OBSERVATIONS UPON THE SIZE OF THE SPLEEN.

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By means of a radiographic technique Barcroft, Harris, Orahovats and Weiss(1) inferred that the spleen in the normal animal at rest was usually much larger than the spleen of the same animal post-mortem, and that the change was caused by the discharge of blood from the spleen pulp. If these observations were correct the alterations in the size of the spleen far exceeded delicate changes which demanded the use of plethysmographic apparatus and should be easily visible to the naked eye. Moreover, as pointed out to one of us, the experiments quoted did not absolutely preclude the possibility that the spleen, when it became longer and broader, also became thinner, although there was no reason to believe that this was the case. They were also criticised in conversation on the ground that effects of extra-abdominal pressure on the spleen had not been excluded. These reasons made it desirable to adopt a technique in which the spleen could be observed directly. Such a technique, however, demanded certain conditions proper to the original method, namely, that the animal should be in normal health and spirits, that it should be free from pain and that it should not be under the influence of an anæsthetic.

At first we carried out a number of experiments in animals in which the spleen, though in the abdomen, could be seen through a celluloid window; in the next series we brought the spleen through the muscular layers, so that when the wound healed the spleen was between the body wall and the window; lastly we dispensed with the window, so that the spleen was situated outside the animal, like the testicle. The last of these procedures proved so much the best that the others will be passed over rather lightly.

Granting that the spleen could be placed in such a position as to render it visible, some sort of judgment must be made of the amount of blood which enters or leaves it under given circumstances. It is possible, if the spleen be under a window, to draw a line above the edge of the spleen, marking out on the window the projection of the spleen on the surface of the window. This may easily be transferred to paper and the area of this projection can be measured. The area of this projection of the spleen on the window will be called in this paper the "spleen area." In the case of a spleen which by operation has been rendered extra-cutaneous a piece of celluloid can be laid over the spleen and a similar projection made. This process is of course a rough one and is not susceptible of dealing with small changes in the size of the organ. But when such a tracing is made, is it possible to discover a relation, even if only approximate, between arterial variation in the bulk of the spleen and consequent variations in the two-dimensional "spleen area"? Amongst the relevant considerations are the following:

(i) If the actual shape of the spleen did not alter, if the increase was uniform in all directions the volume would be proportional to the cube of the square root of the area.

(ii) If the spleen does not increase in thickness to the extent which it increases in length and breadth, cubing the square root of the area will give an exaggerated idea of the change in volume.

(iii) On the other hand it is evident from looking at the spleen that when it becomes distended its angles disappear and its surfaces become rounded. The effect which such changes in shape can produce may be exemplified by an extreme case. Suppose a regular tetrahedron be viewed from above and that its base has an area A; suppose further that the tetrahedron be magnified till its base has an area $2A$: its volume will have increased in the ratio $\frac{(2A)^{\frac{3}{2}}}{A^{\frac{3}{2}}}$. So that if the original area were 100 and the new area 200, the volume would be respectively as 1000 to 2800 (approximately). But now suppose that the new tetrahedron is turned into a sphere, the surface of which passes through the points of the tetrahedron, the volume of the sphere will be of the order of 18,000, or eighteen times the volume of the original tetrahedron. With an irregular object like the spleen, which can alter not only in size, and in the convexity of its surfaces, but also in the convexity of the whole organ (which in the dog and cat is C-shaped), the problem becomes more difficult still. As it seemed less hopeless to obtain a relation between the "spleen area" and the spleen volume by calibration than by calculation, resort was had to injecting blood into the excised spleen and comparing the weight of the spleen with the "spleen area." Four such experiments were performed, two in the dog and two in the cat. All the splenic vessels were carefully tied except the main splenic vein, into which the injection was made; into this a cannula was placed. The animal was bled and its blood defibrinated; the spleen was then rapidly taken out, weighed and placed on the surface of the animal (as the extra-cutaneous spleen would be); blood in known quantities was injected with a syringe and was controlled by weighing the spleen at intervals. Tracings were taken and the "spleen area" observed after each injection. The procedure was varied a little in different experiments but it was of the same general character in all except the last. In that the dog injected its own spleen. The vessels were tied except the main artery and vein. A "bulldog" forceps was placed on the artery. Cannulæ were placed in the two convergent branches of the splenic vein close to where they emerge from the spleen. The nerves were destroyed. When the "bulldog" was removed from the artery, the spleen swelled up to a great size (of which more will be said later). The spleen without loss of blood was cut out, weighed, and then the blood allowed to gush out of the cannula in the veins. It was collected in measuring cylinders. The spleen was also weighed and tracings taken. The volume measurements and the weighings agreed on the assumption that the specific gravity of dogs' blood was 106. The result of these experiments was to show that the total weight of the spleen was almost exactly proportional to the square of the "spleen area." In one case the actual change was slightly below that calculated; in the other three it was above, and above to a rather greater extent. On the whole, therefore, if the gain or loss is computed as being proportional to the change in the square of the spleen area, the quantity of blood entering or leaving the spleen is likely rather to be under- than to be over-estimated.

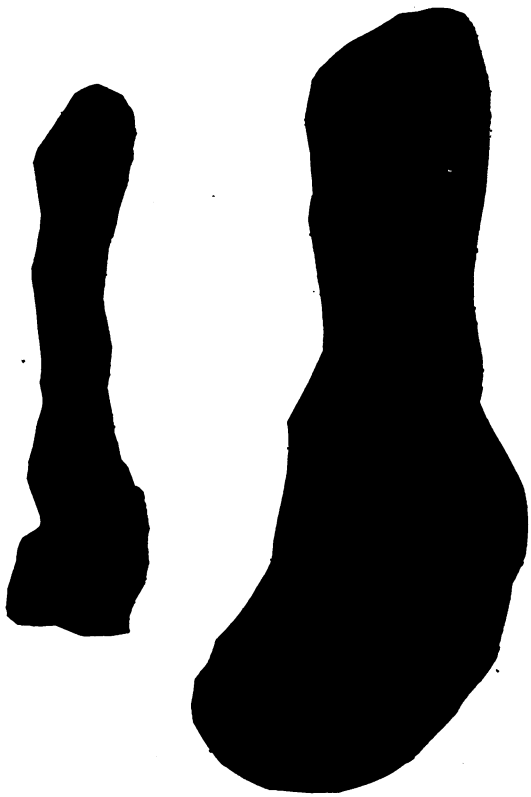
The following example will serve: the weight of the spleen at the end of the experiment was 46 grm. as found by actually weighing the organ. 28 grm. of blood has just been injected (last figure in column 3); it may be presumed that the weight previously was 18 grm. (last figure in column 2 carried back to penultimate in column 4). Comparing the squares of the areas before and after the 28 grm. were added, these areas were found to be in the ratio of 19 to 46, as the weight after the addition of blood was 46 grm. and the computed weight before the addition of the blood was 19 grm. (last figure in column 1). In the case of the last observation but one, the organ was presumed to weigh 18 grm. after a measured quantity of 30 grm. had been withdrawn from it, therefore the weight before the withdrawal was presumed to be 48 grm. The ratio of the square of the surface before the 30 grm. were withdrawn to the square of the surface at the end of the experiment was as 47 : 46 and so on.

1	2		3		4
Weight of spleen computed as 46 × $(\text{surface})^2 \div (\text{surface when weight is 46 grm.})^2$	Weight of spleen arrived at by adding the sum of the weights : blood injections to 46 grm.		Weight of blood injected in each observation		Weight after injection of blood in column 3
29	24	+	5	=	29
32	29	+	16	=	46
46	45	+	52	=	97
88	97	−	10	=	87
80	87	+	21	=	108
91	108	−	60	=	48
47	48	−	30	=	18
19	18	+	28	=	46

All weights in grams.

The most unsatisfactory feature of the above experiments is that the animals used were not those in which the spleen had been outside the body wall for some days or weeks. An attempt was made to calibrate such a spleen, but the numerous anastomoses with the vessels of the body wall, as well as additional ones in the abdomen, rendered the experiment abortive. In calculating the amount of blood gained or lost by the spleens in our experiments we have assumed that the law which governs their relation of spleen area to spleen volume is the same as that in the spleens which we calibrated and that the change in volume is approximately as the square of the change in area.

The extreme limits of the dimensions of the spleen. The figures given above show a four-fold alteration in the size of the spleen. The question naturally arises: what are the limits within which the spleen is capable of varying? In this connection the following comparison is of interest. Two dogs of approximately the same size (16 and 18 kilos. respectively) were used. The first was killed by bleeding. The spleen weighed 33 grm. The second dog, whose spleen to all appearance was approximately the same size to start with, was the subject of an experiment already described (p. 3) in which the nerves were cut and the veins tied. The spleen attained a weight of 365 grm. or eleven times that of the first animal. Probably even so great a weight falls short of the extreme capability of the spleen, for the region of the solar plexus had been much handled and no doubt the arterial pressure was very low. Fig. 1 shows the relative sizes of these two spleens. Other organs of the body are not susceptible of such changes, or indeed of changes of volume at all comparable. Probably the penis is the only organ which can suffer a tenfold increase in volume. Physiologically speaking there seems no reason why the word erectile should not be applied to the spleen. The above experiment is confirmed by another in which the spleen of a 20 kilo. dog attained a computed size of 360 grm. under similar circumstances. In the first case (that of the 18 kilo. dog in which the spleen was actually



33 grams. 59 sq. cm. | 365 grams. 194 sq. cm.

Fig. 1.

weighed when at its largest) the dog probably contained 1·6–1·8 kilos. of blood in all: of which 340 c.c. at least was in the spleen, which held therefore about one-fifth of the entire blood of the animal.

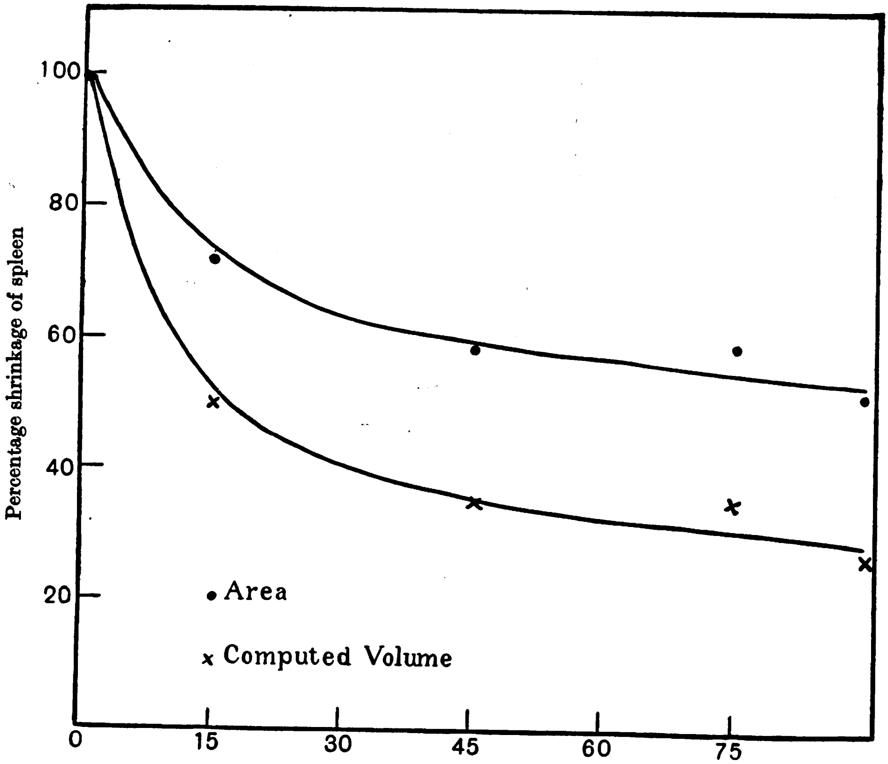
The technique used for the celluloid window experiments need not be referred to in detail; it was that of Katsch and Borchers(2) as modified by Florey and Carleton(3) and Stephens and Florey(4). We also introduced some modifications which seemed important at the time. The series, however, culminated in our discarding the window and merely observing the spleen when situated outside the skin, as will shortly be described. The general results of experiments obtained with the celluloid windows will be found in a preliminary communication in the *Lancet*, 13 March, 1926, also in the *Ergebnisse der Physiologie*, xxv, 1926. The following additional points only may be quoted before passing to the extra-cutaneous spleen.

(*a*) *Exercise on treadmill.* These results are probably accurate to within 5 or 10 p.c.

Cat	1	2	3
Area of spleen, rest	15	16	34 sq. cm.
Area of spleen, exercise	8·5	9	21 sq. cm.
Computed weight, rest	—	—	34 grm.
Computed weight, exercise	—	—	14·8 grm.

Cat 3 post-mortem weight of spleen 8·57 grm., area 16 sq. cm.

(*b*) *Hæmorrhage.* The effect of hæmorrhage is to produce an immediate and approximately uniform diminution in the volume of the spleen. Fig. 2 was obtained by withdrawing 15 c.c. samples of blood from



Hæmorrhage in c.c.

Fig. 2. Effect of hæmorrhage on volume of spleen.

the carotid artery of a cat (under urethane) at ten minute intervals, and represents the percentage diminution in the area and calculated mass of the cat's spleen as the result of such successive hæmorrhages. The result is in close agreement with that of Barcroft, Harris, Orahovats and Weiss(1), namely, that the effect is most marked at the onset of hæmorrhage.

Pneumonia. The onset of pulmonary infection as judged by stethoscopic evidence was always signalised by a gradual shrinkage of the spleen. This progressed steadily for a few days until a reduction in volume of about one-third to one-half had occurred, after which the volume remained almost constant until death.

No reliable conclusions could be drawn from the two cases as to the effects of sepsis at the site of the window, although it seemed that no volume change whatever had occurred.

Administration of oxygen had the effect of brightening the colour of the spleen in pneumonia, in which disease the spleen is dark. Normally administration of oxygen does not affect the colour of the spleen.

Death. The post-mortem spleen irrespective of the mode of death was in all cases much smaller than that during life. Fig. 3 is a photograph of the actual post-mortem spleen placed over the paper template of the same spleen during life one hour previously. The animal in this case was the same one as that employed for Fig. 4, but after it had died of hæmorrhage. This represents a shrinkage to one-third of the volume during life and emphasises the fallacy of visualising the living spleen as being the same-size as the dead one.

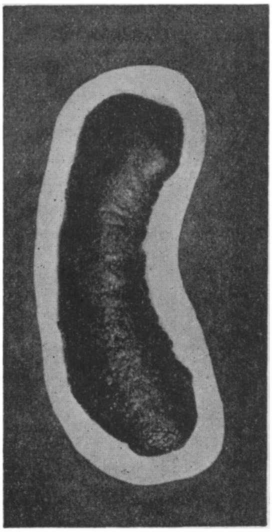


Fig. 3.

THE EXTRA-CUTANEOUS SPLEEN.

Although experimental animals into which celluloid windows have been inserted exhibit a surprising freedom of movement and are apparently very little embarrassed by the presence of the window, nevertheless great difficulties are encountered in endeavouring to make observations over periods of time greater than about two weeks, principally due to the formation of fibrinous material on the inner surface of the window. These difficulties are partly eliminated by the employment of a removable window. On the whole, however, the movable window only succeeded in a minority of cases.

We therefore turned to the extra-cutaneous spleen. It was found possible to bring the spleen through a slit in the abdominal wall and to fix it uncovered outside the skin. The spleen thus retains all of its original vascular, nervous and mesenteric attachments and yet grows as a protrusion outside the body of the animal, just as an ear grows as a protrusion outside the head. The only covering for some weeks is the peritoneal investment which undergoes some degree of thickening and fibrosis. Ultimately epithelium grows over the spleen from the edges.

Sepsis does not occur. Lavage with warm eusol every second day or thereabouts, and lubrication with melted vaseline, suffice to keep the spleen for some months as a cutaneous appendage. Observations were thus made on one animal over a period of three months until terminated during a trial demonstration at Stockholm by an automobile accident. Another dog had his spleen outside his skin for eight months.

Detail of operative procedure. The operation has been performed on cats and dogs, but dogs are the more suitable.

A linear incision about four inches long is made through the shaved and sterilised skin of the animal about one and a half inches below the costal margin. This does not coincide with the long axis of the spleen, but it is necessary to draw the spleen as far towards the cephalic end of the dog as possible, in order that the hind limb may not rub against the organ in running and jumping. The extreme mobility of the spleen in most cases permits of this procedure without difficulty.

A shorter incision is then carried through the entire abdominal musculature, the spleen identified, and the length of its mesenteric attachment estimated. The incision through the muscles may now be enlarged so that the resulting slit will accommodate the mesentery of the spleen. The spleen is then drawn through the aperture, and a series of thick cat-gut stitches inserted, firstly through the entire fascial, muscular and peritoneal layers, then through the splenic mesentery, avoiding the blood vessels, next through the peritoneum, muscle and superficial fascia of the opposite side and finally returned and tied. The tying of these stitches calls for some judgment. Too great laxity will permit a hernial protrusion of viscera; too great pressure on the splenic vessels will cause œdema or infarction of the spleen. Moreover, allowance must be made for some degree of post-operative œdema of the muscle. If the stitches are tied fairly loosely, however, the lips of the incision just touching the mesentery, these difficulties will be satisfactorily avoided.

The retracted skin is best fixed by a continuous plain cat-gut suture around the margins of the incision. In this way the spleen exists in a mushroom-like fashion with a median linear attachment to the animal. If a bare area of superficial fascia is allowed to remain underneath the spleen adhesions are inevitable and the borders of the spleen will be obscured by exuberant granulations, as was found to be the case in our first experiment.

Post-operative treatment. The spleen is apparently entirely non-sensitive; the animal will roll on its spleen without concern, and it therefore becomes necessary to cover the organ with a dressing lubricated with vaseline in order to avoid accidental damage. To prevent the animal from licking or biting its spleen, too, some form of covering is necessary except when actual observations upon running, etc., are being conducted, and the dog should be muzzled when not under observation.

The best method of dressing so far devised has been the following. The spleen is first thoroughly irrigated with warm eusol, then anointed with vaseline previously melted and cooled, and then covered with oil-silk sterilised by boiling.

A thick pad of cotton wool is advisable to prevent injury to the spleen, and this may be bandaged in position. Some form of jacket to prevent access of the tongue is necessary until the dog has grown accustomed to the dressing. New dressings are applied every second or third day.

The method of recording and its accuracy. Fig. 4, to be discussed later,

Fig. 4.

illustrates the superficial area of the external surface of the spleen, *i.e.* the projection of the spleen as seen through a piece of celluloid placed over it. In taking the records the area covering the spleen is marked out on the celluloid with a "grease pencil." The celluloid is then removed and the tracing is transferred to a piece of paper either by pressing the paper on to the celluloid, when the mark adheres partly to the paper, or by transmitting light through the celluloid and paper, and marking the paper in ink over the mark on the celluloid. The area is then measured with a planimeter.

In order to show the error as between several successive readings of the same spleen in the resting condition, the following were taken in quick succession.

Three examples are given:

I. 45, 48, 45, 45, 46 sq. cm. dispersion 7 p.c.
II. 44·5, 44, 44, 45 sq. cm. dispersion 3 p.c.
III. 40, 41, 40, 42 sq. cm. dispersion 5 p.c.

It may be taken that any observed alteration of over 10 p.c. represents a real change in the size of the spleen.

There is a personal equation involved in this measurement of the "surface" which the spleen presents. This equation depends upon the amount of pressure put upon the celluloid when in contact with the spleen. As between two persons it is very constant, at least it was so as between Barcroft and Stephens and the readings are all given, in terms of Barcroft, a correction of 14 p.c. being applied to Stephens' readings. This correction is of course small relatively to the changes in size of which the spleen is capable.

In every case in which the animal has been killed, post-mortems have been carried out and histological sections of the spleen have been made. The post-mortems have not shown any hypertrophy of accessory spleens and the histological preparations have revealed little that is abnormal in the interior of the organ.

On the surface there is a thick deposit of white fibrous tissue and in the case of experiments of longest duration this has been overgrown by an invasion with stratified epithelium from the edges. The spleen has ceased strictly speaking to be extra-cutaneous.

The muscular trabeculæ in some cases give the impression of hypertrophy and certainly there is relatively more muscular tissue than in normal spleens. It is not clear, however, that any real hypertrophy has taken place. Quite probably the shrinkage in the spleen may have taken place at the expense of the spleen pulp rather than the muscle and that the muscle may therefore have got rather "bunched up" and the trabeculæ thickened in that way. There is no real evidence that the spleen contained a greater number of grams of unstriated muscle at the end than at the beginning. For the rest the organ was normal in character.

The size of the spleen. Immediately after the operation the spleen is frequently very large and shrinks considerably in the first few days; when in this condition it must be regarded as hyperæmic, though the cause of this hyperæmia is obscure, there being no evidence of undue pressure on the veins. Nevertheless it is not claimed that the spleen would be so large inside the animal as it is outside during the early days after the operation. This period, however, soon passes off, the spleen acquires a healthy colour and from that point observations on the effect of exercise may be undertaken. After the initial dilatation has passed off the spleen of the resting animal changes little from day to day, but if observed over long intervals of time it is found to be shrinking slowly. The experiment of longest duration so far lasted over eight months, during all of which time the animal appeared to be in perfect health and excellent spirits. Nevertheless the spleen underwent a slow shrinkage, as shown by Fig. 4.

The cause of this gradual diminution in size cannot be definitely stated. As histological preparations have revealed nothing save a covering of scar tissue, one tends to take refuge in the phrase that it is the nature of cicatricial tissue to contract, and there to leave the matter.

Yet there are some other facts which may not be devoid of importance. Not all spleens shrink at the same rate. All dogs which have undergone the operation acquire a reduced hæmoglobin value in their blood. The question arises, is there any relation between the degree of anæmia and the rate of shrinkage of the spleen? These experiments are still being carried out, but the facts may be stated graphically (Fig. 5). They are all taken from observations made during the first six weeks or so after the operation. Ultimately any relation, apparent or real, would probably disappear, because at the end of, say, two months or less the anæmia would probably tend to pass off, but the spleen does not enlarge, if only because it is encapsulated by a dense connective tissue covering.

At the present juncture one must guard against the assumption that because two phenomena can be plotted on the same piece of paper and a curve produced, there is any real causal relation between them. All that the figure shows is that simultaneously two progressive phenomena are taking place, the anæmia and the shrinkage; whether or not they are related can only be decided by experiment, *i.e.* by discovering whether by imposing one factor on the animal, the other factor necessarily follows.

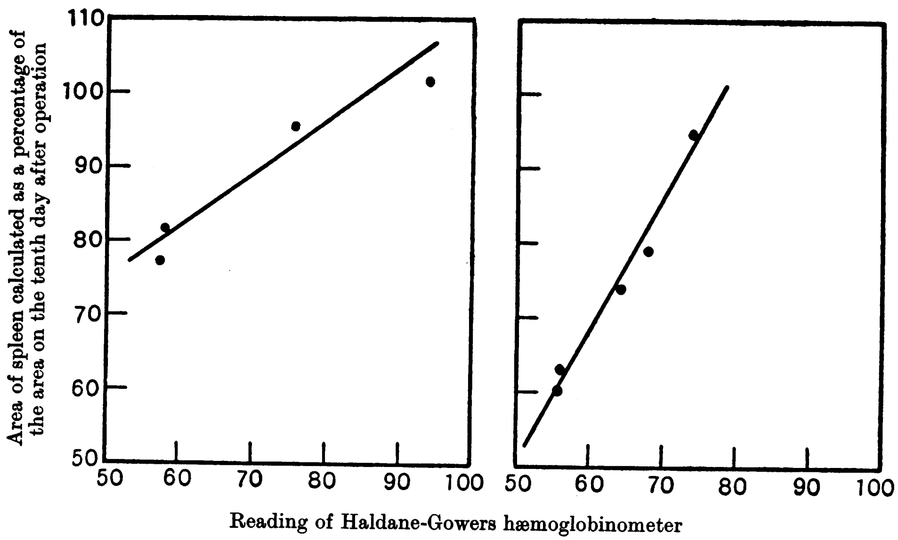


Fig. 5.

This much, however, can be said, namely, that the animals which have become anæmic most rapidly are those in which the spleen has shrunk most rapidly. That merely has been our experience gleaned from the animals which drew our attention to the phenomenon. The subject now demands, and is receiving, systematic work, to be reported on a subsequent occasion.

Relation of area to volume. Within the limits shown above, the size of the resting spleen over short ranges of time is remarkably constant. The result shown earlier in this paper, namely, that the change in volume varies almost exactly as the square of the spleen area applies to the extra-cutaneous spleen; at all events, the changes in volume of the extra-cutaneous spleen are not exaggerated if computed as being proportional to the difference in the square of the areas. Some caution must be used if the changes considered involve long periods of time.

Exercise.

Three forms of exercise have been used:

(1) Running on the level.

(2) Swimming.

(3) Running on the staircase.

The first dog of the present series, "Pongo," was a small Scottish terrier who was easily trained to run about the laboratory grounds after a bicycle, being full of spirit. Our greatest difficulty with her was to get her in a sufficiently quiet mood ever to give us a tracing of what we could regard as a spleen with the animal at rest.

A course of 2000 metres was marked out; the usual time taken for her to run over the course was six minutes. Naturally these figures are very rough, because she did not keep precisely to the course and the time was subject to slight variations.

Inspection of the spleen was even more striking than measurement. It was evident that the shrinkage in thickness was not less than that in length and breadth, and that the spleen which at rest had been deep red became quite pale when discharged. Taking the weights above for what they are worth, the greatest amount of blood which the spleen discharged on exercise was 74 grm. This dog was about four kilos. in weight; its blood volume may therefore have been about 400 c.c. The spleen then discharged 18·5 p.c. of the volume of the blood. There is reason, however (partly on the basis of Cruickshank's, and of Scheunert and Kryzwanek's(13) work, and partly on the basis of work shortly to be published), to believe that the corpuscle value of the blood discharged from the spleen is greater than that in the general circulation. At times the disparity is about 3 : 2 but on the average about 6 : 5. Using the latter figure 74 c.c. of spleen blood would amount to about 86 c.c. of ordinary blood. This calculation would show that about one-fifth of the corpuscles in the body can be held in the spleen and expelled into the blood during exercise.

Effect of running.

Exp.	Date from operation (days)	Distance run (metres)	Computed weight of spleen (grm.) Before	After	Difference
I.	28	500	60	31	29
II.	33	300	66	34	32
III.	32	2000	104	30	64
IV.	34	2000	68	40	28
V.	59	2000	42	12	30
VI.	77	2000	35	16	19

Probable blood volume = 400 c.c.

One suggestive point in the above table is shown by a comparison of experiments II and IV. The sizes of these were much the same at the commencement of the exercise and the same was true of the end, so that there was an equal diminution in size, and that between roughly the same limits; yet the amount of exercise taken in the two experiments was very different. In experiment II the animal ran 300 metres, in experiment IV 2000 metres. The point is noted here, it will be considered later on.

Not every dog takes kindly to running after a bicycle, and one which did not was the next on which we operated. It was necessary to find some form of exercise for her about which she had no option. Having a large tank in the laboratory animal house we turned it into a swimming bath. Its depth was such that when full she could not touch the bottom. Warm water was laid on so as to preclude the possibility of change in the size of the spleen being due to contact with cold water. Also the spleen was supported, which appeared at the time to be an important point. We thought that possibly the act of galloping might cause an irritation to the unsupported organ to which it might respond by contraction. To a strong swimmer the amount of exercise involved in paddling about in a swimming bath appears to be very trivial, but it can easily be increased by tying a suitable weight to the dog. Most of the experiments were carried out at between 34° C. and 37° C., but no great difference appeared in the result even if the water was not heated.

The following table gives some data for the dog which yielded the results on running tabulated above. Some data are given also for another dog which show that the changes in size of the first were not exceptional.

Effect of swimming.

	Date from operation (days)	Temp. of water °C.	Duration of swim (min.)	Computed weight of spleen (grm.)		
				Before	After	Difference
"Pongo"	28	15	3	68	38	30
	31	37	3	74	38	36
	36	36	3	48	24	24
	60	34	6	38	23	15
"Trixie"	10	34	6	152	88	64
	—	34	6	99	49	50
	18	34	10	93	49	44
	25	34	10	61	40	21

Here again the duration of the exercise—as between three and six minutes—did not seem to have any marked effect.

On the other hand the mere fact of being in the bath had little effect upon the spleen because that organ did not shrink, or it only shrank slightly if the water was shallow enough to allow of the dog touching the bottom with her feet.

Effect of swimming.

No.	Days after operation	Time (min.)	Computed weight of spleen (grm.) Before	After	Difference
			Back feet touching bottom		
1	22	4	63	63	0
2	—	4	50	40	10
3*	—	7	50	34	16
			Swimming		
4	28	3	68	38	30
5	36	3	48	24	24

* Water deeper in 3 than in 2.

Staircase experiments. The best subject which we have had for research is a retriever bitch of 14 kilos. The particular advantage which she has over any dog of the terrier type is that she will remain perfectly quiet when told to do so. Thus she will lie on the table with but little attention and, once she understood what to do, would run up or down the laboratory stairs without wishing to wander off into the passages. The total height of the staircase is 14·2 metres, there are 89 steps.

The changes shown in the spleen of this dog after a run upstairs, which took only a very short time, were even more conspicuous than those of the other two animals which we have described. At the bottom of the stairs her spleen was like a somewhat flattened sausage, and was red or purplish in colour according to circumstances. At the top it was pale and flat—almost concave—so that the tracings only give a partial idea of the weight of evidence which shows the extent to which the spleen discharged its contents.

Dog	Day after operation	Exercise	Dimensions of spleen: Before, Area of tracing	Before, Computed weight	After, Area of tracing	After, Computed weight
N	35	Once up staircase	42	102	34	68
	40	Once up staircase	41	98	30·4	54
	47	Once up staircase	41	98	29	42
	43	Once down and up	36	74	20	23
	19	Once down and up	44	104	36	76

The computed loss of blood on these occasions was very nearly the same, namely 22, 27, 30 and 31 grm. respectively.

Only three experiments are given above; in point of fact the routine was gone through many times, the alteration to the eye being sufficient to make the demonstration a most telling one. It is not possible to make any calculation of value as to the relation of the amount of blood expelled to the total blood volume, because on the thirteenth day after the operation the dog removed the dressing in one place and bit off a large portion of the spleen, which appears to be quite insensitive either to tactile or painful stimuli.

At the end of eight months, the spleen, though small, showed the same proportionate contraction as earlier, namely to about two-thirds its area, or about one-half its weight, as running up the stairs.

It shrank also on the animal's being bled to death to about the same, or to a somewhat greater extent.

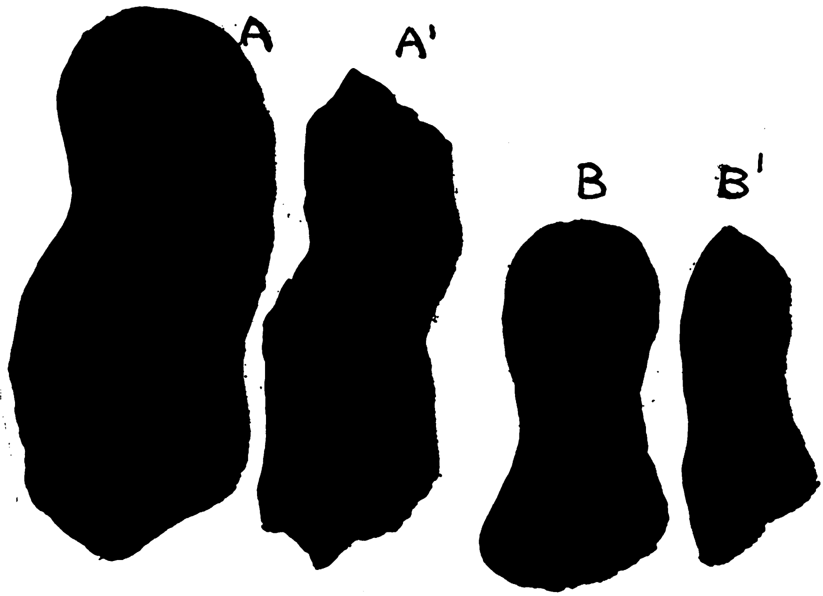


Fig. 6. *A* rest. *A′* after ascent of staircase, 29 October, 1926.
B rest. *B′* after ascent of staircase, 8 May, 1927.

Relation between the amount of exercise and the amount of contraction. On the whole the general rule holds that the greater the amount of exercise, the greater the amount of contraction; thus in the foregoing tables, a longer swimming effort or a longer running course usually produced a greater degree of evacuation of the spleen. The striking point, however, is that the response to very mild exercise is proportionately greater than to severe exercise.

Thus to casual inspection the degree of contraction and pallor of the spleen caused by running up the staircase, involving roughly 200 kilogram-metres of work against gravity differed little from the corresponding effects of running down the staircase in the case of one dog (Nigger); in the case of another (Pongo) a run of 70 metres from one end of the laboratory passage and back caused a computed shrinkage of volume of from 74 to 38 grm. whilst a run of 200 metres after a bicycle only produced shrinkage of from 74 to 28 grm.

The conclusion seems to be that in exercise as in hæmorrhage the most marked effect relatively is in the initial stage, that expulsion of blood into the circulation is far from being a final effort on the part of the organism to adjust the volume of the circulating blood to that of the bed in which the blood circulates; it is in fact a fine adjustment and it can be done without, like most fine adjustments.

Mechanism of the splenic contraction with exercise. Hargis and Mann(6), in a most convincing research, demonstrated the sensitiveness of the spleen to nervous influences in the unanæsthetised animal. de Boer and Carrol(7) showed that the contraction of the spleen which was induced by administration of carbon monoxide, was operated from the central nervous system; in an animal to which the spleen was attached by the nerves only, it contracted when the animal inhaled the gas: the spleen, however, received its blood from a perfusion circuit and introduction of CO into the blood did not lead to contraction of the organ. The contraction caused by exercise is also of nervous origin, or at least principally so. The alteration in size is not marked when the nervous connections are severed. A striking demonstration is furnished by severing all the nerves which run along those branches of the splenic artery that supply the ventral and larger portion of the organ, whilst the nerves which supply the dorsal portion are left intact. This method of demonstration was suggested to us by Dr Anrep. We are indebted to Dr Lim(14) also for sending an account of a very similar experiment performed by him. Our experience has been that on exercise the denervated portion suffers a very slight decrease in area with a certain increase in flabbiness, whilst the intact portion contracts, as does the ordinary spleen, to about two-thirds of its area, during the ordinary staircase experiment. Such a contraction would mean halving the volume.

Fig. 7 shows the effect of one ascent of the Laboratory staircase by a dog in which the top portion of the spleen was innervated normally and the lower portion denervated (as far as is known, for the dog being still alive, no post-mortem has been done on the spleen). The thin horizontal line between *A* and *B* separates the innervated and denervated areas, so far as can be seen. The following are the areas of the two before and after exercise:

	A	*B*	Total
Before exercise	12·5	21·5	34 sq. cm.
	8·5	20·5	29

It appears likely that there is, either due to a lowering of the pressure in the portal vein, or for some other reason, a certain small loss of blood by even the denervated spleen, but that the cause of the principal expulsion of blood in the normal organ is active contraction of the spleen due to nervous excitation. Exactly to what extent this nervous mechanism plays on the splenic musculature as opposed to the musculature of the splenic vessels is uncertain and possibly differs in different spleens: the spleens of some animals contain much more muscle than those of others. The hard consistency of the contracted spleen suggests, however, that the splenic musculature is heavily involved, though that does not exclude the vaso-motor mechanism from participating. Indeed it may be that there is no rigid distinction between the one and the other; the whole musculature of the spleen may with some reason be regarded as of a piece with that of the splenic vessels (the splenic veins are richly endowed with muscle). The principal evidence against such a view is based on the action of drugs, pituitrin acting, according to de Boer and Carroll(8), on the vasomotor mechanism and not on the spleen muscle. Such evidence does not seem to suggest more than that some differentiation has taken place. It does not seem to prove any fundamental disparity in origin. It has been shown by several authors (Bayliss(9), Masuda(10) and Hoet(11)) that the spleen dilates on stimulation of the central end of the depressor; here again there is no evidence as to how far the musculature of the organ as opposed to that of its vessels was involved.

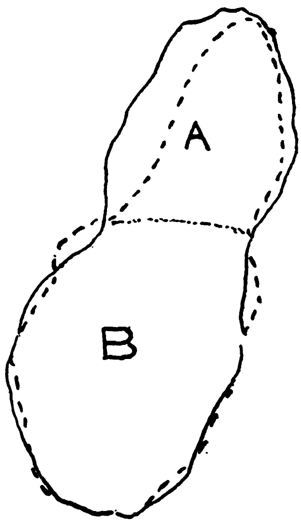


Fig. 7. Rest ———. After ascent of staircase - - - - -.

The speculation that the splenic musculature may be typical of the vasomotor musculature of the visceral area generally leads to the general consideration of the responses of that area to anoxæmic conditions and to the inducement of oxygen want by exercise. That field is too wide to justify exploration on the basis of our present experiments.

So far as the spleen is concerned the investigation of exercise was undertaken because exercise presented a particular form of oxygen want, but a form to meet which the body has become very highly specialised; it is not surprising therefore to find that, as in the cases of the pulse and the respiration, the nervous control of the spleen has learned to anticipate the actual exercise and the spleen will respond to the type of mental agitation which if pushed would culminate in physical violence. Two examples can be furnished: (1) The first is from the dog mentioned above, one half of the spleen of which was denervated. This animal became greatly attached to its keeper whom it always was anxious to follow. If when it was lying on the table in the hands of another attendant the keeper left the room, the denervated portion of the spleen shrank so that the picture approached but did not quite attain to that of the dotted line in Fig. 7. On being released it bounded off the table after the keeper. This reaction could be depended upon absolutely to take place. (2) A dog of an extremely jealous disposition. While she lay on the table her spleen would normally remain of constant area, but if her kennel companion were fondled by the keeper in her presence, her spleen would become smaller and grow pale.

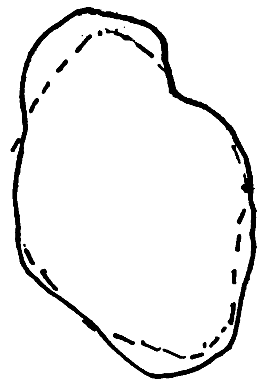


Fig. 8. Size at rest ———. Excited by fondling a rival - - - - - -.

The change in size is shown in Fig. 8.

THE SIGNIFICANCE OF THE SPLENIC CONTRACTION DURING EXERCISE.

(*a*) *In relation to the vascular bed.* The question cannot fail to be asked: to what extent does it matter whether either the blood volume or the volume of circulating hæmoglobin receives a considerable augmentation when exercise is taken? The discussion is a difficult one owing to our almost complete incompetence to express the importance of the factors involved in quantitative terms. We know for instance that if exercise be taken the "vascular bed" in the region exercised increases in volume. If it is a matter of importance (and within unknown limits it must be) that the volume of blood should bear some relation to the capacity of the bed in which it lies, then there would be some merit in having a reservoir of blood outside the circulation which could adjust the volume of circulating blood to the capactiy of the vascular bed. It could then enable the vascular areas in the active organs to open up without unduly employing those in other places. It seems probable that a 20 p.c. variation in the blood volume would go a great way in carrying out these adjustments, though of course it is known that in active exercise other areas than the spleen are drawn upon, *e.g.* the kidney. The body does in fact provide blood for one organ at the expense of another. If in the most active exercise of which an animal is capable, the kidney may be deprived of blood to the extent of being actually injured, how desirable is it that in lesser exercise some store should exist which may be drawn upon without imposing a tax on such important an organ as the kidney.

In a great struggle it is desirable that the animal should overcome as the result of muscular strength, even though a transient albuminuria accompanies the victory. The muscles must have the last ounce of blood which they can get, but on lesser occasions the vital organs may count themselves fortunate that before their blood supply need be drawn upon the spleen can produce a considerable quantity.

(*b*) *In relation to the minute volume of circulating blood.* In the heart lung preparation, the simple way of increasing the output of the heart is to take a jug full of blood and pour the contents into the open venous reservoir. But with a closed circulation blood can only be put into the heart in increased quantity at the expense of what is circulating and therefore prejudices the position for the period of time immediately following.

The spleen pulp, however, being unlike other organs outside the circulation, is, in effect, such a jug. The blood expelled along the splenic vein when the spleen contracts is blood added to the circulating fluid. That may be admitted, but it does not furnish an answer either way as to whether such an action on the part of the spleen is on a scale sufficient appreciably to influence the circulation rate. Nor can an answer be given in our opinion, for it depends upon factors as yet undetermined; the most that can be done is to state the problem more definitely by a numerical example which involves quantities of possibly a reasonable order.

The following example probably flatters the power of the spleen.

Suppose (1) that the blood volume of a man is five litres, (2) that he can expel one litre of blood from his spleen, (3) that this can be expelled in 12 seconds, (4) that his minute volume of blood is five litres at rest. In that case one litre of venus blood reaches the heart in 12 seconds. Starting from zero time, suppose the spleen contracts during 12 seconds and adds a litre of blood, so that two litres of blood reach the heart, one from the general circulation and one from the spleen. The venous inflow to the heart during that time will be doubled, and the arterial outflow during 12 seconds will be doubled. If during that 12 seconds the vascular conditions in the body become so readjusted that the venous inflow during the next 12 seconds (*i.e.* the 13th to the 24th second) becomes doubled because the arterial output is doubled during seconds 1–12, the whole circulation rate will have become doubled until something takes place to alter that condition of affairs. The power of the spleen then depends not merely on the amount of blood which the spleen can contribute relatively to that in the body, but upon the rate at which the blood from the spleen can be thrust into the venous system and also the rate at which a fresh cyclic equilibrium can be established in the circulatory system. These factors are unknown.

Even could the events just set forth take place in the times and on the scale stated, the blood flow would only double. That is a much smaller increase than takes place during active exercise, but a twofold increase forms an appreciable proportion of the whole and if it occurred would bring the splenic contraction within the group of factors which when integrated accounted for the total increase in the circulation rate.

SUMMARY.

1. Methods for placing a celluloid window over the spleen have given fairly satisfactory results for about a fortnight.

2. Much better is the method of making the spleen extra-cutaneous; as such it has been kept under observation for more than eight months. During that time it underwent a gradual diminution in size. Its function remained unimpaired except in so far as functional changes bore a constant proportion to its size.

3. Both methods show that the spleen contracts to about one-half to one-third of its size on exercise and to an even smaller volume on death or severe hæmorrhage.

4. The amount of blood squeezed out during exercise is estimated at its maximum as forming one-fifth of the volume of blood in circulation; the spleen under such circumstances becomes pale.

5. The events described in paragraphs 3 and 4 (above) depend on the integrity of the nervous supply of the spleen.

6. Psychological processes calculated to culminate in violence may also cause the spleen to become pale and to contract somewhat, thus anticipating actual exercise. In any case the contraction of the spleen is

an early event in both hæmorrhage and exercise, rather than a last resort for the acquisition of blood.

7. The significance of the splenic contraction on exercise is discussed (*a*) with regard to the relation of the blood volume to the vascular bed, (*b*) with regard to augmentation of the minute volume of circulating blood.

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REFERENCES.

1. Barcroft, Harris, Orahovats and Weiss. This Journal, 60. p. 443. 1925.
2. Katsch and Borchers. Zeit. f. Exp. Path. und Therap. 12. p. 225. 1913.
3. Florey and Carleton. Journ. of Path. and Bact. 29. p. 98. 1926.
4. Stephens and Florey. Brit. J. Exp. Path. 6. p. 269. 1925.
5. Barcroft. Preliminary Communication. Lancet, p. 544. 1926. See also Ergebnisse der Physiol. 25. p. 88 1926.
6. Hargis and Mann. Amer. Journ. Physiol. 66. p. 116. 1924
7. de Boer and Carrol. This Journal, 59. p. 302. 1924.
8. de Boer and Carrol. This Journal, 59. p. 381. 1924.
9. Bayliss. Quoted in Starling's Principles of Human Physiology, 4th ed. p. 849.
10. Masuda. This Journal, 62. p. 289. 1927.
11. Hoet. Communicated privately.
12. Krogh. Skand. Ariv. 27. p. 227. 1912.
13. Scheunert and Kryzwanek. Pflüger's Archiv, 213. p. 204. 1926.
14. Lim and How communicated this work to the British Medical Association Meeting, Edinburgh, July, 1927. They found a greater effect on exercise than we, which they attributed to adrenalin secretion affecting the denervated portion. Their exercise was much more severe.

For more complete literature see Krumbhaar, Physiol. Reviews, 6. p. 160. 1926, and Emil v. Skramlik, Ergebnisse der Biologie, 2. p. 505. 1927.