EXPERIMENTAL ATHEROSCLEROSIS AND BLOOD PRESSURE IN THE RABBIT.

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The relation between atherosclerosis and blood pressure has engaged the attention of several workers following the statement of Fahr (1) and Van Leersum (2) that rabbits fed on abnormal diet (egg, liver) develop a high blood pressure. The published material that I have examined is disconcerting, due not so much to the variety of methods used to determine the blood pressure, as to the lack of sufficient information concerning the "normal" fluctuations of blood pressure, that is, the fluctuations observed in normal animals with the particular method used. Van Leersum, who claimed to have found a marked elevation of blood pressure under the influence of a liver diet, obtained no lesion whatever in the circulatory apparatus of his rabbits. Again most workers seem to be under the impression that the blood pressure is a constant, that is, that if the blood pressure oscillates in a certain region for a few weeks before the experiment, then any increase above this region that may occur afterwards during any experimental condition is necessarily due to the experimental condition, although these values may be well within values recorded from other normal animals. I have had considerable experience with Van Leersum's method (3, 4) and have given curves which sufficiently illustrate the fallacy of that assumption. For instance, in the graph of Rabbit 48-3 (3) it may be seen that the blood pressure oscillated around 100 mm. Hg for fully 10 months (from November, 1923, to September, 1924) and then rose in September and October, reaching 140 (average) on September 27. If an experiment had been started in the last week of August and this rise had been observed, the observation would have been supported by a good control period, but an inaccurate conclusion could have been drawn. Another type of curve is shown here, and still others may be found in former papers (3–5). To avoid repetition, by "blood pressure" and "normal range" is understood the systolic blood pressure of the rabbit as obtained from a carotid loop (Van Leersum's method) and the range of blood pressure in normal rabbits as determined previously by the writer.

The experimental atherosclerosis of the rabbit has in itself considerable intrinsic interest, so it seemed worth while to repeat the experiment. The literature on cholesterol feeding experiments is voluminous. For an introduction to the subject the reader is referred to the references given here, particularly Schönheimer. In the present work egg yolk was chosen since natural emulsions have been found best suited for the purpose. The rabbits (five in number) were given their usual food (hay, oats, greens) throughout the experiment; the yolk of one or two eggs was mixed with powdered unleavened bread and dried at 37°C., the whole appearing finally as yellowish crisp masses. The animals ate it readily in the beginning, but after some time they seemed to tire of it, so the yolk was diluted with plain water and the stomach tube used. The blood pressure was taken daily in the manner explained elsewhere (3). These five animals were chosen at random. One had been measured as a routine for several months, others for less time. Two of the five received lead carbonate by mouth in additon to the egg yolk. It seems justifiable to report the two cases where lead was also given, D 1, D 10, for the following reasons:

1. The association of lead poisoning and high blood pressure in man has always been considered in clinical medicine.

2. Lead carbonate in the form given to these animals and lead acetate given by stomach tube, in my experience, do not produce high blood pressure in the rabbit (unpublished data).

3. Recent work done in this country on the general subject of lead poisoning throws doubt on the efficiency of absorption of lead by the gastrointestinal canal (summarized in Reference 6).

4. There is no essential fact in the behavior or in the autopsy of these two animals that could be attributed with certainty to lead.

The curves were plotted at the completion of the experiment. The organs of the animals were carefully examined after death. Microscopic examination, however, was not systematically done. A brief analysis of Van Leersum's report (2) will be found at the end of the paper, followed by a note on the results obtained by other methods of measuring the blood pressure.

The essential data on these five animals will be given in the form of condensed protocols.

D 1.—Male, brown, Belgian rabbit. Nov. 30, 1923, carotid loop is made; weight 2.310 kilos. Jan. 7, 1924, blood pressure measurements started; weight 2.625 kilos.

Feb. 26, feeding experiment began; one egg yolk mixed with powdered "Matzos" and 30 mg. of lead carbonate smeared in carrot and fed by hand, daily; weight 2.855 kilos. Mar. 13, daily dose of lead carbonate increased to 60 mg. Mar. 29, daily dose of lead carbonate increased to 80 mg. Apr. 6, best weight 3.360 kilos.

Apr. 14, conjugate motions of eyes and head, toward the right, with drooping of right ear. Lead and egg feeding are interrupted. Apr. 16, same condition; weight 2.660 kilos. Apr. 21, right ear is full of a foul smelling creamy pus; weight 2.410 kilos. Apr. 30, eyes were found to possess well developed corneal arcs. May 11, weight 2.395 kilos. May 17, death in coma.

Total number of yolks consumed, 40. Total amount of lead carbonate given, 2.690 gm.

Blood Pressure.—Highest pressure recorded was 135 mm. Hg on February 7. This animal was one of those which exhibit a phenomenon described in the preceding paper (4) and ascribed to a local constriction of the carotid under the stimulation of the external pressure applied on the cuff. It is well illustrated by the following examples.

Feb. 6, 1924, 10.21 a.m.* 121–71–101–112 = 115–126–125–128 = 125–125–120–123 = 128–124–122–120 = 121–122–124–127 (pulse rate 192).

Mar. 4, 1924, 10.09 a.m.* 103–88–68–0 (15 seconds)–99–92–93–93 = 96–95–93–95 = 76–69–70–89 = 95–93–94–91 = 92–91–97–95 = 95–95–96–93 (pulse rate 168).

* Cuff adjusted to loop.

This phenomenon renders the tabulation or plotting of the figures almost impossible. It appeared throughout the course of the experiment, but not every day. It was absent in the last part of the experiment, when the intracranial complication of the otitis media became evident. The blood pressure during this latter period was, in general, low, oscillating between 70 and 90. The pulse rate varied between

136 (February 23 and April 23) and 220 (January 17) per minute. During the terminal coma the pulse became very irregular, a few beats passing through at 90 mm. Hg.

Autopsy.—Heart: base of large mitral cusp infiltrated with fatty substances. Aorta: large patch of infiltration at opening of arch branches, extending for a short distance into the common root of carotids and into left subclavian. In ascending arch there are a few minute nodular elevations. Nothing in thoracic aorta. In abdominal aorta, several small nodules and two streaks, one at root of celiac trunk, another at level of renal arteries. These two streaks are perpendicular to the axis of the aorta. Remainder of aorta and iliacs, normal. Pulmonary artery shows several elongated patches of moderate size, along posterior wall, parallel to the axis. Left adrenal, 950 mg.; right 860 mg. (weighed on March 26, 1927, in formol in the interval, see discussion). Corneal arcs, bilateral, well formed. Brain: white, thick, purulent exudate at level of tentorium cerebelli, both sides of midline. Right middle ear is filled with a creamy pus.

D 10.—Female, brown rabbit. Dec. 6, 1923, left carotid loop is made; weight 2.280 kilos. Jan. 7, 1924, blood pressure readings started; weight 2.465 kilos.

Feb. 26, feeding experiment began; one egg yolk mixed with powdered "Matzos" and 30 mg. lead carbonate smeared in carrot and fed by hand, daily; weight 2.705 kilos. Mar. 7 and 12, best weight 2.840 kilos. Mar. 13, daily dose of lead carbonate increased to 60 mg. Mar. 26, animal looks sick; weak; egg and lead are withheld. Mar. 27, weight 2.555 kilos. Apr. 16, egg given again; animal has recovered its former appearance and behavior, but not its weight; weight 2.515 kilos. Apr. 26, weight 2.700 kilos.

May 1, egg given through stomach tube; lead carbonate given again, 50 mg. daily. May 2, no corneal arc in either eye. May 15–17, lead carbonate suspended in egg yolk emulsion, stomach tube. May 16, weight 2.350 kilos. May 18, dead.

Total number of egg yolks consumed, 55. Total amount of lead carbonate given, 1.780 gm.

Blood Pressure.—Highest figures recorded before experiment: 146 (January 7, 1924), 150 (January 23), 149 (February 25) with averages for day, 140.4, 140.3, 144.8 respectively. Highest figures recorded during experiment: 152 (March 5), 150 (March 6), 149 (March 11), 148 (March 12), 150 (March 15) with averages for day, 139.6 (30 readings), 143.2, 145.9, 140.6, 143.0 respectively. From March 26 to April 10, the blood pressure reached the lowest level observed during the whole experiment, as low as 87 mm. Hg. This is the same period in which the animal appeared sick, concomitantly with loss in weight, loss in appetite, and, as it will be seen afterwards, increase in the pulse rate. I have no explanation for this. I have seen nothing like it in my experiments with egg yolk alone, or in the other animal which received lead together with the egg, or in several animals which have had lead carbonate or lead acetate alone (unpublished data). The pulse rate during this period oscillated between 216 and 288 per minute. The contrast in the behavior of pulse rate and blood pressure in the three periods, before March 26, from March 26 to April 10, and after April 10, is best seen in tabular form, where I have taken figures corresponding to the two extremes and mean of pulse rate for the respective periods.

Date	Pulse rate per min.	Blood pressure
1924		*mm. Hg*
Jan. 7	264*	138–146
Feb. 5	264	119–130
Feb. 9	272	118–124
Feb. 25	280	139–149
Jan. 9	232	116–138
Jan. 22	240	110–130
Mar. 17	232	130–135
Feb. 11	188	101–116
Mar. 26	216	87–92
Mar. 31	264	98–107
Apr. 5	280	97–101
Apr. 7	288	93–103
Apr. 16	192	102–110
May 1	144	111–126
May 14	240	122–129

* These fast rates are counted by groups of two $\underset{1}{\underline{\downarrow\downarrow}}\ \underset{2}{\underline{\downarrow\downarrow}}\ \underset{3}{\underline{\downarrow\downarrow}}$.

With increasing rates the pulse becomes very rhythmic and this process of computation is accordingly easier.

Autopsy.—Aorta is mottled throughout with yellow spots and streaks, slightly elevated, parallel to the axis of the vessel; somewhat more abundant in arch and thoracic portions. For a distance of 1.5

cm. above the opening of the celiac trunk these small infiltrated areas become confluent. Infiltration from this point downward is less and less marked. Pulmonary artery shows a large, irregular, slightly raised patch, at bifurcation, extending both ways for a short distance. In left kidney there are a few yellow streaks in outer zone of pyramid. Eyes show no visible corneal arcs. Adrenals are large, right weighs 440 mg. (weighed on March 26, 1927, almost 3 years in formaldehyde solution; left adrenal has been split open and a piece of central portion cut off for microscopic examination; there was no obvious difference in size at the time of the autopsy).

D 6.—Female, brown rabbit. Dec. 4, 1923, carotid loop is made; weight 1.785 kilos. Jan. 7, 1924, first blood pressure readings; weight 2.065 kilos.

Feb. 26, feeding experiment began; one egg yolk mixed with powdered "Matzos," daily; weight 2.495 kilos. Apr. 3rd and 4th weeks, rut. Apr. 26, best weight 2.820 kilos. Apr. 30, egg yolk given by stomach tube.

May 2, corneal arc is well formed in right eye, spreading toward center of cornea for a distance of 3 mm. from limbus. In left eye there begins to appear a faint, delicate, white line next to iridocorneal junction. June 28, weight 2.340 kilos.

July 14, two egg yolks by stomach tube, daily. Aug. 19, weight 2.165 kilos.

Aug. 21, animal is cold, wabbly, looks sick. Egg feeding is discontinued. Aug. 22, very weak, cold. Died at 11.30 p.m.

Total number of egg yolks consumed, 210 (without interruption, except isolated days).

Blood Pressure.—Range of figures where highest values were obtained, (*a*) before egg yolk feeding, (*b*) during egg yolk feeding, together with mean of set and pulse rate:

Date	Oscillation	Mean	No. readings	Pulse rate
1924	*mm. Hg*	*mm. Hg*		
(*a*) Jan. 14	128–134	130.4	10	168
Jan. 15	129–134	130.4	10	160
Feb. 26	128–140	133.7	10	
(*b*) Mar. 6	124–139	131.4	20	176
Mar. 27	105–135	115.2	20	160
Apr. 10	118–138	128.6	20	216
Apr. 29	122–143	136.1	19	216
May 3	114–142	134.2	20	216
May 6	120–141	130.9	20	200

Toward the end of May, during the whole of June and first half of July, the pressure reached the lowest values observed during the entire experiment, oscillating between 72 and 108 mm. Hg. Pressure above 120 was recorded on May 17, and the next pressure above 120 was recorded on July 18. The lowest temperature of the room where the measurements were done, was, in this interval of time, 19°C. (May 22, 11.05 a.m.), blood pressure 86–109, mean 97.8 (20 readings), pulse rate 152 per minute. The highest temperature in the same interval, 30°C. (June 24, 3.53 p.m.), blood pressure 85–93, mean 90.1 (10 readings), pulse rate 184 per minute. The thermometer is mounted on the stand of the manometer and the temperature read systematically at the end of the measurements. From July 18 on, the pressure was not as low as in the interval just discussed but reached levels in general not as high as those recorded before May 17, the only exception occurring on August 9, when the pressure rose to 141 after the rabbit moved during the measurement. The fastest pulse rate was 232 per minute, on February 19 and March 12, which did not coincide with the highest pressures. The lowest rate was 128 per minute, on June 28, with a blood pressure between 80 and 86, mean 83.1 (10 readings), and on August 22, when the rabbit was profoundly asthenic and the blood pressure was between 82 mm. and 90 mm. Hg (measured while animal was lying on its side).

Autopsy.—Marked dilatation of heart. Marked atherosis of aorta at root, arch and first portion of thoracic aorta, where intima is thoroughly infiltrated; from here on infiltration is patchy, mainly at the opening of branches, with a rather large patch at the opening of celiac trunk. The infiltration is greatest just beyond the opening of branches, in many instances forming like a crescent on the caudal side of the opening. Lumbar aorta is practically free; iliacs free. Carotids not involved, except at their opening, and excepting a small portion of the root of left carotid (that within loop), in continuation with the aortic infiltration. Aortic leaflets and large mitral cusp are slightly infiltrated. Pulmonary shows a large patch at bifurcation, extending both ways for a short distance. Root of pulmonary is completely free. Profound infiltration of liver, which feels hard, and in many areas is coarse, the capsule in these places being thick and opaque. Spleen large and pale. Adrenals are large; weighed on March 26, 1927 (after almost 3 years in formaldehyde solution), left 550 mg., right 500 mg. Ovaries thoroughly infiltrated, but scarcely larger than normal. Gall bladder much distended, walls not thickened. Bile is very thick. Lungs contain many whitish nodules throughout, not unlike a miliary tuberculosis, and small areas of congestion at both bases. The microscope revealed the presence of a radiating fungus in these nodules.

No. 47-0.—Male, gray, Belgian rabbit. July 7, 1923, left carotid loop is made; weight 2 kilos. July 31, first blood pressure readings. Jan. 12, 1924, weight 2.895 kilos.

Jan. 16, feeding experiment started; one egg yolk mixed with powdered "Matzos," daily. Feb. 16, best weight 3.360 kilos.

Apr. 30, well developed corneal arcs on upper segment of both corneæ. Egg yolk by stomach tube. May 2, corneal arcs spread to lower segments, equatorial parts remaining free. May 11, weight 2.645 kilos. May 12, looks sick. May 15, dead.

Total number of egg yolks consumed, 117 (without interruption).

Concerning the *blood pressure*, see Fig. 1 and discussion.

Autopsy.—Aorta thickened and opaque, intima greatly thickened, yellowish, rough and totally infiltrated from arch down to bifurcation; only spot free from atherosis is the root of the aorta where there is only a small elevated nodule. Pulmonary artery is almost as much involved as the aorta. Root of carotids and iliacs infiltrated. Small amount of fluid in abdomen. Walls of descending and transverse colon are yellowish. Spleen much enlarged, rounded edges, tough. Liver very pale, mottled with yellow. Adrenals very large (left 1.383 gm.; right 1.169 gm. weighed fresh), float in formol solution (10 per cent). Kidneys of normal size and consistency, surface smooth, yellow striations in pyramid following the normal rays; here and there, at base of pyramid, small yellowish nodules. Bilateral purulent pleurisy. Discrete patches of consolidation in left lung. Frozen sections of aorta show the intima to be *twice* as thick as the muscular coat. Histological details are omitted because they add nothing to the present knowledge of this experimental condition.

D 12.—Female, white rabbit. Dec. 10, 1923, left carotid loop is made; weight 1.855 kilos. Jan. 7, 1924, first blood pressure readings; weight 1.970 kilos.

Feb. 26, egg yolk feeding is started; weight 2.435 kilos (one egg yolk in bread, daily).

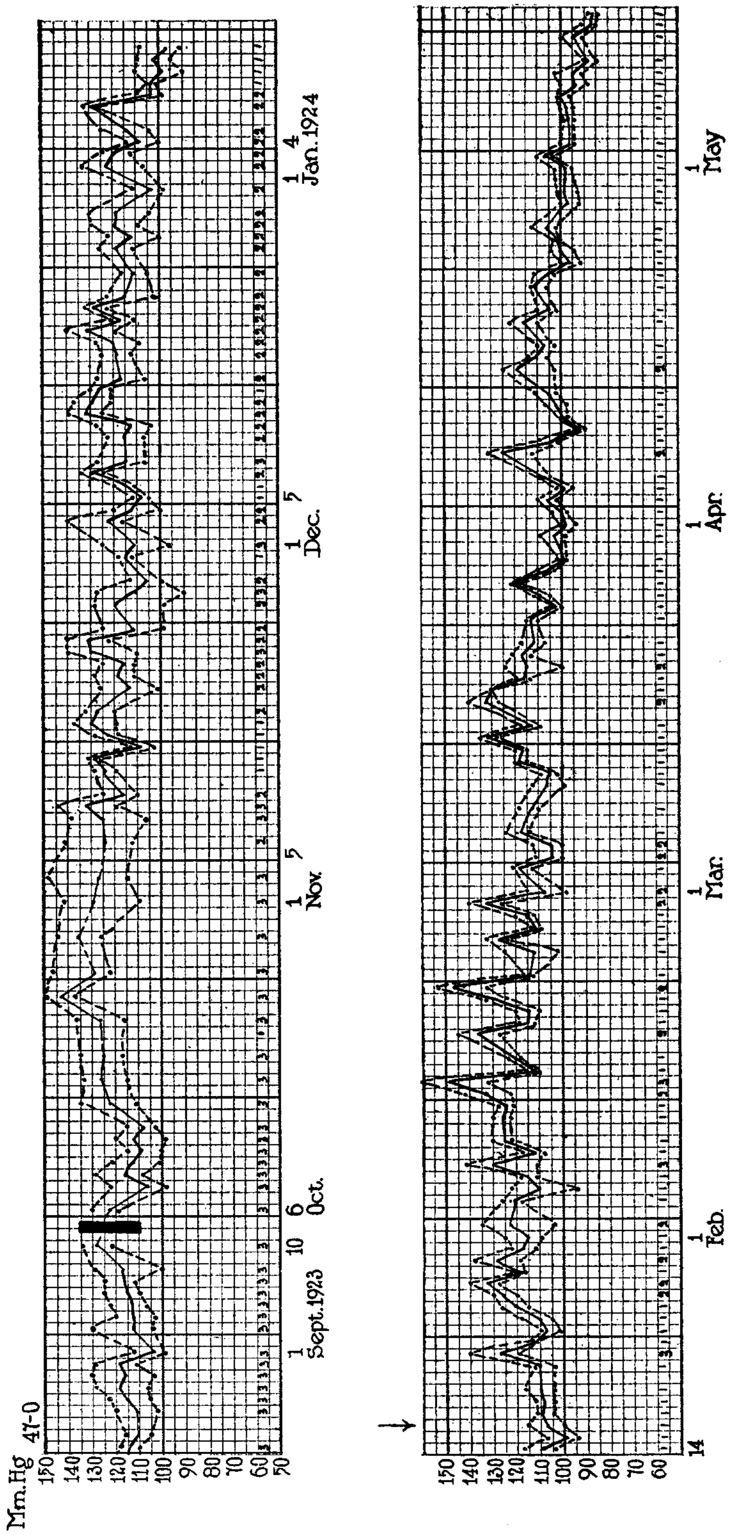

Fig. 1.

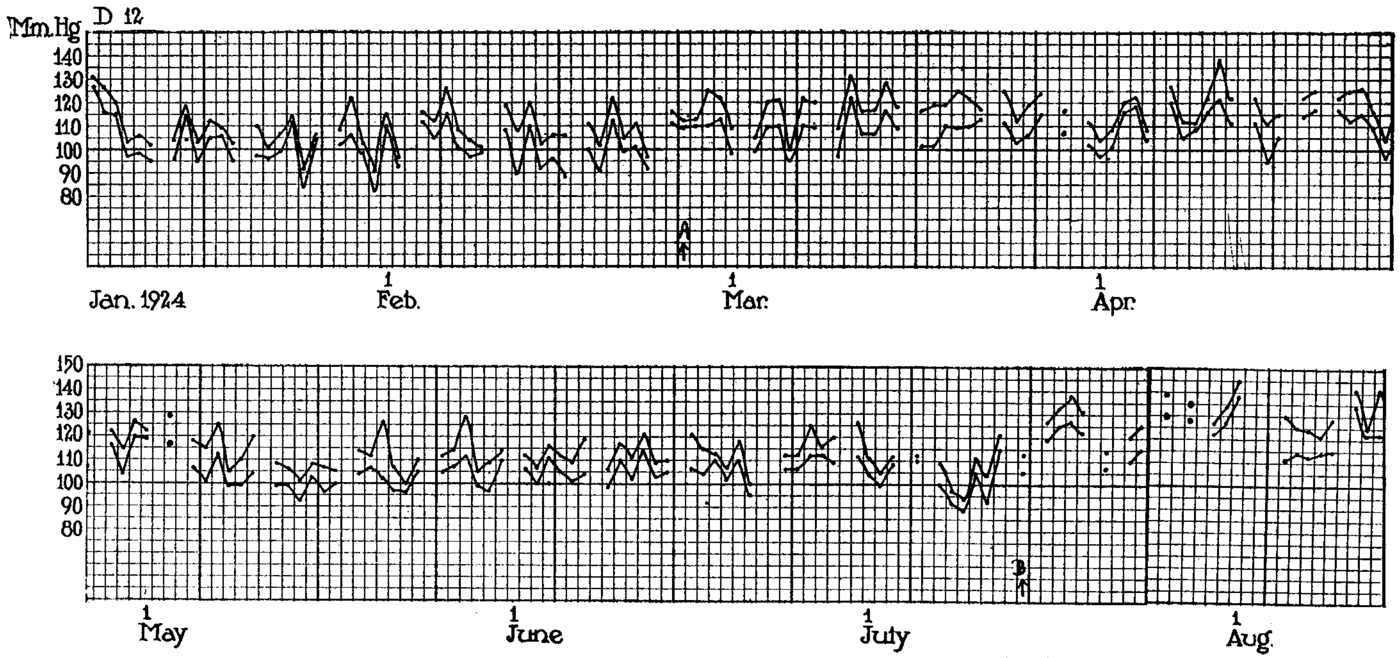
Mm.Hg
D 12
140
130
120
110
100
90
80
A
Jan. 1924
1 Feb.
1 Mar.
1 Apr.
150
140
130
120
110
100
90
80
B
1 May
1 June
1 July
1 Aug.

FIG. 2.

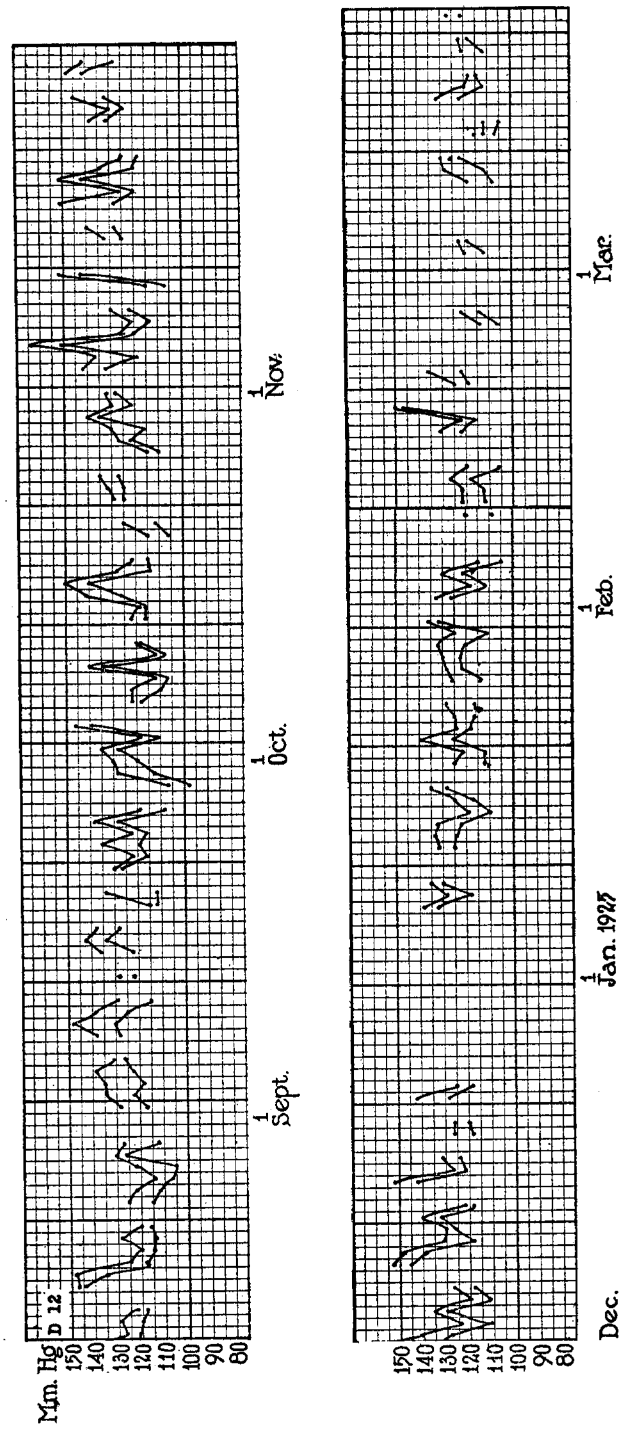
Mm. Hg
D 12
150
140
130
120
110
100
90
80
1 Sept.
1 Oct.
1 Nov.
Dec.
1 Jan. 1925
1 Feb.
1 Mar.

Fig. 3.

Apr. 30, egg yolk in milk, through stomach tube. May 1, egg yolk in water, stomach tube, daily. Weight 2.615 kilos.

July 14, two egg yolks in water given, daily. Dec. 27, weight 3.335 kilos. Mar. 17, 1925, weight 3.220 kilos.

Mar. 23, animal in excellent condition. It was sacrificed (chloroform) because the experiment was considered to have lasted sufficiently long. The animal did not receive any eggs from Dec. 28, 1924, to Jan. 5, 1925, inclusive. Occasional days scattered throughout the experiment were missed also.

Total number of egg yolks consumed, 531.

For *blood pressure* see discussion. Graph is reproduced in Figs. 2 and 3.

Autopsy.— *Aorta* and *pulmonary* artery are opaque, thick. Intima is rough, yellowish, wrinkled. In the pulmonary artery the infiltration is greatest around the bifurcation and extends into its branches. In the aorta the infiltration begins at the ring and extends clear down to the iliacs. All the main trunks are infiltrated a short distance from their root. The arch is dilated, and here the infiltration is greatest. Carotids and iliacs are not infiltrated. Mitral valve is thoroughly infiltrated; tricuspid only slightly. Liver: intense fatty infiltration. Costal margin has made a deep impression on convex surface and in this area the infiltration is greatest. Spleen somewhat enlarged. Transverse colon possesses a distinct yellowish discoloration of the mucosa. Uteri are enlarged, and vagina is greatly dilated (contains abundant mucus); cervices, full of clear, transparent polypi. Uterine mucosa is full of like polypi. Adrenals are large, left weighs 570 mg., right 471 mg., weighed fresh. Kidneys normal in size and shape; surface moderately pitted; cut surface shows a few well defined, large yellow nodules in the boundary zone, somewhat spindle-shaped, with the long axis parallel to the medullary rays; besides these large fatty deposits there are fine yellowish striæ in the medulla.

DISCUSSION.

Autopsy Findings.—All the animals developed varying degrees of atherosclerosis of the aorta; from a few flat clusters scattered here and there (D 1) to a total infiltration from root to bifurcation (No. 47-0 and D 12). The most severe lesion was found in No. 47-0 (117 egg yolks) where the fatty intima formed two-thirds of the total thickness of the aorta (7). D 6, after 210 egg yolks, and D 12, after 531 yolks, had a less profound infiltration, although it extended to the whole length of the aorta in D 12. The least infiltration occurred in D 1, which received the smallest number of eggs (40 eggs). The pulmonary artery was involved in all, the most extensive infiltration occurring again in No. 47-0.

The largest corneal infiltration was seen in D 6, a brown female rabbit. Corneal arcs were absent in D 10 (55 eggs) and D 12 (531 eggs). The former was a brown female rabbit, the latter an albino, female also. Schönheimer (8) concludes, from his experiments, that males are more resistant than females as far as the production of corneal arcs is concerned, and that extensive arcs are obtained only in females after prolonged administration of cholesterol. In accordance with this, the most extensive corneal infiltration was present in a female, but another female had no corneal arcs after 531 eggs. Since this animal was the only albino of the group, and Schönheimer says nothing about the color of the rabbits, this observation should be emphasized. On the other hand, D 10 had no corneal arcs after 55 egg yolks, while D 1, a brown male, had good corneal arcs after 40 eggs.

The fatty infiltration of the liver was most intense in D 12, without any evidence of a cirrhotic process; but in D 6 the liver was firm and in some places distinctly coarse. The spleen was considerably enlarged only in one (No. 47-0). The kidneys showed fatty streaks in the pyramid in three (D 10, No. 47-0, D 12) and discrete clumps in the boundary zone (Bailey) in two (No. 47-0 and D 12).

The adrenals were of good size in all. Unfortunately some of the adrenals were not weighed fresh, but after several years standing in formaldehyde solution. To get an approximate idea as to the effect of such prolonged fixation on the weight, the adrenals of No. 47-0 and D 12, which had been weighed at the time of the autopsy, were weighed again, that is, after an almost equally long formol fixation, with the result that the left gland of No. 47-0 *gained* about 8 per cent (the right had been sectioned and a piece removed for microscopic examination) whereas the adrenals of D 12 *lost* about 4 per cent. If it be assumed that all the other adrenals *gained* 8 per cent (to assume the worst case) and this amount be subtracted from the recorded weight, an average of 667 mg. for the left and somewhat higher for the right is obtained, far greater than the corresponding averages 238 and 221 mg. for fourteen normal rabbits above 2 kilos in body weight and greater than the averages 353 and 341 mg. for fourteen rabbits above 2 kilos in body weight, whose thyroid and parathyroids had been removed by Marine (Stewart and Rogoff (9)). In the extensive statistical work of Brown, Pearce and Van Allen (10) on 645 normal *male* rabbits, mainly from eastern Pennsylvania and the immediate vicinity of New York City, the mean combined weight of the adrenals is given as 0.383 gm. More details are unnecessary, since all these findings have been described before (8). My sole purpose is to show that the present animals actually had the now well recognized picture of experimental cholesteatosis, and that in particular the aorta showed from slight to extreme fatty intimal deposits. None of these aortas showed any gross calcification of the media. Schönheimer observed a marked calcification of the thoracic aorta in one of his animals, and says, very naively, that it is probably not due to the diet, but rather to the use of the stomach tube, which through repeated, short elevations of the blood pressure, may act like adrenalin injections. Although he had Schmidtmann's method (11) at hand, he did not use it to see if there was really an elevation of blood pressure, which if found, could still be ascribed to struggle or excitement, since the animals do not take the tube without resisting, and granting, of course, that adrenalin necrosis is mechanically produced and not due to a toxic action or something else.

The kidney deserves special mention. Bailey has found it more frequently affected than other investigators (Schönheimer). Bailey found the surface pitted in four out of nine animals egg-fed, or in six out of eight whose kidney showed gross cholesterol lesions. I find striæ and nodules also, but scars in the cortex only in one (D 12), a moderate scarring in fact, and although this animal received nine times as many eggs in less than five times as many days as Bailey's Rabbit 7, the xanthomatose lesions appear insignificant when compared with the extraordinary lesions illustrated in Bailey's paper (his Fig. 7, kidney of Rabbit 7). There were no scars in the kidneys of D1 and D10 which received lead carbonate. I am inclined to believe that these xanthomatose formations are secondary to a preexisting scarring of the cortex, a view considered by Bailey himself and by Schönheimer.

Blood Pressure.—Fig. 1 contains *all* the essential data from No. 47-0. The lower broken line represents the lowest reading of the corresponding day, the upper broken line the highest reading. The solid line is the calculated arithmetic mean. No figure has been discarded, but the measurements taken from July 31 to August 23 have been omitted to shorten the graph. The range of pressure in the omitted period covers from 90 mm. to 134 mm. Hg, with a mean of 110.0 (420 readings). The small figures at bottom of the graph multiplied by 10 give the number of readings of each day. No readings were taken from September 11 to October 5 (black bar in graph), or on isolated days (break in sequence of bottom figures). Egg feeding was started on January 16 (arrow in graph). The only difference in the blood pressure curve before and during the egg feeding consists in the wider range of daily oscillations in the former, a difference readily accounted for by the larger number of daily readings. Toward the end, when the animal was obviously sick (see protocol), the blood pressure was low. The blood pressure reached about 150 on two occasions, both before and during egg feeding. The details of these days are as follows:

Oct. 24, 1923, 10.41 a.m. 145–144–146–144–147–143–144–142–141–141 (pulse rate 200).
10.49 a.m. 149–145–143–144–141–143–145–142–143–146 (pulse rate 160).
10.58 a.m. 144–146–142–142–139–137–137–140–139–138 (pulse rate 160).
Nov. 3, 1923, 3.45 p.m. 129–127–126–126 = 127–126–130–130 = 130–130 (pulse rate 200).
3.51 p.m. 146–148–146 = †128–120–115–116 = 116–116–116 (pulse rate 184).
At the point indicated by † animal moved backward, in box.
3.58 p.m. 129–130–129–128 = 121–120–123–125 = 121–123 (pulse rate 176).
Feb. 14, 1924, 12.08 p.m. 160–160–155–153 = 156–150–152–150 = 150–148–147–152 = 152–147–146–142 = 149–152–150–145 (pulse rate 200). Mouth piece removed for a few seconds, cuff in place, animal did not move.
12.17 p.m. 145–142–135–132 = 139–140–149–140 = 144–146 (pulse rate 188).
4.59 p.m. 118–121–122–122 = 128–129–129–131 = 136–134 (pulse rate 184).

5.07 p.m. 155–153–152–150 = 145–144–144–142 = 143–144 (pulse rate 180).

Feb. 22, 1924, 3.18 p.m. 138–133–139–139 = 142–142–145–143 = 150–150–149–144 = 149–150–150–149 = 150–153–153 = 153 (pulse rate 176).

Pulse rate behaved as follows:

Fastest, Aug. 30, 1923, 248 per minute, with a blood pressure between 125 and 130 mm. Hg, mean 126.7 (first 10 readings).

Slowest, Nov. 27, 1923, 128 per minute, blood pressure 90–104, mean 95.3 (1st 10 readings).

Apr. 19, 1924, 128 per minute, blood pressure 102–110, mean 104.8 (10 readings).

Apr. 30, 120 per minute, blood pressure 94–102, mean 98.3 (10 readings).

May 1, 128 per minute, blood pressure 95–102, mean 98.2 (10 readings).

On May 12, 13 and 14, when the animal was suffering from a pleuropneumonia, the pulse rate was 192, 224, 192, respectively, and the blood pressure oscillated between 84 and 100 mm. Hg. The curve illustrates one of those rare animals which cover the whole range from 90 to 150 mm. Hg, that is, almost the totality of the normal fluctuations of blood pressure in the rabbit. Without special indication it would be impossible to pick out the egg feeding period. D 1 with a maximum pressure of 135 mm. Hg and D 6 with a maximum of 143, may be dismissed without further discussion. D 10 does not differ essentially from No. 47-0.

There remains D 12, whose graph is reproduced in Figs. 2 and 3. During January and February, 1924, up to *A* (see Fig. 2), blood pressure oscillated between 83 and 132 mm. Hg, with a mean of 104.9. From *A* to *B*, 139 days (animal receiving one egg yolk daily), the blood pressure oscillated between 88 and 138 mm. Hg, with a mean of 109.4, the oscillations being somewhat greater during March and April than during May and June. From *B* on to the end of the experiment (animal receiving two egg yolks daily), the blood pressure was in general higher and the oscillations became larger, between 98 and 165 mm. Hg, with a mean of 124.0. Pulse rate behaved as follows:

Fastest, Nov. 5, 1924 (resistance), 256 per minute, blood pressure 151–164 mm. Hg, mean 155 (20 readings).

Nov. 17, 256 per minute, blood pressure 128–151, mean 135.3 (20 readings).

Slowest, June 3, 1924, 132 per minute, slightly arythmic, blood pressure 100–107 mm. Hg, mean 102.1 (10 readings).

July 9, 1924, 136 per minute, blood pressure 89–94, mean 91.4 (10 readings).

The protocols of the days of highest blood pressure are as follows:

Nov. 5, 1924, resistance. 2.32 p.m. 164–164–162 = 160–158–157–155 = 154–154–153–151 = 152–152–151–151 = 153–152–153–153 = 152 (pulse rate 256).

Dec. 1, 1924, 10.25 a.m. 165–163–160–155 = 152–153–149–150 = 145–142–142–142 = 142–142–141–139 = 138–140–139–135 (pulse rate 200).

The curve from *B* on to the end is representative of the average blood pressure of a good number of the normal rabbits. It compares well also with the curve of D 46 (Fig. 6, in a previous paper (5)) before and after double adrenalectomy, but oscillates about a lower level than D 65 (Fig. 4, in the same paper (5)), also adrenalectomized. Taken alone, this part of the curve has, therefore, little interest. It gains interest, only when brought into relation with the first part of the curve, and particularly when it is remembered that from *B* on two eggs were given daily instead of one. But, interesting as it may be, is it significant? The answer may be split in two according to the meaning to be attached to the word "significant." If by "significant" is understood "fluctuations beyond the range of blood pressure of normal rabbits in general" the answer is immediate and negative, as appears clearly enough from my former work (3, 4) and from the preceding paragraphs, or from another glance at the normal curve of No. 47-0 (Fig. 1). If by "significant" is meant "fluctuations within the normal range, but high relatively to the fluctuations of the pressure in the same animal during a period of observation, more or less short, before the experiment," (and these constitute the large majority of the claims found in the literature), then the answer is very difficult, perhaps impossible. It was stated at the beginning of this article that there is a fallacy in ascribing any rise in blood pressure to the experiment which is being done, because sometimes rises of pressure without known cause were seen in animals employed for observation for a long time. Moreover, D 12 is the only one of five animals in which such an effect is observed. From *A* to *B* the animal consumed 130 egg yolks and it is highly probable, judging from the other four experiments and from Bailey's experience, that at the end of that time there was already a good infiltration of the aorta. So that from the standpoint of the etiological relation of high blood pressure to the development of atherosclerosis, this observation is not a favorable one, and the other four are no better. Although the pulmonary artery is just as frequently and almost as severely involved as the aorta, I have not seen any report claiming an increased blood pressure in the pulmonary artery of these rabbits. Not knowing how to measure this pressure, I shall not discuss it, but clearly an examination of this fact will have to be considered by those who believe in the mechanical theory of atherosclerosis and especially by those who may find in experimental conditions material for their clinicopathological speculations. Whatever it may be, these experiments, positive as far as the production of atherosclerosis of the aorta is concerned, are far from satisfying the criterion I have given for a pathologic high blood pressure in rabbits (4).

Analysis of Van Leersum's Work (*2*).—Van Leersum does not mention the number of animals examined, or the length of time during which they were examined. He contents himself with saying that with his method of measuring the blood pressure (an excellent method, I believe) he has determined "regularly and for a very long time" the blood pressure oscillations of normal rabbits and of rabbits subjected to liver feeding. But there is no explicit mention of the actual oscillations found.

He says that all values were recorded, but the first ones usually were not taken into account; that only a series of 5 values which did not differ very much from one another were considered admissible (page 416), but this procedure is arbitrary and Van Leersum does not justify it. The criterion which guided him is as follows: "Die im Anfang erhaltenen Werthe jedoch sind wegen der Unruhe des Thieres in der Regel weniger gelichmässig: der Blutdruck ist dann oft bedeutend erhöht oder ermässigt. Während des Messens kommen die Thiere aber allmählich zur Ruhe und wird der Blutdruck gleichmässiger." For instance, on page 417, where a sample of a protocol is given, 11 figures (9 a.m.) are discarded, from 182 to 205 cm. water, and 5 retained, from 195 to 199 (average 196); at 2 p.m., 5 figures are discarded, from 163 to 170, and the average of the retained figures is 159. It results from all this, that Van Leersum's curves do not represent the fluctuations of blood pressure of his animals, but are constructed from sets arbitrarily selected among a wide range of values.

He reports the blood pressure of eight animals to which he gave powdered liver and of four more to which he gave sodium taurocholate or sodium glycocholate.

The summary of his results is as follows: No. 87, "average" blood pressure during normal feeding (1 week), 181.6 cm. water; during liver feeding (another week), 194.5 cm. water. No. 57 was given liver every other week. The values (means) as they appear in the graph (Curve 1) are as follows, normal feeding weeks being placed in parentheses: (163) 158 (152) 164 (156) 156 (144) 164 (162), 173 (159) 162 (156) 158. With the exception of 173, which occurs during the liver feeding, all the others are equal to or less than the first figure 163, which happens during a normal period; for differences of 1 cm. of water (less than 1 mm. of Hg) lie within the experimental error. The instance in which the average blood pressure was 173 is less than the average blood pressure of No. 87 during the control period. Rabbit F, early in October, showed an average of 164 cm. (1 week), and in the 1st week of November, still under normal feeding, 180 cm. In the 1st week of liver feeding the average blood pressure was 173, and in the 4th week 181 cm. Rabbit G, early in October, had an average blood pressure of 162.5 cm.; in the 1st week of November, 169 cm. At this time the liver feeding was started. In the 3rd week of this feeding the pressure averaged 171 cm., and in the 6th week 189 cm. water. Van Leersum adds: "Eine Steigung also von gut 11 pCt." But Rabbit F, just mentioned, jumped from 164 to 180 cm., an increase of 10 per cent, without the help of liver feeding. The other four animals may be considered jointly (A, C, D, 23). The averages during the normal feeding period varied between 141 (D) and 172 (C), the observation lasting 1 week for C, 2 for A and D, and irregularly in July, Sept., and Dec., 1911, for No. 23. The maximum weekly average recorded, *during a diet period*, appears as follows: 206 (A), 219 (C), 216 (D), 198 (23) and *after a liver feeding*, 221 (D), in the week following the cessation of the abnormal nourishment.

Van Leersum discusses these findings in the next 3 pages and then comes to the question of what part of the liver is responsible for this effect on the blood pressure. He tries the bile salts (sodium taurocholate and sodium glycocholate) and says: "ihre Wirkung auf den Circulationsapparat ist eine lähmende und sie vermindern den Blutdruck, wie Versuche an vier Kaninchen mich gelehrt haben, in erheblichem Maasse." The protocols are brief but very instructive. I copy from them the pertinent figures. Blood pressure during control period, average, Rabbit Q, 200 cm. water; Rabbit Z, 221; Rabbit X, 208; Rabbit Y, 220. Blood pressure after about 1 month of bile salt (mixed with the food, carrot), average, Rabbit Q, 168 cm. water; Rabbit Z, 190; Rabbit X, 161; Rabbit Y, 182. In other words, figures like 161, which in the main part of the paper are considered normal, are now interpreted as due to the injurious effect of the bile salts, and figures between 200 and 221 which were interpreted before (Rabbits A, C, D, 28) as due to the effect of liver feeding are now considered normal. If Van Leersum had examined more normal animals and for a longer time, before any experiment was undertaken, he would have interpreted his results differently. I venture this statement because my figures for normal animals include *all* of Van Leersum's figures. 122 cm. water (Curve 6, Nov. 2) and 239 cm. water (Curve 2, Jan. 10) are the lowest and highest figures, respectively, recorded in the papers under discussion, that is, about 90 mm. and 176 mm. Hg respectively. In Fig. 2, Rabbit 489, Graph IV of a previous paper (3), there may be seen a few instances of pressure about 180 (averages, since Van Leersum's figures are averages also) and in Figs. 3 and 4, Rabbit 483, Graphs I, II, and III, there are several below 90.

The analysis may be summarized thus: (1) Van Leersum's range for the normal blood pressure in the rabbit, as recorded by his method, is confirmed; (2) Van Leersum's conclusion concerning the influence of a liver diet on the blood pressure of the rabbit is not substantiated by his data, since the fluctuations of blood pressure he obtained do not surpass his own recorded figures for normal animals.

Note on the Results Obtained by Other Methods.—The work of Fahr, Schmidtmann and Schönheimer, together with an account of the methods used by these authors to measure the blood pressure, has been summarized by Shapiro and Seecof (12), who in their own experiments used Anderson's method. I have answered their criticism of Van Leersum's method elsewhere (4). A few remarks, however, may not be out of place here, particularly since a few reports have appeared subsequently to Shapiro and Seecof's.

1. Van Eweyk and Schmidtmann (11) state that the blood pressure of a healthy rabbit, as recorded by their method, lies between 90 and 100 mm. of Hg. They do not say how many animals they have examined or for how long a period. By comparing their method with the values obtained with a Cowl-Gad's manometer, they find an agreement between their figures and the minimal pressure of the "blutige" measurement. But they add: "Zunächst lassen wir es dahingestellt sein, ob diese Übereinstimmung gesetzmäzig ist oder nicht." In Schmidtmann's first report (13) (six rabbits) the blood pressure was taken *once weekly*. The lowest figure is 88 (Protocol IV) before the experiment, the highest 132 (Protocol VI) in the 2nd and 3rd weeks of liver feeding. In her second report (14), she speaks of feeding experiments (diets rich in cholesterol) on 67 rabbits: in some the blood pressure effect was negative, in the large majority, however, she states that the pressure rose to 120–140 mm. Hg for weeks and even months. Then the blood pressure fell to normal, whereas the blood cholesterol remained high. She explains this fall by assuming an injurious effect of cholesterol on the heart muscle and the vascular system. It is difficult to interpret these data because it is not at all clear what is being measured with that method. The authors think that their figures agree with the minimal pressure of the aorta, but have left "undecided" (their words) how legitimate this agreement is. On the other hand, *one* measurement *once* a week gives entirely too little information on the blood pressure of the rabbit, to draw positive conclusions therefrom.

Schönheimer (8) says that he has also found an increase in blood pressure (Schmidtmann's method) under the influence of cholesterol feeding, but he gives no data, except that the largest increase was from 80 to 112 mm. Hg.

Anitschkow (15) says he has confirmed Schmidtmann's results, but gives no details, not even mentioning the method used.

Deicke (16) reports his findings on 88 rabbits fed on cholesterol or liver. He used Schmidtmann's method, but there is no statement as to how often or how many times the blood pressure was measured. Judging from his graphs, the blood pressure was taken *once a week*, sometimes more than a week apart, sometimes less. The normal curve reproduced shows fluctuations between 96 and 110 mm. The highest values represented in his graphs are, in mm. Hg, 131 (Curve 5, liver feeding), 132 (Curve 6, after intravenous injection of cholesterol solution), 133 (Curve 3, enteral cholesterol), 142 (Curve 2, enteral cholesterol).

Thölldte (17) states that the blood pressure of the rabbit, taken *twice daily* for months (Schmidtmann's method, cholesterol feeding) shows no increase "beyond the physiological fluctuations." Unfortunately he gives no details or figures, so that it is not known what is meant by physiological fluctuations.

2. Anderson's method (18) consists in the recording of the pressure necessary to obliterate the pulsation of one of the ear arteries of the rabbit. The greatest error comes, according to Anderson, from the changes in the caliber of the vessel, but he says that they can be largely controlled by keeping the ears warm. The blood pressure, in his normal animals, ranges between 75 and 90 mm. Hg. That the conditions laid down for the measurements must be closely adhered to is obvious from the recent report of Behrens (19), who devised a method essentially the same as Anderson's, and says that the values obtained in this fashion are very constant and lie about 40 mm. Hg. He does not mention Anderson's work, but quotes the work of Kuraya (20), who, independently of him, designed the same method of blood pressure measurements and obtained the same values (mean blood pressure in healthy animals 35–50 mm. Hg). Shapiro and Seecof (12) used Anderson's method and concluded that "the systolic blood pressure of the central artery of the rabbit's ear averages between 75 and 90 mm. Hg as reported by Anderson." In their table the figures for the controls are not essentially different, ranging between 77 and 105 mm. Hg. They fed lanolin to rabbits which had been subjected to various surgical operations (splenectomy, thyroidectomy, double adrenalectomy and combinations of double adrenalectomy with splenectomy or thyroidectomy). The number of blood pressure readings was very small: from 2 to 6 during the whole course of the experiments. The conclusion reached was that there is no significant hypertension during the developmental stage of experimental lanolin atherosclerosis in rabbits.

SUMMARY.

Egg yolk was fed to five rabbits provided with a good carotid loop (Van Leersum's method). The blood pressure was measured daily

before and during the egg feeding until the animal's death. The duration of the experiment varied from 81 to 391 days. The number of eggs consumed varied from 40 to 531. Two animals received, in addition to the egg yolk, lead carbonate by mouth.

Autopsy findings, blood pressure readings and pulse rate are discussed. Two blood pressure graphs are reproduced. The paper contains a brief analysis of Van Leersum's work on liver feeding and a review of the results obtained by other methods of measuring the blood pressure in the rabbit.

CONCLUSIONS.

1. Van Leersum's range for the normal blood pressure in the rabbit, as recorded by his method, is confirmed.
2. Van Leersum's conclusion concerning the influence of a liver diet on the blood pressure of the rabbit is not substantiated by his data, since the fluctuations of blood pressure he obtained do not surpass his own recorded figures for normal animals.
3. Fluctuations of systolic blood pressure beyond the "normal" range are not *necessary* for the production of experimental atherosclerosis of the aorta in rabbits. Inversely, egg yolk feeding experiments in rabbits in which atherosclerosis of varying degree, even extreme, is obtained, are not accompanied by an elevation of blood pressure *outside* the "normal" range.
4. The fluctuations of blood pressure observed during experimental atherosclerosis do not simulate the condition of essential hypertension in man.

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