blood before hypothalamic stimulation and 0.66 mU after hypothalamic stimulation, and these values may represent the physiological limits of oxytocin in direct contact with tissues. The lactogen-stimulatory effect of 100 mU Pitocin *in vitro* was therefore obtained with an unphysiological level of oxytocin. The Pitocin used in this study was commercially prepared by extraction of posterior pituitaries and, since epinephrine has been identified in the hypothalamus and other neural tissue(20), it is possible that epinephrine might have been present in the preparation. Epinephrine then would account for the increase in lactogen production by the highest Pitocin level.

Summary. Anterior pituitaries from female, hooded Norway rats were cultured in Mixture 199 for 3 days at 35 ± 1°C in 95% O_2-5% CO_2. Neurohumors serotonin, acetylcholine, epinephrine, and norepinephrine were added to the medium at a level of 1, 10, and 100 μg/ml; Pitocin and Pitressin were added at levels of 1, 10, and 100 mU/ml. Serotonin, acetylcholine, norepinephrine, and Pitressin did not alter lactogen production from that of controls. Epinephrine, at a level of 1 μg/ml, increased lactogen production 43% and Pitocin at a level of 100 mU increased lactogen production 53%.

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Influence of Oligodeoxyribonucleotides on Phagocytic Activity of the Reticuloendothelial System.* (30492)

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Enzymatic digests of deoxyribonucleic acid stimulate multiplication and DNA synthesis of Gram-positive bacteria and also enhance antibody formation in animals(1-3). The activity of these digests, which is independent of the source of DNA, is attributable to oligodeoxyribonucleotides and cannot be duplicated by monodeoxyribonucleotides or -sides. The activity of the oligonucleotides can be attributed, at least in part, to a stimulation of kinases involved in nucleoside synthesis (4). Administration of DNA digest with

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sheep erythrocytes results in a striking elevation of the early appearing number of hemolysin-forming cells in the spleen of adult mice(3) and causes an earlier appearance of hemolysin-forming cells in the lymphoid organs of newborn mice(5). These effects were demonstrated with the Jerne technique, which also revealed that the digest-induced acceleration of the immune response is blocked by kinetin riboside(3). DNA digest also increases host-resistance to a number of Gram-negative pathogens(2) and it, therefore, became of interest to determine whether such effects result only from a stimulation of antibody-forming cells or involve, in addition, an effect on the phagocytic activity of the reticuloendothelial system.

Material and methods. Male Ha/ICR mice, weighing 20-22 g, were injected i.v. with a suspension of Günther Wagner C-11/1431a carbon prepared by the method of Biozzi *et al*(6), and used at a dose of 15 mg/100 g body weight as previously described(7). To determine carbon clearance, 0.025 ml blood samples were taken from the retroorbital plexus at 5-minute intervals and residual blood carbon concentration was determined in 0.1% Na_2CO_3 at 675 mμ. Half-time ($t_{1/2}$) of clearance, in minutes, was obtained for each animal from the semi-logarithmic plot of residual blood carbon concentration *vs* time and mean $t_{1/2} \pm$ standard error was computed for each group. The curves were constructed by passing a line of the computed mean $t_{1/2}$ through the mean log of carbon concentration at 5 minutes.

The enzymatic digest of calf thymus DNA (DD) was prepared as previously described (3) and administered only once, partially i.v. and partially i.p. Kinetin riboside (KR) was dissolved in warm 95% ethyl alcohol, diluted with saline, and injected i.p. (2 mg/0.2 ml) immediately prior to injection of DD. Mixtures of the common monodeoxyribonucleotides and -nucleosides were prepared at concentrations equivalent to the DNA, as measured by optical density at 260 mμ, and injected by the same regimen. 6-Mercaptopurine (6-MP), dissolved in 0.05 N NaOH, was injected i.p. (10 mg/kg) daily for 3 days prior to, and on the day of, injection of DD. Precautions to avoid extraneous pyrogen contamination were rigorously observed.

Results. Twenty-four hours after administration of DD, carbon clearance was significantly enhanced ($t_{1/2} = 4.2 \pm 0.3$) in comparison to uninjected controls or controls given saline ($t_{1/2} = 13.6 \pm 0.6$). An enhancement was still noted at 48 hours ($t_{1/2} = 5.5 \pm 0.5$), but stimulatory effects were absent 72 hours after DD injection ($t_{1/2} = 11.6 \pm 1.3$) (Fig. 1).

These stimulatory effects of oligodeoxyribonucleotides were not duplicated by mixtures of the 4 common monodeoxyribonucleotides or -sides (Fig. 2).

Kinetin riboside, which by itself failed to influence clearance ($t_{1/2} = 16.7 \pm 0.6$), caused a partial reversal of the stimulatory effects of DD ($t_{1/2}$ of DD $= 5.3 \pm 0.8$; $t_{1/2}$ of DD + KR $= 10.9 \pm 0.5$; $t_{1/2}$ of controls $= 14.0 \pm 1.1$) (Fig. 3).

Administration of 6-MP, in doses known to affect immunological responsiveness in rabbits and rats, had no influence on clearance in presence or absence of DD (Fig. 4).

Discussion. The demonstration that DD enhances phagocytic activity, in addition to its previously recognized ability to stimulate early events in antibody synthesis, suggests that DD's ability to enhance host-resistance involves alterations of both cellular and humoral factors.

DD behaves in many respects like endotoxin and it has been suggested that part of the endotoxin effects are the consequence of the release of DNA breakdown products from intracellular sites(8). It should be noted that endotoxin effects occur independent of the administration of specific antigen whereas the effects of oligodeoxyribonucleotides on antibody formation require the concurrent injection of specific antigen(3). This requirement for antigen (replaceable by supplementing DD with small amounts of endotoxin) appears to be due to a requirement for altered permeability of the target cells which can be achieved by specific antigen, endotoxin or substances such as chlorpromazine, histamine, cortisone, or nitrogen mustard (HN2). Such requirement for altered permeability does not

appear to exist in the case of phagocytes since, as shown here, they can be stimulated by DD alone.

Unlike endotoxins, oligodeoxyribonucleotides do not cause an early alteration of clearance, either in a positive or a negative way (9). Clearance experiments done with DD-treated mice during the first 3 hours after administration of the stimulator revealed normal clearance rates. This observation is in

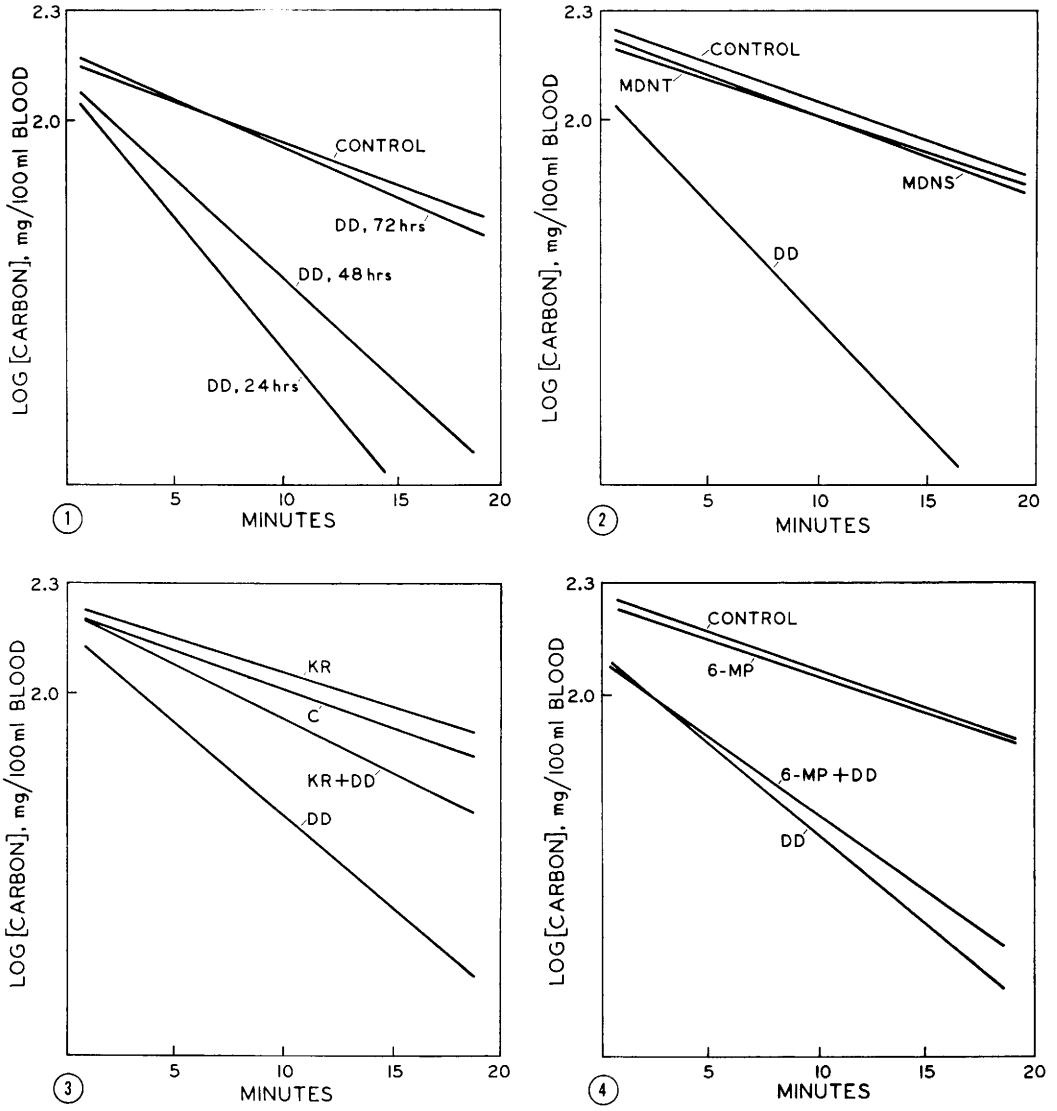

FIG. 1. Clearance of carbon from blood of control mice (12 animals), and in groups of mice tested at 24 hr (8 mice), 48 hr (8 mice), and 72 hr (6 mice) after treatment with oligodeoxyribonucleotides (DD).

FIG. 2. Clearance of carbon from blood of mice treated with mixtures of monodeoxyribonucleotides (MDNT) or monodeoxyribonucleosides (MDNS). Experimental groups contained 4-6 mice, control group 4 mice.

FIG. 3. Clearance of carbon from blood of control mice (5 animals), and in groups of mice 24 hr after treatment with oligodeoxyribonucleotides (DD), kinetin riboside (KR), or DD + KR. All groups contained 5-6 animals.

FIG. 4. Clearance of carbon from blood of mice treated with 6-mercaptopurine (6-MP) in presence or absence of oligodeoxyribonucleotides. Data collected on 4-5 animals in each group, tested 24 hr after treatment.

line with the prior suggestion(8) that endotoxin triggers the release of intracellular factors of which a DD-like material is only one, the factors responsible for early effects on clearance being absent in the case of DD administration.

It has also been suggested previously(8) that the release of stimulatory oligodeoxyribonucleotides may be a natural event, possibly as a consequence of interactions between antigen and antibody on cell surfaces. In view of the present data it can now be suggested that such stimulators may affect not only the multiplication and functioning of antibody-forming cells, but also the activity of cells involved in the uptake and processing of the antigen.

Summary. Oligodeoxyribonucleotides (but not monodeoxyribonucleotides), derived from calf thymus DNA, produce a significant stimulation of the phagocytic activity of the reticuloendothelial system. Kinetin riboside inhibits this stimulation without affecting the normal rate of clearance. This reversal of the effects of oligodeoxyribonucleotides is comparable to that previously observed in studies on stimulated antibody formation.

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Effect of Jensen Tumor on Pancreatic Diabetes in Nonadrenalectomized and Adrenalectomized Rats.*† (30493)

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The author is among several who have reported that certain tumors suppress the glycosuria of diabetic rats and other animals (1). The effect is more striking in steroid diabetes than in pancreatic diabetes(2). The adrenal cortices of the tumor host enlarge several-fold, yet the glycosuria is ameliorated, not exacerbated as would be expected if the tumor caused a state of hypercorticalism. If the adrenals of the tumor host should be incapable of secreting normal amounts of glucocorticoids, this might explain the suppression of glycosuria as due to hypocorticalism and the cortical hypertrophy as a compensatory response to hypocorticalism. A partial test of this hypothesis herein reported, shows that Jensen sarcoma suppresses the glycosuria of adrenalectomized rats given "normalizing" doses of adrenal cortex extract, therefore modification of secretion of corticoids cannot be the only way in which the tumor affects the glycosuria.

Methods. Male Sprague-Dawley rats were partially pancreatectomized at a weight of 275-285 g. The procedure of Ingle and Griffith(3) was modified to remove the pancreas lying between the bile duct and duodenum by gentle aspiration with a fine pipette. Any amount of pancreas can be removed within 10-15 minutes.

Extensive but not "complete" pancreatectomy was done on some animals and 85 to

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