THE EFFECT OF A BRIEF PERIOD OF HIGH BLOOD PRESSURE ON CHOLESTEROL-INDUCED ATHEROMA IN RABBITS

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IN a previous investigation into the effect of high blood pressure on the severity of cholesterol-induced atheroma in rabbits (Bronte-Stewart and Heptinstall, 1954), it was noticed that 2 animals which underwent a spontaneous drop in pressure from their previous high levels had atheroma comparable in severity with those with a high pressure for the whole of the experiment. From this it appeared that a short period of high blood pressure might accentuate the amount of atheroma produced. In the present experiment the truth of this was tested, and it was found that in an 8-week feeding period, a high blood pressure for the first 2 or 3 weeks could increase the amount of aortic atheroma produced, although not to the same extent as in rabbits with a high pressure for the whole 8 weeks.

MATERIALS AND METHODS

Male Chinchilla and New Zealand White rabbits were divided into 3 groups. Group I consisted of 8 rabbits with high blood pressure produced by the removal of the right kidney and the application of a silver clip to the left renal artery 2 weeks later (Pickering and Prinzmetal, 1938). These were the rabbits referred to as Group I in the foregoing paper. The animals were fed diet 18 of Bruce and Parkes (1946) to which cholesterol was added (Heptinstall and Bronte-Stewart, 1954), the diet being adjusted so that each animal consumed between 3·5 and 4·2 g. cholesterol per week. Unlimited water was allowed and the total period of feeding was 8 weeks. Group II consisted of 7 rabbits in which a high blood pressure was produced in the same way and cholesterol feeding started as in Group I. After a period of between 14 and 21 days the silver clips were removed from the renal artery through the abdomen, with a resultant fall in blood pressure within 4 days to normal or near normal levels. Cholesterol feeding was maintained during this time and for the remainder of the 8-week period. Group III was a miscellaneous group of 7 rabbits acting as controls. Three were given a high blood pressure and the clip removed after 14–21 days as in Group II but differing in that no cholesterol was added to the diet during the whole of the period. After the blood pressure had fallen the rabbits were given the cholesterol-supplemented diet for 8 weeks. The remaining 4 were animals which failed to develop a high blood pressure or which showed an insignificant rise after removal of one kidney and the application of a silver clip to the other renal artery. These were fed the cholesterol diet for 8 weeks.

In all groups the animals were weighed and bled for serum total cholesterol estimations once weekly and blood pressures taken twice weekly as in the previous experiment (Heptinstall and Porter, 1957). After 8 weeks on the cholesterol diet the rabbits were killed and post-mortem examination carried out. The amount of atheroma in the aorta was recorded as described in the previous experiment and a " score " given to each rabbit. Adrenal, spleen, liver and carcass were weighed and paraffin and frozen sections made from the main organs.

RESULTS

Level of serum cholesterol

In most cases the serum total cholesterol reached a stable level after 4 weeks' feeding and for the remainder of the experiment remained at or near that level. Tables I and II show the mean levels over the last 4 weeks of feeding. There was no appreciable difference between the means in Groups I and II, while in Group III a higher level was maintained. An occasional rabbit showed a poorer response to feeding than the others and one such animal (number 49 in Group II) had a peak level of only 360 mg./100 ml. with a mean of 280 mg./100 ml.

TABLE I.—*Amount of Aortic Atheroma, Serum Total Cholesterol Level, Adrenal Wt./Carcass Wt. Ratio, Mean Blood Pressure and Mean Rise in Blood Pressure, for Groups I and III*

	Number of rabbit	Aortic atheroma score	Mean serum total cholesterol over last 4 wk. (mg./100 ml.)	Adrenal wt. × 100 / carcass wt.	Mean B.P. (mm. Hg)	Mean rise in B.P. (mm. Hg)
Group I	22	103	1450	0·070	114	42
	17	82	1110	0·042	120	46
	63	66	1590	0·074	105	30
	20	63	960	0·073	99	16
	31	41	1040	0·086	111	33
	48	40	990	0·051	100	28
	50	22	520	0·074	100	21
	15	15	1170	0·073	99	13
		Mean	1104±318	0·068±0·015		
Group III	11	11	760	0·069	81	0
	39	6	1390	0·150	91	12
	35	5	2730	0·075	78	0
	72	5	1350	0·101	80	13
	43	3	500	0·041	81	5
	62	2	1350	0·075	83	4
	37	1	2200	0·100	81	6
		Mean	1470±772	0·087±0·038		

Blood pressures

Rabbits in Group I maintained their high blood pressure (Table I) for the duration of the experiment with the exceptions of numbers 50 and 15 which showed a fall of 10–15 mm. Hg during the final 2 weeks. In spite of this their mean pressures for the duration of the experiment were 100 and 99 mm. Hg respectively.

Rabbits in Group II were allowed to have a high blood pressure for 14–21 days of cholesterol feeding, followed by removal of the clip on the left renal artery. This resulted in a fall in the blood pressure to normal or near normal levels within 4 days. The one exception, number 49, showed a fall of 17 mm. Hg but remained with a pressure 31 mm. above its pre-experiment level for the remainder of the feeding period. Table II shows the blood pressures at various stages of the experiment.

No rabbit in Group III (Table I) with the exception of number 39 exceeded a mean pressure of 83 mm. Hg during the experiment.

TABLE II.—*Amount of Aortic Atheroma, Serum Total Cholesterol Level, Adrenal Wt./Carcass Wt. Ratio, Levels of Blood Pressure at Different Periods for Group II*

Number of rabbit	Aortic atheroma score	Mean serum total cholesterol over last 4 wk. (mg./100 ml.)	Adrenal wt. × 100 / carcass wt.	Period of high B.P. (wk.) and level of B.P. (mm. Hg) Time	B.P.	Period of low B.P. after clip removal (wk.) and level of B.P. (mm. Hg) Time	B.P.	Mean level of B.P. for whole experiment (mm.Hg)	Mean pre-operative B.P. (mm.Hg)
26	45	1230	0·052	2	111	6	88	94	85
8	21	1310	0·078	2	99	6	78	84	80
53	15	930	0·040	3	100	5	83	88	78
44	12	910	0·080	3	100	5	80	89	79
49	8	280	0·060	2	120	6	103	110	72
59	8	1900	0·060	2	101	6	81	87	72
13	2	1340	0·083	3	97	5	84	91	75
Mean		1130±357	0·065±0·018						

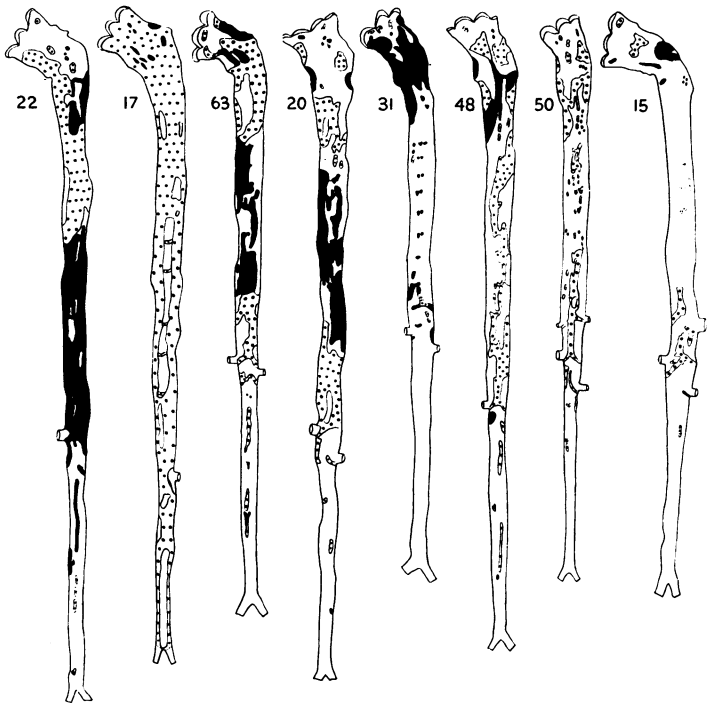

FIG. 1.—Amount of aortic atheroma in Group I (3/5 natural size). Dark areas represent prominently raised plaques and stippled areas slightly elevated plaques in this and in Fig. 2 and 3.

Amount of aortic atheroma

The amount of atheroma in the aorta in all three groups is adequately shown in Tables I and II and Fig. 1, 2 and 3. It will be seen that the amount in Group II is less than in Group I but greater than in Group III when the groups are considered as a whole.

Visceral changes

The usual visceral changes found in cholesterol-fed rabbits were present to a comparable degree in all 3 groups. Only one feature need be stressed and this is the adrenal wt./carcass wt. ratios. This has been shown to be a crude index of cholesterol absorption (Heptinstall and Bronte-Stewart, 1954), and in the present experiment is seen to be equal in Groups I and II but higher in Group III.

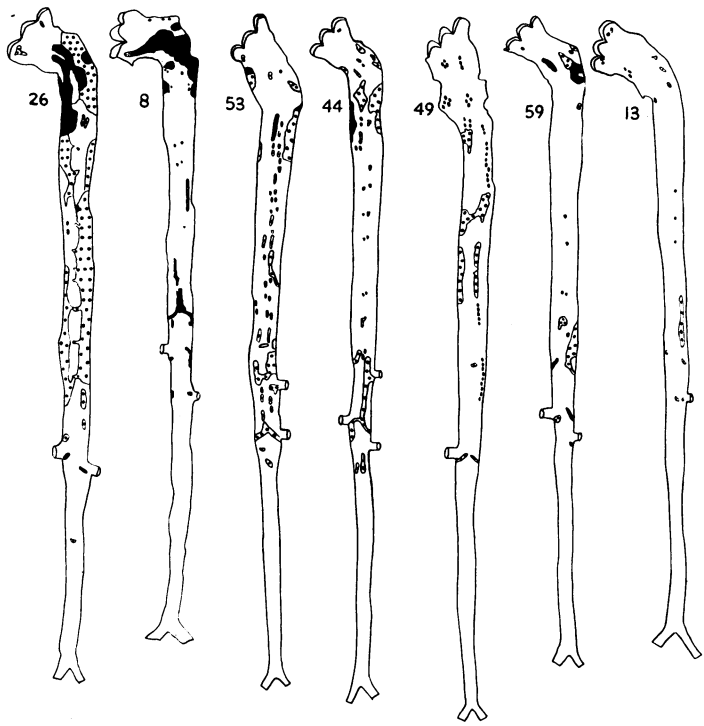

FIG. 2.—Amount of aortic atheroma in Group II (3/5 natural size).

DISCUSSION

To account for the difference in the amount of aortic atheroma in the 3 groups, certain factors can at once be eliminated. The duration and amount of cholesterol feeding were the same in all groups. The amount of cholesterol absorbed and available for deposition in the aorta is reflected in the serum total cholesterol levels and adrenal wt./carcass wt. ratios. Both of these are identical in Groups I and II but somewhat higher in Group III, the control group, a situation which hardly favours Groups I and II. All the animals used were of the same sex and

the proportions of Chinchilla and New Zealand White rabbits were the same in all groups. It is therefore evident that the level of the blood pressure is of the greatest importance in determining the amount of atheroma produced. A raised pressure during the whole of the feeding period produces considerably more aortic atheroma than a normal pressure, confirming the results of a previous experiment (Bronte-Stewart and Heptinstall, 1954). It will also be seen that the 2 rabbits in Group I which showed a terminal fall in pressure (numbers 50 and 15)

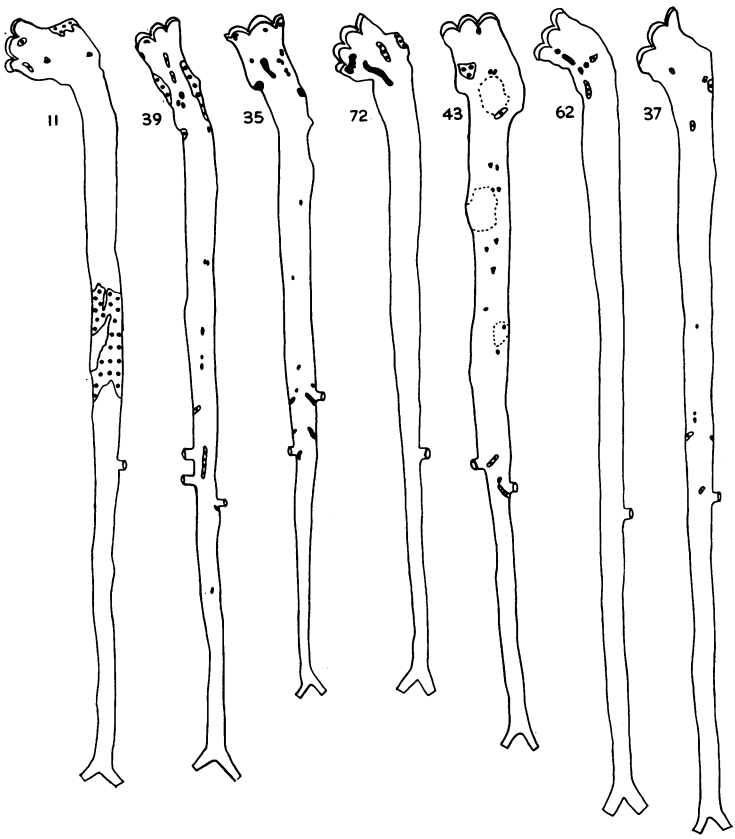

FIG. 3.—Amount of aortic atheroma in Group III (3/5 natural size). Areas enclosed by dotted line in rabbit 43 represent areas of spontaneous medial disease.

are the two with the least amount of atheroma in the group. Rabbits with a brief period of high blood pressure, Group II, show more atheroma than the control Group III but less than Group I. This, in certain measure, confirms what had been suspected in a previous experiment. Briefly it had been found that in a 10-week feeding experiment, 2 rabbits which had maintained blood pressures of 128 and 119 mm. Hg respectively for 5 weeks suddenly showed a drop in pressure. During the last 5 weeks of the experiment the blood pressures of both fluctuated between 88 and 98 mm. Hg. At post-mortem examination both were found to have aortic atheroma comparable with rabbits maintaining a high pressure for

the whole 10-week period. It was therefore decided to repeat this with an even briefer period of high blood pressure. The briefer period was partly conditioned by the observation of Blacket and Sellers (1951) that the fall in pressure is slower the longer the clip is in position before removal. In general in the present experiment the clip was removed 4–5 weeks after application, 2 weeks being allowed to make sure that the pressure was rising before cholesterol feeding was started, and 2–3 weeks on cholesterol being allowed before clip removal. The amount of aortic atheroma found in the rabbits of Group II was not so great as in the 2 rabbits in which the original observation was made, but the present experiment was 2 weeks shorter and the initial high pressure was neither so high nor so long maintained.

The reasons why a brief rise in pressure accentuates atheroma are not altogether clear. If the effect of the high pressure were merely to increase the passage of cholesterol into the vessel wall, then one would expect to see evidence of aortic atheroma at a time corresponding to the end of the period of high pressure. Examination of the aorta of rabbits with a high blood pressure fed cholesterol for 3 weeks shows no gross deposition of cholesterol and only insignificant amounts microscopically. However, it must be stated that examination of the aorta from the endothelial surface as described by Lautsch, McMillan and Duff (1953) has not been attempted, and this might provide information not ordinarily apparent. Local injury might provide an explanation for the increased lipid deposition. Examination of the aorta of rabbits on a normal diet with high blood pressures of a degree comparable with those in the experiment has revealed no obvious structural changes. Also 3 of the rabbits in Group III (numbers 11, 35 and 43) had high blood pressures for short periods prior to their pressures being reduced by clip removal before cholesterol feeding was started. These rabbits showed scores of 11, 5 and 3 which might have been expected to be higher if aortic damage had occurred during the period of high blood pressure. However it is conceivable that injury produced in a vessel wall during cholesterol feeding may be more potent in producing lipid localisation than an injury sustained before cholesterol feeding is started. It may well be as Duff (1954) has speculated, that when the whole organism is flooded with lipids, the smallest structural or functional aberration of the vessel wall may serve to localise the lipid.

Whatever the explanation, the findings serve to emphasise the important rôle played by high blood pressure in the accentuation of cholesterol-induced atheroma.

SUMMARY

Rabbits were divided into 3 groups all of which were fed cholesterol 0·5 g. per day for 8 weeks.

The first group consisted of animals which had a high blood pressure for the whole of the experiment. The second group had a high blood pressure for the first 2 or 3 weeks and a normal or near normal pressure for the remainder of the 8 weeks. The third group had normal or only slightly elevated pressures for the whole duration of the experiment. Aortic atheroma in the second group was greater than in the third but not so great as in the first.

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