter of 3–20 μm. This range of sizes of ashes was obtained through a series of successive deposits and with different settling times, after diluting the volcanic material in fresh water. Phytoplankton were obtained from mass cultures of *Isochrysis galbana*.

CR measurements were conducted as previously described. On this occasion, we used an initial concentration of 50,000 particles mL^{-1}, which consisted of a combination of volcanic ash particles and micro-algae in the following proportions: 100:0, 75:25, 50:50, 25:75 and 0:100. The experimental volume of each aquarium was 4 L. For each treatment were used 3 sets of 10 aquarium tanks each, with each tank holding one adult mussel, approximately 50 ± 3 mm ($n = 30$) in shell length. During each experimental event, we used 2 control aquariums with the same characteristics of the experimental aquaria, but without the animals inside. Constant aeration kept the particles in suspension.

2.7. Standardization and transformation of variables

The measured physiological rates were converted to an individual of “standard” dry tissue weight, in accordance with the following equation (Bayne et al., 1987):

$$Ys = (Ws/We)\, b \times Ye$$

where:

Ys = physiological rate for a weight-standardized individual,
Ws = standard weight,
We = measured (actual) weight,
Ye = actual physiological rate without standardization and,
b = exponent of the regression between the physiological measurement and weight of the experimental specimen.

The following regression exponents were obtained from Navarro and Winter (1982) for *M. chilensis*: 0.62 for CR and 0.74 for OCR.

2.8. Statistical analysis

The Shapiro-Wilk test for normality and the Levene test for homoscedasticity were used to determine whether to evaluate our data using a parametric (ANOVA) or non-parametric (Kruskal-Wallis) test. One-way analysis of variance (ANOVA) followed by Tukey tests were used to determine whether the concentrations of ash particles in the seawater altered CR significantly ($p < 0.05$). The same analysis was used to identify significant differences between CR's depending on the variation in the proportions of ash particles and microalgal cells in the experimental diets. The Tukey test allowed us to determine which diets resulted in significantly different CR's. The non-parametric Kruskal-Wallis test was used to identify significant differences in OCR between treatments.

t- tests were used to determine whether the EI values from the inhalant area (seston) and the pseudofeces samples differed significantly from zero. Moreover, comparisons of dry tissue weights between the experimental mussels at the end of 15-day exposure to diets containing ash particles and those from controls, whose individuals were fed only with microalgae, were made using a *t*-test.

All data were analyzed using the program STATISTICA version 7.

3. Results

3.1. Fifteen day experiments in M. chilensis exposed to diets with volcanic ashes

3.1.1. Survival

No adult or juvenile mussels died during the 15-day exposure period, regardless of the proportion of ash particles incorporated into the diet to which they were exposed.

3.1.2. Physiological measurements: oxygen consumption rates (OCR) and clearance rates (CR)

Adult mussels, after 15 days of exposure to diets including volcanic ash, presented significant reductions in rates of oxygen consumption (OCR) (Fig. 1) (Kruskal Wallis test; $H =_{(5.47)} 18.691$, $p = 0.002$), when they were exposed to 120 mg ashes L^{-1} (0.133 ± 0.072 mg $O_2\,h^{-1}\,g^{-1}$, $p = 0.034$) and 150 mg ashes L^{-1} (0.131 ± 0.128 mg $O_2\,h^{-1}\,g^{-1}$ $p = 0.030$). The control specimens presented OCR's > 3 times higher, with an average OCR (± SD) of 0.421 ± 0.185 mg $O_2\,h^{-1}\,g^{-1}$.

Adults showed significant decreases in clearance rates (Fig. 2, 1-way ANOVA, $F_{(5.56)}$ 3.689, $p = 0.005$) after experiencing the treatments

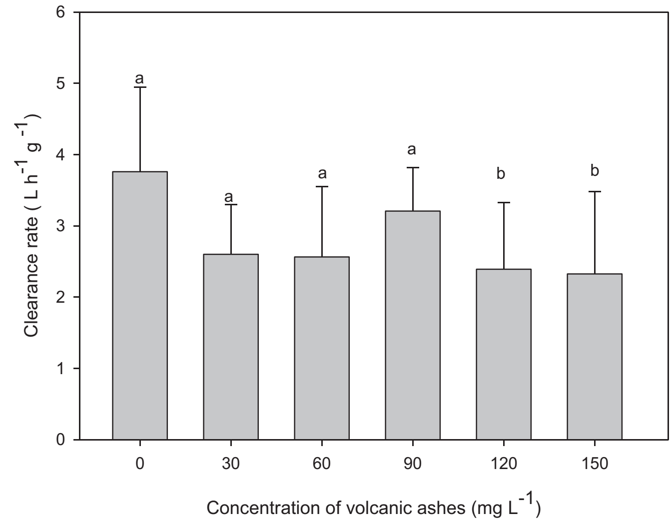

Fig. 2. *Mytilus chilensis*. The impact of suspended volcanic ash on clearance rates in adults, after they had been exposed for 15 days to diets containing a constant concentration of 50,000 cells of *I. galbana* mL^{-1} and different quantities of volcanic ash particles. Each bar represents the mean ± standard deviation per treatment ($n = 12$ individuals per treatment). Different letters at the tops of bars indicate significant differences between means ($p < 0.05$).

with the highest concentrations of volcanic ash: 150 mg L^{-1} (2.324 ± 1.157 L h^{-1} g^{-1}) and 120 mg L^{-1} (2. 390 ± 0.935 L h^{-1} g^{-1}). The test identified Tukey HSD a posteriori differences for the 120 mg L^{-1} concentration ($p = 0.015$) and 150 mg L^{-1} ($p = 0.016$) with respect to the control. In the latter, there was an average CR 3.759 ± 1.188 L h^{-1} g^{-1} ($n = 11$).

For juveniles, in contrast to results obtained for adults, the level of ash content in the diet produced no significant differences in mean oxygen consumption rates (Fig. 3; Kruskal Wallis test; $H = _{(5.70)}$ 2.519, $p = 0.773$), although oxygen consumption was seen to decrease with increased concentrations of volcanic ash particles.

For juveniles, although mean clearance rates (CR) decreased in the presence of increased proportions of volcanic ash, the differences were not statistically significant (1-way ANOVA, $F_{(5,64)} = 0.130$, $p = 0.273$) (Fig. 4).

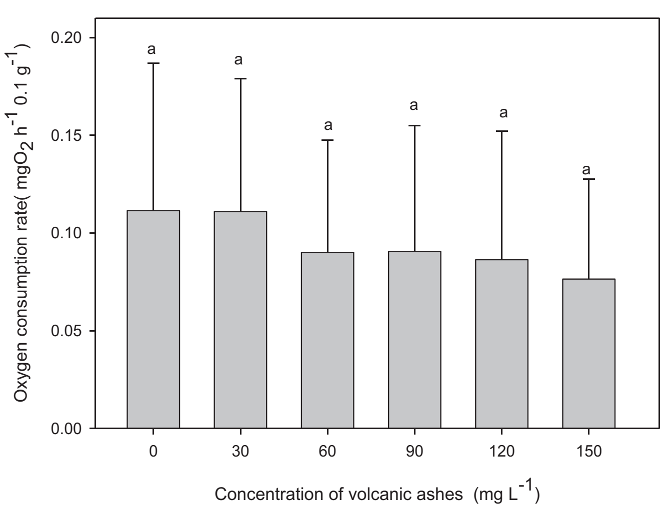

Fig. 3. *Mytilus chilensis*. The impact of suspended volcanic ash on oxygen consumption rates in juveniles, after being exposed for 15 days to diets containing a constant concentration of 50,000 cel *I. galbana* mL^{-1} and different quantities of volcanic ash. Each bar represents the mean ± standard deviation per treatment (total $n = 12$ individuals per treatment). Different letters above bars indicate significant differences between means ($p < 0.05$).

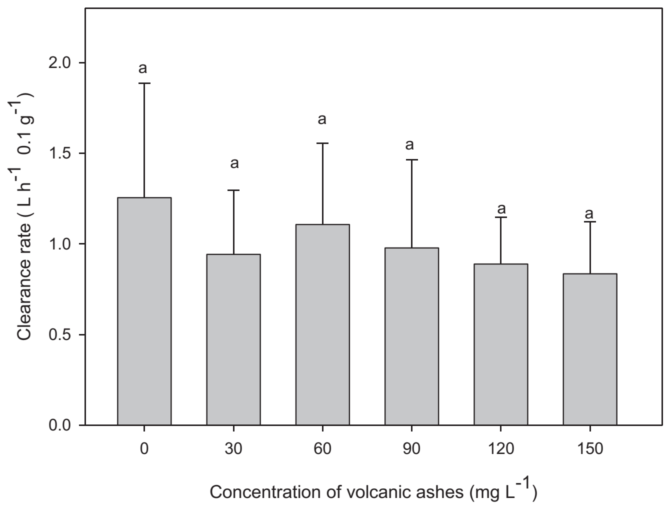

Fig. 4. *Mytilus chilensis*. Effect of volcanic ashes exposure for 15 days on the clearance rate in juveniles. Each bar represents the mean ± standard deviation per treatment (total $n = 12$ individuals per treatment). The different letters above the bars indicate significant differences ($p < 0.05$).

3.1.3. Gill lamella SEM observations

Scanning electronic microscopic analysis of gill lamellae from adult mussels did not reveal any physical damage due to the abrasiveness of the ashes (Fig. 5). However, we did identify a large amount of secreted mucus on the gills of mussels that had been exposed to diets containing 150 mg ashes L^{-1} (lamella from controls: Fig. 5a; lamella from mussels exposed to 150 mg ashes L^{-1}: Fig. 5b). In some cases, volcanic ash material was identified among the gill ciliature (Fig. 5c).

3.1.4. Variations in mussel dry tissue weight

Adult mussels showed significant weight loss during the experimental period of 15 days (Fig. 6). These declines were recorded in mussels exposed to a diet with 120 mg ashes L^{-1} (0.762 ± 0.220 g ind^{-1}; $p = 0.050$, $n = 27$) and 150 mg ashes L^{-1} (0.706 ± 0.312 g ind^{-1}; $p = 0.025$, $n = 27$). Control mussels had an average dry tissue weight of 0.849 (± 0.303) g ind^{-1} ($n = 24$).

Juveniles from all treatments after 15 days of experimentation showed a significant reduction in dry tissue weight with regard to that of the control individuals (Fig. 7; $p < 0.05$ for all treatments).

3.2. Short time feeding experiments with M. chilensis

3.2.1. Election of particles from the seston: Electivity index (EI)

The electivity index (EI) for microalgae was significantly different from zero ($p < 0.001$) for individuals feeding on the 120 mg ashes L^{-1} diet, indicating preferential retention of *I. galbana* for ingestion (Fig. 8). On the other hand, the EI's for mussels feeding on the 120 mg ashes L^{-1} diet and the 30 mg ashes L^{-1} diet were significantly different from zero ($p < 0.001$ and 0.009, respectively), indicating the deliberate rejection of these particles by the animals.

3.2.2. Clearance rate (CR) in diets containing different proportions of microalgae:ashes

One-way ANOVA showed significant differences in CR between diets ($F_{(5128)} = 4.999$, $p < 0.001$). Thus, for diets containing a higher proportion of volcanic ash, the mean CR decreased significantly in comparison to that of individuals feeding on those diets with higher proportions of microalgae (Fig. 9). The Tukey HSD test indicated that CR's were significantly lower for animals feeding on the 100% ash diet compared with values measured for individuals in the 50:50 algae-ashes treatment ($p = 0.036$) and the 100% algae treatment ($p = 0.025$). Mean CR for individuals fed the 70:30 ashes-algae

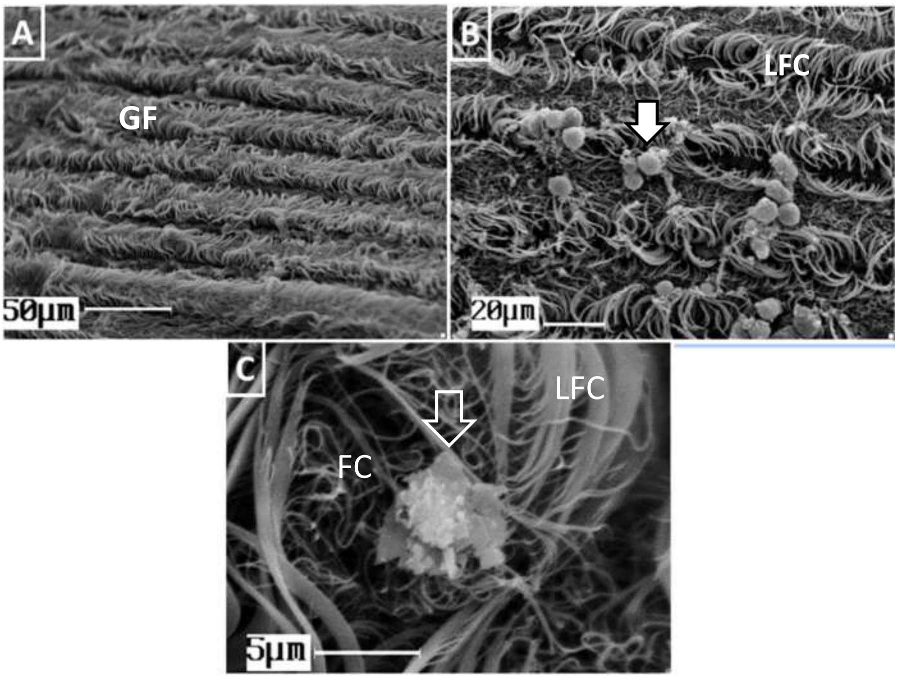

Fig. 5. *Mytilus chilensis*. SEM photomicrographs from gill lamellae of mussels A) control, B) exposed to diet of 150 mg of ash particles + microalgae, which cause the production of a large amount of mucus. (C) Presence of volcanic ash on the frontal cilia of a gill filament. Full arrow: mucous secretions. Empty arrow: ashes on the gill. LFC: lateral frontal cilia, FC: frontal cilia. GF: gill filaments.

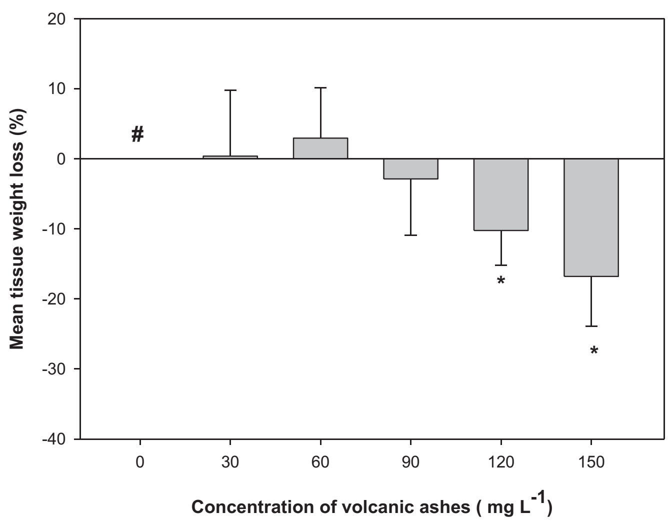

Fig. 6. *Mytilus chilensis*. The impact of suspended volcanic ash on the dry tissue weight in adults after having been exposed during 15 days to diets containing different levels of volcanic ashes. Values are expressed as a percentage of the dry tissue weight of the control mussels. Each bar represents the mean ± standard deviation per treatment (total $n = 30$ individuals per treatment). # = animal treatment controls. * = indicates significant differences from zero ($p < 0.05$).

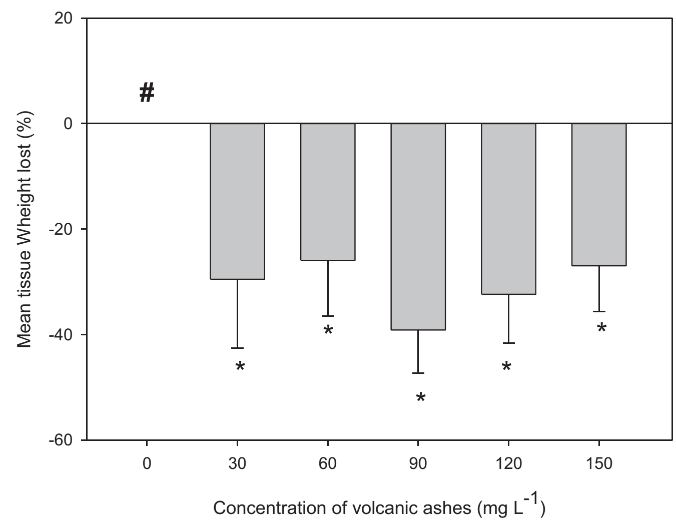

Fig. 7. *Mytilus chilensis*. Variations in dry tissue weight after juveniles had been exposed for 15 days to diets containing different levels of volcanic ash. Values are expressed as percentage of the dry tissue weight of the controls. Each bar represents the mean ± standard deviation ($n = 30$ individuals per treatment) # = animal from control. The asterisks above the bars indicate values different to zero ($p < 0.05$).

treatment was significantly lower than those recorded for individuals fed the 50:50 ashes:algae ($p = 0.013$) or 70% and 100% algae ($p = 0.009$) treatments.

4. Discussion

Our study revealed a number of detrimental impacts of volcanic ash particles in diet on the growth and physiological energetics of *M. chilensis*.

Previous researchers have shown that exposure to volcanic ash in the water column has negative effects on a variety of animals, including mortality via cuticular abrasion in arthropods (Buteler et al., 2011), almost full loss of marine benthic communities (Jewett et al., 2010), and significantly decreased fecundities in cladoceran communities (Wolinsky et al., 2013). The specific responses seem to vary with the ash concentrations to which individuals had been exposed and the size of the experimental animals or their feeding mechanisms. The small freshwater cladoceran *Daphnia commutata* showed visible damage in the stomach due to the abrasiveness of the volcanic ashes in experiments conducted using concentrations of only 5 mg ash L^{-1} (Wolinsky et al., 2013); these crustaceans appear not to have well-developed capacities for particle selection (Kirk, 1991; Jensen et al., 2001). In contrast, a number of suspension-feeding mollusk species have been found capable of pre-ingestive particle selection (e.g. *Cerastoderma edule*, Urrutia et al., 2001; *Mytilus chilensis*, Garrido et al., 2012), allowing individuals to separate the material to be ingested from that which will not be ingested. For this, mussels use principally their buccal palps, which, by producing pseudofeces, are able to eliminate some of the captured material without ingestion (Garrido et al., 2012; Navarro

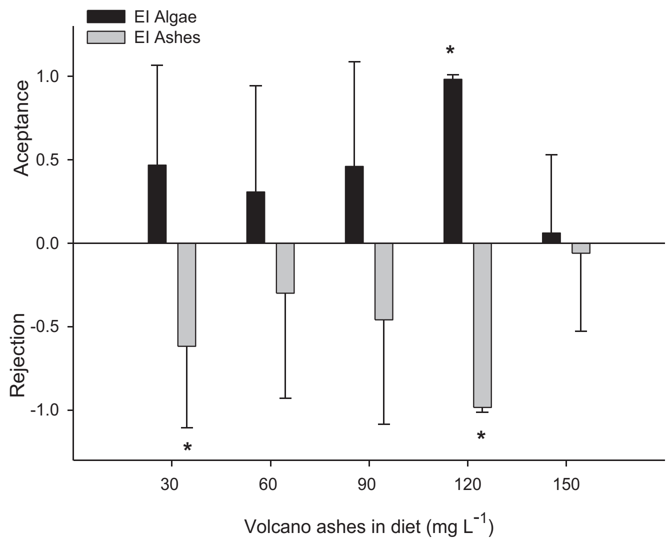

Fig. 8. *Mytilus chilensis*. Electivity index for microalgae and volcanic ashes in the gills of adult mussels exposed to diet with different proportions of algae and ash particles. Each bar represents the mean ± standard deviation per treatment. $n = 6$ per treatment. The asterisks represent significant differences of zero.

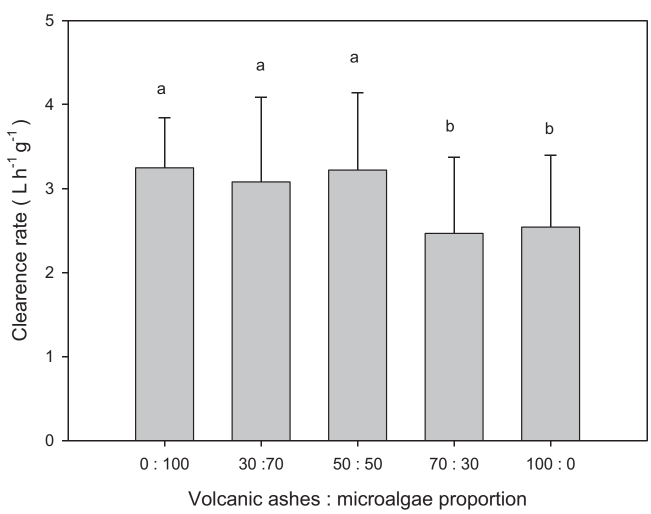

Fig. 9. *Mytilus chilensis*. Clearance rates in adult mussels fed different proportions of volcanic ash particles and microalgae. Each bar shows the mean ± standard deviation, $n = 30$ individuals for treatment. The different letters on the bars indicate significant differences between means ($p < 0.05$).

et al., 1992; Velasco and Navarro, 2002). Thus, the abrasive fraction of seston, or part thereof, does not pass through the digestive system of these mussels, which could explain why during our 15-day experiments when mussels were offered diets consisting of algae and different proportions of ash particles, no juveniles or adults died.

Contact between volcanic ash and the gills of juvenile salmon generated substantial mucus production, resulting in massive mortalities in *Oncorhynchus tshawytscha* and *Oncorhynchus nerka*, presumably through a dramatic reduction in gas exchange capacity (Newcomb and Flagg, 1983). In contrast, we identified no physical gill damage caused by volcanic ashes in our studies with *M. chilensis*. However, electronic microscopy observations showed, particularly in animals exposed to the highest concentrations of ashes, a high production of mucous secretions on the gill filaments. The increase of mucous on the gills was very well correlated with the production of abundant pseudofeces by mussels in the experimental containers. It is well known that pseudofeces production requires substantial mucous production, which the animals' palps use to eject particles to the outside, thereby avoiding their ingestion (Garrido et al., 2012).

Although none of the animals continuously exposed to ash-laden diets for 15 continuous days died, we did record substantial declines in mean mussel dry tissue weight. These losses were recorded for both adults and juveniles, and were associated with substantially reduced clearance rates and pseudofeces production. In adults, mean tissue weights decreased up to 17% for mussels exposed to a concentration of 150 mg ashes L^{-1}, while juvenile mean dry tissue weights declined up to 40% compared to that of control mussels, which were fed only on microalgae. Such substantial weight losses could become life-threatening if ash particles remained in suspension for more than the 15 day period used in our experiments. The presence of inorganic particles in the diet appears to impact the suspension-feeding process in bivalves. For example, when juveniles of the bivalve *Mya arenaria* were exposed to suspended sediment at concentrations of 100 mg L^{-1} and 200 mg L^{-1}, their growth rates were strongly reduced compared with those seen in control animals that were fed only microalgae (Grant and Thorpe, 1991). When *Cerastoderma edule* was fed with a principally inorganic diet, they produced pseudofeces that also contained high levels of inorganic material; the organic content in those pseudofeces was mainly mucus produced during suspension-feeding. The amount of energy lost in producing this pseudofecal mucus depended on both the rate of pseudofeces production and the inorganic content of the filtered material (Urrutia et al., 2001). If the amount of organic loss in the pseudofeces exceeds the energetic benefits of particle selection, resulting in a net loss of energy via excessive mucus production, it would seem more beneficial for the animals to simply reduce their suspension-feeding activity than to continue with the process of active particle selection (Urrutia et al., 2001). In general, mucus production costs in bivalve suspension-feeders have been ignored when calculating the scope for growth (Worrall et al., 1983; Bayne et al., 1987), probably because the principal component of the mucus is water (see review Davies and Hawkins, 1998). However, when substantial quantities of pseudofeces are released by suspension-feeders (Bayne and Newell, 1983; Héral et al., 1983; Davies and Hawkins, 1998), the amount of mucus in pseudofaeces is likely to be energetically costly. For example, considerable net carbon losses were recorded for both *Mytilus edulis* and *Cerastoderma edulis* when individuals were exposed to high seston concentrations, due to the substantial quantities of mucous lost through pseudofeces production (Prins and Smaal, 1989). This mucous cost due to pseudofeces production typically accompanies filtration activity in high turbidity environments (Hawkins and Bayne, 1992). The very high production of pseudofeces evidenced in our experiments is clear evidence of the high load of seston (Iglesias et al., 1992) to which individuals were exposed, and it helps to explain the substantial declines in tissue weights exhibited by the mussels in our experiments.

In our experiments, adult individuals that were exposed to the highest concentration of volcanic ashes presented significant reductions (nearly 35%, compared with control mussels) in their CR after 15 days of experimentation. Similarly, Cheung and Shin (2005) recorded a reduction in the CR and OCR in specimens of *Perna viridis*, after these mussels had been exposed for two weeks to diets containing concentration of suspended sediments as high as 1000 mg L^{-1}. Bricelj and Malouf (1984) found that increasing the sediment load in the base diet of the venerid *Mercenaria mercenaria* to 1 mg L^{-1} caused a 1.3% decrease in CR, and also resulted in a substantial increase in pseudofeces production. Similarly, the juveniles of the Chilean mussel tested in our study showed CR reductions of up to approximately 25% when feeding on all diets containing ash particles. In addition, when *M. chilensis* adults were exposed to the highest ash concentrations tested in our study, we recorded a significant decline in mean oxygen consumption rates with respect to those of the control individuals.

Field studies have also shown that high concentrations of suspended solids cause physiological changes in feeding and respiration of bivalves (Angioni et al., 2010; Grant and Thorpe, 1991). For example, exposure of *Mya arenaria* to suspended sediments reduced rates of oxygen

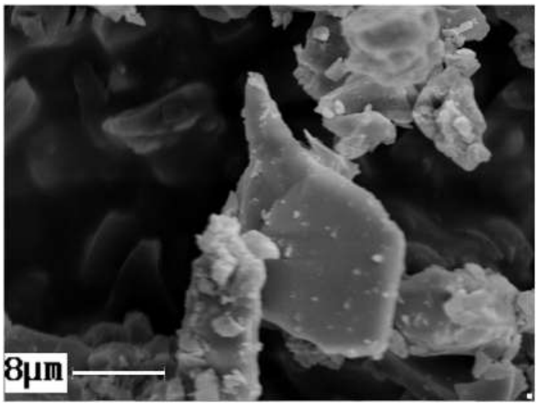

Fig. 10. SEM photomicrograph of volcanic ashes used in our experiments, resulting from the eruption of the Calbuco volcano, South of Chile, in May 2015.

consumption very quickly after the start of the exposure period when they were measured within a short time after exposure, probably reflecting the physiological stress produced by the sediment in diets (Grant and Thorpe, 1991). According to Widdows and Hawkins (1989) and Hawkins et al. (1996), decreased metabolic rates would be an expected response of the animals when dealing with foods of low quantity and/or quality. A similar energy-saving response has been reported for *Mytilus edulis* when individuals adapted their metabolic rates due to the seasonal changes in food availability (Hawkins et al., 1985). Similarly, studies on the clam *M. arenaria* show a logarithmic decrease of OCR with increasing environmental turbidity (Grant and Thorpe, 1991). In our experiments, adults of *M. chilensis* showed significant differences in mean oxygen consumption rate in the treatments with higher ash concentration with respect to those recorded for control individuals. However, we did not see this result in the juveniles of that species.

In general, volcanic ashes are reported to have a vitreous consistency and sharp edges (Caneiro et al., 2011), as confirmed by the electron microscopic observations made in this research (Fig. 10). It therefore seems reasonable to expect detrimental effects on suspension-feeding organisms once the particle concentrations exceed some level. Nevertheless, our scanning electron microscopy observations showed no physical damage to the gills of individuals exposed to diets containing volcanic ash. Although we found no evidence for any physical damage to the gills, we did observe volcanic ash particles among the ciliature, so there is still a possibility of damage from the abrasive effect of the ashes during the filtration process, something that should be examined in future research. Experiments with specimens of the green-lipped mussel *Perna viridis* that were exposed to suspended sediment for 14 days showed that an increase in concentration and in the size of the particles in diet did more damage to the gills, especially on the frontal ciliature of the lamellae. Also, the same authors recorded a depletion of the cilia from ascending and descending lamellae, which appears to be a consequence of the mechanical damage generated by direct contact of the sediments with the front of the lamellar surface (Cheung and Shin, 2005). The extent of damage was also strongly related to sediment particle size, with the largest particles (250 to 500 μm) causing a far greater depletion of frontal cilia than particles of the smallest sizes (particles < 63 μm) (Cheung and Shin, 2005). In our study, when individuals of *M. chilensis* were directly exposed to diets containing different proportions of volcanic ashes and microalgae, CR decreased significantly when ashes constituted over 70% of the diet. In *M. edulis* (Bayne et al., 1989; Bayne et al., 1993; Beninger and St-Jean, 1997a; Hawkins et al., 1996; Widdows et al., 1979) and other molluscs (Cranford and Gordon, 1992; Grant and Thorpe, 1991; Iglesias et al., 1992; MacDonald and Ward, 1994; Cucci et al., 1985; Velasco and Navarro, 2002), CR decreased substantially when the animals were exposed to high concentrations of inorganic material in the diet, possibly reflecting a strategy of reducing energy costs by reducing the production of pseudofeces (Velasco and Navarro, 2002).

Particle selection by suspension-feeding mollusks can optimize rates of food and energy intake (Widdows et al., 1979; Shumway et al., 1997; Espinosa et al., 2010; Iglesias et al., 1992). For example, some bivalves can discriminate between different species of microalgae (Shumway et al., 1997), while others can detect differences in particle organic content (Bayne et al., 1993; Kiørboe and Møhlenberg, 1981; Levinton et al., 2002; MacDonald and Ward, 1994; Newell and Jordan, 1983; Ward et al., 1997). In the present research, we identified clear feeding selectivity, with volcanic ash particles being preferably chosen for rejection as pseudofeces and the microalgae preferably chosen for ingestion. The mechanism for such selection is still unclear. In suspension-feeder bivalves the physicochemical properties of particles appear to be an important factor in the selection process. Newell and Jordan (1983) hypothesized that particle selection is achieved using a chemosensory mechanism that dictates particle retention. For example, with the bivalves *Mytilus edulis* and *Crassostrea virginica* feeding on different kind of microalgae, lectin affinity and wettability (= contact angle) were the strongest predictors of particle selection preferences (Rosa et al., 2017). In concordance with the above, particle selection and rejection by suspension-feeding bivalves seems to involve a specific interaction between carbohydrates on the particle surfaces and lectins within the mucus (Espinosa et al., 2009). Clearly the mechanism employed in capturing and selecting particles made possible the capacity to do selection between the algae *I. galbana* and the volcanic ashes used in our *M. chilensis* experiments. Based on the above, it seems likely that certain physicochemical characteristics of suspended particles can account for the particle selection and rejection that we have identified in our experiments on *M. chilensis*. Increased concentrations of volcanic ash in the diet tended to generate a greater electivity for microalgae. The gastropod *Crepipatella dilatata* behaves similarly, in that a decrease in the relative proportion of microalgae to inorganic particles in suspension leads to relatively higher proportions of microalgae being retained on the gills for ingestion (Chaparro et al., 2013). In the case of *Cerastoderma edule*, physiological sorting and selection mechanism have been found to be especially effective at seston concentrations of $< 100\ mg\ L^{-1}$; and this selection ability is strongly reduced when seston concentrations are higher (Navarro and Widdows, 1997), a situation that is consistent with our findings for *M. chilensis*.

Many species have mechanisms that help them cope with environmental changes that may affect them negatively, and thus cease to be physiological slaves of the environment (Jørgensen, 1990). In summary, in our experiments, the presence of substantial volcanic ash content in the diet was not a lethal stressor for the mussel *M. chilensis*, at least during the 15-day duration of our study. However, there was a clear negative impact on the individuals tested, in that oxygen consumption and clearance rates, and especially tissue weights, were substantially reduced, especially at the highest ash concentrations tested. Ashes did not seem to directly damage the gills in our study, despite the sharp edges and abrasiveness of these particles, but their presence in the diet forced the mussels to actively select particles, resulting in a high production of pseudofeces and a lower overall energy intake; this translates into high energy costs associated with the generation of mucus used in forming the pseudofeces and the rejection of this material to the surrounding seawater. The potential for longer exposure times than the 15-day period tested in this study to result in substantial mortality of juveniles and adults of this species remains to be examined; newly metamorphosed juveniles may be especially susceptible, as they may have a less developed capacity to manage and select particles during feeding, as documented in *Perna canaliculus* (Gui et al., 2016).

Supplementary data to this article can be found online at https://doi.org/10.1016/j.marpolbul.2017.12.024.

Acknowledgements

This work was supported by the Fondo Nacional de Investigación Científica y Tecnológica, Fondecyt-Chile by the grant 1141052.

References

Acuña, K., Duarte, C., Jaramillo, E., Contreras, H., Manzano, M., Navarro, J.M., 2012. Distribución vertical de la macroinfauna asociada a bivalvos en una planicie intermareal sedimentaria del sur de Chile. Rev. Biol. Mar. Oceanogr. 47, 383–393.

Akberali, H.B., Trueman, E.R., 1985. Effects of environmental stress on marine bivalve molluscs. Adv. Mar. Biol. 22, 101–198.

Angioni, S.A., Giansante, C., Ferri, N., 2010. The clam (*Chamelea gallina*): evaluation of the effects of solids suspended in seawater on bivalve molluscs. Vet. Ital. 46, 101–106.

Baker, S.M., Levinton, J.S., 2003. Selective feeding by three native north American freshwater mussels implies food competition with zebra mussels. Hydrobiologia 505, 97–105.

Balseiro, E., Souza, M.S., Olabuenaga, I.S., Wolinski, L., Navarro, M.B., Laspoumaderes, C., Modenutti, B., 2014. Effect of the Puyehue-Cordon Caulle volcanic complex eruption on crustacean zooplankton of Andean lakes. Ecol. Austral 24, 75–82.

Bayne, B.L., Newell, R.C., 1983. Physiological energetics of marine molluscs. In: Saleuddin, A.S.M., Wilbur, K.M. (Eds.), The Mollusca. Physiology, Part 1 4. Academic Press, Toronto, pp. 407–515.

Bayne, B.L., Thompson, R.J., 1970. Some physiological consequences of keeping *Mytilus edulis* in the laboratory. Helgoländer Meeresun. 20, 526–552.

Bayne, B.L., Widdows, J., 1978. The physiological ecology of two populations of *Mytilus edulis* L. Oecologia 37, 137–162.

Bayne, B.L., Hawkins, A.J.S., Navarro, E., 1987. Feeding and digestion by the mussel *Mytilus edulis* L. (Bivalvia: Mollusca) in mixtures of silt and algal cells at low concentrations. J. Exp. Mar. Biol. Ecol. 111, 1–22.

Bayne, B.L., Hawkins, A.J.S., Navarro, E., Iglesias, I.P., 1989. Effects of seston concentration on feeding, digestion and growth in the mussel *Mytilus edulis*. Mar. Ecol. Prog. Ser. 55, 47–54.

Bayne, B.L., Iglesias, J.I.P., Hawkins, A.J.S., Navarro, E., Heral, M., Deslous-Paoli, J.M., 1993. Feeding behavior of the mussel, *Mytilus edulis*: responses to variations in quantity and organic content of the seston. J. Mar. Biol. Assoc. UK 73, 813–829.

Beninger, P.G., Dufour, S.C., 1996. Mucocyte distribution and relationship to particle transport on the pseudolamellibranch gill of *Crassostrea virginica* (Bivalvia: Ostreidae). Mar. Ecol. Prog. Ser. 137, 133–138.

Beninger, P.G., St-Jean, S.D., 1997a. The role of mucus in particle processing by suspension-feeding marine bivalves: unifying principles. Mar. Biol. 129, 389–397.

Beninger, P.G., St-Jean, S.D., 1997b. Particle processing on the labial palps of *Mytilus edulis* and *Placopecten magellanicus* (Mollusca: Bivalvia). Mar. Ecol. Prog. Ser. 147, 117–127.

Beninger, P.G., Veniot, A., Poussart, Y., 1999. Principles of pseudofeces rejection on the bivalve mantle: integration in particle processing. Mar. Ecol. Prog. Ser. 178, 259–269.

Beninger, P.G., Decottignies, P., Guiheneuf, F., Barillé, L., Rincé, Y., 2007. Comparison of particle processing by two introduced suspension feeders: selection in *Crepidula fornicata* and *Crassostrea gigas*. Mar. Ecol. Prog. Ser. 334, 165–177.

Bougrier, S., Hawkins, A.J.S., Héral, M., 1997. Preingestive selection of different microalgal mixtures in *Crassostrea gigas* and *Mytilus edulis*, analysed by flow cytometry. Aquaculture 150, 123–134.

Bricelj, V.M., Malouf, R.E., 1984. Influence of algal and suspended sediment concentrations on the feeding physiology of the hard clam *Mercenaria mercenaria*. Mar. Biol. 84, 155–165.

Buteler, M., Stadler, T., López-García, G.P., Lassa, M.S., 2011. Insecticidal properties of ashes from the volcanic complex Puyehue-Caulle range and their possible environmental impact. Rev. Soc. Entomol. Argent. 70, 149–156.

Caneiro, A., Mogni, L., Serquis, A., Cotaro, C., Wilberger, D., Ayala, C., Daga, R., Poire, D., Scerbo, E., 2011. Análisis de cenizas volcánicas del Cordón Caulle (Complejo volcánico Puyehue-Cordón Caulle). In: Comisión Nacional de Energía Atómica, Informe Cenizas Volcanicas. 11.

Chaparro, O.R., Montory, J.A., Pereda, S.V., Thompson, R.J., Rivera, G., Osores, S.J.A., Segura, C.J., 2013. Particle selection and regulation of particle uptake by the slipper limpet *Crepipatella fecunda*. Mar. Ecol. Prog. Ser. 474, 167–177.

Cheung, S.G., Shin, P.K.S., 2005. Size effects of suspended particles on gill damage in green-lipped mussel *Perna viridis*. Mar. Pollut. Bull. 51, 801–810.

Coughlan, J., 1969. The estimation of filtering rate from clearance of suspensions. Mar. Biol. 2, 356–358.

Cranford, P.J., Gordon, Jr D.C., 1992. The influence of dilute clay suspensions on sea scallop (*Placopecten magellanicus*) feeding activity and tissue growth. Neth. J. Sea Res. 30, 107–120.

Cucci, T.L., Shumway, S.E., Newell, R.C., Selvin, R., Guillard, R.R.L., Yentsch, C.M., 1985. Flow cytometry: a new method for characterization of differential ingestion, digestion and egestion by suspension feeders. Mar. Ecol. Prog. Ser. 24, 201–204.

Davies, M.S., Hawkins, S.J., 1998. Mucus from marine molluscs. Adv. Mar. Biol. 34, 1–71.

Delgado, F., Pritchard, M.E., Ebmeier, S., Gonzalez, P., Lara, L., 2017. Recent unrest (2002–2015) imaged by space geodesy at the highest risk Chilean volcanoes: Villarrica, Llaima, and Calbuco (southern Andes). J. Volcanol. Geotherm. Res. 344, 270–288.

Durant, A.J., Villarosa, G., Rose, W.I., Delmelle, P., Prata, A.J., Viramonte, J.G., 2012. Long-range volcanic ash transport and fallout during the 2008 eruption of Chaitén volcano. Chile. Phys. Chem. Earth. Parts A/B/C 45–46, 50–64.

Espinosa, E.P., Perrigault, M., Ward, J.E., Shumway, S.E., Allam, B., 2009. Lectins associated with the feeding organs of the oyster *Crassostrea virginica* can mediate particle selection. Biol. Bull. 217, 130–141.

Espinosa, E.P., Perrigault, M., Ward, J.E., Shumway, S.E., Allam, B., 2010. Microalgal cell surface carbohydrates as recognition sites for particle sorting in suspension-feeding bivalves. Biol. Bull. 218, 75–86.

Garrido, M.V., Chaparro, O.R., Thompson, R.J., Garrido, O., Navarro, J.M., 2012. Particle sorting and formation and elimination of pseudofaeces in the bivalves *Mulinia edulis* (siphonate) and *Mytilus chilensis* (asiphonate). Mar. Biol. 159, 987–1000.

Grant, J., Thorpe, B., 1991. Effects of suspended sediment on growth, respiration, and excretion of the soft-shell clam (*Mya arenaria*). Can. J. Fish. Aquat. Sci. 48, 1285–1292.

Gui, Y., Kaspar, H.F., Zamora, L.N., Dunphy, B.J., Jeffs, A.G., 2016. Capture efficiency of artificial food particles of post-settlement juveniles of the Greenshell™ mussel, *Perna canaliculus*. Aquaculture 464, 1–7.

Hawkins, A.J.S., Bayne, B.L., 1992. Physiological interrelations, and the regulation of production. In: Gosling, E. (Ed.), The Mussel *Mytilus*: Ecology, Physiology, Genetics and Culture. Elsevier, Amsterdam, pp. 171–222.

Hawkins, A.J.S., Salkeld, P.N., Bayne, B.L., Gnaiger, E., Lowe, D.M., 1985. Feeding and resource allocation in the mussel *Mytilus edulis*: evidence for time-averaged optimization. Mar. Ecol. Prog. Ser. 20, 273–287.

Hawkins, A.J.S., Smith, R.F.M., Bayne, B.L., Heral, M., 1996. Novel observations underlying the fast growth of suspension-feeding shellfish in turbid environments: *Mytilus edulis*. Mar. Ecol. Prog. Ser. 131, 179–190.

Héral, M., Deslous-Paoli, J.-M., Sornin, J.-M., 1983. Transferts énergétiques entre l'huître *Crassostrea gigas* et la nourriture potentielle disponible dans un bassin ostréicole: premiéres approches. Oceanis 9, 169–194.

Hirano, T., Kiyota, M., Aiga, I., 1995. Physical effects of dust on leaf physiology of cucumberr and kidney bean plants. Environ. Pollut. 89, 255–261.

Horwell, C., Baxter, O.J., 2006. The respiratory health hazard of volcanic ash: a review for volcanic risk mitigation. Bull. Volcanol. 69, 1–24.

Hutchinson, S., Hawkins, L.E., 1992. Quantification of the physiological responses of the European flat oyster *Ostrea edulis* L. to temperature and salinity. J. Molluscan Stud. 58, 215–226.

Iglesias, J.I.P., Navarro, E., Jorna, P.A., Armentina, I., 1992. Feeding, particle selection and absorption in cockles *Cerastoderma edule* (L.) exposed to variable conditions of food concentration and quality. J. Exp. Mar. Biol. Ecol. 162, 177–198.

Irlandi, E., Maciá, S., Serafy, J., 1997. Salinity reduction from freshwater canal discharge: effects on mortality and feeding of an urchin (*Lytechinus variegatus*) and a gastropod (*Lithopoma tectum*). Bull. Mar. Sci. 61, 869–879.

Jacobs, J., 1974. Quantitative measurement of food selection. Oecologia 14, 413–417.

Jensen, T.C., Hessen, D.O., Faafeng, B.A., 2001. Biotic and abiotic preferences of the cladoceran invader *Limnosida frontosa*. Hydrobiologia 442, 89–99.

Jewett, S.C., Bodkin, J.L., Chenelot, H., Esslinger, G.G., Hoberg, M.K., 2010. The nearshore benthic community of Kasatochi Island, one year after the 2008 volcanic eruption. Arct. Antarct. Alp. Res. 42, 315–324.

Jørgensen, C.B., 1990. Bivalve Filter Feeding: Hydrodynamics, Bioenergetics, Physiology and Ecology. Fredensborg: Olsen and Fredensborg, Denmark (140pp.).

Jørgensen, C.B., 1996. Bivalve filter feeding revisited. Mar. Ecol. Prog. Ser. 142, 287–302.

Kiørboe, T., Møhlenberg, F., 1981. Particle selection in suspension-feeding bivalves. Mar. Ecol. Prog. Ser. 5, 291–296.

Kiørboe, T., Møhlenberg, F., Nøhr, O., 1980. Feeding, particle selection and carbon absorption in *Mytilus edulis* in different mixtures of algae and resuspended bottom material. Ophelia 19, 193–205.

Kirk, K.L., 1991. Inorganic particles alter competition in grazing plankton - the role of selective feeding. Ecology 72, 915–923.

Klűser, L., Erbertsede, T., Meyer-Arnek, J., 2013. Observation of volcanic ash from Puyehue-Cordon Caulle with IASI. Atmos. Meas. Tech. 6, 35–46.

Levinton, J.S., Ward, J.E., Shumway, S.E., 2002. Feeding responses of the bivalves *Crassostrea gigas* and *Mytilus trossulus* to chemical composition of fresh and aged kelp detritus. Mar. Biol. 141, 367–376.

MacDonald, B.A., Ward, J.E., 1994. Variation in food quality and particle selectivity in the sea scallop *Placopecten magellanicus* (Mollusca: Bivalvia). Mar. Ecol. Prog. Ser. 108, 251–264.

Navarro, J.M., Chaparro, O.R., 2002. Grazing–filtration as feeding mechanisms in motile specimens of *Crepidula fecunda* (Gastropoda: Calyptraeidae). J. Exp. Mar. Biol. Ecol. 270, 111–122.

Navarro, J., Velasco, L., 2002. Feeding physiology of infaunal (*Mulinia edulis*) and epifaunal (*Mytilus chilensis*) bivalves under a wide range of concentrations and qualities of seston. Mar. Ecol. Prog. Ser. 240, 143–155.

Navarro, J.M., Widdows, J., 1997. Feeding physiology of *Cerastoderma edule* in response to a wide range of seston concentrations. Mar. Ecol. Prog. Ser. 152, 175–186.

Navarro, J.M., Winter, J.E., 1982. Ingestion rate, assimilation efficiency and energy balance in *Mytilus chilensis* in relation to body size at different algal concentrations. Mar. Biol. 67, 255–266.

Navarro, E., Iglesias, J.I.P., Ortega, M.M., 1992. Natural sediment as food source for the cockle *Cerastoderma edule* (L.): effect of variable particle concentration on feeding, digestion and the scope for growth. J. Exp. Mar. Biol. Ecol. 156, 69–87.

Navarro, J.M., Gonzalez, L.P., Chaparro, O.R., 2011. Filtering capacity and endoscopic analysis of sympatric infaunal and epifaunal bivalves of southern Chile. J. Mar. Biol. Assoc. UK 91, 1607–1612.

Newcomb, T.T., Flagg, T.A., 1983. Some of effect of Mt. Helens volcanic ash on juvenile salmon smolt. Mar. Fish. Rev. 45, 8–12.

Newell, R.I.E., Jordan, S.J., 1983. Preferential ingestion of organic material by the American oyster *Crassostrea virginica*. Mar. Ecol. Prog. Ser. 13, 47–53.

Newell, C.R., Wildish, D.J., MacDonald, B.A., 2001. The effects of velocity and seston

concentration on the exhalant siphon area, valve gape and filtration rate of the mussel *Mytilus edulis*. J. Exp. Mar. Biol. Ecol. 262, 91–111.
Prins, T.C., Smaal, A.C., 1989. Carbon and nitrogen budgets of the mussel *Mytilus edulis* L. and the cockle *Cerastoderma edule* (L.) in relation to food quality. Sci. Mar. 53, 477–482.
Rosa, M., Ward, J.E., Shumway, S.E., Wikfors, G.H., Pales-Espinosa, E., Allam, B., 2013. Effects of particle surface properties on feeding selectivity in the eastern oyster *Crassostrea virginica* and the blue mussel *Mytilus edulis*. J. Exp. Mar. Biol. Ecol. 446, 320–327.
Rosa, M., Ward, J.E., Holohan, B.A., Shumway, S.E., Wikfors, G.H., 2017. Physicochemical surface properties of microalgae and their combined effects on particle selection by suspension-feeding bivalve molluscs. J. Exp. Mar. Biol. Ecol. 486, 59–68.
Self, S., 2006. The effects and consequences of very large explosive volcanic eruptions. Philos. Trans. R. Soc. A Math. Phys. Eng. Sci. 364, 2073–2097.
SERNAPESCA, 2015. Informe: Actividades de Fiscalización Efectuadas en el año 2014. Servicio Nacional de Pesca y Acuicultura, Ministerio de Economía, Fomento y Turismo, Valparaiso, Chile, pp. 13–14 Páginas.
Shirakawa, M., Fukushima, R., Kyushima, K., 1984. Experimental studies on the effects of Mt. Sakurajima volcanic ashes on the respiratory organs. Jpn J Ind. Health 26, 130–146.
Shumway, S.E., Cucci, T.L., Lesser, M.P., Bourne, N., Bunting, B., 1997. Particle clearance and selection in three species of juvenile scallops. Aquac. Int. 5, 89–99.
Urrutia, M.B., Navarro, E., Ibarrola, I., Iglesias, J.I.P., 2001. Preingestive selection processes in the cockle *Cerastoderma edule*: mucus production related to rejection of pseudofaeces. Mar. Ecol. Prog. Ser. 209, 177–187.
Velasco, L.A., Navarro, J.M., 2002. Feeding physiology of infaunal (*Mulinia edulis*) and epifaunal (*Mytilus chilensis*) bivalves under a wide range of concentrations and qualities of seston. Mar. Ecol. Prog. Ser. 240, 143–155.
Vernier, J.P., Fairlie, T.D., Murray, J.J., Tupper, A., Trepte, C., Winker, D., Pelon, J., Garnier, A., Jumelet, J., Pavolonis, M., Omar, A.H., Powell, K.A., 2013. An advanced system to monitor the 3D structure of diffuse volcanic ash clouds. J. Appl. Meteorol. Climatol. 52, 2125–2138.
Vidal, L., Nesbitt, S.W., Salio, P., Farias, C., Nicora, M.G., Osores, M.S., Mereu, L., Marzano, F.S., 2017. C-band Dual-Polarization Radar Observations of a Massive Volcanic Eruption in South America. IEEE J. Sel. Topics Appl. Earth Observ. Remote. Sens. 10, 960–974.
Ward, J.E., Shumway, S.E., 2004. Separating the grain from the chaff: particle selection in suspension- and deposit-feeding bivalves. J. Exp. Mar. Biol. Ecol. 300, 83–130.
Ward, J.E., Levinton, J.S., Shumway, S.E., Cucci, T., 1997. Site of particle selection in a bivalve mollusc. Nature 390, 131–132.
Widdows, J., Hawkins, A.J.S., 1989. Partitioning of rate of heat dissipation by *Mytilus edulis* into maintenance, feeding, and growth components. Physiol. Zool. 62, 764–784.
Widdows, J., Fieth, P., Worrall, C.M., 1979. Relationships between seston, available food and feeding activity in the common mussel *Mytilus edulis*. Mar. Biol. 50, 195–207.
Wolinsky, L., Laspoumaderes, C., Navarro, M.B., Modenutti, B., Balseiro, E., 2013. The susceptibility of cladocerans in north Andean Patagonian lakes to volcanic ashes. Fresh. Wat. Biol. 58, 1878–1888.
Worrall, C.M., Widdows, J., Lowe, D.M., 1983. Physiological ecology of three populations of the bivalve *Scrobicularia plana*. Mar. Ecol. Prog. Ser. 12, 267–279.