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EFFECT OF ORGANIC LOADING ON GROWTH
CHARACTERISTICS OF ALGAE AND BACTERIA
IN MIXED CULTURE

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1. INTRODUCTION AND METHODS

The growth characteristics of alga *Chlorella vulgaris* and heterotrophic bacteria and their effects on conversion of organic carbon in mixed culture fed batch reactors were studied. Glucose used as a source of organic carbon was fed daily in granular form to provide input concentration ranging from 25 to 700 mg/l. Nutrient composition containing KNO₃ 1000 mg/l, MgSO₄·7H₂O 250 mg/l, K₂HPO₄ 250 mg/l, NaCl 100 mg/l, CaCl₂·2H₂O 10 mg/l, FeSO₄·7H₂O 2 mg/l as well as trace minerals composed of Boron, Manganese, Zinc, Copper and Molybdenum were added at the beginning of culture. The culture was kept at 30°C, and mixing was provided with magnetic stirrers. Light intensity provided was 6000 lux at 12h/12h alternating photo periods of light and darkness. Algae used was *Chlorella vulgaris* and bacterial inoculum used to seed the chemostat reactors was collected from the final sedimentation tank of Sendai city activated sludge treatment plant.

2. RESULTS

In order to analyze the growth kinetic parameters, glucose was assumed to be the only biomass growth limiting substrate. The variation in biomass and culture volume in a fed batch reactor may be defined by the following mass balance equations assuming Monod kinetics is valid:

$$\frac{d(V \cdot X)}{dt} = \mu \cdot V \cdot X \quad \text{for biomass} \quad \text{and} \quad \frac{dV}{dt} = V_s - V_e - V_a \quad \text{for culture volume}$$

where μ = specific growth rate (d⁻¹), biomass concentration (mg/l), t = operation time (d), V = culture volume (l), V_s = volume of substrate added daily (l), V_e = volume of culture lost through evaporation (l) and V_a = volume of sample taken for analysis (l). Because glucose was added in solid form V_s is negligible. Sample volume V_a does not have effect on the biomass concentration and neglecting the effect of V_e , then:

$$\frac{dX}{dt} = \mu \cdot X = \frac{\mu_{\max} \cdot S}{K_s + S}$$

where μ_{\max} = maximum specific growth rate (d⁻¹), S = concentration of growth limiting substrate (mg/l) and K_s = saturation constant (mg glucose/l).

The growth kinetics of algae and bacteria is shown in figure 1. In two reactors R300 and R700 supplied with 300 and 700 mg glucose/l/d, dissolved oxygen was completely depleted from the first day of operation but growth of algae was observed. Although algae is believed to be strictly aerobic organisms, their growth in anaerobic conditions have been repeatedly reported by many researchers such as Stewart and Pearson (1970) and Baas-Becking and Wood (1955). Our data however, indicate that although algae grows under anaerobic conditions, growth rate is significantly reduced but no effect was observed for heterotrophic bacteria. While dissolved oxygen was observed to be a major contributing growth limiting factor for algae during exponential growth phase, beyond exponential growth phase pH was observed to limit growth of algae as well as bacteria particularly at values below 5.2. Heterotrophic bacteria were observed to grow at the rate of 0.54~3.07 d⁻¹, the growth rate increasing with increasing glucose loading rate, while growth rate for algae was between 0.20 and 0.91 d⁻¹. The maximum growth rate for algae and bacteria was respectively 1.58 and 1.70 d⁻¹, but observed saturation constant K_s for algae and bacteria was significantly different at 174 and 27 mg/l respectively. This seems to indicate that during the exponential growth phase either algal activity was more responsible for glucose uptake than bacterial activity or inorganic carbon produced from glucose degradation was insufficient for algal growth. The effectiveness of conversion of organic carbon into biomass was however decreasing with increasing organic loading rate (figure 2).

Algae has been repeatedly reported to prefer unionized dissolved CO₂ for photosynthesis (Tsuzuki *et al.*, 1980; Azov *et al.*, 1982). Most of free CO₂ obtained in algal-bacterial system is derived from

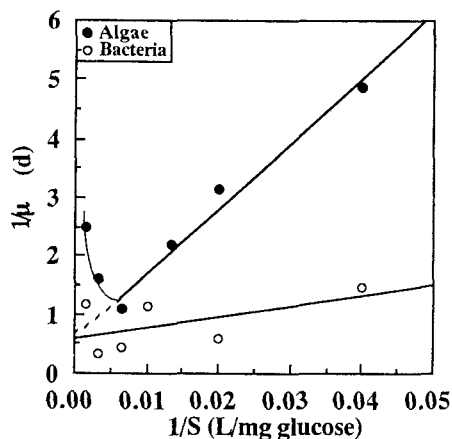


Fig. 1: Lineweaver-Burk plot of dependence of glucose uptake rate by algae and bacteria on glucose loading rate in mixed culture.

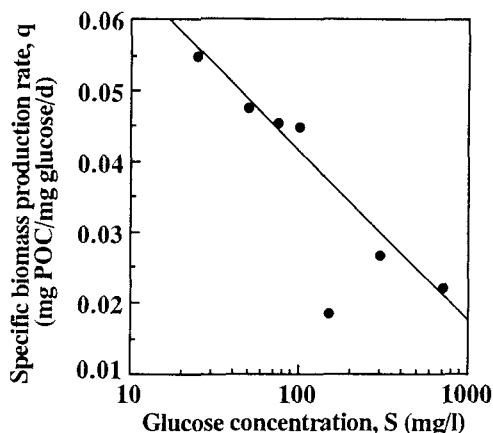


Fig. 2: Dependence of rate of incorporation of carbon into biomass on glucose loading rate.

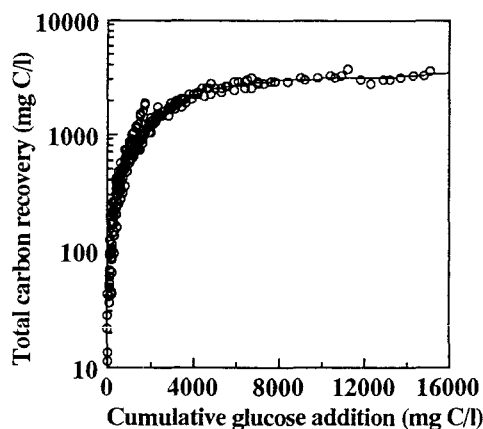


Fig. 3: Effect of glucose loading on gas emission.

bacterial degradation of organic matter. The availability of inorganic carbon although is theoretically proportional to available organic carbon, in practice this will be true if pH is within the favorable range. Our experiments showed that inorganic carbon production increased with increasing organic loading rate but at higher loading rates above 150 mg/l/d sudden decline was observed following volatile fatty acids accumulation resulting in the decline of pH to acidic range.

Figure 3 shows the fraction of carbon lost to the air most probably through emission of CO_2 produced by microbial degradation of organic matter. This indicates that excessive loading of carbon results mostly into formation of gases. The proportion of gas production was observed to be as high as 79% and was increasing with increasing glucose loading rate.

3. CONCLUSIONS

Glucose loading rate was observed to inhibit algal growth rate at loading rate above 150 mg/l/d. Both bacteria and algae were observed to be affected by pH values below 5.2. The efficiency of total biomass production was observed to decrease with increasing glucose loading rate.

4. REFERENCES

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