Effects of Light on the Translocation of Bacterial DNA in *Solanum lycopersicum esc*

We have previously reported[1] results showing that exogenous DNA molecules could migrate in the plant and, after some depolymerization, be taken up by cell nuclei without modification of their primary and secondary structures[2].

In this paper, we studied the effects of light on the amount of DNA absorbed by the plant and on depolymerization during translocation.

Tomato plantlets are developed for 15 days (20 °C, 90% humidity) either in daylight or in darkness. They are subsequently incubated either in light or in darkness in the presence of bacterial DNA. Exogenous labelled DNA (2×10^6 dpm/μg) is extracted from a thymineless strain of *Escherichia coli* (CR 34), cultured on a medium containing ^{3}H-thymine. The stems of the plants are dipped into a solution (200 μg/ml) of *E. coli* ^{3}H-DNA for 6 h. They are then transferred to water for 48 h; (this being the time required for the nuclei to take up the foreign DNA)[2]. The part of the plant submerged in the various solutions is removed before homogenization and the DNA from the remainder of the stem and the cotyledons is extracted by a method already described[3]. This DNA is analysed by centrifuged chromatography on DEAE-cellulose columns[4].

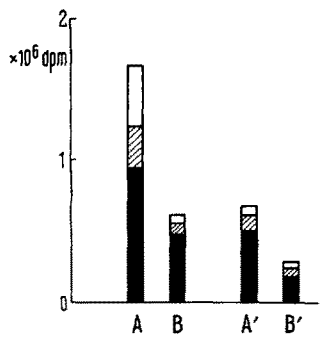

Fig. 1. Radioactivity of exogenous DNA absorbed by plantlets either in light or in darkness (radioactivity is related to 100 μg endogenous DNA). (A) plantlets developed in light and incubated in light. (B) plantlets developed in light and incubated in darkness. (A′) plantlets developed in darkness and incubated in light. (B′) plantlets developed in darkness and incubated in darkness. Every letter corresponds to 3 series of 10 plantlets drawn in black, haching or white. The black series took up the smallest amount of DNA and the white series the largest, the hachures series being intermediate (for instance, for A: the black = 0.9×10^6 dpm; the hachures series = 1.2×10^6 dpm; the white series = 1.6×10^6 dpm).

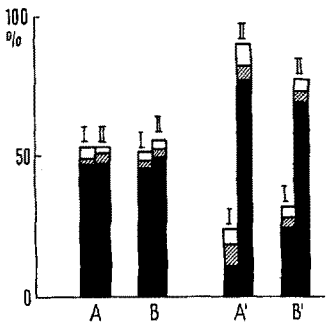

Fig. 2. Percentage relation of fractions of exogenous DNA absorbed by plantlets exposed either to light or to darkness. (Fraction I: molecular weight of $2 \times 10^5 - 1 \times 10^6$). (Fraction II: molecular weight above 1×10^6). (A) plantlets developed in light and incubated in light. (B) plantlets developed in light and incubated in darkness. (A′) plantlets developed in darkness and incubated in light. (B′) plantlets developed in darkness and incubated in darkness. Every letter corresponds to 3 series of 10 plantlets. In black, the smallest percentage of DNA in fractions I and II; in white, the largest amount, the hachures lines being intermediate.

A parallel study was made of the synthesis of endogenous DNA under the same conditions while incubating the plantlets in a solution of tritiated thymidine with the same specific activity as bacterial ^{3}H-DNA.

As shown in Figure 1, under identical incubation conditions, the plantlets developed in daylight take up larger amounts of DNA than those developed in darkness (A compared with A′ and B with B′). Furthermore, light also affects the course of incubation. Under identical development conditions, the plantlets that received bacterial DNA in daylight take up more of this DNA than those that received it in darkness (A compared with B and A′ with B′).

As indicated in Figure 2, the plantlets developed in daylight have a more depolymerized exogenous DNA than the plantlets developed in darkness (compare A and A′, B and B′). The influence of light on the course of incubation does not appear so clearly.

In light, plants incubated in the presence of tritiated thymidine synthesize, during the same time period, an amount of radioactive DNA 15–20 times less than the amount of bacterial DNA absorbed by plantlets developed and incubated in light. Furthermore light or darkness does not seem to affect in any way either the amount of endogenous DNA synthesized or its degree of polymerization.

The fact that under our experimental conditions light affects the translocation of foreign DNA without influencing, in the least, the synthesis of endogenous DNA proves that the 2 phenomena are distinct.

Since temperature and humidity are held constant in all experiments, sweating cannot be held responsible for the major uptake of foreign DNA in light. We may therefore conclude that the translocation of foreign DNA is connected with active transportation.

In order to explain the higher molecular weight of exogenous DNA found in plants cultivated in darkness, we tested in vitro the rates of DNAse I and II in different series of plantlets. Our failing to observe any difference does not exclude the possibility of a photosensitive activity of the DNAse in situ[5].

Résumé. Nous avons étudié l'influence de la lumière sur la translation de l'ADN d'origine bactérienne chez des plantules de tomate. La lumière augmente la prise d'ADN étranger. Cet ADN est, par contre, dans un état moins polymérisé que chez les plantules exposées à l'obscurité. Dans les mêmes conditions expérimentales, la lumière n'a aucune influence sur la synthèse de l'ADN endogène, réalisée à partir de petits précurseurs.

M. STROUN, P. ANKER and J. REMY

Département de Radiobiologie, Laboratoire de Biochemie cellulaire, Centre d'Etude de l'Energie Nucléaire, Mol (Belgium), 8th May 1967.

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[5] This work was done under a Euratom/C.E.N. contract, and was supported by grants from the Belgian Fonds de la Recherche scientifique fondamentale collective.