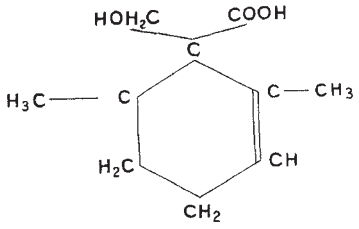
It crystallizes from water as colourless plates or needles (dimorphism) of m.p. 160°. It is slightly soluble in water, readily soluble in ether, alcohol and acetone. It is strongly lævorotatory ($[\alpha]_D = -100°$ in methanol). Oleuropeic acid has the composition $C_{10}H_{16}O_3$. Titration with alkali proves the presence of one carboxyl group per mole and the addition of bromine and the reaction with potassium permanganate proves the presence of a double bond. The infra-red absorption spectrum shows the absorption bands for a hydroxyl group, a carboxyl group and a double bond. Catalytic hydrogenation abolishes the optical activity. $C_{10}H_{18}O_3$, does not show the absorption band for a double bond in infra-red.

Treatment of the oleuropeic acid with 6 *N* sulphuric acid also abolishes the optical activity by eliminating methanol and causing the disappearance of the absorption band of the hydroxyl in infra-red.

Aromatization of the oleuropeic acid with sulphur also eliminates methanol, abolishes the absorption band for hydroxyl and produces in a good yield 2,6-dimethyl benzoic acid (xylic acid). Ozonization of the double bond of oleuropeic acid produces a compound which gives a positive test with sodium nitroprusside (—$CO.CH_3$ group) and reduces Fehling's solution (—CHO group), thus demonstrating the vicinity of the double bond to a methyl group.

These findings clearly established the structure of oleuropeic acid as :



BARUCH SHASHA
JESHAIA LEIBOWITZ
Department of Biological Chemistry,
Hebrew University,
Jerusalem.

[1] Baruch Shasha and Jeshaia Leibowitz, *Bull. Res. Coun. Israel*, **8**, A (1959).

ANIMAL PHYSIOLOGY

Insulin and Hypersensitivity

IT was reported in 1957[1] that a single administration of insulin renders the rat more sensitive to the inflammation produced by injections of egg white or dextran. The mechanism by which insulin exerts this effect is not known, and we have now studied this problem.

In the first experiments, it was confirmed that intraperitoneal injections of soluble insulin (4–12 units/kgm.) given 15 min. before egg white (24 ml./kgm.) or dextran (300 mgm./kgm.) aggravated the anaphylactoid reactions; its speed of onset was increased, and the reaction which developed was so severe that many rats died from intestinal hæmorrhage. Moreover, doses of the inflammatory substances which were non-effective *per se* became effective in the presence of insulin. For example, an intravenous or intraperitoneal dose of 30 mgm./kgm. dextran produced no œdema of the extremities nor blueing of the feet of the rat which had received Evans blue intravenously, yet in the presence of insulin the reaction was intense. Similarly a dose of 2·5 μgm./kgm. dextran injected subcutaneously into the dorsum of one foot produced extensive œdema in the other foot, ears and nose, only when insulin had been previously injected subcutaneously.

Next, the course of anaphylaxis was found to be greatly modified by insulin. Rats were sensitized to egg white[2] and 15 min. before the challenge with the antigen insulin was injected subcutaneously. The shock value of the rats was greatly increased and many animals died. On the other hand, shock was minimal when the insulin was given before the sensitization of the rats. Insulin therefore does not affect the formation of antibodies but renders the animal more sensitive to the antigen—antibody reaction or to the products of this reaction. Pretreatment of the animal with insulin however did not alter the Dale-Schultz reaction of the isolated colon or uterus, and there was only a slight increase in the toxicity of histamine or 5-hydroxytryptamine.

In further experiments, the effect of insulin on the production of œdema in the paw of the rat by the local administration of several agents[3] was investigated. Œdema was measured visually and by a water-displacement method, whilst blueing was studied in rats which had received Evans blue intravenously. Pretreatment of the rat with insulin however failed to modify the response after local injections of histamine (100 μgm.), 5-hydroxytryptamine (0·5 μgm.), polymyxin *B* (1 μgm.) or compound 48/80 (1 μgm.).

The potentiating effect of insulin on the anaphylactoid reaction and on anaphylaxis may be similar to that of thyroxine or *Haemophilus pertussis* vaccine, or when the animal is adrenalectomized or hypophysectomized. Hypoglycæmia is a factor common to these different procedures and may be accelerating the absorption or distribution of egg white or dextran. On the other hand, the target organs may be made more sensitive to the active materials by virtue of a change in the permeability of the endothelial cells in the tissues. Further work on the mechanism of action of insulin by which hypersensitivity may be produced is now in progress.

R. K. SANYAL
Department of Pharmacology,
Darbhanga Medical College,
Bihar, India.

P. S. J. SPENCER
G. B. WEST
Department of Pharmacology,
School of Pharmacy,
29–39 Brunswick Square,
London, W.C.1.
Sept. 11.

[1] Adamkiewicz and Langlois, *J. Physiol.*, **145**, 667 (1959).
[2] Sanyal and West, *J. Physiol.*, **142**, 571 (1958).
[3] Parratt and West, *J. Physiol.*, **139**, 27 (1957).

Cardiac Output in Horses

IN a previous communication we described the determination of the cardiac output in cattle by an injection method using *T*.1824 as the intravascular indicator[1]. The same method was used to determine the cardiac output in horses. The technique used for obtaining arterial blood samples from the brachial artery of horses was that described by Fisher[2], and for convenience the serial arterial blood samples were collected at 2-sec. intervals. Duplicate determinations of the cardiac output were carried out on 10 adult horses which, so far as could be ascertained, had normal cardio-vascular systems. The 10 horses on which the determinations were made were not at the