tendon. GUSTAVSON[1], also, reports that for various collagens, chemical methods indicate that polysaccharides do not stabilize the structure.

Apart from the high value for T_t in the intervertebral disc sample, the results obtained by physical methods show that collagen retains its characteristic thermal behaviour even when associated with large amounts of polysaccharide.

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A mass-spectroscopic study of the EMERSON enhancement effect

During 1955–1958, the late Professor R. EMERSON discovered a synergistic effect on the rate of photosynthesis when algae were simultaneously illuminated with two light beams of different wavelength[1]. The discovery of the EMERSON enhancement effect[2–8] has been interpreted to mean that photosynthesis involves two separate photoreactions, sensitized by two pigment systems. In view of the effect of light on respiration[9], and the limitations of the methods (manometry[1,2] and polarography[3–7]) so far employed, an isotopic study of the EMERSON effect permitting the separation of concurrent evolution and uptake of O_2 during illumination appeared desirable.

We have used the mass spectrometer inlet system of HOCH AND KOK[10] permitting continuous sampling of gases dissolved in liquids. The experimental methods were similar to those described earlier[11]. Light beams of appropriate wavelengths were obtained from a 750-W tungsten lamp, filtered by a 12-in water filter to remove the infrared radiation and isolated by the combination of a second-order Bausch and Lomb interference filters (half bandwidth, 10 mμ) with suitable sharp cut off coloured glasses. In these experiments, the two light beams were brought to focus on the surface of the cuvette at an angle of 30°. Several neutral-density wire screens were used to vary the intensity of the incident light.

A suspension of *Chlorella vulgaris* (growth conditions: 25°; 1000 ft candles; modified Knop's medium; 3% CO_2 in air; 2-day-old culture) served as the experimental material. Heavy oxygen ($^{18}O_2$) was introduced by shaking it with an algal suspension in a syringe.

The rate of photosynthesis caused by a single beam of light, increases linearly with light intensity; then the light curve ($P = f(I)$, where P = rate of photosynthesis and I = intensity of light) bends, and finally saturation is reached. A part of this

relationship is shown in Fig. 1 for light of 650, 710 and 720 mμ. It is important to mention that the unit of light intensity for different wavelengths was not the same. The maximum available intensities (I_{max} for 650 mμ; I_{max}' for 710 mμ and I_{max}'' for 720 mμ) are all represented by 100 on the abscissa in Fig. 1. The light curves do not suggest that different saturation levels are reached at different wavelengths. The curves for photosynthesis (solid points) were obtained by calculation from the rates and concentrations of the $^{16}O_2$ and $^{18}O_2$ exchange in light. They are thus corrected for the respiration in light. However, the curves with open points are corrected for respiration in the dark (before and after illumination); in other words, they are equivalent to photosynthetic rates obtained by manometry or polarography. The same general shape is shown by both curves. The difference between them is a measure of the effect of light on respiration. At 650 mμ, the effect of light is negligible on the O_2 uptake up to $I = 0.08\text{–}0.10 \cdot I_{max}$, I_{max} being the maximum available 650-mμ intensity. This is the linear part of the light curve. Later, the O_2 uptake increases with intensity. In the case of 710-mμ light, the differences between the solid and

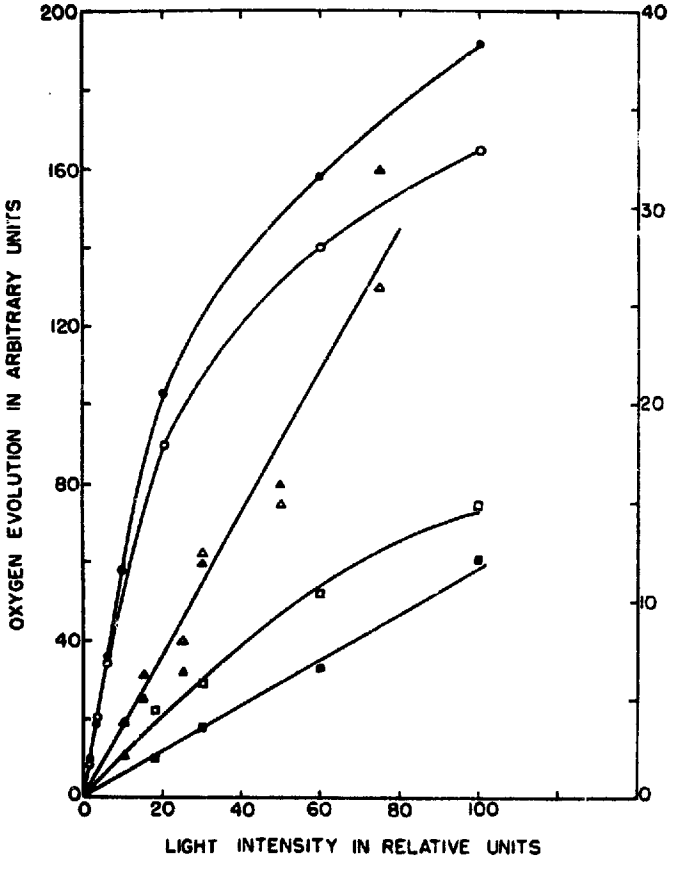

Fig. 1. O_2 evolution in *Chlorella vulgaris* plotted against incident intensities of different wavelengths of light. ●—●, rate of photosynthesis in 650-mμ light corrected for "light respiration"; O—O, rate of photosynthesis in 650-mμ light corrected for dark respiration; ▲—▲, the same for 710-mμ light corrected for light respiration; △—△, the same for 710-mμ light corrected for dark respiration; ■—■, the same for 720-mμ light corrected for light respiration; □—□, the same for 720-mμ light corrected for dark respiration. All the points for 650 and 710-mμ light are averages of 5 experiments. The points for 720-mμ light are averages of 2 experiments. The ordinate on the left hand side is for the rates in the 650-mμ light and that on the right hand side is for both 710 and 720-mμ light. The unit of relative light intensity (on the abscissa) is different at each wavelength; 100 reprents the maxi...m intensity used at each wavelength.

open points are very small. A closer examination reveals that up to $I = 0.30\ I_{max}'$, the solid points are slightly below the open points showing inhibition of O_2 uptake. On the other hand, at $I = 0.80\ I_{max}''$, a stimulation of respiration is seen. In 720-mμ light, the O_2 uptake is clearly inhibited at all the intensities used.

With this preliminary information on the effect of light on respiration in *Chlorella vulgaris* samples, we measured the rates of O_2 evolution in separate and combined beams of light, under varying intensity conditions. Care was taken to stay within the linear part of the light curves. The EMERSON enhancement effect was calculated from the isotopic data (*i.e.* data corrected for light respiration, rather than for dark respiration), according to the following equation:

$$E = \frac{R_{650\,+\,\text{far red}} - R_{650}}{R_{\text{far red}}}$$

where $R_{650\,+\,\text{far red}}$ is the rate of photosynthesis in the combined beams, R_{650} is the rate in 650-mμ light and $R_{\text{far red}}$, the same in the far-red light. The results are shown in Fig. 2. It can be seen from this figure that (a) a maximum EMERSON effect of $E = 8$ was obtained when 650-mμ light of 0.10 I_{max} and 720-mμ light of 0.09 I_{max}'' were combined: (b) if the intensity of far-red light was kept constant, the EMERSON effect increased with increasing intensity of 650-mμ light, and (c) if the 650-mμ light intensity is kept constant, a decrease in the 720-mμ intensity results in an increase in the EMERSON effect. Essentially similar results were obtained when the supplementary light was of 480 mμ.

We have thus confirmed the EMERSON enhancement effect in *Chlorella vulgaris* by a method that can distinguish between the effect of light on respiration and photosynthesis. We further found that light has a differential influence on the O_2 uptake

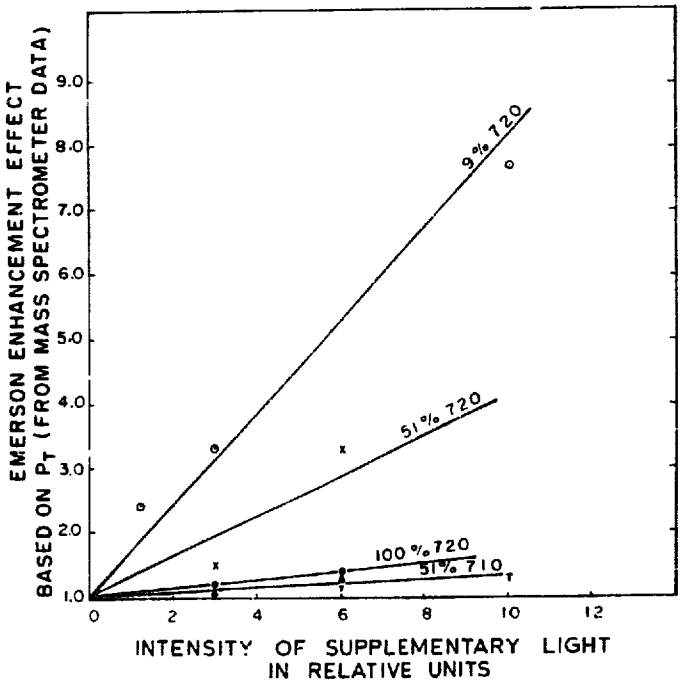

Fig. 2. The EMERSON enhancement effect (E, see equation in the text) plotted as a function of different intensities of supplementary 650-mμ light for varying intensities of far-red light. The numerical values of intensities in this graph are comparable with those in Fig. 1. P_T on the ordinate refers to the rate of total photosynthesis corrected for light respiration.

in *Chlorella vulgaris*. An inhibition by low intensity of far-red light and a stimulation by high intensity 650-mμ red light was observed. These effects on O_2 uptake are similar to those observed with *Anacystis nidulans*[12].

In an independent study, MAYNE AND BROWN[13] have confirmed the enhancement effect in another species of Chlorella—*Chlorella pyrenoidosa* (Emerson's strain 3)—by mass spectrometry; their measurements were made in the gas phase above the liquid. The results presented in this paper, were obtained from measurements made directly on the gases dissolved in the liquid phase[10]. The latter method[10] does not involve complications due to the diffusion barrier presented by the liquid–gas interface.

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