

as gaseous ammonia have large stores of CaCO_3 in the outer shell of the body. Possession of haemocyanin is another characteristic common to both gastropods and isopods. Perhaps this respiratory pigment is optimally adjusted for operation in a medium rich in calcium⁷ as well as in the alkaline milieu required for the maintenance of a gradient along which ammonia can diffuse from blood to tissues¹.

The molecular causes of the seasonal variation in glutaminase activity are not yet clear, mainly because the membrane-bound enzyme has so far withstood attempts to solubilize it without too great a reduction in its activity. In summer it proved to be easier to obtain reproducible curves with the 'G-25' treated homogenate than in winter. A month-by-month check of enzyme activity suggests that at this time of year changes in enzyme activity are due to the interference of a competitive inhibitor (Fig. 2). However, the increase in activity observed at high substrate concentrations in winter is not accompanied by an increase in substrate affinity. The enzyme appears to be part of a complex system that adjusts the rate of ammonia production and the storing capability of terrestrial isopods to the physiological state of the organism which, in turn, is controlled by seasonal variables.

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W. WIESER

Institut für Zoophysiologie der Universität,
Innsbruck

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- Speeg, K. V., and Campbell, J. W., *Amer. J. Physiol.*, **214**, 1392 (1968).
- Wieser, W., and Schweizer, G., *J. Exp. Biol.*, **52**, 267 (1970).
- Hartenstein, R., *Amer. Zoologist*, **8**, 507 (1968).
- Meister, A., *Biochemistry of the Amino Acids*, vols. 1 and 2 (Academic Press, New York and London, 1965).
- Sayre, F. W., and Roberts, E., *J. Biol. Chem.*, **233**, 1128 (1958).
- Wieser, W., Schweizer, G., and Hartenstein, R., *Oecologia*, **3**, 390 (1969).
- Burton, R. F., *Comp. Biochem. Physiol.*, **41A**, 555 (1972).

Possible Adverse Effect of Protozoa on Effluent Purification Systems

EFFLUENT purification systems based on the "activated sludge system" contain a naturally occurring population of microorganisms which can be represented as a continuous-flow culture of either chemostat or plug-flow type. In the ideal chemostat type the culture is completely mixed, whereas in the ideal plug-flow type it flows through the vessel without mixing. Both types of system are used, but the choice seems arbitrary although recent theory suggests that the chemostat system is better¹. In view of the large capital sums now being invested in sewage disposal, the arbitrary design of the activated sludge process² calls for urgent investigation. A case in point is the effect of protozoa on the process.

An effluent is biologically purified by the utilization of the soluble waste as a nutrient or substrate for microbial growth and metabolism. It is assumed that bacteria are the principal agents of soluble waste removal. There are also protozoa which consume the bacteria and thereby contribute to clarification of the effluent. Canale³ and Curds⁴ have analysed the relations of the bacterial prey and the protozoan predator in a chemostat process, and show that the three variables, prey, predator and nutrient (sewage) concentrations, may either approach true steady states or oscillate continuously, depending on the microbial growth constants and the dilution rate of the system. In Fig. 1 steady state values have been plotted as a function of dilution rate using growth constants feasible for protozoa as predators and bacteria as prey. Similar results have been obtained by Curds⁴, using different values for the microbial growth constants. The protozoa are washed out of the system if the dilution rate is above the critical value (0.39 h^{-1}) up to which value, in steady states, the specific growth

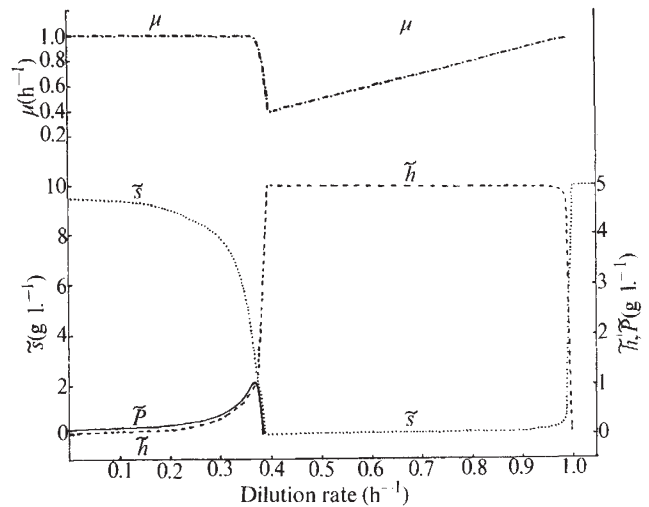


Fig. 1 Steady state values of parameters of a prey and predator culture (for example, bacteria and protozoa) in a chemostat. The fixed parameters were: $\mu_m = 1.0 \text{ h}^{-1}$; $K_s = 0.01 \text{ g l}^{-1}$; $\lambda_m = 0.4 \text{ h}^{-1}$; $K_h = 0.10 \text{ g l}^{-1}$; $Y = 0.5 \text{ g prey g}^{-1} \text{ growth-limiting nutrient}$; $W = 0.5 \text{ g predator g}^{-1} \text{ prey}$; $s_r = 10.0 \text{ g l}^{-1}$. The variables are: \bar{s} = concentration of growth-limiting nutrient for bacteria; \bar{h} = concentration of bacteria; \bar{p} = concentration of protozoa; μ = specific growth rate of bacteria; D = dilution rate. (The tilde signifies a steady state value.) The equations for the system are: $\mu = \mu_m s / (s + K_s)$; $\lambda = \lambda_m h / (h + K_h)$; $dp/dt = (\lambda - D)p$; $dh/dt = (\mu - D)h - \lambda p/W$; $ds/dt = D(s_r - s) - \mu h/Y$. In the steady state $dp/dt = dh/dt = ds/dt = 0$.

rate of the protozoa is equal to the dilution rate. Above the critical dilution rate of the protozoa, only bacteria remain and nutrient utilization is practically complete until near the critical dilution rate for the bacteria (1.0 h^{-1}). It is surprising that when the protozoa are present, the nutrient concentration is high except at dilution rates approaching the critical dilution rate of the protozoa, indicating that predation decreases the number of bacteria in the system, and diminishes the rate of nutrient consumption. Thus it seems that in the presence of a protozoan predator the efficiency of waste (nutrient) removal is significantly less than is potentially possible.

This analysis leads to the conclusion that efficient waste removal combined with subsequent protozoa development may be achieved by two chemostats of differing volumes in series. In the first vessel, of relatively small volume (V_1), the flow rate (f) could be adjusted so that the dilution rate (f/V_1) was higher than the maximum specific growth rate of the protozoa population thus eliminating predatory behaviour and allowing nutrient utilization by the bacteria to achieve its full potential. In the second vessel, of larger volume (V_2), the dilution rate (f/V_2) would be less than the maximum specific growth rate of the protozoa and predation on the incoming bacteria would take place. Using the values of Fig. 1, nutrient utilization would be almost complete with a dilution rate of 0.9 h^{-1} . In the second stage with a dilution rate of 0.3 h^{-1} , the protozoa would consume almost all the bacteria. Hence with a total residence time of 4.4 h in the two-stage process, the nutrient to protozoa conversion would be about 99% efficient. Thus a two-stage system would have the advantages given by the presence of protozoa in an effluent purification system combined with a considerably more efficient mechanism for removal of the waste material.

S. J. PIRT
M. J. BAZIN

Department of Microbiology,
Queen Elizabeth College,
London W8 7AH

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- Abson, J. W., and Clarke, E. I., *Proc. Biochem.*, **6**, 15 (1971).
- Pirt, S. J., *Association of River Authorities Year Book*, 119 (1971).
- Canale, R. P., *Biotech. Bioeng.*, **12**, 353 (1970).
- Curds, C. R., *Water Research*, **5**, 793 (1971).