# NON-THERMAL EFFECT OF MICROWAVE TREATMENT ON ENZYME SUSPENSIONS PART I.: WATER ELECTROLYSIS

***Ágnes Szerencsi, Erika Lakatos, Attila József Kovács, Miklós Neményi***
University of West Hungary, Institute of Biosystems Engineering
Vár 2., Mosonmagyaróvár, 9200, Hungary
e-mail: szerencsia@mtk.nyme.hu; lakatose@mtk.nyme.hu; kovacsaj@mtk.nyme.hu;
nemenyim@mtk.nyme.hu

**ABSTRACT**

In previous researches it was showed that cellulase enzyme activity could be raised by microwave treatment. Also the physiological cell division process of microorganisms could be influenced by the microwave effect that is a higher number of cells could be reached by examining the *Saccharomyces cerevisiae*. As these structures are in aqueous media, the aim of the research was to find the effect of microwave on water that is present in all biological medium. After low-intensity microwave treatment of 1% NaCl water solution electrolysis was carried out in Hofmann's Voltameter. To focus only on the non-thermal effect, as control, an other fluid was warmed up on hotplate by the same heating parameters. We came to the result that the speed of water decomposition of microwave-treated water is higher, than those of the control. The results showed clearly, that microwave has an effect on the water and so indirectly on every substance that is present in water. In the case of samples irradiated by microwave electrolysis was faster even 48 hours after treatments compared to the control samples. Water was able to remember the electromagnetic waves of low intensity microwave treatment, so this result proved the memory effect of water.

## 1. INTRODUCTION

Recently, a lot of research projects focus on the non-thermal effects of microwave. Non-thermal heat-shock can be induced by low-intensity microwave fields (Pomerai et al., 2000). In substances such as water, molecules have permanent electric dipole because of the different electro-negatives of individual atoms. Some biological materials may be affected by very slow energies, such as in the case of hydrogen-bonded structures, in which protons may be displaced at very low energy expense. The molecules gain some energy by the dipolar polarization behavior (Schubert et al., 2005). The dielectric constant of water is very high; therefore water can store the electric field energy (Géczi, 2002). By certain frequence-, power-, and time values there are strong interactions among cells and macromolecules, but beyond these values interactions are negligible (Mátay et al., 2000). Water is an essential aqueous media of physiological processes. It takes part of cell-cell communication and transmits information via electromagnetic waves. These properties are utilized in alternative medicine and in applied researches as well. In previous researches it was found that cellulase enzyme activity could be raised 20% by microwave treatment (Neményi et al. 2008a). Also the physiological cell division process of microorganisms such as in the *Saccharomyces cerevisiae* can be accelerated by examining microwave effect (Grundler et al., 1977; Neményi et al., 2008b).
During our examinations a question aroused in connection with the microwave: does the microwave have a direct or indirect influence? Does it effect the enzyme, the water, or both? The aim of the examination was to detect the effect of microwave irradiation on

water, which is present in the suspension of the enzyme and in all aqueous media around cells. It was found, that microwave has an influence on the electrolytic decomposition of KOH water solution (Dragomir et al, 2007). In order to detect water decomposition affected by microwave, electrolysis was used to the experiments.

## 2. MATERIALS AND METHODS

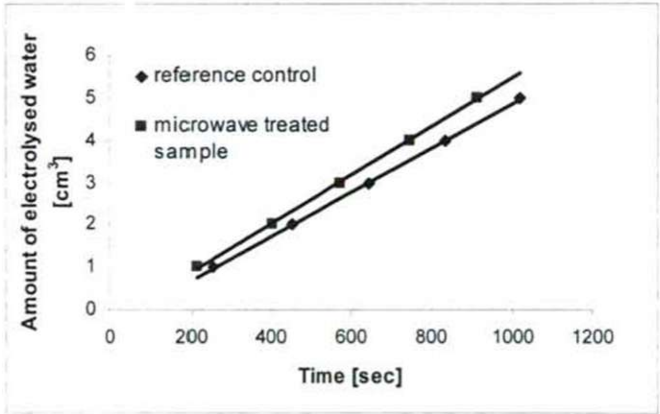
NaCl water solution (1%, 200 ml, 12 °C) was treated by microwave at 100 W, for 50 minutes till the temperature reached 45 °C. To focus on the non-thermal effect of microwave, as control an other solution was heated by the same heating parameters on hotplate. As reference control untreated 1% NaCl water solution was used. A PANASONIC NNF 653 WF domestic microwave oven with a FISO MWS-4 fiber optic thermometer was used for microwave treatments. The power was continuously emitted by the special designed inverter type oven (Lakatos, 2006) in contrast to most of the commercial ovens where the microwave power is pulsated. The treatment parameters were adjusted by a computer connected to the oven. Hence, individual programming could be carried out. Temperature changes of the treated samples were followed by the in-built fiber optic thermometer. This measures temperature based on Fabry-Perot interferometry (Datta and Anantheswaran, 2001). The teflon sample holder (diameter: 85 mm, height: 60 mm, about 340 ml) is placed in the middle of the turntable (Figure 1.). Four other containers (diameter: 38 mm, height: 100 mm of each, about 113 ml) filled with 90 g 12 °C tapwater working as water trap were evenly distributed in radial direction from the center for given distances. The aim of that is to change the inhomogeneous electromagnetic field inside the microwave oven to homogenious field.

*Fiure 1. PANASONIC NNF 653 WF type domestic microwave oven with a FISO MWS-4 fiber optic thermometer on the top of it. Inside: four watertrap containers and the sample holder in the middle of turntable*

After treatment water electrolysis was carried out in Hofmann Voltameter apparatus. In the first series of experiments 24 V was used, then 12 V in the second ones. When current is running through Hofmann's Voltameter, gaseous oxygen forms at the anode and gaseous hydrogen at the cathode. Each gas displaces water and collects at the top of the two outer tubes. The speed of electrolysis of microwave treated sample and those of control can be measured by stopper observing the decreasing level of water in the calibrated tubes. The presence of gas in the collecting tube can be detected with a smoldering match or "glowing wood splint". Oxygen will cause the match to immediately burst into a bright white flame and will burn vigorously, while the presence of hydrogen gas will create a "pop" sound due to the hydrogen rapidly burning. In our research we examined the speed of water electrolysis shortly, 24 and 48 hours after treatment. We measured changes in pH aswell. In another case only distilled water was treated by microwave and just before electrolysis was given NaCl (1%) to the sample.

## 3. RESULTS AND DISCUSSION

The first series of experiments showed (Figure 2.) that there was an obvious difference (13%) in speed of electrolysis by 24 V between microwave treated NaCl water solution and the reference control. From all cases water electrolysis was the most fast, when sample was treated by microwave and 24 V was used in Hofman's Voltameter.




*Fiure 2. Electrolysis of microwave-treated water and that of untreated reference sample – by 24 V*

The second series of experiments (Fig. 3) resulted that electrolysis (12 V) of microwave treated water sample was 10% faster than those of samples heated on hotplate by the same heating parameters. We could determine, that even after 24 and 48 hours of treatment (Fig. 4) the process of water electrolysis was (6% and 4%) faster by microwave irradiated samples than those of the controls. Due to the one-factor analysis of variance the significance level is 95% examining the difference between microwave treatment and hotplate. Comparing the two methods of treatments there were no difference in the measured pH values of the samples. The pH changed neither before nor after microwave treatment. However acidity varied (from: pH =7.10 to: pH=10.28) during water electrolysis. In case when NaCl was given only to the treated distillated water, we could determine minimal difference (3%) in speed of electrolysis between the two methods of treatments.

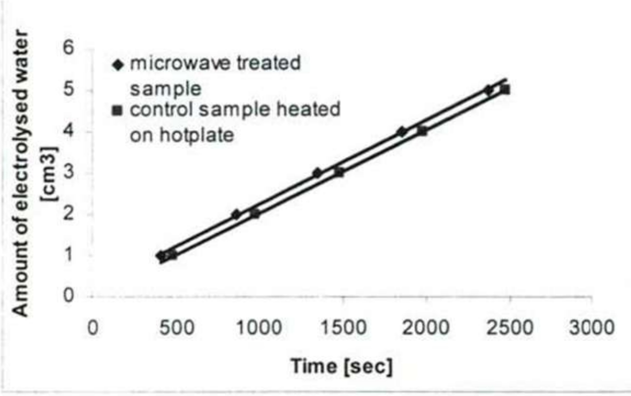



*Figure 3. Comparing the electrolysis of water, treated by microwave, and the control sample, heated on hotplate – by 12 V, shortly after the treatment*

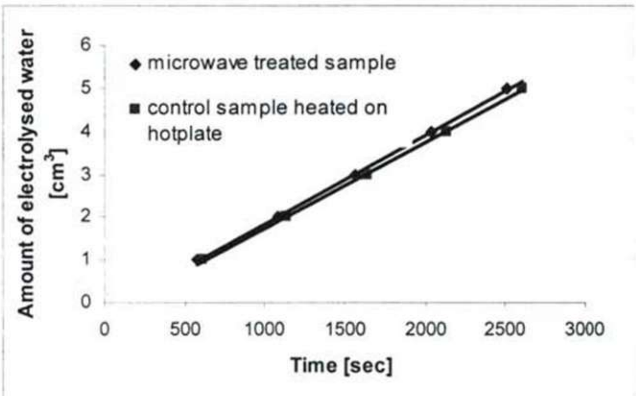



*Figure 4. Comparing the electrolysis of water, treated by microwave, and the control sample, heated on hotplate – by 12 V, 48 hours after the treatment*

The results of all experiments show clearly, that microwave treated water can be electrolysed faster, than water heated on hotplate or water hasn't been treated at all. Consequently less energy is needed to water electrolysis in case when microwave was used. In the research the non-thermal effect of microwave has been showed. It has an effect among different materials on the water aswell. In the course of our experiments it was proved, that water was surely treated by microwave. As water is a basic and unexceedable medium of most materials, it may affect all microwave treated materials in an indirect way. It happens when cellulase enzyme activity was enhanced by microwave treatment, also its medium: the water was treated. This means that it can influence the enzyme indirectly. The experiments show obviously, that water was able to remember the electromagnetic waves of low intensity microwave treatment even 48 hours long. Our results proved the memory effect of water.

## 4. CONCLUSIONS

Thank the results more information about the behaviour of water is available, which is important to carry out further research in connection with microwave treatment. The purpose of future experiments is the examination of microwave treated suspensions of enzymes and microorganisms. This contributes to the expansion of production efficiancy by second generation of bioethanol promoted also by the EU7 framework.

## REFERENCES

1. Datta, A.K., Anantheswaran, R.C. (2001): Handbook of Microwave Technology for Food Applications. Marcel Dekker, Inc., New York.
2. Dragomir R. Stanisavljev, Tomislav D. Grozdić, Milica P. Marčeta Kaninski, Antonije R. Djordjević and Dragica Lj. Stojić (2007): The microwave influence on the electrolytic decomposition of KOH water solution Electrochemistry Communications Vol. 9(5), pp. 901-904.
3. Géczi G. (2002): Összefüggések mezőgazdasági termények dielektromos jellemzőinek meghatározására, (Relationships of determination of dielectric properties of agricultural products), Mezőgazdasági Technika, April, pp. 2-4.
4. Grundler W., Keilman F., Fröhlich H. (1977): Resonant growth rate response of yeast cells irradiated by weak microwaves. Physics letters 62A: 463-466 pp.
5. Lakatos E. (2006): Treatments of liquid foodstuffs with special regard to the effect of microwave on milk. PhD thesis. Mosonmagyarovar, 85-88.
6. Mátay G., Zombory L. (2000): A rádiófrekvenciás sugárzás élettani hatásai és orvosbiológiai alkalmazásai, Műegyetemi Kiadó ISBN 963 420 658 1.
7. Neményi M., Lakatos E., Kovacs A., Szerencsi Á. (2008a): The effect of microwave treatment on cellulase enzyme activity. EurAgEng-International Conference on Agricultural Engineering, Hersonissos, Crete, Greece, 2008 06 23-25. Book of Abstracts-Agricultural&Biosystems Engineering for a Sustainable World, p. 06, Conference Proceedings CD.
8. Neményi M., Lakatos E., Kovács A.J., Szerencsi A. (2008b): Mikrohullámú kezelés hatása az élesztősejtekre (*Saccharomyces cerevisiae*). XXXII. Óvári Tudományos Nap, Agrárműszaki Kutatási és fejlesztési szekció, 2008. 10. 9., Mosonmagyaróvár, Hungary, Előadások és poszterek összefoglaló anyaga, NYME University of West Hungary, Konferencia CD.
9. Pomerai D., Daniells C., David H., Allan J., Duce I., Mutwakil M., Thomas D., Sewel P.,Tattersal J., Jones D., Candido P (2000): Non-thermal heat shock response to microwaves. Nature. 405. pp 417-418.
10. Schubert H., Regier M. (2005): The microwave processing of foods, Woodhead Publishing Limited and CRC Press LCC, Cambridge, England.