The weight of the animals remained approximately at a constant level during the whole experiment.

Discussion. The most striking outcome of these experiments was the delay of 12 days of the time of healing in a second wound produced 6-7 days after the first one (Exp. 1). This interval is judged in comparison with the time of healing of the first wound. Compared to the 2 control groups, the interval would be even greater. This, however, is due to a fact I cannot explain, namely that the healing times of the first wounds in experiments 1-4 were consistently later, by about 4 days, than those of the controls.

It seems to follow from the experiments described, that an inhibiting effect appears during a limited period of the wound healing process, which has its influence on another healing process in the same organism. This effect may indicate the existence of an inhibiting factor appearing during this period. There is normally an inhibition during the last 7 days before closure in the first wound. In Exp. 1 a further inhibition in the second wound was observed which occurred within 1-3 days after the former. This inhibition in the second wound occurred much earlier in the wound healing process than usual, making its appearance during the phase of active proliferation of the granulation-tissue.

This inhibitory influence was not present or was too weak to become clearly visible in the Exp. 2-4, where the inhibiting period of the first wound coincided with an earlier and more active stage of the proliferation of the granulation-tissue of the second wound or with the stage immediately after the production of the second wound.

Summary. 1. Experiments were carried out in which the last phase of the healing process of a primary wound was made to coincide either with the phase of active proliferation of the granulation-tissue in a second wound in the same animal or with the stage of the very beginning of this proliferation. 2. The results obtained show the appearance of an inhibiting effect during a limited period of normal wound healing. This finding may suggest the existence of an inhibiting factor.

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Effect of Age upon Hepatic Synthesis of Cholesterol in Rat.* (19855)

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Although *in vitro* studies have been done on the effect of age on the hepatic synthesis of cholesterol in the rat, no similar study has been done on intact rats. Recently, however, a means was found(1) by which the hepatic synthesis of cholesterol might be gauged in the living unanesthetized rat. It consisted of collection of the daily bile output of a rat and determination of its cholesterol concentration. Other studies(2-4) have confirmed the validity of this technic, in that an increase or decrease of the biliary cholesterol concentration parallels an induced respective increase or decrease in the hepatic synthesis of cholesterol as determined by tracer studies. Accordingly, groups of rats at various ages were subjected to a study in which bile was collected and analyzed for its cholesterol concentration.

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TABLE I. Effect of Age on Bile Cholesterol Concentration.

Age	No. of rats	Wt (g)	Plasma cholesterol conc. (mg/100 ml)	Bile		
				Daily vol (ml)	Cholesterol	
					Conc. (mg/100 ml)	Daily output (mg)
6 wk	9	116 (94-138)*	64.8 (54-88)	7.1 (5.3- 8.9)	23.6 (18.3-31.9) S.E.$_m$: ±1.5 †	1.7 (1 -2.4)
8	6	181 (161-209)	43 (32-53)	11.1 (9 -14)	26.6 (21.6-36) S.E.$_m$: ±2	2.9 (1.9-3.8)
9	12	184 (160-236)	47.7 (40-71)	12 (7.5-14.5)	24.1 (17.6-30) S.E.$_m$: ±5.2	3 (1.8-5.1)
5 mo	12	310 (253-370)	64 (52-82)	14.5 (11 -21)	16.8 (13 -20) S.E.$_m$: ±.23	2.4 (1.7-3.4)
7	10	373 (315-443)	47 (38-51)	15.7 (13.5-23)	12.8 (10.5-15.5) S.E.$_m$: ±.48	2 (1.7-2.8)
11	10	406 (319-497)	46.5 (31-59)	15.3 (11 -24.5)	12.5 (9.7-16.5) S.E.$_m$: ±.7	1.9 (1.5-2.4)

* Figures in parentheses refer to range.
† S.E.$_m$ refers to stand. error of the mean.

Methods. Male rats (Long-Evans strain) from the same colony were fed a stock laboratory ration. Groups of rats were subjected to bile duct cannulation as they attained the various ages indicated in Table I. During the ensuing 24-hour interval in which food was withheld, bile was collected and analyzed for its content of cholesterol according to technics described previously(5). In each instance the plasma cholesterol concentration was determined preoperatively(6).

Results. As shown in Table I, the bile volume output was found to increase somewhat in older rats in association with increased size and weight. Thus, the average bile volume of the immature rats (7.1-11.1 cc/24 hr) was consistently less than that obtained from the mature animals (14.5-15.7 cc/24 hr). Study of the range of values of the plasma cholesterol levels obtained in this investigation, and comparison with those obtained in other experiments performed in this laboratory fails to indicate that age had any significant effect in this respect. On the other hand, the biliary cholesterol concentration at various ages showed significant differences. Much higher average and range of values of biliary cholesterol concentration were found in the younger rats. After the age of 9 weeks, these values consistently were lower than those obtained in the immature rats. Thus, the average biliary concentration of cholesterol in the immature rats varied from 23.6 to 26.6 mg per 100 cc and, in the mature animals varied from 12.5 to 16.8 mg per 100 cc. The daily output of cholesterol in the bile, on the other hand, also is a function of the bile volume. Therefore, the daily output of biliary cholesterol was lower in the youngest group of animals despite the higher concentration of cholesterol.

Discussion. It has been observed in animals that hepatic synthesis of cholesterol depends upon the age and weight of the animal (7). Thus, in studies of cholesterol synthesis conducted in liver slices, an increased rate of synthesis was found in livers obtained from younger animals(8). The results obtained in the present investigation agree with this observation. Thus an increased hepatic synthesis of cholesterol, as reflected by a considerably greater biliary cholesterol *concentration* (1), consistently was noted in those animals in the prepubertal age groups. This was found to decrease with the advent of puberty. The study of cholesterol synthesis using the technic described above, in its agreement with the results obtained from studies employing liver slices(8), lends further proof of the validity of the biliary cholesterol concentrations as an index of hepatic synthesis of cholesterol(1).

Summary. The hepatic synthesis of cholesterol has been studied in the rat at different ages, using a biliary concentration of cholesterol as an index of this factor. The results suggest that the hepatic synthesis of choles-

terol depends upon the age of the animal, with a considerably greater synthesis in the younger rat.

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Metabolism and Urinary Excretion of Several Flavonoid Compounds.* (19856)

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There is considerable disagreement among investigators regarding metabolism and urinary excretion of flavonoid compounds. Fukuda(1) claimed that orally administered rutin appeared unchanged in the urine of rabbits within 20 hours after administration. Garino (2) made similar claims for the dog, but Field and Rekers(3) could not confirm his findings. Porter *et al.*(4) found only traces of rutin in the urine of humans given oral doses of 60 to 2250 mg daily. Clark and MacKay(5) confirmed these findings in humans and also in the rat. Furthermore, they showed that rutin had little effect on urinary total phenol and glucuronide excretion in the rat. It was also pointed out that intestinal destruction of orally administered flavonoids made it improbable that very much of the administered dose could be absorbed and excreted in the urine. Kirtley and Peck(6) reported that hesperidin methyl chalcone was not excreted in the urine in the free form after large oral doses in man. From the above evidence it appears that the oral route of administration results in almost complete destruction of the flavonoid compounds before absorption can take place. However, it is possible that if the compounds were administered by another route, information could be derived concerning the manner in which the body metabolizes and excretes such compounds.

Experimental. Male CFW strain rats weighing an average of 294 g were caged separately in Norwich type metabolism cages. Six animals were used for each compound. Twenty-four hour urine samples were collected daily for 2 days prior to and 5 days after the subcutaneous injection of 100 mg/rat of the following compounds: rutin, hesperidin, hesperidin methyl chalcone, naringin and phloroacetophenone. Chemical analyses were made for the following: free rutin by the aluminum complex method of Porter, *et al.*(4), free hesperidin and naringin by the cyanidin test (5); free hesperidin methyl chalcone by borocitrate reaction(7); free phenol by O'Leary's method(8); ethereal sulfate by the method of Treon and Crutchfield(9); and glucuronide by Dische's method(10).

Results. The results obtained in the anlyses are given in Table I, in which the excretion as degradation products and conjugates is based upon differences between pre- and post-injection values. These are average excretion figures and represent only the first 24 hours

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