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PHYSICOCHEMICAL PROCESSES IN A PHOTOEXCITED GLYCINE SOLUTION

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Data obtained with a photomultiplier (PM), during recording of the radiation from an excited glycine solution, add to the views formed previously regarding its after-luminescence and based on a study of the mitogenetic radiation of glycine by a method of biological detection.

We may perhaps first summarize briefly the original data, full details of which were given in [1-4]. Soon after short-term irradiation (seconds) of a 0.5% glycine solution with light from a mercury-quartz lamp, the intensity of which was reduced by about 4 orders of magnitude, long-term (several hours) radiation arises in the solution in the UV region of the spectrum. This radiation disappears after the solution is heated to 80°C. After dialysis of the irradiated amino acid through collodion film, the radiation is preserved only in the solution in the inner compartment of the container. It was suggested that this radiation is connected with the active influence on the glycine molecule of a polypeptide formed during the first few minutes after photoexcitation. Its concentration, however, is maintained at a very low level, due to the considerable reversibility of the condensation process.

In fact, addition of a small portion of emitting glycine to a much larger volume of fresh solution led to the appearance of radiation in the latter, which behaved in the same way as the radiation of the original solution. These "transfers" could be repeated many times.

It was also shown that during the first minutes after photoexcitation of glycine, its emission spectrum contains bands characteristic of free NH_2 , CO, and OH radicals, which later were replaced by the appearance of a band in the 230 nm region, corresponding to fluorescence of NH_3 . It can thus very probably be concluded that a chemically active polypeptide, an analog of the prosthetic group of the enzyme deaminase, capable of inducing a mild degree of oxidative deamination of the glycine molecule, is synthesized from the initial photodissociation products.

Later facts obtained by the use of a photomultiplier, recording in the visible region of the spectrum, confirmed and, at the same time, supplemented the previous data.

EXPERIMENTAL METHOD

To investigate the fluorescence of glycine, the apparatus illustrated in Fig. 1 was set up. The FÉU-106 PM, with multialkali photocathode, capable of recording light rays within the spectral range from 170 to 830 nm, was used as radiation receiver. The PM was tuned to operate on the pulsed mode (counting photons). The output signal, after appropriate amplification, was recorded by the pulse counters 1 and 2. To reduce thermoemission noise the PM was placed in the chamber 4, packed with dry ice, and in this case the frequency of counting of dark pulses on the plateau of the counting curve did not exceed 1 Hz. To increase the effective area of the photocathode the quartz photon 7 was fixed on the end window of the PM by means of optical glue. Radiation from the cuvette 13 to be investigated was focused on

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TABLE 1. UV Component in Spectrum of Radiation from 0.5% Glycine Solution

Series of experi- ments	Object on which measurements made	Number of measure- ments	$\overline{N}_{qu} - \overline{N}_{g1}$	t	Reliabil- ity of presence of mito- genetic radiation
I	Unirradiated gly-	95	0,25	0,086	0,54
H	Irradiated glycine	181	7,7	3,65	0.999
H	Irradiated glycine	79	2,2	0,7	0,75
ÍV	after heating Transfer of irra- diated glycine	152	8,5	3,7	0,999

TABLE 2. Initial and Final Phases of Radiation from Glycine Solution

Object on which measurements made	Number of measure- ments	Nqu - Ng1	t	Reliability of presence of mito- genetic radiation
Irradiated glycine:				ĺ
First group	95	6,05	2,08	0,96
Second group	86	9,5	2,6	0,99
Transfer of irradiated glycine:				·
First group	76	9,6	2,96	0,995
Second group	76	7,6	2,34	0,98
	i	1	1	1

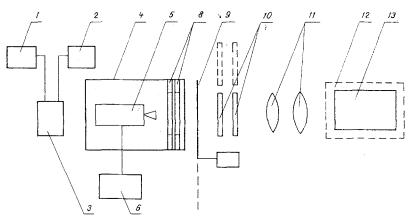


Fig. 1. Scheme of apparatus for recording mitogenetic radiation. 1, 2) pulse counters, 3) amplifier, 4) chamber containing PM, 5) the FÉU-106 PM, 6) high-voltage rectifier, 8) quartz windows, 9) modulator, 10) quartz and glass plates, 11) lenses, 12) thermostat, 13) cuvette containing glycine.

the PM by means of the optical system 11. The radiation was modulated by an obturator consisting of the half-disk 9. Under these circumstances, when the radiation was shut off, pulses from the PM were recorded by the counter 1, but when the radiation could pass through, the output pulses from the PM were led to the counter 2. The useful signal due to the light radiation was determined by subtracting the readings of the counters. In this way it was possible to analyze slow changes in frequency of the dark pulses of the PM during its operation.

Measurements were made as follows. Glass and quartz plates (filters) 10 were placed alternately in the path of the radiation, and the number of pulses from each of them was determined on the counter 2; this number corresponded to 100 dark background pulses of the PM, recorded by counter 1. In this way the useful signal was determined during passage of the radiation either through quartz ($N_{\rm qu}$) or through glass ($N_{\rm gl}$), in the time required to store 100 dark pulses (6-7 min). Usually not more than 12 consecutive measurements of $N_{\rm gl}$ and $N_{\rm qu}$ were made with one portion of 0.5% glycine solution, and for that reason, the necessary statistics were obtained for 15-20 portions, and the results are summarized in Table 1 in accordance with the sequence of measurements.

EXPERIMENTAL RESULTS

The presence of mitogenetic radiation (the UV component in the spectrum of glycine emission) could be judged by the significance of differences between mean values of $N_{\rm qu}$ and $N_{\rm gl}$, estimated by the method of comparison of means, using Student's distribution.

The value of t in Table 1 was calculated by the equation:

$$t = \frac{\overline{N} \operatorname{qu} - \overline{N} \operatorname{gl}}{\sigma \sqrt{\frac{2}{n}}},$$

where o denotes the standard error of measurement and n the number of measurements.

It will be clear from Table 1 that the presence of mitogenetic radiation in the experiments with irradiated glycine and with transfer can be taken as reliably proved. The standard error of the measurements $\sigma \approx 20$, and therefore a sufficiently large number of measurements has to be made. The extensive statistical data obtained by measurements in series II and IV allowed the time course of the radiation to be estimated. For this purpose, the corresponding results were divided into two groups, corresponding to the initial and final phases of the measurements.

The duration of the first group of measurements was 25-30 min, and of the second group 45-60 min. The results of this breakdown are given in Table 2.

During the time indicated above, radiation from the irradiated glycine solution thus continued to increase, whereas after transfer it decreased gently.

These experiments confirm data obtained previously by the use of a biological detector. The intensity of the measured radiation, according to our own estimates, is very low (about 1 quantum/cm²·sec), and in subsequent investigations it will therefore be necessary to increase the efficiency of focusing of this radiation on the light receiver significantly.

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