

# Physiological energetics of the ascidian *Styela clava* in relation to body size and temperature

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## Abstract

The ascidian *Styela clava* is widely distributed in northern China and is thought to be important in the functioning of estuarine systems. This ascidian may experience highly variable physiological conditions, and its physiological responses to these are of interest considering its ecological role. This study presents data on the physiological parameters in relation to body size and temperature (12–28 °C) of *S. clava*. Respiration, excretion, feces, ingestion and absorption energy were positive related to size and its mass exponents (b-values) varied from 0.2930 to 0.7488. Respiration energy increased with increasing, but critical values were found at 20 °C for energy of ingestion and absorption, while 24 °C for energy in feces and excretion. At the range of 16–24 °C, the scope for growth, gross and net growth efficiencies of ascidians increased with increasing temperature. The energy budget equations of different sized ascidians were obtained at different temperatures. Excretion energy shared a minimal fraction in ingestion energy (1.30–2.47%), the ratios of feces energy and respiration energy to ingestion energy were 46.53–64.27% and 10.26–80.75%, respectively. The physiological data obtained in the present study indicated that *S. clava* could adjust its energy budget according to the environment and its physiological conditions to meet their nutritional and energetic demands. In the range of experimental temperature (12–28 °C), 16–20 °C was suitable for the rearing of *S. clava* to achieve optimum development, while deficient metabolic adjustment induced a negative scope for growth of *S. clava* at 28 °C.

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## 1. Introduction

The solitary ascidian, *Styela clava*, distributed widely in Bohai and western Yellow seas, is an efficient filter-feeder capable of depleting the water volume of suspended particles. Large populations of ascidians can affect the environment through phytoplankton consumption, nutrient cycling, and biodeposition (Jørgensen, 1990). The physiological energetics of marine organisms is important for understanding their adaptations to particular habitat conditions. Most physiological studies of filter-feeding animals have been done on bivalves,

and only a few studies have focused on ascidian, especially for *S. clava*. Despite its abundance, few studies have focused on this group, and each study has only addressed one aspect of physiological energetics, such as oxygen consumption or nitrogen excretion (Zhang et al., 2000; Zhang and Fang, 2000). Our current study was particularly concerned with the energy budget of *S. clava* as influenced by body size and water temperature. No information on these aspects of the physiology of this species currently exists in the scientific literature.

A physiological response represents the sum of all cellular and biochemical reactions as influenced by the environment or the animal itself. For this reason, organisms are capable of reflecting any environmental deterioration even before the effects are manifested in the population or community as a whole (Bayne et al., 1985). Organisms encounter and assimilate resources (henceforth termed “food”) from the environment. The energy content of the food is transformed into a

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metabolically inactive storage and distribution form (such as protein or fat) that we term “reserves”. Energy is subsequently extracted from reserves and used for metabolic maintenance, growth and reproduction. The balance energy equation (Winberg, 1956) has been used as a conceptual framework in examining variation in organismal performance. Bayne and Newell (1983) expressed the equations as  $C = F + U + R + P$ , where  $C$  is energy consumed,  $F$  is energy excreted through feces,  $U$  is energy lost as other excretion methods,  $R$  is the metabolism energy, including the metabolic lost of body maintenance ( $R_m$ ) and all other metabolic costs ( $R_r$ ) (Koehn and Bayne, 1989),  $P$  is the growth (increment) energy containing somatic production  $P_g$  and reproductive production (gonads)  $P_r$ , sometimes named scope for growth (SFG) (Warren and Davis, 1967). The equation can be transferred to another style  $P_g + P_r = C \cdot AE - (R_m + R_r)$ , where AE is the efficiency with which consumed energy is absorbed. This energy will vary according to the effects of different extrinsic and intrinsic factors and therefore it is important to calculate the cost of production (in terms of growth) when considering total aquaculture activity.

Feed composition and ingestion are the most important factors to consider in order to balance the growth equation; from the moment where the animal decides whether to ingest or reject each item of food, the amount ingested, and the process of digestion and absorption, after which the animal's physiological systems deal with the influx of nutrients to convert the food into new tissue or metabolic expenditure. In addition, metabolic rate is a major component of the balanced growth equation; this is entirely a loss term that provides a measure of the energetic cost to the system of supporting the animal. Thus, both the measurement and interpretation of any physiological response should be not considered in isolation but in the context of the balanced growth equation or other physiological integrations, considering all the extrinsic and intrinsic factors. Therefore, it is necessary to establish the limits of the extrinsic and intrinsic factors for the validity of a physiological study. The main extrinsic factors are related to fluctuations in the biotic and abiotic conditions within the water column, whereas an intrinsic factor refers to internal characteristic such as body size. The first set of factors can be controlled experimentally, whereas the intrinsic factors are determined from the sampled population. It has been demonstrated that size (intrinsic) and temperature (extrinsic) are two of the most important factors for determining the energetic cost within any particular species (Bayne et al., 1985).

The aim of the present work was to evaluate the physiological responses from different size groups of ascidians in controlled culture conditions, to create a balanced growth equation throughout an allometric relationship, providing a basic framework for physiological research in *S. clava*.

## 2. Materials and methods

### 2.1. Biological material

Specimens of *S. clava* were collected from the intertidal zone of Yantai, Shandong Province in May. The ascidians were

transferred into aquariums to acclimatize at ambient temperature for at least 10 days, and were fed with microalgae *Isochrysis galbana* until the experiments. Then the water temperature was gradually adjusted to experimental temperature at a rate of 0.5 °C per day. Ascidians were kept at the experimental temperature for at least 48 h prior to testing. Five experimental temperatures (12, 16, 20, 24 and 28 °C) and five mass classes of ascidians (1.5–2.0 g, 2.5–3.5 g, 5.0–6.5 g, 8.0–9.5 g and 11.0–14.0 g) were used.

### 2.2. Experimental methods

Filtered seawater of ambient salinity (30±2‰) was delivered into a 400 L header tank continually, and gravity fed through delivering lines to twelve 10 L flow feeding chambers. Flow rate was held constant over time and among the 12 chambers at 30 L h<sup>-1</sup>. An inflow aperture fixed on the bottom of the tank and an outflow aperture fixed 2 cm from the top of each culture chamber maintained water volume at 10.0 L. Each experiment comprised five duplicate culture chambers (contained 20 ascidians) and two controls that were used to determine the concentration of suspended particles in the system and to estimate the quality of particles removed by ascidians. The experimental animals were allowed to acclimate to conditions in the feeding chambers for at least 10 h before testing.

The diet concentration (3.06±0.11 mg POM L<sup>-1</sup>) in the inflow water was regulated by adding laboratory cultured microalgae in the header tank. The diet concentrations in the inflow and outflow water as well as feces were monitored as described by Zhuang et al (2004), and the clearance (CR) and assimilation efficiency (AE) of ascidians were calculated as detailed below.

Immediately after feeding for 10–12 h, the ascidians were divided into two groups randomly to determine their oxygen uptake and ammonia excretion, respectively with the method reported by Zhuang et al (2004). The dry masses of the ascidians used in these experiments were precisely measured after they were dried at 60 °C.

### 2.3. Computing method

#### 2.3.1. Clearance rate (CR)

Clearance rate is estimated using the formula of Coughlan et al. (1992):

$$CR = V \times \frac{\ln[(C_{e0} - C_{e0} \times S_{ed})/C_{et}]}{N \times t}$$

Where  $V$  indicates the experimental seawater volume (L),  $N$  is the number of animals used in the experiment (ind),  $C_{e0}$  and  $C_{et}$  are the diet concentration (mg TPM L<sup>-1</sup>) in the inflow and outflow water of the culture chamber, respectively.  $S_{ed}$  refers to the variation coefficient in diet concentration of controls,  $S_{ed} = \frac{C'_{e0} - C'_{et}}{C'_{e0}}$ , where  $C'_{e0}$  and  $C'_{et}$  are the diet concentration (mg TPM L<sup>-1</sup>) in the inflow and outflow water of the control chamber, respectively, and  $t$  is the duration of the experiment (h).

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