Acid and pepsin responses to gastrin in Heidenhain pouch dogs following bilateral adrenalectomy

ALLAN R. COOKE

*From the V. A. Center and Department of Medicine, UCLA School of Medicine,
Los Angeles, Calif., U.S.A.*

EDITORIAL COMMENT Bilateral adrenalectomy reduced acid output in response to histamine but not to gastrin stimulation.

In a previous study (Cooke, Preshaw, and Grossman, 1966a) bilateral adrenalectomy reduced significantly acid output from Heidenhain pouches in response to submaximal and maximal doses of histamine. Preliminary tests using gastrin stimulation suggested that bilateral adrenalectomy did not reduce the acid output from Heidenhain pouches (Cooke, Preshaw, and Grossman, 1965).

In this study using Heidenhain pouch dogs, evidence is presented (a) that acid output is unaffected following adrenalectomy using gastrin stimulation; (b) that pepsin output is unchanged by adrenalectomy using either gastrin or histamine stimulation; and (c) that acid and pepsin output is increased by giving hydrocortisone to the adrenalectomized animal.

METHODS

Four mongrel dogs weighing between 14 and 20 kg. were used. Under sodium pentobarbital anaesthesia a Heidenhain pouch was made and a Gregory cannula (Gregory, 1958) was inserted. Four weeks after recovery, experiments were started.

The animals were fasted 18 hours before each test. Studies were made during four periods: (1) control studies with adrenals intact; (2) studies following bilateral adrenalectomy while the animals were maintained on desoxycorticosterone acetate (D.O.C.A.) 2.5 mg. intramuscularly (i.m.) every second day together with added NaCl to their food (about 4 g./day). Tests were started about one month after adrenalectomy. The animals were in good health. (3) Studies when adrenalectomized while on D.O.C.A. (2.5 mg. i.m. every second day) plus hydrocortisone 25 mg. intramuscularly (i.m.) every second day; (4) studies when adrenalectomized while on no therapy apart from NaCl added to the food or given

as daily subcutaneous infusions (500 ml. 0.15 M NaCl).

For each experiment a continuous intravenous infusion (30 ml./hr.) of 0.15 M NaCl was commenced and gastric juice was collected for two 15-min. periods to confirm basal levels of secretion. At the end of 30 min. either gastrin or histamine was added to the NaCl to give the desired dosage. Each dose was doubled every 60 min. (histamine) or 75 min. (gastrin).

Gastric juice was collected by gravity drainage every 15 min. The volume was recorded to the nearest 0.1 ml. and the concentration of acid determined by titrating a 0.2 ml. sample of juice with 0.2 N NaOH to pH7 using a glass electrode and an automatic titrator.¹ Pepsin activity was determined using radioiodinated serum albumin as substrate (Grossman and Marks, 1960).

Gastrin was extracted from the pyloric gland mucosa of hog stomachs by a modification of the method of Gregory and Tracy (1961) described by Gillespie and Grossman (1963). A single batch was used throughout the study and the dose is expressed in terms of the wet weight of mucosa from which it was obtained. Histamine doses are expressed in terms of the weight of the dihydrochloride salt.

For each dog the significance of the difference before and after adrenalectomy was determined by the U test of Mann and Whitney (Siegel, 1956). The probabilities derived from the tests in individual dogs were combined by the method of Fisher (1958).

RESULTS

EFFECT OF BILATERAL ADRENALECTOMY ON GASTRIN-STIMULATED ACID AND PEPSIN SECRETION Acid output in response to gastrin was not significantly altered following bilateral adrenalectomy (Fig. 1). The maximal acid output before adrenalectomy was 425 μ Eq./15 min. and after adrenalectomy was 375 μ Eq./15 min. ($P > 0.1$). Pepsin output did not

This work was supported in part by Veterans Administration research funds and by U.S.P.H.S. grant No. AM8354.

¹Autoburet Radiometer, Copenhagen

FIG. 1. Dose response curves to gastrin before and after bilateral adrenalectomy. The scale for doses of gastrin is logarithmic. Each point is the mean acid output of the last two 15-min. periods at each dose, the vertical bars the S.E.M. Each line is the mean of 12 experiments in four dogs.

FIG. 2. Effect of adrenalectomy on acid output, pepsin concentration, and pepsin output in response to gastrin. Each line is the mean of 12 experiments in four dogs.

show any significant alteration after adrenalectomy (Fig. 2).

EFFECT OF BILATERAL ADRENALECTOMY ON HISTAMINE-STIMULATED ACID AND PEPSIN SECRETION Acid output in response to histamine was decreased significantly in all four animals after bilateral adrenalectomy ($P < 0.05$ all doses of histamine). Maximal acid output before adrenalectomy was $890 \mu\text{Eq./15 min.}$ and after adrenalectomy was $415 \mu\text{Eq./15 min.}$ (Fig. 3). Maximal pepsin output

FIG. 3. Effect of adrenalectomy on acid output, pepsin concentration, and pepsin output in response to histamine. Mean of 12 experiments in four dogs.

was not altered by bilateral adrenalectomy (Fig. 3). All the animals collapsed at the highest dose of histamine (6.0 mg./hr.). This effect has been described previously (Cooke *et al.*, 1966a).

EFFECT OF HYDROCORTISONE ON ACID AND PEPSIN OUTPUT FOLLOWING ADRENALECTOMY Each adrenalectomized animal receiving D.O.C.A. plus NaCl was given 100 mg. of hydrocortisone intramuscularly and then maintained on D.O.C.A. plus hydrocortisone 25 mg. i.m. every second day. They were tested five days later. The mean maximal acid output in response to histamine on the fifth day

FIG. 4. Effect of hydrocortisone administration on mean maximal acid and pepsin output in adrenalectomized dogs stimulated with histamine and gastrin. Each point is the mean of four experiments in four dogs.

(1,115 $\mu\text{Eq./15 min.}$) was greater than the pre-adrenalectomy maximal output (890 $\mu\text{Eq./15 min.}$) and on the eighth day the mean maximal acid output was 1,200 $\mu\text{Eq./15 min.}$ (Fig. 4). Over the ensuing days it decreased but from the twentieth day remained about 930–960 $\mu\text{Eq./15 min.}$ The mean maximal acid output in response to gastrin was slower to rise but by the tenth day (600 $\mu\text{Eq./15 min.}$) it was approximately one and a half times greater than the postadrenalectomy (NaCl plus D.O.C.A.) value (375 $\mu\text{Eq./15 min.}$). Mean maximal pepsin output in response to histamine and gastrin was greatest on the fifth and sixth days and thereafter decreased but by the twentieth day onward tended to remain constant (Fig. 4). Mean maximal pepsin output in response to histamine and gastrin was approximately twice control values from the twentieth day onward. After stopping hydrocortisone, acid and pepsin output decreased and returned to within the range of values obtained during the postadrenalectomy (NaCl plus D.O.C.A.) period (Fig. 4).

GASTRIN-STIMULATED ACID OUTPUT WHILE RECEIVING NaCl ONLY When the animals were tested after all therapy except NaCl had been suspended for two weeks, the maximal acid output in response to gastrin was not different from the preadrenalectomy studies ($P > 0.1$) (Fig. 1).

DISCUSSION

There are no published studies of the effect of bilateral adrenalectomy on gastrin-stimulated gastric acid and pepsin secretion.

In the present study bilateral adrenalectomy reduced significantly acid output from Heidenhain pouches in response to histamine (Fig. 3) and confirmed results obtained in a previous study (Cooke *et al.*, 1966a). Various mechanisms have been postulated to explain the reduction in acid output; neither parietal cell atrophy (Baker and Bridgman, 1954; Bralow, Komarov and Shay, 1964) nor metabolic alterations (Ramey and Goldstein, 1957) nor changes in the mucosal barrier (Cooke *et al.*, 1966a) have been demonstrated. In contrast to the decrease in acid output following adrenalectomy using histamine stimulation, acid output was unchanged when gastrin was used as a stimulus (Fig. 1). Before adrenalectomy, histamine was a more potent stimulant of acid secretion than was gastrin. After adrenalectomy, histamine and gastrin were approximately equal stimulants of acid output from dogs with a Heidenhain pouch (Fig. 4). The effect of adrenalectomy on acid output in response to gastrin in the present studies was different from that found in dogs with a gastric fistula (Cooke, Nahrwold, and Grossman, 1967). In those studies (Cooke *et al.*, 1967), acid output stimulated by gastrin was reduced by bilateral adrenalectomy. This difference is unexplained.

Pepsin secretion following adrenalectomy was found to be reduced in the rat (Tuerkischer and Wertheimer, 1945; Bralow *et al.*, 1964). In the dog, McIntosh, Anderson, Duthie, and Forrest (1960) reported that SU 4885, an 11β hydroxylase inhibitor, reduced gastric acid output but not pepsin output. The present findings indicate that pepsin output was unaffected by bilateral adrenalectomy whether histamine or gastrin was used as a stimulus (Fig. 2 and Fig. 3). The difference between the findings of the present investigation and those reported using the rat (Tuerkischer and Wertheimer, 1945; Bralow *et al.*, 1964) may be resolved on a basis of species difference. In the rat, chief cell involution was reported to occur after bilateral adrenalectomy (Abrams and Baker, 1954) but this was not found in the dog many months after hypophysectomy (Jacobson and Magnani, 1964).

In a previous study using histamine stimulation (Cooke *et al.*, 1966a) hydrocortisone given to adrenalectomized dogs produced acid outputs that were greater than preadrenalectomy control by the fifth day then gradually returned to within the range of preadrenalectomy control. These findings have been confirmed in the present study (Fig. 4).

Furthermore, mean maximal acid output in response to gastrin was greater than before adrenalectomy and this effect remained constant throughout the treatment period. In a previous study in dogs with Heidenhain pouches and intact adrenals, hydrocortisone (50 mg i.m. daily) increased mean maximal (Cooke and Grossman, 1966b) acid output in response to gastrin and histamine.

There are no previous studies of the effect of hydrocortisone on gastric secretion of pepsin in the adrenalectomized dog. In the rat, adrenalectomy reduced pepsin secretion and this was restored to preadrenalectomy values by glucocorticoids (Tuerkischer and Wertheimer, 1945). In the present study, hydrocortisone increased pepsin output in adrenalectomized dogs both in response to gastrin as well as to histamine. The response in general was similar to that found in dogs with their adrenals intact (Cooke and Grossman, 1966b).

SUMMARY

The effect of bilateral adrenalectomy on secretion of acid and pepsin by Heidenhain pouches was studied in four dogs. Bilateral adrenalectomy reduced acid output in response to histamine but not to gastrin stimulation. Pepsin output was unchanged by adrenalectomy. Hydrocortisone given to adrenalectomized dogs (25 mg. every second day) increased acid and pepsin output in response to both histamine and gastrin stimulation.

REFERENCES

- Abrams, G. D., and Baker, B. L. (1954). The cytology and secretory activity of gastric zymogenic cells after ablation of ductless glands. *Gastroenterology*, 27, 462-468.
- Baker, B. L., and Bridgman, R. M. (1954). The histology of the gastrointestinal mucosa (rat) after adrenalectomy or administration of adrenocortical hormones. *Amer. J. Anat.*, 94, 363-397.
- Bralow, S. P., Komarov, S. A., and Shay, H. (1964). Effect of total adrenalectomy on gastric secretion in chronic gastric fistula rats. *Amer. J. Physiol.*, 206, 1309-1314.
- Cooke, A. R., Preshaw, R. M., and Grossman, M. I. (1965). Gastric mucosal hydrogen ion transfer and adrenocorticoids. (Abstract) *Clin. Res.*, 13, 253.
- , — (1966a). Effect of adrenalectomy and glucorticoids on the secretion and absorption of the hydrogen ion. *Gastroenterology*, 50, 761-767.
- , Grossman, M. I. (1966b). Effect of hydrocortisone on secretion of acid and pepsin by Heidenhain pouches. *Proc. Soc. exp. Biol. (N.Y.)*, 123, 704-707.
- , Nahrwold, D. L., and Grossman, M. I. (1967). Effect of bilateral adrenalectomy on gastric acid and pepsin secretion from gastric fistulas and Heidenhain pouches in dogs. *Gastroenterology*, 52, 488-493.
- Fisher, R. A. (1958). *Statistical Methods for Research Workers*, 13th ed. Hafner, New York.
- Gillespie, I. E., and Grossman, M. I. (1963). Inhibition of gastric secretion by extracts containing gastrin. *Gastroenterology*, 44, 301-310.
- Gregory, R. A. (1958). Gastric secretory responses after portal venous ligation. *J. Physiol. (Lond.)*, 144, 123-1137.
- , and Tracy, H. J. (1961). The preparation and properties of gastrin. *Ibid.*, 156, 523-543.
- Grossman, M. I., and Marks, I. N. (1960). Secretion of pepsinogen by the pyloric glands of the dog with some observations on the histology of the gastric mucosa. *Gastroenterology*, 38, 343-352.
- Jacobson, E. D., and Magnani, T. J. (1964). Some effects of hypophysectomy on gastrointestinal function and structure. *Gut*, 5, 473-479.
- McIntosh, J. W., Anderson, N., Duthie, H. L., and Forrest, A. P. M. (1960). The effect of an adrenal inhibitor (SU 4885) on gastric secretion in dogs. *Ibid.*, 1, 345-350.
- Ramey, E. R., and Goldstein, M. S. (1957). The adrenal cortex and the sympathetic nervous system. *Physiol. Rev.*, 37, 155-195.
- Siegel, S. (1956). *Nonparametric Statistics for the Behavioral Sciences*. McGraw-Hill, New York.
- Tuerkischer, E., and Wertheimer, E. (1945). Adrenalectomy and gastric secretion. *J. Endocr.*, 4, 143-151.