system is involved in regulation of fat transport due to release of norepinephrine at nerve endings in adipose tissue.

Summary. Concentrations of nonesterified fatty acids and of glucose in plasma were determined before and after infusion of epinephrine and norepinephrine into dogs. At the same concentration and rate of infusion, norepinephrine produced a greater increase in nonesterified fatty acids.

ADDENDUM: Since completion of this manuscript, we found that Laurell, S., Christensson, B., *Acta Physiol. Scand.*, 1958, v44, 248 reported increased plasma NEFA in human subjects following norepinephrine injection.

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Periarteritis in Rats Given Single Injection of 4′-Fluoro-10-methyl-1,2-benzanthracene.* (25040)

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Periarteritis nodosa has been recognized in patients for almost a century(1), and an etiology of hypersensitivity has been invoked (2-4). Thus, the disease sometimes appears to follow administration of foreign sera(4,5), and similar lesions can be induced in experimental animals by foreign proteins(6-11). A number of clinical cases of periarteritis nodosa have been ascribed to hypersensitivity reaction to various drugs (i.e., sulfonamides(12) and iodides(13)). Similar vascular lesions have been induced in experimental animals by a few compounds of low molecular weight. Periarteritis was observed in rats and monkeys stressed by unilateral nephrectomy and high salt diet and then given 9-fluoro- or 9-chloro-2-methylcortisol(14-16). Anemic infarction of lungs was found in rats treated with certain halogenated hydrocarbons such as hexachlorotetrafluorobutane(17). In a study on carcinogenicity of various fluorinated derivatives of 10-methyl-1,2-benzanthracene (10-Me-BA)† all rats injected subcutaneously with 2.14 mg of 4′-fluoro-10-Me-BA died within 8 weeks. A marked degree of vascular changes as found in lungs and kidneys, and pulmonary lesions appeared to cause death. Development of these lesions was followed in a second series of rats, and data for the latter experiment form the subject of this report.

Materials and Methods. Male adult albino rats of Holtzman stock‡ with average initial weight of 250 g were maintained in screen-bottom cages in groups of 2 to 4 and were fed Purina Laboratory Chow *ad lib.* Each rat received a single subcutaneous injection of 0.2 ml of tricaprylin (previously dissolved in petroleum ether and extracted with alkali to remove traces of free acid) or 0.2 ml of tri-

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‡ Holtzman Rat Co., Madison, Wis.

TABLE I. Number of Rats with Periarteritis at Various Times.

Weeks after inj.	Hydrocarbon inj.* None	10-Me-BA, 2 mg	4′-Fluoro-10-Me-BA 2.14 mg	4′-Fluoro-10-Me-BA 1.07 mg
0	0/3†			
1		0/2	0/3	
2	0/2	"	1/4	0/2
3		"	0/3	
4	"	"	4/7	"
5		"	4/5	
6	"	"	2/2	1/2
7		"	"	
8	0/3	0/4		2/4
Total	0/12	0/18	13/26	3/10

* Tricaprylin served as the solvent for subcut. inj. of the hydrocarbons and was also administered to control rats.

† Numerator denotes No. of rats with periarteritis; denominator, No. of rats autopsied.

caprylin containing 2 mg of 10-Me-BA, 2.14 mg of 4′-fluoro-10-Me-BA (equimolar to the 10-Me-BA), or 1.07 mg of 4′-fluoro-10-Me-BA. At weekly intervals for 8 weeks the rats were weighed and representative animals (2-4/group) were killed (Table I). Autopsies were performed and pieces of major tissues, except brain, were fixed in 10% formalin. The sections were cut at 6 μ and stained with hematoxylin-eosin, periodic-acid-Schiff (PAS) reagent(18), and Kingsley stain (19). On microscopic examination the vascular lesions in the organs were graded from 1+ to 4+. Grade 1+ designates very mild lesions with little or no perivascular infiltration. Grade 4+ designates pronounced periarteritis which involves all 3 layers of the vessels and is associated with necrosis and thickening of vessel walls. Grades 2+ and 3+ are of intermediate severity. Only lesions of 2+ or greater severity were considered of diagnostic significance.

Results. The general health of control rats and those injected with 10-Me-BA was good; these rats gained an average of 130 g during the 8-week experimental period. On the other hand, rats injected with 2.14 mg of 4′-fluoro-10-Me-BA gained 26 g during the first 2 weeks and thereafter lost weight slowly. From the fifth to seventh weeks surviving rats averaged 5-10 g below their starting weights. Weights of rats injected with the lower dose of 4′-fluoro derivative were similar to those of control rats until the fifth week; thereafter, their weights remained essentially constant. Rats killed for autopsy 4 to 7 weeks after injection of 2.14 mg of 4′-fluoro-10-Me-BA were generally in poor health; some were cyanotic and appeared to be near death.

The major pathological lesion was periarteritis (Table I) which involved particularly the vessels of lungs and kidneys, but occasionally also, to a lesser extent, the periadrenal and pancreatic arterioles. In the lungs lesions were most pronounced in arteries whereas in kidneys arterioles and capillaries were involved. The lesions were first observed in lungs of a rat killed 2 weeks after injection of higher dose of 4′-fluoro-10-Me-BA, but they were sporadic and poorly developed. By the fourth week 4 of 7 animals examined had 3-4+ lesions in pulmonary and/or renal vessels, and all but one of the animals killed thereafter had advanced lesions. Of the animals injected with 1.07 mg of 4′-fluoro-10-Me-BA 3 of 6 rats examined at 6-8 weeks had these lesions. No vascular lesions were observed in any rats which received injections of tricaprylin alone or tricaprylin containing 2 mg of 10-Me-BA.

Lungs. In the larger arteries there was a marked thickening of all 3 layers (Fig. 1). The intima was characterized by endothelial proliferation which tended to occlude the lumen. The media was edematous and infiltrated by numerous inflammatory cells, of which the majority were neutrophilic, polymorphonuclear leukocytes and fibroblasts. Eosinophilic leukocytes were very scarce, even in sections stained with the Kingsley preparation. Nuclear debris and pyknotic and karyorrhectic nuclei were frequently scattered among the leukocytes. While the internal elastic membrane was fairly distinct, there was no sharp distinction between media and adventitia. Similarly, the outer layer of adventitia blended in with the inflammatory infiltration of adjacent lung tissue. Necrosis was rare and when it occurred it involved only a segment of the arterial wall.

The PAS reaction stained the intima more strongly than other layers. It outlined cell membranes clearly, but did not indicate an excessive deposition of polysaccharides.

Kidneys. Lesions in kidneys selectively involved arterioles and extended into the glom-

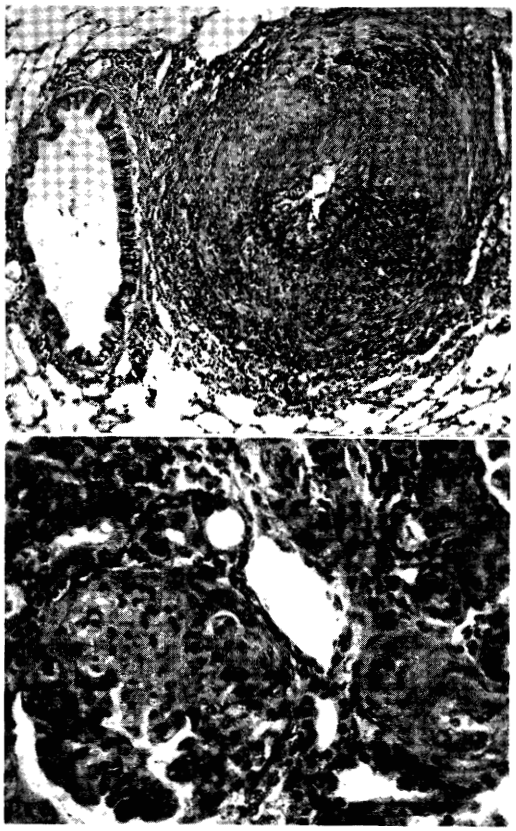

FIG. 1 (top). Periarteritis of a pulmonary artery. Endothelial proliferation, as well as inflammatory infiltration and fibrosis of the media and adventitia are evident. Grade 3+ lesion. (100×, PAS stain.)

FIG. 2 (bottom). Hyaline necrosis of kidney arterioles and capillaries. Afferent arterioles at the right are almost occluded. Upper half of the glomerulus at left shows obliteration of the capillaries. Grade 3+ involvement. (100×, PAS stain.)

erular capillaries (Fig. 2). Arterioles, particularly the afferent arterioles, exhibited hyaline necrosis which tended to obstruct the lumen and extended into the glomerular capillaries, where the degree of involvement varied from only a few loops to the entire glomerulus. In some cases glomeruli were swollen and adherent to outer layer of Bowman's capsule. These lesions are not specific for periarteritis since they have been described in malignant nephrosclerosis and glomerulonephritis as well(20). Fundamental similarities in the pathogenesis of glomerulonephritis and periarteritis have been suggested by Masugi and Sato(6), Rich(4) and others who have stressed their frequent concurrence. We believe that in this case association of glomerular changes with a more typical lesion in the lungs favors a diagnosis of periarteritis.

Renal lesions were most pronounced in rats killed in fourth and fifth weeks and were absent from those killed at 7 and 8 weeks. This observation suggests that the lesion may be reversible. In this connection it is of interest that Hawn and Janeway(21) observed regression of the vascular lesions induced in rabbits by administration of foreign proteins; regression of lesions was correlated with a decreased level of circulating antigen.

Discussion. The etiology of severe and fatal periarteritis which occurs in rats given a single injection of 4′-fluoro-10-Me-BA is not known. Similar lesions were not seen in rats given equimolar doses of the parent hydrocarbon 10-Me-BA, and 3-fluoro-10-Me-BA was also non-toxic under same conditions.§ The stability of C-F bonds of this type and the lack of reports of a similar lesion in fluoride-treated rats suggests that the fluorine atom itself is not involved. Hypersensitivity has been shown to be an etiological factor in periarteritis nodosa, and the possibility that an immunological response may occur in rats given 4′-fluoro-10-Me-BA should also be considered. Williams(22) reported that a human volunteer treated topically at one site with 9,10-dimethyl-1,2-benzanthracene and then, 1 and 2 months later, treated similarly at other sites showed, subsequent to second and third applications, a papulo-vesicular response both at test site and in areas previously painted with the hydrocarbon. No response occurred after first application. Various polycyclic aromatic hydrocarbons(23-26) have been shown to combine with certain tissue proteins of the mouse *in vivo*. If 4′-fluoro-10-Me-BA likewise combines with one or more proteins *in vivo* and should yield an antigenically foreign protein, autoimmunization would result. Such an autoimmunization against altered tissue proteins has been suggested by Green(27) as the basic factor in genesis of cancer by polycyclic hydrocarbons and other chemicals.

Summary. 1. A single subcutaneous injection of 2.14 mg of 4′-fluoro-10-methyl-1,2-benzanthracene in albino rats produced a se-

§ Miller, J. A., Miller, E. C., unpublished data.

vere and fatal periarteritis in 4 to 8 weeks. Similar lesions were seen after injection of one-half as much of this hydrocarbon, but they developed more slowly. Equivalent levels of 10-methyl-1,2-benzanthracene or the solvent alone were ineffective. 2. The most severe lesions were observed in pulmonary arteries; this finding is in accord with the cyanotic conditions of some rats. Hyaline necrotic lesions were observed in renal arterioles and glomerular capillaries. 3. A possible immunological basis for these lesions is discussed.

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Comparative Anti-Inflammatory Efficacy of Topically Applied Steroids on Human Skin. (25041)

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Hundreds of hydrocortisone analogs have been synthesized in the search for more ideal anti-inflammatory agents. It is desirable to evaluate these newer compounds in animals and humans as quickly as possible. This paper describes results obtained using a modification of an unpublished assay method recommended by Brunner and Finkelstein (Research Laboratories, The Toni Co., Chicago). The original method will be reported by these authors.

Method. Immediately prior to testing, 7 accurately weighed steroid samples are separately dissolved in 15 ml tetrahydrofurfuryl alcohol (THFA).* This irritant alcohol serves both as solvent for test steroids and as agent for producing experimental erythema. Potencies of the steroids are determined from their ability, when applied simultaneously with the irritant, to inhibit THFA-induced contact dermatitis. THFA alone is used as control. Twenty-four white male volunteers at least 21 years of age are used for each test. One by 3″ Elastoplast† bandages are used to

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† Duke Labs., South Norwalk, Conn.