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EFFECT OF LIGHT SOURCE FOR GROWTH ON PHOTOSYNTHETIC REACTIONS OF *ANABAENA VARIABILIS* PARTICLES

SHAW S LEE

Department of Agricultural Chemistry, Oregon State University Corvallis, OR 97331, U S A

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Key Word Index—*Anabaena variabilis*, Cyanophyceae, light source, Hill reaction and photophosphorylation

Abstract—Photosynthetic particles were prepared from the blue–green alga *Anabaena variabilis* grown under two different light conditions Photosynthetic activity of particles from fluorescent-light-grown cells was lower than that of particles from tungsten-grown cells The photosynthetic activities of particles from fluorescent-light-grown cells could be increased by moving these cultures to tungsten light The extent of recovery depended on the ratio of fluorescent growth to the tungsten growth period It was concluded that the difference in photosynthetic activities between tungsten and fluorescent grown cell-free preparations is more likely due to a lack of red light rather than to disturbance of pigment contents

THE MANUFACTURE of NADPH and ATP in photosynthesis, grana development, and biosynthesis of chlorophyll have been shown to depend on light [1–3] Artificial tungsten light was first used by Warburg[4] for the cultivation of algae Recently, fluorescent lamps have taken the place of tungsten light, for they emit much less heat and cause no significant changes in incubator temperature Growth studies indicate that fluorescent lamps are suitable for artificial illumination of algae However, it is important to know whether fluorescent light influences Hill reaction and photophosphorylation activities of the algae in a special way

The present communication reports the effect of light source for the growth of a blue–green alga *Anabaena variabilis* on the Hill reaction and photophosphorylation activities in cell-free preparations

RESULTS AND DISCUSSION

The first observation revealed that *Anabaena variabilis* growing in fluorescent light possesses only 50–60% photophosphorylation activity of the organism growing in tungsten light (Table 1) Hill reaction activity of particles from fluorescent-light-grown cells is 30–50% of that of particles from tungsten grown cells in six experiments (Table 1) This clearly indicates that fluorescent light is less effective than tungsten light in producing competence for both photosynthetic phosphorylation and Hill activity

Abbreviations NADP Nicotine adenine dipyridin nucleotide phosphate ATP adenosine triphosphate, TCIP– 2 3 6-trichlorophenol indophenol

[1] ARNON D I ALLEN M B and WHATLEY P R (1954) *J Am Chem Soc* **76**, 6324

[2] VISHNIAC, W and OCHOA, S (1951) *Nature* **167**, 768

[3] ANON (1970) *Electron Microscopy and Plant Ultrastructure* (ROBARDS A W ed), p 105, McGraw-Hill, New York

[4] WARBURG O (1919) *Biochem J* **100**, 230

After cells had been grown in fluorescent light for 4 days, the Hill activity of particles from fluorescent light grown cells could be increased from 30 to 100% of that of particles from tungsten light grown cells by moving those cultures to tungsten light The extent of recovery depends on the ratio of the growth periods spent in fluorescent and tungsten light (Table 1 expts 3–6) 4 days fluorescent-1 day tungsten growth gives particles showing 60–70% of the activity of those obtained from organisms grown in continuous tungsten light Sixty hr fluorescent-81 hr tungsten growth gives particles with 80 100% Hill activity of the control Particles from fluorescent-light-grown cells were usually less active in Hill reaction and photophosphorylation activities than particles from tungsten-light-grown cells However particles from 4 days fluorescent 1 day tungsten-light-grown cells possess 90% of the photophosphorylation activity and 60 70% of the Hill activity of those from tungsten-light-grown cells Particles from 60 hr fluorescent-81 hr tungsten-light-grown cells are almost as active as those from continuous tungsten-light-grown cells (Table 1)

TABLE 1 EFFECT OF LIGHT SOURCE FOR GROWTH ON HILL REACTION AND PHOTOPHOSPHORYLATION ACTIVITIES OF *Anabaena cantabilis* PARTICLES

Expt No	Light source for growth	Hill activity (μmol TPIP reduced/mg chlorophyll/hr)	(%)	ATP synthesized (Δμmol Pi mg chlorophyll hr)	(%)
1	Tungsten	104	100	711	100
	Fluorescent	43	41	396	56
2	Tungsten	202	100	582	100
	Fluorescent	85	42	318	55
	4 days Fluorescent-1 day tungsten	135	67		
3	Tungsten	176	100	460	100
	Fluorescent	86	49	260	57
	4 days Fluorescent-1 day tungsten	110	63	420	91
4	Tungsten	220	100	380	100
	Fluorescent	74	34	230	60
	4 days Fluorescent-1 day tungsten			340	90
	60 hr Fluorescent-60 hr tungsten	145	66		
5	141 hr Tungsten	280	100	500	100
	141 hr Fluorescent	83	30	260	52
	60 hr Fluorescent-81 hr tungsten	230	82	400	80
6	141 hr Tungsten	260	100	600	100
	141 hr Fluorescent	99	38	450	75
	60 hr Fluorescent-81 hr tungsten	275	105	550	92

Hill activity towards NADP by particles from fluorescent-light-grown cells is only half of that by particles from tungsten-light-grown cells Finally particles from 60 hr fluorescent-60 hr tungsten-light-grown cells are as active in reducing NADP as those from continuous tungsten-light-grown cells (Table 2)

Figure 1 shows light saturation curves for Hill reaction activity of tungsten-light-grown and fluorescent-light-grown *Anabaena* particles The Hill activity of particles from tungsten-light-grown cells exceeds that of particles from fluorescent-light-grown cells over the entire range of light intensities employed

The difference in Hill reaction and photophosphorylation activity between cell-free preparations from tungsten-light-grown and fluorescent-light-grown cells cannot be attributed to the disturbance of chlorophyll and carotenoid content[5] since absorption spectra of the two types of particles showed no significant difference in pigment content Instead, this difference in activity is more likely due to a lack of red light since tungsten light

TABLE 2 EFFECT OF LIGHT SOURCE FOR GROWTH ON RATE OF $NADP^+$ REDUCTION

Expt No	Light source for growth	μmol NADP (reduced/mg chlorophyll/hr)	(%)
1	120 hr tungsten	68	100
2	120 hr fluorescent	36	53
3	60 hr fluorescent-60 hr tungsten light	67	99

Reaction mixtures contained in μmoles 10 phosphate buffer (pH 7 8), 70 NaCl, 1 2 $NADP^+$, 90 $MgCl_2$, saturating amount of ferredoxin and *Anabaena* particles containing 20 μg chlorophyll in a final volume of 3 ml

completely restores the photophosphorylation and Hill activities of fluorescent light grown cells (Tables 1 and 2) Particles from organisms grown for 60 hr in fluorescent light followed by 60 hr in tungsten light give the same rate of NADP reduction as those from organisms grown in continuous tungsten light Furthermore, the light saturation

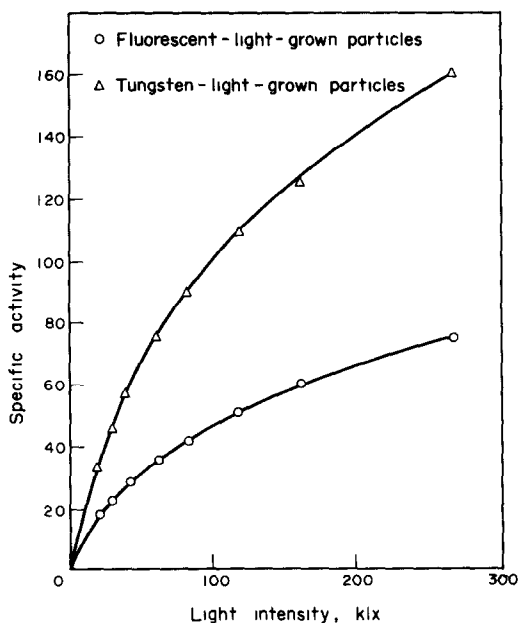

FIG 1 LIGHT SATURATION CURVE OF TUNGSTEN-LIGHT-GROWN AND FLUORESCENT-LIGHT-GROWN *Anabaena* PARTICLES

Sp act was expressed in terms of μmol TCIP dye reduced/mg chlorophyll/hr Assay of TCIP photoreduction is described in the text

[5] ARGUS S (1951) *Physiol Plant* **4**, 742

curve of tungsten-light-grown-cells preparation was quite different from that of fluorescent-light-grown cell preparations, suggesting that the deficiency is in or very close to the photochemical act

Growth experiments (data unpublished) have established that *A. variabilis* grows equally well in tungsten and fluorescent light and it may be concluded that the Hill reaction and photophosphorylation are not growth limiting in this organism

EXPERIMENTAL

The culture of *Anabaena variabilis* used in these experiments was a gift of Dr. J. Meyers. The cells were grown according to the procedure of Kratz and Meyers[6] except the inorganic salt concentration were diminished slightly to prevent precipitation. All cells grown under tungsten and fluorescent light (short wave type) were collected by centrifugation and washed 2× with dist. H_2O and 1× with sucrose-NaCl soln. The particles were prepared according to the method of Susor and Krogmann[7] using sonic oscillation to break the cells. The particles were stored at −15° instead of liq. N_2 temp. Chlorophyll was extracted with 80% MeOH-H_2O and was determined by the method of Macknney.[8] Photophosphorylation was determined by the method of Avron and Jagendorf as modified by Duane and Krogmann.[9] Hill reaction activity to 2,3,6-trichlorophenol indophenol was measured by the change in absorbance at 620 nm as described by Jagendorf.[10] NADP reduction was followed by the increase in absorbance at 340 nm.

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[6] Kratz, W. A. and Myers, J. (1955) *Am. J. Botany* **42**, 282.
[7] Susor, W. A. and Krogmann, D. W. (1964) *Biochim. Biophys. Acta* **88**, 11.
[8] Mackinney, G. (1941) *J. Biol. Chem.* **140**, 315.
[9] Duane, W. C. (1964) Master Thesis, Wayne State University, Detroit.
[10] Jagendorf, A. T. (1956) *Arch. Biochem.* **62**, 141.