

Fig. 2. Release of PF₄ during platelet aggregation by adrenaline (aggregation indicated by the arrows). $\circ ---\circ \circ PRP + saline, +---+ PRP + adrenaline <math>10^{-7}M$, $\bullet ---- \bullet PRP + adrenaline <math>10^{-6}M$, $\bullet ---- \bullet PRP + adrenaline <math>10^{-6}M$.

The second possible explanation is that PF₄ may induce paracoagulation of soluble fibrin monomer complexes occurring in the platelet atmosphere. Formation of fibrin threads between platelets may cause their mutual attachment. In support of these data we may quote microscopic observations showing platelets as centres of fibrin thread formation in the hemostatic plug, and Solum's findings⁹ on the induction of platelet aggregation by fibrin oligomers.

Résumé. Nous avons constaté que le facteur plaquettaire 4 est libéré au début de l'aggrégation des plaquettes par l'ADP et l'adrénaline. Ce phénomène a pu être mis en évidence en étudiant l'activité antihéparinique et l'activité paracoagulante (coagulation non enzymatique de complexes solubles de monomères de fibrine) du facteur plaquettaire 4.

S. Niewiarowski, B. Lipiński, R. Farbiszewski and A. Poplawski

Department of Physiological Chemistry, Medical School, Bialystok 8 (Poland), 20 November 1967.

⁹ N. O. Solum, Scand. J. clin. Lab. Invest. 18, 577 (1966).

Photosynthesis and Respiration II. Effect of 3-(3,4-Dichlorophenyl)-1,1-Dimethylurea and of Partial Pressure of Oxygen on the Rates of Carbon Dioxide Exchange in Light and in Darkness of Detached Wheat Leaves

In our previous work¹, we found entirely different effects of various metabolic inhibitors on the rates of CO_2 evolution in light and in darkness. It is known² that CO_2 evolution in light or photorespiration is greatly dependent on the oxygen concentration in ambient air (stimulation) whereas dark respiration was practically unaffected by an oxygen concentration as low as 1%.

The present study was designed to investigate the simultaneous action of 3-(3,4-dichlorophenyl)-1,1-dimethylurea (DCMU) and of partial pressure of oxygen on the rates of CO₂ exchange in light and in darkness of detached wheat leaves. DCMU is an extremely specific inhibitor of photosynthesis which does not interfere with the dark respiration or fermentation. Therefore, using the DCMU, one can expect that after suspension of CO₂ uptake in light, the rates of CO₂ evolution in light or in darkness must be similar if these 2 processes are identical. Moreover, using oxygen as a stimulating factor of CO₂ output in light, we can observe the effect of partial pressure of oxygen on respiration both in light and in darkness in the absence of photosynthesis.

Detached wheat leaves (*Triticum vulgare* Vill. var. Thatcher) taken from 1-month-old plants were used as experimental material. Measuring the carbon dioxide exchange rates both in light and in darkness, the application

of DCMU to the leaves and other details of the methods used in the present study were identical to those described in our previous publication¹.

The Table shows the simultaneous effect of 10^{-6} and $10^{-6}M$ of DCMU and 1 and 100% of oxygen on the rates of CO₂ exchange of wheat leaves. The rates of apparent photosynthesis (APS) in an atmosphere of 100% oxygen were about 80% lower as compared with those in 1% oxygen. The rates of dark respiration in both atmospheres of oxygen were similar. However, the rates of photorespiration (PR) were very low in 1% O₂ and were about 30 times higher in 100% O₂. DCMU in applied concentrations inhibited the rates of apparent photosynthesis in an atmosphere of 1% O₂ by about 90% as compared with those in control, and completely in the pure oxygen. The rates of dark respiration (DR) were practically the same in the presence or absense of DCMU. After inhibition of

¹ G. Poskuta, C. D. Nelson and G. Krotkov, Pl. Physiol., Lancaster 42, 1187 (1967).

M. L. FORRESTER, G. KROTKOV and C. D. Nelson, Pl. Physiol., Lancaster 41, 422 (1966).

³ H. GAFFRON, Plant Physiology, A Treatise (Ed. F. C. STEWARD; Academic Press, New York and London 1959), vol. 1b, p. 222.

apparent photosynthesis by DCMU, the rates of photorespiration were about 60% lower than in the control. The magnitude of this inhibitory effect of DCMU on photorespiration was similar in 1 as in 100% oxygen.

However, the stimulating effect of oxygen on this part of photorespiration which was not inhibited by DCMU was clearly pronounced. The relative values of the stimulation of photorespiration in 100% O₂ were similar during photosynthetic absorption of CO₂ and after suppression of this process by DCMU.

The results presented above have confirmed and extended our previous studies ^{1,4}, showing that also this part of CO₂ evolution in light, which was not inhibited by DCMU in concentrations applied, represents a different

Effect of 3-(3,4-dichlorophenyl)-1,1.dimethylurea (DCMU) and of oxygen concentration on the rates of apparent photosynthesis (APS), dark respiration (DR) and photorespiration (PR) in detached wheat leaves. Light intensity $90\times10^3~{\rm ergs/cm^2\,sec^{-1}}$.

	μ g CO $_2$ /min g fresh weight			
	$\mathcal{O}_2\%$	APS	DR	PR
Control	1	121.8	13.0	1.0
(without DCMU)	100	22.0	14.6	32.4
10 ⁻⁶ <i>M</i> DCMU	1	12.6	12.0	0.3
	100	0.0	15.9	9.9
Control	1	114.6	14.6	0.6
(without DCMU)	100	21.4	15.6	28.3
10 ^{−5} M DCMU	1	9.4	13.4	0.2
	100	0.0	15.8	7.8

process from dark respiration. This conclusion is supported by the fact that, in the absence of photosynthesis, photorespiration was stimulated by oxygen relatively to the same degree as during photosynthesis. Because of the discrepancy in the response of photorespiration and dark respiration to simultaneous action of DCMU and oxygen concentration, these experiments suggest that CO₂ output in light must come from a different source from that in dark respiration. Moreover, it supports the conclusion made earlier 1,2,5 that dark respiration was inhibited in light and replaced by a photorespiration 6,7.

Zusammenfassung. Es werden der Einfluss des Photosynthesehemmstoffs DCMU und des O₂-Partialdrucks auf die Photosynthese und die Licht- und Dunkelatmung untersucht. Die Ergebnisse zeigen erneut, dass Licht- und Dunkelatmung 2 verschiedene Prozesse sind.

J. Poskuta8

Department of Biology, Queen's University, Kingston (Ontario, Canada), 13 November 1967.

- ⁴ G. Poskuta, C. D. Nelson and G. Krotkov, Pl. Physiol. Proc. Ann. Meetings Univ. of Maryland, College Park 37 (1956).
- E. B. TREGUNNA, G. KROTKOV and C. D. Nelson, Physiologia Pl. 19, 723 (1966).
- 6 The financial support for this work came from the Committee on Extra-Mural Research Projects, Department of Forestry, Ottawa, Ontario.
- Many thanks are due to Prof. G. Krotkov and C. D. Nelson for their interest and discussions during this study.
- ⁸ Present address: Department of Plant Physiology, University of Warsaw (Poland).

Histochemistry of Ovarian 20a-Hydroxysteroid Dehydrogenase in Mature Hypophysectomized Rats

The activity of ovarian 20α -hydroxysteroid dehydrogenase (20α -HSD) can be histochemically demonstrated only in the corpora lutea (C.L.) 4–5 days after their onset in normal cycling rats and 24 h before parturition in pregnant animals ^{1–4}.

A possibility of hypophysial control on the appearance of this enzymatic activity has been suggested in previous papers but it has not yet been clarified.

A blocking effect in the newly formed C.L. was obtained by treatments which inhibit gonadotropic incretion, such as the administration of natural or synthetic estrogens and of progestin-estrogen association as well^{5,6}. On the other hand Balogh et al.^{7,8} were able to induce this enzymatic activity in ovarian interstitial and thecal cells of hypophysectomized immature female rats by administration of HGC and in the C.L. of superovulated rat ovaries by administration of luteinizing hormones.

The task of the present work has been to evaluate the appearance of the 20α -HSD activity in the C.L. of ovaries from mature female rats deprived of their pituitaries. For this purpose the hypophysectomies were performed in rats in the different phases of the estrous cycle, i.e. with newly formed C.L. in different stages of maturation, before the onset of the 20α -HSD activity. In these experiments albino Sprague-Dawley female rats were used, weighing 150 g, with regular 4–5 days estrous cycles,

controlled by daily vaginal smears for at least 3 cycles before making the operation. The animals were hypophysectomized by traspharyngeal approach and killed by decapitation 2, 4, 8 and 10 days after the hypophysectomy. The ovaries from single animals were quickly dissected out and frozen with CO_2 . The 3β -hydroxysteroid dehydrogenase (3β -HSD) and the glucose-6-phosphate-dehydrogenase (G-6-PD) were determined at the same time with the 20α -HSD in order to obtain a more com-

- ¹ K. Balogh Jr., J. Histochem. Cytochem. 12, 670 (1964).
- ² E. Turolla, U. Magrini and M. Gaetani, Experientia 22, 675 (1966).
- ³ M. PUPKIN, H. BRATT, J. WEISZ, C. W. LLOYD and K. BALOGH JR., Endocrinology 79, 316 (1966).
- ⁴ E. Turolla, G. Arcari, U. Magrini and M. Gaetani, Second Int. Congr. Horm. Steroids 1966, Commun. No. 468.
- ⁵ E. Turolla, U. Magrini, M. Gaetani and G. Arcari, Experientia 23, 909 (1967).
- ⁶ E. Turolla, M. Gaetani and G. Baldratti, Folia endocr. 20, 526 (1967).
- ⁷ K. BALOGH, W. R. KIDWELL and W. G. WIEST, Endocrinology 78, 75 (1966).
- ⁸ W. R. Kidwell, K. Balogh Jr. and W. G. Wiest, Endocrinology 79, 352 (1966).