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Evidence of Photon Emission from DNA in Living Systems

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The widespread, if not general, phenomenon of "ultraweak" photon emission (PE) from living cells and organisms, which is different from bioluminescence [1], has been extensively reviewed [2, 3]. The measurement of PE is made with a photomultiplier of high sensitivity in the range between 200 and 800 nm. The living material is kept within kuvettes in a dark chamber before the photomultiplier (for a more detailed description see [4, 5]). With our equipment a photon current density of 2 photons/s/cm^2 can be detected at a significance level of 99.9% within 6 h. Calculations were made with an interfaced computer.

We have now found evidence that chromatin is a photon emitter, using ethidium bromide (EB) as a probe. Probably, DNA is the most important source of "ultraweak" photon emission (or electromagnetic radiation) from living cells.

Fig. 1 shows, as an example, the temporal course of the PE signal (i.e., the total count rate minus background) of cucumber seedlings (*Cucumis sativus* L., cv. "Chinesische Schlangen"). The seedlings have been exposed to aqueous solutions of EB at concentrations between 10^{-2} and 10 mg/l. In order to examine the kinetics of EB uptake and the location of the binding sites in vivo, we analyzed EB-treated plantlets by fluorescence and electron microscopy. The treatment time was 30 s to several h, and some seedlings were, after washing, returned to tap water for 24 h.

An intense and fast binding of EB occurs almost exclusively with chromatin (Fig. 2a). After a few seconds of incubation, the nuclei of the root tips exhibit fluorescence (the material was quickly prepared in the living state). During the first few minutes, EB is transported through the roots and stems, as can be seen by progressive fluorescence of nuclei in more distant cells. After 1–3 h all nuclei of a seedling have bound EB. Since replacement of the EB solution by tap water did not lead to any fading of the fluorescence, EB is evidently irreversibly bound. We also digitized electron micrographs of nuclei by a semi-automatic morphometric system (MOP-Digiplan, Kontron) after incubation with EB (Fig. 2b). The proportion of chromatin in the condensed state decreased and increased, respectively, fairly parallely to the in vitro uncoiling and coiling of isolated DNA (Fig. 3a).

All these observations are compatible with the known intercalation of EB between DNA base pairs [6, 7]. Increasing concentrations of EB have been shown to completely unwind the supercoils in closed circular DNA, while at higher concentrations the supercoils reform but with their twists in opposite direction (Fig. 3a).

If DNA is a photon store, particularly in certain conformational states, then as it unwinds (or as the amount of decondensed chromatin increases) it should release photons. Hence, one expects a relationship between DNA unwinding and the number of released photons. This can be tested with EB binding to DNA. As shown in Fig. 3a, b, a quantitative correspondence in concentration dependence does actually occur (see also legend to Fig. 2). Since only

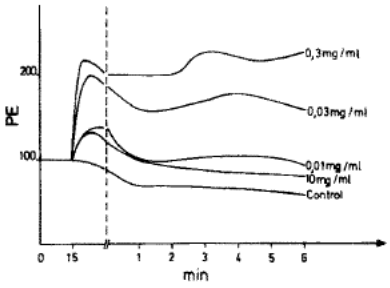

Fig. 1. Ultraweak photon emission from cucumber seedlings after incubation with aqueous solutions of ethidium bromide. The emission increases with EB concentration and decreases at higher concentrations but remains still higher than that from control plantlets. The PE is normalized to 100 counts per second (cps) for all untreated control seedlings (absolute value without background: about 100 cps)

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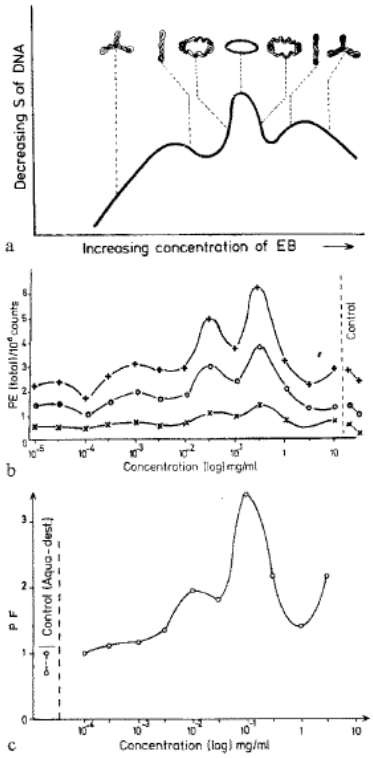

Fig. 3. a) Sedimentation (S) of DNA indicating DNA unwinding after treatment with ethidium bromide at increasing concentrations and restoration of supercoils at higher concentrations (but in opposite rotational direction). The data come from [6] but shows decreasing S as a measure of DNA uncoiling (decondensation). b) The total photon emission of cucumber seedlings shows qualitatively the same dependence on the concentration of ethidium bromide as the uncoiling and recoiling of DNA (a). There is, moreover, a clear correspondence of PE and in vitro DNA uncoiling/coiling to in vivo chromatin decondensation/condensation (Fig. 2b). c) PF, the photon emission increase factor (i.e., the count rate immediately after addition of EB to the count rate before) shows the same dependence on the EB concentration as DNA uncoiling (a) or the total number of counts (b). The plantlets were covered with an aluminium foil except the roots, which rapidly take up EB

the roots take up EB rapidly, the relationship should be more distinctive if either the other parts of the seedlings are covered (for instance with an aluminium foil) or the electromagnetic radiation is measured after a longer time of EB treatment. Actually, the observed kinetics of EB binding are in complete agreement with the theoretical considerations (Fig. 3c). Experiments on other living systems such as bean seedlings (*Phaseolus vulgaris*) and wheat grains (*Triticum aestivum*) confirmed the results obtained with cucumber plantlets. The PE of isolated, pure DNA is, however, too weak to be measured with our instrument.

The results can best be understood in the light of recent developments in DNA spectroscopy, in particular the detection of DNA excimers [8, 9]. These excited dimers provide an attractive potential between two nucleotides after excitation of only one of the monomers. In this way the nucleotides constitute active photon stores [8], operating above or at the laser threshold [10].

The biological significance of the described physical property of DNA is seen in a possible role of photon storage and emission in the control of gene activity, cell metabolism and cell communication [11]. One not understood problem of biology, the well-known C-value paradoxon [12] could also be solved in terms of electromagnetic effects of DNA. It is evident that the basic DNA content of a species is not related to its gene number and evolutionary (or organismic) complexity. Only a very small proportion (about 0.1 and 2%) of DNA operates as genetic material and is organized in nucleotide sequences according to the genetic code [13]. Models have, therefore, been proposed which suggest some regulatory role for the non-protein-coding DNA [14]. Recently, this regulatory role is being seen more in terms of some basic physical mechanisms [15], particularly the coherent electromagnetic interactions between different DNA sections [16], rather than a biochemical store of information. Considering such physical properties of DNA, we hope to find new insights into the mechanisms of phylogenetic and ontogenetic diversification of living matter.

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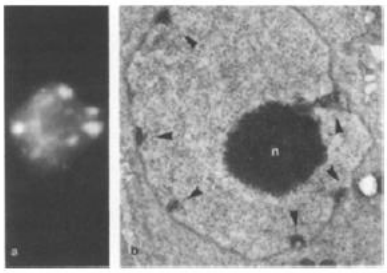

Fig. 2. Fluorescence and electron microscopic images of cucumber cell nuclei after EB treatment in vivo. a) Living, fluorescing root nucleus, 1 min after incubation: there is almost no fluorescence visible over the cytoplasm or cell wall (×900). b) Glutaraldehyde/OsO_4-fixed nucleus of a plantlet which was treated for 1 h with 0.03 mg EB/ml, showing reduced size of chromocenters (heterochromatin; arrowheads). After this treatment, about 3% of the nuclear volume are occupied by heterochromatin, while the controls exhibit 6%, on an average, condensed chromatin (×7000)

chemiluminescence in white blood cells

TABLE 26.1 Sample matrices and methods employed for biomonitoring of PAH-DNA adducts in susceptible populations

Exposure Route	Biological Matrix	Population	Analytical Method	Reference
Inhalation (Cigarette smoke)	White blood cells	Healthy smokers & non-smokers	ELISA	Perera *et al.*, 1987
Inhalation (Cigarette smoke)	White blood cells	Lung cancer patients	^{32}P-postlabelling	van Schooten *et al.*, 1992
Inhalation (Cigarette smoke)	Peripheral leukocytes	Lung cancer patients	ELISA	Tang *et al.*, 1995
Inhalation (Cigarette smoke)	Oral cavity (mouth floor & buccal mucosa)	Healthy subjects	Immunoperoxidase assay	Besaratinia *et al.*, 2000
Inhalation (Cigarette smoke)	White blood cells	Smokers	Immunoassay	Funck-Brentano *et al.*, 2006
Inhalation (Cigarette smoke)	Cervical mucosa	HPV-infected patients	Chemiluminescence assay	Pratt *et al.*, 2007
Inhalation (Environmental tobacco smoke)	Maternal & umbilical cord blood	Pregnant mothers	?	Perera *et al.*, 2007
Inhalation (Polluted air)	Placenta	Pregnant women from chemical & radioactive polluted-areas	Chemiluminescence immune assay	Obolenskaya *et al.*, 2010
Inhalation (Occupational)	White blood cells	Coke oven workers	ELISA	van Schooten *et al.*, 1990
Inhalation (Occupational)	White blood cells	Iron foundry workers	ELISA	Santella *et al.*, 1993