

Influence of Light on the Autospore Number of some Chlorococcal Algae

J. NEČAS

Laboratory of Algology of the Microbiological Institute of the Czechoslovak Academy
of Sciences, Třeboň

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Abstract. The method of cultivation of algae on the surface of solid medium under regulated conditions was used for studying the influence of light on the autospore number with three strains of chlorococcal algae: *Scenedesmus quadricauda*, *Scenedesmus obliquus* and *Chlorella kessleri*. The results of these experiments show that not only light but the type of medium as well, exert a very important influence upon both nuclear and cell division and the whole course of the life cycle of algal cells. The number of autospores produced by single cells can be ascertained reliably on the surface of the solid medium. Owing to this fact the relations in cellular populations can be expressed with greater precision in comparison with the evaluation of cultures in liquid media.

The reproduction of algae is a subject of many algological studies. Different types of reproduction with various species of algae and conditions, inducing or blocking the generation process, eventually enhancing or retarding its course, were studied. Relatively little attention was dedicated (evidently for technical reasons) to the influence of different environmental factors on the number of autospores produced with chlorococcal algae. It follows from the collective and general works of recent years (ERBEN 1962, Ettl and al. 1967) that further works concerning this problem are needed. The control of the generation process of algae by cultivation conditions is one of the first tasks of the working teams engaged in the cultivation of algae on a large scale. This is the reason for more detailed studies dedicated to algal strains suitable for these purposes (e.g. *Chlorella* — MORIMURA 1959, TAMIYA 1964, TAMIYA 1966). They concern predominantly synchronous cultures. TAMIYA (1964) mentions some factors influencing the number of autospores produced by *Chlorella*. TAMIYA (1966) gives a survey of the autospore number to which the cells of some chlorococcal algae divide under the given cultivation parameters. As to *Scenedesmus quadricauda*, the influence of light and cultivation temperature on the number of produced autospores in liquid medium was verified by KOMÁREK and RŮŽIČKA 1969

in our laboratory. The influence of anomalous nutrient relations in medium was confirmed by PŘIBIL and MARVAN (in press).

It has been found in some of our experiments with mutagens that the cultivation of algae on solid media can be successful for these studies. This fact is shown by an experiment demonstrating the influence of light on the number of produced autospores with three strains of chlorococcal algae in this paper.

Material and Methods

Material and Cultivation Technique

The following strains of chlorococcal algae were studied and compared: *Scenedesmus quadricauda* strain GREIFSWALD/15 and *Scenedesmus obliquus* LHOTSKÝ (both strains from our laboratory collection) and *Chlorella kessleri* strain LARG-1 (from the collection of the Laboratory of radiation genetics, Institute of general genetics of Acad. Sci. of USSR in Moscow). Cultures of these strains were pre-cultivated under sterile conditions in cultivation flasks placed on a shaker at 25° in light of lower intensity than the lowest value used in the experiment (about 4 W/m²). The cultures were then inoculated onto the surface of solid medium in Petri dishes.

Cultivation Medium

Two percent agar with mineral nutrition according to Björkman modified by KOMÁREK and RŮŽIČKA (1969) was used as a solid cultivation medium. Yeast autolysate and glucose were added to the solid medium of experimental variants with complete solid medium. The cells (coenobia) were distributed on the surface of the medium in a way enabling a reliable count of the number of autospores from one mother cell. Not only the whole experiments were repeated, but the new distribution of single cells was performed twice or three times, always after one or two life cycles.

Evaluation of the Results

The responses of the cellular populations under the given cultivation conditions were expressed by frequencies of cell divisions to 8 or 4 autospores. Other autospore numbers from one mother cell were relatively less frequent in the cultivation conditions used and therefore they were not involved in the evaluation. The Petri dishes were placed on a cultivation shelf at three illumination levels given by distances from the lighting source: 15, 10 and 7 W/m², and at a temperature of 28° to 30°. Another variant was placed in an incubator in darkness at a temperature of 30°.

Results and Discussion

The method of cultivation on the surface of solid medium is very suitable for evaluating the influence of different environmental factors on the number of autospores into which the cells of chlorococcal algae divide in the gene-

ration phase of their life cycle. This method provides a precise picture of the population of cells growing under given conditions and makes possible the expression of its state by frequencies of types distinguishable in the given population and its characterisation by generally comparable values.

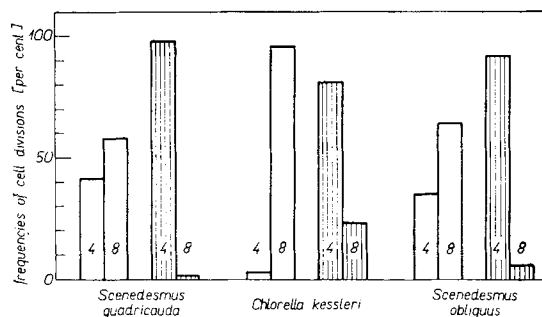


Fig. 1. Percentual frequencies of the four autosporous (4) and eight autosporous (8) cell divisions in light 15 W/m^2 $\square$ and in darkness |||| with three strains of chlorococcal algae (minimal medium).

The average number of autospores corresponding to the number of cell doublings in liquid medium is usually given in synchronous populations by the alternation of cell number in volume unit after one or more life cycles. But its determination is often subject to greater methodological fault. The frequencies of single division types cannot be found in this way. The per-

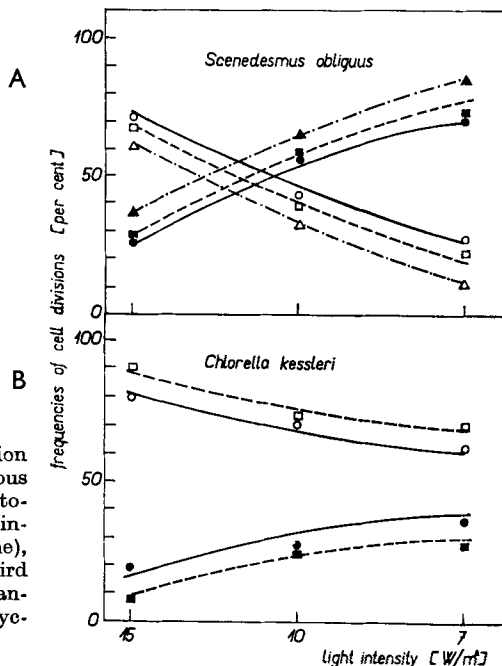


Fig. 2. The dependence of the mutual relation of percentual frequencies of four autosporous cell divisions (full marks) and eight autosporous cell divisions (empty marks) on the intensity of light. The first cell division (full line), the second life cycle (dotted line), the third life cycle (dashed line). (The lines did not change their position significantly in further life cycles.)

centual frequencies of the cell division to 4 and 8 autospores with the strains studied at given conditions are shown in Fig. 1. Their mutual relation characterizes the strain and shows that light enhances the frequencies of cell division to 8 autospores. *Chlorella* is the most sensitive strain from this point of view. The cells of *Scenedesmus obliquus* respond more sensitively to light than the cells of *Scenedesmus quadricauda*. This fact is proved by the results given in Fig. 2A. This diagram also shows a certain adaptation of the

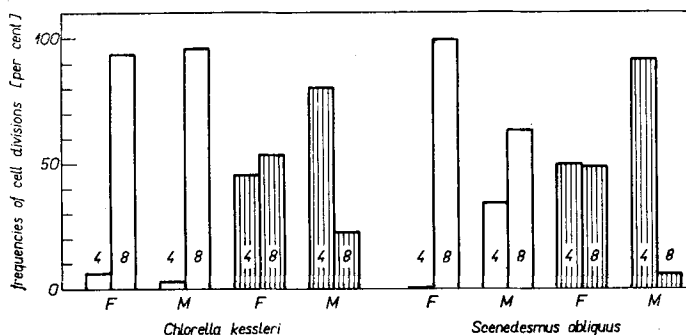


Fig. 3. Percentage frequencies of the four autosporous (4) and eight autosporous (8) cell divisions at the light intensity of 15 W/m² □ and in darkness ▨ with two strains of chlorococcal algae on complete medium (F) and on minimal medium (M).

population to the given conditions. With *Chlorella* (Fig. 2B) the dependence of this frequency relation of both division types on the light intensity is not very expressive. It is due, to a certain degree, to the enhancement of division frequencies to 16 autospores at the increased illumination intensities. Nevertheless, strain differences can be presupposed. The necessity of more detailed studies of the influence of light on the nuclear and cellular division of algal cells is evident. As some experiments were carried out on complete medium as well, it is possible to see in Fig. 3 the interaction influence of medium with light. It follows from this fact, that organic nutrition enhances the division frequencies to a higher autospore number.

The number of autospores into which the cells of chlorococcal algae divide is based on the relation between the nuclear and cellular division. Both processes are temporary and functionally linked together but their mutual rate is determined by different conditions. If it is nuclear division only which is retarded by some factor, but the protoplast can divide into as many cells as nuclei which arise, the number of autospores produced by a cell can be reduced to two in the extreme case. On the contrary the protoplast divides into as many autospores as nuclei which could arise in a given time, if it is the protoplast division that plays the deciding role. It is evident that there is only the question of a mutual temporary shift of successive nuclear division and more or less simultaneous cell division, as it is the case with *Scenedesmus quadricauda*. Greater attention must be paid to the metabolic side of both processes. Nuclear division necessitates an intensive biosynthesis of other materials than are involved in division of the protoplast.

But, it is difficult to presuppose, that it is the metabolic side of both processes only, that can explain the whole process of nuclear and cellular division.

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J. NEČAS. Algologická laboratoř Mikrobiologického ústavu ČSAV, Třeboň: **Vliv světla na počet autospor u některých chlorokokálních řas.** — *Biol. Plant.* **11** : 465–465, 1969.

Metoda kultivace řas na povrchu pevného agarového média má určité výhody vzhledem ke kultivaci v tekutém médiu při studiu vlivu různých faktorů na počet autospor, na něž se rozdělí buňky chlorokokálních řas. Touto metodou jsem studoval vliv světla u tří kmenů: *Scenedesmus quadricauda*, *Scenedesmus obliquus* a *Chlorella kessleri*. Uvedené výsledky ukazují, že nejen světlo, ale také typ média se významně uplatňují v působení na jaderné i buněčné dělení a celý průběh životního cyklu buněk řas. Počet autospor, na něž se jednotlivé buňky dělí, je možno určit přímým pozorováním na povrchu pevného média. Frekvence jednotlivých typů dělení v buněčných populacích mohou být takto zjištěny. V tekutých médiích nejsou možná přímá pozorování v takovém rozsahu.