Clinicopathologic Effects of Chronic Reserpine Administration in Mongrel Dogs

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Abstract □ Twenty-four mongrel dogs were given low oral doses of reserpine twice daily for periods of 1–12 months, and certain clinicopathologic examinations were compared with findings from an equal number of control dogs which received placebos. Prolapsed nictitating membrane, miosis, and diarrhea were the initial clinical signs observed, and most reserpine-treated dogs exhibited the former throughout the study. CNS depression, muscle tremors, and a pronounced Parkinsonism-like syndrome were observed in the reserpine-treated population; these central effects, in contrast to alterations in peripheral autonomic activity, appeared to increase as the duration of reserpine administration lengthened. The hematocrit, hemoglobin content, and total leukocyte counts decreased in the reserpine-treated population, and the mean values were routinely significantly lower ($p < 0.05$) than in the control group. Right ventricular dilatation was diagnosed upon gross necropsy in five of nine dogs that received reserpine for 2–8 months and in three of four dogs that received the drug for 1 year. These findings suggest that the chronic administration of relatively low dose levels of reserpine in dogs may induce marked behavioral and ambulatory abnormalities and possible detrimental effects upon hematologic components and the myocardium.

Keyphrases □ Reserpine, chronic—clinicopathologic effects, dogs □ Antihypertensive agent—chronic reserpine administration, myocardial surveillance □ CNS depression, Parkinsonism-like syndrome—chronic reserpine, dogs □ Myocardial effect—chronic reserpine, dogs

The long-term administration of various dose levels of rauwolfia compounds for the therapeutic management of hypertension and other chronic ailments is well documented (1–4). Treatment periods are often a year in duration and may continue throughout the life of the patient. However, few experimental studies have simulated this chronic treatment schedule, even though the potential toxicity of large acute doses of reserpine is well recognized. Such studies would be important since, in certain circumstances, the prolonged administration of low dose levels of reserpine may also induce toxic responses. For example, clinical studies in man indicated that antihypertensive therapeutic doses of reserpine can adversely alter myocardial function (5) and induce signs of congestive heart failure (6, 7). Premature ventricular contractions and disruption of normal electrocardiographic patterns were likewise associated with therapeutic regimens of reserpine (8, 9). In experimental animals, alterations in myocardial function and structure were observed following the administration of relatively small quantities of reserpine (10, 11). It appeared, therefore, that a long-term study might elucidate certain effects of reserpine that may not be apparent upon short-term administration.

The purposes of the present study were to investigate certain effects of reserpine administered for 1–12 months in mongrel dogs and to correlate these data with information currently available from acute animal experiments as well as clinical studies in man. Relatively low doses of reserpine, less than those suggested for chronic clinical maintenance in the dog (12), were utilized in an attempt to simulate chronic administration of clinical dose levels in man.

MATERIALS AND METHODS

Animals and Treatment Schedule—The dogs used were mongrels received from a commercial supplier. Each dog selected was subjected to the following standards: (*a*) body weight between 8 and 12 kg. or appear as though, upon conditioning, it would be within this range; (*b*) age approximately between 1 and 3 years as determined by dentition; and (*c*) heart and lung sound, with no detectable abnormalities upon auscultation.

After selection, each accepted dog was vaccinated against common canine infectious diseases and treated for endo- and ectoparasites. Forty-eight dogs were selected, half males and half females, and placed in quarantine for at least 30 days prior to the beginning of the study. Approximately 500 g. of Purina dog chow was given once daily and water was available as needed. The dogs were randomly placed into four groups: male or female treated and male or female control. Each dog was maintained in an individual cage, and males and females were housed in separate rooms which were maintained at a constant temperature and a 12-hr. dark and 12-hr. artificial light cycle throughout the study.

A random sample of the tablets[1] contained 0.137 mg. of reserpine per tablet. Preliminary studies, conducted over an 8-week period, indicated that the minimal effective oral dose of reserpine in 10-kg. mongrel dogs (using relaxation of the nictitating membrane as the criterion) was approximately 0.15 mg., b.i.d. In the present study, each treated dog received 0.137 mg. of reserpine *per os*, b.i.d. through Day 21 of the test. At this point, the daily dose was reduced in half to 0.137 mg. per dog, which was continued through Day 62; however, at this time the original dose was reinstituted and maintained throughout the remainder of the study. The average daily dose of reserpine administered to each dog was approximately 26 mcg./kg. The dose varied from 18 to 39 mcg./kg.; however, the outside dose levels represent only a few animals since body weight was similar among the reserpine-treated dogs with little deviation from the group mean (see *Results*). The control dogs received a placebo tablet on the same schedule as the treated animals. The clinical condition of each dog was assessed daily, and body weights were determined every 2 weeks and body temperature twice a week.

Clinical Chemistry—The following clinical hematologic tests were conducted at 2–4-week intervals on most of the dogs: blood urea nitrogen, erythrocyte count, leukocyte count (total and differential), hemoglobin, hematocrit, bilirubin, and blood glucose. Serum glutamic-oxalacetic transaminase was assayed utilizing a radioisotopic method developed in this laboratory[2]. A sample of serum (0.05 ml.) was incubated for 1 hr. at 37° in a phosphate buffer medium containing aspartic acid and α-ketoglutaric acid-U-^{14}C. The incubation mixture was then acidified and passed over a 0.5 × 3-cm. column of Dowex-50 (hydrogen form) exchange resin. The product of the reaction, glutamic acid-U-^{14}C, was retained on the column, subsequently eluted with alkali, and quantitated in a liquid scintillation counter.

Twelve of the reserpine-treated and 12 control dogs (six of each sex) were randomly selected for glucose tolerance tests, and the re-

[1] Supplied and assayed by Ciba Pharmaceutical Co., Summit, N. J.
[2] Unpublished study of R. J. Ertel *et al.*

maining 12 treated and 12 controls were employed every 4 weeks for adrenocortical function tests utilizing β^{1-24}-corticotrophin[3]. For glucose tolerance determinations, the dogs received an intravenous dose of 1 g./kg. of sterile glucose solution (50%) following the collection of the initial preglucose-load blood sample. Venous blood samples were withdrawn at 15 and 60 min. after loading, and blood glucose was determined utilizing prepared reagents[4].

The analysis of adrenocortical function was performed essentially as described by Wood *et al.* (13). The animals received a single dose of 250 mcg. of β^{1-24}-corticotrophin after a predrug blood sample was obtained. Thirty minutes after the administration of the synthetic corticotrophin, an additional blood sample was withdrawn, and the adrenal function was evaluated on the basis of the mcg./100 ml. increase in serum corticosteroids. Fluorometric analysis of serum corticosteroids was performed according to the method of Nielsen and Asfeldt (14), which is based on the principle that cortisol, the major corticosteroid, and corticosterone have the same fluorescent intensity 5 min. after development of fluorescence by the addition of ethanolic sulfuric acid solution in the final step. The effects of reserpine on serum sodium and potassium were determined utilizing an atomic absorption spectrophotometer (Perkin-Elmer).

Gross and Histopathologic Studies—The animals were sacrificed using pentobarbital administered by intravenous injection. Two dogs, one male and one female, were sacrificed from the treated and control groups each month for the first 5 months following the initiation of reserpine treatment, except at 3 months when one treated and two controls were studied. In addition, two treated and two control dogs were sacrificed after 8 months, and the remaining animals were sacrificed at the termination of the study. Gross necropsy examinations were conducted on the 11 reserpine-treated dogs that were sacrificed through the 8th month, on four dogs treated with reserpine for 1 year, and on control dogs. Changes in cardiac structure were determined utilizing a method that involves macroscopic evaluation of the relative size and consistencies of myocardial areas during postmortem examination (15). The following tissues were fixed in 10% buffered formalin and prepared for histopathologic examination utilizing hematoxylin and eosin staining: heart, lung, stomach, small intestine, adrenals, kidneys, liver, testes, ovaries, and pituitary glands.

Additional cardiovascular and biochemical studies were conducted throughout the investigation; details of these phases will be reported elsewhere.

RESULTS

Clinical Signs—Minimal variations were observed in body temperature and body weights in the control and treated populations throughout the 1-year study. The mean body temperature varied between 38.2 and 39.0° in both groups. The mean body weight ($\bar{X} \pm SE$) in the reserpine-treated group was 9.9 ± 0.57 kg. at the beginning of the study and 10.38 ± 0.57 kg. at the termination. The mean value was below 10 kg. on only five occasions throughout the study, and the maximum value did not exceed 10.73 ± 0.83 kg. In the control group, similar variations were observed, and the body weight was 10.2 ± 0.4 kg. at the beginning and 11.63 ± 0.69 kg. at the termination of the study. The differences between the mean body weights of the treated and control populations at the beginning and at the termination of the study were not significant ($p > 0.98$ and $p > 0.1$, respectively).

Cardiovascular studies[5] showed no significant differences in peripheral arterial pressure, heart rate, or blood volumes between control and treated groups prior to initiation of reserpine administration. Chronic administration of reserpine did not induce significant changes in the indirectly measured (obtained from the left forelimb using a Korotkoff microphone) arterial blood pressure of unanesthetized dogs. The heart rate was significantly depressed in the treated dogs after 2 weeks of reserpine administration, and this effect persisted throughout the study.

Relaxation of the nictitating membrane was the initial detectable clinical sign observed in the reserpine-treated dogs beginning on the 3rd day of reserpine administration. Within 2 months, 70–95% of the treated dogs exhibited this sign, which persisted throughout the study (Fig. 1). In the initial 6 months of the study, 5–8 mm. of the third eyelid was usually visible; however, after the 6th or 7th month, less of the structure was observed. Persistent prolapsed nictitating membrane was not seen in control dogs.

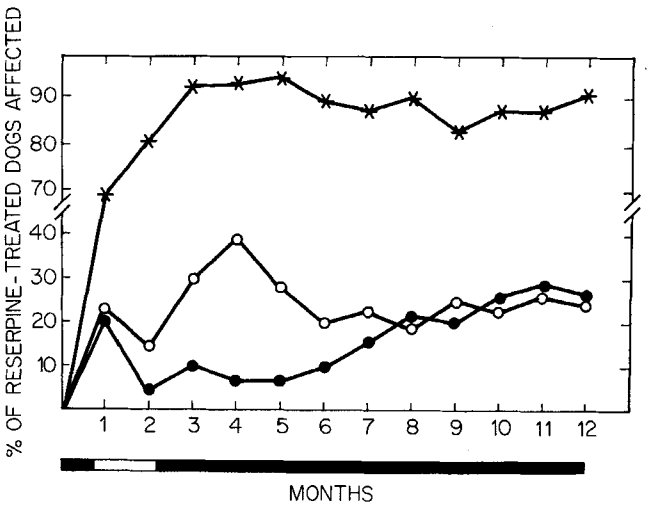

Figure 1—*Clinical signs observed in reserpine-treated dogs. Each value represents a monthly mean of clinical signs observed daily. Key:* *—*, *relaxed nictitating membrane;* ○—○, *tremors;* ●—●, *depression;* ■, *0.137 mg. reserpine, p.o., b.i.d.; and* □, *0.137 mg. reserpine, p.o., daily.*

The general behavioral patterns of the dogs demonstrated a form of sedation–depression within the initial month of the study, characterized by inactivity, recumbency, frequent narcosis, and minimal interest in stimuli from other activity within the animal room. The sedation–depression state was initially observed in three of the 24 treated dogs after 8 days of reserpine administration; however, the incidence of this condition rapidly increased through the 3rd and 4th weeks, when 33% of the treated dogs were affected. (As shown in Fig. 1, the monthly mean was 20%.) The incidence of sedation decreased, after a 1-week delay, during the 6-week period of reduced dosage (see *Methods*). However, upon reinstitution of the original dose of reserpine, the condition was again observed within a week and increased in incidence through the remainder of the study (Fig. 1).

Pupillary size determinations, measured with a millimeter rule under constant illumination, revealed miosis in four treated dogs after 4 days of reserpine administration. This condition was observed in 20–80% of the reserpine-treated population through 2 months of treatment; after this time, reliable results could not be determined due to individual variation. During this same period, miosis was diagnosed in less than 15% of the control dogs.

Blepharoptosis was occasionally observed in treated dogs only after the initial 3–4 months of reserpine administration; this sign was neither persistent nor pronounced, and it became unnoticeable upon stimulation of the animal.

Diarrhea was observed in the first 2 weeks of the study in six of the reserpine-treated dogs. It lasted only a few days and recurred in only one treated dog throughout the 1-year study.

Resting muscle tremors were noticed after a week of reserpine administration in approximately 25% of the reserpine-treated dogs. After a reduction in the incidence and severity of tremors during the period of decreased dosage, the incidence again increased through the 4th month when approximately 40% of the treated dogs were affected. By the 6th month and throughout the remainder of the study, between 20 and 30% of the reserpine-treated population exhibited resting tremors (Fig. 1). A larger percentage of reserpine-treated dogs often exhibited physiological tremors upon removal from their cages; however, only those individuals exhibiting resting tremors while in the cage undisturbed were classified as positive for this sign.

A prominent Parkinsonism-like syndrome was observed in three dogs after 3, 7, and 8 months, respectively, of reserpine treatment. This condition was characterized by severe skeletal muscle tremors, head and limb shaking, ataxia, difficulty in ambula-

[3] Synacthen, Cortrosyn.
[4] Diagnostest, Dow Chemical Co.
[5] Unpublished data of B. S. Jandhyala *et al.*

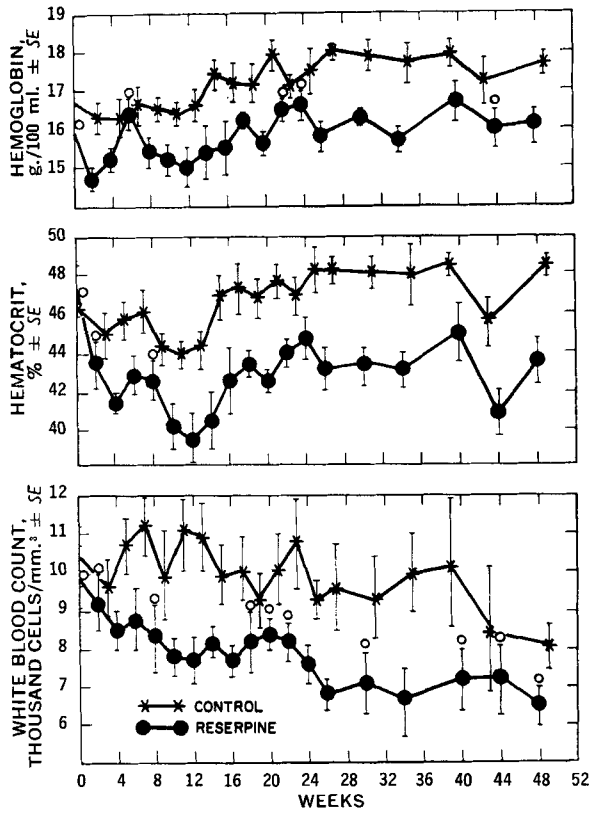

Figure 2—*Effects of chronic reserpine administration on certain hematologic components in dogs.* O = *not significantly different from respective control values* ($p > 0.05$). *All other values of the reserpine-treated group were significantly less than respective control values* ($p < 0.05$).

tion, and severe depression. A similar, though less severe, syndrome was periodically observed in other dogs. Reserpine administration was stopped in the dog exhibiting the Parkinsonism-like signs after 3 months of treatment, and the dog returned to normal behavioral patterns in approximately 10 days. However, when the original dose of reserpine was reinstituted, the animal again exhibited marked depression, severe tremors, head shaking, diarrhea, and ambulatory difficulties within 10 days. Reserpine administration was again halted, and a complete remission of clinical signs was observed in 2 weeks after which the animal was deleted from the study.

Reserpine administration was continued in the dog exhibiting Parkinsonism-like signs after 7 months of treatment. After 1 month of this syndrome, the dog became even more unresponsive. It was persistently recumbent and semicomatose in appearance, and extreme coaxing was necessary to induce standing or walking. Walking appeared very difficult and was stilt-legged and shuffling in manner and extremely slow. Additional withdrawal, nonambulation, and depression occurred until death after 9 months of reserpine administration. Autolytic changes observed upon necropsy precluded pathologic evaluation of this animal. The reserpine dosage was decreased by 50% in the dog exhibiting the Parkinsonism-like syndrome after 8 months of treatment, and a gradual decrease in severity of the syndrome was observed. However, the dog continued to exhibit pronounced depression and tremors while regaining body weight.

Hematologic Studies—The data summarized in Fig. 2 show that the most significant alterations produced by chronic reserpine administration were in the hemoglobin content, hematocrit, and total leukocyte count. Throughout the study, the hemoglobin content and hematocrit content were consistently and significantly lower than corresponding control values, although total erythrocyte counts were not significantly altered. The mean total leukocyte count of the reserpine-treated group was consistantly lower than the total leukocyte count of the control group. This reduction in the total leukocyte count was not associated with a significant alteration of the relative percentages of monocytes, lymphocytes, or granulocytes.

Analysis of blood urea nitrogen, glucose, and bilirubin revealed no significant differences in these values throughout the study. The mean serum glutamic-oxalacetic transaminase activity ranged between 7.5 and 11 μM/min./l. in both groups of dogs throughout the period of study, and no significant differences were observed. Aside from an initial rise in serum K^+ observed in the reserpine group during the first 4 weeks of the program, the mean serum K^+ ranged between 4 and 5 meq./l. in both groups of dogs through the remainder of the study. Serum Na^+ levels in both the reserpine and control groups ranged between a mean of 135 and 145 meq./l. throughout the experimental period, and there were no consistently significant differences. The glucose tolerance tests were compared statistically on the basis of the sum of the three glucose values; that is, the sum of 0-time, 15-min., and 1-hr. postload glucose values. Aside from a significantly lower ($p < 0.05$) glucose tolerance sum (GTS) in the reserpine group at Weeks 4 and 6, there were no significant differences in the GTS throughout the remainder of the experimental period. There were no significant differences between the reserpine-treated and respective control group values either in the resting corticosteroid levels or in the increase in serum levels following administration of synthetic corticotrophin. The resting mean ($\pm SE$) serum corticosteroid level varied from 3.75 ± 0.5 to 4.70 ± 0.7 mcg./100 ml. in the control group and from 2.93 ± 0.4 to 4.22 ± 0.3 mcg./100 ml. in the reserpine-treated dogs. Following the administration of β^{1-24}-corticotrophin, the mean ($\pm SE$) increase in serum corticosteroid levels varied from 7.76 ± 0.8 to 8.87 ± 1.24 mcg./100 ml. in the control dogs and from 6.03 ± 0.7 to 7.24 ± 0.7 mcg./100 ml. in the reserpine-treated group.

Pathologic Findings—Right ventricular dilatation was diagnosed in eight of the 15 reserpine-treated dogs that were necropsied. Abnormal changes in other cardiac structures and chambers were not noticed. The right ventricular finding was observed initially in two dogs after 2 months of treatment, in one dog after 3 months, and again in two dogs after 8 months of reserpine administration. This lesion was also observed in three of four dogs treated with reserpine for 1 year (Table I). The gross diagnosis of right ventricular dilatation was based primarily upon the enlarged, rounded, and flaccid appearance of the right ventricle (Fig. 3), although a relatively thin wall and enlarged lumen were also usually noted. The degree of right ventricular involvement in this example is representative of the condition observed in other treated dogs and differs markedly from the normal right ventricle (16). The gross appearance of right ventricular dilatation was observed in only two of 24 control dogs (one at 4 and one at 12 months), and the degree of flaccidity observed in the right ventricular wall of reserpine-treated dogs was not seen in the hearts from these two controls. The incidence of this lesion was significantly greater ($p < 0.01$) in the reserpine-treated group when the data obtained from the sacrifice periods were pooled (Table I). In the dogs treated with reserpine for 2–8 months, focal fatty infiltration was often observed upon histopathologic examination of the right-side dilated hearts. However, control dogs occasionally exhibited the same findings, and this histologic lesion was, therefore, classified as incidental. Chronic interstitial nephritis and chronic peribronchial pneumonia also were occasionally seen in the control and treated dogs. These findings did not appear to be correlated with the incidence of right ventricular changes and were considered incidental findings. All other tissues examined appeared normal.

Table I—Incidence of Right Ventricular Dilatation in Hearts of Dogs Treated Chronically with Reserpine

Months	Treated (n = 15) Positive	Treated (n = 15) Negative	Control (n = 24) Positive	Control (n = 24) Negative
1	0	2	0	2
2	2	0	0	2
3	1	0	0	2
4	0	2	1	1
5	0	2	0	2
8	2	0	0	2
12	3	1	1	11
Total	8[a]	7	2	22

[a] Significant difference between the reserpine-treated and control group, $p < 0.01$ (χ^2-analysis with Yates correction factor).

DISCUSSION

One purpose of the present investigation was to simulate chronic administration of clinical dose levels of reserpine in man; therefore, the dosages utilized bear discussion. Various quantities of reserpine have been studied in experimental and clinical situations in both man and animal. Following early experimental studies in animals, Earl (17) advocated the clinical use of 10–20 mcg./kg. for producing tranquilizing effects in dogs. Repeated doses of 50–100 mcg./kg. were reported to produce good effects in a much shorter time, with only an increased incidence of intestinal disturbances mentioned. Rauwolfia alkaloids have not been used extensively in the clinical practice of veterinary medicine, but 10–20 mcg./kg. of reserpine was suggested as an optimal daily oral dosage for ataractic effects in dogs (12). Gassner *et al.* (18), however, concluded that greater quantities of reserpine, 22–66 mcg./kg., were the most effective dosages in treating hypertension and other disorders in the same species. It was also mentioned that up to 35 mcg./kg. was well tolerated by dogs and that they could be chronically maintained on a daily oral dosage of up to 100 mcg./kg. of reserpine (12). It seems justifiable, therefore, to consider the daily oral administration of 18–39 mcg./kg., as in the present investigation, as a relatively low dose level in the dog.

In man, small amounts of reserpine, 0.1–0.6 mg., were administered daily in fixed-ratio combination with other antihypertensive agents for prolonged treatment of high blood pressure (2–4). Increased dosages, up to 1–2 mg. per day, however, were administered chronically when reserpine was the sole antihypertensive agent studied (1, 19). Also, daily doses of 10 mg. and often more were used in the management of mental disorders (1, 7). These reported quantities represent, considering a 70-kg. individual, a dose range of approximately 1.5–138 mcg./kg. per day. It is submitted, therefore, that the dose level used in the present investigation represents relatively low daily oral dosages.

The daily oral administration of 0.274 mg./dog of reserpine induced clinical evidence of the pharmacologic effects of the compound upon adrenergic neuronal function. The signs observed in the initial weeks of the investigation included a relaxed nictitating membrane, a decreased pupillary diameter, increased intestinal activity, and CNS depression. These effects can be attributed to the well-documented depletion of endogenous amines by reserpine (1, 20–22) with, in some instances, resulting parasympathetic dominance (23). These observations are in accord with Plummer *et al.* (23), who stated that the pupil and nictitating membrane of the dog are rapidly responsive to small amounts of reserpine, and with Earl (17), who suggested diarrhea as an indication of the attainment of effective reserpine dosage in the dog. However, with the exception of CNS depression, which steadily increased in incidence after a daily dosage of 0.274 mg. was reinstituted at the end of the 2nd month (Fig. 1), the severity of reserpine-induced effects upon adrenergic activity appeared to wane. For instance, the incidence of diarrhea was of short duration, and a decreased pupillary diameter was not detectable after the initial 2 months of the investigation due to the introduction of individual variability. Also, the extent of relaxation of the nictitating membrane appeared to decrease after 6–7 months of reserpine administration, even though the percentage of dogs affected remained the same. It is possible, therefore, that the peripheral effects of reserpine decline even though daily administration of the agent is continued. In contrast, adverse clinical manifestations of reserpine administration appeared to increase with the chronic daily administration of the drug.

Adverse clinical effects observed in this study were primarily severe CNS depression and a pronounced Parkinsonism-like syndrome. The presence of these effects in response to large doses of reserpine is well documented; however, this study demonstrates that the chronic daily administration of relatively small amounts of reserpine can likewise induce this characteristic syndrome in certain instances. A pronounced Parkinsonism-like condition with severe CNS depression was consistently present in only three dogs after 3, 7, and 8 months, respectively, of reserpine administration, although other dogs periodically exhibited less severe but similar signs after the initial few months of the study. This obvious clinical syndrome indicates the potentiality of toxic responses in certain, apparently susceptible, individuals following prolonged administration of relatively small amounts of the drug, even

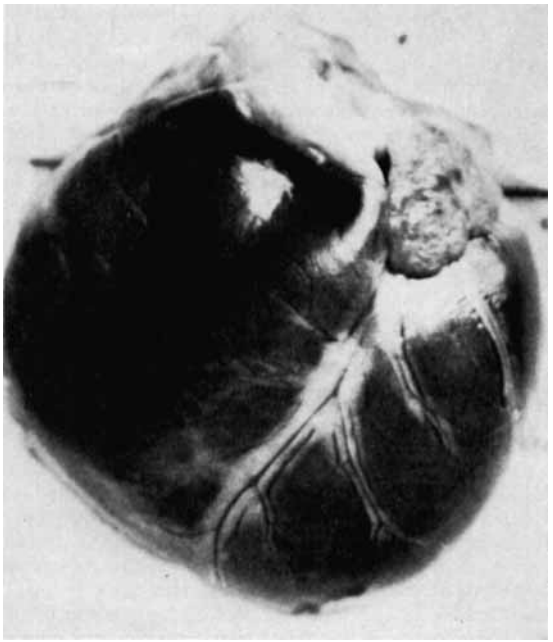

Figure 3—*Right ventricular dilatation observed in a reserpine-treated dog.*

though the mean group values for muscle tremors and CNS depression were not excessively high. Since both reserpine-induced CNS depression and Parkinsonism are probably related to the balance of brain amine levels, it appears that central pharmacologic actions of the drug, as determined by clinical evaluation, may be augmented by repeated administration whereas peripheral autonomic effects appear to decline during repeated dosage.

A possible adverse myocardial effect was another finding in the present investigation. In dogs necropsied at intervals throughout the 1-year study, right ventricular dilatation was observed in eight of 15 reserpine-treated dogs, whereas the lesion was seen in only two of 24 controls. Ventricular dilatation may be occasionally observed in clinically normal dogs following sacrifice, and this may explain the incidence of the finding in the two control dogs. However, in the present study, this represents less than 10% of the control population and, as previously mentioned, the lesion was diagnosed in approximately 50% of the treated dogs. Furthermore, pooled data obtained from all sacrifice periods indicate that the incidence in the two groups was statistically different ($p < 0.01$). This finding is in agreement with previous reports that reserpine can induce adverse alterations in myocardial function and structure in animals and, by inference, in man. Cohen *et al.* (5) examined cardiac effects of the chronic administration of 0.25 and 0.5 mg. of reserpine in patients with essential hypertension. Their study indicated that therapeutically administered reserpine may decrease cardiac output, increase atrioventricular conduction, and augment second-degree heart block during induced tachycardia in a hypertensive population. Similar antihypertensive–therapeutic doses of reserpine induced signs of cardiac failure in man (6, 7). Disruption of normal electrocardiographic patterns and premature ventricular contractions likewise were associated with therapeutic dose levels of reserpine (8, 9).

In dogs with experimental heart block, Roberts and Modell (24) observed that reserpine (0.1 mg./kg.) resulted in a high incidence of fatalities. These authors concluded that death was probably due to heart failure since cardiac enlargement, ascites, and pulmonary edema were observed upon gross necropsy examination. Withrington and Zaimis (25) and Zaimis (26) reported that circulatory failure was present in cats 24 hr. following the administration of 1 mg./kg. of reserpine. Furthermore, upon thoracotomy of these animals, the authors observed that the hearts were enlarged, flabby, and soft, and the right side appeared more severely affected. In later experiments, Withrington and Zaimis (10) studied the effects of chronic administration of doses of reserpine "of a similar magnitude to those administered to man." They concluded that the daily intraperitoneal injection of 10 mcg./kg. of reserpine in

cats for 5–26 weeks induced evidence of alterations in myocardial function.

Direct evidence of impairment of myocardial function by relatively high concentrations of reserpine, which appeared unrelated to catecholamine depletion, was presented by Nayler (27), utilizing the isolated toad ventricular strip. Histopathologic evidence of alterations of myocardial structure in cats treated with large doses of reserpine was reported by Scott and Zaimis (28). Also, marked mitochondrial swelling and fragmentation were seen upon electron microscopy of the myocardium following the daily administration of only 25 mcg./kg. of reserpine in dogs (11). It appears, therefore, that various dose levels of reserpine are capable of inducing deleterious changes upon the heart of man and animal. The present finding of right ventricular dilatation agrees with prior reports of similar lesions observed in dogs and cats. More important, it indicates that the long-term administration of relatively low doses of reserpine may introduce the possibility of adverse myocardial effects.

The hematologic determinations conducted in the present investigation generally remained within the normal limits of recognized canine blood values. However, in reserpine-treated dogs, the hematocrit and hemoglobin content were consistently and significantly lower ($p < 0.05$) than control determinations, and the total leukocyte count was consistently lower in the treated population. These findings are in contrast with the report by Earl (17), who observed no effect of chronic reserpine administration upon hematologic components in dogs; however, reserpine was administered for only 5 days a week rather than daily, as in the present study. The present hematologic findings are in agreement with Marley and Pare (7), who observed a reduced hemoglobin value and an increased plasma portion of the hematocrit following reserpine treatment in man, and, by inference, with West *et al.* (29), who observed a reserpine-mediated lack of terminal leukocytosis in leukemic mice.

Marley and Pare (7) suggested that reserpine produced a hemodilution *via* fluid retention and they rejected the possibility of altered hematopoiesis. However, impaired hematopoietic processes could induce a microcytic condition which would have given similar results and may explain the current findings. Also, the peripheral edema observed by these investigators may have been a sequela to the reserpine-induced cardiac failure rather than an indication of fluid retention leading to cardiac decompensation, as suggested. Blood volumes were unaffected by reserpine in the present study[6]; it appears, therefore, that fluid retention is an insignificant factor in reserpine-induced hematologic alterations. It is possible that the present hematologic findings induced by reserpine can be explained on the basis of an overall direct or indirect cytotoxic action of the agent, which has been suggested by previous reports (29–31). Likewise, a direct cytotoxic action of reserpine could explain the mechanism of its myocardial effect, which remains undetermined.

Reserpine is commonly administered on a chronic basis for the treatment of hypertension in man. Thus, a pathologic condition(s) of the cardiovascular system already exists upon initiation of reserpine therapy. In the present study, clinical evidence of cardiac failure was not observed. However, the incidence of cardiopathy, as detected by macroscopic examination, supports previous reports which suggest that rauwolfia preparations should be used cautiously in patients with preexisting cardiac irregularities (5–7). The findings of the present study, therefore, reinforce the need for patient surveillance during reserpine therapy. Furthermore, it is suggested that particular emphasis on surveillance of myocardial characteristics be considered during prolonged periods of administration of even small doses of reserpine.

[6] Unpublished data of B. S. Jandhyala *et al.*

REFERENCES

(1) A. R. Krogsgaard, "On Reserpine, with Particular Reference to Its Use in Hypertension," Munksgaard, Copenhagen, Denmark, 1961.

(2) H. Ausubel and M. L. Levine, *Curr. Ther. Res.*, **9,** 29(1967).

(3) D. K. Bloomfield and J. L. Cangiano, *ibid.*, **11,** 351(1969).

(4) J. L. Kalliomaki and A. Kasanen, *ibid.*, **11,** 344(1969).

(5) S. I. Cohen, M. W. Young, S. H. Lau, J. I. Haft, and A. N. Damato, *Circulation*, **37,** 738(1968).

(6) G. A. Perera, *J. Amer. Med. Ass.*, **159,** 439(1955).

(7) E. Marley and C. M. B. Pare, *Brit. Med. J.*, **1,** 267(1956).

(8) B. N. Wilson and N. A. Wimberley, *J. Amer. Med. Ass.*, **159,** 1363(1955).

(9) C. J. Schreader and M. M. Etzl, *ibid.*, **162,** 1256(1956).

(10) P. Withrington and E. Zaimis, *Cardiovasc. Res.*, **1,** 52(1967).

(11) D. E. L. Wilcken, D. Brender, C. D. Shorey, and G. J. MacDonald, *Science*, **157,** 1332(1967).

(12) N. H. Booth, in "Veterinary Pharmacology and Therapeutics," 3rd ed., L. M. Jones, Ed., Iowa State University Press, Ames, Iowa, 1965, pp. 215, 216.

(13) J. B. Wood, A. W. Frankland, V. H. T. James, and J. Landon, *Lancet*, **1,** 243(1965).

(14) E. Nielsen and V. H. Asfeldt, *Scand. J. Clin. Lab. Invest.*, **20,** 185(1967).

(15) T. Andre, *Nord. Vet. Med.*, **7,** 905(1955).

(16) M. E. Miller, "Anatomy of the Dog," 1st ed., W. B. Saunders, Philadelphia, Pa., 1964, p. 271.

(17) A. E. Earl, *J. Amer. Vet. Med. Ass.*, **129,** 227(1956).

(18) F. X. Gassner, V. D. Stauffer, K. W. Smith, J. T. Bennett, and J. R. Naylor, personal communication, quoted by A. E. Earl in *J. Amer. Vet. Med. Ass.*, **129,** 227(1956).

(19) A. D. Dennison, P. T. White, R. B. Moore, and W. J. Pierce, *Neurology*, **5,** 56(1955).

(20) B. B. Brodie, E. G. Tomich, R. Kuntzman, and P. A. Shore, *J. Pharmacol. Exp. Ther.*, **119,** 461(1957).

(21) J. H. Burn and M. J. Rand, *J. Physiol. (London)*, **144,** 314 (1958).

(22) E. N. Terry, R. H. Clauss, L. R. Rouen, and W. Redisch, *J. Cardiovasc. Surg.*, **10,** 287(1969).

(23) A. J. Plummer, A. Earl, J. Schneider, J. Trapol, and W. Barrett, *Ann. N. Y. Acad. Sci.*, **59,** 8(1954).

(24) J. Roberts and W. Modell, *Circ. Res.*, **9,** 171(1961).

(25) P. Withrington and E. Zaimis, *Brit. J. Pharmacol.*, **17,** 380(1961).

(26) E. Zaimis, *Nature*, **192,** 521(1961).

(27) W. G. Nayler, *J. Pharmacol. Exp. Ther.*, **139,** 222(1963).

(28) M. G. Scott and E. Zaimis, *J. Physiol. (London)*, **162,** 8P (1962).

(29) W. L. West, G. M. Baird, J. D. Steward, and S. N. Pradhan, *J. Pharmacol. Exp. Ther.*, **131,** 171(1961).

(30) A. Goldin, R. M. Burton, S. R. Humphreys, and J. M. Venditti, *Science*, **125,** 156(1957).

(31) G. A. Robison, S. Kruger, and F. W. Schueler, *Fed. Proc.*, **18,** 439(1959).

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