In the present experiment ^{3}H-pregnenolone along with the acetyl-CoA was added to the incubation medium. Pregnenolone is generally considered to be a necessary intermediate in the conversion of acetate via cholesterol to progesterone. Pregnenolone however had a ^{3}H/^{14}C ratio higher than progesterone. This may indicate that during the incubation period only part of extracellular ^{3}H-pregnenolone had reached the steroid converting enzymes. By the extraction procedure intracellular pregnenolone was mixed with extracellular ^{3}H-pregnenolone. But progesterone had a ^{3}H/^{14}C ratio higher than DHEA. If both metabolites were derived from the same precursor – intracellular pregnenolone – they should have exhibited the same ^{3}H/^{14} ratio. The higher ^{14}C content of DHEA suggests ^{14}C-precursors other than pregnenolone for DHEA. A direct conversion of cholesterol into DHEA via 17,20α-dihydroxy-cholesterol[7] and via 17α-hydroxy-pregnenolone[8-10] has been demonstrated by other investigators[11].

Zusammenfassung. Nach In-vitro-Inkubation vǫn Nebennierenschnitten eines Rhesusaffen mit 1-^{14}C-acetyl-CoA und 7α-^{3}H-Pregnenolon konnte man Pregnenolon, DHEA und Progesteron isolieren, die alle sowohl ^{3}H- als auch ^{14}C-Aktivität enthielten. Es ergaben sich Hinweise für verschiedene Biosynthesewege für Pregnenolon und DHEA.

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Some Effects of Laser upon the Bones

The biological effects of laser upon various body tissues are studied with increasing interest, because of the broader use of laser in clinical practice. Up to the present time, little attention was paid to the influences of laser pulsations on bones. In the scanty reports available, some such observations are mentioned. Perforations or excavations of bones are understandable after transmissions of high-power laser pulses[1-3], but according to some reports, also much lower intensities (10–50 J) were sufficient to cause char of bone periosteum with necrotic areas of bone tissues beneath it[4]. But a surprisingly normal macroscopic appearance of bones was reported even after higher intensities of laser radiations[5,6]. Some initial degrees of damage can hardly be found in the bones macroscopically and sometimes not even microscopically. When investigating the influences of various other physical agents on the skeleton[7], we noticed the usefulness of isotopic methods with ^{45}Ca as a tracer, in detecting unconspicuous aberrations in the bone tissue. Therefore, we tried to investigate the influence of laser pulses on the bones in this manner.

Male Wistar rats, weighing 120–130 g, were subjected to 3 laser pulses from a CO_2-laser, working in the IR-spectral area with $\lambda = 10.6$ µm. The 4 mm broad beam reached the area of the knee of the right extremity in expositions, lasting 3 sec each, with 3 sec long intervals. The whole amount of energy applied to the skin surface was estimated to be 9 J. Non-irradiated controls and irradiated animals were kept under the same conditions. After 4, 11, 18, 33, 46, 60, 74 and 102 days post irradiation, groups of 5 animals from each category were formed (i.e. irradiated and non-irradiated). Each animal was given 20 µCi $^{45}CaCl_2$ in 1.0 ml saline i.p. After 48 h the animals were killed by ether narcosis and both tibiae were liberated from surrounding tissues. The bones were investigated chemically for the content of calcium, and dosimetrically for their content of ^{45}Ca in the same manner as reported previously[7,8]. The 48-h uptake was expressed as average of all 5 homologous bone samples.

Although there was no significant difference in the weight of dried bone substance of the tibia (Figure 1) in irradiated and non-irradiated animals, the uptake of radioactive calcium was different (Figure 2). During the first 3 weeks, there were less noticeable differences in this uptake; after this time distinct deviations of the tracer uptake started in the irradiated as well as the non-irradiated legs of experimental animals. The appearance of the irradiated skin and the gross appearance of the bones during preparation were normal. The Table gives a summary of statistical significances (*t*-test) of values, expressed in Figure 2.

Like various other physical agents (e.g. X-rays, electric current, heat, cold, ultrasound)[7], laser radiations caused disturbances not only in the calcium metabolism in directly irradiated bones, but even in the opposite legs. This was true in spite of the fact that the skin burns healed without complications during the first week after irradia-

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Tibia	Days 4	11	18	33	46	60	74	102
Irradiated: control	> 0.05	> 0.05	> 0.05	< 0.05	> 0.05	< 0.05	< 0.05	> 0.05
Opposite: control	> 0.05	> 0.05	> 0.05	< 0.05	< 0.05	< 0.01	< 0.05	> 0.05

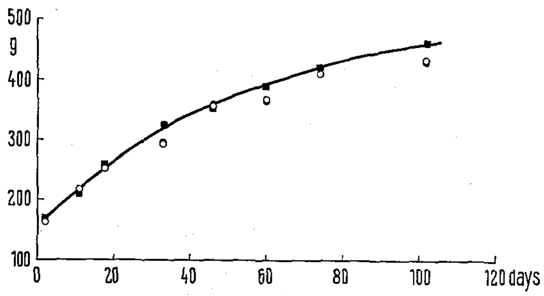

Fig. 1. Weights of dried bone substances from the whole tibia in healthy controls (■), in irradiated legs (○) and in opposite legs (●) of irradiated animals.

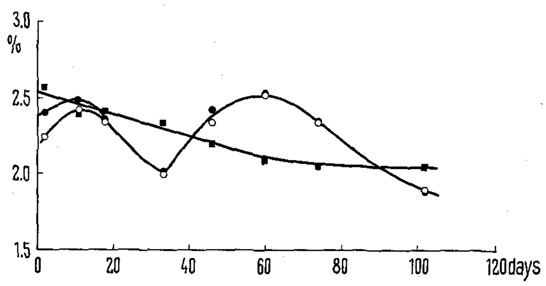

Fig. 2. The 48 h ^{45}Ca-uptake in the whole tibia expressed in per cent of the ^{45}Ca-dosis, given to the animals.

tion and that there were no gross macroscopic pathological findings in the bones. The onset of this metabolic aberration was relatively late.

From this experiment some evidence was gained about metabolic mineral disturbances in the bones, caused by even such small intensities of laser pulses, as used here. With respect to increasing uses of laser in various therapeutic interventions, the possible harmful effects on the bones are not neglectible and some further investigations about the permissible levels of energies used in clinical practice are justified.

Zusammenfassung. Bei männlichen Wistarratten wurden nach 3 Laserpulsen (Energie 9 J) deutliche metabolische Abweichungen vom 48-h-^{45}Ca-Empfang in den Knochen festgestellt, die einige Monate andauerten. Das makroskopische Aussehen der Knochen war dabei normal. Eine Bestimmung der zulässigen Energien der Laserstrahlen scheint für die klinische Praxis notwendig zu sein.

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The Influence of Sodium Salicylate on the Formation of Inorganic Phosphate in Human and Rabbit Erythrocytes in vitro

The rate of erythrocyte glycolysis is markedly influenced by the composition of the suspending medium[1]. Furthermore, it has been shown[2] that great differences occur in the rate of glucose consumption and lactate production of aliquots of the same erythrocyte sample suspended in different media. The rate of glycolysis of erythrocytes suspended in $0.1 M$ potassium phosphate buffer pH 7.4 containing $0.01 M$ glucose was 2–3 times that of cells suspended in Tyrode-Locke's solution containing $0.01 M$ glucose. Inorganic phosphate is an important factor in regulating the rate of erythrocyte glycolysis[3–6] and by increasing its concentration in the incubation medium corresponding increases were seen in erythrocyte glucose consumption and lactate production[7].

Sodium salicylate in concentrations ranging from 1 to $5 \times 10^{-3} M$ increased glucose consumption and lactate production by both human and rabbit erythrocytes washed and suspended in Tyrode-Locke's solution containing $0.01 M$ glucose[2]. The present investigation concerns the effect of sodium salicylate on inorganic phosphate formation in erythrocytes.

Human venous blood was obtained by venipuncture of the antecubital vein while rabbit blood was withdrawn

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