



ELECTRONIC TRANSFERENCE ASSESSMENT OF THE REDOX PROCESSES AT CARBON ELECTRODES COATED WITH *GEOBACTER SULFURREDUCTENS* THAT GROWN AT DIFFERENT TEMPERATURES

L. PEIXOTO*, A. F. S. Santos, I. Machado, A. M. Sousa, A. G. Brito, P. Parpot, M. O. Pereira, R. Nogueira

* luciana.peixoto@deb.uminho.pt

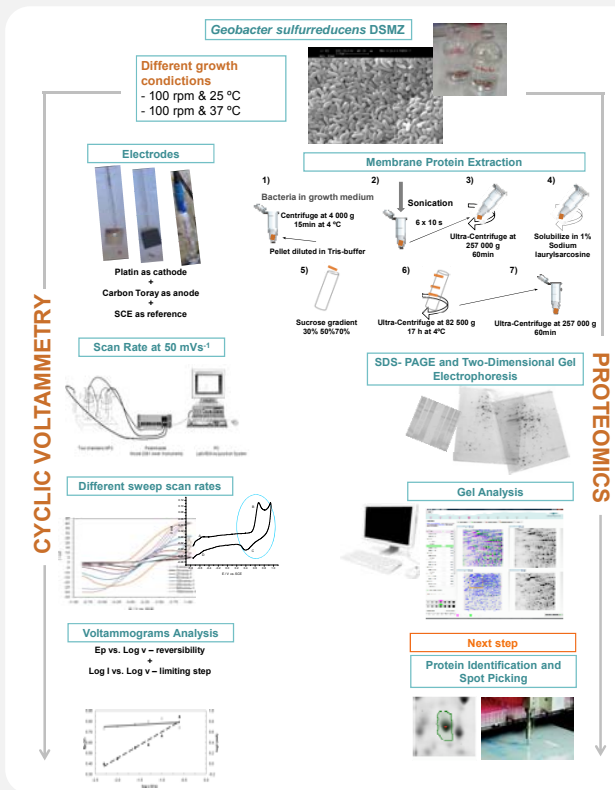


University of Minho
School of Engineering
Biological Engineering Center

INTRODUCTION

Geobacter sulfurreducens is a bacteria that can transfer electrons directly to the electrode from different external membrane cytochromes. Each cytochrome is associated with a range of redox potentials, being energetically more favourable than some others (Logan and Regan, 2006). Different bacterial growth conditions, such as temperature, may influence the prevalence of certain cytochromes in the external membrane (Peixoto et al. 2011). The aim of this work was to evaluate the effect of the growth temperature on the electrochemical behavior of *G. sulfurreducens*.

METHODOLOGY



RESULTS AND DISCUSSION

CYCLIC VOLTAMMETRY

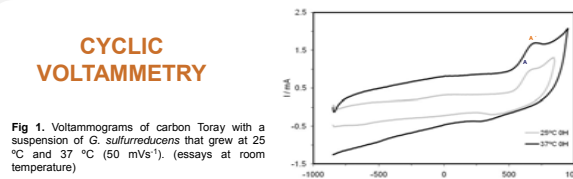


Fig 1. Voltammograms of carbon Toray with a suspension of *G. sulfurreducens* that grew at 25 °C and 37 °C (50 mVs⁻¹). (essays at room temperature)



Fig 2. Voltammograms of carbon Toray with a suspension of *G. sulfurreducens* that grew at 25 °C and 37 °C (25 mVs⁻¹), after 24 hours stabilize (a). (essays at room temperature). Comparison with the result obtained in time 0 h (b).

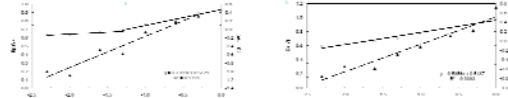


Fig 3. log I vs. log v (▲) and E versus log v (○) curves for the oxidation (Fig. 1) of a pure culture of *G. sulfurreducens* in suspension that grew at 25 °C (a) and 37 °C (b).

Table 1 Electrochemical data obtained for bacteria grown at two different temperature. (room temperature essays).

Electron Transfer Mechanisms		
25 °C 0h	- Reversible charge transfer between 5 mVs and 50 mVs and irreversible to higher sweeps scan rates. - Mixed control	25 °C 24 h at room T
37 °C 0h	- Irreversible charge transfer - Limited by diffusion	37 °C 24 h at room T
		37 °C 48 h transfer Last 24 h at 37 °C

- For Bacteria that growth at different temperatures, the oxidation peaks potentials and current intensities were different;
- Higher current intensities were found for bacteria grown at higher temperatures;
- the potential of the oxidation peak obtained with bacteria grown at higher temperature was more anodic, thus more energy was required;
- At lower sweep rates, it was possible to observe two oxidation processes, that are better defined in bacteria grown at 25 °C;
- Comparing the oxidation potential with literature (Richter et al. 2009), it was possible to conclude that different types of cytochromes can be established as responsible for heterogeneous electronic transfer.

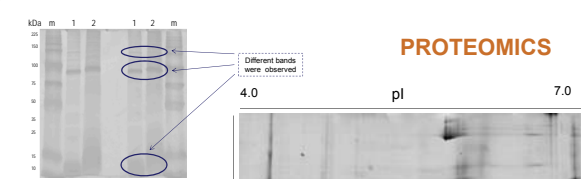


Fig 4. 12% SDS-PAGE of *G. sulfurreducens* proteins (1.6 µg), that grown at 25 °C (1) and at 37 °C (2); Molecular weight markers (m).



Fig 5. 2-DE of outer membrane proteins of *G. sulfurreducens* (protein load: 200 µg). Proteins that revealed differential expression from bacteria that grow at 25 °C and 37 °C are indicated by their index number given in the Table. Proteins were visualized by silver staining.

PROTEOMICS

Table 2 Normalised intensity of spots in the proteome profile of *G. sulfurreducens* grown at 25 °C and 37 °C. Protein spots were considered to display significant quantitative differences if they fulfilled the following criteria: p values ≤ 0.05 (t-test); detection threshold, average volume ≥ 20 (n = 3); differential tolerance, fold change ≥ 2

Spot #	Average normalized volumes mean (SD)	
	25 °C	37 °C
93	1293,967	4751,629
101	5612,445	1,983e+004
111	7505,605	2,135e+004
195	485,974	1623,547
203	233,095	633,443
207	759,898	1962,325
241	636,831	1622,581
247	2487,584	3847,489
266	455,310	3266,062
286	1281,760	299,479

- About 100 spots discriminated on the different conditions;
- Gel analysis of Outer Membrane Proteome at different growth conditions revealed:
- Up-regulation of 9 spots at 37 °C;
- Down-regulation of just one spot at 37 °C;

The spot picking and protein identification are in progress.

CONCLUSIONS

The difference observed in voltammetric study can be related to the structural differences in bacteria that grown at different temperatures. Changes in the *Geobacter sulfurreducens* growth temperature promotes different protein expression that can be responsible for different redox centers.

REFERENCES
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Qian, X. (2009). "Investigation of Fe(II) reduction in *Geobacter sulfurreducens* characterization of outer surface associated electron transfer components." PhD Dissertations.
Richter, H., Nevin, K.P., Daniel, H.J., Lowy, A., Lovley, D.R. and Tender, L.M. 2009. "Cyclic voltammetry of biofilms of wild type and mutant *Geobacter sulfurreducens* on fuel cell anodes indicates possible roles of OmcB, OmcZ, type IV pili, and protons in extracellular electron transfer." *Energy Environ Sci* 2, 506-516.
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