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Role of physico-chemical environment on gastropod assemblages at hydrothermal vents on the East Pacific Rise (13°N/EPR)

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Abstract:

Deep-sea hydrothermal vents display extreme and highly variable environmental conditions that are expected to be among the most important factors structuring associated benthic populations and communities. We tested this assumption, focusing on the distribution of gastropods, as well as on the demographic population structure and reproductive biology of one dominant gastropod species in zones characterized by alvinellid polychaetes and vestimentiferan tubeworms. A total of 14 biological samples from both types of habitats were collected at three sites on the East Pacific Rise 13°N vent field in May 2002. At all vents except one, the physico-chemical environment was described in two steps: (1) pH, total sulphide and reduced iron concentrations have been measured *in situ* in *Alvinella* habitats and correlations to temperature were assessed at the scale of each sampled vent; and (2) assuming the consistency of these relationships within a single edifice, ranges of physico-chemical factors were estimated for each biological sample from the corresponding fine scale temperature measurements. A total of 11 gastropod species were identified from all samples and 2 main faunal assemblages were distinguished: one dominated by *Lepetodrilus elevatus* in the alvinellid zone as well as in the vestimentiferan zone, and one dominated by the peltospirids *Nodopelta heminoda, N. subnoda* and *Peltospira operculata* confined to the alvinellid zone. Peltospirid gastropods were dominant over lepetodrilid gastropods in the more acidic, sulphide-richer, and hotter environments. Although this pattern could be related to specific physiological tolerances to temperature and sulphide toxicity, the weak correlation between community structure and physico-chemical variables suggests that additional factors are also involved. Particularly, the low species richness and the overwhelming dominance of *L. elevatus* in one faunal assemblage suggest that this species may outcompete peltospirids and greatly affect community structure. This hypothesis is supported by large differences in the demographic structure and reproductive biology of *L. elevatus* between the 2 faunal assemblages.

Keywords: physico-chemical environment; gastropod assemblages; hydrothermal vents; East Pacific Rise

INTRODUCTION

Deep-sea hydrothermal vents host highly productive communities fueled primarily by chemosynthetic microbial production and characterized by low species richness but high proportion of endemic species, and a large biomass in contrast with the surrounding deep-sea floor. The limited lifespan of vent sites, related to sea-floor spreading rate and frequent volcanic eruptions, leads to the creation of ephemeral and transient habitats, patchily distributed around vent sites separated from 10s of meters to 100s of kilometers. Within a single vent site, the high spatio-temporal variability of environmental conditions in terms of temperature, pH, and concentrations of oxygen, sulphide and metals (Johnson et al., 1994; Sarradin et al., 1998; Le Bris et al., 2003, 2006b) produces a mosaic of habitats with contrasted biological characteristics (Sarrazin et al., 1997; Shank et al., 1998; Sarrazin & Juniper, 1999). Organisms are distributed in different assemblages around the vent, and their composition varies in space and time in relation with the decreasing gradient of fluid exposure, the physical structure of the mineral substrate and the temporal dynamics of vent colonization (Sarrazin et al., 1997; Shank et al., 1998). On the East Pacific Rise (EPR), 4 main megafaunal assemblages have been described: (i) alvinellid polychaete colonies restricted to the most active areas of chimney walls with high-temperature emissions, (ii) vestimentiferan tubeworm assemblages in recent and active diffuse flow areas, (iii) bivalve assemblages in moderate and older diffuse flow areas and (iv) suspension-feeder assemblages dominated by serpulid polychaetes and barnacles at the periphery of vents in seawater with little or no hydrothermal influence (Jollivet, 1996; Shank et al., 1998).

The influence of environmental factors in shaping hydrothermal communities appears quite complex, and some non-exclusive hypotheses are still debated. According to the correspondence between physico-chemical gradients and faunal zonation, several ecological studies have related the variability of faunal composition in space and time to changes in

environmental conditions, putting much emphasis on physiological tolerances and nutritional requirements of organisms (Sarrazin et al., 1997; Shank et al., 1998). In this context, two physico-chemical parameters were commonly referred to as potentially determinant: temperature (Bates et al., 2005; Mills et al., 2007), and sulphide which is both a major electron donor for chemoautotrophic microbes and a potent poison for aerobic organisms (Childress & Fisher, 1992). However, more than total sulphide concentration, it was pointed out that differences in chemical speciation of sulphide among habitats may be a key-factor driving the distribution of species (Luther et al., 2001). Although total acid-volatile sulphide concentrations ($H_2S + HS^- + FeS(aq)$) was shown to be at least 5 times higher in an *Alvinella pompejana* colony than in a *Riftia pachyptila* clump, these authors found that FeS(aq) was the dominant sulphide phase in the former habitat while free sulphide ($H_2S + HS^-$) was the major form in the latter. FeS formation was therefore proposed to act as a sulphide detoxification mechanism in *Alvinella* colonies. The importance of this process depends largely on the dissolved iron to sulphide ratio which is known to be highly variable among habitats (Le Bris & Gaill, 2007; Le Bris et al., 2006a), resulting of end-member fluid composition variability in space and time (Von Damm & Lilley, 2004). Conversely, Govenar et al. (2005) showed that the structure and composition of the epifaunal community associated with different *R. pachyptila* clumps were remarkably similar between sites and independent of sulphide and iron concentrations. Likewise, Mullineaux et al. (2000) suggested that the settlement of the two vestimentiferan species, *R. pachyptila* and *Oasisia alvinae*, was independent of tolerances to physico-chemical conditions but rather facilitated by the occurrence of *Tevnia jerichonana*. While biotic interactions between organisms (i.e. facilitation, competition, predation) could be major determinants of community structure (Micheli et al., 2002), they were shown to vary along the gradient of flow intensity with facilitation processes occurring at the periphery of vents, where animal density is lower, and inhibition processes occurring in the high diffuse

vent flow areas (Mullineaux et al., 2003). More recently, Mills et al. (2007) suggested that most hydrothermal gastropod species are not exclusive to a specific megafaunal zone as they may occupy specific microhabitats.

In other respects, physico-chemical conditions could also play a significant role on population dynamics of deep-sea hydrothermal species (e.g. growth, survivorship, reproduction) although very few studies addressed directly these questions (Mullineaux et al., 1998; Copley et al., 2003; Kelly & Metaxas, 2007). While reproductive cycles of lepetodrilids were described to be quasi-continuous within females and asynchronous among females (Pendlebury, 2004), Kelly & Metaxas (2007) reported that the rate of gametogenesis of *Lepetodrilus fucensis* could vary between habitat types. By controlling the abundance and the turnover of local populations, one can expect that these spatial variations in biological features may affect the intensity of biological interactions and the structure of benthic communities.

On hydrothermal vents of the East Pacific Rise, the alvinellid polychaetes *A. pompejana* and *A. caudata* inhabit the surface of active sulphide structures, presumably in the most hypoxic and toxic conditions found in these environments (Le Bris & Gaill, 2007), and are known as the very first macrofaunal colonizers of new chimney habitats (Pradillon et al., 2005b; Taylor et al., 1999). In these extreme environmental conditions, biological adaptations and community structure are expected to be mostly driven by biogeochemical processes (Luther et al., 2001). However, most studies on processes involved in hydrothermal community structure were conducted on mussel beds or vestimentiferan clumps (Micheli et al., 2002; Mullineaux et al., 2003; Tsurimi & Tunnicliffe, 2003; Van Dover, 2003; Govenar et al., 2004; Dreyer et al., 2005) and faunal distribution in *Alvinella* colonies remains poorly known (Desbruyères et al., 1998). Focusing on gastropods, which represent a major part of vent fauna in terms of density and diversity in different habitats (Jollivet, 1996; Mills et al.,

2007) the aims of the present study were: (1) to better characterize the physico-chemical variability of habitats at the surface of *A. pompejana* colonies from the 13°N-EPR vent field, from the assessment of, both, fine scale temperature ranges and correlations of temperature with chemical factors that were measured *in situ*; pH, sulphide and iron concentrations ranges were thus determined over spatial scales as relevant as possible to the conditions experienced by gastropods; (2) to relate these environmental ranges to the variability in the composition of faunal assemblages at different scales; and (3) to assess the relationships between population biology and environmental conditions with *Lepetodrilus elevatus* as an example. This species was chosen as it displayed a wide distribution in hydrothermal habitats (Mills et al., 2007) and was reported to be a highly competitive species in hydrothermal faunal assemblages (Micheli et al., 2002). Additional samples from *R. pachyptila* clumps were also analysed for comparison with *Alvinella* habitats.

MATERIALS AND METHODS

Study sites

All physico-chemical and biological data were collected using the ROV Victor 6000 during the PHARE cruise carried out in May 2002 on the 13°N vent field along the East Pacific Rise (EPR). A hierarchical sampling method involving three spatial scales was undertaken for biological sampling (Table 1): (1) three vent sites, Genesis, Parigo and Elsa located at a depth of ~ 2620 m and spaced by 100s of meters, (2) two active sulphide structures (here referred to as ‘vents’) within each site at a scale of 10s of meters, and (3) one to six samples within each structure at a scale of meters. On the Genesis site, the PP12 vent is a small diffuser colonized by dense colonies of *Alvinella pompejana* at the top and *Riftia pachyptila* clumps around. On the opposite, the Hot 2 vent consisted in a large diffuse flow area on the side of a cliff with dense colonies of *R. pachyptila* and no *A. pompejana* colony

present. The Parigo site was composed of three sulphide edifices including two small diffusers and one tall chimney. The PP-Ph05(1) diffuser is covered by dense *A. pompejana* colonies largely mixed with *R. pachyptila* tubes while the PP-Ph05(2) vent is a high chimney with large colonies of *A. pompejana* or uncolonized surfaces on the upper half and *R. pachyptila* clumps on its basis. On the Elsa site, PP-Ph01 vent is a massive black smoker inhabited by large colonies of *A. pompejana*, at the top and intermediate height, and the occurrence of large uncolonized surfaces. The Hot 3 vent is a 3 meter diameter white smoker with dense colonies of *A. pompejana* at the top and *R. pachyptila* clumps around (Pradillon et al., 2005a).

Chemical analysis and definition of environmental parameters

In a first step, extensive *in situ* measurements of temperature, pH, total sulphide and reduced iron in the environment surrounding the gastropods, were conducted for each vent during different dives, using the submersible flow analyser Alchimist (AnaLyseur CHIMIque In SiTu) combined to temperature and pH probes (Le Bris et al., 2000). Colorimetric flow injection analysis (FIA) methods were used to detect the most labile fraction of acid volatile sulphide S(-II) and labile forms of ferrous iron, Fe(II). S(-II) includes free sulphide forms, aqueous iron sulphide forms and freshly precipitated iron sulphide colloids (i.e. HS^-, H_2S, FeS(aq), $Fe(HS)^+$, FeS(am)) (Le Bris et al., 2003). *In situ* pH measurements were made using an autonomous deep-sea sensor (MICREL, France) equipped with a combined glass electrode and a miniaturized thermocouple that was specifically designed for these hydrothermal environments (Le Bris et al., 2001). Due to logistical constraints, no chemical data was available for the *Riftia* zone at Genesis Hot 2 vent which was therefore excluded from the analysis on the relationships between biological and environmental data.

For each vent, this first step allowed to assess the relationships between temperature and sulphide, iron, and pH respectively, assuming the conservative mixing of a local hydrothermal fluid and seawater for a single alvinellid aggregation. This conservative mixing assumption was shown to be consistent for the water layer at the surface of an alvinellid colony in the vicinity of a local source (Le Bris et al., 2005). Hence, iron and sulphide concentrations could be estimated for each sample using the linear correlations established for the corresponding hydrothermal structure. Similarly, pH was estimated from empirical logarithmic correlations. In a second step, just before biological sampling, time-series measurements of temperature over duration ranging from 1 to 50 min were performed at 1 to 5 sampling locations in the biological sampling area using the Pt100 temperature probe of the ROV Victor 6000 (Table 1). To ensure that they are representative of the micro-habitats surrounding the organisms, time-series were selected from the close-up video imagery acquired simultaneously, and only data corresponding to a probe position ~ 0-2 cm above the surface of the *Alvinella* colony were retained. For each biological sample, mean, maximum and minimum temperature were then defined while mean, minimal and maximal values of pH, sulphide and iron were calculated from the correlations previously established for each vent.

Biological samples collection

All biological samples were obtained from *Alvinella pompejana* colonies, except two of them which corresponded to the collection of *Riftia pachyptila* clumps. Fauna was collected using the hydraulic arm of the ROV, occasionally completed with the ROV suction device, on an area from ~ 400 to 700 cm^2. On board, samples were washed on a 1 mm mesh sieve and fixed with 10% neutral formalin in seawater. In the laboratory, all gastropods

specimens were sorted, identified to the species level when possible and then transferred to 70° ethanol.

Analysis of community structure

Multivariate analyses were performed with the software Primer v.5 in order to group the samples according to their faunal composition using the Bray-Curtis similarity index calculated from standardized and square-root transformed species-abundance data. The initial standardization consists in calculating the relative abundance of each species by dividing each count by the total abundance of all individuals in the sample, and consequently removes any effect due to differences in sample volumes (Clarke & Warwick, 2001). Data were presented using two complementary graphic approaches: cluster using group-average linking, and non-metric multi-dimensional scaling (NMDS). For each faunal assemblage, species-effort accumulation curves with 95% confidence intervals were generated from species-abundance data using EstimateS v.7.5 (Colwell, 2005) to compare biodiversity distribution from samples of very different size.

In order to assess the environment influence on the community structure, faunal data were linked to environmental factors using the BIO-ENV procedure within the Primer program (Clarke & Ainsworth, 1993). The different steps of this procedure are as follows. A biotic matrix based on Bray-Curtis similarity index from faunal data and abiotic matrices based on the Euclidian distance from environmental factors are established. While the among-samples similarity matrix was calculated once, the equivalent matrix on abiotic data was computed many times at different levels of complexity (i.e. k variables at a time, where k = 1,2,3…., n). The best matches of biological and environmental matrices at increasing levels of complexity were measured using the Spearman rank correlation coefficient (ρ_ω). Minimal values of temperature, S(-II) and Fe(II), and maximal pH were excluded from the analysis as

they were assumed to not be significant for organisms distribution. The natural turbulence of the environment and the difficulties for precise positioning of probes restricted the reliable definition of these extrema at the organism-scale. It can be reasonably assumed that they should represent surrounding seawater conditions, as shown in other vent habitats where background seawater conditions are recovered within a few centimeters from invertebrate aggregations (Johnson et al., 1988; Le Bris et al., 2006b). To ensure that colinearity among the 8 remaining environmental variables (i.e. mean and maximal values of temperature, S(-II) and Fe(II), and mean and minimal pH) did not affect results, 2 of them which have mutual correlations above 0.75 were excluded from the analysis: minimal pH and mean temperature.

Population biology of Lepetodrilus elevatus

The demographic population structure of *Lepetodrilus elevatus* was analysed for the Genesis site and the Parigo PP-Ph05(1) vent where this species was in sufficient number (i.e. > 100 individuals). The curvilinear shell length (L_{curv}), defined as the total length from the anterior edge of the shell to the lip of the protoconch, was used as a size index (Sadosky et al., 2002). Measurements were conducted under the 'Image tool' image analysis software (University of Texas, http://www.uthscsa.edu/dig/download/html) with an error fixed at 0.14 mm. All specimens in a sample were measured except for the two very large samples from the collection of *Riftia pachyptila* clumps for which a random subsample of ~500 individuals was used. Size-frequency histograms were constructed using a size class of 0.4 mm according to three criteria (Jollivet et al., 2000) : (1) most size-classes must contain at least 5 individuals; (2) the number of empty adjacent classes must be minimized; and (3) the size-class interval has to be much greater than the error of measurement. Size-frequency distributions were compared among samples using a Kruskall-Wallis multi-sample test (Zar, 1999).

All previously measured individuals with a curvilinear length > 3mm were sexed to determine the sex-ratio which was compared to a theoretical sex-ratio 1:1 with a χ^2 goodness-of-fit test. Males were identified by the presence of a large penis, modified from the left cephalic tentacle. In addition, sexual maturity of females was assessed from histological examination of gonads (Pendlebury, 2004; Kelly & Metaxas, 2007). Depending on the sample size, 1 to 11 females per vent were analysed. Body of females stored in 70° ethanol were removed from their shell, dehydrated in 100° ethanol for at least 6 hours, cleared in xylene for 6 hours and embedded in paraffin wax in a 70°C oven for approximately 12 hours. Individuals were then set in wax blocks and 2 to 3 serial sections of 7 µm thickness were obtained with a microtome. Sections were stained using the picro indigo carmin method which stains nucleus in brown and cytoplasm in green (Gabe, 1968). For each female, 14 to 229 oocytes, in which the nuclei were visible, were measured from images captured under a microscope. As packing of the oocytes can severely distort the oocyte shape, Feret diameter was calculated from the measure of the oocyte area using the Lucia software (Laboratory Imaging Ltd.). Intra- and intersample synchrony of female reproductive development was determined using a Kruskall-Wallis multi-sample test to compare the oocyte size-frequency distributions. When significant differences occurred, a multiple range test using the Dunn-Nemenyi procedure was performed (Zar, 1999).

RESULTS

Physico-chemical environment

At the surface of alvinellid colonies, the mean temperature varied from 6 to 32°C among samples (Table 2). It was globally higher for the Genesis PP12 samples, varying between 25 and 32°C, and lower for the Parigo samples, ranging between 7 and 12°C at PP-Ph05(1), and 15 and 16°C at PP-Ph05(2). At the Elsa site, large differences in mean

temperature were observed between the samples from the two vents. While it did not exceed 6°C at PP-Ph01, it reached 24 and 20°C respectively for the 2 samples from PP-Hot3. The range of temperature oscillations was highly variable but tended to be wider as the average temperature increased. Thus, for the Genesis PP12 vent, which displayed the highest mean value, the temperature fluctuated between 5 and 69°C over a few minutes. Likewise, at the Elsa PP-Hot3 vent, temperature variations could reach 45°C whereas they did not exceed 27°C for the Parigo site samples. As discussed in Le Bris & Gaill (2007), such dispersion of temperature data can be easily explained by, both, turbulent mixing of hot fluid and cold seawater above the openings of *Alvinella* tubes and weak instabilities in the position of the probe within the steep gradients characterizing these habitats. Note that temperature variations recorded in the present study were independent of time series duration ($R^2 = 0.0629$; $n = 14$; $p = 0.3872$).

Estimated mean pH corresponding to samples were lower than regular seawater pH with acidic to slightly acidic values (i.e. 6.0-6.6) at the Genesis PP12 and Elsa PP-Hot3 vents and near-neutral to alkaline values (i.e.7.4-7.8) at the Parigo site and the Elsa PP-Ph01 vent (Table 2). Estimated mean sulphide concentrations ranged from 164 to 406 μmol l^{-1} with the highest values corresponding to samples of Parigo PP-Ph05(2), Genesis PP12 and Elsa PP-Hot3 vents. Reflecting temperature variability, large sulphide ranges were defined for each sample (e.g. ranging between 24 and 927 μmol l^{-1} for the sample E3-b). Iron concentrations greatly differed among sites with a large contrast between the Genesis-Parigo sites and the Elsa site. While iron was strongly depleted in the habitat sampled at Genesis (below the level of detection of our *in situ* analysis method) and displayed only moderate concentration in the environment of Parigo (less than 29 μmol l^{-1}), this compound reached much higher concentrations at the Elsa PP-Hot 3 vent (between 270 and 339 μmol l^{-1}). At Elsa PP-Ph01, the estimated average concentration revealed to be low but the large range of iron content

indicated that it could be found punctually at high concentrations. Rather than a iron-depleted fluid such as observed at Genesis, the low mean iron level in this case reflected a weak contribution of the hydrothermal fluid to the environment, on average, as indicated by a low mean temperature.

As compared to the large variability of physico-chemical conditions encountered when considering the whole architecture of the alvinellid aggregation (Le Bris et al., 2005), the variability of physico-chemical parameters was quite low within a vent at the surface of the alvinellid zone. Conversely, substantial variability in habitat physico-chemical conditions could be observed among different vents within a site. As an example, two types of habitat were considered at the Parigo and Elsa sites: one with lower temperature, near neutral pH and moderate sulphide concentrations (PP-Ph05-1 and PP-Ph01) versus one with higher temperature, slightly acidic pH and higher sulphide concentrations (PP-Ph05-2 and PP-Hot3). Major differences among sites were due to variations in iron concentrations.

In *Riftia* clumps, if the sample from Genesis Hot 2 displayed the lowest mean value of temperature, the thermal range in sample P1-e from Parigo site did not differ from neighbouring samples collected in *Alvinella* colonies on the same vent (Table 2). According to our model assuming that temperature correlation with chemical parameters is conserved at site scale, similar chemical features for these two environments at Parigo were inferred from similar temperatures.

Faunal composition

A total of 11 gastropod species were identified in the 14 samples (Table 3). Five species (i.e. *Lepetodrilus elevatus, L. pustulosus, Nodopelta heminoda, N. subnoda* and *Peltospira operculata*) were found in 9 to 13 samples and represented between 87 and 100%

of the total number of individuals in each sample. By contrast, only 2 species (i.e. *Pachydermia laevis* and *Hirtopelta hirta*) were found in only one sample.

The cluster analysis showing the percentage similarity of faunal composition for each sample is given on Figure 1A. Samples separated into two well-defined faunal groups. The cluster 1 included all the samples from the Parigo PP-Ph05(1) vent as well as the G2-a sample collected in a *Riftia* clump and exhibited high internal similarity. It was largely dominated by *L. elevatus* which accounted for more than 80% of individuals. The cluster 2 was composed of samples from Genesis-PP12, Parigo PP-Ph05(2) and Elsa vent sites, which displayed a large proportion of peltospirid gastropods (i.e. *P. operculata*, *N. subnoda*, *N. heminoda*). However, the low internal similarity between samples of this cluster, just over 50 %, testified of a high heterogeneity of faunal composition. For example, the samples P2-a, E1-a and E3-a were characterized by a high proportion of *P. operculata* (54.5 to 61.8 % of individuals) and the presence of *N. subnoda* (15.2 to 32.7 % of individuals) while the samples G12-a, G12-b, P2-b and E3-b were mainly composed of a mixture of both species of *Nodopelta* (23.5 to 84.1 % of individuals) and *L. elevatus* (8.3 to 47.1 % of individuals). Species-effort curves suggested that habitats related to the group 2, characterized by a large proportion of peltospirids, have higher species richness than habitats related to group 1, largely dominated by *L. elevatus* (Figure 2). Nevertheless one can note that no species-effort curve had reached the asymptote.

The BIO-ENV procedure provided the best matching of faunal groups to physico-chemical patterns by considering combinations of environmental variables at increasing levels of complexity. When only one variable was considered, mean sulphide concentration appeared to be the most explanatory variable with a ρ_w of 0.373. The next best variables were mean pH (ρ_w = 0.358) and maximal temperature (ρ_w = 0.233). The overall optimum combination involved these 3 variables (ρ_w = 0.401). Superimposing these environmental data

onto the NMDS performed on faunal composition highlighted the influence of these physico-chemical variables in shaping gastropod communities (Figure 1B). The cluster 1 was associated with lower sulphide concentrations and maximal temperatures, and a higher pH. In contrast, the cluster 2 tended to be generally associated with higher maximal temperatures and sulphide concentrations and a lower pH. Nevertheless, a large disparity in physico-chemical conditions occurred within this cluster. As an example, the highest and the lowest mean sulphide concentrations reported in the data set (406 and 53 μmol l^{-1}) were observed for two samples from this cluster, E3-a and E1-a respectively (Table 2).

Biology of Lepetodrilus elevatus

The population structure of *L. elevatus* was analysed on only 8 samples from 3 vents (i.e. Parigo PP-Ph05(1), Genesis PP12, Genesis Hot2) for which individuals were in sufficient number (Figure 3). The curvilinear shell length ranged from 1.09 to 11.27 mm. Size-frequency distributions were highly variable among samples (Kruskall-Wallis H statistic = 430.61, df = 7, $p < 10^{-3}$). Multiple range tests using the Dunn-Nemenyi procedure identified 4 groups of samples: (i) G12-a, (ii) G2-a, (iii) P1-b, P1-c, P1-d and P1-f, and (iv) P1-a and P1-e. These latter samples differed from the other Parigo samples by a higher abundance of larger individuals (> 7 mm). Furthermore, the G12-a sample was distinguishable from other samples by a dominance of small individuals. The proportion of individuals < 5 mm reached 85.8 % in G12-a while it varied between 14.7 and 48.6 % in other samples.

For all samples the sex-ratio did not differ significantly from the theoretical balanced 1:1 sex-ratio (χ^2 goodness-of-fit test; $p>0.05$). In the gonad, oocyte diameter ranged from 7.3 to 113.9 μm. Two types of germinal cells were observed: (1) small oogonies and previtellogenic oocytes with a large nucleus, a basophilic cytoplasm and a size < 40 μm and (2) vitellogenic oocytes with a large cytoplasm containing yolk granules and a size > 40 μm.

Most females exhibited a common pattern in oocyte size distributions with a first major peak of oogonia and previtellogenic oocytes and a second minor peak of vitellogenic oocytes (Figure 4). However, oocyte size-frequency distributions differed significantly at the different spatial scales analysed in this study, suggesting asynchronous reproduction among vents and among females within each vent (Figure 5). Among vents, significant differences in size distributions (Kruskall-Wallis H statistic = 246.28, df = 4, $p < 10^{-3}$) were mainly due to the individuals from the Genesis PP12 vent which exhibited a smaller mean oocyte size than individuals from the other vents ($p < 0.05$). This result could be explained by the animal size as the 7 individuals from the Genesis PP12 vent were the smallest individuals analysed in this study (i.e. curvilinear shell length ranging from 3.71 to 5.48 μm) and were mainly characterized by a large dominance of previtellogenic oocytes (Figure 4). Indeed, while there was a significant correlation between the curvilinear shell length and the percentage of vitellogenic oocytes per female when all females were considered ($R^2 = 0.2564$, n = 72, $p < 10^{-3}$), the correlation became non significant when the individuals from the Genesis PP12 vent site were removed ($R^2 = 0.0578$, n = 65, p = 0.054). Significant differences in oocyte size distributions were also reported among samples within a vent on the example of Parigo PP-Ph05(1) vent (Kruskall-Wallis H statistic = 62.79, df = 5, $p < 10^{-3}$) and among females within a sample (sample P1-a: Kruskall-Wallis H statistic = 70.97, df = 10, $p < 10^{-3}$; sample P1-d: Kruskall-Wallis H statistic = 51.61, df = 9, $p < 10^{-3}$; sample P1-f: Kruskall-Wallis H statistic = 150.36, df = 10, $p < 10^{-3}$). However, multiple range tests using the Dunn-Nemenyi procedure indicated that 54 to 60 % of the females examined in each sample showed no significant difference in oocyte size-frequency distributions.

DISCUSSION

At the scale of a biological sample, a large variability of physico-chemical conditions was reported, consistently with the steep chemical gradients already described over scales of centimetres and seconds to hours in hydrothermal habitats (Johnson et al., 1988; Chevaldonné et al., 1991; Sarradin et al., 1998; Le Bris et al., 2005). From this observation it was suggested that animals would occasionally deal with high temperature, high sulphide concentrations and low pH. However, the estimated amplitude of thermal and chemical ranges at the surface of the alvinellid colony where the gastropods are located are still substantially lower than depicted for the general *Alvinella pompejana* environment (Sarradin et al., 1998; Luther et al., 2001; Le Bris et al., 2003; Le Bris et al., 2005). This is consistent with the synthesis of temperature ranges reported for this environment in Le Bris & Gaill (2007) that underlined much milder conditions at the surface of the colony than among the tubes of *Alvinella* spp. that composed the interface between the chimney wall and seawater. Still, close-up video recordings confirmed that organisms sometimes bathed in turbulent shimmering water, where they can alternately experience, in few seconds, conditions that only slightly depart from seawater and conditions with a more significant hydrothermal influence. Considering the similar temperature ranges, no large variation in the range of physico-chemical conditions was expected among samples in the *Alvinella* habitat, within a single vent, suggesting that the habitat sampled was quite homogeneous. Within a single *Alvinella* aggregation, Le Bris et al. (2005) highlighted substantial discrepancies at small-scale but they were mostly reported when comparing different microenvironments in the matrix surrounding *Alvinella* tubes and the interior of the tubes. Comparatively, measurements from the water layer overlying the surface of the colony, where gastropods live, appear more consistent with the conservative mixing model, at least in the vicinity of venting sources (Le Bris et al., 2005). Larger environmental differences were documented among samples from different vents within a site. Thus, at the Parigo and Elsa sites, one vent was characterized by lower temperature and

423 sulphide concentrations, and higher pH than the other one. Among sites, the most significant
424 result in terms of spatial variations in habitat chemistry regarded iron which distinguished the
425 iron-rich Elsa site from the iron-depleted Genesis and Parigo sites. Such variations in the Fe:S
426 ratio which mainly depends on the end-member vent fluid composition have been already
427 highlighted for the 13°N/EPR hydrothermal vent field (Le Bris et al., 2003, Le Bris & Gaill,
428 2007).

429 While thermal conditions are commonly reported to be strongly contrasted in different
430 hydrothermal habitats (Shank et al., 1998; Luther et al., 2001; Le Bris et al., 2003), no
431 difference in temperature was observed between the sole *Riftia*-dominated sample and
432 *Alvinella*-dominated samples at Parigo PP-Ph05(1) vent. Again, this is not so surprising
433 considering that the surface of *Alvinella* tube aggregation displays a much more limited
434 thermal range than the bulk of the colony. The chemical conditions could however be much
435 more contrasted than assumed here between these habitats as the consistency of the
436 temperature correlation with sulphide, iron and pH may only be valid when comparing
437 alvinellid environments. Large discrepancies in the sulphide-temperature correlation have
438 been observed between alvinellid and *Riftia* aggregations located less than one meter apart
439 (Le Bris et al., 2006a).

440 Similarly to environmental conditions, gastropod community structure was mainly
441 similar among samples from a single vent but variable among vents and sites, being either
442 dominated by peltospirids (i.e. *Peltospira operculata*, *Nodopelta heminoda* and *N. subnoda*)
443 at Elsa, Genesis-PP12 and Parigo PP-Ph05(2) or lepetodrilids (i.e. *Lepetodrilus elevatus*) at
444 Parigo PP-Ph05(1) and Genesis-Hot 2. Nevertheless, within the peltospirids-dominated vents,
445 the relative proportion of *P. operculata* and *Nodopelta* spp. as well as the abundance of *L.*
446 *elevatus* could be highly variable among samples. While numerous studies have already
447 highlighted changes in habitat temperature and chemistry as primary driving forces for

448 changes in hydrothermal community composition at the site or vent field-scale (Sarrazin et
449 al., 1997; Shank et al., 1998; Sarrazin & Juniper, 1999), our study showed that for gastropods
450 the most explanatory environmental variable considered alone was the mean sulphide
451 concentration followed by the mean pH and maximal temperature. If the best matching of
452 faunal composition to environment involved these 3 physico-chemical variables, the addition
453 of pH and temperature improved only slightly the link between environmental data and
454 gastropod distribution patterns.

455 The role of these 3 environmental variables on species distribution may be related to
456 physiological adaptations and nutritional requirements as generally suggested for
457 hydrothermal fauna. Sulphide is thought to be the primary energy source for chemosynthetic
458 bacterial primary production in these environments (Childress & Fisher, 1992) but is also
459 known to be deleterious to all aerobic organisms (Visman, 1991). Hydrothermal animals thus
460 have to deal with conflicting constraints related to this compound. Likewise, temperature can
461 act directly on vent fauna according to their thermal tolerance. Lee (2003) experimentally
462 showed that *Lepetodrilus fucensis* and *Depressigyra globulus*, 2 common gastropods in
463 alvinellid colonies at the Juan de Fuca Ridge (Northeast Pacific), could not stand short
464 exposure to temperature exceeding 30-35°C and 35-40°C respectively. Moreover, Bates et al.
465 (2005) suggested from field observations and experiments that temperature had a significant
466 influence on the distribution patterns of 3 gastropod species (i.e. *L. fucensis*, *D. globulus* and
467 *Provanna variabilis*) from the Juan de Fuca Ridge. While the former two species occupied
468 near-vent habitats with a temperature between 5 and 13°C, the latter one was reported in areas
469 with significantly lower temperature from 4 to 11 °C. Although the direct influence of pH on
470 vent fauna physiology is less documented, this parameter is highly relevant to assess the
471 impact of habitat condition on vent communities as it mainly affects the distribution of
472 sulphide in different chemical forms of contrasted biological effects (Le Bris et al., 2003).

The sulphide toxicity and bioavailability mostly depends on the relative proportions of the free sulphide and iron-associated forms (Luther et al., 2001; Le Bris et al., 2003). According to their iron concentration and pH range, the quality of the habitats sampled in this study could be classified in terms of relative toxicity. At low to negligible iron concentrations, free sulphide forms (H_2S and HS^-) constitute the dominant sulphide species. The acidity constant (pKa) of H_2S being close to 7, sulphide should be mostly present as the most toxic neutral species, H_2S, in the higher temperature and more acidic habitat (i.e. Genesis PP-12) while it would be mostly present as the less toxic anionic species, HS^-, in the lower temperature and less acidic habitat (i.e. Parigo PP-Ph05(1)). Conversely, the high iron concentrations observed at Elsa site would decrease sulphide environmental toxicity, even in this acidic habitat by the formation of FeS.

A first assumption to explain the differential distribution between peltospirids and lepetodrilids species at 13°N could be their different thermal tolerance. Such assumption is in accordance with recent observations performed at 9°50’ N which showed different thermal boundaries between lepetodrilids mainly associated with the vestimentiferan habitat and peltospirids confined to alvinellids habitat (Mills et al., 2007). However, if *L. elevatus* was abundant (more than 50 % of individuals) in samples with maximal temperature not exceeding 29°C, it was also reported in samples where maximal temperature reached 69°C, suggesting that this species could be subject to short-term exposures to high temperature. The better tolerance of *Nodopelta* spp. and *P. operculata* to sulphide toxicity, as compared to *L. elevatus* might provide another explanation to the distribution observed. In this case, the former species will be favored in habitats characterized by higher sulphide concentrations and temperature, and more acidic conditions. However, gastropods community structure in the presumably more toxic environment, Genesis-PP12, did not highly differ from the structure reported in samples from sulphide-rich but less toxic environment like Elsa PP-Hot3 and

498 Parigo PP-Ph05(2) vents. Govenar et al. (2005) also highlighted high similarity in epifaunal
499 community structure including lepetodrilids in *Riftia pachyptila* clumps of contrasted sulphide
500 and iron ranges

501 Despite heuristic and conceptual interest, the BIO-ENV procedure mainly remains an
502 exploratory tool to assess the relationship between multivariate community structure and
503 environmental variables (Clarke & Warwick, 2001). Even if conclusions cannot be supported
504 by significance tests given the lack of model assumptions underlying the procedure, the low
505 values of ρ_w ranging from 0.373 to 0.401 suggested that chemical variables as measured in
506 this study only poorly explained gastropod distribution patterns. Two non-exclusive
507 hypotheses could be proposed: (1) limitations in the ability to discriminate habitats in terms of
508 physico-chemical conditions experienced by organisms and (2) a more complex interplay
509 between abiotic factors impact and biological interactions.

510 Chemical parameters were not measured simultaneously with temperature prior to
511 animal collections, but were extrapolated from temperature measurements assuming a
512 conservative behavior in the mixing interface between the source fluid and ambient seawater
513 at the scale of a single vent edifice. These limitations may have reduced the validity of our
514 extrapolations as some discrepancies in the temperature relationship with chemical factors
515 have been reported at site scale for various types of hydrothermal habitats (Johnson et al.,
516 1988; Le Bris et al., 2000, 2005, 2006b). Indeed, several processes can alter the relationships
517 between temperature and chemical parameters including biological consumption of sulphides
518 in mussel beds and *Riftia* clumps (Johnson et al., 1994; Le Bris et al., 2006b) and conductive
519 thermal exchanges in *Alvinella* colonies (Le Bris et al., 2005). Over similar temperature
520 ranges, highly different sulphide contents may indeed characterize adjacent *Riftia* clumps and
521 *Alvinella* colonies on a single chimney (Luther et al., 2001; Le Bris et al., 2003, 2006b), and
522 the lack of chemical differences supposed at Parigo PP-Ph05(1) vent between both habitats

523 may be erroneous in the present study. In the *Alvinella* habitat, if temperature is not a relevant
524 tracer of fluid mixing, the processes involved vary among distinct micro-environments (Le
525 Bris et al., 2005). In the inner-tube environment, the unexpected combination of a high
526 temperature and a high pH is mostly explained by a conductive heating of a seawater-
527 dominated mix through the tube walls whereas, in the medium surrounding the tubes, a
528 conductive cooling of the warm and low pH fluid occurs when it passes through the thickness
529 of the worm colony. By contrast, at the surface of the colonies, a detailed analysis of the
530 relationship between temperature and pH, considered as a reliable tracer of the vent fluid
531 contribution, suggested that the conservative mixing hypothesis is acceptable in a first
532 approximation (Le Bris et al., 2005). For this peculiar environment on which most of the
533 present study focused, the use of empirical correlations between temperature and chemical
534 parameters could be assumed to have greatly improved the general characterization of
535 habitats, as compared to those only based on temperature measurements. On the other hand,
536 even if close-up video imagery was used to ensure that ranges and mean values described the
537 environment of gastropods at the surface of *Alvinella* colonies, the measurement strategy used
538 for this study did not allow us to characterize fine-scale temperature variability in micro-
539 niches at the individual scale (i.e. cm) as reported in Di Meo-Savoie et al. (2004), Bates et al.
540 (2005) and Le Bris et al. (2005). Indeed, some hydrothermal gastropods may occupy
541 microhabitats that differ from the general surrounding physico-chemical environment at the
542 surface of *Alvinella* colonies (Mills et al., 2007).

543 A better assessment of temporal variability would be also required to appreciate the
544 factors that could influence species distribution. Temperature in vent habitats fluctuates at
545 different time scales, in response to turbulent mixing of hydrothermal fluid and ambient
546 seawater which produces rapid pulsations and brief spikes at second to minute scales, to tidal
547 motion at hour to day scales, and to variations in bottom currents and hydrothermal emissions

548 at longer scale (Shank et al., 1998; Chevaldonné et al., 1991; Tivey et al., 2002). In the
549 present study, time series measurements of temperature performed only at short timescale,
550 from about 1 to 44 minutes, could imperfectly describe thermal conditions encountered by
551 organisms. A continuous record of temperature over a week on an *A. pompejana* colony at
552 Elsa PP-Hot3 vent showed that temperature mostly ranged between 10 and 20°C but could
553 display peaks of temperature reaching 25-27°C for a duration of several hours (Pradillon et
554 al., 2005a). Nevertheless, measurements carried out at different dates at Parigo PP-Ph05(1)
555 vent in the present study provided comparable range of temperature variations and no
556 significant relationship occurred between temperature fluctuations and time series duration,
557 suggesting some stability over time. Likewise, the Fe:S ratio in end-member fluid which is
558 largely modulated by subsurface processes, was also reported to be highly variable in time in
559 relation to variations in fluid emission, mainly at monthly to yearly scales (Shank et al., 1998;
560 Von Damm & Lilley, 2004). The distinction between the iron-rich Elsa site and the iron-
561 depleted Genesis and Parigo sites depicted in the present study has already been mentioned by
562 Le Bris et al. (2003) from measurements performed 3 years before (Le Bris et al., 2003).
563 Thus, for mobile species like gastropods which can actively respond to long-term variations in
564 the physico-chemical environment, we can expect that our data are sufficiently representative
565 of the global range of environmental parameters actually experienced by the organisms at
566 scales ranging at least from minutes to weeks.

567 In addition to the organism physiological tolerances to the physico-chemical
568 environment, the gastropod community structure could also rely on biological interactions
569 between species, including predation and competition for a limiting food and space resource.
570 Over a limited range of environmental conditions, the relative dominance of one species
571 would in part depend on its abilities to outcompete other species according to its reproductive
572 and growth potential. In this context, the most striking result of the present study is the lower

573 species richness in the lower temperature and sulphide concentrations vent (Parigo PP-
574 Ph05(1)), because of the overwhelming dominance of *L. elevatus* in *Alvinella*-dominated as
575 well as *Riftia*-dominated habitats. In the *Riftia* habitat, manipulative field-experiments already
576 showed that *L. elevatus* could strongly modify the community structure, and exert negative
577 influences on sessile and mobile colonists by physically dislodging their recruits (Micheli et
578 al., 2002; Mullineaux et al., 2003). Different results on the demography and reproduction of
579 *L. elevatus* in relation to physico-chemical conditions may support the hypothesis that this
580 species might well develop in a less extreme environment and influence peltospirid
581 populations.

582 The significant variation of size-frequency distributions of *L. elevatus* among vents
583 could result from numerous factors including (i) sampling bias related to small sample size,
584 (ii) spatial and temporal variations in larval supply (Metaxas, 2004), (iii) site- and size-
585 specific growth rate (Mullineaux et al., 1998) or (iv) site-and size-specific mortality rate.
586 Nevertheless, the main difference was due to the only sample collected in a high temperature
587 and high sulphide toxicity environment at Genesis PP12 vent which differed significantly
588 from all other samples by a very small proportion of large individuals. Furthermore,
589 Mullineaux et al. (1998) or Sadosky et al. (2002) have shown that recruitment of this species
590 is generally coherent at the vent field scale. Thus, even if the factors mentioned above cannot
591 be ruled out, this pattern may suggest that physico-chemical conditions could alter the
592 demography of *L. elevatus*. The very high proportion of small individuals at Genesis PP12
593 vent may result from broader physiological tolerances and higher survivorship of juveniles
594 than adults in marginal habitat as already reported by Mullineaux et al. (1998). In terms of
595 reproduction, heterogeneous size-frequency distributions of oocytes among mature females
596 confirmed that gametogenesis was asynchronous at vent as well as at sample scale
597 (Pendlebury, 2004), and seemed independent of the physico-chemical environment. However,

598 the positive relationship between the animal size and its sexual maturity, defined as the
599 proportion of vitellogenic oocytes, indicated that most individuals from the Genesis PP12
600 vent were immature and did not participate to the local reproductive effort. Along the Juan de
601 Fuca Ridge, Kelly & Metaxas (2007) observed that *L. fucensis* gametogenic maturity did not
602 vary between actively venting habitats but was significant lower in senescent areas according
603 to variation in energy supply. Here, while the environment at the Genesis site may constitute
604 the upper limit for *L. elevatus* to develop, the cooler habitats seem to be optimal, so that the
605 females can maximize their reproductive output. While the lack of replicate in the most toxic
606 habitat impedes a global statistical analysis over the range of physico-chemical conditions
607 encountered in the present study, those results suggest that *L. elevatus* may be highly
608 competitive in the lower temperature and less toxic environmental conditions. Even if
609 peltospirid gastropods may probably survive in lower temperature, as at Elsa PP-Ph01 vent,
610 habitat colonization by *L. elevatus* could exclude them since environmental conditions are
611 favorable to the development of this latter species in *Alvinella*- as well as in *Riftia*-dominated
612 habitat.

613 In describing the different hydrothermal communities structure and diversity patterns,
614 only a few studies to date have included locally defined chemical parameters of ecological
615 relevance to identify processes governing the observed patterns (Sarrazin et al., 1997; Shank
616 et al., 1998; Sarrazin & Juniper, 1999; Govenar et al., 2005). Despite the inabilities to actually
617 perform quantitative samples in the high-temperature hydrothermal environment, the present
618 study reported for the first time the influence of environmental chemistry on epifaunal
619 assemblages in different alvinellid colonies at vents along the East Pacific Rise, focusing on
620 gastropods. The main physico-chemical parameters governing the habitat quality and
621 consequently the community structure were, in a decreasing order of importance, mean
622 sulphide concentration, mean pH and maximal temperature. Peltospirid gastropods (i.e.

623 *Nodopelta* spp. and *P. operculata*) were then dominant in the more acidic, higher temperature
624 and richer sulphide vents than lepetodrilid gastropods (i.e. *Lepetodrilus elevatus*) (Figure 6).
625 Although this pattern could result from different specific physiological tolerances and
626 nutritional requirements, the occurrence of all common species over a wide range of physico-
627 chemical conditions as well as the low correlation between biological community structure
628 and physico-chemical parameters suggests that other factors may be responsible for
629 community composition in *Alvinella* colonies. In particular, the lower richness resulting from
630 the dominance of *L. elevatus* in the lower temperature and sulphide concentrations habitat
631 suggests that the later species may outcompete other species in such environmental
632 conditions. Thus, in these conditions, gastropod community structure did not differ between
633 *Alvinella* colonies and *Riftia* clumps. If field samples remain essential to describe patterns and
634 make assumptions about processes involved, further experimental manipulative shipboard and
635 field studies should be necessary to identify the ways of biogeochemical processes on
636 community structure and separate unambiguously the relative contributions of physiological
637 tolerance, nutritional requirement and biological interactions.
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639
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Table 1. *Location of the study sites and biological samples. For each sample, the number of temperature sampling points and the duration of temperature measurements used to characterize the physico-chemical habitat are given. * indicates samples collected in* Riftia *clumps.*

Vent site	Vent	Sample	Latitude	Longitude	Date (2002)	Number of temperature sampling points	Total duration of temperature measurements (min:s)
Genesis	PP12	G12-a	12°48,632'N	103°56,426'W	May 25	3	1:01
		G12-b			May 25	5	6:44
	Hot2	G2-a*	12°48,635'N	103°56,404'W	May 30	4	49:18
Parigo	PP-Ph05(1)	P1-a	12°48,585'N	103°56,400'W	May 19	4	4:53
		P1-b			May 20	4	42:41
		P1-c			May 22	2	8:37
		P1-d			May 22	1	1:46
		P1-e*			May 22	4	17:22
		P1-f			May 23	5	43:38
	PP-Ph05(2)	P2-a	12°48,620'N	103°56,390'W	May 30	2	22:41
		P2-b			May 30	1	9:42
Elsa	PP-Ph01	E1-a	12°48,150'N	103°56,267'W	May 29	4	26:31
	PP-Hot3	E3-a	12°48,145'N	103°56,266'W	May 29	3	6:56
		E3-b			May 29	5	11:53

Table 2. *Temperature (mean ± SD) and chemical parameters (mean) of the different samples from the 13°N/EPR hydrothermal vent field. Range values are given in brackets. For each sample, temperature was measured at different sampling points (see Table 1) while chemical parameters were calculated from geochemical modelling. See text for explanation. nd: not determined. * indicates samples collected in* Riftia *clumps.*

Vent site	**GENESIS**			**PARIGO**								**ELSA**		
Site	PP12		Hot 2	PP-Ph05(1)						PP-Ph05(2)		PP-Ph01	PP-Hot3	
Sample	G12-a	G12-b	G2-a*	P1-a	P1-b	P1-c	P1-d	P1-e*	P1-f	P2-a	P2-b	E1-a	E3-a	E3-b
Temperature (°C)	32.3±11.2 (22.0-48.0)	25.4±19.8 (5.0-69.0)	4.9±1.20 (2.5-7.9)	12.4±3.7 (2.7-18.1)	9.5±2.3 (2.7-18.1)	7.3±1.8 (3.7-12.2)	7.1±0.9 (5.2-8.5)	8.0±3.7 (2.8-16.1)	11.2±6.5 (2.0-29.0)	16.0±4.0 (7.0-25.0)	15.0±3.0 (7.0-20.0)	6.2±2.3 (2.0-22.0)	23.8±12.6 (5.5-50.3)	20.4±12.9 (4.4-50.3)
pH	6.0 (5.8-6.2)	6.1 (5.5-7.1)	nd	7.5 (7.2-8.0)	7.5 (7.2-8.0)	7.8 (7.5-7.9)	7.8 (7.7-7.9)	7.7 (7.3-8.0)	7.6 (6.4-8.0)	7.4 (6.8-7.8)	7.4 (7.1-7.8)	7.4 (6.7-8.0)	6.5 (6.0-7.5)	6.6 (6.0-7.7)
S-II ($\mu mol.l^{-1}$)	394 (263-594)	306 (46-861)	nd	315 (39-477)	234 (39-477)	169 (68-310)	164 (111-204)	191 (42-421)	282 (20-788)	404 (167-667)	385 (156-534)	53 (0-600)	406 (45-927)	339 (24-927)
Fe II ($\mu mol.l^{-1}$)	0	0	nd	22 (0-35)	15 (0-35)	9 (0-21)	9 (4-12)	11 (0-31)	19 (0-62)	29 (9-52)	28 (8-40)	6 (0-581)	339 (0-867)	270 (0-867)

Table 3. *Number of individuals (N), species richness (SR) and species-abundance list of gastropods collected in the different samples from the 13°N/EPR hydrothermal vent field. * indicates samples collected in* Riftia *clumps.*

Vent site	GENESIS			PARIGO								ELSA		
Vent	PP12		Hot 2	PP-Ph05 (1)						PP-Ph05 (2)		PP-Ph01	PP-Hot3	
Sample	G12-a	G12-b	G2-a*	P1-a	P1-b	P1-c	P1-d	P1-e*	P1-f	P2-a	P2-b	E1-a	E3-a	E3-b
N	348	157	10,848	164	129	281	250	8,258	522	27	17	55	33	91
SR	8	9	6	4	6	5	6	3	2	5	3	4	8	8
Lepetodrilus elevatus	149	13	10,589	158	106	250	224	8,179	509	2	8	0	4	33
Lepetodrilus pustulosus	4	1	205	3	2	9	12	76	13	0	0	0	1	1
Lepetodrilus cristatus	0	1	13	0	1	0	0	0	0	0	0	1	1	7
Nodopelta subnoda	89	68	0	0	7	2	2	0	0	5	0	18	5	22
Nodopelta heminoda	74	64	0	1	8	18	6	0	0	2	4	2	1	22
Peltospira operculata	27	7	0	2	5	2	5	3	0	15	5	34	18	2
Peltospira delicata	1	1	1	0	0	0	0	0	0	3	0	0	2	2
Cyathermia naticoides	2	0	11	0	0	0	1	0	0	0	0	0	1	2
Pachydermia laevis	2	0	0	0	0	0	0	0	0	0	0	0	0	0
Hirtopelta hirta	0	2	0	0	0	0	0	0	0	0	0	0	0	0
Rhynchopelta concentrica	0	1	28	0	0	0	0	0	0	0	0	0	0	0

Figure captions

Figure 1. Bray-Curtis similarity among the 14 gastropod samples collected in *Alvinella* colonies and *Riftia* clumps at 13°N/EPR hydrothermal vent field. Abundance data were standardised to number of individuals in the sample and square-root transformed. (A) Group average sorting dendrogram. (B) Non-metric Multidimensional Scaling (NMDS). Relationships between the 2 major faunal groups identified by the cluster analysis and physico-chemical variables were reported on the NMDS plots. Size of the bubbles is proportional to the value of the physico-chemical parameters for each sample (see Table 2). * on the group average sorting dendrogram indicates samples collected in *Riftia* clumps.

Figure 2. Species-effort curves with 95 % confidence intervals generated with the EstimateS v.7.5. software from species abundance data of each faunal assemblage identified by the group average sorting dendrogram. These curves provide an estimate of expected species richness for random subsets sampled in the total species pool of a faunal assemblage.

Figure 3. Size-frequency distributions of the curvilinear shell length of *Lepetodrilus elevatus* for different samples collected in *Alvinella* colonies and *Riftia* clumps from the 13°N/EPR hydrothermal vent field. N = number of measured individuals. * indicates samples collected in *Riftia* clumps.

Figure 4. Oocyte size-frequency distributions for *Lepetodrilus elevatus* from different vents and sizes. L: curvilinear shell length; N = number of measured oocytes.

Figure 5. Mean oocyte Feret diameter (± 95% confidence limits) (μm) of *Lepetodrilus elevatus* at different spatial scales. A, between vents. B, between samples from the Parigo PP-

Ph05(1) vent. C, D and E, between females from samples P1-a, P1-d and P1-f respectively. Numbers near the mean give the number of individuals and measured oocytes respectively used to calculate mean oocyte diameter. Similar letters indicate no statistical difference among oocyte size distributions ($p>0.05$) calculated from the multiple range test using the Dunn-Nemenyi procedure. * indicates samples collected in *Riftia* clumps.

Figure 6. Schematic representation of the influence of physico-chemical variables on the gastropod community structure for the different vents from the 13°N/EPR hydrothermal vent field. Toxicity of the different habitats was determined from the interactions between total sulphide concentrations, pH, and iron concentrations. For each habitat, dominant gastropod species are indicated.

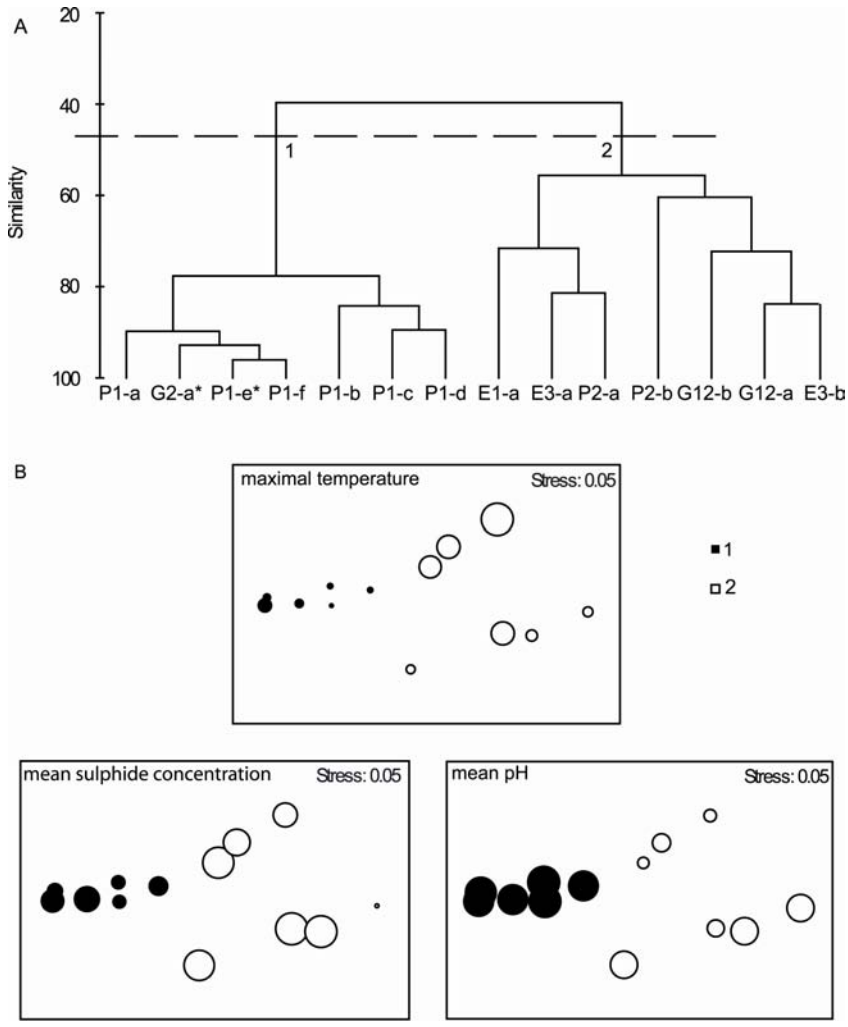
A
20
40
60
80
100
Similarity
1
2
P1-a G2-a* P1-e* P1-f P1-b P1-c P1-d E1-a E3-a P2-a P2-b G12-b G12-a E3-b
B
maximal temperature
Stress: 0.05
1
2
mean sulphide concentration
Stress: 0.05
mean pH
Stress: 0.05

Figure 1

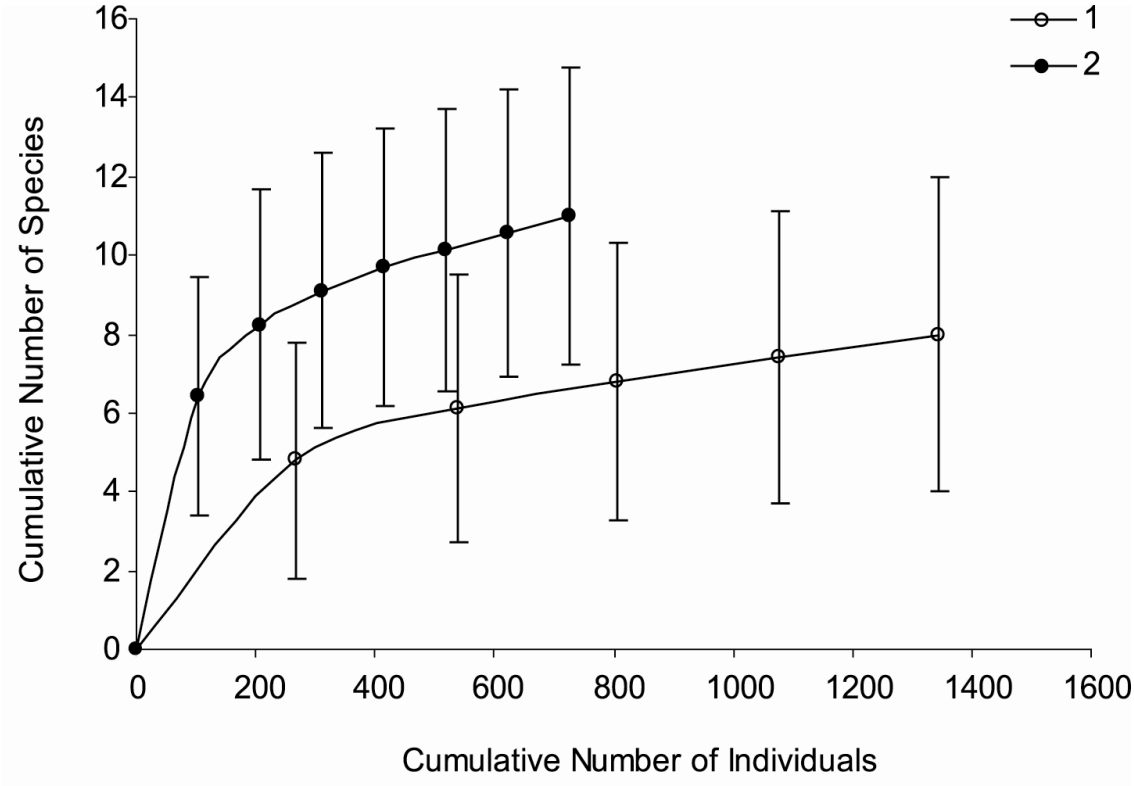
16
14
12
10
8
6
4
2
0
Cumulative Number of Species
0
200
400
600
800
1000
1200
1400
1600
Cumulative Number of Individuals
1
2

Figure 2

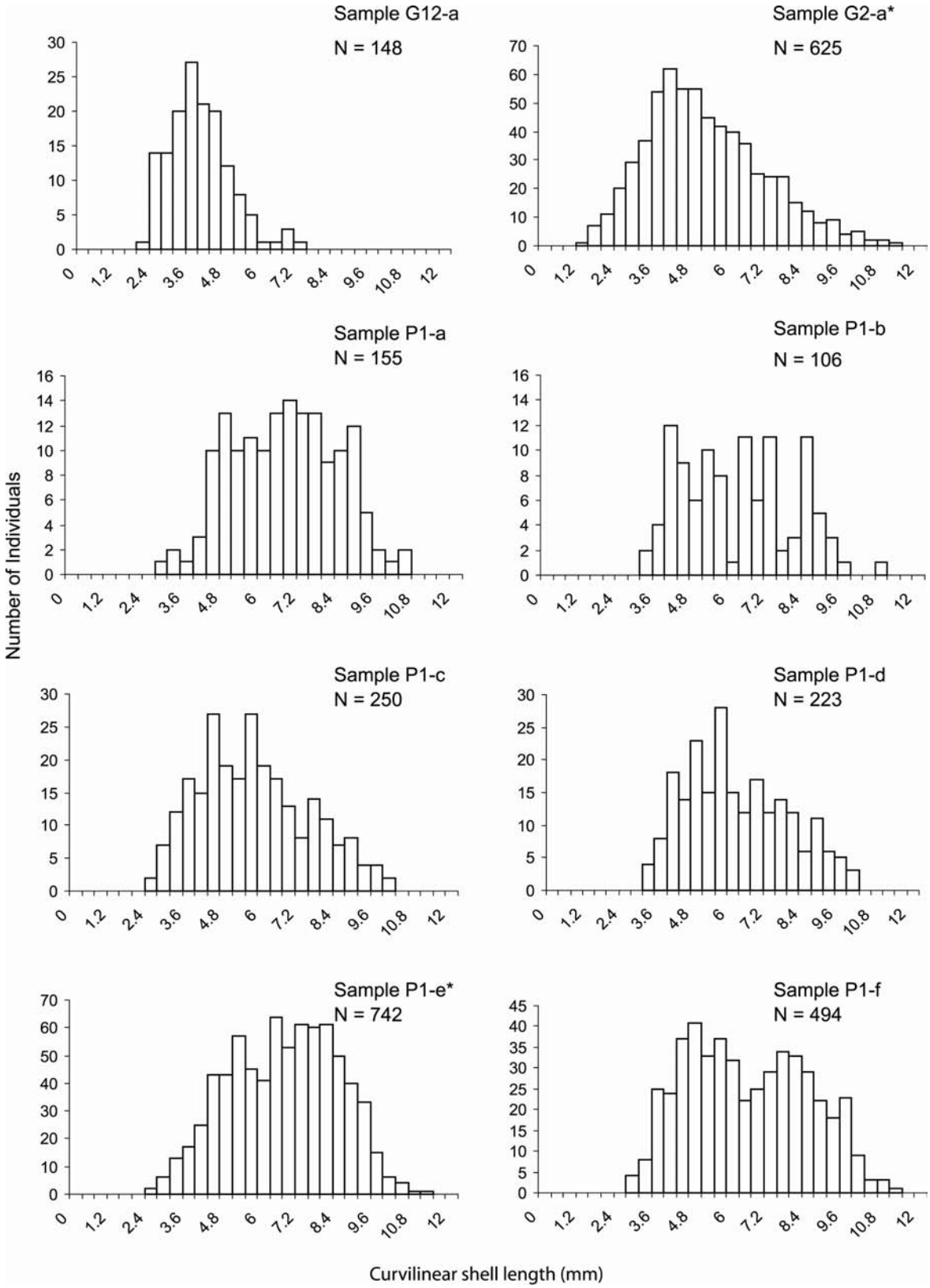
Sample G12-a
N = 148
Sample G2-a*
N = 625
Sample P1-a
N = 155
Sample P1-b
N = 106
Sample P1-c
N = 250
Sample P1-d
N = 223
Sample P1-e*
N = 742
Sample P1-f
N = 494
Number of Individuals
Curvilinear shell length (mm)

Figure 3

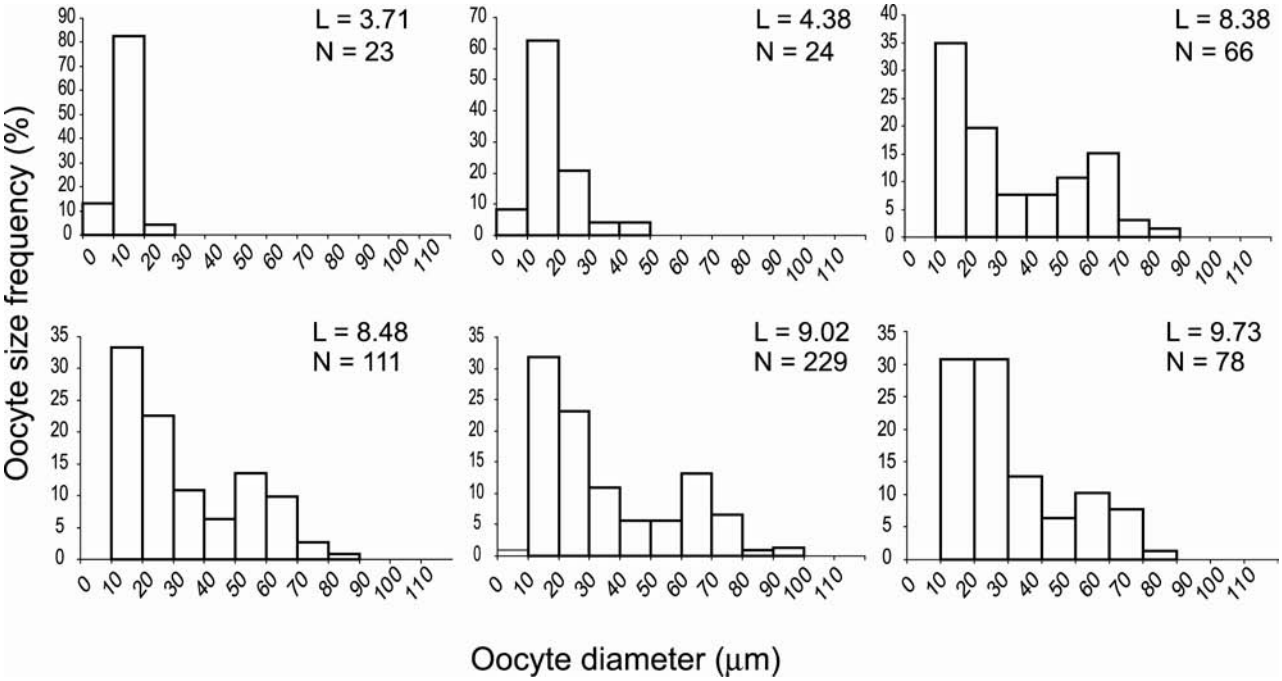
L = 3.71
N = 23
L = 4.38
N = 24
L = 8.38
N = 66
L = 8.48
N = 111
L = 9.02
N = 229
L = 9.73
N = 78
Oocyte size frequency (%)
Oocyte diameter (µm)

Figure 4

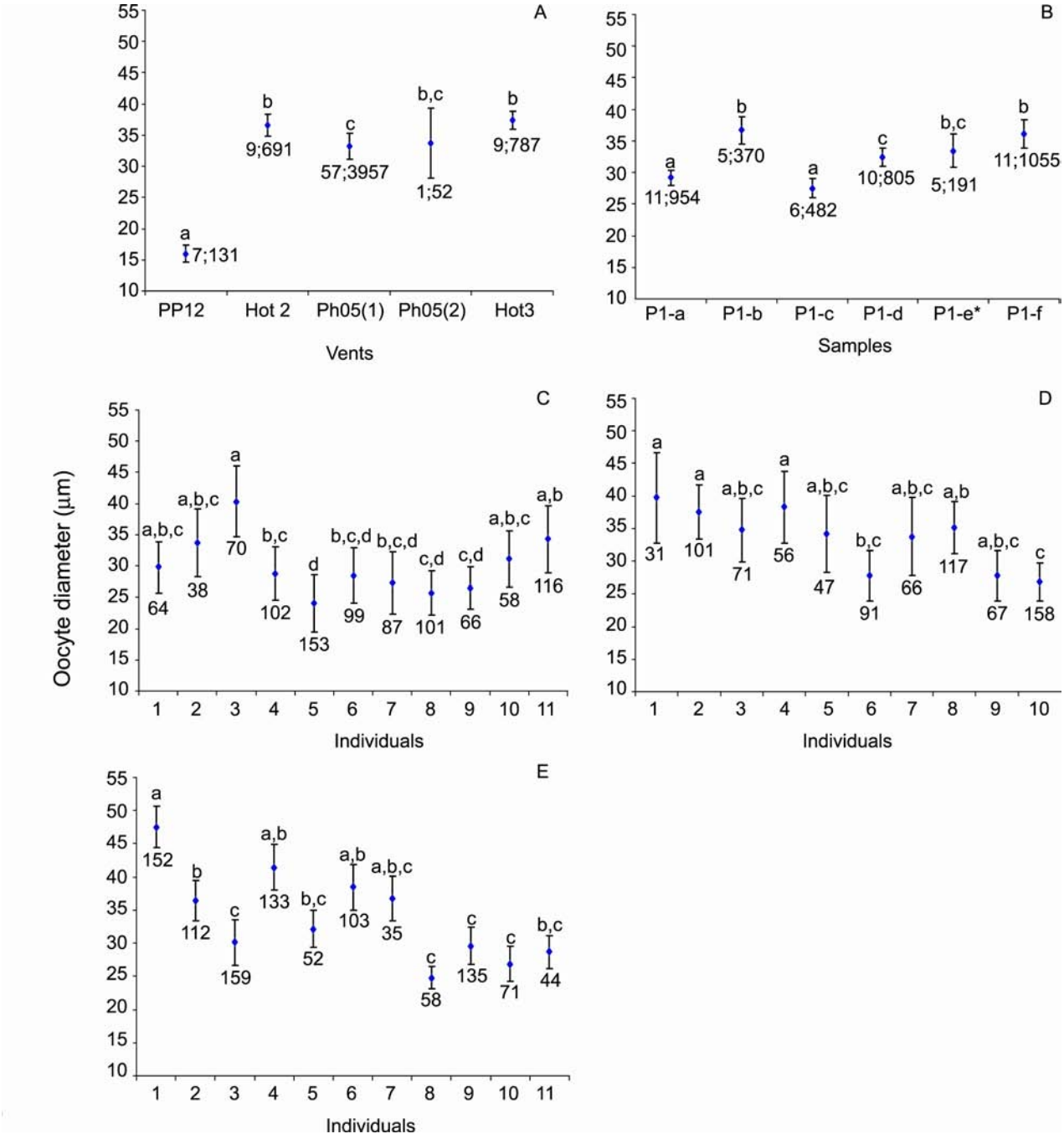
A
B
C
D
E
Oocyte diameter (µm)
Vents
PP12
Hot 2
Ph05(1)
Ph05(2)
Hot3
Samples
P1-a
P1-b
P1-c
P1-d
P1-e*
P1-f
Individuals
7;131
9;691
57;3957
1;52
9;787
11;954
5;370
6;482
10;805
5;191
11;1055
64
38
70
102
153
99
87
101
66
58
116
31
101
71
56
47
91
66
117
67
158
152
112
159
133
52
103
35
58
135
71
44

Figure 5

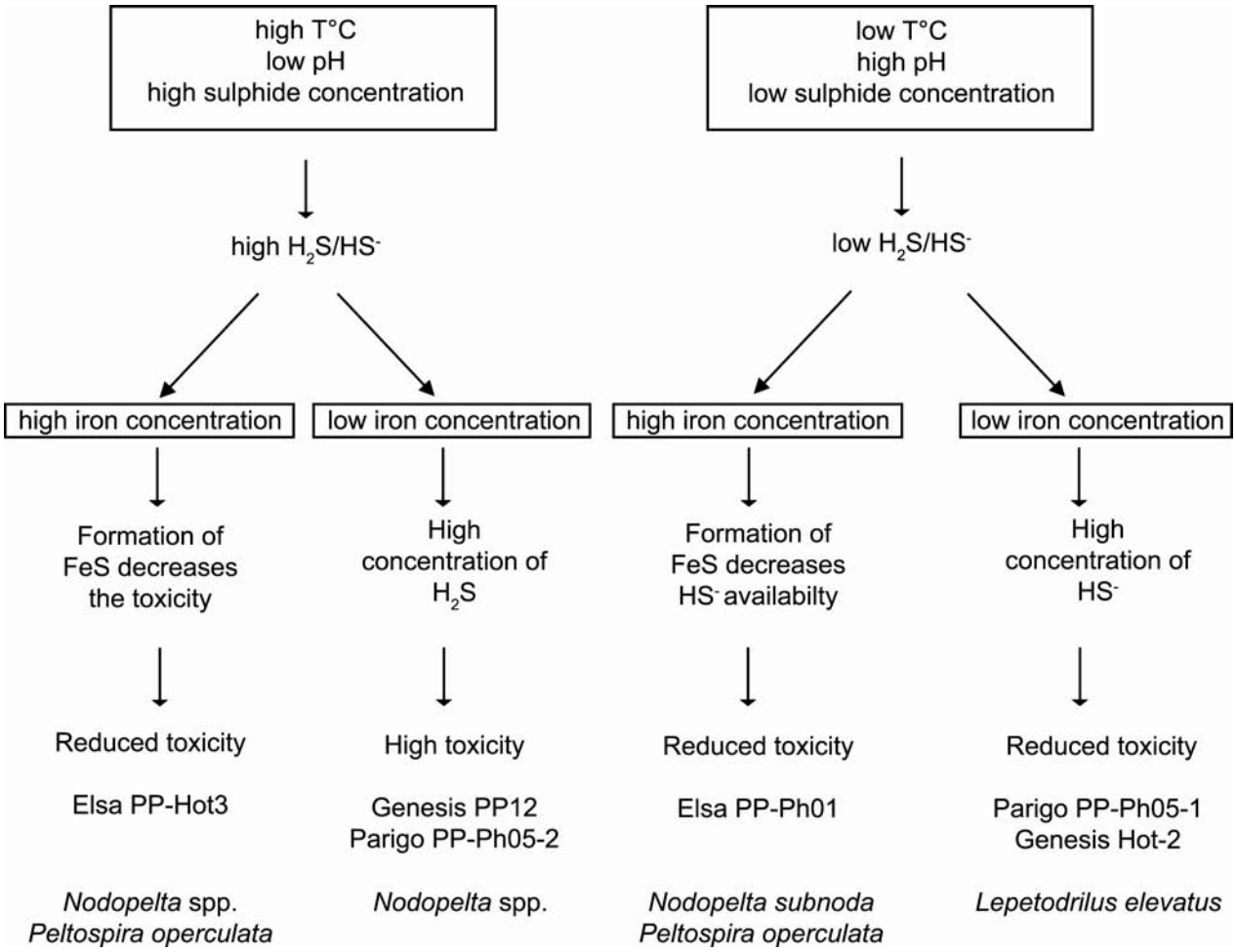
high T°C
low pH
high sulphide concentration
low T°C
high pH
low sulphide concentration
high H_2S/HS^-
low H_2S/HS^-
high iron concentration
low iron concentration
high iron concentration
low iron concentration
Formation of FeS decreases the toxicity
High concentration of H_2S
Formation of FeS decreases HS^- availabilty
High concentration of HS^-
Reduced toxicity
High toxicity
Reduced toxicity
Reduced toxicity
Elsa PP-Hot3
Genesis PP12
Parigo PP-Ph05-2
Elsa PP-Ph01
Parigo PP-Ph05-1
Genesis Hot-2
Nodopelta spp.
Peltospira operculata
Nodopelta spp.
Nodopelta subnoda
Peltospira operculata
Lepetodrilus elevatus

Figure 6