per electrophoresis indicated that they possessed the properties of oxytocin and arginine vasopressin. The amino acid composition of the oxytocin and vasopressin isolated from the human glands was determined. It is evident that the oxytocin and vasopressin found in the human pituitary are identified as oxytocin and arginine vasopressin.

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## Effect of Cortisone and X-radiation on RNA and Glycogen Content of Rat Hepatocytes.* (24155)

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It was previously observed that DNA content of rat liver nuclei decreased both during cortisone administration and following x-radiation. In the former instance, the fall was progressive with continuing administration of the hormone; in the latter, there was a return to normal 5 days after x-radiation. Furthermore, daily cortisone administration to rats previously exposed to radiation caused a fall in DNA/nucleus and prevented the return to normal values at 5 days. A hypothesis was also presented to account partially for these observations. It was suggested that the apparent losses and increases in DNA/nucleus were due to depolymerization and repolymerization of the DNA(1). It appeared pertinent to study the effects of cortisone and/or x-radiation on the RNA and glycogen content of rat liver.

*Materials and methods.* Male rats of Wistar strain,†, weighing between 100-120 g were fed *ad libitum* on Lablox R diet.‡ Four groups of animals were studied: Group 1 received no treatment and served as control; Group 2 was injected with cortisone intramuscularly (25 mg/day) for one to 5 days and sacrificed after appropriate fast: Group 3 was exposed to x-radiation (1300 r) and killed from one to 5 days after x-radiation; Group 4 was cortisone-treated post-x-radiation; cortisone administration started within one hour after x-radiation and was given for one to 5 days. Details regarding cortisone dosage, frequency of administration, dosage and conditions of x-radiation and other precautionary steps were identical with those described previously(1). All animals were fasted 24 hours before removal of livers and were sacrificed between 9:00 and 10:00 a.m. to obviate diurnal variations in metabolism.

*Cellular disruption and chemical analyses.* At intervals after appropriate treatment, fasted animals were sacrificed and a 20% homogenate of liver pulp was prepared in slightly alkaline saline (containing 2 ml of 0.1 N NaOH/L of 0.85% NaCl W/V). Nucleic acids were extracted from a portion of the brei by procedure of Schneider(2) using hot trichloracetic acid (TCA), following removal of acid soluble materials with cold 5%

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TABLE I. Effect of X-radiation and/or Cortisone on RNA and Glycogen Content of Rat Liver.*

| Days after initiation of treatment§ | Cortisone (Group 2) RNA† | Cortisone (Group 2) Glycogen‡ | X-radiation (Group 3) RNA | X-radiation (Group 3) Glycogen | X-radiation and cortisone (Group 4) RNA | X-radiation and cortisone (Group 4) Glycogen |
|---|---|---|---|---|---|---|
| | mg/liver | | | | | |
| 0‖ | 19.8 ± .6 (12) | 23.8 ± .7 (12) | 19.8 ± .6 (12) | 23.8 ± .7 (12) | 19.8 ± .6 (12) | 23.8 ± .7 (12) |
| 1 | 24.4 ± 1.0 (10) | 74.4 ± 3.0 (10) | 24.4 ± .9 (10) | 49.5 ± 2.4 (10) | 28.6 ± 1.3 (10) | 188.1 ± 10.2 (10) |
| 2 | 28.0 ± 1.2 (10) | 181.4 ± 8.2 (10) | 22.9 ± 1.1 (10) | 71.6 ± 3.1 (10) | 33.1 ± 1.1 (10) | 302.9 ± 10.9 (10) |
| 3 | 27.0 ± 1.5 (10) | 357.5 ±14.6 (10) | 20.9 ± .9 (10) | 105.0 ± 4.2 (10) | 29.5 ± 1.7 (10) | 206.4 ± 10.5 (10) |
| 4 | 22.5 ± 1.1 (9) | 386.0 ±16.6 (9) | 16.4 ± 1.2 (10) | 12.9 ± .7 (10) | 21.6 ± 1.5 (11) | 180.1 ± 8.5 (11) |
| 5 | 21.0 ± .8 (9) | 353.0 ±14.6 (9) | 17.0 ± 1.8 (11) | 20.1 ± 1.5 (11) | 17.9 ± 1.4 (10) | 162.2 ± 8.2 (10) |

* Mean and stand. error of mean. Figures in parentheses refer to No. of experiments.
† RNA—Ribose.
‡ Glycogen values expressed as total reducing carbohydrate.
§ Days after x-radiation or days cortisone administered.
‖ Initial control Group I.

TCA. The TCA extract was analyzed for ribose by the orcinol reaction(3). Glycogen was determined on another portion of brei (4). The accompanying Table uses the term RNA to indicate nucleic acid-ribose, since this was the material determined. Glycogen content is expressed as total reducing carbohydrate. In view of alterations in liver weights following cortisone administration or exposure to x-radiation(1), the RNA and glycogen values are expressed as mg/liver.

*Results.* The data are presented in Table I. Total RNA increased and returned to normal in the last 3 groups of animals. While maximum values were observed on second day in Groups 2 and 4, x-radiation (Group 3) stimulated maximum RNA increase at 24 hours. However, in x-radiation, the increase was smallest, and the values returned to normal earliest. X-radiation, in conjunction with cortisone, showed largest accumulation of RNA, but even in this group of animals, RNA values had returned to normal by fifth day.

Data on glycogen accumulation may also be observed in Table I. X-radiation caused liver glycogen to increase for 3 days following exposure and to decrease sharply on days 4 and 5. Following cortisone administration, however, liver glycogen increased for 3 days and leveled off at very high value on days 3 to 5. When animals previously subjected to x-radiation received cortisone, glycogen content rose but not to the levels observed in animals receiving similar amounts of cortisone without radiation. It is apparent from these data that x-radiation modified the usual rate of accumulation of glycogen induced by cortisone. Acute x-radiation has been reported to decrease drastically liver glycogen content(5).

*Discussion.* It appears worth while to compare alterations on RNA and glycogen content obtained in the present study with those of DNA/nucleus obtained in a previous report(1). Such a comparison is warranted, since experimental conditions in both cases are similar and since these agents caused alteration in DNA content of rat hepatocytes. In animals which received cortisone alone, the maximum increase of RNA was obtained 24 hours prior to the time when a significant fall in concentration of DNA was observed, day 3. However, in animals which received x-radiation or both cortisone and x-radiation, maximum accumulation of RNA was found 48 hours earlier than when the fall in DNA became significant, days 3 and 5, respectively. The rapid increase in RNA content of rat liver to different levels under various conditions of treatment in our investigation would

imply either an increased rate of synthesis or decreased rate of destruction. The increases in RNA content observed might also be nonspecific, since administration of massive doses of estradiol and progesterone(6) to rats and thyroxine to rats(7) or mice(8) produces a similar rise in RNA content of liver. However, failure of RNA concentration to increase beyond the third day after x-radiation and or cortisone administration can be correlated with onset of a fall in DNA content. This may also be a reflection of interference with intracellular biochemical organization resulting from depolymerization of DNA. Allfrey and co-workers(9) reported that *in vitro* protein synthesis by isolated thymus nuclei, a reaction dependent on structural integrity of DNA, is inhibited when cortisone is added to the reaction mixture.

Accumulation of glycogen in cortisone-treated group reached a peak level (day 4) 24 hours after the fall in DNA had become significant. In animals exposed to x-radiation, the maximum deposition of glycogen in liver occurred on day 3, when DNA/nucleus was also minimum. When maximum accumulation of glycogen was observed in animals receiving both cortisone and x-radiation (day 2), DNA/nucleus was not different from that found in untreated control. In view of lack of certainty concerning the locus of action of cortisone(10-15), as well as paucity of information relevant to effects of x-radiation on enzymes involved in gluconeogenesis, any explanation of the manner by which x-radiation modifies glycogen accumulation by cortisone does not appear pertinent at the present time.

*Summary.* Rats were treated with cortisone, x-radiation and both agents in combination, and the effects noted on RNA and glycogen content of liver. The following observations were made on concentrations of RNA and glycogen relative to that of DNA: (1) Under various conditions of treatment, RNA content increased promptly to different degrees and returned to normal values at different times. Comparison of RNA content/liver in the present study, with those of DNA/nucleus in previous study, indicates that, in animals which received cortisone alone, the maximum increase in RNA was obtained 24 hours prior to a fall in concentration of DNA. However, accumulation of RNA in rats which received x-radiation or both cortisone and x-radiation was 48 hours earlier than when the fall of DNA became significant. (2) X-radiation or cortisone administration caused glycogen accumulation in liver, the former transiently, the latter in more dramatic and prolonged manner. X-radiation, prior to administration of cortisone, modified the response and decreased glycogen accumulation. Changes in glycogen content in contradistinction to RNA could not be correlated with those of DNA.

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