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THE EFFECT OF ABRUPT CHANGES IN SALINITY ON THE SFG OF THE MUSSELS

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ABSTRACT

Mytilus edulis of over one year old were exposed to three different proportion of fresh and salt water for seven days. The objective of the study is to evaluate Scope For Growth (SFG), using the physiological responses of mussels as a potential monitor of the impact of environmental stresses. The feeding rates, respiration rates, food absorption efficiencies, and SFG of the mussels were measured following the seven day exposure period. Clearance rates relatively showed a small decrease with exposure to decreasing salinities, but it was not statistically significant. Absorption efficiencies of the mussels in the three groups were generally not different. Respiration rates among the three groups were not affected by, but changed consistently with increasing proportions of fresh water. The SFG among the three groups did not differ significantly by Anova, but the P value (0.069) is very close to the level considered significant (0.05). This study suggests that the decreasing salinity "stressed" the mussels.

Keywords : Scope For Growth (SFG), environmental toxicology

I. Introduction

Environmental stresses can affect the physiological functions of *M. edulis* such as its growth and reproduction. It is difficult to measure bivalve molluscs' growth and production directly because a large proportion of the total production can be lost in the form of gametes (Bayne *et al.* 1985; Widdows and Johnson 1988). It is impractical to measure the tissue somatic growth or weight change of an individual due to the presence of the shell, and also because growth takes a long time to occur (Widdows and Johnson 1988; Widdows 1992). It is also not sufficient to measure changes in shell length because shell growth and tissue growth are not necessarily directly related (Widdows and Johnson 1988; Widdows 1992). For example, shell growth can continue during starvation while tissue mass declines. Therefore, the ratio of shell length to tissue mass can not be used to estimate other growth components, such as somatic and gonadal contributions (Bayne and Newell

1983; Bayne et al. 1985; Widdows 1992). Bivalve growth is also difficult to quantify and interpret in relation to environmental pollution due to the complication of separating nutritional effects from toxicant effects (Widdows and Donkin 1992).

Many of the problems mentioned above can be overcome by measuring "Scope For Growth", a fundamental physiological calculation of all living organisms (Bayne and Newell 1983; Bayne et al. 1985; Capuzzo 1988). Scope For Growth (SFG) is the whole integrated response of an organism to its total environmental stimuli, including both natural and anthropogenic stressors (Bayne et al. 1985; Widdows and Johnson 1988). SFG is determined by measuring the energy ingested which is available for growth and reproduction by subtracting the amount of energy respired and excreted from the amount of energy absorbed from the food (Bayne et al. 1985; Widdows and Johnson 1988).

SFG can range from positive values, under the best conditions when there is optimum energy available for growth and production of gametes, to negative values, when the animal is severely stressed and is utilizing its body reserves for maintenance metabolism (Widdows 1985b; Widdows and Johnson 1988). SFG is particularly useful as a physiological stress index in assessing the biological effects of pollution, especially when it is combined with an analysis of chemical contaminants in the bivalve's tissues (Lack and Widdows 1986). An analysis of chemical contaminants provides insight into the causes of toxicity and which chemical compositions produce adverse biological responses which affect changes in growth rate (Widdows 1992). Advances in SFG monitoring procedures using bivalve mussels have allowed scientists to distinguish small gradients of pollution effects that were not detectable a decade ago (Bayne et al. 1988). Thus, SFG has been proposed as a valuable pollution monitoring tool (Bayne et al. 1985). Moreover, SFG can be measured over a short period of time and is cost effective as it does not require expensive equipment (Johnson 1988). It can be measured in the field using a mobile laboratory (Widdows et al. 1987) or in the laboratory under standardized conditions (Widdows and Johnson 1988).

II. Study Objectives

The main objective of this study was to assess SFG, using the physiological responses of mussels (*M. edulis*), as a potential monitor of the impact of environmental stresses caused by pulp waste. This would provide insight into the growth process and how it might be disrupted by environmental stresses and pollution in a controlled experiment.

Data obtained from this study will also provide a reference for future biomonitoring studies and will contribute to study in waste management and marine environmental protection.

III. Material and Methods

3.1.. Experimental organisms

Mussels for Experiment were collected in summer months and their shell

lengths ranged from 61 to 67.5 mm. All mussels were over one year old, and were obtained from a "clean" population. Individuals were cleaned of epibionts, detritus and fouling organisms under cold running freshwater. Mussels then were acclimated in a flowing seawater tank, and fed with *Isochrysis galbana* Green, temperature 12^0 C, the salinity was approximately 32 ‰, in the laboratory for one week (Widdows *et al.* 1983; Widdows and Johnson 1988).

After this acclimatization periode, mussels were exposed to various concentrations of freshwater for seven days. The exposures were conducted under static systems. Eight mussels were used for each treatment in Experiments. Individual mussels were numbered using a white water- resistant marker pen. After exposure in Experiment, the physiological responses of the test animals (clearance rate, food absorption efficiency, respiration and excretion rate) were determined in a static system.

3.2. Experiment

This experiment was used to determine the effect of freshwater (the decline in salinity) on SFG of mussels. Mussels (*M. edulis*) were divided into three groups. Each group consisted of 8 mussels (61 - 67.5 mm shell length). Group 1 was used as a control and exposed to 100 % seawater. Group 2 was exposed to 30 % freshwater and 70 % seawater, and group 3 to 70 % freshwater and 30 % seawater. All groups were exposed for seven days. The salinity of 100 % seawater was approximately 32 ‰. 30 % freshwater was approximately 23 ‰, and the salinity of 70 % freshwater was approximately 10 ‰.

3.3. Physiological measurement:

Physiological rates were converted to mass-specific rates for mussels of 1 g dry weight using appropriate weight exponents (b) in the allometric equation (Widdows and Johnson 1988). This experiment used 1.000 weight exponent because the range of weights of individual mussels was small and the weight was close to 1 g. Individual physiological responses were converted into energy equivalents and used in the balanced energy equation:

$$P = C - R - U - F$$

where:

P = production of both somatic tissue and gametes
C = total consumption of food energy
R = respiratory energy expenditure
U = energy loss as excreta
F = faecal energy loss

In this study, U was ignored because it is usually not significant in the calculation of SFG in energy units (Widdows 1991b).

Calculation of C, and R (all $J.g^{-1}.h^{-1}$) is as follows:

C = clearance rate ($L.\ g^{-1}.h^{-1}$) * food energy value ($J.\ L^{-1}$) * absorption efficiency.

R = Vo_2 ($ml\ O_2.\ g^{-1}.h^{-1}$) * 20.33 $J.ml^{-1}\ O_2$

(Widdows and Johnson, 1988)

The clearance rate (C.R.) by individual mussels was calculated using the following formula:

$$C.R. = 2 \text{ liters} * \frac{(\log_e C_0 - \log_e C_t)}{\text{time interval (hours)}}$$

To calculate the absorption efficiency (AE) was using the following equation:

$$AE = \frac{(F-E)}{(1-E) * F} * 100$$

In this study, the food energy value was calculated to be 9.522 $J.L^{-1}$ which was converted from the data of Whyte (1987).

The rate of oxygen consumption is calculated as follows:

$$Vo_2 = 60\,[C(t_0) - C(t_x)]\,(V_r - V_a)/(t_x - t_0)$$

IV. Results

Physiological responses, including clearance rates, respiration rates and food absorption efficiencies of *M. edulis* after exposure to three concentrations of freshwater; 0 % (control), 30 % and 70 %, for seven days, are presented in Table 1. In all stages of the experiment, *M. edulis,* in both the control and freshwater treatments, displayed widely open shell valves, representative of mussels in an unstressed state.

4.1. Clearance rates and absorption efficiencies

As shown in Table 1, mussels appeared to show a relatively small decline in the clearance rate with exposure to increasing concentrations of freshwater. The clearance rates of mussels from El_{30} were 7 % lower than the rates of mussels from C1 (control), but this reduction was not statistically significant (Table 2). The clearance rates of mussels from El_{70} were 17 % lower than the rates of mussels from C1, but this was also not statistically significant (Table 2). Compared to C1 and El_{30}, mussels from El_{70} showed the lowest feeding rates.

Absorption efficiencies of mussels in the three groups were generally not different. There was no consistent change in the value with increasing proportions of freshwater (Table 1).

4.2. Respiration rates

Respiration rates or oxygen consumption among the three groups of mussels were similar. Respiration rates were not affected by, but changed consistently with increasing concentrations of freshwater (Table 3). Analyses of variance (Table 5) confirmed that the treatment had no significant effect on the respiration rates of mussels ($p>0.05$).

4.3. Scope For Growth (SFG)

The physiological processes representing components of the balanced energy equation were converted to energy equivalents in order to calculate Scope For Growth and growth efficiencies for mussels from each group (Table 4).

After seven days of exposure there appeared to be a reduction in the SFG of mussels exposed to 30 % freshwater ($E1_{30}$) relative to the control group (C1), but this was not statistically significant (Table 5). Mussels exposed to 70 % freshwater ($E1_{70}$) had positive SFG values and growth efficiencies that were also statistically not significantly different from the control mussels, although mussels from the control group had slightly higher SFG values (Table 5).

Although SFG in the three groups did not differ significantly by ANOVA (Table 5), the P value (0.069) is very close to the level considered significant (0.05). In addition, there were very clear trends in clearance rates, respiration rates and in SFG, to indicate that the decreasing salinity "stressed" the animals. Figure 1 shows a histogram of SFG of *M. edulis* as a function of salinity.

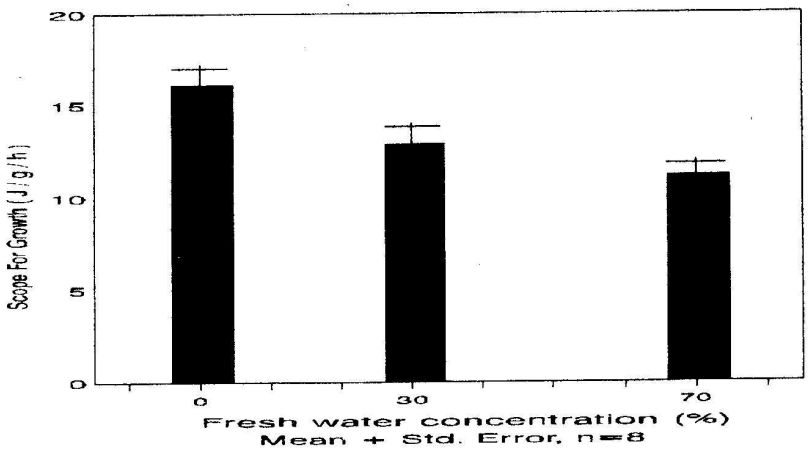

Figure 1.
Scope For Growth ($J.g^{-1}.h^{-1}$, Mean and Standard Error) in *Mytilus edulis* ater Exposure to Three Different Proportions of Fresh and Salt Water for Seven Days

Table 1:

Physiological responses of *Mytilus edulis* after exposure to three different concentrations of freshwater for seven days (mean ± SE, n = 8).

Group number	Shell length (mm)	Dry tissue (g)	Feeding rates		Absorption Efficiency	Respiration rate	
			Clearance ($L.g^{-1}.h^{-1}$)	Ingestion ($J.g^{-1}.h^{-1}$)		($mmolO_2.g^{-1}.h^{-1}$)	($J.g^{-1}.h^{-1}$)
C1	64.75±0.76	1.007±0.09	4.915±0.230	46.805±2.192	0.45±0.02	10.914±1.039	4.977±0.474
$E1_{30}$	63.95±0.29	0.968±0.08	4.577±0.266	43.580±2.531	0.41±0.02	11.050±0.729	5.039±0.333
$E1_{70}$	63.44±0.60	0.911±0.05	4.082±0.227	38.870±2.157	0.42±0.02	11.405±0.894	5.200±0.408

C1 = control (100 % seawater + 0 % freshwater)
$E1_{30}$ = 70 % seawater + 30 % freshwater
$E1_{70}$ = 30 % seawater + 70 % freshwater

Table 2:

Statistical analyses (ANOVA) of energy consumed ($J.g^{-1}.h^{-1}$) calculated from clearance rate of *Mytilus edulis* after exposure to three different concentrations of freshwater for seven days.

Source	D.F	Sum of Squares	Mean Squares	F Ratio	P Probability
Between Treatments	2	254.81	127.40	2.63	0.09
Within Treatments	21	1015.41	48.35		
Total	23	1270.22			

Table 3.

Statistical analyses (ANOVA) of energy consumed ($J.g^{-1}.h^{-1}$) calculated from respiration rate of *Mytilus edulis* after exposure to three different concentrations of freshwater for seven days.

Source	D.F	Sum of Squares	Mean Squares	F Ratio	P Probability
Between Treatments	2	0.21	0.11	0.77	0.93
Within Treatments	21	32.08	1.53		
Total	23	32.29			

Table 4

Components of the energy budget and Scope For Growth ($J.g^{-1}.h^{-1}$) of *Mytilus edulis* from the three groups after seven days exposure to three different concentrations of freshwater (mean ± SE, n = 8).

Group number	Energy absorbed A	Energy respired R	Scope for growth SFG = A-R	Net Growth efficiency (A-R)/A
C1	21.062 ± 0.987	4.9766 ± 0.506	16.085 ± 0.935	0.76
$E1_{30}$	17.868 ± 1.038	5.0389 ± 0.355	12.829 ± 1.006	0.72
$E1_{70}$	16.325 + 0.906	5.2005 + 0.436	11.125 + 0.704	0.68

Table 5

Statistical analyses (ANOVA) of Scope For Growth of *Mytilus edulis* ($J.g^{-1}.h^{-1}$) after seven days exposure to three different concentrations of freshwater.

Source	D.F	Sum of Squares	Mean Squares	F Ratio	P Probability
Between Treatments	2	101.6399	50.8200	7.993	0.069
Within Treatments	21	133.4141	6.3531		
Total	23	235.0541			

V. Discussion

This experiment was conducted to assess how physiological energetics or SFG (including its components) of mussels might be affected by freshwater (abrupt salinity changes). In this study, mussels were exposed to seawater diluted with freshwater, which resulted in salinities of approximately 23 ‰ and 10 ‰.

In this experiment mussels were expected to experience stress when exposed to freshwater due to a decline in salinity. Stress was expected to be greatest when the concentration of freshwater was highest. The results of this experiment indicate that mussels exposed to various concentrations of freshwater show a slight decrease in SFG, however, this reduction is not statistically significant (Table 4).

Widdows (1985a) found that adult mussels exposed to an abrupt decline in salinity from 30 ‰ to 15 ‰ showed significantly decreased physiological responses, including decreased clearance rate, oxygen consumption and SFG. After 24 to 48 hours these mussels closed their valves and their haemolymph osmolality declined gradually. Oxygen consumption and clearance rate ceased immediately upon transfer to 15 ‰. The same experiment also showed that each physiological response had a different rate of adaptation to an abrupt salinity change. A steady-state was achieved first in haemolymph osmolality, followed by the rate of oxygen consumption, clearance rate, food absorption and finally SFG. This is in agreement with earlier observations by other authors (Shumway 1977a,b; Davenport and Fletcher 1978) who noted that mussels closed their valves at salinities below 20 ‰. Davenport (1979) later found that the exhalent siphon closed below 20 ‰ and there was only partial valve closure. This would result in the break of feeding activity, while oxygen uptake continued at a reduced rate.

In this present study the physiological responses of mussels exposed to freshwater were only measured after the seventh day of exposure, instead of every day due to the lack of equipment available in the laboratory. Physiological measurements, including respiration rates, clearance rates, food absorption efficiency and SFG after seven days of exposure showed insignificant declines. These results are not in agreement with those of the previous studies, however, this probably reflects differences in the duration of the studies. After seven days of exposure those mussels probably would have adapted to the changes in salinity, and therefore, reached a steady-state. Widdows (1985a) noted that within 24 hours the respiration rate had returned to the initial rate recorded at 30 ‰, and within eight days the clearance rate had reached a steady-state not significantly different from the rate at 30 ‰. According to Widdows (1985a), SFG was negative for six days after an abrupt decrease in salinity but then rapidly increased to a positive steady-state value by the eighth day.

Alternatively, differences between these results and those of previous studies could possibly be due to differing experimental conditions and procedures. For example, the duration of exposure may have varied and test solutions may or may not have been renewed.

VI. Conclusion

Responses by individual mussels to changes in the environment, including the addition of pollutants, can be measured as physiological events. Physiological responses can be used as general indices of the effects of pollution because they relate directly to the fitness of the individual (Bayne, 1986). Physiological indices, such as clearance rate, respiration rate and Scope For Growth, showed a response to abrupt changes in salinity.

This study demonstrated that SFG can be measured over a short period of time. It also demonstrated that these measurements are cost effective, as they do not require expensive equipment, and can be measured conveniently in the laboratory under standardized condition. SFG measurement also can be applied elsewhere, in temperate, subtropical and tropical countries using local bivalves.

Clearly this study is very preliminary and further work is required. It would be useful to examine the rate and extent of physiological responses of mussels to many more concentrations of pollution in order to find the best correlation between Scope For Growth and the toxicants. It would also be desirable to examine the rate and extent of physiological responses of mussels to selected toxicants in order to understand the toxicants' mode of action, and possibly to predict the biological consequences in terms of Scope For Growth.

Chemicals' tissue concentrations of the mussels should also be examined to determine how they correlate to functional abnormalities and to Scope For Growth. The relationship between tissue concentration and physiological responses derived from such studies could not only facilitate the identification of chemical contaminants causing effects recorded in the environment, but also enable biological effects to be predicted from environmental levels of contaminants in water and body tissues.

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