

## EFFECTS OF CATHODIC POLARISATION ON CARBOHYDRATE METABOLISM IN *AMPHORA COFFEAIFORMIS*, A MARINE FOULING DIATOM

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Attachment, growth and carbohydrate metabolism of *Amphora coffeaeformis* cells on cathodically polarised and non-polarised 304 stainless steel were evaluated when cells were grown under continuous illumination at 18°C for 8 d in medium prepared in synthetic seawater. Attachment to cathodically polarised steel was very poor as evident from lower values of cell dry weight, organic carbon chlorophyll *a* and protein. Levels of these decreased with time indicating the absence of algal growth on the stainless steel under these conditions. In contrast, on non-polarised steel, the attachment of *A. coffeaeformis* cells was higher by a factor of 10 to 100. The observed non-linear increase in algal cell dry weight, organic carbon, chl *a* and protein with time indicate active growth of the algal cells on non-polarised steel. Although carbohydrate and protein were the most abundant constituents on both types of steel, there were apparent differences in their relative distribution. Carbohydrate was the most abundant organic constituent in the cells from non-polarised steel, whereas protein was the major organic component in the algal cells from cathodically polarised steel. Such differences were also evident in protein:carbohydrate, protein:acid soluble carbohydrate, carbohydrate:chl *a* and protein:chl *a* ratios in the cells from cathodically polarised and non-polarised steel. Carbohydrate composition data indicate 10 to 100 times enrichment of acid soluble carbohydrates in the cells from non-polarised steel. The observed increase in acid soluble carbohydrate was due to increased synthesis of this fraction in cells from non-polarised steel. Under cathodic polarisation no such increase was observed. The consistent decrease in the acid soluble carbohydrate and increase in the protein:acid soluble carbohydrate ratio in cells from cathodically polarised steel suggest preferential loss of acid soluble carbohydrate. It is possible that this may play an important role in enhancing the effectiveness of cathodic protection in the control of fouling and/or corrosion of metals immersed in the marine environment.

**KEY WORDS:** *Amphora coffeaeformis*, carbohydrate metabolism, cathodic polarisation, stainless steel, microfouling.

### INTRODUCTION

The formation of a complex layer of attached organisms due to a series of discrete chemical and biological events is known as biofouling. In the marine environment surfaces adsorb organic matter thereby conditioning them (Loeb & Neihof, 1977; Characklis & Escher, 1988; Zutic & Tomaic, 1988). A number of compounds including glycoproteins (Baier, 1980), humic materials (Loeb & Neihof, 1975) and unspecified macromolecules (Zaidi *et al.*, 1984) have been implicated as conditioning agents. The presence of these compounds affect surface charge (Loeb & Neihof, 1977; Zutic & Tomaic, 1988), and surface wettability (Dexter *et al.*, 1975). These properties play an

important role in the subsequent attachment and growth of microfouling organisms including bacteria, fungi, protozoa and diatoms. Furthermore, the formation and succession of fouling organisms in a community are affected by a variety of environmental and biological factors and by the components of the surface substratum (Characklis & Escher, 1988; Fletcher, 1988).

Such colonization, that takes place each time a surface is cleaned or repaired, can have a considerable influence on the physical and chemical conditions at the surface (Terry & Edyvean, 1984; Edyvean, 1984) and can be detrimental to the activity of antifouling compounds (Daniel & Chamberlain, 1981).

The methods used to protect submerged steel structures from corrosion are cathodic protection, non-toxic paint coatings or a combination of both. Cathodic protection is effected by the introduction of an electrical current to counteract the normal electrochemical corrosion reaction. The steel is forced to become cathodic and thus prevented from corrosion. Two different systems are employed as sources of the protective current, *viz.* a system of sacrificial anodes, or, a DC electrical current which is driven through the structure. Both systems generate high pH at the metal surface due to oxygen reduction forming hydroxyl ions, and pH values up to 11.5 have been recorded (Smith & Matson, 1975). Since the solubility of most inorganic compounds, especially calcium and magnesium carbonates decreases with the increasing pH, calcareous deposits ("scale") will form on the protected steel. This scale is insoluble at normal seawater pH and by acting as a barrier to oxygen, is an aid to the prevention of metal corrosion.

Electrochemical polarisation may alter many of these above parameters, especially hydrogen ion concentration, and thus may influence the attachment and growth of microfouling organisms. Earlier studies have considered the effect of cathodic polarisation on the attachment of microorganisms, especially bacteria, to metal surfaces (Gordon *et al.*, 1981; Dhar & Bockris, 1981; Dhar *et al.*, 1982; Edyvean & Moss, 1986; Little *et al.*, 1988; Mollica *et al.*, 1989; Guezennec, 1991).

Whilst colonizing surfaces, microorganisms synthesise polysaccharides (Corpe, 1974; Costerton *et al.*, 1987; Ford & Mitchell, 1990; Neu & Marshall, 1991). Moreover, the involvement of polysaccharides in biofouling and biocorrosion processes has been documented (Geesey *et al.*, 1986; Jolley *et al.*, 1988). Despite their importance in biofouling and biocorrosion processes, nothing is known about the influence of cathodic polarisation on carbohydrate metabolism in fouling microorganisms, including diatoms.

The diatom *Amphora coffeaeformis* is common in the microfouling community on non-toxic and toxic surfaces in the marine environment and therefore was used as a model organism for the present studies.

In this paper, data on the attachment, growth and carbohydrate metabolism of *A. coffeaeformis* on cathodically polarised and non-polarised 304 stainless steel are presented and discussed. Although, cathodic polarisation is not generally used on stainless steel, the use of this metal provides a relatively inert surface for settlement studies in comparison with other metals which do not have the same surface passivity.

## MATERIALS AND METHODS

### *Test Panels*

Test panels (10 × 7.2 cm) of 304 stainless steel were washed with detergent solution, rinsed several times with tap water followed by double distilled water and then rinsed with analytical grade methanol and dried at 60°C.

### *Culture and Growth condition*

*Amphora coffeaeformis* var *perpusilla* (Grunow) Cleve was isolated and cultured as described by Bhosle *et al.* (1993).

### *Attachment, Growth and Carbohydrate Production on Non-polarised 304 Stainless Steel*

Experiments to assess attachment, growth and carbohydrate production by *A. coffeaeformis* on non-polarised steel were carried out in 5 dm<sup>3</sup> PVC tanks containing 1 dm<sup>3</sup> of f/2 medium (Guillard & Ryther, 1992) prepared with Instant Ocean artificial seawater (Aquarium Systems, USA). Each tank had 4 replicate panels. To each tank 100 cm<sup>3</sup> of 48 h old culture of *A. coffeaeformis* were added. After inoculation, the tanks were kept under continuous illumination at 18°C for 2, 4, 6 and 8 d.

### *Attachment, Growth and Carbohydrate Production on Cathodically Polarised 304 Stainless Steel*

The test cells consisted of 5 dm<sup>3</sup> PVC tanks equipped with a graphite counter electrode, Standard Calomel Electrode (SCE) (reference) and working electrodes in 1 dm<sup>3</sup> of f/2 medium prepared as above. The test cell was connected by a saturated potassium chloride agar salt bridge to another cell containing a graphite counter electrode. Each tank was inoculated as above by adding 100 cm<sup>3</sup> of a 48 h old culture of *A. coffeaeformis*. Metal panels were then polarised using a potentiostat set to maintain a predetermined potential of the working electrode (WE) to within  $\pm 0.2$  mV. The potential applied to the 304 stainless steel panels was fixed at  $-700$  mV/SCE over a period of 2, 4, 6 and 8 d.

### *Sampling for Analysis*

At each sampling time *viz.* 2, 4, 6 & 8 d, 4 cathodically polarised and 4 non-polarised stainless steel panels were removed from the PVC tanks and scraped individually using a nylon brush and Instant Ocean artificial seawater (Bhosle *et al.*, 1989a; Bhosle *et al.*, 1990 a,b). The volume of the scraped material was adjusted and a known volume (1 to 5 cm<sup>3</sup>) filtered through pre ignited (450°C, 12 h) and preweighed GF/C glass filters (25 mm, 1.0  $\mu$ m pore size). Replicate (2–6) samples were then analyzed for cell dry weight, carbon, chl *a*, proteins, carbohydrate and carbohydrate fractions.

### *Analytical Methods*

Cell dry weight was determined after drying the filters to constant weight in an oven at 50°C. The filters were cooled and weighed on a microbalance. Chl *a* was determined spectrophotometrically after extracting with dimethylsulphoxide (Shoaf & Holden, 1976; Liem, 1976). Cell carbon was analysed following the method of Parsons *et al.* (1984). Protein was analysed by the bicinchoninic acid method (Anon, 1987) after extracting with 0.1 N NaOH. Carbohydrate fractions, dilute acid (0.1 N H<sub>2</sub>SO<sub>4</sub>) soluble, dilute alkali (0.1 N NaOH) soluble and residual carbohydrate were extracted as described earlier (Bhosle & Dhople, 1988; Bhosle *et al.*, 1989b). Total carbohydrate and carbohydrate fractions were estimated by the phenol-sulphuric acid method (Dubois *et al.*, 1956). Carbohydrate carbon and protein carbon were estimated from carbohydrate and protein values by multiplying with 0.40 and 0.47 respectively.

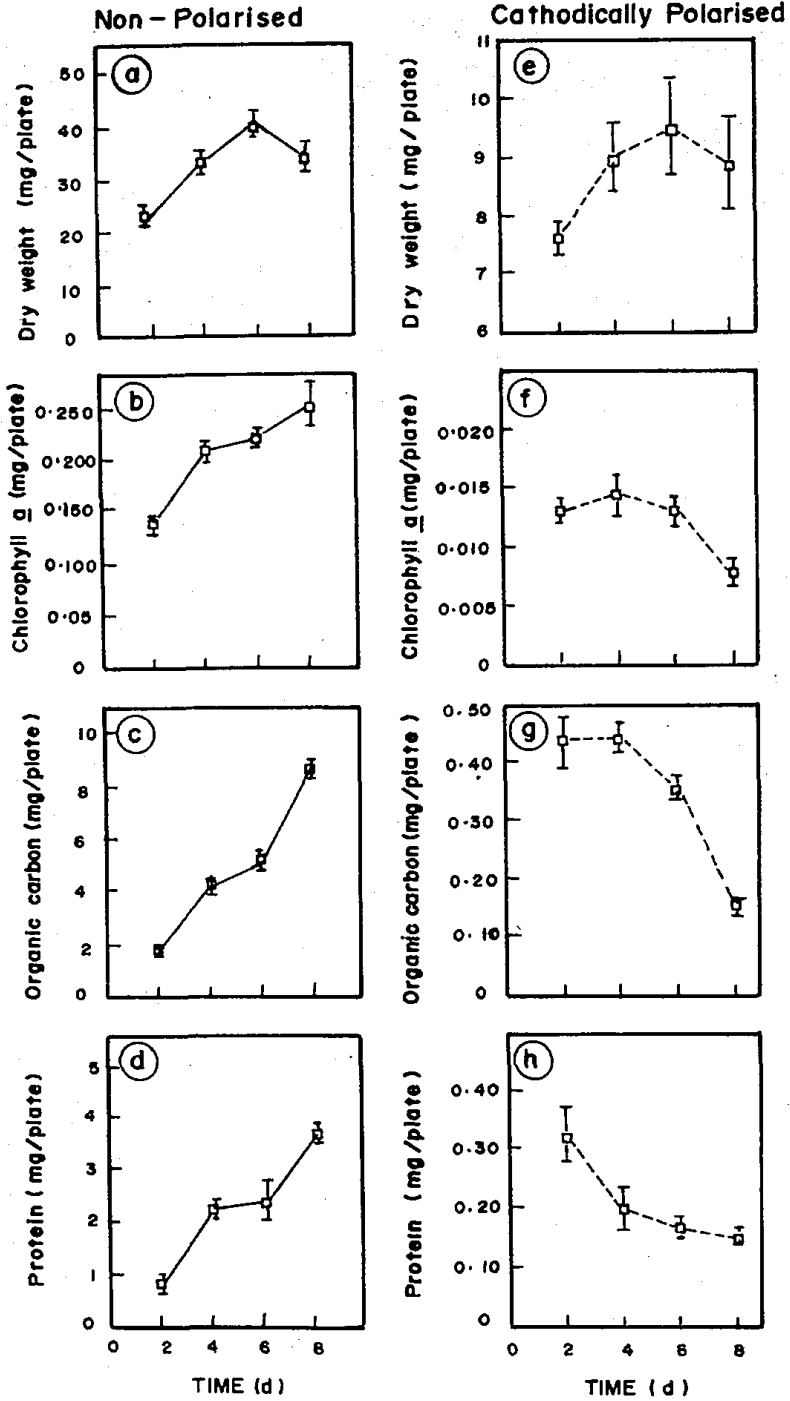


Fig. 1 Variation in *Amphora coffeaeformis* biomass on non-polarised (a to d) and cathodically polarised (e to h) 304 stainless steel as a function of incubation period. a,e=carbohydrate; b,f=chl *a*; c,g=organic carbon; d,h=protein. Bar =  $\pm 2$  SD

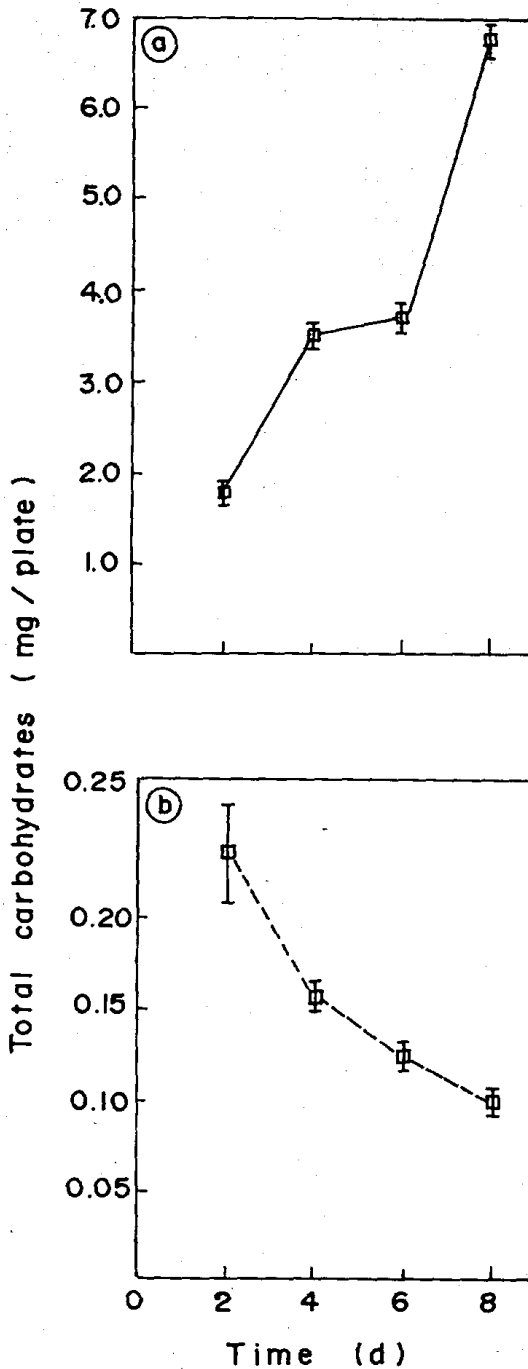


Fig. 2 Changes in the total carbohydrate of *Amphora coffeaeformis* cells attached to non-polarised (a) and cathodically polarised (b) 304 stainless steel

#### Scanning Electron Microscopy (SEM)

Unprotected and cathodically protected panels ( $0.3 \times 1$  mm) subjected to experimental conditions as described above, were retrieved at 2 d intervals over a period of 8 d.

**Table 1** Carbohydrate composition ( $\mu\text{g}/\text{plate}$ ) of *Amphora coffeaeformis* cells attached to non-polarised and cathodically polarised 304 stainless steel panels when grown in f/2 medium under continuous illumination at 18°C for 8 d

Time (d)	Non-polarised			Cathodically polarised		
	Acid	Alkali	Residual	Acid	Alkali	Residual
2	1426 $\pm$ 3.25 (0.22%)	286 $\pm$ 65.00 (0.72%)	65 $\pm$ 5.20 (8.00%)	167.2 $\pm$ 19.50 (11.62%)	44.20 $\pm$ 5.20 (11.76%)	22.88 $\pm$ 4.12 (18.00%)
4	2707 $\pm$ 42.25 (1.56%)	568 $\pm$ 35.75 (6.28%)	213 $\pm$ 5.00 (2.34%)	100.1 $\pm$ 6.50 (6.49%)	40.30 $\pm$ 14.3 (35.48%)	18.20 $\pm$ 0.52 (2.85%)
6	2859 $\pm$ 19.50 (0.82%)	676 $\pm$ 26.00 (3.84%)	111.80 $\pm$ 23.40 (20.93%)	77.50 $\pm$ 7.80 (10.00%)	36.25 $\pm$ 8.15 (22.48%)	11.25 $\pm$ 1.80 (16.00%)
8	5349 $\pm$ 6.50 (0.12%)	1131 $\pm$ 156 (13.79%)	356 $\pm$ 18.20 (5.11%)	62.58 $\pm$ 5.72 (9.14%)	32.86 $\pm$ 3.12 (9.59%)	10.62 $\pm$ 0.52 (4.89%)

Concentration  $\pm$  standard deviation; figures in parenthesis = percentage coefficient variation

**Table 2** Carbohydrate group composition expressed as a percentage of the total carbohydrates of *Amphora coffeaeformis* cells attached to non-polarised and cathodically polarised 304 stainless steel panels when grown in f/2 medium under continuous illumination at 18°C for 8 d

Time (d)	Non-polarised			Cathodically polarised		
	Acid %	Alkali %	Residual %	Acid %	Alkali %	Residual %
0	75.60	17.92	6.47	75.60	17.92	6.47
2	80.25	16.09	3.60	71.36	18.86	9.76
4	77.58	16.30	6.11	63.11	25.41	11.47
6	78.39	21.47	3.55	62.00	29.00	9.00
8	78.24	16.54	5.21	59.00	30.98	10.01

**Table 3** Carbohydrate and protein carbon as a percentage of the cell dry weight and total carbon of *Amphora coffeaeformis* cells attached to non-polarised and cathodically polarised 304 stainless steel panels when grown in f/2 medium under continuous illumination at 18°C for 8 d

Time (d)	Non-polarised				Cathodically polarised			
	Dry weight		Carbon		Dry weight		Carbon	
	a	b	a	b	a	b	a	b
0	2.56	1.76	36.63	25.00	2.57	1.76	36.63	25.00
2	3.08	1.68	45.23	24.72	1.22	1.99	21.62	35.24
4	4.19	3.19	39.61	30.14	0.70	1.05	14.30	21.28
6	3.60	2.70	33.88	25.46	0.52	0.84	13.95	22.31
8	7.85	4.98	37.10	23.53	0.47	0.82	27.71	48.60

a = carbohydrate carbon %; b = protein carbon %.

**Table 4** Carbon (POC): chlorophyll *a* (Chl *a*), total carbohydrate (PCHO): chlorophyll *a* (Chl *a*) and protein (PP): chlorophyll *a* (Chl *a*) ratios of *Amphora coffeaeformis* cells attached to non-polarised and cathodically polarised 304 steel panels when grown in f/2 medium under continuous illumination at 18°C for 8 d

Time (d)	Non-polarised			Cathodically polarised		
	POC/Chl <i>a</i>	PCHO/Chl <i>a</i>	PP/Chl <i>a</i>	POC/Chl <i>a</i>	PCHO/Chl <i>a</i>	PP/Chl <i>a</i>
2	11.29	12.77	5.94	33.34	18.03	25.01
4	16.96	16.79	10.87	31.16	11.14	14.11
6	19.49	16.51	10.56	27.51	9.60	13.08
8	28.51	26.44	14.28	19.61	13.60	20.29

**Table 5** Protein (PP): total carbohydrate (PCHO) and protein (PP): acid soluble carbohydrate (AC-CHO) ratios of *Amphora coffeaeformis* cells attached to non-polarised and cathodically polarised 304 stainless steel when grown in f/2 medium under continuous illumination at 18°C for 8 d

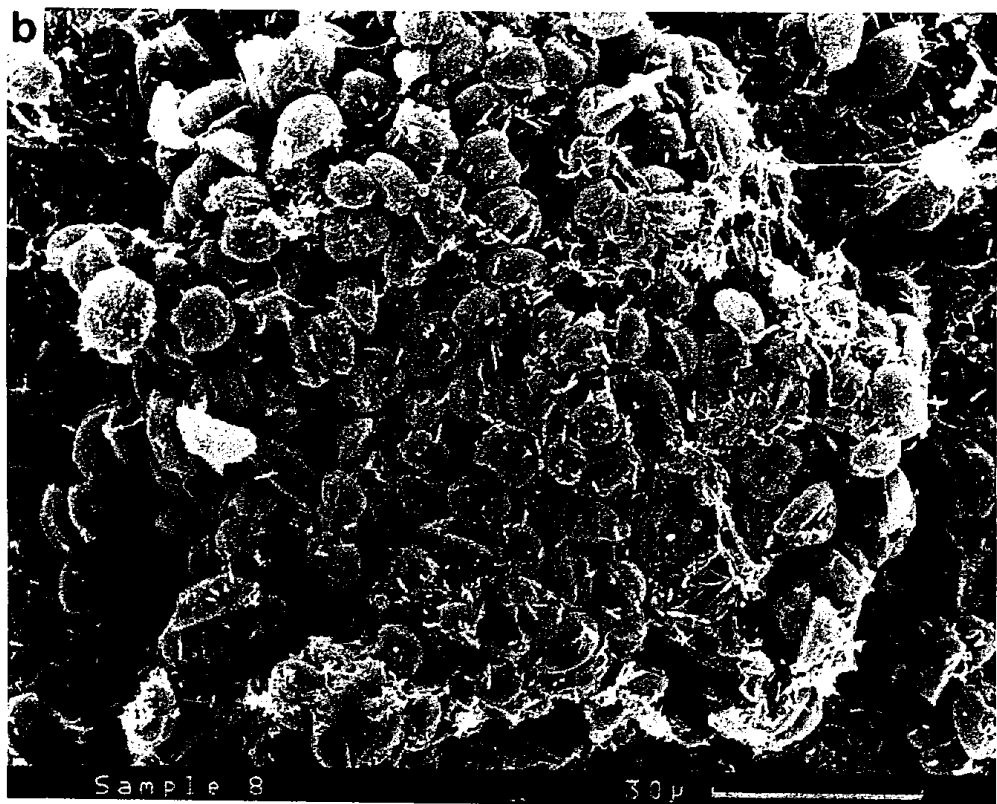
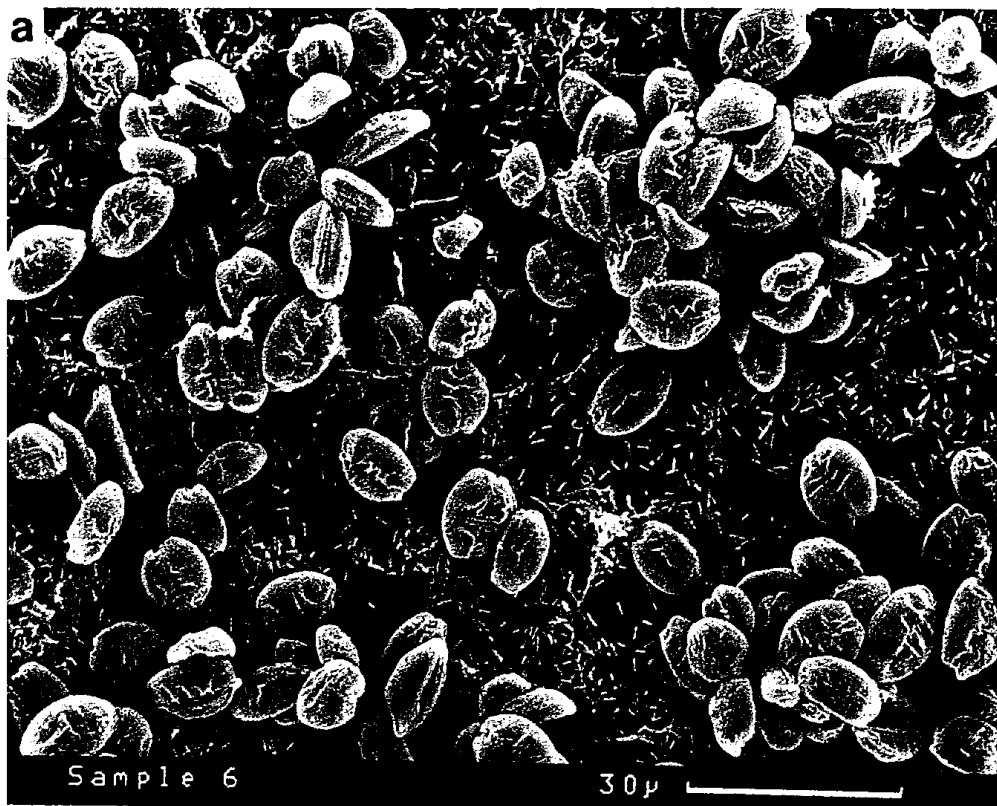
Time (d)	Non-polarised		Cathodically polarised	
	PP/PCHO	PP/AC-CHO	PP/PCHO	PP/AC-CHO
2	0.46	0.58	1.38	1.94
4	0.64	0.83	1.20	2.00
6	0.64	0.81	1.36	2.19
8	0.54	0.69	1.49	2.59

The panels were fixed in 1% osmium tetroxide in filtered (0.2 µm pore size) seawater for 1 h. They were then dehydrated through an acetone:water series beginning with 20% acetone and ending with 100% acetone. Samples were then critical point dried, sputter coated with gold using a Polaron Sputter Coater, and examined with a Camscan SEM (Cambridge, UK).

#### *Attachment, Growth and Carbohydrate Production on Non-polarised 304 Stainless Steel*

On non-polarised steel algal biomass increased non-linearly during the period of cultivation and the maximum was observed on the 8th day after inoculation. This was evident from the increase in the cell dry weight, carbon, chl *a* and algal protein (Fig. 1a to d). However, cell dry weight peaked on the 6th day and decreased thereafter (Fig. 1a). Cell carbohydrate (Fig. 2a) showed a pattern similar to that observed for cell carbon, chl *a* and protein. It was the most abundant organic constituent and was found to accumulate during growth of the alga.

When cell carbohydrate was fractionated into acid soluble, alkali soluble and residual carbohydrate all three fractions showed an increase with the period of cultivation (Table 1). The acid soluble carbohydrate was the most abundant constituent of the alga (Table 2), whereas, alkali soluble and residual carbohydrate





were minor constituents contributing about 20% to the total carbohydrate (Table 2). When expressed as a percentage of cell dry weight and carbon, carbohydrate varied from 3 to 7% and 33 to 45%, respectively (Table 3). Carbon:chl *a*, total cell carbohydrate:chl *a*, and protein:chl *a* ratios are shown in Table 4. These ratios were generally low and increased during the period of cultivation. Protein:carbohydrate and protein:acid soluble carbohydrate ratios were low and showed some decrease on the 8th day of the experimental period (Table 5).

#### *Attachment, Growth and Carbohydrate Production on Cathodically Polarised 304 Stainless Steel*

When steel was polarised using a potential of  $-700$  mV, attachment of *A. coffeaeformis* was very poor and no appreciable growth was observed. Moreover, under cathodic polarisation algal biomass showed a progressive decrease with cultivation period, seen in the decreases in cell dry weight, carbon, chl *a* and protein (Figs 1e to h), and the minimum was observed on the 8th day after inoculation.

Cell carbohydrate decreased in abundance (Fig. 2b) and group composition data suggest that all fractions, *viz.* acid soluble, alkali soluble and residual carbohydrate decreased in algal cells attached to cathodically polarised steel (Table 1). Amongst these, the decrease in acid soluble carbohydrate was very pronounced, and this preferential loss resulted in a greater abundance of structural carbohydrates, which together contributed 28 to 41% to the total carbohydrate of the alga (Table 2).

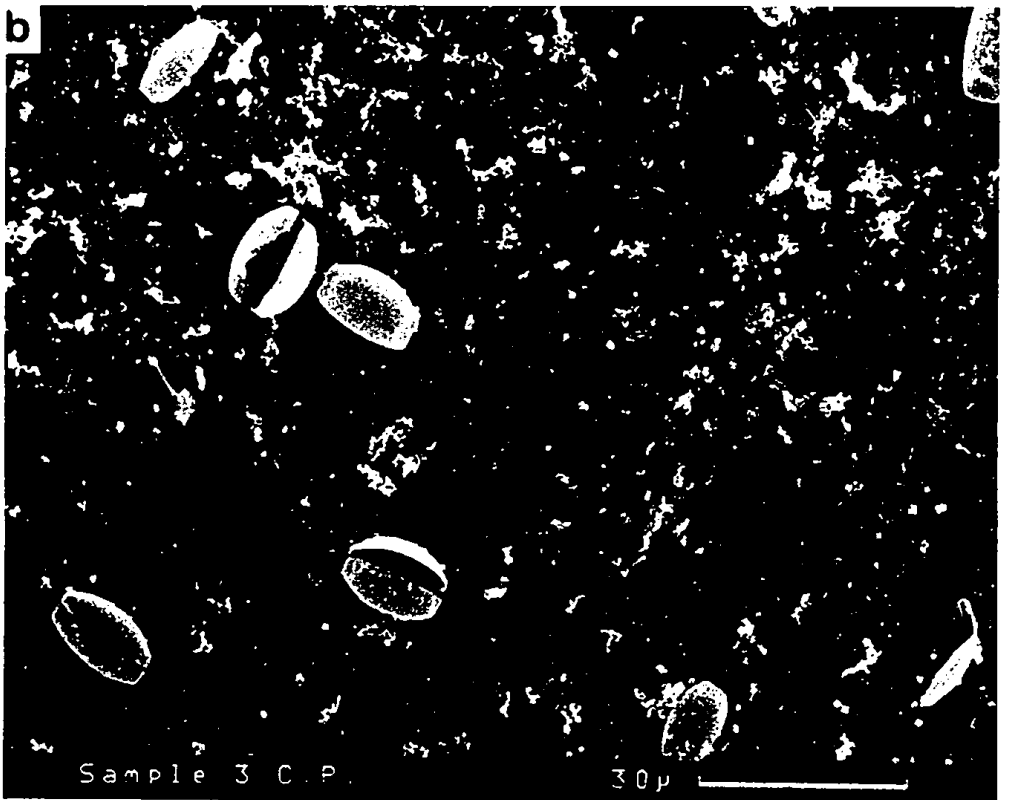
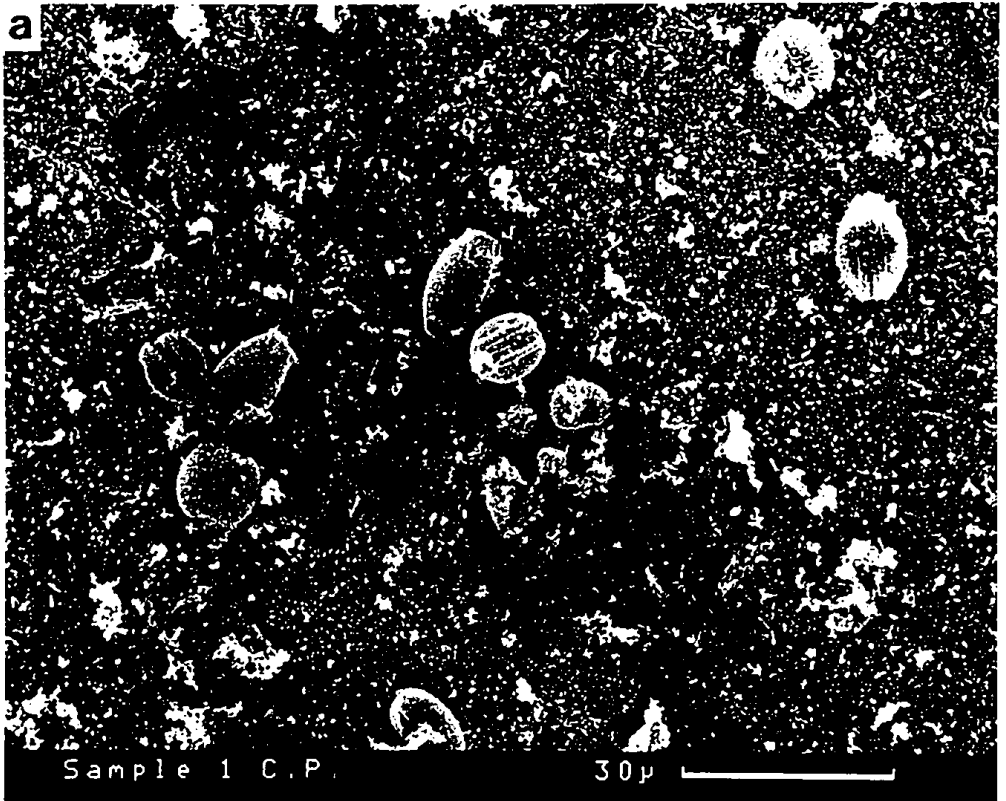
The carbohydrate contribution to cell dry weight and organic carbon ranged from 0.47 to 1.22 and 14 to 27%, respectively when expressed as a percentage of algal cell dry weight and carbon (Table 3). Protein was the most abundant organic constituent (Table 3), and carbon:Chl *a*, total carbohydrate:chl *a*, and protein:chl *a* ratios were relatively high and generally showed decreasing trends, with some exceptions (Table 4). More pronounced differences were observed for the protein:acid soluble carbohydrate ratio (Table 5), which was relatively high and generally increased with the cultivation period (Table 5).

#### SEM

In the case of non-polarised steel, attachment of large numbers of algal cells occurred and abundance increased with the period of cultivation (Fig. 3a, b). In contrast, on cathodically polarised steel, attachment of *A. coffeaeformis* cells was very poor. Furthermore, no appreciable increase in the abundance of the algal cells with the period of cultivation was observed (Figs 4a, b). Some bacterial contamination was present.

#### DISCUSSION

*A. coffeaeformis* biomass increased non-linearly on non-polarised steel when grown in f/2 medium under continuous illumination. The algal cells attained maximum biomass on the 8th day of the cultivation period. The increase in production of cell



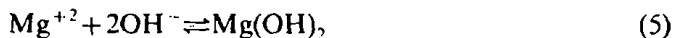
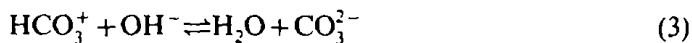
carbon, chl *a* and protein as a function of time (Fig. 1b to d) represents net synthesis as no attempt was made to account for losses due to respiration *etc.*

When the steel was cathodically polarised using a potential of  $-700$  mV, the biomass of attached cells was less (Fig. 1e to h). Cathodically protected systems generate a high pH at the metal surface due to oxygen reduction forming hydroxyl ions leading to deposition of calcareous deposits. Since the increase in the cell dry weight was not accompanied by an increase in organic constituents it can be concluded that the increase in dry weight was due to the formation of calcareous deposits on the cathodically protected steel. Evidence that *A. coffeaeformis* did not grow on the cathodically protected steel comes from the reduced levels of chl *a*, carbon, protein and carbohydrate (Figs 1f to h & 2). The effect of cathodic polarisation on the algal cells may be attributed to enhancement of the cathodic reactions that normally take place at the surface of a freely corroding metal, and to additional reactions that are induced by cathodic polarisation (Gordon *et al.*, 1981; Little *et al.*, 1988).

An impressed cathodic potential causes the following reactions:



These reactions cause an increase in the alkalinity that results in a shift in the equilibrium of all chemical reactions involving calcium, magnesium and bicarbonate ions.



In addition to inorganic deposits [Eqs (4), (5)], attachment to metal surfaces can be influenced by cathodic polarisation. For example, more bacteria attached to cathodically polarised copper than to unpolarised copper (Gordon *et al.*, 1981). In contrast, Dhar *et al.* (1982) reported that the number of adsorbed bacteria on tin oxide and titanium cathodes decrease with an increase in cathodic potential. They proposed that an increase in interfacial pH negated the contribution from long range Van der Waal's and double layer interactions between bacteria and the electrode surface. Furthermore, pulsed cathodic polarisation of steel retarded biofouling in seawater due to an increase in the interfacial pH as a result of cathodic consumption of oxygen [Eqn (1)] (Littauer & Jennings, 1968). As numerous processes occur at cathodically polarised surfaces, the exact mechanisms involved in controlling the attachment and growth of microorganisms to such surfaces are difficult to define. Nevertheless, it appears that pH plays a major role.

Microbial attachment and growth are influenced by surface charge and surface conditioning processes (Loeb & Neihof, 1977; Characklis & Escher, 1988; Zutic & Tomaic, 1988). Both these processes are also influenced by cathodic polarisation, thereby affecting attachment of microorganisms to the surfaces (Marshall 1973; Corpe, 1974; Fletcher & Loeb, 1979; Gordon *et al.*, 1981; Little *et al.*, 1988).

Fig. 4 Scanning electron micrographs showing attachment of *Amphora coffeaeformis* cells to cathodically polarised 304 stainless steel. a = 2d; b = 6d.

The charge imposed on a metal surface depends on the extent and direction of polarisation, and on the nature of the metal. The interfacial pH at a polarised surface and the electrochemical reaction products have a similar dependence. The surface charge of microbial cells, on the other hand, depends on interfacial pH. The pH determines the degree of protonation of ionogenic groups associated with the cell wall or capsular material and thereby the interaction with the surface. The resistance of microorganisms to electrochemical reaction, on the other hand, does not depend primarily upon the ionogenic groups, but is more likely to depend on the physiology of the organisms. It is likely that some of these factors might have influenced the decrease in the attachment and biomass of *A. coffeaeformis* cells on the cathodically protected steel.

Whilst colonizing surfaces, microorganisms produce exopolymeric substances (EPS), the major organic constituents of which are generally polysaccharides (Corpe, 1970; Costerton *et al.*, 1987). As these compounds play an important role in biofouling and biocorrosion processes (Geesey *et al.*, 1986; Jolley *et al.*, 1988; Ford & Mitchell, 1990), the influence of cathodic polarisation on carbohydrate metabolism in *A. coffeaeformis* was assessed.

Scanning electron micrographs from both the cathodically polarised and the non-polarised steel showed the presence of some bacterial rods as contaminants. Since this might have had some influence on the carbohydrate and protein determinations the data have been used only for a relative comparison of the biochemical composition of algal cells grown on cathodically polarised and non-polarised steel.

Carbohydrate was the most abundant organic constituent of algal cells attached to non-polarised steel (Table 3). Availability of nitrogen affects the biochemical composition of algae. For example, nitrogen starvation enhanced the production of cellular and extracellular carbohydrate (Myklestad & Haug, 1972; Myklestad, 1977; Hama, 1988; Bhosle *et al.*, 1993). Thus nutrient deficient cultures will enhance their carbohydrate and/or lipid at the expense of protein. *A. coffeaeformis* cells attached to non-polarised steel produced greatly increased amounts of carbohydrate and the protein:carbohydrate ratio (Table 5) suggests that nutrient deficient conditions may have prevailed.

The relative content of protein and carbohydrate in algae is a function of nutrient supply. The protein:carbohydrate ratio of algal cells attached to non-polarised steel was very low compared to that of cells from cathodically polarised steel (Table 5). The low protein:carbohydrate ratio in cells from non-polarised steel reflected maximum rates of net carbohydrate production with relatively low rates of protein synthesis. Conversely, in cells from cathodically polarised steel, lower rates of carbohydrate production led to higher protein:carbohydrate ratio (Table 5). This was further confirmed from the carbohydrate percentage data and the carbohydrate:chl *a* and protein:chl *a* ratios obtained for cells attached to cathodically polarised and non-polarised steel. This implies that the physiology of *A. coffeaeformis* cells attached to non-polarised and cathodically polarised steel was very different. Further evidence for this was provided by the carbohydrate composition data of cells obtained from the two types of steel.

Cells of *A. coffeaeformis* obtained from cathodically polarised and non-polarised steel showed large differences in carbohydrate group composition data (Tables 1 & 2), although acid soluble carbohydrate was the most abundant constituent of the total carbohydrates in cells from both types of steel. The relative contribution of acid soluble carbohydrate to total carbohydrate was 77 to 80% and 59 to 71% in cells from non-polarised and cathodically polarised steel respectively. Interestingly, the

concentration of acid soluble carbohydrate decreased, probably due to utilization by the alga (and/or bacteria) or to cell lysis during cathodic polarisation (Table 2). The protein:acid soluble carbohydrate ratio tended to increase with an increase in the experimental period. This implies that the acid extractable carbohydrate decayed at a much faster rate than protein in the cells on cathodically polarised steel.

The preferential loss of acid soluble carbohydrate in cells on cathodically polarised steel results in higher levels of structural carbohydrates such as alkali soluble and residual carbohydrates (28 to 40%) (Table 2). From the consistent increase in these carbohydrates in cells even under cathodic polarisation, it is clear that they are not favoured substrates for algal respiration and/or bacterial degradation. It can therefore be concluded that acid soluble carbohydrates were preferentially utilized for algal respiration and/or bacterial degradation during cathodic polarisation, leaving the structural carbohydrate in the algal cells.

Similar studies utilizing other diatoms and microorganisms as well as studies in the natural environment should improve understanding of the mode of action of cathodic protection of metal surfaces.

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