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Effects of triclosan exposure on the energy budget of *Ruditapes philippinarum* and *R. decussatus* under climate change scenarios

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Running head: Triclosan alters the energy budget in two competing bivalves

Abstract

We built a simulation model based on Dynamic Energy Budget theory (DEB) to assess the growth and reproductive potential of the native European clam *Ruditapes decussatus* and the introduced Manila clam *Ruditapes philippinarum* under current temperature and pH conditions in a Portuguese estuary and under those forecasted for the end of the 21st c. The climate change scenario RCP8.5 predicts temperature increase of 3ºC and a pH decrease of 0.4 units. The model was run under additional conditions of exposure to the emerging contaminant triclosan (TCS) and in the absence of this compound. The parameters of the DEB model were calibrated with the results of laboratory experiments complemented with data from the literature available for these two important commercial shellfish resources. For each species and experimental condition (eight combinations), we used data from the experiments to produce estimates for the key parameters controlling food intake flux, assimilation flux, somatic maintenance flux and energy at the initial simulation time. The results showed that the growth and reproductive potential of both species would be compromised under future climate conditions, but the effect of TCS exposure had a higher impact on the energy budget than forecasted temperature and pH variations. The egg production of *R. philippinarum* was projected to suffer a more marked reduction with exposure to TCS, regardless of the climatic factor, while the native *R. decussatus* appeared more resilient to environmental causes of stress. The results suggest a likely decrease in the rates of expansion of the introduced *R. philippinarum* in European waters, and negative effects on fisheries and aquaculture production of exposure to emerging contaminants (e.g., TCS) and climate change.

1. Introduction

Human activities are at the origin of two main hazards that marine species are exposed in the 21st century: changes in physical water parameters (particularly temperature and pH) and the impacts from exposure to chemicals (Manciocco et al., 2014). Both hazards constitute major stress factors for marine organisms (Pörtner and Farrell, 2008; Wittmann and Pörtner, 2013; Fent et al., 2006). In addition to the single impacts of individual stressors, the synergistic action of increased temperature and ocean acidification (OA) on the thermal tolerance of marine ectotherms was hypothesized by Pörtner and Farrell (2008) and shown experimentally by Lannig et al. (2010). For instance, the latter found that standard metabolic rate increased faster in *Crassostrea gigas* exposed to increased temperature and pCO_2 than in oysters subjected only to elevated temperature.

Currently, there is an increasing concern for the consequences of emerging contaminants (ECs), which include pharmaceuticals and personal care products (PPCPs), on humans and aquatic wildlife (Fent et al., 2006; Manciocco et al., 2014). The interaction of ECs with physical parameters (temperature, salinity, pH) can further compound the hazard. It has been demonstrated that water pH variations can influence the toxicity of ionisable drugs (Boström and Berglund, 2015). In the review on the combined effects of climate change and marine pollutants, Cabral et al. (2019) reported that synergistic effects predominate in 2/3 of the cases when compared to additive or antagonistic effects. These authors discuss how a temperature increase in marine systems, due to global warming, is likely to result in increased metabolic rates in ectotherms and higher food demands, thereby increasing their exposure to contaminants and potentially associated harmful effects.

ECs (mostly pharmaceuticals) are bioactive molecules, designed in some cases with a specific mode of action, but in many cases they, or their by-products, also have the capacity to interact with the energetic and biotransformation pathways of aquatic species (Dann and Hontela, 2011; Fernandes et al., 2013; Smith et al., 2012; Ribalta and Solé, 2014). As an example, triclosan (CAS 3380-34-5), the focus of this study, is a chlorophenol antibiotic widely used in PPCPs (Ricart et al., 2010) and is a suspected lipid and endocrine disruptor (Halden and Paull, 2005; Bilal et al., 2020). It is

considered a chemical of concern based on environmental indicators and has been detected in marine waters at concentrations of 1 μg/L (Ortiz de Garcia et al., 2013; Lolić et al., 2015). Bivalves, including clams, can bioaccumulate PPCPs (Gatidou et al., 2020; Ismail et al., 2014), which can interfere in the organism's metabolic processes modulated by temperature and pH (Almeida et al., 2014; Freitas et al., 2015).

Information on the consequences of environmental changes from exposure to ECs is still limited, and in some cases contradictory, among other reasons because not all marine ectotherms react equally to human-induced hazards. The results of experimental exposure of mussels (*Mytilus galloprovincialis*) to certain pharmaceuticals and endocrine disruptors by Serra-Compte et al. (2018) showed that warming and acidification may increase the bioconcentration factor of some of the tested compounds. Moreover, acidification decreases the mussel's ability to metabolize contaminants. Contaminant-specific and species-specific responses need to be taken into account when upscaling the effects of pollution and climate change to the population or ecosystem levels (Cabral et al., 2019).

In particular, the tolerance to environmental stressors may differ between native and exotic species (Velez et al., 2017; Peric et al., 2020), which may favor the establishment of invasive species in vulnerable ecosystems, such as the coastal zones (Darling and Carlton, 2018). A significant example of invasion capability among bivalves is the Pacific oyster (*Crassostrea gigas*), which was introduced for aquaculture purposes in countries of both sides of the Atlantic sea. This species has expanded its range in the eastern coast of the United States and Atlantic Europe, threatening the native oysters *Crassostrea virginica* and *Ostrea edulis,* respectively, due to its broad thermal tolerance and physiology (Anglès d'Auriac et al., 2017; Pack et al., 2021).

One way of understanding the long-term impact of stressors on organisms is by quantitative analysis by means of mathematical models of their effect on the organism's metabolism. In this context, the Dynamic Energy Budget (DEB) model (Kooijman, 2010) is a powerful tool to model the changes on metabolic rates (such as feeding, growth, or reproduction) due to the additional energetic demands of organisms under stress (Flye-Sainte-Marie et al., 2009, Mónaco and McQuaid, 2018).

Costa et al. (2020a, b) conducted laboratory experiments to investigate changes in physiological and metabolic rates in two model marine bivalves of fisheries and aquaculture importance (the European native *Ruditapes decussatus* and the introduced *Ruditapes philippinarum*) subjected to current and forecast temperature and pH conditions for the end of the 21st century and the presence of two emerging contaminants (triclosan and diclofenac). The non-indigenous Manila clam, *R. philippinarum*, was introduced from Indo-Pacific waters for economic purposes in the 1970s in Europe (Flye-Sainte-Marie et al., 2009) as a cultured species more economically profitable than its native counterpart, the European clam, *R. decussatus*. There are evidences that Manila clam is expanding in its invaded range and seems to have reached levels of abundance comparable to or even exceeding the native clam in the Iberian Peninsula (Juanes et al., 2012; Velez et al. 2015).

The present study aimed to evaluate, in energetic terms, the metabolic cost of stress in clams (physical and chemical, from data obtained under experimental conditions) of the expected temperature increase of 3 ºC and decrease in 0.4 units of pH, projected under the more pessimistic RCP8.5 IPCC scenario for the world ocean for 2100 (IPCC, 2019), and the combined effect of exposure to triclosan. The analysis is based on the parameterization of a DEB model separately for the two closely related clams, with the objective of assessing their differential response with regards to Climate Change and ECs in these model organisms of high interest in fisheries and aquaculture.

2. Material and Methods

2.1 Sample collection and experimental set-up

Individuals of *R. decussatus* and *R. philippinarum* of comparable biometrics (average shell length of 4.4 cm and 3.9 cm ShL, respectively), were collected in October of 2018 from shallow (< 3 m depth) banks of the Mira channel of the Ria de Aveiro (NW coast

of Portugal), where pollution from anthropogenic sources is relatively low (Freitas et al., 2014). Clams were acclimated over two weeks to laboratory conditions: temperature 17 °C, salinity 30, and photoperiod 12 h light : 12 h dark (Costa et al., 2020a).

After these two initial weeks, clams were divided and placed in one of two scenarios for 30 days (experimental period I): A) actual field conditions at the time of sampling, autumn: pH 8.1, 17 °C; and B) mimicking forecasted CC conditions of pH decrease and temperature increase by the end of the century (pH 7.7, 20 °C). After this 30-day period, the experiment continued (experimental period II) for 7 additional days and included exposure to triclosan (TCS) (nominally 1 μg/L) in half of the acclimated specimens at the two conditions and for each species making a total of eight possible combinations (2 climate factors × 2 chemical exposures × 2 species), with three replicate aquaria each. During the seven days that included TCS treatment (experimental period II) water from all tanks (control and spiked) was renewed every two days, followed by re-establishing the targeted TCS concentration. The dose of TCS chosen simulates water concentration of environmental concern (1 μg/L) and falls within the range assayed in similar studies (Neuparth et al., 2014). At the end of the experimental phase, two clams per replicate aquarium (n=6 individuals per experimental condition and species) from each of the eight experimental conditions were used to evaluate the respiration rate (RR, mg/h/g dry weight-DW) and energy reserves (glycogen, protein, lipid content, mg/g fresh weight-FW). The full experimental protocol can be found in Costa et al. (2020a, b).

2.2 Dynamic Energy Budget Model

A simulation model based on Dynamic Energy Budget (DEB, Kooijman, 2010) was built to assess the growth and reproductive potential of a 4-cm ShL model individual of *R. philippinarum* and *R. decussatus* along 730 days (2 years). The DEB modelling framework was selected for its ability to capture the underlying physiological processes (growth, reproduction and maintenance) based on first principles that are applicable across different taxa and ontogenetic stages (Kooijman et al., 2020) and the availability of reasonably complete parameter sets in the literature (Sousa et al., 2010; Monaco et al., 2014, AmP collection https://www.bio.vu.nl/thb/deb/deblab/add_my_pet). The

performance of *R. philippinarum* and *R. decussatus* was examined under four scenarios: ambient temperature and pH (17 ℃ in autumn, pH=8.1), increased temperature, and reduced pH (20 ℃ in autumn, pH=7.7), both in the presence or absence of triclosan (Fig. 1). Following other applications of the DEB theory to bivalves (e.g., Flye-Sainte-Marie et al., 2009; Mónaco and McQuaid, 2019), we adopted the *abj* DEB model (i.e., assuming the equations of a standard animal with acceleration growth between birth and metamorphosis (Kooijman, 2010)). In this extension of the standard model, the life cycle of an animal is divided into four main stages: embryo (non-feeding stage), larva (feeding, non-reproductive stage), juvenile (feeding, non-reproductive stage), and adult (feeding, reproductive stage). The stages are separated by the birth, metamorphosis, and puberty transformations. In the *abj* model the "juvenile" stage is divided in two phases: larva (prior to metamorphosis) and juvenile *stricto sensu* (after metamorphosis) to account for differences of growth during the pre-adult stage.

At each stage, the energy of an individual can be apportioned in three compartments: structure (somatic tissue, *V*), reserves (*E*), and maturity (*H*) (Kooijman et al., 2008). Two of these compartments, structure and reserve, contribute to mass, with an additional compartment, the reproduction buffer, also adding to the animal's mass and energy budgets. In DEB *abj* models, the energy of the reproduction buffer is allocated to the gametes at the moment of reproduction. The amount of reserve at any point in time is the balance between energy assimilated from food (assimilation flux $\dot{p}_A$, Appendix A) and energy mobilized to meet costs (according to flux $\dot{p}_C$). The flux of energy from reserves is transferred to somatic maintenance ($\dot{p}_M$) + growth ($\dot{p}_G$) and to maturity ($\dot{p}_J$) + reproduction buffer ($\dot{p}_R$). The proportion of $\dot{p}_C$ allocated to somatic maintenance and growth is assumed to be, in DEB theory, a constant ratio κ ("kappa-rule"). The complement of the mobilized reserves (1-κ) is allocated to maturity and reproduction. The reproduction buffer models the accumulation of invested energy for reproduction, and empties after each spawning event. DEB models give priority to maintenance, drawing energy from the reproduction buffer or structure if maintenance costs cannot be met with ingested food. Appendix A provides details on the DEB model.

The actual parameter values used in the here presented simulation model (Table 1, Table 2) combine parameters on maintenance rate coefficient and energy contents from

the experiments in Costa et al. (2020a, b), complemented with literature parameters (Flye-Sainte-Marie et al., 2009; https://www.bio.vu.nl/thb/deb/deblab/add_my_pet) and the parameter estimation process outlined in Marques et al. (2020). In general, the parameter estimation procedure consists of executing a series of species-specific MATLAB script files containing the experimental data on, for instance, growth ("mydata" file), energy parameters determined experimentally, such as energy contents ("pars_init" file), and a file containing the DEB model description ("predict" file). We downloaded the most recent DEB model parameter estimates from the add_my_pet collection for our two species (with dates of 15 June 2019 for *R. philippinarum* and 15 July 2017 for *R. decussatus*). For each species and experimental condition (eight combinations), we added the newly obtained experimental data to the files and re-estimated the key parameters controlling food intake flux ($\{\dot{p}_{Xm}\}$), assimilation flux ($\{\dot{p}_{Am}\}$), somatic maintenance flux ($[\dot{p}_M]$),]) and energy at the initial simulation time ($E_{t=1}$) (Table 1). With these parameters and a simulation model projecting the DEB model equations, we examined the processes of energy allocation to growth and, indirectly via de κ-rule, to reproduction under different possible future scenarios (warming and acidification with or without TCS, against a control scenario of no change).

The simulations were carried out with an adaptation of the R code made available by Monaco and McQuaid (2019) at https://github.com/cristianmonaco/DEB-model-application-Mussel-zonation-and-reproductive-potential. The simulations encompassed 730 days (2 years), taking the current temperature regime similar to the ambient conditions in the Ria de Aveiro along the year: water temperature 17 ºC in autumn, 14 ºC in winter, 15ºC in spring, and 20 ºC in summer (Fig. 1) (summarized from http://riadeaveiro.web.ua.pt/index.php/ria-de-aveiro/parametros-abioticos, University of Aveiro, Laboratory of Ichthyology, Fisheries and Aquaculture). These temperatures were increased by 3 ºC in the scenarios simulating the conditions at the end of the 21st century. The conditions of pH were 8.1 in the current scenario and 7.7 under the future scenario. Fig. 1 summarizes the simulation conditions along one year.

The simulations comprised 2 years at daily scale (730 time steps, *t*), based on the mean energy content (sum of glycogen, lipids, and protein at the start of the simulation,

E[t=1]). At each time step the value of the fluxes and state variables in Annex1 were computed. Considering that *R. philippinarum* spawns between May and November-December in Portugal (Moura et al., 2018), we initiated the simulations at t=1 corresponding to 1st November with post-spawning individuals under "autumn regime." Likewise, for *R. decussatus* we initiated the model runs on 1st September with post-spawning individuals as this species spawns from July to September in the Atlantic Iberian peninsula (Urrutia et al., 1999). Individuals in the model were allowed to grow when $\kappa \cdot \dot{p}_C > \dot{p}_M$, i.e., the fraction of mobilized reserve was larger than the maintenance costs. Investment in reproduction occurred when $(1-\kappa) \cdot \dot{p}_C > \dot{p}_J$, i.e., the fraction mobilized for reproduction or maturity was larger than the costs of maturity maintenance. Spawning (emptying the reproduction buffer in the model) occurred from days 184 to 365 of each year (i.e., 1st May to 30 October) in *R philippinarum* and 245 to 365 in *R decussatus* (that is, 1st of June to 30 September).

3. Results

The feeding, assimilation and maintenance rates, converted from the experiments to energy rates, are shown in Table 1. For all of these parameters, the one to experience a more significant variation due to both stressors was the somatic maintenance rate. In *R. philippinarum*, maintenance rate $[\dot{p}_M]$ decreased by 67% (from 24.611 J d^{-1} cm^{-3} under normal conditions to 8.204 J d^{-1} cm^{-3}) in the presence of TCS, and up to 80% under high temperature / low pH (from 20.509 J d^{-1} cm^{-3} to 4.102 J d^{-1} cm^{-3}). In *R. decussatus*, the contrary was observed, with no large differences in maintenance rates at ambient temperature and an increase of 60% when exposed to TCS under the climate change scenario (from 10.255 to 16.407 J d^{-1} cm^{-3}).

Feeding and assimilation are intimately related in the equations of the DEB model. Assimilation decreased in *R. philippinarum* un-exposed to TCS (Table 1), and the combined temperature / acidification effect, together with TCS exposure, contributed to the highest decrease in assimilation rates $\{\dot{p}_{Am}\}$ by more than 80%: from 13.476 J d^{-1} cm^{-2} to 2.246 J d^{-1} cm^{-2}. In *R. decussatus* the reduction in assimilation rates was less marked, of the order of 30% in the worst case (from 9.125 J d^{-1} cm^{-2} under ambient

conditions to 6.083 J d^{-1} cm^{-2} under temperature / acidification regime), with limited effect of TCS exposure.

In *R. philippinarum*, energy contents (glycogen + protein + lipid) at the start of the simulation E$_{t=1}$ were enhanced by ca. 20% under the warmer / more acidic treatment condition, but the effect of TCS was negligible (Table 1). In *R. decussatus*, the highest energy content was displayed under ambient conditions in the absence of TCS, and decreased by ca. 30% at high temp / low pH, from 5686.97 J cm^{-3} to ca. 4100 J cm^{-3}, regardless of exposure to TCS.

Table 2 shows the values of other parameters necessary to run the simulation DEB model. In general, the energy contents of *R. decussatus* were higher than in *R. philippinarum* at the different life stages (E$_0$, E$_H^b$, E$_H^i$, E$_H^p$) and maximum (E$_m$).

The results of the DEB application are shown in Fig. 2 focusing on the indicators shell length, body dry mass, gonad dry mass and daily egg production, averaged over the 730 days (2 year) projection time. In both species, the warmer and more acidic conditions forecasted for the end of the 21st century would yield larger individuals, especially in *R. philippinarum* (mass increase of 16% against <1% for *R. decussatus*) but gonad mass and eggs produced did not vary significantly (Fig. 2). Conversely, the addition of TCS implied an important reduction of growth (shell length, body mass) in both species. It was more marked in *R. philippinarum* than in *R. decussatus*. In *R. philippinarum*, shell length decreased by 25% from 6.70 to 5.38 cm, but the corresponding decrease in mass was 50% larger, from 2.71 g$_{DW}$ to 1.88 g$_{DW}$ (Fig. 2). Even larger decreases were observed in the Manila clam under warmer and more acidic conditions in the presence of TCS: reduction of 33% in shell length and 60% in mass (Fig. 2), respectively. The effect of TCS on reproductive output was very significant in *R. philippinarum*, with a reduction of gonad mass and daily egg production of ca. 80% in present or forecasted scenarios with TCS (Fig. 2).

For *R. decussatus*, the effect of TCS on growth was less severe than in *R. philippinarum*, according to model outputs (Fig. 2). Growth in shell length was depressed when simulating exposure to TCS both under ambient conditions (15% reduction) and under warmer / more acidic conditions (10% reduction). Growth in terms

of mass is projected to decrease by 35% and 25%, respectively (Fig. 2). Gonad mass and egg production did not vary significantly between the present ambient and warmer / more acidic scenarios (0.19 g_{DW} and 0.18 g_{DW}, respectively, with a moderate decrease in daily eggs of only 10%) despite the 80% decrease of the Manila clam. In the native European clam, the addition of TCS produced a reduction of gonad mass under the ambient conditions of 32%, but the egg production is projected to increase by 6% under predicted climate change conditions (Fig. 2).

4. Discussion

The projection with the DEB model considering the parameters related to feeding, assimilation, maintenance and reserves showed contrasting results in *R. philippinarum* and *R. decussatus*, suggesting that the response of two co-generic bivalves can be different when exposed to the same stressors. The parameters estimated revealed that the maximum feeding ($\{\dot{p}_{Xm}\}$) and assimilation rates ($\{\dot{p}_{Am}\}$) were appreciably depressed in *R. philippinarum* under TCS exposure, regardless of temperature / acidification condition. These results suggest that this species responds to the chemical stressor by reducing food intake. Likewise, TCS exposure resulted in a strong reduction in maintenance costs. However, energy contents were not appreciably reduced by TCS exposure and even increased under the addition of climate change stressors , suggesting that the species does not mobilize reserves under stress. In *R. decussatus*, feeding and assimilation rates were not reduced by exposure to TCS, while the effect of warming and acidification caused only a slight reduction in these rates, suggesting that this species keeps feeding under stress. Maintenance costs did not vary in *R. decussatus* as much as in *R. philippinarum* under the four scenarios, but energy contents were the highest in the control (ambient, no TCS scenario), suggesting that the mobilization of reserves in *R. decussatus* is higher under stress than under normal conditions. .

The selected parameters were gathered and contrasted in terms of two main consequences: somatic growth (shell length and body dry mass) and reproductive output (gonadal weight and daily egg production), both of high economic relevance in shellfish production. In *R. philippinarum* somatic growth and reproduction would be severely

depressed in the scenarios of TCS exposure, while the projected effects of warmer / more acidic conditions, without TCS, would benefit this invasive species, with higher somatic growth and practically no change in reproductive output. In *R. decussatus* instead, we observed almost no effect of exclusively climate change projected conditions on somatic growth or reproductive output (the latter is inherently lower in *R. decussatus* than in *R. philippinarum*). The drug TCS would depress somatic growth in *R. decussatus* but to a lesser extent than in *R. philippinarum*.

As a common feature, DEB projections at the end of the simulation period showed that the reproductive output would be affected by exposure to an environmentally realistic TCS concentration of 1 μg/L. This result is consistent with the reported role of the antimicrobial agent TCS as an endocrine disruptor (Dann and Hontela, 2011; Bilal et al., 2020) and TCS has been shown to interfere in *in vitro* models with the enzymatic system involved in xenobiotic, lipid and hormone metabolism in mammals (Morrisseau et al., 2009; Lian et al., 2018) and in bivalves (Solé and Sánchez-Hernández, 2018). TCS (and other endocrine disruptors) can be accumulated in mussels (Gatidou et al., 2010; Kookana et al., 2013; Freitas et al., 2019; Pirone et al., 2019) and in the clams *R. decussatus* and *R. philippinarum* (Costa et al., 2020b). Nonetheless, in mussels (*Mytilus galloprovincialis*) experimentally exposed to TCS for 20 days under increased temperature and acidic conditions the bioconcentration factor (BCF) of this chemical was not enhanced (Serra-Compte et al., 2018). In *R. philippinarum*, but not *R. decussatus*, maintenance rates (measured as respiration) in the present experimental conditions decreased under exposure to TCS, but not under high temperature and low pH conditions; this trend is also observed with other stressors, such as diclofenac (Costa et al. 2020a) and nanoparticles (De Marchi et al., 2019), which suggests a higher response to the xenobiotics in the invasive species and resilience by lowering metabolic rates in the native clam.

Overall, bivalves inhabiting coastal areas have developed behavioral, physiological and biochemical strategies to cope with the simultaneous presence of multiple environmental stressors (i.e., contaminants, severe/ salinity, pH, or temperature fluctuations: Lannig et al., 2010; Carregosa et al., 2014; Serra-Compte et al., 2018). The native species may be more adapted to changes in their established environment and therefore be more ecologically competitive. However, the cultivation of the invasive,

economically more rewarding species such as *R. philippinarum* or *Crassostrea gigas*, can facilitate their dispersal and can compromise the ecological competitiveness of the native species. Laboratory studies contrasting the performance of bivalves in front of anthropogenic stressors (i.e., climate change factors and/or pollution) show species- and context-dependent outcomes (Carregosa et al., 2014; Domínguez et al., 2020; Mónaco and McQuaid, 2018; 2019; Perić et al., 2020).

The study of Carregosa et al. (2014) reported that the invasive *R. philippinarum* had higher survival rates than the native species *Venerupis corrugata* when subjected to stress due to low salinity. Domínguez et al. (2020) compared these same two species and a further two bivalves, *R. decussatus* and *Cerastoderma edule*, collected in a nearby area. Their results evidenced a complex species-specific pattern of behavioral and physiological responses to salinity stress that ultimately may reduce growth and increase predation. When considering the complexity of behavioral and physiological responses to a combination of stressors (with possible synergies) and the effects at the level of population growth and reproductive output, it is not clear if invasive bivalves will always outcompete native species. For instance, Perić et al. (2020) concluded that the exotic *Crassostrea gigas* is less capable of tolerating exposure to copper concentrations than the native *Ostrea eaulis* in the Adriatic Sea.

Scaling up from laboratory experiments to model-based results, a DEB model application to two mussels in South Africa showed that the exotic species would not necessarily outcompete the native one under climate change scenarios. The studies of Monaco and McQuaid (2018, 2019) in South Africa with a pair of intertidal mussels species, the native *Perna perna* and the invasive *Mytilus galloprovincialis*, indicated that the reproductive potential of both species was likely to increase under future temperature scenarios, although this beneficial effect was less evident in the invasive bivalve. Thus, the expansion potential of the invasive bivalve species may be compromised, if, as shown also in our DEB model application, anthropogenic factors, such as emerging contaminants, override the effect of climatic change. However, quick adaptation of invasive populations to new conditions cannot be ruled out, as seen with the highly invasive mussel *Musculista senhousia*, when exposed to low pH levels for several generations that resulted in increased reproductive fitness (Zhao et al., 2019).

As noted, the consequences of TCS exposure and projected climate change conditions would also affect somatic growth of both clams' species, with important consequences on the production by the aquaculture sector. Extrapolating from our results on somatic growth (Fig. 2), the economic implications of this growth reduction would be more severe in the invasive clam *R. philippinarum*, which represents 88% of the clam production in Europe (both species combined: 36 000 t in 2016, EUROSTAT[1]) and 80% in economic value (both species combined: 191 million € in 2016, EUROSTAT). In addition to depressed somatic growth due to TCS exposure under projected climate conditions, these clams, as well as other bivalves, bioaccumulate TCS (Costa et al. 2020b; Gatidou et al. 2010), and consumption of contaminated shellfish will pose increasing risks to human health (Bilal et al. 2020) and marine predators (Dann and Hontela, 2011).

5. Conclusions:

The consequence of TCS exposure is projected to be more negative on somatic growth and reproduction than climate change alone on both clam species (*R. philippinarum* and *R. decussatus*). Moreover, the synergy between emerging contaminants and climate change is not strong in these species because of the overriding TCS effect. Under warmer / more acidic conditions, the growth and reproductive output of the native clam *R. decussatus* would be less severely impacted than in the exotic *R. philippinarum*. In the long run, these adaptive resilience mechanisms may benefit the native species, in front of the invasive one, under chemical and physical anthropogenic pressure, even though Manila clam cultivation has been encouraged in recent decades in European markets.

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[1] EUROSTAT fisheries and aquaculture data viewer,
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Acknowledgements

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Appendix A – Dynamic Energy Budget Model

Dynamic Energy Budget (DEB) theory is detailed in the Kooijman (2010), with the parameter estimation techniques exposed in Kooijman et al. (2008; 2020). The model describes the general dynamics of growth and energy reserves of an animal in time, with several alternative parameterizations depending on hypotheses made on the animal ontogeny and data available. Here we used the standard DEB model with one parameter extension (metabolic acceleration) to account for growth differences from the larval to juvenile stage. The model bivalve is defined by energy (E) or mass (M) in each compartment: structure (V: structural volume), reserve (E), or maturity (H). We used energy in Joules (J) and structural volume in cm^3. Metabolic fluxes were calculated per day (d^{-1}). In the DEB model, the energy ingested from food (X) is first allocated to the reserve compartment (A "assimilation") and the flux of energy from reserves (C "mobilization") is the transferred to somatic maintenance (M) + growth (G) and to maturity (J) + reproduction buffer (R), according to a constant ratio κ ("kappa-rule". Fig. A.1). The energy flows and the dynamics of the state variables are given in Table A.1.
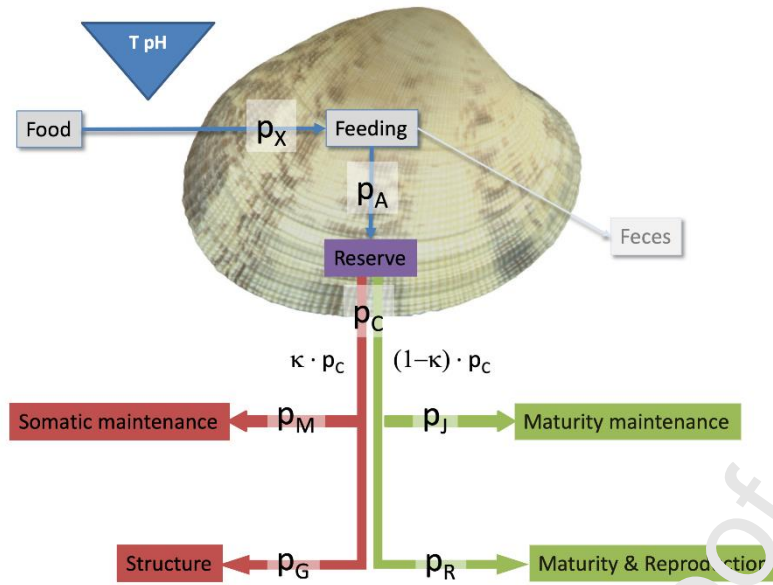
Fig. A.1. Summary of the main allocation rules and fluxes of energy in the DEB model applied to a model bivalve. Note that food was kept constant and abundant (f=1) but the clams were reared under different temperature and pH experimental conditions and exposure (or not) to triclosan.

Table A.1. DEB model equations describing the fluxes and dynamics of state variables in Fig. A.1. All fluxes were corrected by the thermal sensitivity equation on Arrhenius thermodynamics, as necessary (Kooijman 2010, chapter 1). Based on Tables 1, 2, 3 of Monaco and McQuaid (2019). The parameters estimated are shown in Table 1 of the main text.

Energy flux (dot) or state variable	equation	symbol
Ingestion flux (J d^{-1})	$$\dot{p}_X = \{\dot{p}_{Xm}\}V^{2/3}f$$	$\{\dot{p}_{Xm}\}$: Ingestion rate (J cm^{-2} d^{-1}) V : structural volume (cm^3) f : constant, abundant food, assumed $f=1$.
Assimilation flux (J d^{-1})	$$S_M = \frac{\min\left(V^{\frac{1}{3}}, L_J\right)}{L_b}$$ $$\{\dot{p}_{Am}\} = ae\{\dot{p}_{Xm}\}S_M$$ $$\dot{p}_A = \{\dot{p}_{Am}\}V^{2/3}f$$	S_M : metabolic acceleration factor L_b , L_J = structural length at birth and metamorphosis (cm) ae : assimilation efficiency $\{\dot{p}_{Am}\}$: Assimilation rate (J cm^{-2} d^{-1})
Mobilization flux (J d^{-1})	$$\dot{p}_C = \frac{E/V}{[E_G] + \kappa E/V}\left(\frac{[E_G]\{\dot{p}_{Am}\} V^{2/3}}{[E_m]}\right) + [\dot{p}_M]V$$	E : Energy in reserves (J) $[E_G]$: cost of structure (J cm^{-3} d^{-1}) κ : fraction of

		mobilized reserve allocated to soma *[E_m]* : maximum reserve capacity (J cm^{-3} d^{-1}) $[\dot{p}_M]$: cost of somatic maintenance (J cm^{-3} d^{-1})
Somatic maintenance (J d^{-1})	$$\dot{p}_M = [\dot{p}_M]V$$	
Somatic growth (J d^{-1})	$$\dot{p}_G = \kappa\dot{p}_C - \dot{p}_M$$	
Reproductive maintenance or maturity (J d^{-1})	$$\dot{p}_J = \min(V. V_p)[\dot{p}_M]\left(\frac{1-\kappa}{\kappa}\right)$$	V_p : structural volume at puberty (cm^3)
Reproduction (J d^{-1})	$$\dot{p}_R = (1-\kappa)\dot{p}_C - \dot{p}_J$$	
Reserve dynamics (J d^{-1})	$$\frac{dE}{dt} = \dot{p}_A - \dot{p}_C$$	
Structure dynamics (J d^{-1})	$$\frac{dV}{dt} = \frac{\dot{p}_G}{[E_G]}$$	
Reproduction buffer dynamics (J d^{-1})	$$\frac{dE_R}{dt} = \dot{p}_R\kappa_R$$	κ_R : fraction allocated to reproduction buffer
Biological metrics in physical length or dry mass		
shell length (cm)	$L_W = V^{1/3} / \delta_M$	δ_M = shape correction factor
soma dry mass (g)	$W_{sd} = V\ d_V + E\ \rho_E$	dV = density of

		structure (g cm^{-3}) ρ_E = inverse of energy contents of dry mass (g J^{-1})
gonad dry mass (g)	$W_{gd} = E_R\, \rho_E$	
Gonadosomatic index (GSI)	$GSI = W_{gd} / W_{sd}$	

The simulations were carried out with an adaptation of Monaco and McQuaid (2019) R code. https://github.com/cristianmonaco/DEB-model-application-Mussel-zonation-and-reproductive-potential

CRediT author statement

Francesc Maynou: Conceptualization, Methodology, Writing- Original draft preparation. **Silvana Costa:** Methodology, Investigation, Writing - Review & Editing **Rosa Freitas:** Supervision, Writing - Review & Editing, Funding acquisition **Montserrat Solé:** Writing - Review & Editing, Funding acquisition

Declaration of interests

☒ The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

☐The authors declare the following financial interests/personal relationships which may be considered as potential competing interests:
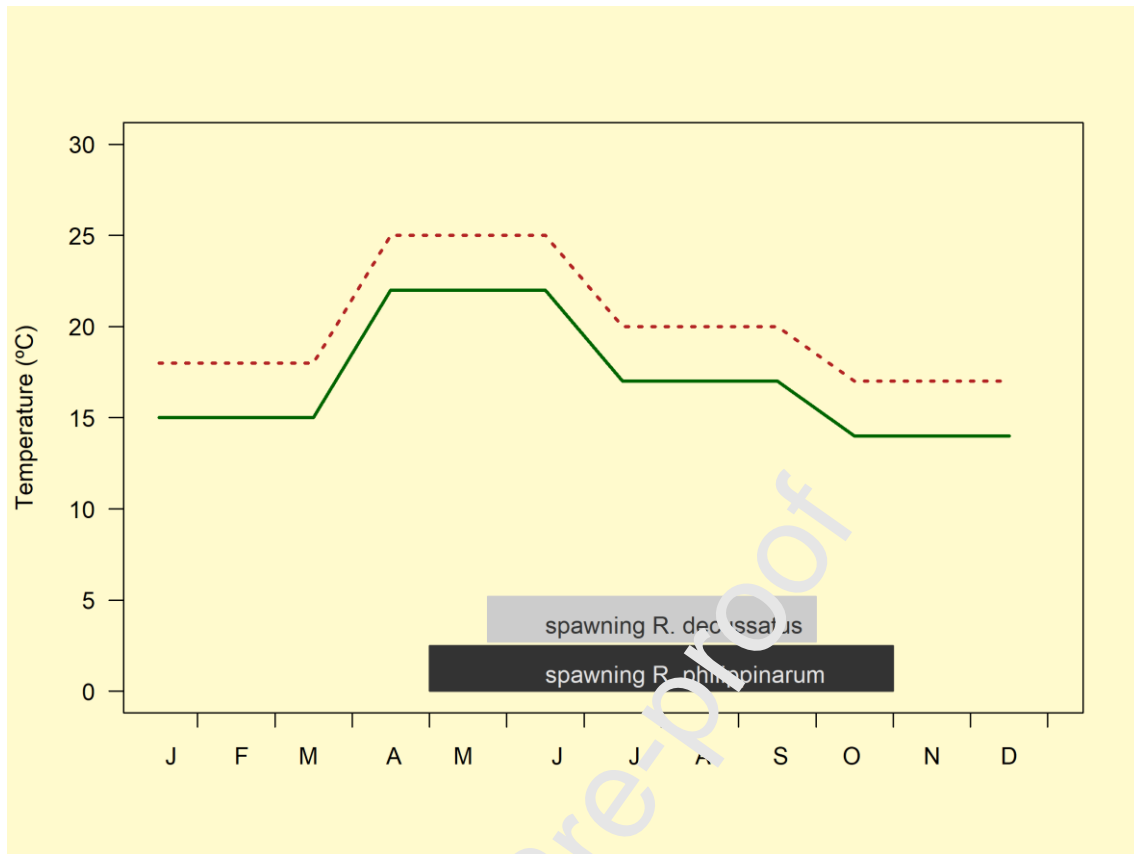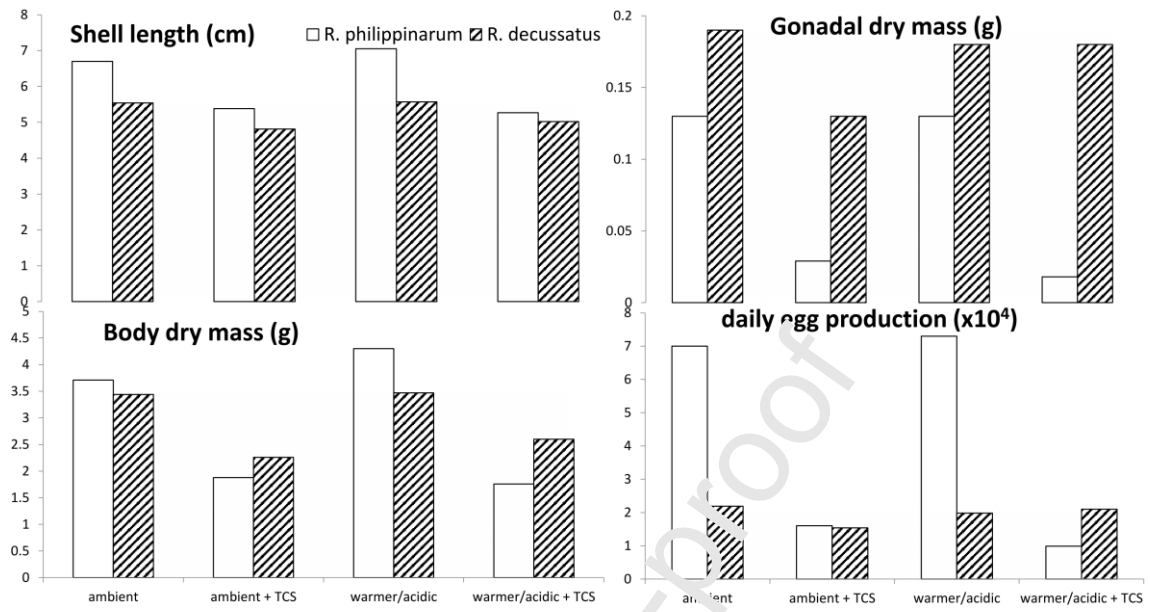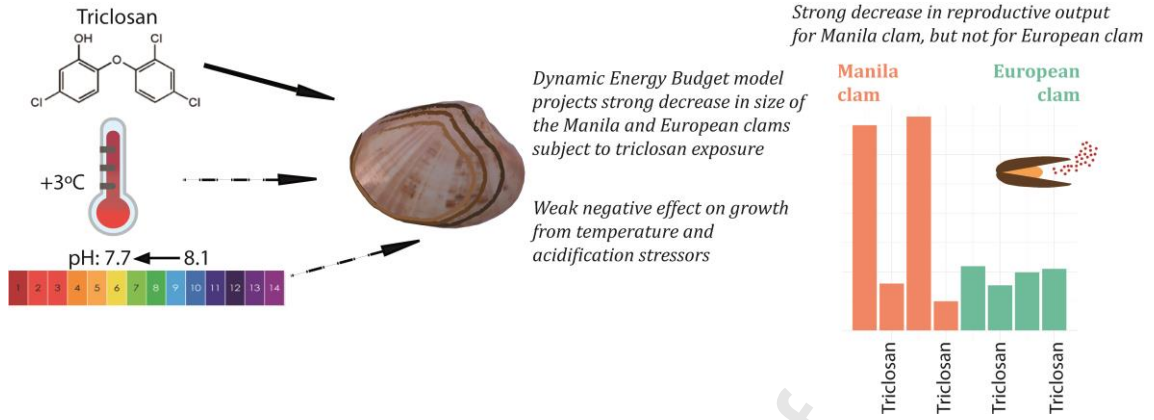
Figure 1

Figure 2

Table 1. Parameters of the DEB model for *Ruditapes philippinarum* and *Ruditapes decussatus* based on add_my_pet, Flye-Sainte-Marie et al. (2009) and estimated here from Costa et al. (2020a, b) experiments. Refer to Table A.1 for the symbols. Ambient: 17º C, pH 8.1; Climate Change (CC): 20º C, pH 7.7.

	Ruditapes philippinarum	*Ruditapes decussatus*
$\{\dot{p}_{Xm}\}$ (J d^{-1} cm^{-2}) ambient, no TCS	16.845	11.406
ambient, TCS	5.615	10.342
CC, no TCS	14.037	7.604
CC, TCS	2.807	9.733
$\{\dot{p}_{Am}\}$ (J d^{-1} cm^{-2}) ambient, no TCS	13.476	9.125
ambient, TCS	4.492	8.2'3
CC, no TCS	11.230	6.083
CC, TCS	2.246	7.787
$[\dot{p}_M]$ (J d^{-1} cm^{-3}) ambient, no TCS	24.611	15.382
ambient, TCS	8.204	14.433
CC, no TCS	20.509	10.255
CC, TCS	4.102	16.407
$E_{t=1}$ (J cm^{-3}) ambient, no TCS	4346.83	5686.97
ambient, TCS	4292.92	4772.01
CC, no TCS	5222.39	4094.55
CC, TCS	5989.37	4147.55

Table 2. Parameters taken from the literature Flye-Sainte-Marie et al. 2009; Urrutia et al. 1999) or from the *addmypet* database (https://www.bio.vu.nl/thb/deb/deblab/add_my_pet) and kept constant through the simulation horizon.

	Ruditapes philippinarum	*Ruditapes decussatus*
d_V (g cm^{-3})	0.100	0.100
δ_M	0.427	0.514
y_{VE} (J J^{-1})	0.976	0.976
$\dot{v}$ (cm d^{-1})	5.82 x 10^{-3}	1.32 x 10^{-2}
$\dot{k}_J$ (d^{-1})	2.00 x 10^{-3}	2.00 x 10^{-3}
κ	0.975	0.950
κ_R	0.950	0.950
g	4.100	4.208
[E_G] (J cm^{-3})	2357.884	2357.884
E_m (J cm^{-3})	589.718	1496.06
ρE (g J^{-1})	9.83 x 10^{-5}	7.49 x 10^{-5}
L_b (cm)	0.003	0.04
L_j (cm)	0.019	0.031
L_p (cm)	0.715	1.081
L_i (cm)	2.13	3.496
a_b (d)	2.409	1.925
a_j (d)	18.436	15.236
a_p (d)	399.284	354.99
a_m (d)	2059.800	2936
E_0 (J)	6.71 x 10^{-5}	1.18 x 10^{-3}
E_H^b (J)	4.48 x 10^{-7}	5.12 x 10^{-6}
E_H^j (J)	1.34 10^{-4}	5.21 x 10^{-6}
E_H^p (J)	8.885	27.1
T_A (º K)	6071	6071
T_L (º K)	290.15	290.15
T_H (º K)	300	300
T_{AL} (º K)	30424	30424
T_{AH} (º K)	299859	299859
T_{ref} (º K)	293.15	293.15

Graphical abstract

Highlights:

- Environmental triclosan levels alter the reproductive output of *R. philippinarum*
- Environmental triclosan levels reduce body mass in *R. philippinarum*
- *R. decussatus* growth was resilient to environmental changes
- Worst case scenario (TCS and climate change) will affect Manila clam production