

## MAMMALIAN CELL RESPONSE TO THE APPLICATION OF INTENSE BURST SINUSOIDAL ELECTRIC FIELDS

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Biological effects of intense pulsed electric fields (PEF) have been intensively investigated in the last decade. PEFs are capable of giving biological targets unique stress dependent on its pulse duration, rise time and field strength. In general, nanosecond pulsed electric fields (nsPEF) cause intracellular effects, while micro-to-millisecond long pulses affect the cell membrane. We are using an intense burst sinusoidal electric field (IBSEF) instead of rectangular pulses that have wide frequency band. The use of IBSEFs, which have narrow frequency band, allows to give well-defined electric fields to biological targets, helping us discuss the biological effect in the frequency domain [2]. In the previous work [3], we have experimentally demonstrated that the application of non-thermal 100 kV/m IBSEFs induce the breakage of DNAs in Chinese hamster ovary cells, and the breakage depends on the frequency and the strength of the field.

Here, we discuss the mechanism of the field induced DNA breakage by means of fluorescent molecular probes in addition to comet assay. The DNA breakage can be caused by direct physical effect of electric field or secondary biological effect eventually executed by nuclease. We have investigated the dependence of DNA breakage on the culture time after the exposure to the IBSEF by means of comet assay, which shows the DNA breakage promoted by biological processes. The comet tail pattern, which indicates the degree of the DNA breakage, is promoted gradually within 1 hour after the exposure to the IBSEF. This temporal behavior depends on the frequency of the field. This experiment implies that the field-induced DNA breakage is rather secondary effect. Also we have investigated the activity in mitochondria and intracellular calcium concentration using fluorescent molecular probes.

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