CELLS AND ELECTRODE, STICKING TOGETHER IN SEAWATER

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Abstract

Potential controlled electrode surface propertiesoffer a possibility for direct studiesof non-specific interactions between living cells, non-living particles during aggregation processes or biofilm formation in seawater. Our approach is based on measuring electrical signals of double-layer charge displacements caused by adhesion of single cells or particles. Unicellular marine alga, *dunaliella tertiolecta* of micrometer size and flexible cell enveloppe was used as a model particle. The dropping mercury electrode acted *in situ* adhesion sensor and the electrochemical technique of chronoamperometry allowed precise measurement of the spread cell-electrode interface area and the distance of the closest approach of a cell.

Keywords : algal cells, electrochemistry, fouling, interface.

Introduction

Microorganisms in nature generally live and grow in aggregated forms such as biofilms and flocks. Appearence of biofilms in natural and technological environments causes severe problems. We still do not understand completely how cells sense the surface no the underlying mechanism of adhesion (1). Employing mercury electrode as in situ sensor offers detection/identification of single algal cells at freshly exposed electrode/seawater interface. Mercury electrode with dynamic growth and hydrophobicity can mimic natural fluid interfaces while adhesion force can be fine tuned by the changing applied potential (2).

Experimental

Electrochemical measurements : Dropping mercury electrode was used because of its well-defined interfacial properties and renewable surface. The electrochemical experiments were performed in a standard Methrom vessel 3-electrode system. The mercury electrode was directly immersed in cell suspension in oxygen-free aqueous 0.1 M NaCl (pH at 8.2) solution or in seawater. *Cell culture : Dunaliella tertiolecta* Butcher cells are suitable for electrochemical detection because of their size, membrane properties, euryhaline nature, and their ability to form stable suspensions of single cells due to their pronounced motility and low stickness. The cells (maximum dimension 6-12 μm) were grown in seawater enriched with F-2 medium in batch culture. Cells were separated after 8 days of growth with mild centrifugation and washed several time with filtered seawater. Vialability of cells was controlled by microscopic observation of cell motility.

Results and discussion

Attractive interaction between a cell and electrode results in a double layer charge displacement as represented by a scheme in Figure 1. The transient flow of current reflects the dynamics of adhesive contact formation and subsequent spreading of a cell. The signals of individual cells from suspension differ only slightly in the peak current and duration, indicating attachments from a nearly monodisperse particle population. The rate of adhesion and spreading of cells is enhanced by the hydrodynamic regime of electrodeís growing fluid interface. Adhesion signals of cells appeared at characteristic potential range, while outside of this potential range the cells act as inert particles (even in dense suspension). Potential range for cell adhesion depends on surface cell properties and their aging. The spike-shaped signals have the peak current in μA range, duration of 5-10 ms and displaced charge in nC range. Surprising similarities to adhesion signals of droplets of liquid hydrocarbons suggest that collective properties of cell exterior govern the dynamics of adhesion and rate of spreading, with fluidity playing a major role. The electrochemical technique thus allows a precise measurement of the contact area between the cell and the substrate (Table 1).

TABLE 1. Contact areas (A_C) of algal cells of different species and sources (A, B) at the positively and the negatively charged electrode surface.

ALGAL CELLS	LENGTH /µm	CONTACT σ_{Hg} = +3.8 µC/cm²	INTERFACE/ x 10^{-4} cm² σ_{Hg} = -6.5 µC/cm²
D. tertiolecta (A)	6-12	2.88 ± 0.53	1.73 ± 0.23
D. tertiolecta (B)	6-9	1.25 ± 0.23	0.68 ± 0.15
I. galbana	3.7-7.5	1.02 ± 0.6	0.57 ± 0.3
C. maculata	12-20	7.83 ± 3.67	3.64 ± 0.81

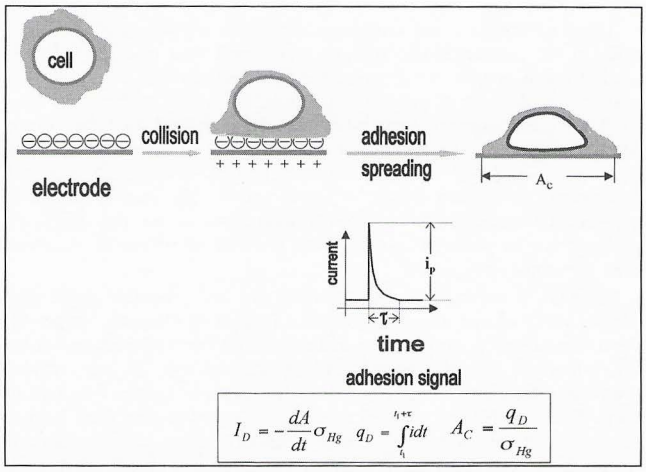

Figure 1. Attractive between cell and positively charged mercury electrode in seawater. The adhesion signal is caused by the double-layer charge displacement from the contact area A_C.

The variation in A_C values can be ascribed to the distribution of cell sizes in the culture. The contact interface area, A_C, exceeds cross-section area of a free cell by two orders of magnitude. Evidently, *D. tertiolecta* cell ruptured during the spreading process. It is known for vesicles that strong adhesion always leads to vesicle rupture (2). The distance of the closest approach of an adhered cell can be estimated with certainty as equal or smaller than the outer Helmholtz plane i.e. 0.3-0.5 nm.

Conclusion

Our results demonstrate a general significance of adhesion phenomena in single cell-substrate interactions in seawater. The characteristic potential range of adhesion can serve to study the interplay of complex surface forces involved in soft particle interactions in seawater and resulting ìstickiness coefficientî. This electrochemical approach also meets requirements for *in situ* single particle analysis (3).

References

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