Evidence for a Monophasic Response in Aldosterone and Cortisol Secretion to Hemorrhage (35739)

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It is well established that acute hemorrhage is accompanied by an increase in the rate of secretion of aldosterone (1–6) and the mechanisms involved have been extensively investigated. In recent studies by Fabre *et al.* (7), it was suggested that acute progressive hemorrhage in dogs resulted in an initial rise in aldosterone secretion, followed by a decrease in the secretion rate and that with further hemorrhage the aldosterone output was increased again, before a second preterminal decrease in the secretion rate occurred. Such a biphasic response to hemorrhage might suggest the existence of a more complicated control system than that now believed (2, 6) to regulate aldosterone secretion during hemorrhage. The present experiments were undertaken to reinvestigate the possibility of a biphasic response in aldosterone secretion to hemorrhage. Changes in cortisol secretion were studied simultaneously to determine if the response varied between aldosterone and cortisol.

Materials and Methods. Two series of experiments were performed. In the first group (series A), six female mongrel dogs, ranging from 13.0 to 23.8 kg of body weight, were used. Each dog was anesthetized with pentobarbital (30 mg/kg of body weight), and additional pentobarbital was administered as needed throughout the experiment. A polyvinyl chloride catheter was inserted into the left adrenolumbar vein distal to the adrenal gland and a loose ligature was placed around the vessel between the adrenal gland and the inferior vena cava; when this ligature was tightened, venous blood from the adrenal flowed through the catheter (1, 3, 4). Arterial blood pressure was measured by means of an aortic catheter with a Statham pressure transducer and a Sanborn model 7700 recorder.

At the beginning of the acute experiment, the ligature was tightened and the adrenal venous effluent was collected continuously. At the end of every 7.5 min time interval, a sample of approximately 10 ml of blood was collected in a graduated tube for analysis of the concentrations of aldosterone and cortisol. Toward the end of the experiment, sufficient adrenal venous blood for steroid analysis could not be obtained within the 7.5 min period and the interval between collections was lengthened. Between collection of samples, adrenal venous blood continued to flow into a graduated cylinder for quantitation of blood loss. At the beginning of each collection period, an arterial blood sample was obtained for the determination of plasma concentrations of sodium and potassium. Each experiment was terminated when the animal died or when the adrenal venous flow became less than 0.33ml/min.

In the second group of experiments (series B), the adrenal venous catheter was placed under sterile conditions 3 days before the acute experiment was performed. This design was used to reduce or to prevent the influence of surgical stress in augmenting the plasma level of ACTH and adrenal steroid secretion rates. Otherwise, the experiment design was the same as that employed in series A.

Plasma sodium and potassium concentrations were determined by flame photometry. The concentrations of aldosterone and cortisol in the adrenal venous plasma were measured by the double isotope derivative method of Kliman and Peterson (8). Because of the large number of samples obtained, only

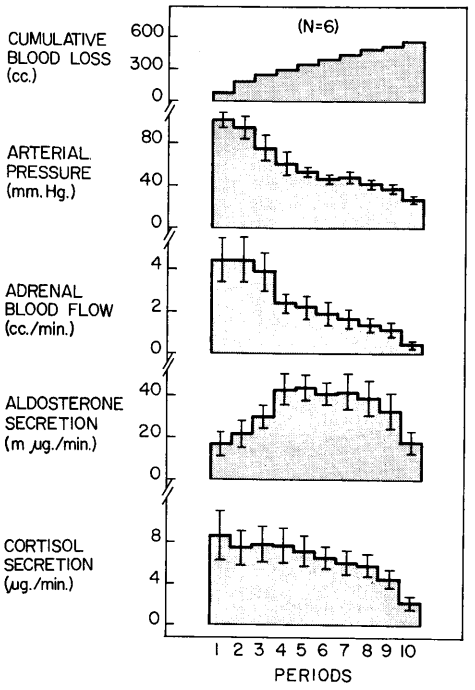

FIG. 1. Means (±SEM) for blood loss, mean arterial blood pressure, adrenal blood flow, and adrenal steroid secretion rates during each of 10 equal time intervals of the series A (acute catheter placement) experiments.

every other sample was analyzed for adrenal steroids.

Results. There was considerable variation in the duration of each individual experiment, which ranged from 154 to 208 min in series A and from 128 to 438 min in series B. Consequently, presentation of the data in composite form required the normalization of the time of the experiments. This was accomplished by dividing the duration of each experiment into 10 equal time intervals and determining an average value for each function studied in each experiment at each time interval. The data are presented in Figs. 1 and 2 with the means (± SEM) for the various functions measured for each of the 10 time intervals in the series A and B experiments, respectively. The continuous blood loss resulted in a progressive decrease in the arterial blood pressure in both experimental series. Adrenal blood flow also decreased during each experiment.

The secretion of aldosterone increased with hemorrhage in the series A (Fig. 1) and reached a near maximal level during the fourth period, remained at this level through the eighth time interval and decreased in the final two periods. Evaluation by the Student's t test for paired observations (9) revealed that the aldosterone secretion rates during the time intervals 4 through 8 were significantly higher ($p<0.05$) than during period 1; also, aldosterone secretion during the final period (period 10) was significantly lower ($p<0.05$) than during periods 4 through 9. In series B, again the rate of aldosterone secretion increased progressively with blood loss and reached a maximum value during the sixth period. The secretion of aldosterone remained at this elevated level until the final period when a decrease occurred. The secretion of aldosterone during time periods 6 through 9 was significantly greater ($p<0.05$) than during period 1. Secretion of aldosterone in period 10 was not significantly different when tested against other time periods. Cor-

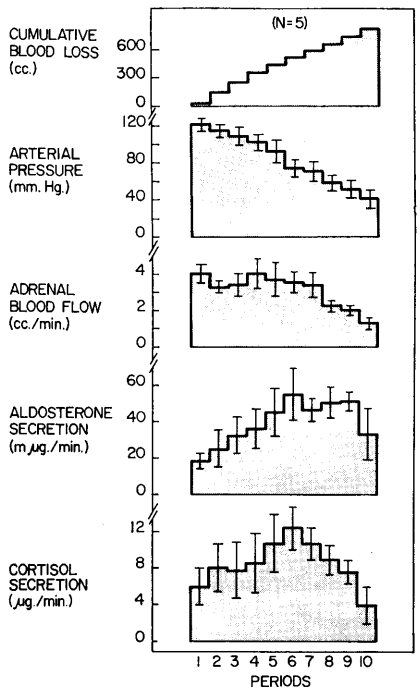

FIG. 2. Means (±SEM) for blood loss, mean arterial blood pressure, adrenal blood flow, and adrenal steroid secretion rates during each of 10 equal time intervals of the series B (chronic catheter placement) experiments.

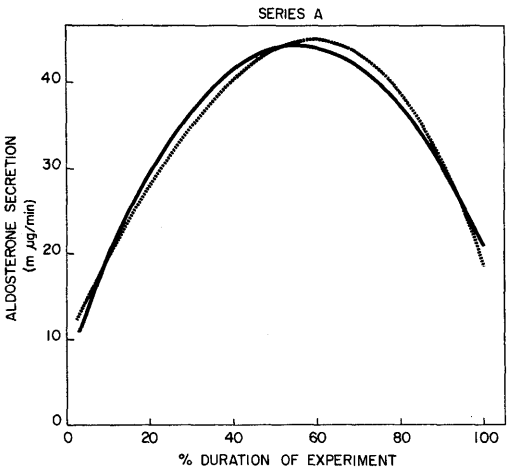

FIG. 3. Plots of the second order (—); and fourth order (- - - -) polynomial equations for the time course of the aldosterone secretion for all dogs in the series A experiments. The duration of each experiment is normalized to 100%.

tisol secretion in series A showed a slow progressive decline during the experiment; whereas in the series B, cortisol secretion increased with hemorrhage until the sixth period after which a progressive decrease occurred. In both experimental series, hemorrhage had no significant effect on the plasma concentrations of sodium or potassium.

Since our primary interest was in the time course of changes in the rate of aldosterone secretion, further data analysis was performed to determine more accurately these changes. After normalizing the duration of each experiment by dividing the time at which each sample was taken by the time at which the last adrenal venous sample was obtained (thus time for each experiment varied from 0 to 1 unit), the data were analyzed on an IBM 360/50 digital computer by use of a least-mean-squares subroutine. Second and fourth order polynomials were examined to determine their effectiveness in fitting the data for all dogs in the series A and B experiments. The resulting curves are presented in Figs. 3 and 4.

Discussion. The present study confirms the observations reported by many other workers (1–6) that there is an increased rate of aldosterone secretion in response to hemorrhage. In the present study, the increase in cortisol secretion observed in the series *B* experiments is also in agreement with similar observations of others (1, 2, 4, 6). Failure of cortisol secretion to increase in response to hemorrhage in the series A experiments probably reflects the state of stress of the animal at the beginning of the experiment due to the acute surgical procedures related to placement of the adrenal catheter. The decrease in both aldosterone and cortisol observed toward the end of the experiment (see Figs. 1 and 2) might be related to the low rate of adrenal blood flow present during this time. Other investigators have demonstrated that the rate of cortisol secretion by the canine adrenal is influenced by the adrenal blood flow rate (10, 11). A similar decrease in aldosterone secretion with declining adrenal blood flow during severe hemorrhage has been reported by Fabre *et al.* (7). Also, the rate of corticosterone secretion in rats is dependent on the rate of adrenal blood flow (12).

It has recently been reported by Fabre *et al.* (7) that acute progressive hemorrhage in the dog results in a biphasic response in the secretion of aldosterone. These workers, by use of an experimental design similar to that present in our series A experiments, reported finding an early increase in aldosterone secre-

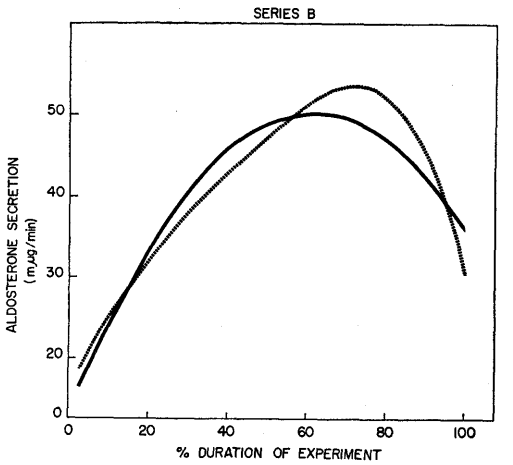

FIG. 4. Plots of the second order (—); and fourth order (- - - -) polynomial equations for the time course of the aldosterone secretion for all dogs in the series B experiments. The duration of each experiment is normalized to 100%.

tion during progressive hemorrhage; further hemorrhage was reported to produce a decline in aldosterone secretion, which was followed by a second rise in the secretion rate. As the degree of hemorrhage became severe and adrenal blood flow fell, a terminal decrease in the aldosterone secretion occurred. The data from the present study failed to demonstrate such a biphasic aldosterone response. It should be emphasized, again, that in the present study the acute response to hemorrhage was determined in two series of dogs. In series A, the response was evaluated in surgically stressed dogs as Fabre *et al.* (7) did; and in series B, the dogs were anesthetized 3 days after placement of the catheter after the immediate effects of surgical stress had subsided. In the series A experiments, aldosterone secretion during the sixth period was slightly less than during either the fifth or the seventh periods; similarly, the data in Fig. 2 show that in the series B experiments, aldosterone secretion during period 7 was slightly less than that during either the period immediately preceding or following. However, these apparent differences were small and were not significant. Therefore, these data cannot be interpreted as evidence for a biphasic response. Rather, the time course of the aldosterone secretion in the present studies suggests that the response is monophasic, since progressive hemorrhage produced an increase in aldosterone secretion during the early periods, a steady elevated rate of secretion and a fall in the secretion rate preterminally.

Further support for the hypothesis that the aldosterone response is monophasic rather than biphasic was obtained from the computer analysis of the data. The graphs describing the computed least-squared second and fourth order equations for the time course of aldosterone secretion are presented in Figs. 3 and 4 for the series A and B experiments, respectively. In both experiments the coefficients of the fourth order equation were such that terms greater than second order did not contribute significantly to the overall curve. This is a strong indication that the experimental curves are truly monophasic since higher order terms in a fourth order equation are needed to describe a biphasic response. Figures 3 and 4 show that for both the series A and B experiments the fourth order curves have only one peak, and in Fig. 3 (series A experiments) the second and fourth order curves almost coincide.

Evaluation of the data obtained from some of the individual dogs revealed results that appear similar to the individual data reported by Fabre *et al.* (7), except that considerably less fluctuation in aldosterone secretion occurred in the present experiments. The maximum and minimum areas in the normalized data of the present study were not coincident among the individual dogs. In our experiments, as in those of Fabre *et al.* the peaks and troughs in the curves from some dogs which seem to make the aldosterone response appear biphasic were usually determined by a single high or low value. Such apparent fluctuations in the rate of aldosterone secretion might be due to errors in the analysis of aldosterone. Failure to find a biphasic response in the present study raises serious question that such a phenomenon, if indeed it does exist, has any appreciable importance in reflecting factors which control the rate of secretion of aldosterone during hemorrhage.

Summary. The acute response in aldosterone and cortisol secretion in continuous progressive hemorrhage was studied in two series of dogs. In series A ($N = 6$), the animals were anesthetized, the catheters were inserted for collecting adrenal venous blood and recording pressure and the acute experiment was performed. In series B ($N = 5$), the adrenal venous catheter was placed 3 days before the acute experiment was done to minimize the effects of surgical stress on ACTH release and steroid secretion. In both series of animals, a clear-cut monophasic response to hemorrhage was observed for both aldosterone and cortisol secretion.

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