

Die präparative Abtrennung der Komponente A aus diesem Gemisch wurde gaschromatographisch in zwei Arbeitsgängen vollzogen. Ein erstes Chromatogramm an einer Kolonne von Emulphor 0 ($T = 130^\circ\text{C}$) ergab ein noch nicht ganz homogenes Produkt. Nach nochmaliger Reinigung an einer Kolonne von Apiezon L ($T = 110^\circ\text{C}$) wurden 60 mg einer gaschromatographisch praktisch einheitlichen Substanz mit den nachfolgend beschriebenen Eigenschaften erhalten.

IR-Spektrum: Banden bei 3480 (s), 3080 (w), 1840 (w), 1647 (w), 1467 (m), 1415 (m), 1370 (s), 1292 (w), 1235 (m), 1190 (Schulter), 1150 (s), 1028 (w), 991 (m), 949 (s), 918 (s), 890 (m), 726 (w), 685 (w) cm^{-1} , praktisch identisch mit dem IR-Spektrum von authentischem 2-Methyl-3-buten-2-ol.

Retentionsindices⁵: $I_{130}^A = 576$; $I_{130}^P = 904$ (Vergleichsprodukt: $I_{130}^A = 576$; $I_{130}^P = 892$). *p*-Nitrobenzoat: schwach hellgelb gefärbte Tafelchen aus Hexan vom Smp. 114 bis 115°C ; Mischprobe mit Vergleichsprodukt (Smp. 115 bis $115,5^\circ\text{C}$) ohne Depression.

$\text{C}_{12}\text{H}_{18}\text{O}_4\text{N}$ Ber. C 61,27 H 5,57% Gef. C 61,08 H 5,65% Das in KBr aufgenommene IR-Spektrum des *p*-Nitrobenzoats war mit jenem des authentischen Vergleichspräparats deckungsgleich.

b) 3-Methyl-2-buten-1-ol (β , β -Dimethyl-allylalkohol) (II) aus Himbeeröl. Ein anlässlich einer eingehenden Untersuchung von Himbeeröl durch Girardierung carbonylfrei erhaltenes Neutralprodukt wurde in der üblichen Weise mit 3,5-Dinitro-benzoylchlorid umgesetzt und das Gemisch der 3,5-Dinitro-benzoate an der 50fachen Menge Kieselgur/Bentonit 1:4 chromatographiert. Mit Chloroform-Essigester (7:3) eluierte eine Fraktion, die sich im Papierchromatogramm (System DMF/Decalin⁶) als ein Gemisch von 3 Komponenten erwies. Mit Hilfe der präparativen Papierchromatographie auf 70 Blättern von je etwa 2 mg Substanz, im System DMF/Cyclohexan⁷ konnte ein einheitliches 3,5-Dinitro-benzoat isoliert werden, welches nach viermaligem Umkristallisieren aus Petroläther einen konstanten Smp. von $69,5\text{--}71^\circ\text{C}$ aufwies.

$\text{C}_{12}\text{H}_{18}\text{O}_6\text{N}_2$ Ber. C 51,43 H 4,32% Gef. C 51,41 H 4,69% Die Mischprobe mit dem 3,5-Dinitro-benzoat von authentischem 3-Methyl-2-buten-1-ol (Smp. 71°C) zeigte keine Depression des Smp. Ebenso erwiesen sich die beiden 3,5-Dinitro-benzoate als identisch auf Grund der IR-Spektren (in KBr) und der R_f -Werte im Papierchromatogramm (System DMF/Decalin). Die Möglichkeit, dass ursprünglich der tertiäre Alkohol I im Naturprodukt vorgelegen und sich dieser während der Aufarbeitung in den primären Alkohol umgelagert hätte, trifft nicht zu, denn ein orientierender Versuch zeigte, dass der tertiäre Alkohol I unter den bei der Girardierung angewandten Bedingungen (Zersetzung mit Essigsäure) nicht in II umgelagert wird (Nachweis über 3,5-Dinitro-benzoat, Smp. 108°C)⁸.

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Summary

2-Methyl-3-buten-2-ol has been isolated from the oil of French lavender and 3-methyl-2-buten-1-ol from the oil of raspberry.

⁵ Vgl. E. KOVATS, Helv. chim. Acta 41, 1915 (1958).

⁶ E. SUNDT und M. WINTER, Analyt. Chem. 29, 851 (1957).

⁷ Ausführung wie mit Decalin. Vergleiche ⁶. Cyclohexan als mobile Phase trennt im allgemeinen etwas schlechter als Decalin, bietet aber Vorteile bei der nachfolgenden Extraktion der Blätter.

⁸ Die Analysen wurden in den mikroanalytischen Laboratorien der ETH (Leitung W. MANSER) und der Firma Firmenich & Cie. (Leitung Dr. E. PALLUY) ausgeführt.

Erythrocyte Glucose-6-Phosphate Dehydrogenase Deficiency and Thalassemia

In family studies on patients suffering from Favism (the clinical syndrome caused by ingestion of the seeds or inhalation of the pollen from the flowers of *Vicia faba*), MARCOLONGO¹ noted that the Thalassemia abnormality was often found amongst the relatives of patients.

It is now well known that the erythrocytes in favism and in drug-induced hemolytic anemia are deficient in the enzyme glucose-6-phosphate dehydrogenase (CARSON, FLANAGAN, ICKES, and ALVING²; SANSONE and SEGNI³; SZEINBERG, SHEBA, and ADAM⁴). The presence of this enzyme may be detected by incubating hemolysates with glucose-6-phosphate and triphosphopyridine nucleotide (Coenzyme II) using the dye brilliant cresyl blue as an indicator of the reaction (DICKENS and GLOCK⁵; MOTULSKY and CAMPBELL⁶).

Using this simple method, blood samples from 7 subjects with Thalassemia major, 16 with Thalassemia minor (parents or relatives of subjects suffering from Thalassemia major or Hemoglobin E – Thalassemia), 6 with Hemoglobin E – Thalassemia and 6 with Hemoglobin-H Disease (Hemoglobin H – Thalassemia, MOTULSKY⁷) have been studied. Absence of the enzyme was found only in 3 of the cases with Hemoglobin-H Disease (all Chinese). In one instance, when it was possible to study the parents of a child with Hemoglobin-H Disease and glucose-6-phosphate dehydrogenase deficiency, the enzyme was deficient in the father and exhibited 'intermediate' activity in the mother, and neither showed Hemoglobin-H Disease. Amongst blood samples containing a variety of abnormal hemoglobins – 5 umbilical cord samples containing the hemoglobin of FESSAS and PAPASPYROU⁸, 1 umbilical cord sample containing hemoglobin 'Alexandra' (FESSAS, MASTROKALOS, and POSTIROPOULOS⁹; VELLA, AGER, and LEHMANN¹⁰), 16 containing hemoglobins A and E, 1 containing hemoglobins A and L, and 2 containing only hemoglobin E – no instance of glucose-6-phosphate dehydrogenase deficiency was found. In one sample (from a Chinese male child hospitalised for hemolytic anemia 15 days after birth) the enzyme deficiency was associated with the presence of the hemoglobin of FESSAS and PAPASPYROU.

¹ F. MARCOLONGO, Minerva Med. (Torino) 44, 1963 (1953); Rec. Progr. Med. int. 15, 137 (1953).

² P. E. CARSON, C. L. FLANAGAN, C. E. ICKES, and A. S. ALVING, Science 124, 484 (1956).

³ G. SANSONE and G. SEGNI, Boll. Soc. ital. Biol. sper. 34, 327 (1958).

⁴ A. SZEINBERG, C. SHEBA, and A. ADAM, Nature 181, 1256 (1958).

⁵ F. DICKENS and G. E. GLOCK, Biochem. J. 50, 81 (1951).

⁶ A. G. MOTULSKY and J. M. CAMPBELL, in F. VELLA, J. Med. Malaya 13, 298 (1959).

⁷ A. G. MOTULSKY, Nature 178, 1055 (1956).

⁸ Ph. FESSAS and A. PAPASPYROU, Science 126, 199 (1957).

⁹ Ph. FESSAS, M. MASTROKALOS, and G. POSTIROPOULOS, Nature 183, 30 (1959).

¹⁰ F. VELLA, J. A. M. AGER, and H. LEHMANN, Nature 183, 31 (1959).

It is not unlikely that the association of the factors for Thalassemia and for absence of glucose-6-phosphate dehydrogenase activity in erythrocytes presumably present in the families studied by MARCOLONGO was fortuitous, since both Thalassemia and Favism are common findings in Sardinia. The presence of this defect in 3 out of the 6 cases of Hemoglobin-H disease studied is interesting, particularly in view of the finding that absence of glucose-6-phosphate dehydrogenase activity was found in 2.22% of 225 healthy Chinese male blood donors and in 2.63% of 76 Chinese male new-borns studied (VELLA). Absence of this enzyme in some cases of Hemoglobin-H disease may explain the severity of hemolysis occasionally seen in some patients.

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Résumé

L'activité de la glucose-6-phosphate déhydrogénase était normale dans les érythrocytes obtenus de sujets atteints de Thalassémie (majeure ou mineure) d'Hémoglobine E – Thalassémie et dans des érythrocytes contenant une variété d'hémoglobines anormales. Chez trois des six sujets atteints d'Hémoglobine H – Thalassémie, la glucose-6-phosphate déhydrogénase était absente.

Effect of Testosterone on the Nuclear Volume of Exorbital Lacrimal Glands of the White Rat (*Glandula lacrimalis praeparotidea*)

Recent studies have demonstrated that the exorbital lacrimal glands (Loewenthal's glands) of the white rat present a sexual dimorphism and that their morphology undergoes modifications after castration and/or administration of male sex hormones (GABE¹; BAQUICHE^{2,3}; DUCOMMUN *et al.*⁴). Similar observations were made by others, who have however erroneously considered this glands as a parotid gland, or as a part of it, owing to the close topographical relationships between these two structures (PARHON *et al.*⁵; CAVALLERO and PINI⁶).

Caryometric studies on exorbital lacrimal glands of the rat have been published by COLLIN and FLORENTIN⁷, COLLIN and GRUJIC⁸ and TEIR⁹. However, in these researches the attempt was made to determine the variability of the nuclear volumes and their distribution in normal conditions. Conversely, it has been our aim to establish whether castration and/or administration of testosterone propionate in the male rat is capable of inducing changes in the nuclear volumes of Loewenthal's glands, in order to give further support to the suggested rôle of these glands as a target organ of male sex hormones.

Twenty adult male white rats of the same strain, about two months old and weighing 150–200 g were employed; they were fed a standard diet and had free access to water.

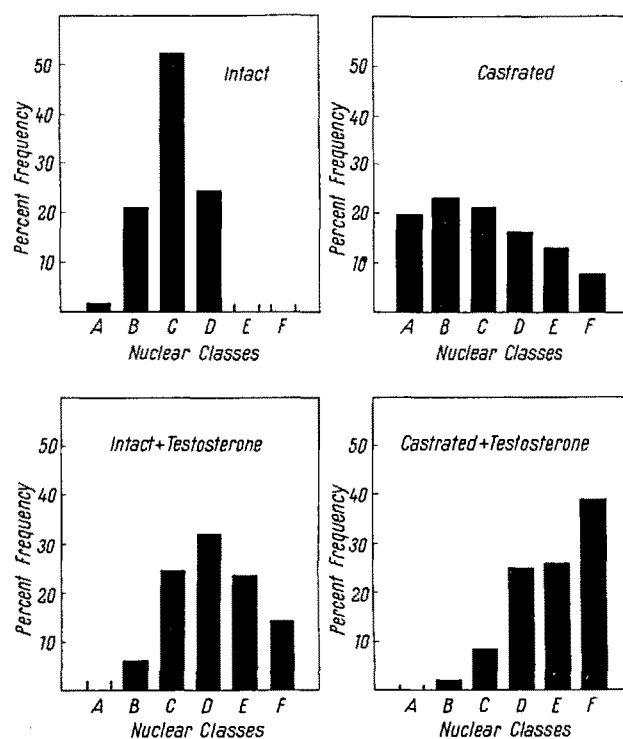
Two experimental groups of animals were considered: the first one was composed of intact rats, with and without testosterone treatment; in the second set, castrated rats, untreated and treated with the hormone, were examined, the orchietomy having been performed 60 days before the beginning of the experiment. Testosterone propionate was given subcutaneously, dissolved in Tween 80, 1 mg daily for 20 days.

At the end of the experimental period the animals were killed by decapitation; the exorbital lacrimal glands were removed, fixed in Bouin's fluid, and embedded in paraffin. Nuclear volumes were measured in 10 μ sections, stained with hematoxylin-eosin. For the measurements, only regular nuclei were selected, 300 nuclei for each gland, and two perpendicular diameters measured at 2000 magnification. The volume V of the nucleus was calculated by means of the formula:

$$V = \pi/6 [D_1 + D_2]^3/2,$$

where D_1 and D_2 are the two nuclear diameters considered. The values were grouped in to six classes, starting from a volume of less than 120 μ^3 up to a volume of more than 700 μ^3 , and the relative percent frequency was calculated. The following nuclear classes were established: $A = < 120 \mu^3$; $B = 121-199 \mu^3$; $C = 200-299 \mu^3$; $D = 300$ to $449 \mu^3$; $E = 450-699 \mu^3$; $F = > 700 \mu^3$.

The results of the experiments are shown by histograms. Figure 1 refers to intact, untreated animals; it appears that normally the highest incidence of the nuclear volumes falls in the class between 200 and 299 μ^3 (mean nuclear volume 279 μ^3). Untreated rats, castrated two months earlier, present a wide dispersion of nuclear volumes, as shown by Figure 2, the highest incidence being at a lower level than in intact animals (mean nuclear volume 320 μ^3). Figure 3 and 4 refer to testosterone-treated rats; they



¹ M. GABE, C. R. Soc. Biol., Paris 149, 223 (1955).

² M. BAQUICHE, Thèse Univ. Genève 5, 1 (1958).

³ M. BAQUICHE, Acta anat. 36, 247 (1959).

⁴ P. DUCOMMUN *et al.*, Acta endocrinol. 30, 78 (1959).

⁵ C. I. PARHON *et al.*, Bul. Sci. Seš. St. med. 7, 3 (1955).

⁶ C. CAVALLERO and C. E. PINI, Atti Soc. lomb. Sci. med. biol. 13, 196 (1958).

⁷ R. COLLIN and P. FLORENTIN, C. R. Soc. Biol., Paris 104, 1279 (1930).

⁸ R. COLLIN and M. GRUJIC, C. R. Soc. Biol., Paris 109, 472 (1932).

⁹ H. TEIR, Acta path. microbiol. scand., Suppl. 58 (1944).