

Compararison of efficiency of the carbon, titanium and TiRuO₂ electrodes in water disinfection

Rodolfo Tolentino-Bisneto¹
Ederio D. Bidoia¹

Received on: March 10, 2002
Accepted on: June 5, 2002

TOLENTINO-BISNETO, Rodolfo; BIDOIA, Ederio D. Compararison of efficiency of the carbon, titanium and TiRuO₂ electrodes in water disinfection. *Salusvita*, Bauru, v. 21, n. 2, p. 75-82, 2002.

ABSTRACT

The disinfection of drinking water is very important for public health. Conventional process of disinfection of drinking water requires addition of substances in the water, which can generate toxic compounds, such as the trihalomethanes (THM). The trihalomethanes are formed by reaction of the chlorine with the organic substances present in water. The electrolytic treatment can replace the addition of chlorine during water disinfection. This treatment does not require addition of substances. Also it is easily automatized and requires a small space. This treatment has low operational cost. The aim of this study is to compare the electrolytic treatment using carbon titanium and TiRuO₂ electrodes in the *Bacillus subtilis*'s survival ratio. *B. subtilis*'s suspensions, in phosphate buffer pH 7.2, were electrolyzed by a carbon, titanium or TiRuO₂ electrodes as cathode. A platinum foil covered by a dialysis membrane was used as anode. After the application of 0.60A DC, at different times of electrolysis, in the *B. subtilis*'s suspension were determinate: the survival ratio, the pH and the temperature. The results allow to conclude that the carbon electrode reduces the survival ratio of the *B. subtilis* about 99.999% after 30 min of electrolytic treatment and the titanium and TiRuO₂ electrodes did not show significant reduction in the survival ratio after 45 min of electrolysis.

KEY WORDS: Disinfection, *Bacillus subtilis*, electrolysis, carbon, titanium, TiRuO₂

¹ Department of
Biochemistry and
Microbiology- Institute
of Biosciences of Rio
Claro - UNESP

Address:
Av. 24-A, 1515, Bela
vista, Rio Claro-SP.
CEP 13506-900
Telefone: (19) 526-4135
Fax: (19) 526-4137
e-mail:
ederio@rc.unesp.br

INTRODUCTION

Pathogenic microorganisms are a constant in most water sources and its elimination in water supply systems is important from the public health point of view (TOMINAGA; MIDIO, 1999). According to the World Health Organization 80% of the all diseases in third world countries are caused by contaminated water (TOMINAGA; MIDIO, 1999).

The standard process of water disinfection includes the addition of chemical substances that can produce toxic compounds affecting health of the consumers. Chloration is the most common method employed nowadays for disinfection, which includes the addition of free chlorine (Cl_2) or substances that release chlorine in the water in order to promote disinfection. Although cheap and efficient, this method produces trialomethane when chlorine is exposed to organic compounds in the water (TOMINAGA; MIDIO, 1999). Trialomethanes are carcinogenic besides other toxicological effects (TOMINAGA; MIDIO, 1999).

Traditionally, the electrolytic treatment has been used in treating wastewater. Reasons for that are the versatility, efficiency, easy automatization, environmental compatibility and low operational cost of this process (ANGELIS et. al., 1998). Furthermore, this method can be associated to biological treatment decreasing the period of effluents retention or modifying persistent substances of difficult biodegradation in material easier to be degradable or more biocompatible (ANGELIS et. al., 1998).

The electrolytic treatment may be an alternative to the traditional process of drinking water disinfection. Besides the advantages to public health the electrolytic treatment is a clean procedure since it does not require addition of substances (LUBICKI; JAYARAM, 1996), which can be used in industries of high technology that need real pure water to their procedures.

From the environmental point of view the electrolytic treatment has many advantages on chloration (LUBICKI; JAYARAM, 1996) among them, a reduced formation of trialomethanes favoring both the environment and health of people using such water.

The aim of this study was to compare the effect of carbon, titanium and TiRuO_2 electrodes on the viability of *Bacillus subtilis*.

MATERIAL AND METHODS

Preparing the inoculum

Cultures of *B. subtilis* CCT 2576 were obtained from two consecutives transpositions. The first in a nutrient agar solid medium NA (nutrient agar) incubated at 28°C for 24h. The second in a liquid culture medium NC (nutrient broth) incubated at room temperature under stirring for 24h.

TOLENTINO-BISNETO, Rodolfo; BIDOIA, Ederio D. Compararison of efficiency of the carbon, titanium and TiRuO_2 electrodes in water disinfection. *Salusvita*, Bauru, v. 21, n. 2, p. 75-82, 2002.

TOLENTINO-
BISNETO,
Rodolfo;
BIDOIA, Ederio
D. Comparison
of efficiency of the
carbon, titanium
and TiRuO_2 elec-
trodes in water
disinfection.
Salusvita, Bauru,
v. 21, n. 2,
p. 75-82, 2002.

Electrolytic treatment

The suspensions of *B. subtilis* were obtained after the addition of an aliquot of 2mL of inoculum to 0.2M phosphate buffer with pH 7.2, sterile and chlorete free. These suspensions were electrolyzed with spongi-
nous carbon electrodes manufactured by Tokai Carbon Co.[®] from Japan, titanium electrodes or titanium electrodes covered with titanium oxide and rutene oxide (TiRuO_2). As cathode it was used a polycrystalline platinum electrode manufactured by Aldrich (99.98% m/m) covered with a dialysis membrane. In addition, the membrane was filled with 2ml of 0.2M phosphate buffer with pH 7.2, sterile and chlorete free. By covering the cathode it is possible to prevent its effect during disinfection.

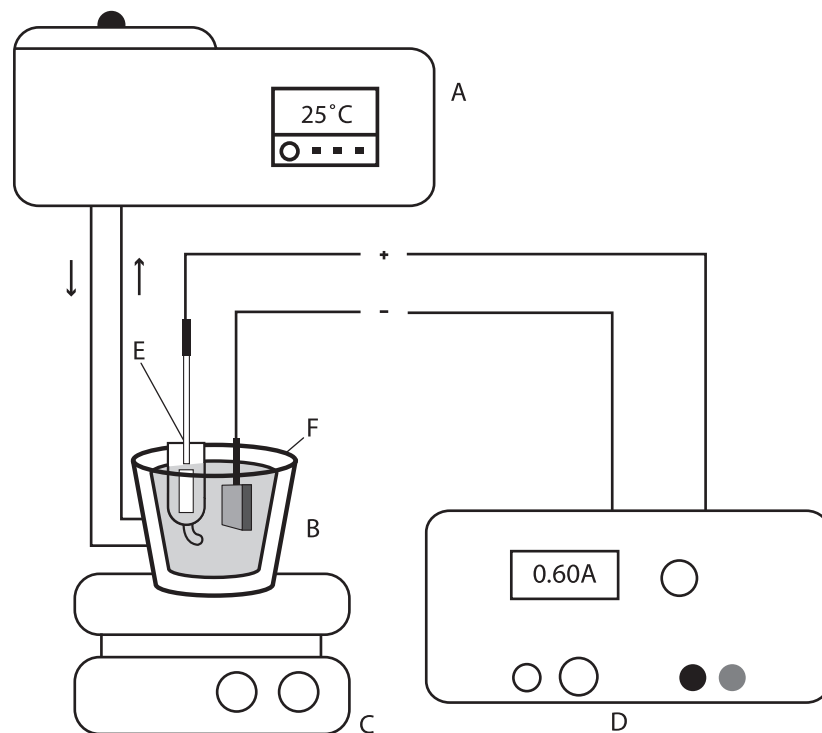


FIGURE 1: Diagram of the treatment system. A: thermostated bath; B: electrolytic cell; C: magnetic stirrer; D: DC power source; E: platinum anode covered with dialysis membrane; F: carbon, titanium or TiRuO_2 cathode.

The suspensions were electrolyzed at a continuous current of 0.60A in a batchwise type system in different periods. The electrolytic cell was maintained under constant refrigeration in order to keep the temperature of the system under 27°C. During the electrolysis the suspensions was continuously stirred. It was determined the pH, temperature and amount of viable cells of the electrolyzed suspensions.

RESULTS AND DISCUSSION

The carbon electrode was more efficient in reducing the viability of *B. subtilis* (FIGURE 2) than the titanium (FIGURE 3) and TiRuO₂ electrodes (FIGURE 4) when used as cathodes. Whereas the treatment using carbon electrode attained a reduction of bacillary viability near to zero in 30 minutes, the treatment with titanium and TiRuO₂ electrodes did not show a significant reduction until 45 minutes after starting the procedure.

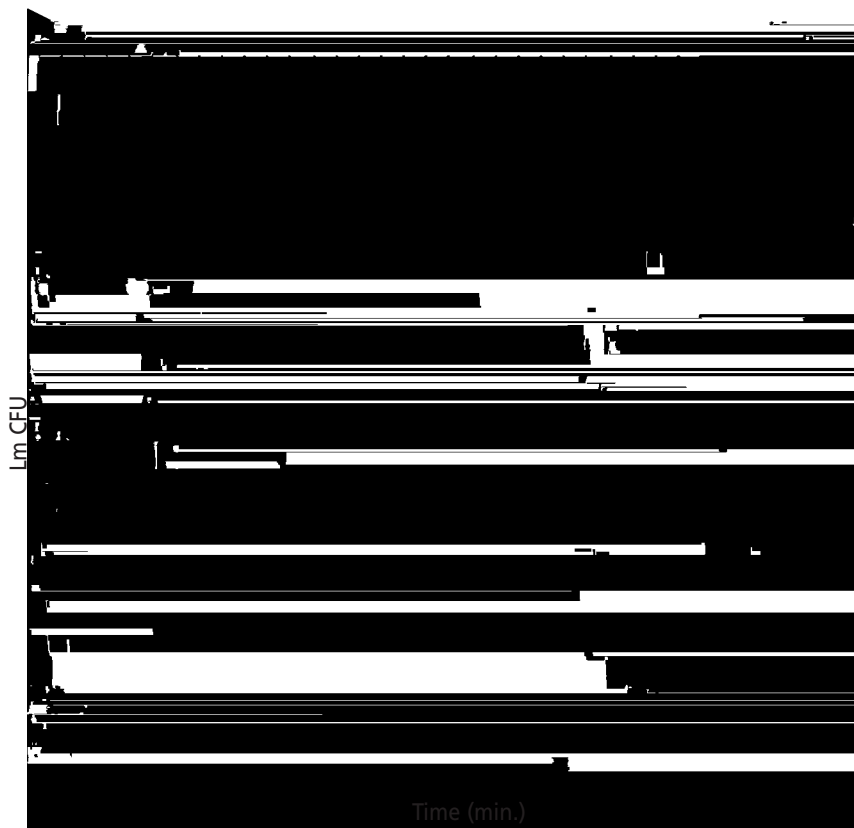


FIGURE 2: *Bacillus subtilis* colony forming units (CFU) (●) and pH (□) related to the period of electrolysis. Glassy carbon electrode as cathode and platinum covered with a dialysis membrane acting as anode. Current 0.60A. Tension 12.5 to 10.1V. Phosphate buffer pH 7.2.

TOLENTINO-
BISNETO,
Rodolfo;
BIDOIA, Ederio
D. Compararison
of efficiency of the
carbon, titanium
and TiRuO₂ elec-
trodes in water
disinfection.
Salusvita, Bauru,
v. 21, n. 2,
p. 75-82, 2002.

TOLENTINO-
BISNETO,
Rodolfo;
BIDOIA, Ederio
D. CompararIson
of efficiency of the
carbon, titanium
and TiRuO₂ elec-
trodes in water
disinfection.
Salusvita, Bauru,
v. 21, n. 2,
p. 75-82, 2002.

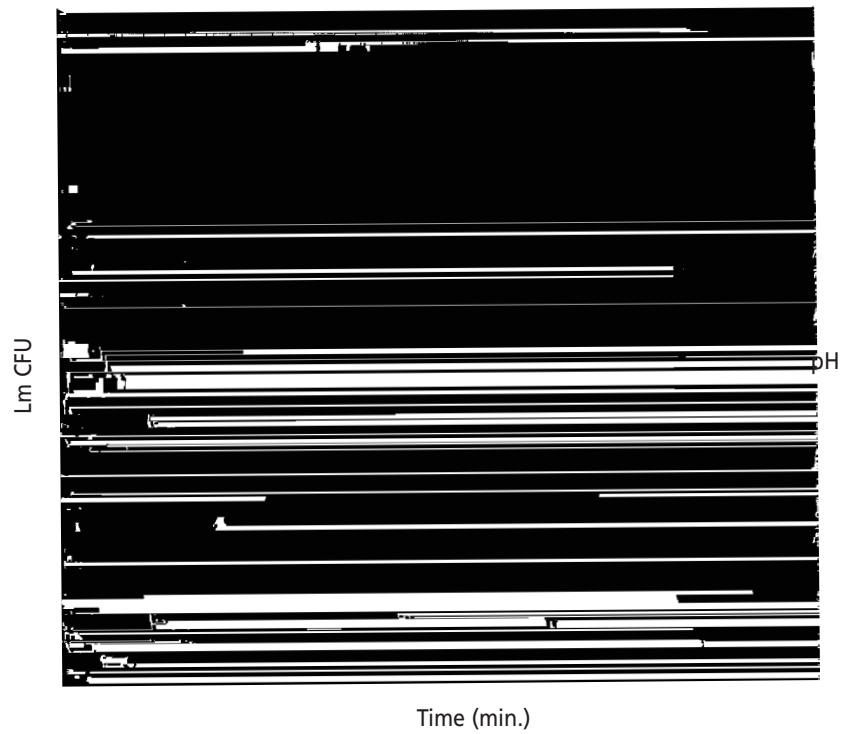


FIGURE 3: *Bacillus subtilis* colony forming units (CFU) (●) and pH (□) related to the period of electrolysis. Titanium electrode as cathode and platinum covered with a dialysis membrane acting as anode. Current 0.60A. Tension 13.0 to 8.7V. Phosphate buffer pH 7.2.

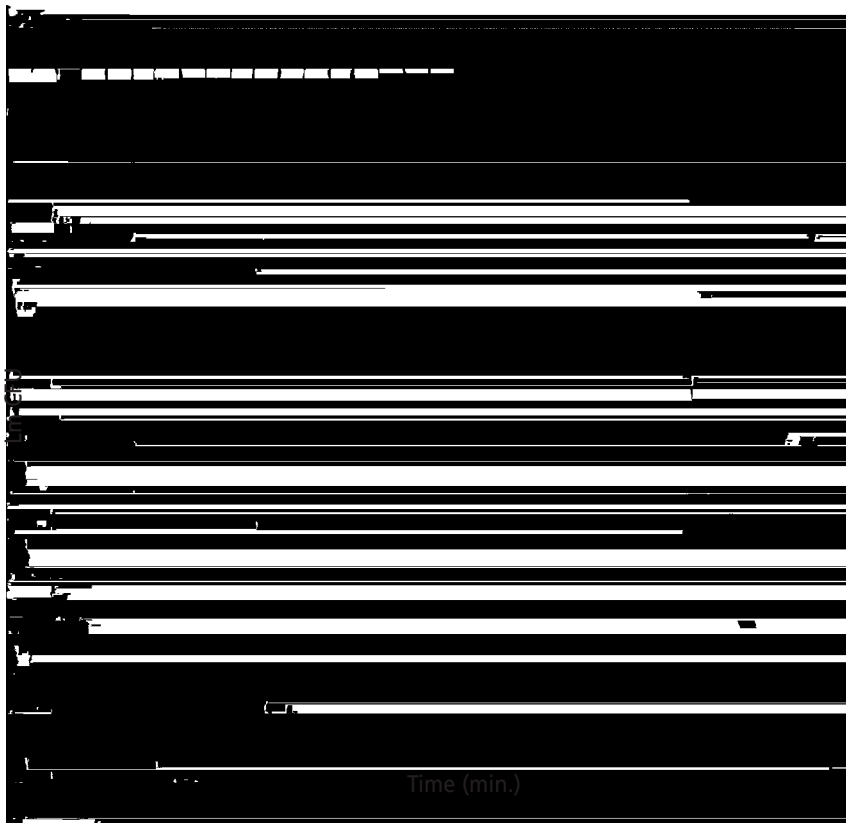


FIGURE 4: *Bacillus subtilis* colony forming units (CFU) (●) and pH (□) related to the period of electrolysis. TiRuO₂ electrode as cathode and platinum covered with a dialysis membrane acting as anode. Current 0.60A. Tension 12.5 to 10.1V. Phosphate buffer with pH 7.2.

The death of *B. subtilis* was due to charge transfer between the electrode and the microorganism (PATERMARAKIS; FOUTOUKINDIS, 1990; NAKASONO et al., 1992; 1993; BRATFICH et al., 1999; TOLENTINO-BISNETO; BIDOIA, 2000 b) associated to electroporation (LUBICKI; JAYARAM, 1996; FRIENDRICH et al., 1998; LEE; TAI, 1999; XUE et al., 2000).

High potentials applied to the cell promote destruction of the cytoplasmic membrane or, at least, an increase in membrane permeability and decrease in selectivity (LUBICKI; JAYARAM, 1996; FRIENDRICH et al., 1998; LEE; TAI, 1999). This phenomenon is called electroporation, which can be irreversible or reversible. In the former there is rupture in the cell membrane leading to cell death and in the latter the membrane rupture is minimal, cell death is not mandatory and the permeability of the membrane is highly increased (LUBICKI; JAYARAM, 1996; FRIENDRICH et al., 1998; LEE; TAI, 1999).

The efficiency of the carbon electrode in reducing the viability of *B. subtilis* may be due to the adsorption of the bacilli to the electrode, what

TOLENTINO-BISNETO, Rodolfo; BIDOIA, Ederio D. Compararison of efficiency of the carbon, titanium and TiRuO₂ electrodes in water disinfection. *Salusvita*, Bauru, v. 21, n. 2, p. 75-82, 2002.

TOLENTINO-BISNETO, Rodolfo; BIDOIA, Ederio D. Compararison of efficiency of the carbon, titanium and TiRuO₂ electrodes in water disinfection. *Salusvita*, Bauru, v. 21, n. 2, p. 75-82, 2002.

does not occur in the titanium and TiRuO₂ electrodes. Since the microorganism is adhered to the electrode the charge transfer between the electrode and bacteria is more effective and at the same time the bacilli is exposed to the electric field for a longer period.

Cellular death was not caused by variation in the solution pH since the solution was buffered and kept within the range supported by the bacteria (TOLENTINO-BISNETO; BIDOIA, 2000 a). The disinfection by chloride formation or by-products (PARELEUX; SICARD, 1970; STONER et al., 1982; PATERMARAKIS; FOUTOUKINDIS, 1990) was not considered since the system was chlorete free. Furthermore, death did not occur due to oxidation of cellular substances, such as the coenzyme A (MATSUNAGA et al., 1992; NAKASONO et al., 1992; OKOCHI et al., 1999) because a dialysis membrane isolated the anode and the *B. subtilis* had no contact with this electrode.

CONCLUSION

Results lead to the conclusion that the carbon electrode reduces the viability of the bacilli in more than 99.999% in 30 minutes electrolysis. Titanium and TiRuO₂ electrodes did not produce a significant reduction before 45 minutes of treatment.

It is possible to conclude that the *B. subtilis* become unviable due to the transfer of electrons between the cell wall of the microorganism and the electrode associated to the phenomenon of electroporation. The cell unviabilization was enhanced in the carbon electrode due to the adsorption of the bacilli to the surface of the electrode.

ACKNOWLEDGMENTS

CAPES, FUNDUNESP, CNPq e FAPESP.

BIBLIOGRAPHIC REFERENCES

1. ANGELIS, D. F. et. al. Eletrólise de resíduos poluidores: I - Efluentes de uma indústria liofilizadora de condimentos. *Química Nova*, v. 21, n. 1, p. 20-24, 1998.
2. BRATFICH, O. J. et. al. Electrolysis of cell suspensions of *Bacillus subtilis* (ATCC-9372) and of *Saccharomyces cerevisiae* (Fleischmann Royal) using cast-iron electrodes. *Brazilian Journal of Ecology*, v. 3, p. 83-84, 1999.
3. FRIEDRICH, U. et. al. High efficiency electrotransfection with aluminum electrodes using microsecond controlled pulses. *Bioelectrochemistry and Bioenergetics*, v. 47, p. 103-111, 1998.
4. LEE, S. W.; TAI, Y. C. A micro cell lysis device. *Sens. and Actuat.*, v. 73, p. 74-79, 1999.

5. LUBICK, P.; JAYARAM, S. High voltage pulse application for the destruction of Gram-negative bacterium *Yersunia enterocolitica*. *Bioelectrochemistry and Bioenergetics*, v. 43, p. 135-141, 1997.
6. MATSUNAGA, T. et. al. Disinfection of drinking water by using a novel electrochemical reactor employing carbon-cloth electrodes. *Applied Environmental Microbiology*, v. 58, p. 686-689, 1992.
7. NAKASONO, S. et. al. Electrochemical disinfection of marine bacteria attached on plastic electrode. *Bioelectrochemistry and Bioenergetics*, v. 27, p. 191-198, 1992.
8. NAKASONO, S. et. al. Electrochemical prevention of marine biofouling with a carbon-chloroprene sheet. *Applied Environmental Microbiology*, v. 59-11, p. 3757-3752, 1993.
9. OKOCHI, M.; NAKAMURA, N.; MATSUNAGA, T. Electrochemical killing of microorganisms using the oxidized form of ferrocenemonocarboxylic acid. *Electrochimica Acta*, VII. 44, p. 3795-3799, 1999.
10. PATERMARAKIS, G.; FOUNTOUKIDIS, E. Disinfection of water by electrochemical treatment. *Water Research*, VII. 24, n. 12, p. 1491-1496, 1990.
11. PARELIEUX, A.; SICARD, N. Lethal effects of Electric Current on *Escherichia coli* *Applied Environmental Microbiology*, v. 19, p. 421-421, 1970.
12. STONER, G. E. et. al. The mechanism of low frequency AC electrochemical disinfection. *Bioelectrochemistry and Bioenergetics*, VII. 9, p. 229-243, 1982.
13. TOLENTINO-BISNETO, R.; BIDOIA, E. D. Efeito do pH na desinfecção de águas de abastecimento por eletrólise utilizando eletrodos de carbono. *IV simpósio de iniciação científica do Campus de Rio Claro – Unesp*. p. 13. 2000.
14. TOLENTINO-BISNETO, R.; BIDOIA, E. D. Desinfecção de águas de consumo através de eletrólise utilizando eletrodos de carbono. *XII Congresso de iniciação científica da UNESP*. São José do Rio Preto-SP. p. 14. 17 a 20 de outubro de 2000.
15. TOMINAGA, M. Y.; MIDIO, A. F. Exposição humana a trihalometanos presentes em águas tratadas. *Revista Saúde Pública*, São Paulo, VII. 33, n. 4, p. 413-421, 1999.
16. XUE, G. P.; JOHNSON, J. S. DALRYMPLE, B. P. High osmolarity improves the electro-transformation efficiency of Gram-positive bacteria *Bacillus subtilis* and *Bacillus licheniformis*. *Journal of Microbiological Methods*, VII. 34, p. 183-191, 1999.

TOLENTINO-BISNETO, Rodolfo; BIDOIA, Ederio D. Compararison of efficiency of the carbon, titanium and TiRuO₂ electrodes in water disinfection. *Salusvita*, Bauru, v. 21, n. 2, p. 75-82, 2002.