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Physiological Studies

(Second Series)

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Physiological Studies

(Second Series)

By

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[*From the* PROCEEDINGS OF THE ROYAL SOCIETY, B. Vol. 90]

The Mechanism and Control of Fibrillation in the Mammalian Heart.

By Professor J. A. MACWILLIAM, F.R.S.

(Received June 6, 1918.)

The results of the present investigation are founded on a very extended study of the subject, carried on from time to time during the past 30 years, in the course of very numerous experiments (hundreds) on the mammalian heart.

These results establish the conclusion that in fibrillation there is an essential change in the manner of conduction of the excitation process in the cardiac musculature; the relation of this change to the excitability of the muscle determines the appearance and characters of the different forms of "fibrillar" action that may be observed. The conduction of the excitation is essentially altered, inasmuch as it is propagated along the muscular fibre systems or fasciculi, instead of travelling directly through the muscular substance, without obvious regard to the arrangement of the fibres, as in the normal beat of the heart.* Fascicular dissociation is an essential feature of fibrillation, which is, strictly speaking, a condition of "fasciculation" rather than "fibrillation." The essential change in conduction may be induced in very different ways. The state of fibrillation is rendered persistent by a disturbance in the normal relations of conduction time and refractory period in the cardiac musculature, resulting in the establishment of a mechanism of circulating excitations.

The cat's heart was the one most largely investigated, but those of rabbits, guinea-pigs, rats, etc., were also employed. The heart action was usually examined and recorded with the thorax open, while artificial respiration, by means of a pump or by continuous insufflation of the lungs with oxygen, was maintained. A myocardiograph of the type described by Cushny† was employed, arterial blood pressure or pulse being often registered at the same time. Intra-cardiac pressure records were often made from the auricles and the ventricles on the principles described by Frank. Anæsthesia was maintained by chloroform, ether, urethane, morphia, chloretone, paraldehyde, or combinations of these. In a number of experiments the method of decapitation was used. The perfused heart was frequently utilised, records being

* See the electrocardiographic evidence advanced by Lewis and Rothschild, 'Phil. Trans.,' vol. 206, p. 181 (1915).

† 'Heart,' vol. 2, p. 1 (1910–11).

made by (*a*) the myocardiograph, used in the same way as with the heart *in situ*, and (*b*) by a rubber bag placed in the left ventricle and connected with a Hürthle manometer, the system being filled with liquid. All the tracings are to be read from left to right; they are all ventricular (L.V. of cat) records except where otherwise noted.* The time is shown in seconds.

For the more accurate use of faradic currents, a Kronecker's inductorium was employed, with two volts in the primary circuit; the values of the units stated are to be taken as obtained with this E.M.F. in each case. For obtaining series of shocks at different rates, a Brodie cut-out arrangement was used, giving either make or break shocks at regular intervals; these shocks were recorded on the tracings by an electrical signal. The shocks were often applied through the myocardiograph, so that they traversed a considerable amount of the cardiac substance; at other times they were sent through electrodes about 1 mm. apart, etc.

The Conduction of the Excitation in Fibrillation.

Instead of travelling uniformly right through the mass of muscle without evident regard to the direction of the fasciculi or bands of muscle, as under normal conditions, the excitation wave in fibrillation travels most easily along the complexly-arranged fasciculi, there being an impairment or failure of propagation at most of the inter-fascicular connections. Such a mode of propagation of the rapidly-recurring contraction waves may be clearly perceived on direct inspection of the heart, and on palpation of the ventricles the apical portion being held between the finger and thumb with varying degrees of light pressure. In the latter case, instead of the normal uniform hardening of the muscular wall at systole, there is a striking want of synchronism in the hardening of the constituent fasciculi, short contraction waves in rapid succession hardening different sets of fibres, while others are relaxed and soft, the contracted ones momentarily standing out and giving a characteristic "wiry" feeling among the quiescent fasciculi; the impression of an incessant turmoil of dissociated or in-coördinated activity is a vivid one. The myocardiograph record shows a series of rapid irregular oscillations, varying to some extent from place to place in rate and in range of excursion. Similar records are obtained from the perfused heart.

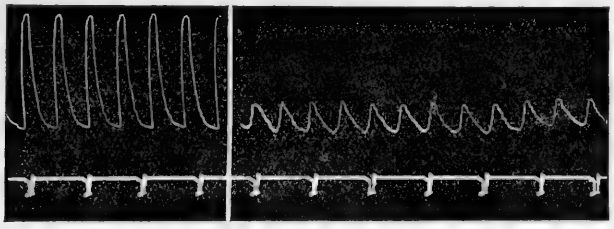
The failure of normal conduction may be induced in two ways: (1) by depressing agencies acting directly on conductivity, and causing more or less extensive blocking in the most susceptible parts, the inter-fascicular junctions, while the intra-fascicular connections remain functional. This effect may be produced even with a moderate or slow succession of

* Upward movement of the ventricular lever = systole.

contractions, but is greatly favoured by rapidity of sequence of the contractions. Such depressing agencies are of various kinds—cooling, intra-vascular injection of potassium salts, bile, over-doses of many drugs, etc., including some substances that are in suitable doses useful as remedial agents promoting recovery from fibrillation; (2) by excessive rapidity of excitation, *e.g.*, by electrical stimulation. This (2) may be the sole cause of the alteration in conduction, or it may co-operate with a depressing influence acting directly on conduction, *i.e.* a combination of (1) and (2) is specially effective.

Change in Mode of Conduction due to Direct Depression.

Fibrillar Beats.—That depression of conductivity is of fundamental importance is evidenced by the fact that individual beats may be "fibrillar" in character (fig. 1). This is strikingly realised on palpation; instead of the



A. B.

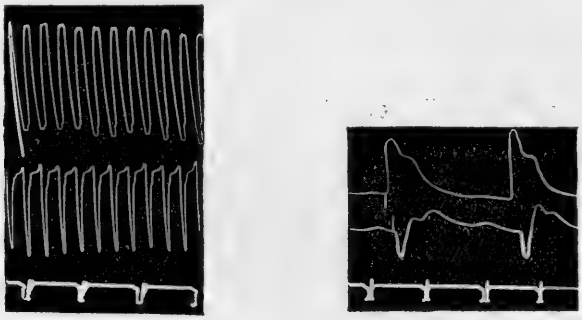
FIG. 1.—The systolic movement of the lever is upward. A = normal beats. B = fibrillar beats, which are strikingly wiry on palpation.

usual sensation of uniform hardening at each systole, the contraction is felt to be passing in asynchronous fashion along the different systems of fasciculi or bands of fibres, some feeling firm and contracted, with the characteristic wiry feeling, while others are soft and relaxed. On the surface of the ventricles the contraction wave is visibly slowed, and in the auricles this may be very strikingly evident in its progress over the muscle.* In this condition the nature of the ventricular beat is similar, whether it occurs in response to an impulse travelling down the A–V. conducting system, or is excited by a direct stimulus applied to the outer surface of the ventricles. The fascicular dissociation is evident even when the impulse is distributed through the endings of the Purkinje system of fibres (fig. 2).

Fibrillar beats are often able to give considerable excursions of the recording lever, and they are often able to pump out a very appreciable amount of blood into the aorta. The contraction and relaxation phases are

* In the ventricles waves can often be plainly seen entering at or emerging from the vortex.

both prolonged; the systolic power is relatively small. The individual beats are quite discrete; there is a very definite interval, varying in duration, of



A. B.

FIG. 2.—A shows normal curves, the upper one ventricular (systolic movement upward) and the lower auricular (systolic movement downward). B shows two fibrillar beats at a later phase of the experiment. Simultaneous points are marked by short vertical lines at the first beat. The Au. and V. contract together; the excitation apparently originates in the A–V. junctional tissues.

complete quiescence between them (fig. 2). The excitability of the cardiac muscle is low when such separate beats are present; the refractory period is long. The occurrence of these fibrillar beats shows that the "fibrillar" mode of contraction is not essentially dependent on or necessarily associated with rapidity of succession at all, though the latter is a very striking feature of typical "fibrillation," giving complexity of movement, complete in-coördination, and mechanical ineffectiveness as regards expulsive power.

Continuous Series of Fibrillar Beats as seen in a More Excitable Heart.

When the excitability is at a higher level, or when stimulation is applied to make the fibrillar beats follow one another more quickly, a continuous succession of contraction waves appears; one fibrillar beat excites another, and they are thus strung in a series, constituting a slow coarse fibrillation (fig. 3). The rate depends on the excitability of the muscle, the degree of

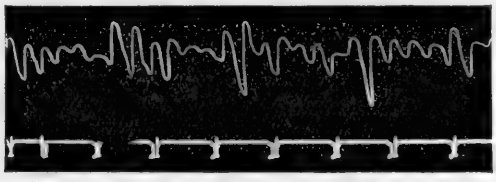


FIG. 3.—Continuous irregular series of fibrillar beats, each beat exciting a subsequent one through the mechanism of circulating excitation. An overdose (intra-vascular) of sodium carbonate induced this condition.

dissociation varies with the rate of succession—the faster the rate the higher the grade of dissociation. In some cases the depression of conduction may be of such a degree that a beat coming after a long interval may show no distinct sign of dissociation by inspection or palpation, whereas, when a quick series occurs, each beat is markedly dissociated, giving the characteristic " wiry " feeling on palpation (fig. 4). When the excitability of such a

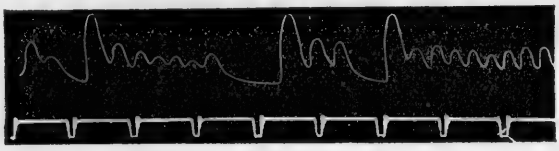


FIG. 4.—The quick series of beats are fibrillar in character. The larger beats coming after long intervals do not show evidence (on palpation) of that character.

heart gradually rises, *e.g.*, under the influence of massage, improved nutrition, certain remedial drugs, removal of depressing influences, etc., the rate of continuous movement may increase, with an accompanying increase in the grade of dissociation.

There is a very definite gradation from (*a*) the phase of discrete fibrillar beats, through (*b*) slow and then quicker series of successive contraction waves, up to (*c*) the rapid and mechanically ineffective oscillations of typical fibrillation. The increase in rate depends on the augmented responsiveness of the more excitable muscle. The degree of asynchronism or dissociation increases with the rise in the rate of succession, the partial blocking between the larger fasciculi or bands and layers of fibres giving the lower grade of dissociation seen in slow coarse fibrillation, while the higher grades of dissociation between fasciculi are present in the condition of rapid fine fibrillation.

Similarly with diminishing excitability and conductivity, a downward gradation may be observed from typical fibrillation, through grades of slower and coarser fibrillation, to the phase of individual fibrillar beats.

Change in Mode of Conduction Due to Excessive Rapidity of Excitation.

When the rate of beat is excessively accelerated by a series of induction shocks of increasing rapidity, a gradation of changes is observable as the rate of succession rises. The individual contractions become briefer and gradually give smaller and smaller excursions of the recording lever. Inspection shows evidence of dissociation becoming very pronounced at the higher rates, so as to bear a close resemblance to the familiar appearance of the ventricular surface in typical fibrillation. Palpation at the same time reveals increasing degrees of asynchronism as the rate rises, until the characteristic wiry

wriggling feeling, practically indistinguishable from that of true fibrillation, becomes very marked, instead of the solid push normally given to the palpating finger. These phenomena are obviously due to the rapid series of short contraction waves traversing, at relatively slowed rates, the various layers, bands or fasciculi of the ventricular musculature according to the lower or higher grades of inter-fascicular blocking and dissociation that are present, thus giving asynchronous contractions at different parts of the thickness of the muscular walls. These changes in their various grades are attended by related degrees of lowering of the arterial pressure, and by auricular acceleration and irregularity. At high rates the force and range of the contractions become small, the output from the ventricles is cut down and a great fall of arterial pressure results.

When the rapidly stimulated ventricles have been brought into the condition above described—presenting many features of resemblance to true fibrillation but not identical in mechanism as will be explained later—diminishing rates of excitation are attended by graded changes of converse order—slower succession of contractions, less dissociation, quicker conduction, apparent coarsening of the oscillations and a gradual return, as the rate falls, to the characters of normal beats.

Pseudo-fibrillation and Fibrillation.

The above-described condition into which the ventricles may be brought by rapidity of excitation (graduated series of shocks or faradic currents of suitable strength) short of the rate necessary to induce true fibrillation, may for convenience be termed pseudo-fibrillation (figs. 5 and 6). As regards the evidence afforded by inspection, palpation, tracings of the oscillations, fall of blood-pressure, etc., the two conditions may be difficult or impossible of distinction, but they differ strikingly as regards persistence; pseudo-fibrillation ceases immediately or at varying short periods after the cessation of the stimulation, while true fibrillation in ordinary circumstances, in the absence of remedial measures, goes on as a rule to the death of the heart. (The duration of pseudo-fibrillation after cessation of the stimulation varies according to the excitability of the stimulated area, the strength and duration of the stimulating current, etc.) The difference depends on the fact that in true fibrillation a mechanism of circulating excitation has been established, whereas in pseudo-fibrillation this is not so. The latter condition depends on the emanation of an excessively rapid series of excitation waves from the area of stimulation; these short waves travelling at reduced speed over the interlaced fasciculi give rise to the condition described. But as soon as the issue of excitations from the stimulated area ceases, the disturbance ceases and the conditions revert to the

normal. The pseudo-fibrillation at once ceases when the stimulated area is disconnected from the rest of the muscle, *e.g.*, by forcible clamping, etc., or

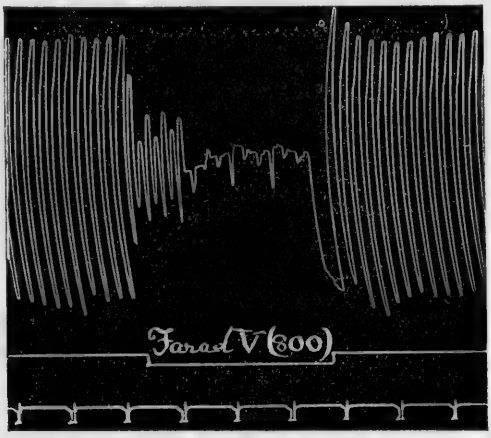


FIG. 5.—Rabbit's heart (R.V.). Faradisation with 800 units induced first a rapid tachycardia, then pseudo-fibrillation which promptly stops at the end of the faradisation. A blood-pressure record taken at the same time showed a great fall, with minute oscillations showing on the tracing.

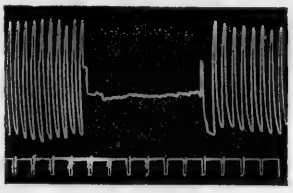


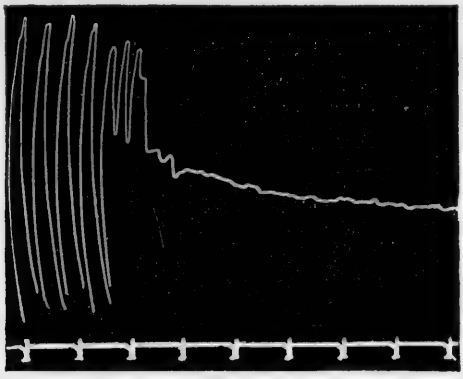
FIG. 6.—Pseudo-fibrillation induced almost immediately in fully developed form by faradisation ; it ends with a larger oscillation when the stimulation ceases.

when it is cut off—as may be done in the perfused heart—or when it is rapidly cooled. In pseudo-fibrillation there has not been established in the mass of the muscle outside the stimulated region a mechanism which ensures the continuance of the movement after the impulses emanating from the excited area have ceased or have been excluded—in striking contrast to what holds good in the case of true fibrillation. This method of differentiating between pseudo-fibrillation and fibrillation may be more easily applied in the case of the auricles, by isolation of the appendix after the stimulation has been applied to the tip.

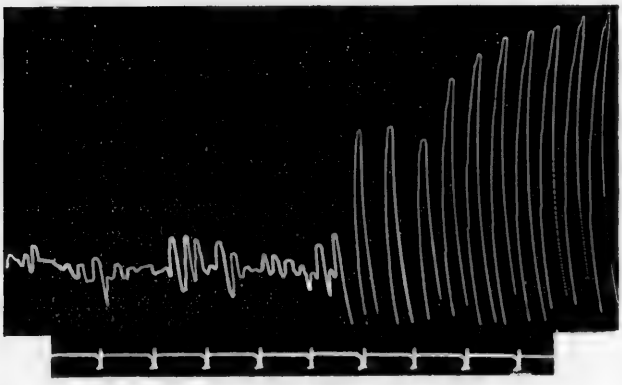
Mode of Recovery from Fibrillation.

When the ventricles are recovering from the state of typical fibrillation, with the aid of massage and of drugs, as stated later, the oscillations visible

on the surface become more vigorous and clearly much coarser, the dissociation becoming much less fine and larger groups of fasciculi contracting together; there is evidently an extension of conduction through inter-fascicular junctions that were formerly blocked. On palpation the muscular substance feels of good tone, and the gradation from fineness to coarseness of fibrillation is very clearly realised—the sensation of universal turmoil due to the fine rapid dissociated twitchings throughout the ventricular walls grading into more vigorous contraction waves of coarser type, and these again into beats giving the normal feeling of uniform hardening of the muscle (figs. 7 and 8).



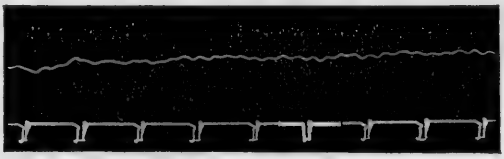
A.



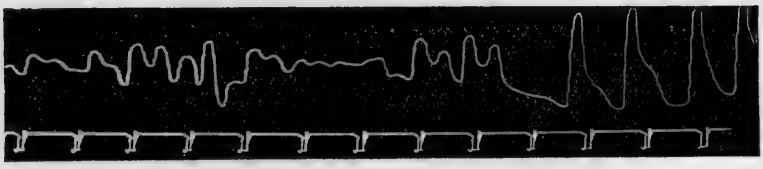
B.

Fig. 7.—Spontaneous recovery from fibrillation in 30 seconds, preceded by coarsening of the fibrillar movement. Urethane, 2·5 grm., had been given hypodermically, in addition to chloroform. In A, the fibrillation was caused by shocks sent into the ventricle at the rate of 480 per minute. A brief tachycardia precedes the fibrillation. In B, recovery is seen, preceded by slower and coarser oscillations.

In the case of a heart which is showing individual fibrillar beats of the nature already described the process of recovery under the influence of



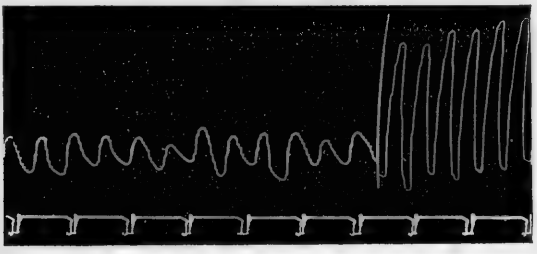
A.



B.

FIG. 8.—R.V. recorded. Fibrillation from application of faradic current (200 units). A is taken 7 seconds after beginning of fibrillation. B shows recovery occurring after fibrillation had lasted 75 seconds, massage being done at intervals. Adrenaline, 0·27 mgrm., had been injected previously, and this probably favoured recovery. Marked coarsening of the movement (followed by a long pause) is seen prior to recovery.

massage, removal of depressing influences, etc., is usually a more elaborate one. The phase of slow coarse fibrillation has to be passed through, with a gradual increase in the rate and the grade of dissociation as excitability is restored; this leads up to the condition of rapid fine fibrillation—from which recovery occurs in the fashion stated above. But treatment with certain doses of adrenaline, etc., may sometimes change the fibrillar beats into co-ordinated ones without a transition through the various phases just enumerated (fig. 9).



A. B.

FIG. 9.—A shows slow coarse fibrillation—a series of irregular fibrillar beats. B is taken shortly after the injection of 0·2 mgrm. adrenalin into the L.V. (1 in 5000 solution used). The fibrillar beats are changed into normal ones.

The coarsening of the rapid oscillation in the process of recovery is quite different from a coarse slow movement that is not on the way to recovery at all and where the muscle is lax and feeble. It is also different from the apparent coarsening with slowing of the oscillations in the graphic record due, as direct inspection of the heart shows, to irregular summation of fine feeble twitchings which are present with a high degree of dissociation and which may gradually become weakened to extinction. It is important to correlate the information derived from (*a*) inspection, (*b*) palpation, and (*c*) graphic records.

Rates of Stimulation Necessary to Establish the Mechanism of Circulating Excitations.

With excitable ventricles in good condition high rates of excitation by induction shocks of moderate strength are necessary to overpass the phase of pseudo-fibrillation and induce true fibrillation, *e.g.*, single induction shocks at rates of 450–500 per minute are commonly effective, but the duration of the application of the series of shocks has an influence in this respect; with longer application lower rates may suffice. When faradic currents are employed the current has to be of such a strength and duration as to raise the rate of responsive contractions to about the above rates. Beyond such rates the state of pseudo-fibrillation is not as a rule maintained, but gives place to true fibrillation as soon as the mechanism of circulating excitation has been established, this point being often recognisable on the tracing by a change from the rapid and more or less irregular curves of small excursion that are present during rapid tachycardia or pseudo-fibrillation to the much smaller and entirely irregular oscillations of true fibrillation (fig. 11).

The conductivity of the muscle plays an essential part in regard to the rate of stimulation needed to cause fibrillation; the necessary rate is not a constant or absolute one, but varies much in relation to the state of the conductivity at the time. The lower the conducting power, the lower is the rate of stimulation required to establish the circulating mechanism, since under these conditions the normal relations between conduction time and refractory period are more readily upset, a relatively low grade of acceleration sufficing to cause slowed excitation waves to reach different parts of the fascicular systems after the refractory period is over in these situations. Agents that depress conduction, *e.g.*, potassium salts, bile, cooling, etc., can be used in such a way and to such a degree as not to induce fibrillation by themselves, but to render the muscle prone to fibrillate with unusually low rates of excitation. Thus the minimal rate of stimulation which induces true fibrillation affords an indication of the state of conductivity. In

conditions of greatly depressed conduction power stimuli not faster than rates commonly seen when the heart is beating in co-ordinated fashion may cause fibrillation. The rate of oscillation when fibrillation is established in such hearts is naturally a slow one, as the excitability is commonly reduced as well as the conductivity.

In such conditions of depressed ventricular conductivity, it is sometimes, though rarely, possible to excite ventricular fibrillation by faradisation of the auricles or of the sino-auricular junction in the region of the S.A. node. Such a result has been quite definitely obtained in a very few cases. The A–V. conducting mechanism was apparently able to transmit a series of impulses to the ventricles sufficient to excite in the latter the relatively low degree of acceleration necessary, in presence of their lowered conductivity, to establish the circulating mechanism.

Rates of Oscillation in Fibrillation.

As has been stated, the rates of oscillation are usually high when fibrillation is induced, and they remain high for some time; if massage is employed, quick oscillation may be maintained for an hour or more. But when, in the absence of massage, etc., the excitability of the muscle becomes lowered, as happens even with massage after a variable time under the usual experimental conditions—the rate of oscillation falls markedly, the less excitable muscle being unable to give such rapid responses to the circulating excitations. And in conditions where the excitability is depressed when fibrillation is induced, the rate of oscillation is, from the beginning, very much slower than usual; such rates as about 280, 250, 240, 140, etc., being seen, *i.e.* rates sometimes below the rhythm of a normally-beating heart when acting rapidly. It must be noted that the graphic records of the oscillations have to be interpreted with caution. For the oscillations caused by contraction waves coursing along the interlaced fasciculi are very complex and irregular and do not denote the succession of contractions in any one fasciculus. Still, the rates observed are, within certain limits, quite definite and significant, though on account of the irregularity precise figures may not be obtainable. Such records must be controlled by the methods of inspection and palpation, and, as a rule, yield results that are in accordance with the evidence afforded by the latter methods.

Influence of Duration of Stimulation.

When electrical stimulation, *e.g.*, faradisation, is used to excite fibrillation, its efficiency shows a marked relation to the duration of its application, as well as to the strength of the current; a longer application, *e.g.*, 10 seconds,

may elicit persistent fibrillation when a shorter one, *e.g.*, 3 seconds, only causes a rapid tachycardia or pseudo-fibrillation. The greater effect of the more prolonged application may be ascribed to at least two factors:—

1. The time needed for the current to produce its full effect in the way of acceleration of the succession of contractions. With suitable strengths of current, the tracings clearly show an increasing acceleration for some little time after the beginning of the application, the excursions become more rapid and smaller until, when the circulating mechanism is established, fibrillation supervenes with its very irregular oscillations. With strong currents the characters of fibrillation may become manifest in the tracing immediately or almost immediately. It is evident that, with relatively weak currents, some time is needed to get up the full rate, with its influence in promoting fibrillation by shortening the refractory period and slowing and impairing the propagation of the excitations.

2. A continuance for some time of the rapid succession of contractions may be assumed to promote fatigue in the more vulnerable parts of the inter-fascicular connections (in analogy to what is known of fatigue of the A–V. conducting mechanism) by an unduly early repetition of an impulse to be conducted. Continuance of the stimulating current after the circulating mechanism has been established seems to be of no importance.

Parallelism between Auricles and Ventricles.

There are close analogies between the behaviour of the auricular and the ventricular muscular systems as regards (1) the occurrence of single contraction waves passing slowly through the muscle, constituting fibrillar beats in the ventricles, and (2) the development of (*a*) regular tachycardias, (*b*) irregular tachycardias, (*c*) pseudo-fibrillation, and (*d*) fibrillation, as results of graduated artificial stimulation.

The persistence or non-persistence of fibrillar movements is clearly explicable on the same principles in both auricles and ventricles—by the altered relation between conduction and refractory period—and the mode of conduction in fibrillation is, as in the ventricles, a fascicular one, depending on the presence of more or less extensive blocking in the inter-fascicular connections. Slow coarse fibrillation may be seen in the auricles as in the ventricles, and separate waves of contraction sweeping over the auricles in irregular fashion, more or less resembling what have been described as fibrillar beats in the ventricles, are often very striking in conditions of depressed conductivity; the progress of the greatly slowed wave can be followed by the eye with the greatest ease. And, with some increase of excitability, the wave of excitation may excite another, just as in the

ventricles, and so set up a continuous slow series—slow here also because of obviously depressed excitability, as shown by diminished readiness to respond to stimuli of definite strengths.

Pseudo-Fibrillation and Fibrillation in the Auricles.

Under gradually increasing electrical stimulation, the auricles, like the ventricles, show higher and higher grades of disturbance: (1) extra-systoles, (2) regular tachycardia, (3) irregular tachycardia, (4) pseudo-fibrillation, and, at least in certain conditions of the auricular muscle, (5) fibrillation. The gradually increasing rate of auricular response rises through the grades of tachycardia or flutter, with diminishing range of lever excursions, up to a condition of rapid tremulous movement (pseudo-fibrillation), with irregular succession and range of oscillations more or less closely approximating to the characters of true fibrillation and often hard to distinguish with certainty from the latter, either by inspection of the auricles or in the tracings, though in pseudo-fibrillation the oscillations are commonly larger and of a less high grade of irregularity than in fibrillation. The movement may last for variable periods after the stimulation has been discontinued.

A ready method of discriminating between the two conditions is afforded by the experiment of isolating the stimulated area (by clamping, etc.). Tachycardia or pseudo-fibrillation is at once arrested, while true fibrillation is not affected.

In the majority of the animals examined special conditions are necessary in the auricular muscle for the production of true fibrillation with its essential mechanism by faradisation, etc., the stimulation *per se* is not, as a rule, sufficient in the easier conditions of quick conduction normally present in the auricles. Contractions in very rapid sequence, *e.g.*, 500–600 or more per minute, may be excited without establishing the mechanism of persistent fibrillation. Certain conditions involving an alteration of conductivity without a great lowering of excitability, are often effective in determining the occurrence of fibrillation, *e.g.*, vagus influence, defective blood supply, certain phases in the action of some drugs, such as chloroform, paraldehyde, pilocarpine, etc.

"Spontaneous" fibrillation, *i.e.* when the precise exciting cause cannot be defined, depends no doubt on the presence of irritation *plus* an altered state of conductivity. The latter is sometimes supplied, under experimental conditions, by the tonic influence of the vagus centre exercised through either the right or the left vagus, as can be seen when only one nerve is intact; section of the nerve in such cases is speedily followed by recovery from fibrillation which may have persisted during the whole preceding part of the

experiment, or at least since the heart was exposed. Such vagus control has not appeared as a common cause of auricular fibrillation in these experiments, but in some instances its influence has been unmistakable.

The simplest and most easily available method of producing true auricular fibrillation for a time is by a combination of electrical stimulation and vagus stimulation. Rapid tachycardia set up by electrical stimulation is converted by vagus influence into true fibrillation which persists as long as the vagus influence is maintained in sufficient strength to provide the condition in the auricular musculature necessary for the keeping up of circulating excitation; the fibrillation so excited goes on under vagus influence long after the electrical stimulation has been discontinued; the latter may indeed have been applied only for a second or two. Under vagus influence the fibrillation oscillations, though very rapid, become greatly weakened, the irregular movements of the recording lever becoming minute. With pretty strong vagus control this weakening may go on to invisibility, so that the auricles look entirely quiescent, even when their surface is scrutinised with a lens. As the vagus influence wears off during prolonged stimulation of the nerve, very fine fibrillation oscillations again begin to become perceptible, and these gradually gain in vigour and range until after a variable time the normal type of beat replaces the fibrillation movement.

A similar sequence of events, more quickly passed through, is evident when vagus stimulation is diminished or discontinued instead of the influence of the nerve being allowed to wear off during continued stimulation. What evidently occurs in these cases when the auricles become motionless under vagus influence, is that the mechanism of circulating excitation goes on working in spite of the inhibitory influence which cuts down the mechanical response to invisibility; there is no true inhibition of the essential mechanism of fibrillation.

The experiment may be done in another way. Instead of first exciting the tachycardia and then stimulating the vagus, the latter may be brought into action first so as to reduce the auricles to complete quiescence; during this period an electrical current is applied briefly (*e.g.*, for one or two seconds) to the auricle; a fine tremulous (fibrillation) movement of small range may at once appear and continue until the vagus influence wanes or is discontinued.

Mechanism of Circulating Excitations without Contractions.

But if the vagus is strongly inhibiting the muscle when the electrical current is briefly applied, there may be no visible effect at all; the auricles remain perfectly motionless until the vagus control has become weakened, when the fine tremulous movement usually appears and gradually gains in

vigour as in the former experiment, after a time giving place to normal action. What has happened in this case is that the electrical stimulation, falling within the period of vagus influence, is effective in setting up the mechanism of circular excitation, while the latter finds no expression in contractile movement on account of the mechanical response to excitation being kept in abeyance by the vagus inhibitory power. When the latter wanes and the mechanical response again becomes manifest, the circulating excitations are attended by the circulating contractions of visible fibrillation.

When the electrical stimulation is applied in the foregoing way without apparent effect on the inhibited auricles, the subsequent appearance and development of fibrillation as described above is not affected by the stimulated area (*e.g.*, auricular appendix) being isolated from the rest of the auricle shortly after the brief application of the stimulating current and while the auricles are still kept in complete quiescence by the vagus; the subsequent fibrillation involves the whole of the auricular muscle, apart from the isolated area. It is plain that the mechanism of excitation necessary for fibrillation has been established in the mass of the auricular muscle, and that it is independent of a continued emission of impulses from the stimulated area—now isolated. In these experiments the isolation was effected (*a*) by clamping off or (*b*) by section, after a weak clip or a ligature not too tightly drawn had been applied along the base of the appendix to prevent hæmorrhage. In some cases rapid cooling of the stimulated area was employed instead of isolation. Control experiments were made to determine that the methods used do not themselves cause fibrillation in the conditions present, under vagus influence, etc.* The vagus evidently can act more strongly on auricular contraction force, if not also on conductivity, than on excitability, for the latter property must remain functional (though depressed) in auricles that respond by subsequently manifested fibrillation movements to an electrical stimulus applied during the period of mechanical quiescence of the muscle.

As a rule, as stated above, the auricular muscle is not sufficiently depressed by vagus influence to prevent excitation occurring in response to adequate stimulation, or to stop the circulation of excitations once this mechanism has been established, though the normally-associated mechanical response may be cut down to the point of invisibility. But in some instances the vagus seems to be able to act so strongly on excitability that after electrical stimulation during the vagus period, fibrillation does not gradually appear in the usual way as the vagus control is passing off, but visible action recommences

* Under certain conditions it is clear that mechanical stimulation may sometimes excite auricular fibrillation.

in the form of slowed auricular *beats*. This is to be ascribed to the vagus acting more strongly than usual on excitability, in addition to the usual effects on contraction force and conductivity.

When the influence of the vagus in converting a rapid tachycardia or flutter into fibrillation was first studied, the question naturally arose as to whether the changes visible on inspection and in the graphic records might not be due simply to the cutting down of the force of the rapidly-recurring contractions, the mechanical limitation of the range of movement associated with distension of the auricular chambers, etc. But the clamping-off experiment brings out there is an essential difference in the mechanisms in the two cases.

The vagus alters or depresses conductivity in the auricles in such a way that the inter-fascicular connections are unable to functionate normally when the succession of excitations is much accelerated. (Distinct from this is the question of the power of the vagus to slow the conduction along the main transmitting paths in the auricles.) Certain other depressant agencies have an influence on the inter-fascicular connections in the ventricles (already described), which resembles that of the vagus in the auricles, and these agencies, when acting in great intensity, may have the further result of causing obvious and striking retardation in the passage of the contraction wave both in the ventricles and the auricles, even when the sequence is not a rapid one, but may indeed be slower than the normal.

Some Differences in the Behaviour of Auricles and Ventricles.

While the analogies between the various phenomena are very close in the auricles and ventricles, certain points of difference may be noted.

1. Electrical stimulation of strength adequate to give a sufficiently excessive rate of beat is, by itself, a ready means of exciting ventricular fibrillation, though, as has been stated, the addition of some influence depressing conductivity causes fibrillation to develop when the rate of beat is not nearly so rapid as would otherwise be required. Auricular fibrillation, on the other hand, is not, in most cases when the heart is in good condition, excited by electrical stimulation *per se*, but requires an alteration of conductivity (in the sense already defined) by some other agency, *e.g.*, vagus influence, defective nutrition, toxic substances, etc. The reason of this difference is probably to be found in conduction being less easily upset in the auricles with their simpler structure and easier conditions of rapid conduction, as compared with the highly elaborate ventricular architecture with the much slower rate of conduction in the ventricular muscle proper—apart from the Purkinje system.

2. The relation of the vagus to fibrillation is quite different in auricles and ventricles; in the auricles the vagus favours fibrillation in the presence of some irritation, *e.g.*, electrical stimulation; in the ventricles vagus influence can often be clearly shown to retard or prevent fibrillation, while not able to remove the latter once it has been established. The difference is due to the stronger action of the vagus on conductivity than on excitability, as a rule, in the auricles; this naturally promotes fibrillation. In the ventricles, on the other hand, in regard to these two properties, the main, if not the sole, incidence of the vagus influence is on excitability; this, of course, tends to repress the development of fibrillation. Pilocarpine, in suitable doses, acts similarly to the vagus, and its relation to fibrillation in auricles and ventricles is to be explained on the same lines.

3. Some drugs and toxic substances, etc., have a different incidence on the auricles and ventricles respectively both in regard to promoting and retarding fibrillation.

Confirmation of Former Views.

So long ago as 1887 the writer* put forward the view that the essential mechanism of typical fibrillation is explicable not simply as an excessive acceleration of rate *per se* or on the assumption of a mechanism of a different nature, in the sense of muscular *v.* nervous, from that concerned in the normal beat, but in a disturbance in the relation between the refractory period and the conduction time in the cardiac musculature; that when this relation is upset by shortening of the refractory period or lengthening of the conduction time or a combination of such changes, the excitation wave, in spreading over the muscular systems, reaches fibres in which the refractory period has already ended and further excitation occurs; the co-ordinated beat is thus abolished and replaced by a rapid and continued series of in-coördinated fibrillar contractions. The alteration in conduction—the passage of the slowed contraction waves in peristaltic fashion along the various complexly-arranged bundles of the ventricular wall at different points of time was described—and also the important fact that single beats may in certain circumstances be fibrillar in character.

Control of Ventricular Fibrillation.

The various actions of different agencies, in promoting or retarding the development of fibrillation and of removing it after it has been established, are to be explained by their incidence on the functions of conduction and excitability and the effects which they bring about in the relations of these functions in different conditions of the cardiac muscle (as a whole) and in the different conditions that may obtain in the auricles and ventricles respectively.

* 'Journal of Physiology,' vol. 8, p. 296 (1887).

Any influence which depresses excitability without depressing—at least proportionately—the function of conduction naturally tends to be in some measure protective against the occurrence of fibrillation and favourable to recovery from that condition when once it has been established. A diminution of excitability opposes the attainment of acceleration sufficient to determine fibrillation; it also diminishes the responsiveness of the muscular fasciculi to circulating excitations. (The control of auricular fibrillation which differs in some respects from that of ventricular fibrillation will be dealt with elsewhere.) Similarly any agency which improves conductivity without unduly exalting excitability is inimical to the mechanism of circulating excitation. Obviously a combination of a depressing influence on excitability with the maintenance of a high level of conductivity would afford the most favourable condition for protection or recovery. Concurrent depressions or elevations of excitability and conductivity in proportionate degree naturally have no specific influence on the question of fibrillation. The agencies which operate successfully in opposing the development of fibrillation—either spontaneous (*i.e.* from unknown causes) or excited artificially by drugs, electrical stimulation, etc.—are often effective in restoring the normal action after fibrillation has been established. Remedies for fibrillation have commonly, in these experiments, been injected into the cavity of the left ventricle through the apex by means of a slender needle; sometimes intravenous injection (external jugular, etc.) was used, massage of the heart being done in both cases, while the artificial respiration is of course maintained. Smaller doses were sufficient by the intra-ventricular mode of injection. Approximately isotonic solutions were used, warmed to body temperature. The doses stated are for cats, usually weighing 2–3 kilos. but sometimes more.

Urethane.—Doses varying between 0·025 and 0·25 grm. injected into the left ventricle were found effective in removing fibrillation in very numerous experiments (fig. 10); 3 per cent. solutions were commonly used for

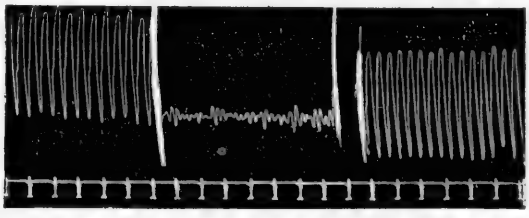
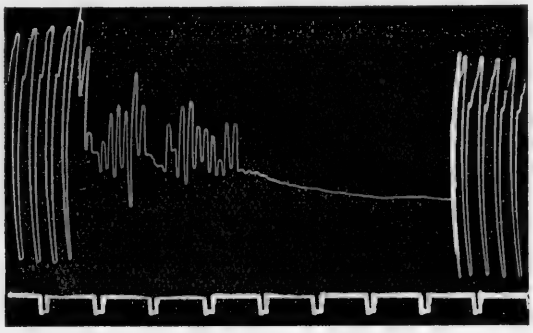


Fig. 10.—The middle portion of the tracing shows fibrillation caused by strong faradisation (5000 units). After it had lasted for 2 minutes (with occasional massage) 0·05 grm. urethane was injected into the L.V. The restored action is seen in the right-hand portion.

intra-cardiac or intra-vascular injections. Hypodermic doses of 0·5 grm. per kilogramme and upwards (given in 25-per-cent. solution, etc.) have a pronounced influence in protecting against fibrillation in light chloroform anæsthesia and in diminishing, though not always obviating, the danger of adrenaline fibrillation in the same grade of anæsthesia. Sufficient time has to be allowed for absorption before the effects are tested. Smaller doses suffice for this purpose when given by intra-vascular (*e.g.*, saphenous vein) injection.

Strontium Chloride was given in doses of 0·01—0·06 grm., a 1-per-cent. solution in dilute Ringer's fluid being usually employed.* Especially when applied at an early phase of the fibrillation this remedy often succeeded very well, and the condition of the heart and circulation were excellent afterwards (fig. 11). In other cases after fibrillation had lasted for a long time and other



A. B.

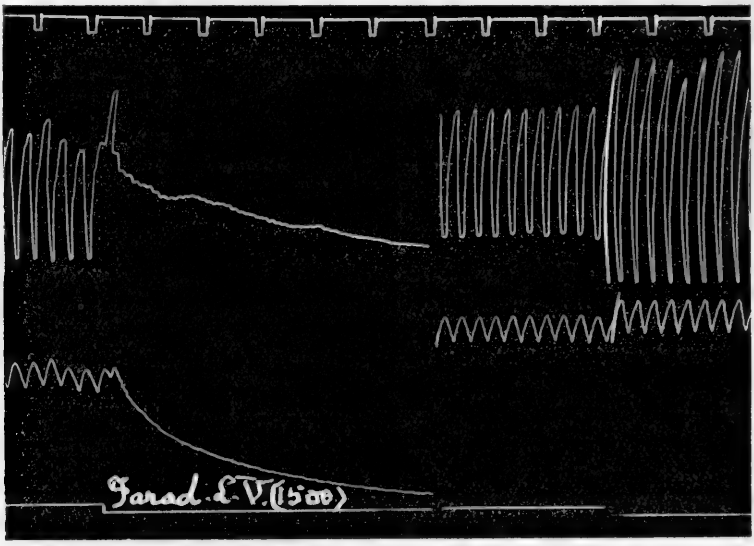
FIG. 11.—A, fibrillation, preceded by period of tachycardia and pseudo-fibrillation, from faradising with 500 units. Injection of 0·06 grm. strontium chloride was followed in 30 seconds by restoration of the normal action, shown in B, taken shortly after recovery. Soon afterwards faradisation with 1000 units again caused fibrillation; recovery followed injection of 0·03 grm., with the usual massage.

measures had been unsuccessful, this salt sometimes speedily induced recovery. Fibrillation, in its various phases, caused by potassium salts is, as might be expected, specially amenable to treatment with strontium in doses varying according to the toxic dose of potassium.

Adrenalin.—Solutions of 1 in 10,000 or 1 in 5,000 were commonly used; sometimes as strong as 1 in 1,000; in Ringer's fluid in each case. The dose varied from 0·1 to 1 mgrm. Successful results were very frequent in fibrillation which had been induced in various ways—by electrical stimulation, chloroform, adrenalin injection during light chloroform anæsthesia (the

* The amounts here stated are of strontium chloride crystals ($SrCl_2+6H_2O$). The doses of the anhydrous salt would be represented by about 60 per cent. of the above amounts.

chloroform-adrenalin reaction described by Levy and abundantly illustrated in this investigation), intravenous injection of potassium salts, etc. In many instances fibrillation has been induced by a small dose (*e.g.*, 0·1 mgrm.) of adrenalin and remedied by the intraventricular injection of a very large dose (up to 1 mgrm.), the state of the heart and circulation remaining good afterwards (fig. 12). The excitability and conductivity of the muscle are



A. B. C.

Fig. 12.—The upper tracing is from the left ventricle, the lower indicates the blood-pressure. In A, fibrillation caused by faradisation with 1500 units lasted 6 minutes, recovery following injection of 0·5 mgrm. adrenalin in three doses. B is shortly after recovery. C, taken 1 minute later, shows much increase in the range of the lever excursions. Note that the blood-pressure is still elevated.

enhanced by a small injection and as early effects of a large injection; subsequently a pronounced depression of excitability occurs—shown in many cases by a great diminution in responsiveness when tested by graduated faradic currents; stimulation, that formerly induced fibrillation readily, now fails to do so even when strengthened to many times its former intensity. Diminished sensitiveness to faradic currents is often pronounced, while the blood-pressure is still elevated and the heart is beating very strongly. Adrenalin can thus act in two ways: (*a*) by reducing excitability, and (*b*) by improving conduction.

Hirudin.—Injections* (into the saphenous vein) of about 8–10 mgrm. per

* Doses of 0·3–0·5 mgrm. were often effective in removing fibrillation injected into the L.V. The solution of hirudin used generally contained 1 mgrm. in each cubic centimetre of Ringer's fluid.

kilogramme of body-weight showed striking effects in opposing the development of fibrillation, either "spontaneously" or in response to electrical stimulation, etc. Even powerful faradisation (often several thousand units) caused only a pseudo-fibrillation, ceasing almost immediately or lasting only a short time (seconds) after the stoppage of the current, or a true fibrillation, which is spontaneously recovered from—on account of the diminished responsiveness of the muscle to the circulating excitations.

Pilocarpine.—Intravenous injection (into jugular, etc.) of 0·0025 grm. (with massage of the ventricles) was often effective in arresting ventricular fibrillation. There was a good deal of variation in regard to this result; there seemed to be a parallelism between the efficiency of pilocarpine in this respect and the activity of vagus inhibition in the particular heart in question—as tested by stimulation of the vagus in the neck or, preferably, the inhibitory area on the dorsal aspect of the auricles. Though vagus stimulation has not been found to arrest fibrillation once it has been established, it has shown notable effects in opposing the development of fibrillation in certain circumstances. And pilocarpine is much more potent than the vagus, though its influence is in the same direction and of the same nature in many respects at least.

Similar remedies were found applicable to the perfused heart, also, a little of the solution of urethane, adrenalin, etc., being injected into the tube leading to the aorta; very small doses usually sufficed.

In some instances, where ventricular fibrillation does not yield so readily as usual to a single remedy, combinations such as urethane and adrenalin, or these followed by strontium chloride, prove very effective. After such treatment the ventricles commonly show a remarkably great resistance to electrical stimulation as far as the induction of fibrillation is concerned, very powerful currents up to 7,000–10,000 units, etc., often causing only pseudo-fibrillation, and, if true fibrillation, with its special mechanism, is induced, it very frequently shows spontaneous recovery after variable periods, frequently without any massage or with massage for some seconds. The difficulty in exciting fibrillation, and its notable tendency to recover, are often very striking, and are to be accounted for, in the main at least, by the diminished responsiveness of the muscle induced by the drugs.

Some relations of different remedial agents to special conditions of the heart may be noted. In very excitable hearts that have fibrillated, depression of excitability is the primary requirement. On the other hand, when direct depression of conductivity (*e.g.*, by potassium salts, bile, cooling, etc.) is the predominant factor in any particular heart, remedies calculated to enhance this function are obviously indicated, whether they act (*a*) by

direct improvement of conductivity or (*b*) secondarily through the slowing of the rate of succession which they may induce, *i.e.* by lowering excitability, provided that this effect is not attended by a proportionate lowering of conductivity. Adrenalin is notably useful in this respect, as indicated by the remarkable improvement in conduction often seen under its influence, especially evident in the auricles, where a strikingly slow contraction wave, present during gravely depressed conduction, may be replaced by an approximation or a return to the normal type. Hence the special utility of adrenalin in dealing with forms of slow coarse fibrillation, already described, and also with fibrillar beats—unless the damage in the latter case has been carried to an irreparable stage (fig. 9).

The success of the above-mentioned methods of obtaining recovery from typical fibrillation, induced by means that did not permanently damage the heart, has been such that in recent years of experimentation there has not been failure in any instance.

For valued assistance in some of the experiments of this investigation, I have to record my thanks to Drs. G. Spencer Melvin and J. R. Murray.

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THE EFFECT OF ALCOHOL ON THE DIGESTION OF FIBRIN AND CASEINOGEN BY TRYPSIN.

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(*Received May 19th, 1919.*)

The behaviour of extracts of pancreas or pure pancreatic juice under different conditions has led various observers to conclude that the pancreas contains a number of proteolytic enzymes. It was shown by Fermi [1890] that after treatment with mercuric chloride, salicylic acid and various other substances, trypsin lost its power of digesting fibrin but would still digest gelatin. Vernon [1901] arguing from the varying sensitiveness of pancreatic extracts towards sodium carbonate, concluded that "trypsin" was really a mixture of enzymes of different degrees of stability, the more sensitive enzymes being destroyed first. Vernon only tested the digestive power of trypsin on raw fibrin in this connection however. In a later paper Vernon [1903, 2] states that pancreatic extracts contain an erepsin as well as trypsin. Pollak [1905] using different preparations of enzymes found that the relative amounts of serum and gelatin digested varied enormously in different cases. He also found that after treatment with hydrochloric acid trypsin lost its power of acting on serum, but was still about as active as ever on gelatin. Pollak concluded that extracts of pancreas contained in addition to trypsin (to which the action on serum was due) a special enzyme which acted only on gelatin. To this enzyme Pollak gave the name of glutinase.

According to Ascoli and Neppi [1908], however, this assumption of a special enzyme acting on gelatin is unjustified, as they find that slight variations in the reaction of the medium affect the digestion of different proteins to different degrees. Mays [1906] after a long series of experiments remarks that the presence of two proteolytic enzymes in pancreatic extracts can only be proved when it is possible to make a separation of the enzymes. It had been previously shown by Bayliss and Starling [1903] that pancreatic juice as secreted contains no trypsin (as tested on coagulated egg white), but contains a weak enzyme like erepsin. This has some action on caseinogen, but very slight. The erepsin has a slight action on fresh fibrin but practically none on fibrin which has been heated to 70°. It may here be mentioned that Long and Barton [1914] state that raw fibrin even when very carefully purified may soon become liquid owing to autolysis.

In later papers Fermi [1913, 1914] contests the theory that some proteolytic enzymes have a specific action, and maintains that all proteolytic enzymes have a general action on all proteins.

Slight differences of behaviour of trypsin towards different proteins under the same conditions have also been noted by Berg and Gies [1906], Porter [1910], Long and Hull [1917], but not much importance seems to have been attached to the facts. Others such as Glaessner and Stauber [1910] and Auerbach and Pick [1912] find differences between the proteolytic and peptolytic actions of trypsin, but in these cases possibly some of the action was due to the pancreatic erepsin also.

It seems to have been assumed, however, by all the authors quoted and by others such as Hedin [1905] that trypsin is the enzyme responsible for the digestion of fibrin and caseinogen, especially in experiments lasting only a few hours.

The action of alcohol on trypsin has been variously stated. Fermi and Pernossi [1894] using Mett's tubes filled with gelatin found that in presence of alcohol trypsin had more digestive action than in presence of water only. The percentage of alcohol used is not stated. Chittenden and Mendel [1896] found that the action of trypsin on fibrin was markedly inhibited by alcohol, but did not test the action on any other substrate. Dastre [1896] found that trypsin still digested fibrin and boiled albumin in presence of 15 to 20 per cent. of alcohol, while Gizelt [1906, 1, 2] states that 20 % alcohol totally inhibits trypsin. According to Bayliss [1915] trypsin will digest gliadin even in presence of 80 % alcohol, the action in this case being due to the trypsin in suspension. Vernon [1903, 1] noted that dilute alcohol had a considerable inhibitory effect on the digestion of raw fibrin by trypsin.

As dilute alcohol is frequently used in making extracts of various digestive organs, it is important to know how the digestive action is affected thereby.

Experimental Details.

The experiments were carried out as described previously [Edie, 1914]. Ox fibrin after being finely minced and thoroughly washed was suspended in water and gradually heated to 85°. The fibrin was then pressed dry and preserved in glycerol and a little chloroform until required. The caseinogen was a 3 % solution in 1 % sodium carbonate. The pancreatic extracts were prepared by finely mincing sheep's pancreas and extracting with chloroform water for about a fortnight. The extract was then filtered and a little chloroform added as a preservative.

The digestion was carried on at 37° in small flasks, a small measured quantity of chloroform being added to exclude bacterial action in every case. When fibrin was used, the amount of digestion was estimated by filtering off the undissolved fibrin and determining the nitrogen in the filtrate by a Kjeldahl determination. When caseinogen was the substrate, the amount of

digestion was found by precipitation with tannic acid and subsequent estimation of the nitrogen in the filtrate. Controls showed that the sodium carbonate alone had no digestive action whatever either on fibrin or caseinogen. The following are typical results showing the effect of dilute alcohol on the digestion of fibrin and caseinogen by trypsin.

			Digestion in cc. of $N/10$ nitrogen
1.	(*a*) 1 c.c. trypsin, 20 c.c. 10 % alcohol, 20 c.c. 1 % Na_2CO_3		4·6
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. 1 % Na_2CO_3		17·1
	1·4 g. fibrin added. Digestion 3 hours.		
	(*a*) 1 c.c. trypsin, 20 c.c. 10 % alcohol, 20 c.c. caseinogen		23·8
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. caseinogen		23·6
	Digestion 1 hour.		
2.	(*a*) 1 c.c. trypsin, 20 c.c. 12 % alcohol, 20 c.c. 1 % Na_2CO_3		4·6
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. 1 % Na_2CO_3		13·8
	1 g. fibrin. Digestion 2·75 hours.		
	(*a*) 1 c.c. trypsin, 20 c.c. 12 % alcohol, 20 c.c. caseinogen		24·1
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. caseinogen		24·3
	Digestion 1·25 hours.		
3.	(*a*) 1 c.c. trypsin, 20 c.c. 10 % alcohol, 20 c.c. 1 % Na_2CO_3		3·8
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. 1 % Na_2CO_3		14·1
	1·3 g. fibrin. Digestion 2 hours.		
	(*a*) 1 c.c. trypsin, 20 c.c. 10 % alcohol, 20 c.c. caseinogen		27·9
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. caseinogen		27·0
	Digestion 1·25 hours.		
4.	(*a*) 1 c.c. trypsin, 20 c.c. 10 % alcohol, 20 c.c. 1 % Na_2CO_3		8·1
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. 1 % Na_2CO_3		23·6
	1·5 g. fibrin. Digestion 2·25 hours.		
	(*a*) 1 c.c. trypsin, 20 c.c. 10 % alcohol, 20 c.c. caseinogen		30·0
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. caseinogen		29·3
	Digestion 1 hour.		
5.	(*a*) 1 c.c. trypsin, 20 c.c. 8 % alcohol, 20 c.c. 1 % Na_2CO_3		5·1
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. 1 % Na_2CO_3		12·1
	1 g. fibrin. Digestion 3 hours.		
	(*a*) 1 c.c. trypsin, 20 c.c. 8 % alcohol, 20 c.c. caseinogen		30·6
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. caseinogen		30·6
	Digestion 1·25 hours.		
6.	(*a*) 1 c.c. trypsin, 20 c.c. 8 % alcohol, 20 c.c. 1 % Na_2CO_3		4·9
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. 1 % Na_2CO_3		12·1
	1 g. fibrin. Digestion 3 hours.		
	(*a*) 1 c.c. trypsin, 20 c.c. 8 % alcohol, 20 c.c. caseinogen		27·7
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. caseinogen		28·0
	Digestion 1 hour.		
7.	(*a*) 1 c.c. trypsin, 20 c.c. 6 % alcohol, 20 c.c. 1 % Na_2CO_3		11·5
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. 1 % Na_2CO_3		20·3
	1 g. fibrin. Digestion 3 hours.		
	(*a*) 1 c.c. trypsin, 20 c.c. 6 % alcohol, 20 c.c. caseinogen		22·2
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. caseinogen		22·0
	Digestion 1 hour.		

				Digestion in cc. of $N/10$ nitrogen
8.	(*a*)	1 c.c. trypsin, 20 c.c. 6 % alcohol,	20 c.c. 1 % Na_2CO_3	12·5
	(*b*)	1 c.c. trypsin, 20 c.c. water,	20 c.c. 1 % Na_2CO_3	22·2
		1 g. fibrin. Digestion 3 hours.		
	(*a*)	1 c.c. trypsin, 20 c.c. 6 % alcohol,	20 c.c. caseinogen	25·0
	(*b*)	1 c.c. trypsin, 20 c.c. water,	20 c.c. caseinogen	24·6
		Digestion 1 hour.		
9	(*a*)	5 c.c. trypsin, 20 c.c. 16 % alcohol,	20 c.c. 1 % Na_2CO_3	7·1
	(*b*)	5 c.c. trypsin, 20 c.c. water,	20 c.c. 1 % Na_2CO_3	18·2
		1 g. fibrin. Digestion 2 hours.		
	(*a*)	5 c.c. trypsin, 20 c.c. 16 % alcohol,	20 c.c. caseinogen	31·6
	(*b*)	5 c.c. trypsin, 20 c.c. water,	20 c.c. caseinogen	31·9
		Digestion 1 hour.		
10.	(*a*)	5 c.c. trypsin, 20 c.c. 13 % alcohol,	20 c.c. 1 % Na_2CO_3	14·0
	(*b*)	5 c.c. trypsin, 20 c.c. water,	20 c.c. 1 % Na_2CO_3	25·2
		1·3 g. fibrin. Digestion 3 hours.		
	(*a*)	5 c.c. trypsin, 20 c.c. 13 % alcohol,	20 c.c. caseinogen	34·5
	(*b*)	5 c.c. trypsin, 20 c.c. water,	20 c.c. caseinogen	34·6
		Digestion 1·25 hours.		
11.	(*a*)	5 c.c. trypsin, 20 c.c. 14 % alcohol,	20 c.c. 1 % Na_2CO_3	13·7
	(*b*)	5 c.c. trypsin, 20 c.c. water,	20 c.c. 1 % Na_2CO_3	26·8
		1·2 g. fibrin. Digestion 2·5 hours.		
	(*a*)	5 c.c. trypsin, 20 c.c. 14 % alcohol,	20 c.c. caseinogen	32·5
	(*b*)	5 c.c. trypsin, 20 c.c. water,	20 c.c. caseinogen	32·5
		Digestion 1 hour.		
12.	(*a*)	5 c.c. trypsin, 20 c.c. 14 % alcohol,	20 c.c. 1 % Na_2CO_3	10·8
	(*b*)	5 c.c. trypsin, 20 c.c. water,	20 c.c. 1 % Na_2CO_3	24·6
		1 g. fibrin. Digestion 2 hours.		
	(*a*)	5 c.c. trypsin, 20 c.c. 14 % alcohol,	20 c.c. caseinogen	30·0
	(*b*)	5 c.c. trypsin, 20 c.c. water,	20 c.c. caseinogen	30·1
		Digestion 1 hour.		

These experiments are sufficient to show that alcohol, when present in percentages varying from 3 to 7, has a very marked inhibitory effect on the digestion of fibrin by trypsin but no such effect on the digestion of caseinogen. The amount of fibrin digested under these conditions varied from about 25 to 50 % of the amount digested in absence of alcohol, the proportion varying somewhat with different trypsin solutions and with varying percentages of alcohol. In no case was there any appreciable difference in the amount of caseinogen digested, beyond the limits of experimental error.

With higher percentages of alcohol the digestion of fibrin was in some cases entirely stopped, a fair amount of caseinogen still being digested, however.

				Digestion in c.c. of $N/10$ nitrogen
13.	(*a*)	1 c.c. trypsin, 20 c.c. 25 % alcohol,	20 c.c. 1 % Na_2CO_3	1·3
	(*b*)	1 c.c. trypsin, 20 c.c. water,	20 c.c. 1 % Na_2CO_3	27·2
		1 g. fibrin. Digestion 3 hours.		
	(*a*)	1 c.c. trypsin, 20 c.c. 25 % alcohol,	20 c.c. caseinogen	20·0
	(*b*)	1 c.c. trypsin, 20 c.c. water,	20 c.c. caseinogen	23·7
		Digestion 1 hour.		

		Digestion in cc. of $N/10$ nitrogen
14.	(*a*) 1 c.c. trypsin, 20 c.c. 25 % alcohol, 20 c.c. 1 % Na_2CO_3	4·8
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. 1 % Na_2CO_3	25·2
	0·8 g. fibrin. Digestion 3 hours.	
	(*a*) 1 c.c. trypsin, 20 c.c. 25 % alcohol, 20 c.c. caseinogen	32·8
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. caseinogen	37·9
	Digestion 1·75 hours.	
15.	(*a*) 1 c.c. trypsin, 20 c.c. 50 % alcohol, 20 c.c. 1 % Na_2CO_3	0·0
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. 1 % Na_2CO_3	23·6
	1 g. fibrin. Digestion 3 hours.	
	(*a*) 1 c.c. trypsin, 20 c.c. 50 % alcohol, 20 c.c. caseinogen	5·3
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. caseinogen	25·2
	Digestion 1 hour.	
16.	(*a*) 1 c.c. trypsin, 20 c.c. 50 % alcohol, 20 c.c. 1% Na_2CO_3	0·0
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. 1 % Na_2CO_3	23·4
	1·2 g. fibrin. Digestion 3 hours.	
	(*a*) 1 c.c. trypsin, 20 c.c. 50 % alcohol, 20 c.c. caseinogen	4·6
	(*b*) 1 c.c. trypsin, 20 c.c. water, 20 c.c. caseinogen	28·2
	Digestion 1 hour.	

These experiments show that in presence of 25 % of alcohol the digestion of fibrin by trypsin is entirely inhibited, while digestion of caseinogen still proceeds to a limited extent. In presence of 12 % alcohol the amount of fibrin digested is from 10 to 20 % of the control, while the caseinogen digested amounts to about 85 % of the control.

Trypsin is well known to be very unstable under some circumstances, and it was considered possible that contact with dilute alcohol for some time might lead to an actual destruction of that part of the enzyme molecule which digests fibrin. The following experiments were carried out to test such a theory.

		Digestion in c.c. of $N/10$ nitrogen
17.	(*a*) 20 c.c. trypsin, 15 c.c. 15 % alcohol (*b*) 20 c.c. trypsin, 15 c.c. water } kept at 37° C. for 3 hours	
	2 c.c. of (*a*), 40 c.c. 0·5 % Na_2CO_3	31·2
	2 c.c. of (*b*), 40 c.c. 0·5 % Na_2CO_3	31·0
	1·3 g. fibrin. Digestion 3 hours.	
18.	(*a*) 20 c.c. trypsin, 5 c.c. 30 % alcohol (*b*) 20 c.c. trypsin, 5 c.c. water } kept at 37° C. for 3 hours	
	2 c.c. of (*a*), 40 c.c. 0·5 % Na_2CO_3	18·2
	2 c.c. of (*b*), 40 c.c. 0·5 % Na_2CO_3	18·3
	1·2 g. fibrin. Digestion 2 hours.	
19.	(*a*) 40 c.c. trypsin, 10 c.c. 30 % alcohol (*b*) 40 c.c. trypsin, 10 c.c. water } kept at 37° C. for 3 hours	
	1 c.c. of (*a*), 40 c.c. 0·5 % Na_2CO_3	14·0
	1 c.c. of (*b*), 40 c.c. 0·5 % Na_2CO_3	14·2
	1 g. fibrin. Digestion 3 hours.	
20.	(*a*) 15 c.c. trypsin, 10 c.c. 15 % alcohol (*b*) 15 c.c. trypsin, 10 c.c. water } kept at 37° C. for 3 hours	
	2 c.c. of (*a*), 40 c.c. 0·5 % Na_2CO_3	17·4
	2 c.c. of (*b*), 40 c.c. 0·5 % Na_2CO_3	17·0
	1 g. fibrin. Digestion 2·75 hours.	

		Digestion in cc. of $N/10$ nitrogen
21.	(*a*) 15 c.c. trypsin, 10 c.c. 15 % alcohol (*b*) 15 c.c. trypsin, 10 c.c. water } kept at 37° C. for 3 hours	
	2 c.c. of (*a*), 40 c.c. 0·5 % Na_2CO_3	16·3
	2 c.c. of (*b*), 40 c.c. 0·5 % Na_2CO_3	16·2
	1 g. fibrin. Digestion 2·75 hours.	

No destruction whatever of the trypsin is caused by the action of 6 % alcohol, although the digestive action of the enzyme is reduced to 30 % or less of the normal amount by the presence of this proportion of alcohol.

A solid substrate such as fibrin might be rendered less digestible by prolonged treatment with concentrated alcohol, owing to the hardening thus brought about. Alcohol of under 30 %, however, could hardly be supposed to have such an effect, and a few experiments showed that after treatment with dilute alcohol fibrin was no less digestible by trypsin than previously.

		Digestion in c.c. of $N/10$ nitrogen
22.	(1) Fibrin + 10 % alcohol (2) Fibrin + water } kept at 37° C. for 3 hours	
	1 c.c. trypsin, 40 c.c. 0·5 % Na_2CO_3, 1 g. fibrin (1)	19·3
	1 c.c. trypsin, 40 c.c. 0·5 % Na_2CO_3, 1 g. fibrin (2)	18·1
	Digestion 2·5 hours.	
23.	(1) Fibrin + 10 % alcohol (2) Fibrin + 10 % alcohol } kept at 37° C. for 19 hours	
	1 c.c. trypsin, 40 c.c. 0·5 % Na_2CO_3, 1 g. fibrin (1)	22·9
	1 c.c. trypsin, 40 c.c. 0·5 % Na_2CO_3, 1 g. fibrin (2)	20·2
	Digestion 3 hours.	

The fibrin which was to be treated with alcohol in these experiments was first washed with alcohol in order to remove any adherent moisture. It will be seen that after treatment with 10 % alcohol fibrin is apparently slightly more readily attacked by trypsin than previously.

The action of trypsin on fibrin and on caseinogen is affected by dilute alcohol to such different degrees that it is reasonable to suppose either that there are two enzymes concerned in the digestion of these proteins or that different groups of the same enzyme molecule take part in the hydrolysis of the different proteins. In the latter case the groups which digest fibrin are very much more easily inhibited by alcohol than the groups which digest caseinogen.

The theory that different side chains in the molecule of an enzyme are responsible for different functions is used to explain the zymoid modification of enzymes. Some observers also, for example, Nencki and Sieber [1901], hold that the behaviour of pepsin and rennin under varying conditions can best be explained on the theory that only one enzyme is concerned here, with different side chains responsible for the proteolytic and milk coagulating functions. Vernon [1903, 1] also considers this probable in the case of the milk coagulating and proteolytic actions of trypsin.

Hitherto it has apparently been assumed that one enzyme "trypsin" is responsible for the digestion of fibrin and caseinogen by pancreatic extracts.

In this case the function is the same (hydrolysis of a protein to form simpler products), but it would seem that different side chains may be necessary for the hydrolysis of different proteins.

Summary.

Alcohol when present to the extent of 3 % and upwards markedly inhibits the action of trypsin on fibrin. The digestion of caseinogen by trypsin is not affected until the concentration reaches 10 %. The action of alcohol is not due to the destruction of the trypsin, since on suitable dilution of the mixture of trypsin and alcohol the digestion of fibrin is as great as in the control.

Fibrin is not rendered less digestible by contact with dilute alcohol, but seems to be slightly more readily dissolved by trypsin than previously.

If "trypsin" is a single enzyme the digestion of fibrin and caseinogen is probably carried on by different side chains, those digesting fibrin being much more readily affected by alcohol than the others.

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FURTHER OBSERVATIONS ON THE DIGESTION OF FIBRIN AND CASEINOGEN BY TRYPSIN

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In a previous paper [Edie, 1919] it was shown that the activity of trypsin as measured by its digestive action on fibrin and on caseinogen is affected to such a different degree by alcohol as to make it seem either that two enzymes are concerned, or, if only one enzyme, that the two substrates are acted on by different groups or side chains, one group being much more sensitive than the other. The effect of heat on the digestion of fibrin and caseinogen by trypsin was next studied. It had been found previously [Edie, 1914] that trypsin when boiled in acid solution still retains much or in some cases all of its power of hydrolysing caseinogen, but in only one case was its action on fibrin tested after heating in this way. Digestion of the fibrin was still noticed, but the amount even in the case of the unheated trypsin was so small that a fresh series of experiments was undertaken.

The trypsin solutions were generally prepared by extracting finely minced sheep's pancreas with water and a little chloroform for 10 to 14 days and filtering. A little chloroform was then added as a preservative. The experiments were carried out as previously detailed.

In the experiments described in the previous paper it had been found that in presence of $N/25$ to $N/50$ HCl the trypsin solutions used retained 60 to 100 % of their original digestive power, as tested on caseinogen, after being heated to 100° for three minutes. The same treatment was applied to the trypsin solutions in this series of experiments and the results are shown in the table.

In making these extracts, one part of pancreas was extracted with two parts of water in each case.

In each experiment 1 cc. trypsin + 40 cc. 0·5 % Na_2CO_3 + 1 g. fibrin were used on the one hand, digestion being for three hours, and 1 cc. trypsin + 40 cc. 1·5 % caseinogen in 0·5 % Na_2CO_3 on the other hand, digestion being for one hour.

The amount of digestion was estimated by precipitating unchanged caseinogen with tannic acid, or by filtering off the undissolved fibrin in the different sets of experiments respectively, and determining the nitrogen in

the filtrate by Kjeldahl's method. Control experiments were always carried out at the same time.

No.	Reaction (HCl)	Treatment	Fibrin digested (in cc. of $N/10$ nitrogen)	Caseinogen digested
1	$N/55$	3 min. at 100°	0·0	1·2
		Control	20·1	28·6
2	$N/20$	1·5 min. at 60°	0·0	3·1
		Control	21·9	34·7
3	$N/20$	1·5 min. at 60°	0·0	5·1
		Control	22·1	30·9
4	$N/30$	1 min. at 85°	0·0	1·9
		Control	7·1	22·4
5	$N/30$	1 min. at 65°	0·0	3·2
		Control	8·6	22·9
6	$N/20$	0·75 min. at 100°	0·0	3·2
		Control	21·2	34·6
7	$N/20$	0·75 min. at 66°	2·2	19·5
		Control	16·6	34·3
8	$N/20$	1·5 min. at 66°	1·1	8·3
		Control	16·2	31·8
9	$N/40$	1·5 min. at 75°	3·4	17·0
		Control	17·4	37·4

In the above experiments it will be seen that even in acid solution the pancreatic extracts used generally lost practically all their power to digest fibrin even when heated only to 60° for a minute and a half. The power to digest caseinogen was not so completely lost, especially in the last three experiments, but it was so markedly reduced, compared with what had been previously found, that the matter merited further investigation.

The extracts used above were aqueous, so a few experiments were carried out to compare the effect of heat on aqueous and alcoholic (20 %) extracts of the same pancreas. The experiments were carried out in the same way as those previously noted.

No.	Reaction (HCl)	Extract	Treatment	Fibrin digested cc.	Caseinogen digested cc.
10	$N/40$	Aqueous	1·5 min. at 75°	3·4	17·0
		,,	Control	17·4	37·4
		Alcohol	1·5 min. at 75°	17·3	29·3
		,,	Control	31·0	47·2
11	$N/50$	Alcohol	1 min. at 100°	23·4	34·4
		,,	Control	27·9	42·8
12	$N/40$	Aqueous	1 min. at 100°	0·0	—
		,,	Control	11·8	—
		Alcohol	1 min. at 100°	21·1	—
		,,	Control	23·1	—
13	$N/40$	Aqueous	1 min. at 100°	0·4	—
		,,	Control	11·7	—
		Alcohol	1 min. at 100°	14·0	—
		,,	Control	21·5	—

It will be seen that the alcoholic extracts retain very much more of their power to digest fibrin after being heated than do the corresponding aqueous extracts. The amount of protein and other nitrogenous substances was practically the same in the two sets of extracts. It might be supposed that the alcohol itself in some way protected the trypsin from destruction when

heated, the lower boiling point of alcohol preventing the alcoholic extract from reaching such a high temperature in the water-bath as the corresponding aqueous extract would reach. No such protection was found to be afforded by alcohol, however. Corresponding aqueous and alcoholic extracts were diluted with alcohol and water respectively so that the percentage of alcohol was the same in both. They were then heated to the same extent and tested on fibrin.

No.	Reaction (HCl)	Extract	Treatment	Fibrin digested cc.
14	*N*/40	Alcoholic	1 min. at 100°	21·1
		,,	Control	23·1
		Aqueous, alcohol added afterwards	1 min. at 100°	0·0
		,,	Control	18·9
15	*N*/40	Alcoholic	1 min. at 100°	14·0
		,,	Control	21·5
		Aqueous, alcohol added afterwards	1 min. at 100°	0·0
		,,	Control	7·9

These experiments show that the addition of alcohol to an aqueous extract of pancreas does not afford any protection against heat to the trypsin as measured by its action on fibrin.

In the original experiments on the resistance of trypsin solutions to heat, the solutions, when heated, contained only a very small amount of nitrogen, not more, in some cases, than 0·02 %. The pancreatic extracts used in the experiments now described were very much richer in nitrogen and generally contained 15 to 20 times as much as the older extracts. This corresponds to a considerable amount of protein in the solution and as protein is known to form a loose compound with hydrochloric acid it was decided to try the effect of considerably higher amounts of acid in the solutions to be heated.

It was now found that the protection afforded to trypsin solutions when heated depends on the amount of acid present, and the more protein there is in solution, the more hydrochloric acid must be added to prevent the trypsin being destroyed by heat.

The following experiments show this increasing protection with increase of acid:

No.	Reaction (HCl)	Treatment	Fibrin digested cc.	Caseinogen digested cc.
16	*N*/50	1 min. at 100°	5·6	21·1
		Control	16·9	39·1
17	*N*/23	1 min. at 100°	—	25·1
		Control	—	35·7
18	*N*/17	1·5 min. at 100°	7·4	26·8
		Control	7·6	26·7
19	*N*/15	3 min. at 100°	10·9	26·2
		Control	10·9	26·4
20	*N*/15	2 min. at 100°	3·5	19·2
		Control	11·6	28·7
21	*N*/12	2 min. at 100°	8·4	14·8
		Control	12·2	15·6

Experiments 16 to 19 show the effect of using increasing amounts of hydrochloric acid with the same trypsin preparation, and experiments 20 and 21 show this again with another sample of trypsin.

Similar results have been obtained with many aqueous extracts of pancreas prepared in the laboratory, and it may be stated generally that the higher the proportion of nitrogen contained in such an extract the larger the amount of hydrochloric acid which must be added in order to prevent destruction of the trypsin by heat.

In a few cases, as for example experiments 18 and 19, it was found that the protection against heat afforded by a certain amount of acid was the same as regards digestion both of fibrin and of caseinogen. Usually, however, the destruction of the fibrin digesting power was considerably greater than that of the caseinogen digesting power.

It has been pointed out by Mellanby and Woolley [1913] that in acid of the strength of 0·05 *N* (HCl) trypsin is slowly destroyed at 16° and more rapidly at 35°. Apparently at room temperature about half the trypsin is destroyed in four hours, and two-thirds is destroyed in a day. The activity of the trypsin in their experiments was measured by its power of coagulating calcified milk. Other references to the effect of hydrochloric acid on trypsin at moderate temperatures have been mentioned in a previous paper [Edie, 1914]. It was also found by Lénard [1914] that if trypsin is rendered inactive by addition of acid, only a trace of its activity is restored by neutralising and then adding alkali. These observers appear only to have tested the activity of the trypsin on one substrate, but in the following experiments the action of hydrochloric acid on trypsin at room or body temperature has been tested as regards the power to digest both fibrin and caseinogen. In these experiments alcoholic (15 %) extracts of pig's pancreas were used. These were practically neutral. In every case 1 cc. of the original trypsin was compared with that quantity of the trypsin + acid which would contain 1 cc. of trypsin originally, and the solutions so adjusted as to contain the same amount of sodium chloride. Digestion both of fibrin and of caseinogen was carried on in presence of 0·5 % sodium carbonate, 1 g. fibrin or 0·6 g. caseinogen being used, in about 40 cc. of fluid. The amount of digestion is expressed, as usual, in cc. of *N*/10 nitrogen.

22. 10 cc. trypsin + 20 cc. *N* HCl. Kept at 36° for 12 min. 20 cc. *N* NaOH then added.
Digestion by control 21·6 cc. fibrin, 38·2 cc. caseinogen.
„ treated trypsin 0·0 cc. „ 8·8 cc. „

23. 20 cc. trypsin + 10 cc. *N* HCl. Room temperature for 4 days. 10 cc. *N* NaOH added.
Digestion by control 12·1 cc. fibrin, 31·6 cc. caseinogen.
„ treated trypsin 0·0 cc. „ 5·4 cc. „

24. 40 cc. trypsin + 20 cc. *N* HCl. Room temperature for 11 days. 20 cc. *N* NaOH added.
Digestion by control 11·7 cc. fibrin, 38·7 cc. caseinogen.
„ treated trypsin 0·0 cc. „ 5·6 cc. „

It will be seen from these experiments that the power of trypsin to digest fibrin is destroyed considerably more readily in acid solution at moderate

temperatures than is the power to digest caseinogen. It is also seen that the power to digest caseinogen withstands a considerably higher percentage of hydrochloric acid than has generally been supposed, the strength of acid being $N/3$ in these experiments and several others with similar results. In one experiment, after 24 hours at room temperature in $N/3$ hydrochloric acid, the trypsin still retained about 10 % of its original fibrin digesting power, but otherwise no fibrin was digested at all after treatment of the trypsin with acid of this strength for a day or upwards.

On the whole, then, treatment of trypsin solutions with hydrochloric acid either at high or low temperatures shows that the power to digest fibrin is more readily destroyed than the power to digest caseinogen. This bears out the theory discussed previously [Edie, 1919] that in some respects the fibrin digesting power is the more subject to outside influences and again points to the hydrolysis of fibrin and of caseinogen being carried out by different side chains, those digesting caseinogen being the more stable.

In my previous paper [1919], the work of Fermi was referred to as showing that after treatment with various reagents trypsin would no longer digest fibrin but would still digest gelatin. Pollak was also mentioned as finding that with different enzyme preparations the relative amounts of serum and gelatin digested varied enormously. I have also found that the relative amounts of fibrin and caseinogen digested by different trypsin solutions vary very much, and this without subjecting the enzyme to treatment of any kind.

Thus, three enzyme solutions were prepared in exactly the same way, by extracting minced sheep's pancreas with three times its weight of water for 14 days and filtering. These were compared at the same time. 1 cc. of each trypsin was taken, with 40 cc. 0·5 % Na_2CO_3 and 1 g. fibrin on the one hand, and 1 cc. trypsin with 40 cc. 1·5 % caseinogen in 0·5 % Na_2CO_3 on the other hand. The amounts of digestion in the three cases were as follows:

Enzyme	Fibrin digested in 2 hours. cc.	Caseinogen digested in 1 hour. cc.
1	12·4	33·7
2	16·1	32·9
3	3·3	17·6

The differences in the relative amounts of fibrin and caseinogen digested by these enzymes, especially Nos. 2 and 3, are very marked. Similar results were frequently noticed in other cases, the general rule being that considerable amounts of caseinogen were digested even though a particular enzyme solution had little or almost no action on fibrin.

When enzyme solutions were kept for some time it was found that the fibrin digesting power as a rule diminished to a very much greater extent than the caseinogen digesting power. For example, a freshly prepared trypsin, under the usual conditions, digested 16·1 cc. fibrin and 32·9 cc. caseinogen. In 15 months, under the same conditions, this trypsin digested 4·8 cc. fibrin and 18·5 cc. caseinogen.

In one extreme case I examined a solution of trypsin which had been in the laboratory for over ten years. It had no digestive action on fibrin at all, but still digested caseinogen to the extent of 26·6 cc. under the usual conditions.

These facts afford further evidence that the digestion of fibrin and of caseinogen by pancreatic extracts is either due to different enzymes or at least to different side chains if only one enzyme is involved. In my previous paper [1914] I mentioned that the power to digest caseinogen seemed to be less affected by heat than the power to coagulate milk, which was taken as the measure of activity of trypsin by Mellanby and Woolley. I further suggested that different sets of side chains might be responsible for these different functions.

In a later paper [1914] Mellanby and Woolley take exception to my suggestion and say "Pancreatic rennin and trypsin are identical. In fact the coagulation of milk by trypsin is an expression of a general law that all proteolytic ferments coagulate milk provided sufficient calcium be contained in it." These authors further say "The unique fact that the ferment or ferments in pancreatic juice which digest protein and coagulate milk should withstand boiling in acid solution is practically conclusive proof that the two actions are produced by one and the same substance." If this assumption of Mellanby and Woolley is correct, however, then the milk coagulating power and the power to hydrolyse both fibrin and caseinogen should presumably be quite parallel in their behaviour. The experiments detailed in the present paper, and those described previously [Edie, 1919], however, tend to show that the digestion of fibrin and of caseinogen, if carried out by one enzyme, involves at least two sets of groups of the enzyme molecule, and therefore cannot be said really to be produced by the same substance in the sense evidently meant by Mellanby and Woolley.

I have also carried out some experiments comparing the milk coagulating power of pancreatic extracts with their proteolytic power, and shall now deal with these.

25. To 20 cc. of pancreatic extract (alcoholic) was added 0·5 cc. *N* HCl. Half of this was then heated to 100° for 1 minute and filtered.

1 g. fibrin. Digestion 2·75 hours at 37°.
(*a*) 1 cc. trypsin, 40 cc. 0·5 % Na_2CO_3. Digestion 21·0 cc. *N*/10 nitrogen.
(*b*) 1 cc. trypsin (heated), 40 cc. 0·5 % Na_2CO_3. Digestion 10·2 cc. *N*/10 nitrogen.
To 20 cc. milk was added 1 cc. of trypsin (1) fresh trypsin.
" " " " " (2) heated trypsin.
(1) Complete coagulation in 6 minutes at 37°.
(2) No coagulation in 3 hours.

26. Similar to last experiment.

1 g. fibrin. Digestion 2·5 hours.
(*a*) 1 cc. trypsin, 40 cc. 0·5 % Na_2CO_3. Digestion 26·2 cc.
(*b*) 1 cc. trypsin (heated), 40 cc. 0·5 % Na_2CO_3. Digestion 8·1 cc.
(1) 20 cc. milk, 1 cc. fresh trypsin. Complete coagulation in 5 minutes.
(2) 20 cc. milk, 1 cc. heated trypsin. No coagulation in 2 hours.

From these two experiments it will be seen that though the heated trypsin is still able to digest a considerable amount of fibrin, its milk coagulating power, if any, is now quite negligible.

27. 20 cc. trypsin + 0·5 cc. *N* HCl. Half kept at 100° for 1 minute and filtered.

(*a*) 1 cc. trypsin, 20 cc. milk. Digestion 39·3 cc.

(*b*) 1 cc. trypsin (heated), 20 cc. milk. Digestion 4·6 cc.

Digestion 1 hour. Tannic acid added and digestion estimated as in the usual caseinogen experiments.

(1) 20 cc. milk, 1 cc. fresh trypsin. Complete coagulation in 5 minutes.

(2) 20 cc. milk, 1 cc. heated trypsin. No coagulation in 2 hours.

28. Similar to last experiment.

(*a*) 1 cc. trypsin, 20 cc. milk. Digestion 38·4 cc.

(*b*) 1 cc. trypsin (heated), 20 cc. milk. Digestion 3·9 cc.

(1) 20 cc. milk, 1 cc. fresh trypsin. Complete coagulation in 5 minutes.

(2) 20 cc. milk, 1 cc. heated trypsin. No coagulation in 2 hours.

These two experiments confirm Nos. 25 and 26 in showing that the milk coagulating power of pancreatic extracts is more readily destroyed by heat than the proteolytic power.

It was noticed that the coagulated casein gradually dissolved under the influence of the fresh trypsin, digestion of this protein taking place rapidly even in neutral solution.

More striking differences are found between the milk coagulating power and the proteolytic action of pancreatic extracts under certain conditions without subjecting these to any such drastic treatment as heating to 100° involves. Edkins [1891] found that fresh, active pancreatic extracts were not so active in altering milk so as to produce Roberts' "metacasein" reaction as were older extracts, but that the proteolytic action was greater in the fresh extracts. Edkins suggested that the production of the metacasein reaction might be an aspect of the proteolytic enzyme of the pancreas. Halliburton and Brodie [1896] confirmed what had been pointed out by Benger, that freshly prepared extract of pig's pancreas had very little curdling action on milk, but acquired this property on being kept a considerable time. They accounted for this fact by supposing that the trypsin at first masks or hinders the milk curdling enzyme, but that the former enzyme deteriorates more quickly and so finally allows the rennin to reveal its presence. Vernon [1901] obtained similar results and found that the ratio of rennin value to tryptic value varied largely in different extracts of pancreas and also in the same extracts at different times. In alcoholic extracts the ratio usually became higher as the extract became older, the tryptic value deteriorating more rapidly than the rennin. In glycerol extracts, however, the ratio diminished after say nine weeks, owing to the trypsin being liberated more slowly from its zymogen than the rennin. Some glycerol extracts which were very rich in trypsin gave practically no clot at all, as though the clot were dissolved nearly as fast as it was formed. In a later paper, Vernon [1903] found that rennin and trypsin were precipitated from pancreatic extracts to practically

the same extent when excess of alcohol was added. He considered that in the case of trypsin some groups have the property of coagulating milk and others have the proteolytic power.

I have tested a number of pancreatic extracts at different stages and record some of the principal results below.

In each case sheep's pancreas was used. It was finely minced and extracted with two and a half times its weight of water and a little chloroform. The experiments were similar to Nos. 25 to 28 in technique.

29. Pancreas extracted for two days and extract then tested.
1 cc. trypsin, 40 cc. 0·5 % Na_2CO_3, 1 g. fibrin. 14·9 cc. digested in 3 hours.
1 cc. trypsin, 40 cc. caseinogen (1·5 % in 0·5 % Na_2CO_3). 44·6 cc. digested in 1 hour.
1 cc. trypsin, 40 cc. milk. 23·3 cc. digested in 0·5 hour.
1 cc. trypsin + 20 cc. milk. No coagulation in 0·5 hour. No coagulation on now adding an active coagulating extract, as the caseinogen had been changed (as can be seen above) into products which no longer give a coagulum with rennin.

30. Pancreas extracted for 1·5 hours and extract then tested.
26·4 cc. fibrin digested in 3 hours.
41·8 cc. caseinogen digested in 1 hour.
No coagulation of milk, but much digestion (tannic acid).

31. Pancreas extracted for 3 days and extract then tested.
26·7 cc. caseinogen (milk used) digested in 0·5 hour.
No coagulation of milk.

32. Pancreas extracted for 3 days and extract then tested.
31·0 cc. caseinogen digested in 0·5 hour.
No coagulation of milk, but much digestion.

These four experiments show that freshly prepared aqueous extracts of pancreas generally do not coagulate milk, but are very active proteolytic agents, both on fibrin and on caseinogen.

In a few cases I have found that the filtrate after three hours' extraction coagulated milk rapidly, but this was exceptional.

It has already been mentioned that Halliburton and Brodie accounted for the fact that fresh extracts have very little curdling action on milk by supposing that trypsin deteriorates more quickly and in a few days or weeks allows the rennin to reveal its presence. Vernon seems to suggest that a clot is formed but is dissolved almost at once. In the extracts which I used, the coagulating power seemed to be fully developed within 15 days, but I never found the proteolytic power to diminish as rapidly as would have to be the case if the above suggestion accounted for all the facts.

The extract used in experiment 30 was tested again for proteolytic power when five weeks old. Complete coagulation of milk now took place within four minutes, and the amount of caseinogen digested was now 44·7 cc., this being actually slightly more than it digested at first. If in the first case a clot were formed but almost immediately redissolved, this should have been still more the case when the proteolytic power had increased, instead of which complete coagulation rapidly took place. In other cases the milk coagulating power seemed to have developed completely within four days, the proteolytic

power being practically the same as at first and there was no evidence whatever in support of the view that with extracts a few hours old coagulation really takes place but the clot is redissolved almost instantly. These last experiments once more show that pancreatic extracts differ greatly in their milk coagulating and proteolytic powers. This again points to there being either two or more separate enzymes present, which develop at very different rates from their zymogens, or at least the groups which are responsible for the different functions develop their properties quite independently.

Summary.

1. The amount of acid required in order to protect trypsin from destruction by heat depends on the amount of protein present. The more protein in solution, the more acid required. If not enough acid is present to afford complete protection to the trypsin, the fibrin digesting power is usually destroyed by heat to a considerably greater extent than the power to digest caseinogen.

2. Hydrochloric acid at moderate temperatures also destroys the fibrin digesting power considerably more rapidly than the caseinogen digesting power.

3. The relative amounts of fibrin and caseinogen digested vary very much in different pancreatic extracts.

4. The milk coagulating power of pancreatic extracts is more easily destroyed by heat than the proteolytic power.

5. Generally, but not always, freshly prepared pancreatic extracts have no milk coagulating power. These extracts are always actively proteolytic, but the proteolytic power does not fall off so rapidly as to justify the assumption that the non-appearance of a coagulum with milk is due to the coagulum being really formed but instantly redissolved. All these facts point to the proteolytic and milk coagulating powers of pancreatic extracts being due to a number of distinct enzymes, or, if only one enzyme is concerned, to the different functions being due to different groups of the molecule.

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A NOTE ON THE QUESTION OF THE IDENTITY OF GASTRIC RENNIN AND PEPSIN.

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Much has been written on the question of the identity of gastric rennin and pepsin, and two theories have been brought forward. One, first associated with Pavlov [Pavlov and Parastschuk, 1904], is that pepsin and rennin are identical, Savjalov [1905] and Gewin [1907] holding that coagulation is the first stage in the digestion of milk by pepsin. The identity theory is based on the parallelism between the behaviour of the proteolytic and milk coagulating actions of gastric extracts under different conditions. The other theory is that the enzymes are different. There are two possibilities here, one, put forward by Nencki and Sieber [1901] and others, being that "pepsin" consists of a large molecule with different side chains, one set of which digests protein in acid solution, while another set is responsible for the coagulation of milk in neutral solution. A second possibility is that there are two distinct enzymes involved in the two functions. This is the theory which has been mainly developed by Hammarsten [1908]. It is difficult to distinguish experimentally between these two possibilities, and indeed the present state of our knowledge of the constitution of enzymes renders a distinction hardly practicable. Both depend on the fact that by suitable treatment the two actions can be separated from one another, so that a solution may be obtained which coagulates milk but has no proteolytic action, and on the other hand it is also possible to obtain an active proteolytic solution which has no milk coagulating properties.

Porter [1911] in support of Hammarsten's theory found that various commercial preparations, while coagulating milk, were actually anti-peptic. A full discussion of the subject is given by Oppenheimer [1913].

When investigating the development of enzymes from foetal life onwards I tested the properties of extracts of stomachs of young rabbits and compared these with similar extracts from adult animals. The differences found are recorded in this note.

According to Oppenheimer, pepsin is already present in the stomach of rabbits before birth, while according to Gmelin [1902] rennin is absent from the stomach of new-born animals. Other observers find that rennin develops

very rapidly after birth. Rakoczy [1910, 1911] and Van Hasselt [1910] found that in the case of calves the rennin disappeared rapidly during the first month after birth, while the pepsin increased greatly. The youngest animal employed by Rakoczy was apparently nine days old, but Van Hasselt does not state the exact age of the animals he used. In my experiments the stomachs of rabbits were taken as soon as possible after birth, washed out thoroughly and ground up with twice their weight of water. A little chloroform was added and after three days the liquid was filtered off and the filtrate tested. The stomachs of adult rabbits were tested in exactly the same way. The extracts were subjected to no further treatment with acid, sodium chloride or other substances such as were used by Hammarsten and others with a view to destroying the proteolytic or milk coagulating action as the case might be.

In the coagulating experiments 1 cc. of extract together with 5 cc. of milk was kept in a water-bath at 37° and examined every five minutes. Coagulation was considered complete when the test tube could be inverted without disturbance of the contents. Experiments were always repeated three times at least.

The proteolytic experiments were carried out on fibrin which had been finely minced, thoroughly washed and heated to 85°. 1 cc. extract + 40 cc. $N/20$ HCl + 1 g. fibrin were kept at 37° for a certain time and then filtered, the nitrogen being determined in the filtrate by Kjeldahl's method. Controls (HCl + fibrin) were also done and allowance was made for the amount of nitrogen originally present in the extracts.

The results are expressed as the number of cc. of decinormal nitrogen obtained from the fibrin digested.

Results of experiments:

A. New-born rabbits (each represents a different litter):

No.	Fibrin digested in 2 hours	Coagulation time
1	0·0 cc.	12 minutes
2	,,	18 ,,
3	,,	30 ,,
4	,,	12 ,,
5	,,	13 ,,
6	,,	22 ,,

B. Adult rabbits:

No.	Fibrin digested in 1 hour	Coagulation time
1	4·6 cc.	No coagulation in 2 hours
2	8·0	,, ,, 2 ,,
3	9·3	,, ,, 1 hour
4	23·8	,, ,, 2 hours
5	8·4	,, ,, 2 ,,

These results show the very wide differences which exist between the gastric extracts prepared from young and adult rabbits respectively with regard to their content of rennin and pepsin.

In some cases the extract from young rabbits' stomachs was incubated with acid and fibrin for 18 hours without any appreciable amount of digestion taking place.

At the end of every coagulation experiment, in the case of the adult extracts, a few drops of active rennin were added to the mixture of extract + milk. Coagulation now always took place, proving, if need be, that the absence of coagulation at first was not due to lack of calcium or other deficiency in the milk, but to lack of rennin in the extract. These facts, so far as they go, seem to argue against pepsin and rennin being identical. The extracts were exactly similar in mode of preparation and it is unlikely that one set would contain any inhibitory substance (for example removable by dialysis) which would be absent from the other. No stomach, either of young or adult rabbit, was an exception to the rule that in young animals we get rennin but no pepsin, and as the animal reaches adult life the rennin entirely disappears but pepsin is now found in the stomach.

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V. DISTRIBUTION OF ENZYMES IN THE ALIMENTARY CANAL OF THE CHICKEN.

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The presence of lactase in the intestines of animals and the non-adaptation of the pancreas and intestine to lactase by feeding with lactose was investigated by Plimmer [1906]. Lactase was always found to be absent from the intestine of chickens. A diet containing lactose had been used by us [1921] in feeding chickens from birth for a period of over three months. Examination of the birds' excreta showed that reducing sugar was absent therefrom, a fact which indicated that the sugar was assimilated. Assimilation of disaccharides is usually preceded by hydrolysis to monosaccharides, which would imply the presence of lactase in the alimentary canal, either in the intestine by adaptation or in some other part. The intestines of the cockerels in this group of birds were therefore examined, after they were killed, for the presence of lactase: it was not found to be present, and the non-adaptation of this organ was verified. If hydrolysis of lactose previous to assimilation occur, it must take place in some other part of the gut. The crop, pancreas and proventriculus were tested and lactase in small amount was detected in the crop. The investigation was then extended to the presence of other enzymes, as no information could be found in the literature about their occurrence in the alimentary canal of birds. The enquiry did not extend to the detection of all known enzymes, but was limited to those concerned in the digestion of the common foodstuffs.

Experimental.

The methods of preparing the enzyme solutions and detecting the presence of enzymes were in general in accordance with those usually adopted; in many cases a longer time of action (up to seven or ten days) was allowed, and in the case of the sucroclastic enzymes, proteins etc. were removed before testing for the reducing sugar formed by their action.

The various parts of the alimentary canal were always taken from chickens killed the same day, or not later than the day previously; on account of the

small size of the crop, proventriculus and pancreas, the organs from four to eight birds were collected and examined together. A single small intestine provided sufficient material, but in most experiments several were combined as the whole series of sucro- or proteo-clastic enzymes were tested for simultaneously. Separate tests were made for lactase. At least two experiments were made with each part, except the caeca.

Preparation of enzyme solutions.

The pancreas, on removal, was cut up into small pieces and ground with sand in a mortar; the ground mass was put into glycerol in which it was kept for several days in the presence of a few cc. of toluene. The solution was then prepared by diluting with rather more than an equal volume of water and filtering from sand, etc.

The other parts of the alimentary canal were cut open and washed with running water to remove the contents. The mucous membrane was scraped off, ground up with sand and water and extracted for 24–48 hours with water in the presence of a little toluene to prevent putrefaction. The aqueous portion was strained off through cloth to remove sand and larger pieces and used for testing for enzymes.

It was not possible to scrape off mucous membrane from the inside of the proventriculus. The organ is glandular, covered with numerous small teats, which, on pressing with a scalpel, emit a yellowish, viscous, distinctly acid secretion. This secretion was the material actually used after grinding with sand and mixing with water. Nothing could be scraped off the gizzard, the interior surface of which resembled parchment.

Detection of enzymes.

(*a*) *Diastase and invertase.* As substrates 100 cc. of 1 % starch solution and 50 cc. of 3 % cane sugar solution were used. Two portions were measured out with a pipette in separate flasks; a known volume of enzyme solution was added to one, and the same volume of boiled enzyme solution, after cooling, to the other; 2 or 3 cc. of toluene were added to each, the flasks corked and put into an incubator at 37° for one or more days. A test for starch by the iodine reaction was made from time to time with a drop removed from the mixture. At the end of the reaction time, the mixtures were washed into a 250 cc. measuring flask, a slight excess of colloidal ferric hydroxide added, any excess of the latter removed by a few crystals of magnesium sulphate, the volumes made up to the mark, the solutions filtered and reducing sugar tested for by the complete reduction of 10 cc. of Fehling's solution. The control solutions containing boiled enzyme did not reduce, or only gave a slight reduction due to sugar present in the extract.

(*b*) *Lactase.* The detection of lactase was carried out in a similar way to that of diastase and invertase, using 50 cc. of 4 % lactose solution as substrate. The enzyme and control mixtures were put directly into 250 cc. measuring flasks

and made up to volume after clearing with colloidal ferric hydroxide and magnesium sulphate. The reducing sugar was estimated by the reduction of 10 cc. of Fehling's solution. The observed difference in reading indicated whether hydrolysis had or had not occurred. No difference in reading was observed in the case of the intestine or proventriculus, but a small though distinct difference was always noticed in the case of the crop extract; it varied from 0·2 to 0·5 cc. in a total of 10 or 10·1 cc. This slight difference indicated an hydrolysis of 10–20 % of the lactose.

(*c*) *Lipase.* This enzyme was not looked for except in the case of the pancreas. Two exactly equal portions of oil in separate test tubes were made just alkaline to phenolphthalein with 0·1 *N* caustic soda. Enzyme and boiled enzyme solution were added. On keeping at 37° and occasionally shaking, the pink colour of the tube containing enzyme solution disappeared and it was restored by adding a few drops of the soda. This could be repeated several times and altogether from 1–2 cc. of alkali were added; the control tube did not change colour.

(*d*) *Proteoclastic enzymes.* Proteoclastic enzymes were detected by their action on Congo-red fibrin in neutral, acid and alkaline media. In the first case, a definite volume of enzyme solution and the same volume of boiled enzyme solution were put into separate flasks; in the other cases the same volumes of enzyme and boiled enzyme solutions were mixed with an equal volume of 0·2 *N* hydrochloric acid or 0·2 *N* sodium carbonate solution in separate flasks; 1 g. of Congo-red fibrin and 2 cc. of toluene were added to each and the several flasks were put in an incubator at 37° for one to seven days. Solution of Congo-red fibrin, which, in the case of hydrolysis, generally occurred in one or two days, was taken as indication of the presence of proteoclastic enzyme; solution did not occur in those flasks with boiled enzyme solution. No investigation was made of the products of the hydrolytic action.

Results.

The presence or absence of enzymes in the various parts of the alimentary canal is most easily seen from the following table:

	Crop	Proventriculus	Pancreas	Intestine whole	Duodenum	Ileum	Caeca
Invertase	0	0	.	+	.	.	0
Diastase	+	0	+	+	.	.	+
Lactase	+	0	.	0	.	.	.
Lipase	.	.	+	.	.	.	.
Proteoclastic in neutral	0	0	+slight	0	0	0	0
,, acid	+slight	+	+less rapid	+	+	+	0
,, alkaline media	0	0	+rapid	+slight	+	+slight	0

The distribution of the sucroclastic enzymes corresponds in most particulars with that in the animal; most animals have invertase in the intestine, lactase is present in some, absent in others: diastase and lipase are generally

present in the pancreas of animals. The proteoclastic enzymes show a difference: the animal has trypsin acting in alkaline media; the chicken in both alkaline and acid media. The intestine of the chicken has an enzyme acting most rapidly in acid medium, less rapidly in alkali. The proteoclastic enzyme of the proventriculus acts only in acid medium; the organ corresponds to the stomach of animals. The caeca, as expected, had no enzyme of this group, but contained diastase.

We wish to thank Prof. J. A. MacWilliam, F.R.S., for kindly allowing us to carry out these experiments in his laboratory.

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VI. THE AMINO-ACIDS OF FLESH.

THE DI-AMINO-ACID CONTENT OF RABBIT, CHICKEN, OX, HORSE, SHEEP AND PIG MUSCLE.

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A long series of food analyses has recently been made by Plimmer [1921, 1], who points out that by the ordinary routine method of analysis, in which the amount of protein is estimated by multiplying the nitrogen content by 6·25, no discrimination is made between the flesh of different animals. The protein of one animal is regarded as being the same as that of another. The work of Emil Fischer and Kossel and their pupils has definitely proved that the various proteins differ very widely in their composition as regards the amino-acids, and this difference is emphasised by the experiments on the food value of the individual amino-acids by Hopkins in conjunction with Willcock and Ackroyd, by Osborne and Mendel and other American investigators[1]. These chemical and biological differences are sufficient evidence that quality of protein in nutrition must be taken into consideration.

Complete analyses of the protein of the muscle of the ox, chicken, halibut and scallop have been made by Osborne and Heyl [1908] and Osborne and Jones [1909], and Drummond [1916] has made some analyses of muscular tissue by Van Slyke's method. Both the more complete analyses by Osborne and co-workers and those by Drummond do not show any marked difference in the amino-acid content of the various muscle proteins. The flesh of various animals shows such distinct appearances, different both to the eye and palate, that it seems probable that greater differences may exist, and that there may be smaller differences in the flesh from various parts of the same animal's carcase, such as back and leg. Some further amino-acid analyses have therefore been made.

The methods of protein analysis are far from perfect: Fischer's ester method for the mono-amino-acids, as he pointed out, is not quantitative: Kossel and Patten's method for the di-amino-acids, in spite of the numerous manipulations, is generally considered to be fairly accurate, but it has been largely superseded by Van Slyke's method which gives higher values for these amino-acids. Van Slyke's method also possesses the advantage of requiring only small amounts of protein and is more rapidly carried out. This method of protein analysis has been used in these experiments, since it was chiefly

[1] See summary by Plimmer [1921, 2].

desired to compare the muscle protein of several animals with a view to more complete data at a later time. A comparison of these results with those by Kossel's method has been made in a few cases. The results indicate that differences exist in the amino-acid content of the various muscle proteins. Duplicate analyses were always carried out; frequently these analyses were not so concordant as was expected. This inconsistency of the results was under investigation by Plimmer [1916] who tested the arginine determination; other details of the method are now being studied.

Experimental.

In the case of the smaller animals (rabbit, chicken) opportunity was taken of comparing the flesh of different parts of the body of the same animal. In other cases the flesh was taken from the thigh. The mode of operation was the same throughout. The flesh (about 350 g.) was freed from inside fat, minced and put into about 2 litres of boiling water containing 0·1 % acetic acid and heated for about ten minutes so as to coagulate the protein and remove the extractives. The liquid was poured off and the coagulated protein squeezed dry in a cloth. This procedure was repeated twice. The coagulated protein (about 200 g.) was then digested with 1 g. pepsin in 2 litres of 0·1*N* HCl, so as to separate nucleins, indigestible matter, etc. After digestion, which usually occupied about ten days at 37° the liquid was filtered off and the total nitrogen estimated. A portion containing about 6 g. of protein was then hydrolysed by boiling with hydrochloric acid added to the liquid so as to make a concentration of 20 %. The hydrolysis was carried on for 36 hours. The hydrolysed solution was evaporated to dryness *in vacuo*, made up to 250 cc. and two samples of 100 cc. were analysed by Van Slyke's method. This was performed as described except for the arginine estimation which was effected by Plimmer's modification [1916]. In the earlier experiments it was impossible to make determinations of amide N owing to the facilities for vacuum distillation not being adequate. The analyses were made in duplicate and the percentage has been calculated from the average.

Table I. *Nitrogen percentages.*

			Di-amino-acids						Mono-amino-acids			
	Amide N	Humin N	Total N	Amino N	Non-amino N	Argi-nine N	Histi-dine N	Lysine N	Total N	Amino N	Non-amino N	Total N of hydro-lysed solution
Rabbit, back	—	—	45·7	21·5	24·0	15	19	11·5	49	—	—	94·7
„ fore limb	—	—	44·1	17·9	27·7	8·8	30·9	5·5	50·7	—	—	94·8
„ hind limb	—	—	44·8	18·4	26·6	13	25	5·8	56·3	—	—	101·1
Chicken, breast	6·9	3	27	9	18	10	13	2	61·1	49·7	11·4	98·0
„ legs	5·5	1·3	25·5	15	10·5	8	7	11	68·5	66·6	1·9	100·8
Beef	6·3	0·5	28·5	15	13·5	13·3	5	11·2	55	26·8	28·2	90·3
Horse	2·9	0·9	37·1	18·8	18·3	14·9	10·5	11·6	70	58	11·9	110·9
Mutton	6·5	0·5	38·3	22·3	15·6	15	18	4·3	54	52	2	99·3
Pork	6·4	1·2	28·2	13·3	15	14	7	7	57	53	4	92·8

Relatively little difference can be observed from the figures for the different meats. The amide N is almost similar, in each case averaging about 6·0 % of the total N.

Table II. *Percentages of amino-acids. Giving the amount of amino-acids in* 100 *g. of protein.*

	Arginine	Histidine	Lysine	Total di-amino N
Rabbit, back	8	10	13	31
„ fore limb	5	19	5	29
„ hind limb	7	15	5	27
Chicken, breast	6	8	1	15
„ legs	4	4	10	18
Beef	7	3	10	20
Horse	7	6	9	22
Mutton	7	11	4	22
Pork	7	4	6	17

Humin N shows a difference. It is, if anything, higher in the white meats, *e.g.* breast of chicken 3 %, legs 1·3 %, pork 1·2 %, than in the red meats, where the average is 0·5 %, except in the horse, where 0·9 % was found. The explanation of this slightly higher value may be that the animal was not properly bled on slaughter.

Lysine figures are, with the exception of mutton, higher for the red meats, averaging about 11 %, while, of the white meats, rabbit limbs show only 5·5 %, chicken breast 2 % and pork 7 %.

Gortner and Holm [1920, 1], working with mixtures of pure amino-acids, have shown that tryptophan, and in the presence of aldehyde also tyrosine, and their analogues are the only known amino-acids which go to form humin. There is therefore no connection between the humin content and the lysine content of the meats; this is exemplified especially in the chicken, where the humin is high, and the lysine is low in the breast; and humin is low and lysine high in the legs. It may perhaps be mentioned that in the preparation of the di-amino-acids by the method of Kossel and Patten a distinct yellowish colouring adheres to the lysine portion.

At the same time too much reliance must not be placed on the humin as an estimation of tryptophan and tyrosine. Gortner and Holm [1920, 2] and Thomas [1921] have shown that tyrosine and tryptophan which go to form humin are not necessarily the only substances giving a reaction with the phenol reagent of Folin and Denis. Estimations of substances giving the blue colour with this reagent were made during the progress of this work, both before the removal of the humin and afterwards. In the case of chicken breast, a white meat, the readings before removal of the humin represented 4 % "tyrosine" whereas after its removal the readings represented 3·5 %. In the case of beef however—a red meat—the difference was greater, the former reading being 3·5 % and the latter 2·1 %, yet the humin N was much lower in the case of beef.

The arginine figures are more constant at about 14 or 15 % except in rabbit fore limb and chicken legs where the average is 8 %.

The histidine figures are less satisfactory, and exhibit perhaps a weak point in the method. In this connection it is of interest to point out that in the cases of abnormally high histidine the figures for the non-amino N are lower than normal and *vice versa*, *e.g.* beef 5 % histidine, 28 % non-amino N,

mutton 18 % histidine, 2 % non-amino N, while in other cases this observation cannot be made. This may be due, either to incomplete precipitation of the histidine by the phosphotungstic acid, or to washing. Work in this connection is in progress.

It is not possible to draw any conclusions from the figures of the mono-amino fraction, which account for about 55 to 60 % of the total N.

The average percentage of the di-amino N is 35.

Comparison with former work on the hydrolysis of meat is difficult, because, with the exception of Drummond [1916] on chicken meat, the other figures relate to the method of Kossel, which generally gives lower results than the Van Slyke method.

The above figures for chicken breast agree in the main with those of Drummond, his total hexone bases N 27·26 being the same as that above. The arginine figures are within 1 % and he records having used the same modification of that process as mentioned above. The figures for histidine and lysine are discordant, Drummond finding 8·45 and 9·81 respectively, while the total N of the mono-amino fraction is 4 % higher than that found by Drummond.

In order to compare the figures of Osborne and co-workers with the above it is necessary to refer to the percentages not of total N but of actual arginine, histidine and lysine. The figures for arginine are generally constant within 1 %, those for histidine are higher than Osborne's, while the lysine figures, owing to the calculation in Van Slyke's method, are dependent on the histidine values. Apart from the arginine values, only the beef of the present sets has given results comparable with those of Osborne, who found 7·5 % arginine, 1·8 % histidine and 7·6 % lysine against 6·8 % arginine, 2·6 % histidine and 9·6 % lysine in this experiment.

Summary.

1. Determinations have been made of the di-amino-acids of the protein of the flesh muscle of rabbit, chicken, ox, horse, sheep, pig by Van Slyke's method.

2. The red meats show a higher lysine content than the white meats.

I wish to take this opportunity of expressing my gratitude to Dr Plimmer, who suggested this work, for his kindness and guidance throughout the time I was under him, and also to Professor J. A. MacWilliam, F.R.S., for so kindly placing his laboratory at my disposal.

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ABNORMAL LEFT CORONARY ARTERY OF OX HEART COMMUNICATING DIRECTLY WITH THE CAVITY OF THE LEFT VENTRICLE NEAR THE APEX

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A hitherto undescribed abnormality was observed in an ox heart received by the above department about the end of October, 1921. Externally, the heart showed at the apex of the left ventricle a circular cyst-like structure. The heart was then held, with the cut end of the aorta pointing upwards, under a tap of running water. The water was allowed to run gently into the aorta, and the cyst-like structure was observed to bulge with fluid. On closer examination, a tubular vessel of arterial type about the calibre of one's middle finger was observed in the interventricular groove between the aorta and this structure at the apex of the left ventricle. This vessel appeared to follow the usual course of the descending branch of the left coronary artery towards the apex of the heart. It should be noted that, anteriorly, over the cyst-like dilatation, the ventricular muscle was quite deficient, and seemed to have been displaced by this abnormal structure.

Heart: weight with attachments of great vessels and fat, 95 ounces.

[Weight of another normal ox heart, 89 ounces.]

Length of heart, 24 cm.
Width of heart, 19 cm. } during rigor.

The auricles looked normal. The thickness of the walls of the left ventricle during rigor was about 5 cm. and did not differ materially from that of the normal ox heart.

With regard to the previous history and health of the animal, the following facts were obtained:

Age, rising 3 years old. Never off feed; very good feeder; always active; walked to sale from the farm ($\frac{3}{4}$ mile). Proportion of beef to live weight, fair average.

The calibre and thickness of the walls of the following vessels are given for the purpose of comparison:

Vessel	Calibre	Thickness of wall
Aorta (about 8 cm. beyond the valves) ...	3·0 cm.	8 mm.
Innominate artery (at its origin)	1·75 „	5 „
Abnormal artery (at its origin above the cusp)	1·5 „	1·5 „

It will be observed that, while the calibres of the innominate artery and abnormal artery are approximately equal, the wall of the latter is much thinner than that of the former.

Further dissection showed that this abnormal vessel arose about 1·25 cm. above the middle of the left posterior cusp of the aortic valve. The tip of the middle finger could be inserted into the vessel at its origin. A coronary artery, smaller in size, arose above the anterior cusp, but no artery arose above the right posterior cusp. In the normal ox heart the calibres of the two coronaries differ considerably, the left being the larger, but the normal left coronary artery did not admit the tip of the middle finger.

No other abnormal opening was noted at the base of the aorta. The aortic wall was healthy and the aortic valves appeared healthy, and, when tested by means of a stream of water directed into the cut end of the aorta, proved competent.

The abnormal vessel passed forward between the left auricular appendix and the pulmonary artery. The first branch came off the main vessel about 3·5 cm. from the cusp, and ran transversely outwards in the left auriculo-ventricular groove. Its calibre appeared similar to that of the normal right coronary artery. This branch was evidently the transverse branch of the left coronary artery and was of normal size.

The abnormal vessel passed along the interventricular septum giving off numerous small branches to the septum without any marked diminution in calibre until it reached just above the apex of the left ventricle anteriorly where it dilated into a cyst-like structure roughly conical in shape with its base anterior and its apex on a level with the inner surface of the left ventricle. With regard to the dimensions of the above structure, the diameter of the base of the cone was about 7 cm. while the height of the cone was about 6 cm. It will be noted that the height of this structure is practically equal to the thickness of the wall of the left ventricle.

The wall of the cone-shaped structure appeared similar in structure to the wall of the abnormal vessel but slightly thinner. It was lined by smooth endothelium, and its base was quite uncovered by cardiac muscular fibres, being apparently in direct relationship with the pericardium. The rest of the wall of the cone-shaped structure was attached firmly to the muscle of the left ventricle throughout its entire thickness.

The walls of the abnormal vessel and the dilated portion were apparently continuous. Anteriorly, where the wall of the dilated portion was not attached to the wall of the ventricle, the epicardium passed directly on to the wall of the dilated portion. At first, the union was not firm, the two being held together by loose tissue, but at a distance of about 2 cm. from the place where the ventricular muscle became deficient the wall of the dilated portion and the epicardium became fused apparently into one.

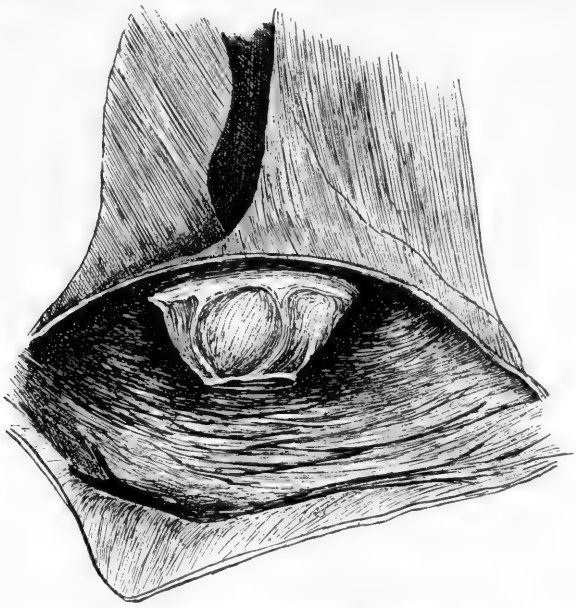
The cavity communicated directly with the left ventricle near its apex by a circular aperture, which was sufficiently large to admit the middle finger, and was guarded by a valve-like structure. Examination showed that the latter consisted of:

(1) an inner fibrous ring, diameter 1·5 cm., forming the circumference of the aperture,

(2) an outer fibrous ring, 3·5 cm. in diameter,

(3) thin fibrous material covered by smooth endothelium, stretching between the two rings and thickened by six or seven fibrous bands running radially between the two circular fibrous rings.

No other abnormality, developmental or acquired, was noted in the right or left chambers of the heart. The interventricular septum did not appear to be in any way abnormal, and the coronary veins both right and left were small. They did not appear to be enlarged on either side of the heart.



Lateral view of valve-like structure between cavity of left ventricle above and cyst-like dilatation below.

From its origin and course, this abnormal vessel was taken to be a left coronary artery—the abnormality affecting more particularly the descending branch of the left coronary artery. The dilatation of the terminal portion of the vessel at the apex was difficult to explain. This dilatation might have been due wholly to the developmental abnormality, or it might have been acquired mainly. If the latter supposition was correct, the dilatation would have been of the nature of an aneurismal dilatation. Support might be lent to this view by the fact that, when the dilated portion was distended with water, it was noted that at two or three places the wall was much thinned. The distension might have been brought about by the escape of blood from the left ventricle throughout the greater part of systole before the opening between the cavities of the left ventricle and the dilated part was closed by the contraction of the left ventricle towards the end of systole.

Developmentally, no explanation of the abnormal coronary and communication with the left ventricle has been suggested.

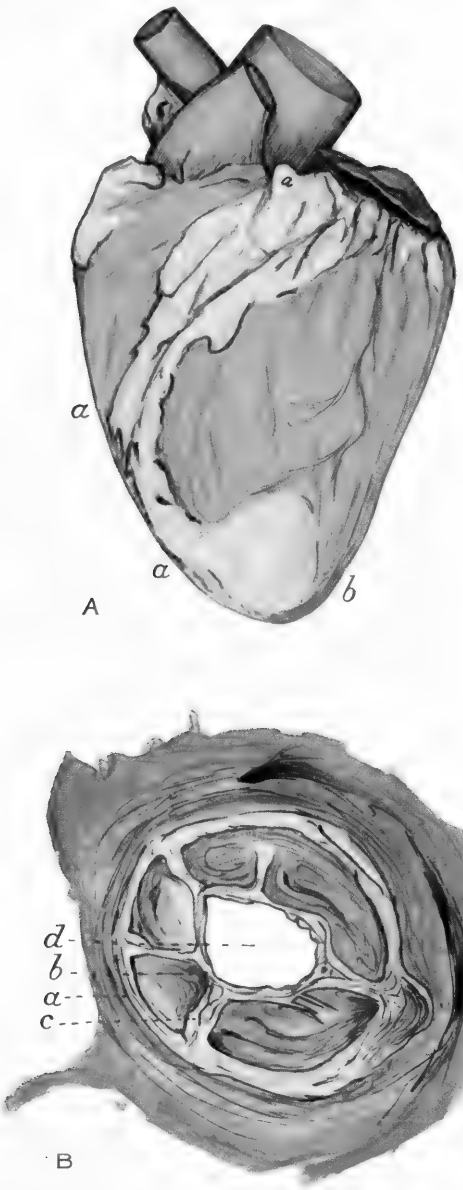


Fig. A. *a*, *a*, *a*, track of abnormal descending branch of the left coronary artery; *b*, cyst-like dilatation at apex.

Fig. B. Wall of dilated part at apex has been opened and held back.

Appearance presented by the pseudo-valvular structure on looking at the apex of the heart: (*a*) external fibrous ring; (*b*) internal fibrous ring; (*c*) radial fibrous band; (*d*) communication between the left ventricle and the abnormal coronary.

The only related case on record was described by Mr H. Blakeway in the *Journal of Anatomy*, vol. LII, p. 354. The heart in this case—a child which lived 36 hours—had amongst other abnormalities no direct communication between the left ventricle and the aorta, but an indirect one by means of the anterior interventricular branch of the left coronary artery. Mr Blakeway considered the question of the possibility of the origin of the abnormal communication between the aorta and the left ventricle as being due to some developmental peculiarity of the bulbus cordis. He, however, rejected this consideration.

The actual course taken by the blood in the abnormal ox heart forms an interesting speculation. Before the heart went into rigor, the left ventricle was artificially compressed above the apex to imitate systole, and at the same time a stream of fluid under pressure was directed against the pseudo-valvular opening by means of a tube introduced through the aorta past the aortic valves. Practically no fluid escaped into the dilated part. Again, fluid was allowed to run into the aorta. The aortic valve being competent, most of the fluid passed along the abnormal channel. The fluid entered the cavity of the left ventricle (the left ventricle being empty) through the pseudo-valvular opening, if the left ventricle was not compressed.

It would appear that during the greater part of systole leakage took place directly from the left ventricle to the dilatation at the apex. In all probability, the opening between the left ventricle and the dilated portion would not be closed by ventricular contraction except towards the end of systole. During diastole, unless the pseudo-valvular structure acted as an efficient valve, there must have been free communication between the aorta and the interior of the left ventricle, and the diastolic pressure in the abnormal vessel and in the interior of the left ventricle must have been equal to the pressure in the aorta. After the wall of the distended part at the apex of the left ventricle was laid open, a strong stream of fluid was directed against the valve-like structure. It appeared to act as an efficient valve except when the left ventricle was distended or relaxed.

As bearing on the accepted relation between increased diastolic intraventricular pressure and dilatation and the striking development of dilatation and hypertrophy in aortic regurgitation in man, it is noteworthy that in this ox regurgitation into the left ventricle with its concomitant high diastolic pressure was not associated with appreciable dilatation or hypertrophy.

The following notes give a brief account of the microscopical appearance of the parts of which sections were made:

(1) *Ventricle* (left).

Muscle, healthy. Nothing abnormal noted.

(2) *Innominate artery.*

Intima, healthy.

Media, towards inner part of media regularly arranged bundles of plain

muscle circularly disposed and elastic fibres; towards outer part of media, amongst the circularly disposed plain muscle and elastic fibres irregularly arranged groups of plain muscle, many running longitudinally.

(3) *Wall of abnormal coronary artery.*

Endothelium, healthy.

Subendothelial elastic layer, quite well marked. Wall varies in thickness, the thinner parts being at most one-half the thickness of the thicker portions.

Thicker portions: large amount of plain muscle arranged in bundles; rather granular looking elastic tissue between the bundles—apparently split longitudinally in places.

Thinner portions: much less plain muscle than the preceding; towards the centre of one portion of the media, small oval-shaped area which does not stain well: nuclei stain fairly well but are variable in shape. The elastic tissue apparently shows large coarse granules and it appears to become fragmented transversely into more or less elliptical portions.

Externa, well marked.

(4) *Wall of dilated portion of abnormal coronary artery.*

Two layers: (1) External, epicardium;

(2) Internal, part corresponding to wall of the abnormal coronary artery; practically no plain muscle or elastic fibres; rather degenerate-looking connective tissue showing nuclei which stain fairly well, fibrils, and perhaps "ghost-like" elastic fibres.

Interior to the above is a fairly thick endothelial and subendothelial layer showing connective tissue and elastic tissue arranged parallel to the inner surface; nuclei stain well.

Endothelium appears to show proliferation of its cells, the deeper layers of which show signs of organisation.

(5) *Lining of cavity and subjacent myocardium.*

(*a*) Endothelium, normal; no proliferation.

(*b*) Subendothelial elastic layer, fairly well marked.

(*c*) Layer of more or less homogeneous tissue taking up eosin stain, no sign of elastic or muscle fibres.

(*d*) More or less continuous layer of about $\frac{1}{6}$th thickness of (*c*), consisting of heart muscle fibres and white fibrous connective tissue.

(*e*) Vascular layer, thinner than (*d*).

(*f*) Heart muscle proper.

(6) *Junction of dilated end of abnormal coronary artery with ventricle.*

Epicardium and wall of distended portion can be seen separated by heart muscle; the heart muscle ceases and the epicardium and the wall fuse loosely at first, but firmly within a distance of 2 cm. from the point where the heart muscle ceases; epicardium at the point where the heart muscle ceases becomes much thinned quite abruptly, and continues thin for about a distance of

1·5 cm. as it lies in direct relationship with the wall of the dilated portion; it then becomes thicker, approximating to its original size.

(7) *Branch of abnormal coronary artery.*

Healthy arterial wall.

(8) *Branch of right coronary artery.*

Nothing abnormal to be noted.

(9) *and* (10) *Coronary arteries from normal ox heart.*

Left: the larger vessel; wall of left varies in thickness considerably.

Conclusions in regard to:

I. Course taken by blood during life in the abnormal vessel.

(1) *During systole.* Probably regurgitation occurred from the left ventricle throughout the greater part of systole through the abnormal communication at the apex. This would give rise to a pulse wave apart from the question of the quantity of blood regurgitated. Another pulse wave would be sent along the abnormal vessel from the aorta. In this way the abnormal vessel would be subjected to strain, and the cyst-like part would be subjected probably to the greatest strain. The movement of blood in the abnormal vessel would perhaps be from the apex of the left ventricle towards the aorta.

(2) *During diastole.* The blood-flow in the abnormal vessel would in all likelihood be from the aorta to the left ventricle. The diastolic pressure in the left ventricle and in the abnormal coronary would be high, viz. aortic pressure. Hence conditions would be favourable for increased strain on the abnormal vessel and dilated part during diastole and on the dilated part more particularly during diastole. On the whole, there would be a relative stagnation of blood in the abnormal vessel.

II. Respective proportions of the abnormality, congenital and acquired.

It would appear clear that the communication between the left ventricle and the coronary was developmental wholly. The pseudo-valvular structure must have been present before birth as a congenital peculiarity.

That the abnormal coronary and dilated portion were subjected to abnormal pressures and in consequence became expanded is concluded from the following:

(1) Varying thickness of the wall of the abnormal coronary and dilated part.

(2) The wall does not show the typical structure of a normal artery.

(3) Irregular arrangement of bundles of unstriped muscle etc. in wall.

(4) Evidence of impaired nutrition of portions of the wall of the abnormal vessel.

(5) High diastolic pressure.

In all probability, had the animal not been killed, it would have died at some period of rupture into the pericardial sac through one of the thinned portions of the wall of the dilated portion at the apex.

I am indebted to Professor J. A. MacWilliam for his help and permission to publish the above, and to Mr George C. Kelly for the sketches.

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SOME APPLICATIONS OF PHYSIOLOGY TO MEDICINE.

I.—SENSORY PHENOMENA ASSOCIATED WITH DEFECTIVE BLOOD SUPPLY TO WORKING MUSCLES.

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ACCURATE knowledge of the effects of defective blood supply to the various tissues and organs is obviously of great importance in view of the innumerable conditions of stress, derangement, and disease in which this factor comes into play, with manifold results in the way of disturbed or impaired functions in the different systems of the body. "Defective supply" naturally covers different conditions—quantitative deficiency in normal constituents, or the presence of abnormal and injurious constituents, or inadequacy as regards the volume, pressure, and rapidity of flow of normal blood. This communication deals with the last-named—certain effects of deficiency in the supply of normal blood to normal muscles.

Many impairments of functional activity from more or less extensive interference with blood supply have long been known, such as the weakening of the heart muscle from deficient coronary supply and the common occurrence of fibrillation after sudden coronary obstruction; the effects on the brain in the form of giddiness, faintness, or loss of consciousness; and the primarily exciting and secondarily depressing influences exercised powerfully on the medulla (respiratory, vasomotor, and cardio-inhibitory centres, etc.) and on the spinal centres from sufficiently extensive or sudden acute lack of blood supply; also the derangement or stoppage of kidney function from similar interference.

Various observations are on record dealing with the functional behaviour of excised organs and muscles artificially perfused with blood or in the exsanguine condition, and also observations on the effects of artificial interference with the blood supply of organs and muscles *in situ* in animal experiments. Under such conditions there is of course no information obtainable as to sensory phenomena attendant on altered blood supply in the conditions of rest and activity.

The present inquiry deals with the behaviour of human muscles temporarily deprived of their blood supply while their normal innervation remains intact; the sensory phenomena

recognizable in the states of rest and activity are examined and brought into relation with other functional conditions, such as changes in contractile power, etc.

Methods of Experiment.

The forearm was investigated (*a*) while the normal circulation was going on, and (*b*) when the blood supply was stopped, the limb either retaining its blood in a stationary condition or being rendered exsanguine before the circulation was arrested —that is, the "congested arm" and the "ischaemic" arm were examined with arrested circulation. The circulation was stopped by a blood pressure armlet applied to the upper arm, which was rapidly pumped up to a constricting pressure much above what was necessary to produce arterial obliteration in the particular individual examined—that is, an armlet pressure largely exceeding the systolic pressure. When this was done in the usual way, as for the measurement of systolic blood pressure, a "congested arm" was obtained containing a large amount of stationary blood shut off from the general circulation, the veins becoming prominent and tense. To obtain the bloodless or ischaemic arm an elastic bandage was first applied to the hand and arm, and removed after the armlet had been pumped up as described.

In the congested arm the sensory phenomena are naturally complex, being partly attributable to conditions attendant on the arrest of the circulation as influencing the muscles, etc., and partly to the discomfort caused by the venous turgescence. To avoid the latter complication the method of the ischaemic arm is employed; the sensations induced by muscular activity in presence of acute want of blood can then be examined.

Under these conditions muscular action was tested in various ways. Graphic records of the flexor muscle of the middle finger were made by means of a Mosso's ergograph, the voluntary flexion movements being made in regular series—one in one second or in two seconds, etc.—timed by a metronome, while the weight lifted at each contraction varied in different experiments from 1 to 3 kg. The behaviour of the muscle in different conditions, the amounts of mechanical work done as measured in kilogram-metres, the development of fatigue, etc., were graphically recorded, while the sensations associated with different phases were noted. The results as regards fatigue, etc., will be described elsewhere, the present communication having to do with the sensory phenomena.

Another method is to use a series of grasping movements with the hand, bringing them to bear on a dynamometer or a dynamograph; this method is in some respects less precise than the preceding.

Another mode of experiment was to use the abductor indicis muscle, working against the resistance of a strong elastic band embracing the fingers; successive abduction movements of the fingers were then made in regular series; only the hand was rendered ischaemic in this case, the armlet being applied to the forearm. The hand was supported on a

table with the palmar surface downwards. Graphic records can be obtained by making the movements of the finger inscribe on a moving smoked paper.

Ischaemia of the Resting Arm.

In observations made by these methods it was found that simple deprivation of blood in the ischaemic limb for periods up to twenty minutes caused no great sensory effects, only coldness in the bloodless part, with an inclination to shift the position of the limb, and a certain amount of discomfort from the continued constriction by the obliterating armlet; the absence of pain is to be noted.

Muscular Action in the Ischaemic Arm.

Muscular action in the ischaemic limb soon becomes painful, and when carried to the point of "fatigue" is acutely painful. "Fatigue" is indicated by inability to go on executing contraction movements even of greatly reduced range. This index of "fatigue" is convenient for comparing the state of matters in normal and ischaemic muscles, though it does not represent inability of the muscle to do more mechanical work in more favourable circumstances—for example, with less resistance opposing the contraction, a lighter weight to lift, etc. It is a useful index of the stage of enfeeblement of the voluntary contractile power with which the sensory manifestations in muscles under different conditions can be correlated. The actual time necessary to induce fatigue and the number of movements that can be executed prior to this point are of course largely influenced by the weight used; with a sufficiently light weight the movements can be kept up for hours without the occurrence of fatigue in the normal arm while the circulation is intact. Under normal conditions the phenomena of fatigue as shown by ergograph records are well known. The associated sensations as the fatigue point is approached take the form of a sense of increased effort being necessary to raise the weight even for a short distance, an increasing disinclination to go on making the successive efforts, aching or dull pain in the central part of the forearm, etc. We have often found a certain amount of local tenderness to pressure in the fatigued muscles, lasting for some little time after action has been discontinued.

In the ischaemic arm the fatigue point is reached much more rapidly, often in one-half or one-third the time needed in the normal arm, with a proportionate diminution in the number of contractions executed, the more rapid development of extensive weakening at a relatively early stage, etc. Pain develops and by the time the fatigue point is reached becomes severe; further efforts at contraction movements lead to distressingly acute pain and the desire for relief becomes urgent, while there is a strong disinclination to attempt further efforts.

Distribution and Characters of the Pain.

The pain is felt over the flexor aspect of the forearm and is most intense in the central part of the forearm; it is

specially marked from wrist to elbow along the line of the flexor digitorum sublimis. It seems to be centred in the belly of the working muscle with a good deal of spreading, but there is, as a rule, no referred pain in more distant parts; in one subject pain in the palm of the hand was complained of. The pain goes on increasing progressively while contractile activity is kept up; there is no remission, as may sometimes occur markedly in the normal arm, where, working with a suitable load, decided aching may develop at a comparative early stage, to pass off more or less completely at a later stage.

It is to be noted that the pain, increasing to almost intolerable severity in some of these experiments, arises from exercise of a comparatively small amount of muscular tissue —the limited portion of the flexor muscle engaged in moving a single finger—in presence of an acute lack of blood supply, involving urgent want of oxygen (anoxaemia) and its consequences, with excessive accumulation of metabolic products, acids, and other bodies. The pain is no doubt protective in character, tending to limitation of effort and shielding the muscle from being spurred on to further and injurious activity. Discontinuance of further effort for short periods does not remove the pain, but it is almost immediately relieved—in a few seconds—by readmission of blood into the limb by removal of the obliterating pressure of the armlet. Contractile energy, on the other hand, recovers gradually and slowly; it takes some time to be fully re-established, and even then is apt to fail more readily than before on repetition of the experiment. It is evident that the pain and the depression of contraction force do not run parallel in the ischaemic arm.

Relation of Pain to Weakening of Contraction Force.

The conclusion just stated is supported by the fact that in the ischaemic arm the development of pain in the course of a successive series of contractions is much greater in proportion to the weakening of contraction force than in the arm with intact circulation; with an equally extensive cutting down of the energy of movement in the two types of arm, as shown by the ergograph tracings, there was sharp pain in the ischaemic arm at a stage when there was only a tired feeling with some aching in the normal arm; pain and weakening of contractile force were differently related to one another in the two cases.

It may be noted that in the normal arm slight aching or local tenderness may last for some little time after the exercise of the flexor muscle (as recorded by the ergograph) has been discontinued, while in the ischaemic arm the sharp pain disappears quickly on re-establishment of the circulation. There is reason to believe that in fatigue following severe muscular exertion under normal conditions (for example, football, etc.) the muscular aching and tenderness, felt for a considerable length of time afterwards, especially in individuals out of training, are dependent on a mechanism of production that is not identical with that of the pain caused by working an ischaemic muscle.

The production of severe pain from a small amount of skeletal muscle working with its blood supply cut off recalls the agonizing pain excited by excessive contraction of a small amount of unstriped muscle in a bit of bile duct in gall-stone colic, or of ureter in renal colic, etc. Of course it does not follow that the mechanism of pain production is similar in the two kinds of muscle—the unstriated and the striated.

Observations on the Abductor Indicis Muscle.

Experiments with the abductor indicis muscle gave results essentially similar to those described above. For example, in an experiment when a certain strength of elastic band was used to resist the abduction movement, a series of about 240 movements could be carried out in the normal state at the rate of one per second before the "fatigue" point was reached—that is, the point where any abduction movement failed to occur against the resistance of the band; this was attended by only slight discomfort and aching—where the latter was present at all. In the ischaemic hand the fatigue point was reached at about one hundred contractions—that is, in less than two minutes, as compared with four minutes in the normal state; this was attended by pain, which spread more or less over the dorsum of the hand, though most sharply felt in the working muscle. Stoppage of the efforts at abduction for a minute did not lead to removal of the pain, but the latter was promptly relieved by re-establishment of the circulation; contractile power recovered much more slowly, and was more easily fatigued subsequently. Some minutes later the hand was again rendered ischaemic, and kept in that condition with quiescent muscles for ten minutes; the hand became cold, but there was no pain, simple ischaemia having, as described above, no appreciable effect in this respect. Abduction movements of the index finger were then performed as before; there was painful fatigue after about sixty-five movements; the pain was removed as before by readmission of the blood. The usual well-known flushing occurred after the period of ischaemia; sensations of tingling gradually developed somewhat later.

Effects of Continuous Muscular Tension.

Experiments were also performed with the middle finger flexor muscle kept voluntarily contracted to sustain the ergograph weight at a certain level instead of making a series of consecutive lifting efforts as already described; graphic records of the behaviour of the muscle were made. Continuous motor effect failed to preserve the initial level beyond a certain time, which varied according to the weight employed, etc.; then came a general progressive decline, varied by minor irregularities in the slope of the tracing, until after a time the weight sank back to the resting position. This "fatigue" is attended by comparatively little subjective disturbance, even in the ischaemic arm. There was disinclination to keep up the tension of the muscle, which seems to need more and more voluntary effort, with some discomfort and aching—the latter felt chiefly in the upper arm and the finger—probably attributable not to the muscle itself but to

the mechanical conditions connected with the fixed position of the limb and pressure on the skin of the finger by the loop at the end of the cord which supports the weight. There is evidently a notable difference as regards pain production between an alternately contracting and relaxing muscle doing mechanical work and the condition of sustained tension necessary to maintain the weight at certain levels. Similar results were obtained with the abductor indicis.

Relation to Pains of Angina Pectoris, Intermittent Claudication, etc.

It need hardly be pointed out that the foregoing observations have a close bearing on the problems associated with the production of the pain of angina pectoris, showing as they do how readily acute pain can be excited in skeletal muscle working with lack of blood supply, the pain developing while the contractile power, though to some extent weakened, is still sufficient to execute movements of considerable range and energy—that is, long before complete fatigue.

There is every reason to believe that processes of the same nature, with a similar production of pain of varying grades of severity, up to the agonizing suffering of fully developed angina, occur in cardiac muscle compelled to work with a blood supply that is inadequate—absolutely or relatively to the amount of work which the arm has to perform. Sir James Mackenzie has emphasized the conception of anginal pain as an expression of exhaustion of the cardiac muscle, commonly associated with a defective coronary blood supply and a susceptible nervous system. He has laid stress on the production of the symptoms of heart failure—pain, breathlessness, giddiness, faintness—as expressions of impaired functions of organs which fail to receive a blood supply adequate to the needs of their normal activities in consequence of a defective output of blood from the heart, the latter itself suffering from insufficient blood supply to its muscular walls; heart failure is thus recognized, not by direct examination of the organ itself, but by the functional effects of diminished blood supply to various organs.

It may be added that the results of the present experiments have an obvious application to the phenomena of the condition called "intermittent claudication," as seen in the legs of men and horses, in which muscular exertion is interrupted by attacks of pain, loss of power, coldness of the limbs, etc. These symptoms can be definitely explained: in consequence of blocking of the main artery, or disease or spasm of the vascular walls, the blood stream has been reduced to such an extent that, while it may suffice to supply the muscles in the resting state, it is quite inadequate for their greater requirements during activity—the results (pain, etc.) of the defective blood supply are of the same nature and mechanism of production as those demonstrable in the ischaemic arm of the healthy subject.

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A METHOD OF ESTIMATION OF DIASTASE IN BLOOD.*

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THERE is a general acceptance of the view that estimation of blood diastase is of distinct diagnostic value, and it is unfortunate that certain features of recent methods render them not entirely satisfactory. Stocks (1916), and later Harrison and Lawrence (1923), adopting Wohlgemuth's method, incubate a series of dilutions of blood serum, or blood plasma, with starch and obtain their results from the colour reaction which occurs on the addition of iodine. In the first place it is very difficult to obtain in general practice a sufficiency of serum for the purpose of the test (from 1 c.c. to 3 c.c.) without veni-puncture, and, in the second place, sera do occur occasionally of which the tint definitely masks the delicacy of the colour reaction. Myers and Killian (1917), using the Lewis and Benedict method of blood sugar estimation, determine the amount of sugar formed from a known quantity of starch by 2 c.c. blood after incubation for a definite period, calculating their results in terms of the percentage of starch reduced. Here again the question of veni-puncture arises, and in addition that of the inaccuracies which the picric acid methods of sugar estimation present as compared with more recent means of blood-sugar determination.

In view of these difficulties the problem of the estimation of blood diastase was approached, and, by an adaptation of MacLean's (1919) method of blood-sugar estimation, a means has been evolved which has the advantage of simplicity and accuracy, while it necessitates the minimum of discomfort to the patient. It also permits of an almost concurrent reading of the blood-sugar and the blood-diastase figures, dispensing with the need for two different techniques. For the method proposed 0·2 c.c. blood only is required—an amount which can easily be obtained by pricking the ear or the finger. The final result depends upon a determination of the patient's blood sugar by MacLean's method, and upon a second determination after 0·2 c.c. of the patient's blood has been incubated for half an hour at 37°C. with 1 c.c. of a 0·1 per cent. solution of starch.

The Effect on Blood Sugar of Incubation at 37° C.

In view of the fact that the literature of the subject gives most discordant views on the problem of glycolysis in blood, it was thought best to make an investigation of the question. It was found that under the experimental conditions described below no glycolysis was in evidence. Freshly drawn blood to the amount of 0·2 c.c. was pipetted into a small Erlenmeyer flask containing 2·8 c.c. normal saline solution. The suspension was incubated for half an hour in a water-bath at an accurately maintained temperature of 37°C. During this period a direct control estimation of the sugar in another sample of 0·2 c.c. was done. When the incubation time was completed the amount of sugar

* Work carried out in the tenure of a Carnegie Research Assistantship.

present in the specimen was estimated. Table I shows the results in six cases taken from some forty.

TABLE I.—*The Effect of Incubation on the Sugar in 0·2 c.c. Blood.*

Case.	Control percentage.	Percentage after incubation.
1	·081	·082
2	·097	·097
3	·102	·106
4	·13	·13
5	·097	·097
6	·131	·134

No appreciable change in the sugar content occurs on incubation of 0·2 c.c. blood in 2·8 c.c. saline solution at 37° C. for half an hour. It should be stated here that throughout the experiment and also throughout the entire diastase investigation, strict aseptic precautions were observed to exclude the possibility of bacterial action during incubation.

THE PROPOSED METHOD DESCRIBED.

Into one of two 100 c.c. Erlenmeyer flasks 1·8 c.c. of 0·9 per cent. saline solution and 1 c.c. of 0·1 per cent. starch solution (*i. e.*, 1 mg. starch) is accurately pipetted, while into the other (control flask) exactly 2·8 c.c. of 0·9 per cent. saline solution is introduced. 0·2 c.c. blood is withdrawn from the finger into a special MacLean pipette* and carefully ejected *into* the fluid of the first flask, the point of the pipette being held beneath the surface of the solution, while the flask is held at an angle. The pipette is rendered free from blood by repeated washing with the clear fluid into which the blood has just been delivered. The flask is then gently shaken with a circular movement so as to mix thoroughly the blood and the solution. A second sample of 0·2 c.c. blood is similarly delivered and washed into the control flask. Both flasks, provided with rubber stoppers, fitted with capillary points, are placed in a water-bath, the temperature of which is accurately maintained at 37° C. Incubation is allowed to proceed for exactly half an hour, at the end of which time the flasks are removed and 21 c.c. of MacLean's acid sodium sulphate solution is added. In the case of the first flask the addition should be made immediately on removal from the water-bath so as to stop the action of the diastase. The subsequent steps in the estimation are precisely as described by MacLean. Briefly the treatment is as follows: The flasks are heated till the boiling-point is just reached. 1 c.c. dialysed iron is added to each and after cooling under the water tap the contents are filtered. To 20 c.c. of each filtrate is added 2 c.c. alkaline copper solution. The resulting solutions are then boiled for six minutes over a flame suitably adjusted to effect distinct boiling in one minute forty seconds. At the end of that period the flasks are immediately plunged into cold water and cooled thoroughly. 2 c.c. of 75 per cent. HCl (or H_2SO_4) are added, and after effervescence has finished and after standing for one minute with occasional agitation, the iodine content of the solutions is found by titration with N/400 sodium thiosulphate. During titration, in the case of the first flask, a variation of colour is seen ranging from dark amber to

* To be obtained from Hawksley & Son, Wigmore St., London.

leaden and to pale blue, the final disappearance of which marks the completion of titration; in some cases when the blue colour is very faint the end-point may be rendered more distinct by the addition of a drop of 3 per cent. starch solution. The amount of sodium thiosulphate used is noted and its exact glucose equivalent ascertained from the glucose-thiosulphate table, or very much better, from a plotted graph. The second flask is treated in exactly the same manner except that starch must be added to complete the titration.

Calculation.

The result is calculated as the percentage of soluble starch transformed to sugar (calculated as glucose) by the 0·2 c.c. blood employed. The amount of starch used is 1 mg., and the difference between the sugar contents, measured in fractions of a milligramme, of the two samples of 0·2 c.c. blood will be equivalent to the amount of starch reduced to sugar.

Suppose that, reading from the glucose-thiosulphate graph, the sugar content of the control preparation is 0·164 mg., while for the starch preparation it is 0·259 mg. Since the amount of filtrate used corresponds to $\frac{4}{5}$ of 0·2 c.c. blood, that amount of blood would contain 0·205 mg. sugar in the one case and 0·323 mg. sugar in the other. The difference, 0·118, is equivalent to the amount of starch transformed to sugar by incubation with 0·2 c.c. blood, so that the diastatic index in this case is 11·8. Allowance should, of course, be made for any slight reducing action of the soluble starch. During the course of this investigation the starch solutions were repeatedly tested for any such action, and in no case was it found necessary to make correction.

In view of the fact that glycolysis does not occur on incubation of 0·2 c.c. blood under the conditions of the experiment, there appears to be no good reason for running a control. The half hour during which the starch preparation is incubating may be very conveniently occupied in making a direct estimation of freshly-drawn blood.

DETAILS OF METHOD AND RESULTS.

Accuracy.—It may seem superfluous to call attention to the need for accuracy in this estimation, but in view of the fact that determinations are made in milligrammes and fractions of milligrammes, perhaps a few points which lend themselves to precision are worthy of note. Cleanliness and attention to aseptic precautions should be maintained throughout. Burettes, flasks and pipettes should be thoroughly cleaned. Antiformin or a solution of potassium bichromate in sulphuric acid is a very useful help. Standardised burettes and pipettes should be used. During the process of the boiling of the blood filtrate and the copper solution, the manometer should be carefully watched for any change in gas pressure and any necessary adjustment made.

Preparation of the starch solution.—Lintner's soluble starch was used throughout this investigation. The solvent was physiological salt solution prepared with water, double (glass) distilled and practically neutral to rosolic acid. The presence of the salt solution prevents hæmolysis, and tends to accelerate the action of diastase. A litre of 0·9 per cent. NaCl is prepared with re-distilled water, and 0·2 grm. of soluble starch weighed out and suspended in about 5 c.c. of the saline solution. Sufficient saline to make 200 c.c. is measured out and heated in a large flask almost to boiling-point,

when the starch suspension is carefully added drop by drop, the contents of the flask being shaken after each addition. A reflux condenser is then attached, and the solution allowed to boil for about ten minutes. The resulting starch solution is homogeneous and transparent, and allows of an intimate association of the enzyme and substrate. It is not advisable to submit the small quantity of starch to prolonged boiling, as is recommended by certain authors (Sherman, Kendal and Clark, 1910), who, however, use a larger quantity of starch. Table II shows how the diastatic index tends to be lowered when starch which has been boiled for some time is used in the test.

TABLE II.—*Showing Variations in Diastatic Activity when* 0·2 *c.c. Blood is treated with Starch Solutions boiled for Different Lengths of Time.*

Case.	Starch solution boiled for— ¼ hour.	1 hour.	2 hours.
	Reducing sugar formed.		
1	·251 mg.	·230 mg.	·200 mg.
2	·180 „	·145 „	·127 „
3	·230 „	·165 „	·16 „
4	·108 „	·098 „	·08 „
5	·143 „	— „	·035 „

The starch solution should be made with great care, and should be freshly prepared every second or third day. No advantage seems to be gained in the use of glycogen instead of starch (Table III).

TABLE III.—*Showing Comparative Activity of Diastase on Starch and Glycogen.*

Case.	Blood sugar.	Sugar formed. 0·1 per cent. starch.	0·1 per cent. glycogen.
	per cent.	mg.	mg.
O. B—	·13	·200	·130
H. M—	·10	·087	·08
E. A—	·11	·141	·14
A. T—	·09	·093	·083
A. I—	·195	·315	·320

The diastatic figures of six normal subjects are given in Table IV. The figure seems to vary in different individuals, the average being about 8 to 10—a figure somewhat lower than that of Myers and Killian (1917), and similar to that of Stocks (1916). From data obtained it would seem that the normal index should not exceed 15.

TABLE IV.—*Showing the Diastatic Activity of Blood in Normal Cases.*

Case.	Age.	Sex.	Diastase.	Blood sugar.
R. R—	30	M.	8·5	·102 per cent.
E. A—	22	M.	9·0	·097 „
A. T—	33	M.	11·8	·134 „
A. G—	34	M.	8·8	·102 „
C. R—	30	M.	7·1	·131 „
W. S—	30	M.	6·5	·106 „

Table V gives the diastatic activity of three cases of diabetes. In the case D. G—, cardiac complications and pulmonary fibrosis were present.

TABLE V.—*The Diastatic Activity of the Blood in three Abnormal Cases.*

Case.	Age.	Sex.	Date.	Blood diastase.	Blood sugar per cent.	Urine diastase.	Urine sugar per cent.	Diet.	Remarks.
A. I—	39	M.	13/12/22	24·5	·19	—	·5	Restricted	—
D. G—	28	M.	16/2/23	14·5	·111	6·6	·25	Not restricted	No albumin in urine.
A. C—	30	M.	8/3/23	2·375	·122	10	·66	Restricted	No albumin in urine.
				3·125	·111	12	·4		

In the course of this investigation certain points have arisen which seem to suggest that the hourly diastatic activity of the blood should not be represented by a constant figure as has been suggested by some. An investigation of the matter has already been commenced and certain observations have been made.

ADVANTAGES OF THE PROPOSED METHOD.

Sugar estimations with picric acid very frequently give results which are decidedly too high. This may be due to—

(*a*) The disturbing effect of creatinine, which varies in amount pathologically and physiologically. In certain renal conditions creatinine is found in excess. On ingestion of food it may be increased, as has been confirmed by many observers, in particular Rose and Dimmitt (1916), who showed that the excess may continue for some days after ingestion. On muscular exercise an increase may be found if measured at a short interval after exercise (Schulz, 1912). No doubt other metabolic influences may have an effect on the creatinine content.

(*b*) The presence of an unknown substance or substances in the red blood-cells, as demonstrated by De Wesselow (1919), who found marked variation in the sugar concentrations depending on the number of red blood-corpuscles, and obviously not due to creatinine.

The present method has the advantage of avoiding these sources of inaccuracy. In addition, its simplicity, rapidity and cheapness are notable considerations; the avoidance of the necessity of using such expensive apparatus as a colorimeter, as in Benedict's and Folin's methods, is important.*

I am indebted to Prof. J. A. MacWilliam and to Prof. Hugh MacLean for much kindly help and criticism.

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* The necessary equipment for the present method can be obtained from Hawksley & Sons, Wigmore Street, London, for about three pounds.

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SOME APPLICATIONS OF PHYSIOLOGY TO MEDICINE.

II.—VENTRICULAR FIBRILLATION AND SUDDEN DEATH.*

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IT may be permissible to recall that in the pages of this JOURNAL[1] thirty-four years ago I brought forward a new view as to the causation of sudden death by a previously unrecognized form of failure of the heart's action in man—a view fundamentally different from those entertained up to that time. In the course of a long series of experiments on the mammalian heart, sudden deaths, occurring under varied experimental conditions, were found to be invariably associated with a very definite mechanism of failure entirely different in character from what had hitherto been believed to be present in cases of sudden dissolution in man depending on cardiac failure. Little attention was given to the new view for many years. At that time the current conception of the relations of the experimental physiology of the heart to practical medicine was widely removed from what it now is, thanks very largely to the work of Sir James Mackenzie and his associates and followers; it was not then recognized that most of the disturbances that have been experimentally induced in the mammalian heart (for example, fibrillation, flutter, heart-block, extra-systoles of various types, rhythms of abnormal origin, alternation of the heart beat, etc.) have their clinical counterparts in the manifold derangements of function in diseased conditions in man.

*Part I was published in the BRITISH MEDICAL JOURNAL of January 13th, 1923 (p. 51).

In more recent times the view that ventricular fibrillation is a cause of sudden death in man has been accepted by numerous observers. Sir James Mackenzie[2] writes of sudden death in auricular fibrillation: "It has appeared to me probable that in these cases the ventricle has passed into fibrillation as MacWilliam suggested." Again, referring to sudden death from heart failure: "The cause of sudden death is almost certainly due to the onset of an abnormal rhythm, probably ventricular fibrillation." Sir Thomas Lewis[3] states that "We have now the strongest *a priori* reasons for believing that sudden and unexpected death comes to many patients in a manner suggested by MacWilliam in 1889." The causation of death in this way has also been recognized by Hering[4] and many other observers.

Direct electrocardiographic curves indicative of ventricular fibrillation at the moment of death have been recorded, notably by an American observer, Halsey.[5] Opportunities for the gaining of such direct evidence are naturally scanty, but the indirect evidence that has accumulated is sufficient to show that this is, in all probability, not only a common cause but the usual cause of sudden and unexpected death of cardiac origin.

Relation of Death from Fibrillation to Ordinary Myocardial Failure.

The mode of death described in this article is a failure of the heart's action essentially different from cardiac (or myocardial) failure in the sense of exhaustion of the contractile power of the cardiac muscle; a verdict in the latter sense would usually be, in cases of sudden death, fallacious. While such exhaustion of contractility is, of course, of common occurrence in disease, it is a gradual process, leading to more and more marked impairment of the pumping power necessary to maintain a good circulation and to respond to the increased demands of muscular exercise, etc. There is no ground for the assumption that a *sudden* loss of power can occur—that is, that muscular fibres endowed with contractility adequate for a tolerably good blood pressure and blood flow should abruptly become enfeebled or paralysed, apart, of course, from the sudden action of violent poisons or such gross causes as asphyxia, obstruction of coronary supply, haemorrhage, etc. What really happens in the supervention of ventricular fibrillation is a misapplication of contractile energy thrown away in a turmoil of fruitless activity, a disastrous change occurring in muscle that may be already more or less limited in power, or that may, on the other hand, be possessed of vigour more than sufficient for the ordinary needs of the circulation. This often occurs in a heart showing no failure of rhythmicity, excitability, or contractility.

The inadequacy of former explanations of sudden death by failure of the heart to contract and expel its contents,

asystole, etc., need hardly be emphasized; such explanations assumed a failure of rhythm or a sudden loss of muscular power for which there is no warrant. Before the experimental investigation of the mammalian heart had shown the actual mode of abrupt and complete failure of the ventricular pump by the occurrence of fibrillation, the observed phenomena of sudden death by syncope — the sudden abolition of pulsation and all other manifestations of the beating of the heart—naturally led to the belief that the end had come through a simple failure of contractility. It was not realized how extraordinarily resistant and enduring the contractile power of the heart is, even under experimental conditions of exposure, manipulation, and severe strain of various kinds.

Sudden Death and the Pathological Changes found post Mortem.

It is well known that in many cases where death is believed to have resulted from cardiac failure the heart has been found *post mortem* to present structural characters apparently little if at all removed from the normal. An elaborate study of sudden death and the pathological conditions associated with it was made by Brouardel and Benham.[6] In this book, extending to more than 300 pages, a great deal is to be found as to numerous and varied morbid conditions and structural changes in various organs, etc., in cases where death has occurred suddenly, while it is stated that in some cases no lesion is found. Elaborate details of dead-house anatomy are presented, but no explanation is given, or indeed attempted, as to how vital function has suddenly broken down, when up to that point in many cases, in spite of pathological conditions that have often been present, the individual has been able to go about his affairs with fair or good activity of body and mind. It is obvious that such structural alterations (coronary lesions, myocardial degenerative changes, etc.) as were found after death were, up to the sudden catastrophe, quite compatible with tolerable efficiency of the functions necessary for the maintenance of a more or less active life; to determine death an abrupt change must have occurred — a process fundamentally different from such slow impairment or limitation of function as may have been present up to the final disaster.

Some Characters of Ventricular Fibrillation.

The inception of ventricular fibrillation is a sudden event, though very often preceded by more or less complex disturbances in the normal action. There is an abrupt replacement of the effective systole by a continuous turmoil of inco-ordinated activity in the muscular bundles, excessively rapid small contractions, each of short duration, coursing over the intercommunicating muscular fasciculi, so that, while some portions are contracted, others are relaxed; the

result is mere oscillation or inco-ordinated quivering of the ventricular walls, with complete loss of the expulsive power normally brought to bear on the contained blood by the strong and simultaneous state of mechanical tension present in all the muscular fibres during the normal systole. The effect on the circulation resembles that of absolute stoppage of the ventricular beat with complete cessation of its muscular activity, such as may be caused by the introduction of certain poisonous agents, etc. While fibrillation is of universal occurrence, under certain conditions, in all warm-blooded animals—both mammals and birds—its tendency to persist when once established varies greatly, being much greater in the higher mammalian types. In some animals among the lower mammals (rat, rabbit, etc.), as well as in some birds, spontaneous recovery from fibrillation frequently occurs, but in the higher mammals, and probably in man, the condition of true fibrillation seems to be invariably fatal —in the absence of the remedial measure of cardiac "massage," which may, in animals at least, be supplemented by the administration of certain drugs, while artificial respiration is maintained. Some instances of assumed spontaneous recovery in the higher mammals and in man are probably not cases where true fibrillation had been fully established, but a related though essentially different condition, which may easily be mistaken for true fibrillation.

The essential feature of fibrillation is the establishment of a mechanism of circulating excitation in the musculature, depending on a derangement of the normal relations of (1) the time taken for conduction of the excitation wave over the ventricular muscle, and (2) the refractory period of the individual fibres. If the conduction time is unduly prolonged, or the refractory period is relatively too short, re-excitation is apt to occur when the excitation wave reaches fibres that have already recovered sufficiently after the previous excitation to respond again; the excitation wave can then circulate through the complexly arranged intercommunicating fasciculi; after a time it becomes feebler and slower as exhaustion develops, until in a few minutes all visible movement becomes extinguished. If rhythmical compression (massage) of the ventricles is employed (while artificial respiration is kept up) the fibrillation may be maintained for prolonged periods (an hour or longer), with ultimate recovery under favourable conditions; and such a heart may show regular and vigorous action for the remainder of a long experiment extending over hours.

So long ago as 1887[7] I described this mechanism as a peristaltic contraction wave along the complexly arranged and intercommunicating muscular bundles, in contradistinction to the normal beat.

"The peristaltic contraction travelling along such a structure as the ventricular wall must reach adjacent muscle bundles at different points of time, and since these bundles are connected with one another by anastomosing branches, the contraction would naturally be propagated from one contracting fibre to another over which the

contraction wave had already passed. Hence, if the fibres are sufficiently excitable and ready to respond to contraction waves reaching them, there would evidently be a more or less rapid series of contractions in each muscular bundle in consequence of the successive contraction waves reaching that bundle from different directions along its fibres of anastomosis with other bundles. Hence the movement would tend to go on until the excitability of the muscular tissue had been lowered so that it failed to respond with a rapid series of contractions. Then there might be some isolated peristaltic contractions, such as I have often seen after the cessation of the fibrillar movement."

These conclusions were confirmed and extended in a paper in 1918,[8] while in the interval similar views had been advanced and supported by experimental evidence — by Mines[9] (1913) in the case of the frog heart and by Garrey[10] (1914) in the mammalian heart. Suggestive experiments had been made by Mayer[11] (1908) on medusa, etc.

Relations of Refractory Period and Conduction Time.

If the fundamental importance of the relation between the duration of the refractory period and the conduction time of the muscle is kept in view it is easy to understand how the mechanism of circulating excitation may come into operation under very diverse conditions affecting the ventricles. Any influence cutting down the refractory period or lengthening the conduction time disproportionately must naturally tend to favour the process of re-excitation; a combination of such changes is, of course, still more effective. Hence the development of fibrillation is witnessed at one time as an apparently " spontaneous " event in a vigorous heart manifesting signs of extreme irritability (for example, from chloroform, digitalis, etc.) or in a normal heart subjected to stimulation —excessive rapidity of excitation playing an essential part by shortening the refractory period and slowing the conduction time. At another time fibrillation appears in a heart that presents features of grave depression, diminished contraction force, loss of tone, lessened excitability, and— what is the determining factor in this case—pronounced slowing of the propagation of the excitatory wave, relatively to the duration of the refractory period; such may be seen in poisoning by potassium salts, extreme cooling, etc.; thus it is often a terminal, or approximately terminal, phase in the dying heart. In some depressed hearts there is a decided liability to fibrillation from mechanical disturbance, external pressure on the ventricles, incising the pericardium so as to remove its support from a relaxed ventricular wall, etc. In either case, whether fibrillation is a manifestation of perverted irritability or of abnormal depression, the same explanation of disturbed relationship between the processes named holds good.

In accordance with this conception it is readily intelligible that the *absolute* values of refractory period and conduction time may undergo extensive variation without fibrillation being set up; if both of these factors vary proportionately

the conditions of circulating excitation do not arise. Thus artificial cooling of the heart, stopping short of a certain extreme point (about 23° C. in the perfused cat's heart), does not induce fibrillation, there being a concurrent lengthening of refractory period and conduction time as the cooling goes on—within the limit stated.

Conversely there may be, as results of a rise of temperature, etc., a marked shortening of the refractory period without fibrillation occurring, the rate of conduction of the excitatory impulse being also accelerated. The essential dependence of the mechanism of fibrillation on the factors named makes it clear why there should be no constant or necessary relation between the incidence of fibrillation, and even great alterations in contractile force, tone, etc. Dilatation of the ventricles is credited by Levy with a protective influence; this view is negatived by various observed facts—among others the proneness to fibrillation seen in ventricles weakened, relaxed, and distended as a result of potassium poisoning; and conversely, the marked resistance or insusceptibility to fibrillation in ventricles that are of small volume, acting strongly and rapidly and well emptied at each beat, in sequence to a large dose of adrenaline, etc. So long as the essential relationship, described above, is not upset a heart may be profoundly influenced in many ways without fibrillating: it may beat very rapidly or very slowly, regularly or very irregularly, powerfully or feebly; it may be well emptied or imperfectly emptied at each beat—with consequent distension in the latter case; it may be very sensitive or very dull to direct stimulation, and its muscle may be lax or firm, etc.

Pseudo-Fibrillation.

Apart from the disastrous event of true fibrillation there is also to be observed under certain experimental conditions (for example, rapid artificial excitation by a series of electrical shocks, mechanical stimulation, etc.) a temporary condition in the ventricles presenting many points of resemblance to true fibrillation—a degree of inco-ordination or asynchronous contraction of the musculature, recognizable on inspection and on palpation of the ventricular substance, attended by great reduction in the range of contraction movement and very little expulsion of blood at each beat, a great fall in arterial pressure, and a failure of recognizable pulsation in the peripheral arteries, etc. It is impossible to distinguish this condition from true fibrillation by examination of the arterial pulse.

Usually this condition is soon recovered from when the artificial stimulation is discontinued, though it may persist for variable periods. It apparently differs essentially from true fibrillation in its dependence on a rapid series of excitatory impulses emanating from a stimulated area or "spontaneously" from one or more irritable foci, these impulses forcing a succession of ventricular beats at a rate

incompatible with their normal characters; cessation of these excitatory impulses is speedily followed by a reversion to the more moderate rates that are compatible with the normal type of beat. The non-persistence of the abnormal condition is due to the fact that the mechanism of circulating excitation, characteristic of true fibrillation, has not been fully established. To this condition I have applied the term " pseudo-fibrillation." It is probable that most, if not all, the alleged instances of transient fibrillation in man where recovery occurred—apart from the application of cardiac massage—were examples of pseudo-fibrillation as recognized experimentally; this would account for the features observed in man during the brief attack, and for the subsequent recovery.

Such is probably the explanation of such a case as that described by Robinson and Bredeck[12] in which there were repeated syncopal attacks simulating those of the Adams-Stokes syndrome; in one of these attacks an electrocardiographic record was obtained and showed characters apparently resembling in some measure those of ventricular fibrillation—followed by recovery. In another case F. M. Smith[13] obtained an electrical record suggestive of fibrillation, the attack being recovered from within a minute. Hoffmann[14] has also published an electrical curve of a temporary condition evidently approximating to, but not reaching, the fully developed condition of fibrillation. There is also a case reported by Kerr and Bender[15] of temporary fibrillation or a closely allied condition (under quinidine therapy) where records were obtained.

The Question of Recovery from True Fibrillation.

As has been already stated, spontaneous recovery from ventricular fibrillation is very frequently seen in the hearts of the lower mammals—for example, rat—but recovery is increasingly difficult and rare in the higher forms. In the cat and rabbit, after true fibrillation has lasted for periods of considerable duration (minutes), the ventricles may, without treatment, sometimes show cessation of the fibrillation movement and " spontaneous " recovery of the power to execute weak co-ordinated beats, with more or less definite pumping out of blood from their chambers into the arteries. This recovery occurs too late as a rule, if not invariably, to provide for continuance of the life of the animal, for the collapse of the circulation has lasted too long for restoration of the vital functions. Most instances of true fibrillation do not show this sort of ventricular " recovery "; the fibrillation movement usually becomes slower and feebler and gradually flickers out in the dying muscular fibres.

In the human subject it is probable that true fibrillation is (apart from treatment by cardiac massage, etc.) usually if not invariably fatal, though it seems possible that such recovery of separate though feeble ventricular beats too late

for the survival of the individual may occur—if one may put this interpretation on the phenomena of some cases that have been recorded by Sir William Osler,[16] cases presenting in striking fashion the external features associated with the occurrence of fibrillation. Referring to sudden death in some anginal subjects, he writes:

"Possibly in some act combining intense emotion with muscular effort there is a rapid change, a sudden unconsciousness, a stony stare, a slight change in the facial expression, and then with two or three gasps all is over; no pulse is felt at the wrist; the respiration stops; but even when the patient is apparently dead a feeble heart impulse may be felt or faint heart sounds heard."

In one of these cases where slowly recurring respiratory gasps occurred over some minutes, cardio-puncture with a long thin aspirator needle showed movements indicating heart beats at gradually diminishing rates—52, 44, 32 per minute—ceasing fifty minutes after collapse and forty-five minutes after the last inspiratory gasp. (It may be remarked that the phenomena here described by Osler were not such as to be suggestive of reflex vagus inhibition.)

Osler's description of such deaths may be compared with what Sir Thomas Lewis[17] says of deaths from ventricular fibrillation, occurring in subjects of auricular fibrillation:

"From time to time a sudden and unexpected catastrophe happens; regarded as convalescent, the patient is sitting in bed, chatting or feeding maybe. A nurse in charge, or perhaps a neighbouring patient, hears a cry or choking sound; the patient falls back on the pillows intensely pale, there are a few gasping respirations, a little convulsive movement, and the pulseless patient, rapidly becoming livid, is still."

Relation of the A-V Junctional Tissues to Ventricular Fibrillation.

Unlike auricular fibrillation, which, as is well known, may go on over a long term of years, ventricular fibrillation is promptly fatal, involving as it does an abrupt and complete abolition of the pumping action of the ventricles and a speedy cessation of the circulation. The effect of auricular fibrillation is to impose a more or less extensive limitation to the efficiency of the heart in two ways—by the absence of the normal action of auricular systole in completing the filling of the ventricular force-pump, and still more by the rapid and disorderly lead given to the ventricles by the rapid and irregular excitations transmitted from the fibrillating auricles to the ventricles; the latter involves a wasteful and exhausting expenditure of the energy of the ventricular muscle, attended by relatively poor results in the way of maintaining the circulation.

Fortunately the structure and properties of the junctional tissues between auricles and ventricles are such as to afford a most important protection against transmission of the fibrillation from the auricles to the ventricles. In default of such protection the onset of auricular fibrillation would necessarily be fatal by extension to the ventricles. It is

known from experiment that in the auricular and ventricular muscle, with their complexly arranged bundles of fibres, fibrillation can be propagated across an artificial isthmus, made by incision, connecting portions of the musculature, only as long as the isthmus is of sufficient breadth; in the junctional mechanism, comprising the A-V node with its special properties, and the narrow A-V bundle with its longitudinally arranged fibres terminating in the Purkinje network on the inner surface of the ventricles, the fibrillation process fails to be transmitted (as such) either from auricles to ventricles or from ventricles to auricles. The junctional mechanism, while able to conduct impulses sufficiently fast to give ventricular beats at rates much above those of the normal heart under resting conditions or in moderate exercise, is not able to transmit the vastly more rapid and characteristic series of excitations of fibrillation. It is only under certain very special conditions, where the susceptibility of the ventricles to fibrillation (at relatively slow rates of excitation) is extraordinarily great, that the writer has in rare instances seen any experimental evidence of the possibility of ventricular fibrillation being excited from the auricles by impulses passing through the A-V bundle. The possibility of such occurring under pathological conditions in man is worthy of consideration.

The Purkinje network formed in the interior of the ventricular walls by the arborizations of the fibres of the A-V bundle form a sort of distributing board over which the normal impulses descending the bundle spread swiftly so as to be delivered almost simultaneously to the myocardium at the various parts of the ventricular cavities, bringing about the normal co-ordinated contraction of the ventricular fibres at each beat. Such mode of excitation is obviously unfavourable to the development of the fibrillation mechanism, but the latter might under certain conditions be promoted by an abnormal delivery of irregular and aberrant exciting impulses, dependent on defects in the branches of the bundle or its terminal arborizations, such partially interrupted or distorted impulses impinging upon the myocardium at different points in abnormal fashion—alterations in timing or direction of the impulses as might readily be determined by morbid conditions in some parts of the conducting network. An exaggeration of these functional aberrations may easily be favoured by circumstances making an extra call on the heart, such as emotion or muscular effort, or by the succeeding phase of reaction with its increase of vagus control and other changes.

There is some clinical and pathological evidence that points to the operation of such causes, resulting in ventricular fibrillation and sudden death in the human subject, though the particular mode of causation indicated above does not seem to have been suggested. Thus Nuzum[18] described a case of sudden death in a man, aged 38, who had shown no definite signs of ill health and where there

were no adequate microscopic *post-mortem* findings to account for the fatal issue, apart from marked alterations present in the branches of the A-V bundle in the shape of fatty infiltration or replacement, the Purkinje fibres being beset with fatty droplets, etc.

Sapegno,[19] from a study of seventy-two cases, inferred that rapid unexpected death may be due to acute or chronic lesion of the bundle fibres, the acute changes being predominant in these fibres when both they and the ventricular myocardium were affected. He cites an instance of sudden death in a girl twelve days after recovery from typhoid fever, not associated with the acute changes sometimes seen in the myocardium after typhoid, but with lipomatosis in the bundle fibres—the cell substance being largely replaced by fat—from the point of division of the main stem to the point where the two main divisions decrease rapidly in size. Monckeberg[20] described two cases of sudden death associated with lesions of the Purkinje fibres. One was a diphtheria case, where no gross changes were found in the myocardium, but fatty changes were shown by staining with Scharlach R in the main stem, branches, and subdivisions; the myocardium was fat-free. In the other case (trephining of skull, etc.) there were more pronounced fatty changes in the bundle fibres than in the myocardium, though some of the Purkinje fibres seemed to have remained normal.

It need hardly be recalled that several observers have described certain abnormal features in electrical records from the heart as being indicative of defective conduction in the main branches, bundle block (Lewis and others), or in the intraventricular Purkinje network (arborization block)—bizarre ventricular complexes with notching or splintering of the deflections, prolongation of the duration of the Q R S group oscillations, inversion of the T wave, etc., in addition to a remarkably low altitude of the curves in arborization block. (Oppenheimer and Rothschild,[21] Carter,[22] Willius,[23] and others.)

Exciting Causes of Ventricular Fibrillation.

In the many observed cases where all the facts point to ventricular fibrillation as the immediate cause of sudden death the common association of muscular exertion or emotional excitement is notable. The ever-recurring reports of sudden deaths during or shortly after exertion, in persons who up to the fatal issue had been able to pursue their usual avocations, emphasize the importance of the conditions attendant on muscular effort—those involving an increased demand on the powers of the heart and more or less stress on the organ. This is brought about in various ways: by the augmentation of rate and force and irritability through the agency of the cardiac nerves (diminution or suspension of vagus control and excitation of the cardiac sympathetic augmentor fibres), increased arterial pressure presenting greater resistance to the pumping out of the ventricular contents,

increased diastolic filling due to more rapid inflow from the venae cavae, etc. Similar changes attend emotional excitement, with the exception of the greatly increased venous return to the heart depending on the pumping action of the working muscles during exertion driving on the blood in the veins.

While the normal heart is not injured by such changes, and in virtue of its great reserve power easily responds to increased demands on it, a heart that is temporarily or permanently in an abnormal condition of excessive susceptibility is apt to be thrown into fibrillation.

Susceptibility to Ventricular Fibrillation.

Both in healthy and diseased animals notable differences in the ease with which fibrillation may be induced were seen under experimental conditions that were apparently similar. And a heart may sometimes be seen, under altered conditions, to pass from the susceptible condition to a stable one, which may be exceedingly resistant to the induction of fibrillation by various forms of stimulation that are usually very effective. Or a change in the opposite sense may take place; or there may be variation from one phase to another more than once in the course of a single experiment in the case of a healthy heart subjected to certain abnormal influences. On the other hand, there are many cases where the abnormal susceptibility is a persistent one associated with altered nutritive conditions, toxic agencies, etc., in the muscle.

In healthy animals, cats particularly, a great susceptibility to fibrillation may be established by the administration of chloroform; the relation of this condition to the phase of light chloroform anaesthesia has been specially worked out by Levy, most (though not the whole) of whose results are in agreement with those obtained by the present writer over a long series of years. It is after a deeper phase of chloroform anaesthesia that the lighter phase is apt to be attended by the marked susceptibility referred to.

In the latter condition fibrillation is often readily induced by stimulation of afferent nerves in various ways—resulting in reflex contraction of skeletal muscles, disturbance of respiration, rise of blood pressure, increased rate and force of the heart with increased return of blood through the great veins, etc.—in short, the same group of changes that occur in muscular effort, and brought about in similar fashion through the instrumentality of the vagus and cardiac augmentor nerves, excitation of the respiratory and vasomotor centres, and the mechanical action of the skeletal muscles in propelling blood more rapidly back to the heart by the veins—increasing the rate of its diastolic filling and its output and work per minute very largely.

It is plain that it is through these changes that the sudden fibrillation of the ventricles is determined in a susceptible heart under chloroform, and there is a strong *a priori* case

for the presence of a similar mechanism in the occurrence of fibrillation and sudden death (apart from chloroform) during or shortly after muscular effort—granted a condition of abnormal susceptibility in the ventricular muscle.

In addition to chloroform there are various other toxic agencies capable of establishing the hypersensitive state. Referring to drugs, it is well known from experimental evidence (adduced by Cushny and others) that bodies of the digitalis series have a powerful influence in this direction—as also have barium salts, etc.—and when pushed to extremity kill by causing fibrillation. A whole series of chemical substances might be cited as having well defined effects in this direction. And some abnormal metabolic products may well exercise a similar influence in this respect on the cardiac muscle. The development of the hypersensitive condition is not necessarily attended by any recognizable structural alteration. A prominent part in the setting up of abnormal susceptibility to fibrillation must be assigned to defective coronary blood supply.

Some Effects of Experimental Coronary Obstruction.

It is unnecessary to recall in detail the long series of experimental investigations at the hands of many workers which have demonstrated the frequent occurrence of fibrillation as a result of ligation of a coronary artery or one of its larger branches. The evidence available leaves little, if any, room for doubt that death from sudden coronary obstruction in man is due to fibrillation; the clinical features of many recorded cases are very significant, taken in conjunction with the very definite facts established by experiment in animals. The reason of the difference between non-fatal and suddenly fatal coronary obstruction is usually to be found in the non-occurrence or occurrence of ventricular fibrillation in the different cases.

It has been found experimentally that coronary occlusion, if sufficient to prove fatal, may do so in more than one way: (1) it may kill rapidly (minutes) from acute ischaemia causing ventricular fibrillation, or (2) failing this, it leads to damaged nutrition with degenerative changes (anaemic necrosis, fibrosis, etc.); these changes, apart from leading in rare instances to rupture of the heart, naturally diminish the contractile efficiency in degrees varying according to the severity of the anaemia and its distribution. Recovery may occur and life be prolonged indefinitely; or death may be suddenly caused by the supervention of fibrillation. It is easy to understand how altered functional relations in the tissues of the damaged area may lend themselves under certain conditions to the decisive upset of the normal relations of refractory period and conduction time in the ventricular walls. It remains to be seen whether the tendency to fibrillation after coronary ligation is dependent mainly on the conditions induced in the Purkinje system or in the ordinary myocardium or in both of these.

The significance of some of these results of experimental physiology does not seem to have been fully realized in relation to their bearing on the human subject. In this connexion the convincing researches of W. T. Porter[24] (1896), Baumgarten[25] (1899), Miller and Matthews[26] (1909), and F. M. Smith[27] (1918) are specially relevant. Numerous observations show that in presence of the defective blood supply following ligation the abnormal conditions that develop in the anaemic areas can not only predispose and lead up—often after months—to fibrillation, either during muscular exertion or during rest, but that the abrupt onset of fatal fibrillation may come without preceding signs of cardiac failure, as tested by exercise, or without immediately premonitory evidences of cardiac disturbance in the shape of extra-systolic irregularities, tachycardia, etc. Thus fibrillation can occur without apparent exciting cause and quite apart from the sequence commonly observed—extra-systoles, tachycardia, and fibrillation—when death suddenly comes as an early event after coronary ligation. In other cases, at a much later time, disturbances of rhythm and evidences of heart failure present themselves, increasing in intensity and eventuating in fibrillation. Or temporary irregularities may have developed at varying periods after ligation—to give place later to regular rhythm, and then after weeks or months to fibrillation and sudden death.

Sudden and Unexpected Death during Rest.

The difficulty of explanation of such deaths has long been felt, in the absence of recognized conditions (effort and excitement) tending to make an extra call on the heart and of such powerful afferent excitation as might be assumed to be provocative of reflex inhibition of intensity and duration sufficient to be fatal. Recourse has constantly been had to the verdict of " failure of the heart's action," though the sudden collapse in cardiac efficiency remains unaccounted for, *post-mortem* examination often affording no explanation of the abrupt ending of life. Sir Clifford Allbutt,[27] while suggesting vagus inhibition as a mode of death during anginal attacks, writes with regard to the class of deaths now under consideration—during conditions of rest, apart from anginal attacks, and where no exciting cause is apparent:

" But the riddle, which I have done so little to read, is the frequent suddenness of death in one who, having scarcely known illness, expires under no extraordinary effort; or in the peace of his own bed or chair passes silently away. The reading of this riddle is not yet."

In this relationship the facts that have been stated with regard to the more or less remote effects of experimental interference with the coronary blood supply by ligation of a branch are obviously of profound significance, showing as they do that ordinary life can go on for prolonged periods (months), either with or without signs of cardiac disturbance, until a sudden ending comes by fibrillation, sometimes

during muscular effort, but often apart from this, in the absence of recognizable exciting cause or in presence of causes too trivial to have any effect under ordinary conditions. And, apart from sudden obstruction, there is no reason to doubt that a gradual interference with the blood supply as a result of coronary disease can, by damaging the nutrition and altering the properties of the muscle, lead to an abnormal susceptibility to fibrillation.

The applicability to man of these results is naturally easy in view of the widespread tendency to serious impoverishment of the blood supply (local or general) of the cardiac muscle in the later half of life, the period when deaths of the class under consideration usually occur. In the light of these facts we have a rational basis for many unexplained disasters—fatal events that otherwise remain shrouded in mystery.

While the effects of limitation of the blood supply are proved by abundant and convincing evidence, other agencies, such as perverted nutrition, toxic influences, generative changes, etc., can be effective causes. Interference with normal functioning of the Purkinje fibres on the inside of the ventricles comes into question as well as alterations in the ordinary myocardium. Fibrillation may be determined at a certain point of time by a sudden aggravation or cumulation of the toxic condition, etc., aided, it may be, by the incidence of some disturbance of the vascular system too slight to produce any serious effects except in the specially predisposed condition.

It is to be borne in mind that the conditions disposing to and leading up to fibrillation need not pervade the whole of the ventricular musculature, but may be limited to a certain amount of that tissue, as after obstruction of a coronary branch and in other conditions; changes in restricted areas can set up fibrillation, which involves the rest of the muscle, healthy as the great bulk of it may be. It is readily intelligible that such limited changes may naturally be associated with little or no recognizable alteration in the force of the ventricular beat or its general efficiency. Thus there may be little or no warning of the impending catastrophe, even at a time when the mine has been laid and only a spark is needed to precipitate the explosion.

Abnormal Cardio-vascular Variations of Obscure Origin.

Under certain conditions of cardio-vascular instability—usually occurring in association with morbid states of the arterial system—irregular tides of circulatory change, often obscure as regards their exciting causes, are sometimes recognizable; one manifestation of these is found in the extensive variations of arterial pressure as measured under similar conditions from day to day or at shorter intervals. These variations are sometimes, but not necessarily, associated with discrepancies in the sphygmomanometer

readings from different limbs, depending on local causes—the presence or absence of strong contraction in the large arteries of the respective limbs. Actual rises of general arterial pressure involve cardiac changes in addition to vascular constriction, etc. It is evident that disturbances of this kind may be influential with regard to the onset of anginal attacks or of sudden death in subjects where a special predisposition exists.

The Question of Coronary Spasm.

With regard to sudden interference with coronary blood supply, apart from the rare accident of embolism and the less rare occurrence of thrombosis, there arises the question of spasmodic contraction—an old hypothesis as applied to the explanation of anginal attacks. The obscure cardio-vascular disturbances already referred to might be invoked to account for the onset of some anginal attacks during rest; the attack might be determined by antecedent unrecognized changes in blood pressure and heart action. But there are on record cases in which such attacks during a period of rest are found to be unattended by elevation of the blood pressure or recognizable changes in the heart's action. Again, there are instances among anginal subjects who have varying periods of relatively low and high pressures, where no greater tendency to angina has been found in the phases of high pressure; a notable example of this has been recorded by Sir James Mackenzie. In such cases the idea of coronary spasm has commended itself to many observers, supported by such analogies as the extreme arterial constriction of Raynaud's disease, the thickening and narrowing of the temporal artery on the same side as the pain in migraine and the occasional association of Raynaud's disease and migraine, the occurrence (sometimes in relatively youthful subjects) of transitory aphasias, hemiplegias, etc., attributed to an acute temporary anaemia or ischaemia from extreme constriction of a cerebral artery, and sometimes associated with migraine. Again, there is the so-called "abdominal angina," which has been correlated with spasmodic contraction of sclerosed mesenteric arteries. Sir William Osler,[28] referring to arteries in general, states that in a certain stage of sclerosis arteries are very prone to spasm—a view repeatedly urged by Professor W. Russell[29] and supported by Pal[30] and many others.

It is evident that a temporarily excessive contraction of some part of the coronary system (especially in cases where the blood supply is already reduced or minimal) would induce an ischaemic condition which might be responsible for the onset of fibrillation or of an anginal attack; in this way an apparently unprovoked paroxysm of pain during rest might be accounted for and also its equally unexplained passing off after variable periods. It might also be surmised that amyl nitrite may relax a constricted coronary as part of its general vascular effect, with relief of pain

which may last after the general blood pressure has again risen to its former level. The spasm hypothesis has been subjected to searching criticism by Sir Clifford Allbutt, who among many other considerations states that amyl nitrite gives no relief in transitory hemiplegias and aphasias. There is also the occurrence of dyspnoea in coronary obstruction from sudden thrombosis or embolism (as established *post mortem*) and its absence as a necessary feature of typical angina; but a possible explanation of this difference can be suggested.

Evidence of the occasional presence of strong contraction of large arteries (brachial, etc.) in diseased conditions in man was obtained by the present writer, in conjunction with Professor G. Spencer Melvin[31] and Dr. J. E. Kesson,[32] in an investigation of blood pressure a number of years ago, and surviving sclerosed arteries from the legs of old horses were found to show extraordinarily intense contraction, causing complete obliteration of their lumen and an enormous resistance to attempts to force blood through them.

In relation to the question of arterial spasm, doubt as to the existence, at least in effective degree, of vasomotor innervation of the coronary arteries is not a consideration of decisive moment. For there is no proof that the forms of excessive contraction now under discussion—for example, in the brachial artery or the horse's leg, etc.—are vasomotor phenomena; it is more probable that they are directly dependent on morbid conditions present in the arterial muscle at the time.

It may be argued that the foregoing considerations point to the feasibility of the hypothesis of coronary spasm, in view of there being no sufficient reason to assume that the coronary vessels—specially prone as they are to sclerotic changes—should in diseased states be immune from such functional disturbances as seem to occur in other arteries. But the question is far from being closed.

Fatal and Non-Fatal Angina.

It is necessary to discriminate clearly between the separate questions of (1) the mechanism of pain production in anginal attacks, and (2) the mechanism of death occurring during or between attacks, sometimes without warning.

The striking tendency of the graver forms of angina to terminate in sudden death need not be emphasized. There is every reason to believe that, in many cases at least, the end comes by ventricular fibrillation, and that the different issue in fatal and non-fatal cases hangs on the supervention of fibrillation in the former and its absence in the latter. The development of fibrillation as a frequent and characteristic result of defective coronary blood supply, as demonstrated experimentally, has already been described; and

we know, from abundant pathological evidence in man, the association of coronary and myocardial impairment with fatal angina. Further, the clinical features of many recorded anginal cases, where sudden death took place either in an attack of pain or apart from such, are very noteworthy in their similarity to those attendant on death by ventricular fibrillation. The significance of these facts taken together need not be enlarged upon.

If we accept the view, which has commended itself to many observers, that an important factor in the production of pain in angina is to be found in the heart muscle working with a defective blood supply, it becomes plain how increased demands on the organ by muscular effort or emotional stress may excite an attack by leading to a relative anaemia, the blood supply, which was sufficient during rest to ensure the absence of pain, now becoming inadequate for the muscle. Such a conception might be brought into relation with the results obtained in an ischaemic limb where, on working a muscle, acute pain is caused long before the fatigue point (as indicated by inability to raise the weight in ergograph experiments) is reached—a mechanism of pain production apparently different from that present in a muscle working to fatigue while its normal circulation is going on (MacWilliam and Webster[33]).

It may be conceived that in the close and striking association of angina and sudden death we see the working of distinct but related mechanisms, based in part at least on a common underlying process, essentially similar in character but differing in intensity and in the fatal or non-fatal issues, these issues being no doubt also influenced by other conditions which affect the results of the fundamental process. A conception of this kind would include two categories of dangerous anginal conditions—one with more or less transient attacks of pain of varying grades of severity, the other with the process, common to the two categories, going further in some directions, attaining greater intensity, and culminating in ventricular fibrillation. But the pros and cons of the vexed question of pain production in angina are beyond the scope of this paper.

In connexion with the well known fact that pronounced coronary sclerosis very frequently exists without angina, it has to be borne in mind that an essential point is, not the structural change in the arterial wall, but the amount of actual defect of blood supply through the sclerosed vessels from greater or less narrowing of their channels, the presence or absence of contraction in their muscular coats, the state of the capillary field, etc. Much depends no doubt on the more or less gradual development of obstructive change, the establishment of more or less efficient collateral circulation, etc. It is known that life may go on after the gradual development of complete occlusion of one coronary while the other may be found to be very greatly reduced in calibre; also after complete blocking of a large branch.

Collateral circulation obviously plays an important part; it is now well known that the coronary branches are not end arteries, as was at one time believed, but have numerous anastomotic connexions. Further, as described by Gross,[34] the blood vessels (arteriae telae adiposae) of the subepicardial fat, which increases in amount as life advances, can in some measure exercise a compensating influence, supplying a considerable amount of blood to the subjacent muscle. Belonging to this system are delicate parallel vessels accompanying (at some distance) the main coronary branches, as well as a feltwork of vessels in the fat of the auriculoventricular groove. Gross emphasizes the importance of a relative anaemia of the muscular walls of the right heart in old age, as bearing on failure in pneumonia, etc.; he suggests a variation of the adage that a man is " as old as his arteries " to " as old as his right coronary artery."

It is noteworthy that when the main trunks and large branches of the coronaries are the seat of pronounced sclerotic changes the intramuscular twigs and finer ramifications may remain practically unaffected. The extent and efficiency of the capillary system are obviously of prime importance.

Syncope from Ventricular Standstill due to Heart-block.

In cases of heart-block, Adams-Stokes syndrome, etc., where death occurs suddenly, it is uncertain whether simple stoppage of the ventricular beat in the state of diastolic relaxation always lasts long enough to kill by paralysis of the nerve centres, following the phases of unconsciousness and convulsive phenomena. The time needed in man for irretrievable damage of these centres by acute anaemia is not known; in the ordinary experimental animals it is relatively long—a number of minutes. Of course, there may sometimes be morbid conditions present in man which would shorten the time that circulatory arrest can be survived. But in view of the associated structural damage present in the Adams-Stokes syndrome, the possibility of the ventricular standstill terminating in fibrillation in some instances must not be overlooked, though there seems to be at present no actual evidence of this happening in man. On the other hand, it is true that a fall of blood pressure, such as accompanies ventricular standstill, exercises a restraining influence on the development of fibrillation under certain conditions; but this does not always hold good under other conditions—for example, fibrillation sometimes develops in the gravely depressed or dying heart, notwithstanding the fact of excessively low blood pressure. In any case it must be concluded that only a fraction of cases of sudden death can possibly be attributed to ventricular standstill depending on the relatively rare condition of heart-block.

Syncope during Tachycardia.

There is strong reason to believe that the fibrillation mechanism is operative in many cases of sudden death

associated with ventricular tachycardia. The myocardial conditions underlying tachycardia are closely related to those on which fibrillation is dependent, and there is abundant evidence that the former may develop into the latter. The excessive rate of beat—whatever be the origin of the tachycardia—is in itself favourable to this development, since it involves shortening of the refractory period and lengthening of the conduction time. The rapidity of succession of contractions—whether arising in the ventricles themselves or transmitted with abnormal frequency from the auricles as in auricular flutter—that the ventricles can stand without fibrillating varies much in different conditions; when the conductivity is already depressed and the conduction time long, a much lower grade of acceleration naturally suffices to establish fibrillation, as can be demonstrated experimentally.

From the work of many observers we know that in certain hearts (for instance, after coronary ligation, etc.) there is often a characteristic sequence of events illustrative of the close relations of tachycardia and fibrillation—extra-systoles, first singly, then in irregular runs, more or less continuous tachycardia, and finally fibrillation. Apart from the supervention of fibrillation it is known that the fall of blood pressure attendant on tachycardia is compatible with life for very considerable periods; there have been recoveries after periods of excessively low blood pressure attended by unconsciousness, etc., for hours. It remains to be seen whether the fall of blood pressure is often or ever sufficient *per se* to kill, or whether the fatal issue is always determined by the occurrence of fibrillation. There are no grounds for accepting vagus inhibition as a mode of sudden death during tachycardia. The vagus is known to lose effectiveness in this condition. Auricular flutter, etc., may induce unconsciousness lasting for hours without causing death; a very small blood supply can suffice to keep the nerve centres alive, as Leonard Hill showed many years ago. The absence of fibrillation is an essential feature in the recovery from ordinary cases of fainting due to temporary vascular relaxation due to vasomotor failure or to vagal inhibition, etc.

Status Lymphaticus, Electrical Shock, Digitalis.

A possible development of the mechanism of fibrillation is worthy of consideration in connexion with the sudden and unexplained deaths of the status lymphaticus, occurring, as they often do, in the absence of any recognized causation. The features of some recorded examples would fit in with the known phenomena of fibrillation—for example, such cases as have shown an abrupt abolition of the signs of heart action while the respiratory movements persisted for some little time, in marked contrast to the order of events in death by asphyxia.

Fibrillation is one of the modes of death in electrical

shock, and according to Jex-Blake[35] it is operative in death from lightning. There is convincing evidence—experimental and clinical—that the same mechanism is responsible for sudden death during overdosing with bodies of the digitalis series.

Sudden Death in Aortic Regurgitation.

The frequency of absolutely sudden death in this condition has long been recognized. In view of the usual coronary and myocardial involvements, the causation of the fatal issue—sometimes occurring without antecedent signs of cardiac failure—may naturally be ascribed to ventricular fibrillation. There seem to be no good grounds for the assumption of protracted vagus inhibition as an effective cause.

Reflex Cardiac Inhibition.

Reflex vagus inhibition has in the past been freely invoked to account for sudden death in many diseased conditions and even in healthy persons—for example, from a violent blow on the epigastrium, etc. On the experimental side extended investigations on a great number of healthy animals and a considerable number of diseased ones have failed to lend support to the hypothesis; it has usually been found impossible to stop the heart long enough to kill by reflex inhibition or even by strong direct stimulation of the vagus, escape of the heart or the ventricles usually occurring much too soon for death to be caused by circulatory arrest. The conclusion has been reached by different observers that the possibilities of a fatal issue in this way have, to say the least, been greatly exaggerated, and that there is no sufficient ground for assuming reflex inhibition *per se* to be a frequent or important mode of death.* In some instances where the vagal hypothesis had met with a large measure of acceptance—for example, in cases of sudden death during an early phase of ordinary chloroform anaesthesia—the view has not proved to be tenable, since such deaths have been shown to be essentially due to an altogether different mechanism—ventricular fibrillation.

On the other hand, the possibility of increased susceptibility to vagus inhibition under certain abnormal conditions must be borne in mind. In Embley's[36] work on chloroform it was found that under special conditions, in dogs after a large dose of morphine, the inhalation of strong chloroform vapour may cause great slowing of the heart, fall of blood pressure, and stoppage of respiration—consequences evidently depending on excessive vagus action, and obviated or removed by exclusion of such action by section of the vagi or by atropine. But the conditions present in

* Sudden death during operative procedures in the thoracic cavity (thoracocentesis, etc.) seems, in the light of the work of Capps and D. D. Lewis, to depend on fall of blood pressure due to vasomotor changes rather than to reflex cardiac inhibition (*Arch. of Int. Medicine*, 1907, cxxxiv, 868).

these experiments differ widely from those of simple chloroform anaesthesia as ordinarily conducted in man. It is well known that in dogs morphine tends to exaggerate the controlling influence exercised by the vagus centre over the heart.

In Laslett's[37] well known case it was clear that vagus inhibition induced repeated syncopic attacks, causing cardiac standstill of the whole heart, sometimes lasting for periods of six to eight seconds, but not long enough to cause death; atropine was found to be effective as a counteracting agent.

In this connexion certain observations by Sir Hugh Anderson, cited by Sir Clifford Allbutt,[38] are very noteworthy. These were on cats in which the cardiac augmentor nerves were cut, by the stellate ganglia being excised some time previously. It was found that swinging the animal in the air caused pronounced slowing of the heart—for example, from 120 down to 40 in the case of *old* cats.* (It may be remarked that such degrees of slowing were not at all dangerous, and probably did not even cause much lowering of the systolic blood pressure.) But a remarkable tendency to sudden death was observed in these animals. No evidence is stated to show whether such deaths were actually due to extraordinarily prolonged cardiac standstill, or to the supervention of some other change—for example, ventricular fibrillation, to which cats are known to be specially prone under various conditions. So far as the available evidence goes, there is nothing to indicate that the observed slowing was more threatening to life than similar slowing as seen often in common cases of non-fatal syncope in man.

With reference to possible applications of indications afforded by such experiments to the human subject it has to be remarked that we know of no clinical condition in man where there is reason to believe that conditions at all resembling those stated above are ever present—consequences of an interruption of the various paths that traverse the stellate ganglia, loss of the nerve cells contained in them, etc.† Experimental investigation shows that the cardiac augmentor nerves are very persistent in their action, extremely resistant against drugs and various abnormal conditions, and demonstrably capable of strikingly effective action in many gravely depressed states of the cardiac muscle—in contrast to the vagus functions, which are well

* Sir Clifford Allbutt predicated an increased potency of the vagus in old and damaged hearts. Gilbert (*Arch. of Int. Med.*, 1923, xxxi, 423) has recently found in old people a more ready response of the vagus to digital compression—an age effect, apart from pathological cause. The mechanism of digital pressure is undecided—whether it acts directly by stimulation of efferent (inhibitory) fibres or reflexly by excitation of afferent fibres. There seems to be no proof of actual danger to life in this way.

† Jonnesco's operation for angina pectoris is an example, resection of the lower cervical and the first thoracic sympathetic ganglion being done with the object of interrupting afferent paths from the heart. Jonnesco recommends the bilateral operation, regarding it as harmless; he does not seem to have recognized any such dangers as were noted in Anderson's experiments. (Jonnesco, *Bull. de l'Acad. de Méd.*, Paris, 1920, lxxx, 93; *Presse Méd.*, 1921, xxix, 193; Ibid., 1922, xxx, 353.)

known to be readily diminished or cut out altogether by various chemical agencies, etc.

In many cases of common syncope vagus slowing of the heart down to 50, 40, etc., a minute is a feature, but such slowing is wholly insufficient to account for the fall of blood pressure and the loss of consciousness; there are other factors concerned. Such cases do not have a fatal issue. Some instances of this condition were described by Lewis[38] a few years ago in subjects of "iritable heart." Pretty extensive cardiac slowing is quite compatible with a fairly good blood pressure—vastly higher than what is necessary for the continuance of life. Pronounced slowing (without danger to life as a rule) is, of course, familiar in some forms of violent pain—for example, in biliary and renal colic, etc.

The conditions under which sudden death most commonly occurs—namely, muscular exertion—are not favourable to prolonged vagus inhibition, but on the contrary are associated with reduction of the normal vagus control over the heart and concomitant activity of the augmentor nerves, leading to acceleration with increased force of the beats, etc.—conditions favouring the development of fibrillation in predisposed subjects. The same holds good generally in emotional excitement, apart from the very brief standstill (or prolongation of diastole) which may be caused by sudden fright.

Vagus Inhibition succeeded by Fibrillation.

There is another possibility with reference to the effects of vagal inhibition. Experiment has shown that attacks of auricular fibrillation of varying duration sometimes develop after a period of inhibition, sometimes after one or more recommencing auricular beats have occurred. The development of fibrillation in these instances is definitely related to the occurrence of the preceding phase of inhibition induced by vagus stimulation.

Fibrillation of the ventricles has also been seen following vagus standstill, but this is a rare phenomenon as compared with the development of auricular fibrillation in similar circumstances. In the course of researches over a great many years I have obtained records of only a very few examples. Still these are enough to indicate that, in presence of undue ventricular susceptibility, fibrillation may sometimes be determined in this way in some of the manifold varieties of abnormal conditions that can occur in man. Thus vagus inhibition, much too brief to be dangerous through the standstill induced, may possibly involve a mortal issue through a succeeding fibrillation.

Fibrillation or Reflex Inhibition in Sudden Death in Anginal Subjects.

The following considerations may be cited in favour of the fibrillation view of death in angina.

1. The presence of coronary and myocardial conditions

which are known in certain circumstances to predispose to fibrillation. The invariable or almost invariable occurrence of coronary lesions in cases of *fatal* angina is a matter of general agreement. Sir Clifford Allbutt has adduced a wealth of facts and considerations marshalled with his usual skill, in favour of his view of the aortic origin—as contrasted with coronary and myocardial origin—of anginal *pain*. But this veteran clinician at the same time recognizes that the question of a *fatal issue* to an anginal attack is essentially associated with the condition of the myocardium.

2. The recognition by numerous observers, in some anginal attacks, of acceleration of the heart's action with irregularities, extra-systoles, etc.; such are known in many conditions to herald the onset of fibrillation. Among others, Windle recorded a fatal attack of angina in which the heart rate rose from 75 to 150 and became very irregular.

3. Though some slight slowing of the heart may occur in an anginal attack, there seems to be no direct evidence of the occurrence of pronounced inhibition, such as might, if somewhat intensified, threaten a suddenly fatal issue; the degrees of inhibitory slowing observed have been far removed from determining circulatory failure or even causing any considerable fall of blood pressure. There seems to be no relation between the severity and duration of the pain and the tendency to die in the paroxysm.

4. Death often occurring at the beginning of an anginal attack or in one that is relatively slight as regards pain, etc., is probably of the same mechanism as absolutely sudden death occurring between attacks or in persons who are not subjects of angina; the considerations bearing on such deaths are probably applicable to deaths *during* anginal attacks.

5. In the case of death between attacks or during relatively slight pain there is no evidence of such powerful afferent excitation as might be supposed to produce cardiac inhibition of such intensity and duration as to be fatal.

Of course it would be rash to dogmatize at the present time on an exclusive application of one mechanism as being the only one operative in all instances of anginal death; it may be that one or other form is present under different conditions. But there is a strong case for fibrillation as a common mode of death in anginal subjects, whatever the precise mechanism of *pain* production in angina may be.

Conclusions as to Sudden and Unexpected Death of Cardiac Origin.

Rupture of the heart is a very rare accident. Simple standstill of the ventricles in complete heart-block can only be a rare cause—assuming that such standstill may sometimes kill without fibrillation as the terminal event. As

has been stated, it is open to grave doubt whether reflex vagal inhibition *per se*—that is, without the supervention of fibrillation—is responsible for many deaths. Blocking of the mitral orifice by a thrombus and embolism of the pulmonary artery are known to be of very rare occurrence. (Thrombosis and embolism of the coronary arteries kill, as has been already stated, by fibrillation.)

The old idea of a heart that is working with fair efficiency abruptly " failing to contract " against excessive resistance may be set aside as untenable, in the absence of the sudden action of violent poisons, etc.

There remains the conclusion that the great majority of absolutely sudden deaths are to be ascribed to ventricular fibrillation.

Symptoms of Ventricular Fibrillation in Man and Animals.

The similarity between the group of symptoms associated with many cases of sudden death in man and those attendant on ventricular fibrillation experimentally induced in animals is indeed striking. The abrupt abolition of pulse, cardiac impulse, and heart sounds, the sudden fall of blood pressure, unconsciousness, muscular relaxation often preceded by a brief phase of rigidity or convulsive movement, dilatation of the pupils, and the continuance of slow deep respiratory movements, are identical in animals and in the human subject, while in the latter the speedy replacement of the initial intense pallor by lividity or marked cyanosis is a notable feature. The amount of colour in the face, together with the occurrence of several respirations after the collapse, have sometimes made onlookers somewhat incredulous that death has taken place. The occurrence in some cases of premonitory features such as extra-systolic irregularities, bouts of tachycardia, etc., is significant. It is important that, in the collection of evidence as to unexpected and unexplained death, special attention should be devoted to ascertaining the occurrence of the group of associated features just stated; these have been very definitely recognized in many cases of sudden death where accurate observations have been made. Some such cases have come under the direct observation of the writer.

No doubt some of these phenomena are common to certain other forms of sudden circulatory failure—for example, from heart-block, collapse induced by violent afferent impulses (blow on epigastrium, etc.), or in certain cases of auricular flutter, etc. But there are special features in some of these—for instance, as regards the behaviour of the respiratory centre, etc.

Protective and Remedial Agencies.

In animal experimentation—on cats, which are remarkably liable to ventricular fibrillation—the use of certain drugs has been found by the present writer[40] (working in

conjunction with Professor Spencer Melvin and Dr. J. R. Murray) to have decidedly beneficial effects, both in the way of protection against the onset of persistent fibrillation (for example, against faradic currents one hundred times as strong as are usually effective) and—in combination with cardiac massage—as regards recovery from the actual attack. These methods are at present only applicable under experimental conditions. But they give some ground for hope that, with fuller knowledge of the conditions that influence the inception and persistence of fibrillation, much may be possible in the future as regards the warding off of such catastrophic happenings as often bring life to an unexpected and sudden close from failure of cardiac function, occurring often in persons whose hearts are far from being worn out, but on the contrary are endowed with myocardial power amply sufficient not only for quiet existence but not infrequently for the demands of considerable bodily and mental activity.

There is reason to believe that in man, as in animals, an undue susceptibility to fibrillation is sometimes a temporary phenomenon depending on circumstances that may be more or less markedly transitory, though no doubt it is very often a persistent condition depending on abnormal changes in the ventricular musculature; in the latter case immunity from sudden death must in large measure depend on avoidance of the directly provocative causes of fibrillation in a predisposed heart, such as sudden muscular exertion, especially when accompanied by emotional stress, etc.

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SOME APPLICATIONS OF PHYSIOLOGY TO MEDICINE.

III.—BLOOD PRESSURE AND HEART ACTION IN SLEEP AND DREAMS:

THEIR RELATION TO HAEMORRHAGES, ANGINA, AND SUDDEN DEATH.*

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THIS is an important, and in some of its aspects an almost unexplored, field of study, with an obvious bearing on many questions. Precise data on the subject are naturally somewhat difficult to obtain. The present paper contains some results of slowly accumulating observations carried on by the writer as opportunities presented themselves over a long series of years.

Changes in Normal Sleep.

The slowing of the pulse rate (noted by Galen) and the respiration during sleep has long been known to be accompanied by a lowering of bodily temperature, a great reduction in metabolic activity and heat production, depression of reflexes, diminished secretion, etc. There is general agreement as to a definite lowering of the systolic blood pressure, varying in different conditions and as recorded by different observers, but often amounting to 15 to 30 mm. Hg at the end of two hours' sleep; the pressure gradually rises in the later portion of the night's sleep. Greater reductions have been noted in persons with high pressures in the daytime. Thus some years ago Brooks

*A communication on this subject was made to the International Physiological Congress at Edinburgh on July 26th, 1923. Part I of this series was published on January 13th, 1923 (p. 51), Part II on August 11th, 1923 (p. 215).

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and Carroll in 39 " hypertonic " subjects with an average systolic pressure of 204 mm. found a fall of 44 mm. after two hours' sleep; at the moment of waking it rose 22 mm. from the level present in sleep. More recently, in the last year or two, C. Müller in normal persons found the systolic pressure to be down to 94 mm. in men and 88 mm. in women during sleep, after a small dose of veronal. Blume, in men and women with moderate day pressures, recorded falls of 15 mm. and 21 mm. respectively, while in those with high day pressures the falls averaged 31 mm. and 39 mm. Much importance has been attached by some writers to this reduction of pressure; it has even been regarded, though on very insufficient grounds, as the determining cause of sleep.

It must be borne in mind that in the recumbent position a fall of aortic pressure would be necessary to prevent the pressure in the cerebral arteries from being higher in the horizontal than in the erect position—from the influence of gravity, the hydrostatic factor of the weight of the column of blood between the levels of the heart and the brain. Allowance for this factor in the recumbent posture cuts down the observed lowering of arterial pressure to a comparatively small amount, probably much too small to play the potent rôle that has been ascribed to it in the production of sleep. Further, the crude analogy of unconsciousness caused by an arrest or sudden great diminution in the blood supply to the brain cannot be regarded as valid when applied to the induction of the normal process of sleep.

Diminished vascularity of the brain substance during sleep has been described by various writers (Durham and others) on the ground of direct observations on exposed portions of the brain surface. A similar change has been inferred from the plethysmographic records obtained by Mosso and his followers, who found evidence of an increased volume of blood in the limbs (arm, etc.) during sleep, and assumed this to be attended by lessened vascularity of the brain, the converse conditions being present after awaking. But this hypothesis has to be revised in view of Leonard Hill's work establishing the practical accuracy of the old Monro-Kellie doctrine that the amount of blood within the skull is a constant quantity, whilst its distribution in arteries, capillaries, and veins respectively varies in different conditions. Weber's more recent work indicates that when there is less blood in the limbs there is more in the abdomen, not in the brain.

Plethysmographic observations have clearly shown responsiveness to stimuli during sleep, inducing alterations in the volume of a limb and showing certain changes in the distribution of the blood in the vascular system. But such observations give no information as to the state of the aortic blood pressure, upon which the pressure in the cerebral arteries depends.

Relations of the Period of Sleep to Some Diseased Conditions.

In accordance with the accepted view that the vital activities, as indicated by heart action, respiration, blood pressure, temperature, and general metabolism, reach their low-water mark in the early hours of the morning, it is easily intelligible that death from illnesses involving progressive exhaustion and a gradual running down of the machinery of life should often take place in that period. Statistics are available which bear this out. Thus Schneider (Berlin), in a total of nearly 58,000 deaths, found that deaths were most common between 4 and 7 a.m. Watson and Finlayson (Glasgow), dealing with records of nearly 14,000 deaths, fixed the highest mortality between 5 and 6 a.m.

Many phenomena of disease, aggravation of morbid conditions and symptoms in the night, can be brought into relation with the general lowering of vital activities during sleep—for example, some respiratory troubles which may in some cases be associated with the depression in respiration naturally occurring in that period, the reduced sensitiveness of the respiratory centre to the normal excitation by CO_2 with the consequent modification in the state of the blood, heightening of the grade of acidosis which may be present, development of Cheyne-Stokes respiration, etc., often associated with attacks of severe dyspnoea, etc.

Incidence of Haemorrhages, Anginal Attacks, and Sudden Death in the Night.

In contrast with the associations of depressed functions during sleep as affecting some of the manifestations of disease there is another class of phenomena for which a different interpretation is required, for they obviously do not lend themselves to explanation by the lowered vital activities of nightly rest and sleep.

In connexion with the subject of haemorrhages of various kinds and their times of occurrence and mechanism, questions arise. The time incidence of many vascular ruptures is naturally accounted for by the conditions prevailing at the moment of their occurrence—rise of blood pressure and increased stress on the walls of the vessels determining rupture at the weakest part—for example, muscular effort, the influence of gravity in certain postures, abdominal straining, etc.

But why should a weakened vessel give way during the period of nocturnal rest and sleep, since a lowered blood pressure is naturally protective against rupture? Why cerebral haemorrhage should frequently occur in the night and in sleep is a question that was asked long ago by Sir Samuel Wilks and apparently never answered. In view of the lowering of blood pressure and a diminished blood flow through the brain in sleep, why should a cerebral vessel

burst at that time? A similar question has to be dealt with in the case of pulmonary haemorrhage, which, as is well known, is frequently nocturnal in its incidence. The same applies to gastro-intestinal haemorrhages.

It is, of course, a matter of familiar knowledge that true anginal pain occurring in the daytime is commonly associated with exertion or excitement involving raised blood pressure and an increased call upon the heart, the pain diminishing or passing off with cessation of the muscular effort or emotional disturbance, reduction of the blood pressure by amyl nitrite, etc. But it is also well known that anginal pain sometimes seizes the patient in the quiet of the night, awakening him from sleep. What is to be put down as determining the onset in these cases?

Again, we know that sudden death in the night is not rare, sometimes coming thus to persons who have shown little or no evidence of serious departure from the level of their ordinary health, or at least nothing to warrant the expectation of so sudden a termination. In a former paper considerations were advanced in support of the view that the usual mechanism of such deaths is to be found in fibrillation of the ventricles, occurring in a heart which has become specially susceptible as a result of defective coronary blood supply, degenerative changes, toxic influences, etc. But, granted such predisposition, what is the exciting cause that precipitates the sudden and unforeseen disaster in the night-time?

Recognition of Two Different Conditions in Sleep.

The results obtained in the present investigation lead to the conclusion that in considering the subject of sleep we have to deal with two distinct conditions, which have strikingly different associations as far as nervous, circulatory, respiratory, and other functions are concerned: (1) undisturbed or sound sleep, attended by lowering of blood pressure, heart and respiratory rates, etc., and (2) disturbed sleep, modified by reflex excitations, dreams, nightmare, etc., sometimes accompanied by extensive rises of blood pressure (hitherto not recognized), increased heart action, changes in respiration, and various reflex effects. The circulatory changes in disturbed sleep are sometimes so very pronounced that it is somewhat remarkable that they should so long have escaped observation. So far as the present writer knows, the occurrence of marked rises in blood pressure during sleep has not even been suggested —apart from the fact that no actual measurements have been recorded. No doubt paucity of opportunities and difficulties in observation have stood in the way. But the considerations as regards the occurrence of haemorrhages, etc., in the night (stated in the earlier part of this paper) give distinct indications of the probability of important blood pressure changes being present in some instances.

Disturbed Sleep.

In connexion with the circulatory and the other phenomena of disturbed sleep there are various categories with regard to the degree in which the subject is able to recall his dreams or is conscious of the disturbances after awaking.

1. There is no recollection of the disturbed sleep or dreaming condition, though the presence of such was clearly shown by observations on the sleeper—occurrence of muttering, talking, groaning, movements of the face, fingers, etc.; reflex disturbances were evidently active in pronounced degree.

2. On awaking there is a sense of the sleep having been uncomfortable and troubled, but there is no recollection of dreaming having occurred.

3. The fact of dreaming is remembered, but not the definite sequence of the dream.

4. Vivid dreams remembered in great detail.

In all the above categories cardio-vascular disturbances, etc., have been recognized in more or less marked degree. These disturbances disappear at various periods after awaking—often in a variable number of minutes.

Distribution of the Changes in Disturbed Sleep.

The incidence of the recorded disturbances upon the various systems varies widely in different instances.

The heart's action may be specially affected by the impinging of nervous impulses on the cardiac regulating centres in the medulla, etc. There may be much acceleration of the pulse with comparatively little elevation of arterial pressure; there may be a strong cardiac impulse, with or without sensations of palpitation.* On the other hand, the arterial tone may be mainly influenced, vaso-constriction being chiefly instrumental in causing a large rise of pressure, attended in some instances by moderate or slow heart rates; in accordance with Marey's law the high blood pressure acts by increasing the controlling influence of the vagus centre over the rate of beat. A strong cardiac impulse and a large pulse wave may be prominently in evidence. Again, both heart and arteries may be markedly influenced, giving a high blood pressure with strong and rapid cardiac beats, powerful cardiac impulse, etc.

The respiration is sometimes much altered in the way of augmentation or irregularity, but there is no constant association between these changes and the circulatory disturbances; there may be marked respiratory changes while

* In contrast with the slowed pulse rate which is normal in the small hours of the morning, there is sometimes an acceleration at this period, apart from the development of abnormal rhythms, true tachycardias, etc., and without evidence of the more extensive disturbances of blood pressure, etc., described in this paper. Such acceleration is probably ascribable to reflex influences which have become operative during the preceding period of sleep in the earlier part of the night.

there is little or no evidence of circulatory alteration. Long ago Hammond described notable respiratory disturbances in dreams while describing the pulse as being unaffected, except in regard to slight irregularity ascribable to the respiratory alterations. In addition to the circulation and respiration the disturbances of troubled sleep may extend, in varying degree, over other systems, somatic and visceral, as evidenced by sweating, tremors, vomiting after awaking, etc. It is obvious that such disturbances acting on various functions in different ways may be responsible for important effects in some conditions of disease.

A notable feature (remarked long ago by Hughlings Jackson) is the absence (apart from somnambulism) of large movements of the limbs, etc., even during dreams of vigorous exertion, while movements of fingers, lips, etc., may occur, contraction of distal muscles being practicable while proximal ones fail. It would seem that impulses from the cerebral cortex can sometimes reach the medullary centres (cardiac, respiratory, vasomotor, sweat, etc.) while failing to activate the large muscles of the limbs even during dreams with strong emotional content.

The Dreaming State in the Dog.

The phenomena observed in the human subject are evidently paralleled by what is recognizable in the healthy dog during dreams of hunting, etc., with the familiar movements of toes and paws, tail and ears, biting action, series of subdued barks, etc. The heart is often rapid and irregular with inhibitory pauses, bouts of acceleration, etc., while a violent cardiac impulse may be perceived; respiration is frequently hurried and irregular, with gasps, etc. The knee-jerk may be increased—as Lombard noted in man during a dream of active movement. It has not been found practicable to get actual measurements of blood pressure that are satisfactory, for disappearance of the changes present in the dreaming state is very quick when the animal awakes. But the finger on an artery has sometimes given unequivocal evidence of a rise of blood pressure.

Some Characters of the Nervous Disturbances.

The extent and intensity of the functional disturbances which may be set up during troubled sleep and the dreaming state are remarkable, though quite intelligible in view of the diminution or suspension of the control normally exercised in the waking state by higher neural mechanisms, which come to be more or less completely in abeyance during sleep; released from such control the lower mechanisms are apt to give exaggerated responses to stimuli which would have comparatively little effect in the daytime. Thus afferent impulses (somatic or visceral)

which would have only slight and quite different effects in the waking state may call forth complex and pronounced reflex responses. The influence of afferent impulses in provoking and shaping the course of dreams need not be emphasized. Potent in this respect are impulses from the viscera which in the waking state would only be productive of slight sensations of discomfort—headache, nausea, etc.; in sleep elaborate responses may be set up, especially when unrestrained emotional processes are called into action with their resultant effects on both cerebro-spinal and autonomic innervation—excitation of sympathetic, etc. Such emotional excitation is apt to reach a high grade of intensity from lack of the balance and restraint normally exercised by the fully active mechanisms of ordinary consciousness, especially by those subserving the higher levels of mental function acquired through experience after the infantile stage of life.

The suddenness of development of the functional disturbances in blood pressure, heart action, etc., in the dreaming state is an important feature. As is well known, most dreams are of very brief duration as regards the actual time occupied, a number of seconds or a very few minutes often sufficing for a dream which is subjectively a long and complicated one—for example, an apparently long and varied dream has been recorded as running its course between the beginning and the ending of a clock striking midnight. The associated functional disturbances may thus be set up with unusual abruptness, as compared with the waking state—as, for instance, in ordinary muscular exercise. It follows that there is little or no time for the coming into play of the various adjustments and compensations in the circulatory and respiratory systems, etc., that are operative in muscular exercise; in the latter the rise of arterial pressure is checked by gradual dilatation of the vessels of the skin and the working muscles, while the heart accommodates itself with the aid of increased coronary blood supply, etc. Thus the blood pressure rise in certain dreams may be both large and steep in ascent. The call on the heart, through its nervous apparatus, etc., may also be a sudden one.

Influence of the Recumbent Posture.

So far as the rupture of a weakened cerebral artery is in question the hydrostatic factor in the recumbent position is an added consideration; the weight of the column of blood between the levels of the head and the heart, which reduces the cerebral artery pressure in the standing position, is now largely out of action; with a given aortic pressure the pressure in a cerebral artery is naturally higher by a very appreciable amount (varying according to the elevation of the head) in the recumbent than in the erect posture, and the danger of a cerebral

haemorrhage during a rise of aortic pressure is necessarily increased.

Observations on Blood Pressure, etc.

The subjects examined were persons mostly between the ages of 30 and 65, all, so far as was known, without organic disease of the circulatory system. The observations were made quickly after the awakening of the subject, the apparatus having been kept in readiness for immediate use. Systolic blood pressure was measured by the auditory

Hour of Observation.	Pulse Rate.	Systolic Pressure.	Diastolic Pressure.	Pulse Pressure.
1.0 a.m.	75			
7.0	62			
8.0	65	110	75	35
8.20 breakfast (in bed)				
8.50	75			
9.15	68–70	125	75	50
9.30	70	125	75	50
9.45	70	125	75	50
11.30	70	125	80	45
12 noon. Subject got up				
12.25 p.m. Sitting quietly	85			
12.55	81			
12 midnight. In bed after first sleep (somewhat disturbed)	70	140	80	60
Next morning—				
6.0 Subject awoke with feeling of disturbed sleep but no memory of definite dream. Arterial sounds (auditory method) very loud and the murmurish phase very pronounced.	62	182	105	77
Another reading two or three minutes later	—	165	95	70
6.15	60	145	90	55
6.30	58–60	115	70	45
8.0	60	130	80	50
10.30	76	120	70	50

During the day the pulse rate was generally 76 to 80.

and tactile methods, diastolic pressure by the auditory method. On occasions when no measurements could be made convincing evidence of the occurrence of extensive changes was obtained by ordinary digital examination of arteries, palpation of the cardiac impulse, etc. The following are some of the examples of the sort of observations made and the nature of the results obtained.

Subject No. 1.

The effect of walking upstairs (twenty steps) was compared in this person with the disturbances occurring in sleep. The pulse rate was raised from 80 to 90-95, the systolic from 120 to 140, and the diastolic pressure from 80 to 90; the observations were made while the ascent was being continued, not after its cessation. Ordinary walking exercise on a fairly level road caused comparatively slight changes in the heart rate and the blood pressure. In the same subject some days previously atropine (1/50 grain hypodermically) raised the pulse rate from 81-82 to 130, with systolic and diastolic pressures of 135 and 75-80 respectively, as compared with 115 and 70 before atropine. Abdominal straining (expulsive efforts) raised the systolic blood pressure only a few millimetres.

It is evident that in this individual the stress on the circulatory system was vastly greater during such disturbed sleep as is described above, than under the conditions of ordinary easy life with avoidance of sudden violent effort, emotional excitement, etc. The actual height of pressure attained during the disturbed sleep was, no doubt, decidedly higher than as measured after awaking when it is declining with some rapidity.

Subject No. 2.

Subject in bed in afternoon. While asleep he had lain on the right side with the right arm pressed on by the head so that the right radial pulse was abolished. The left radial pulse was found to be very fast and the artery large and strikingly tense, immediately after awaking; these unmistakable evidences of an extensive rise of blood pressure speedily passed off. The sleeper reported having been under the influence of a pronounced nightmare, with the illusion of his lying prone near the door of a house while he heard a visitor approaching along the drive; he had vivid and distressing sensations of ineffectual efforts to rise. The nightmare was no doubt determined by the posture, the pressure on the arm, and the ischaemia caused. After awaking numbness and tingling were felt in the right hand, while the radial artery became very large and the skin flushed—evidently after-effects of the ischaemia.

Subject No. 3.

A subject, who had some symptoms of gastro-intestinal disturbance but was pursuing his usual avocations, had in the course of a night of broken sleep a dream in which he felt lively resentment at the irritating conduct of an official on a public occasion —a vivid dream but not distinctly a nightmare; there was no sense of fear, oppression, ineffectual effort, etc. On his awaking it was found that there was no sense of palpitation, no sweating, and no subjective alteration in respiratory sensations; no marked change in the respiratory movements was observed. But the cardiac impulse was greatly increased in force and felt over a larger area than usual in this person. The pulse was accelerated from a normal rate of 70-80 to 90-95. But the most notable change was a greatly raised blood pressure with an extensive pulse pressure, as shown by digital examination of the radials; the arteries were large and tense, obliteration difficult, and the range of the pressure variations at each beat palpably large. When examined fifteen minutes later these altered conditions were practically gone.

Some hours later another dream took place, the details of which were not clearly remembered. Similar phenomena, in

somewhat less pronounced degree, were recognized; these virtually disappeared in a few minutes.

In one dream systolic pressure rose from 130 to over 200

Emotion, Motor Effort, and Gastro-intestinal Disturbance.

While the most striking cardio-vascular effects are naturally present in dreams with a strong emotional content, it is to be noted that a vivid dream of active movement (cycling, for example) without sensations of nightmare, etc., may cause a pronounced rise of blood pressure. Thus in an instance of this kind the pulse tension was greatly increased and the pulse pressure was extensive while the heart rate remained at 72-75; the elevation of blood pressure was evidently brought about mainly by vaso-constriction.

It is noteworthy that the amount of disturbance (circulatory, respiratory, etc.) associated with vivid and alarming dreams varies greatly in different individuals and even in the same individual under different conditions; the effects are sometimes remarkably slight in the case of dreams that are at other times attended by very pronounced effects of the kinds described above. There is reason to believe that the presence of some gastro-intestinal disturbance at the time may sometimes play a part in facilitating the development of the more marked effects on circulation, respiration, etc.

Dangers of the Circulatory Disturbances.

These cardio-vascular changes, involving sudden demands on the heart's power with great alterations in its rate and force and a steep and sometimes very extensive rise in blood pressure, are quite harmless in the healthy individual. Vivid dreams, involving hurrying to catch trains, etc., with failure to do so, are common in many persons and sometimes persistently recurrent — with no injurious consequences apparently. But the case is obviously very much otherwise with a damaged vascular system, life going on under conditions which afford only a narrow margin of safety. There may be a myocardium abnormal in certain functional respects, whether or not these be attended by recognizable structural alterations with or without obvious coronary lesions, giving a susceptibility to ventricular fibrillation, or, on the other hand, a defective arterial tree with localized weakenings (by miliary aneurysms, etc.) in the brain vessels, tuberculous damage in the lungs, ulcerative conditions in the gastro-intestinal tract, etc., where haemorrhage may readily be determined. In a heart susceptible to fibrillation a sudden call on the heart during muscular exertion and excitement in the waking state is often fatal; in the disturbed conditions of sleep and dreaming a similar mechanism is sometimes brought suddenly and strongly into action— diminution of vagus control and, especially under emotional

stress, stimulation of the cardiac sympathetic together with a high blood pressure—conditions which favour ventricular fibrillation.

A possible discharge of adrenaline into the circulation under emotional excitation also comes into question, though the importance of such discharge or its existence has been denied by Stewart and Rogoff, in opposition to the well known work of Cannon and de la Paz. The time incidence of attacks of anginal pain may obviously be determined by similar conditions.

Conclusions.

It is clear that the foregoing facts must be taken as profoundly modifying the simple conception of night as the time of rest, and sleep as a condition in which quiescence prevails and recuperative changes go on, restoring the bodily and mental capacities which have become more or less reduced at the end of the hours of work and wakefulness—a period of repose also attended by sedative and beneficial effects on many morbid conditions. This conception, while true as regards undisturbed or sound sleep, has to be qualified by the consideration that night and sleep are occasionally the season of acute reflex and emotional disturbances which, in the peculiar conditions present, induce very pronounced effects on the circulatory system, throwing a formidable strain upon its weak points, whether these be cardiac, with susceptibility to fibrillation or anginal pain, etc., or arterial, with risk of rupture.

In this way the individual may, during the nocturnal period of assumed repose, be subjected to suddenly developed stresses, as estimated by the rise of blood pressure (even as measured after awaking when it is falling) and the evidences of increased heart action, far beyond what is involved in ordinary muscular exercise gradually initiated—for example, walking, cycling, slow ascent of stairs, straining, or mental excitement in certain degrees. Thus haemorrhages, the onset of anginal attacks, and other disturbances in the night can be readily accounted for; also sudden death, probably due to ventricular fibrillation in most instances

In the light of these observations it is easy to understand how in certain circumstances death may come like a thief in the night to a susceptible person living with circulatory conditions that approach the danger line, though these conditions may, in favourable circumstances and barring fresh developments, be compatible with many years of moderately active life.

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BLOOD PRESSURES IN MAN UNDER NORMAL AND PATHOLOGICAL CONDITIONS

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Long after accurate measurements of blood-pressure had been practised on experimental animals, the study of blood-pressure in man remained virtually a sealed book. Various early methods were tried without reliable results; it was not until Riva Rocci (112) and somewhat later L. Hill and Barnard (63) introduced the armlet method that systolic pressure estimation became really practicable. Even after this method had superseded the earlier attempts of Mosso, Gaertner, Von Basch and others, many results were rendered more or less inaccurate by imperfection in technique, too narrow armlets, etc., while the reliance on systolic pressure alone gave very inadequate and often misleading information as regards the state of the circulation. The later development of diastolic pressure estimation, especially by the auscultatory method, marked a great advance in the usefulness of the study of blood pressure. The adoption of the standard breadth of armlet or cuff as a result of Von Recklinghausen's (109) work was an important step.

As regards oscillatory methods the technique and the principles involved have lost much of their interest and relevancy, since their practical application has receded in importance in view of the development and general adoption of the auscultatory method. The superiority of the latter has become widely recognised, on the grounds of simplicity, quickness and accuracy, as compared with the more cumbrous apparatus and the more difficult and variable interpretation of the oscillatory records, different readings of pressure often being made from the same records by different observers of considerable experience or even by the same observers at different times—difficulties examined by Melvin and Murray (97) and others. A good many workers using oscillatory methods have found the Pachon oscillometer with its visual indications preferable to the Erlanger apparatus with its graphic records. The more recent Pachon apparatus has a Gallavardin armlet

with two independent pressure bags applied to the upper arm instead of the wrist; this is the best form of oscillatory apparatus at present available.

Experience has emphasised the importance of combining the tactile systolic index with the auscultatory systolic as a *routine procedure*, recommended by MacWilliam and Melvin (90) in 1914; the latter index should always show a higher value when the estimation is correctly made. This of course necessitates the use of one of the various forms of apparatus that provide for the retention of the auditory receiver in position, leaving the hands free, e.g., the Baumanometer, the Tycos, the Laubrey sphygmophone, the Oliver tambour, etc. The checking of the auscultatory systolic by the tactile systolic is essential for more than one purpose—as a guarantee of the proper functioning of the auditory apparatus, and in guarding against error which may sometimes arise after prolonged or repeated armlet compression. Repetition may be needed to correct the disturbances in pressure due to excitement, etc., at the first compression; as is well known, subsequent readings are frequently decidedly lower—until a constant level is reached, the residual pressure. Some persons under pathological conditions are specially liable to show a decided rise of pressure from repeated or continued compression especially when the arm becomes congested distally to the compressing armlet. Effects (reflex, etc.) from repeated compression by the armlet come into question, described by Gallavardin with Haour (53) and Tixier (54) as involving different types of pressure changes, rises, falls, etc.

The present writer has found important disturbances of the auditory indications in a certain number of subjects, as a result of compression which involves marked turgescence of the hand and forearm—a cutting down of the systolic index with a rise of the diastolic. Sometimes at a later stage of prolonged compression there is enfeeblement or disappearance (at variable points) of all the sounds below the upper region of sound, in the neighbourhood of the systolic level. Such disturbances may occur while the actual blood pressure is not changed —as shown by the tactile systolic index remaining unaltered; thus incorrect measurements including an unduly restricted pulse pressure may be obtained in these cases by the use of the auscultatory method alone—unchecked by the tactile method. Some subjects are exceptionally susceptible to the development of such disturbances; the significance of these differences in behaviour has not been determined.

Another purpose for which repeated compression has been used is to

reduce strong tonic contraction which may sometimes be present in thickened arteries, giving a certain resistance to obliteration and leading to an over-estimation (to some extent) of the actual intra-arterial pressure. But a better method is to close the brachial artery for three minutes by digital pressure upon it, as recommended by MacWilliam and Melvin, instead of constricting the whole limb by the armlet and leading to venous turgescence and the associated tendency to error described above—as especially applicable to a certain (limited) number of subjects. Experiments (unpublished) in this Laboratory show that in some instances at least the trouble is caused mainly by congestion of the limb, not simply by pressure on the brachial artery. Digital compression while removing the abnormal resistance to obliteration of the pulse does not induce the auscultatory error referred to above.

When the auscultatory method is rendered difficult by noisy surroundings or by impaired hearing in the observer, the vibratory method of Ehret (38) a modification of the tactile method, can be usefully combined—a finger being applied to the artery on the distal side of the auditory tambour, to detect the vibration associated with the sound at the diastolic level. This method is strongly recommended by Gallavardin (51). It is much simpler than checking by oscillatory methods. But difficulties are present in fat subjects with deep brachials, small calibre, and cases (especially aortic regurgitation) where the change in the vibration constituting the diastolic index is less definite than usual.

As regards the mechanism of the sounds, the subject of various conflicting views (Gittings (55), Erlanger (39), MacWilliam and Melvin, L. Hill and others) there have been interesting investigations by Gallavardin and Barbier (52), (8) who describe two zones in the curve of sound; 1, in the upper half of the curve with maximum near the systolic index and murmurs caused by whorls in the blood current as it passes through the compressed area of the artery and gets into a region of lower pressure distally to the armlet; 2, in the lower half of the curve with maximum near the diastolic index, the sounds here originating in the vessel wall and related to sympathetic nerve influence on the arterial wall. Attempts to interpret the meanings of the notable variations in the character of the sounds in different subjects, the duration of the phases and the changes induced by armlet compression have been made by Gallavardin with Haour, Barbier (52) and Tixier, and by Tixier (126), B. Smith (116) Sorapure (117) and others, following

observations by Ettinger, Goodman and Howell (57), (130) Warfield and others. The characters of the sounds in different conditions are so varied and striking that useful information as to circulatory states may very possibly be derived from them when they are better understood.

It need hardly be emphasised that the Hg manometer is the reliable means of measuring pressures; it is only when frequently checked against this instrument that other forms (aneroids, etc.) can be taken as giving valid evidence. In regard to diastolic pressure it may be noted that the reading taken when the armlet pressure is being raised is often appreciably lower (5 mm., etc.) than when taken during deflation; in some subjects the difference shows more than in others. With reference to the systolic index the difference in the readings by the auscultatory method and those by the tactile method have been estimated at 5–14 mm. The experience of the present writer agrees with the lower values, usually only a few millimeters.

Blood pressures in young adults. In 1914 Melvin and Murray (97) established by accurate methods normal values of both systolic and diastolic pressures in healthy young male adults (sitting posture), 59 medical students, average age 20–9. As regards systolic pressure only three were up to 130 mm. (viz., 130, 134 and 135) while five were slightly below 100 mm., the average came out at 112 mm. Of the diastolic pressures 28 were at 60 to 70 mm., 19 at 70 to 80 mm. and 12 at 50 to 60 mm. The pulse pressures gave an average value of 46 mm. Subsequent observations on very large numbers of subjects by various observers in different parts of the world have given S. and D. values higher as a rule and sometimes with wider ranges of variation. Bearing on this difference the observations of Alvarez (4) and of Burlage (23) (to be stated presently) as to a lowering of pressure in the early years of adult life are suggestive, as they include the ages dealt with by Melvin and Murray. Sorapure (117) examining 769 British soldiers also found a systolic maximum at 19 to 22 years followed by a slight fall, as Stocks and Karn (121) did after a systolic maximum at 19 to 20. It may be remarked that while large numbers are of course necessary for statistical purpose, classification, etc., reliance on pressure measurement on a single occasion is apt to introduce sources of error, in view of the universally recognised tendency of single examinations to give results disturbed by temporary causes, nervous excitement, etc. More precise results, as regards the real pressure levels in individuals are obtainable by a more intensive study of smaller numbers, by repeated examinations under carefully ascertained and controlled conditions.

Among more recent investigations Alvarez (4) and his associates made observations on systolic pressure (tactile method) on a very large number of University students (6000 men and 8934 women). In women the average was 11 mm. lower than in men. In men (reclining after tepid shower bath) the pressures grouped mainly about 127 mm. at age of 16 and 118 mm. at 30; in women (standing) 118 mm. at 16, 111 mm. at 24, 117 at 40. There is thus a noteworthy lowering of systolic pressure in the early years of adult life, the average dropping from age 17 to 21 in men and remaining at about the same level till after 50; in women falling from 17 to 25, rising after 25 and especially after the age of 40. After 45 the average pressure is higher in women than in men. A fall of pressure was also noticed by Burlage who made observations on 1700 girls by the auscultatory method; he found a systolic pressure of 104 mm. at 9 years, 124 at 14 and 15, falling to 114 mm. at 18, then constant to 26. Alvarez notes that relatively high pressure is common in young men—45 per cent over 130 mm., 22 per cent over 140 mm.; in young women, 12 per cent over 130 mm. and 2 per cent over 140 mm. Evidence of the occurrence of comparatively high pressures in some young men is also to be found in the results of Barach and Marks (7), Lee (81) and others. Alvarez pronounces his extended investigation as not entirely satisfactory in establishing normal systolic pressure standards for young men on account of the lack of homogeneity, without arriving at any definite explanation of the results, the possible causes of which he discusses—low pressures in 1918, raised in 1919, gradual return in 1920–21. Further there is the disturbing observation that the averages even in 1918 were considerably above those of high school boys of the same age in the same year.

Conception and Bulatao (29), examining 717 subjects (average age 28) in the Phillipines, found in males S.115, D.79; in females S.116, D.83. Pulse rate a little over 72. In Denmark, Faber (43) in 1000 healthy soldiers (ages 20 to 25) by the Riva Rocci method (recumbent) found S. pressures of 110 to 130 mm. in 80 per cent, and higher or lower, 84 to 156 mm., in 20 per cent. Emphasis is laid on these great variations of systolic pressure in healthy men. Men of greater weight showed higher pressures than others of the same height; with equal weights blood pressure is lower, though the differences were slight, in men of greater height. What are termed the "overfat" averaged 123 mm. as compared with 117 mm. for the "underfat." Diastolic pressures are not recorded. The systolic pressures found by Faber agree with those obtained by Tavaststjerna (124), whose average was 117 mm.

Addis (1) examined nearly 400 subjects in two categories, the pressures being taken in the recumbent posture—1, under basal conditions; systolic average 99, diastolic 71. 2, Under other conditions, food taken, walking, etc., systolic average 127, diastolic 78.

The relation of blood pressures to age. There is general agreement as to the presence of lower systolic and diastolic pressures in childhood, the differences from the adult being more marked in the systolic levels with a consequent diminution of the pulse pressures.

The work of Judson and Nicholson (71), Melvin and Murray, and Faber and James (44) may be referred to, also the more recent observations of Stocks and Karn (120). In connection with the smaller pulse pressures the quicker pulse rate of children has to be taken into account, tending to make the product of P.R. $\times$ P.P. approximate to what holds good in the adult.

The available evidence shows that from the very early phase of life there is a progressive steady rise of pressure, apparently a function of increasing age, up to the onset of puberty, then an acceleration of the rise up to the ages of 17 to 20. It is to be noted that there are decided differences between the results of Judson and Nicholson and of Faber and James on American boys, and those of Stocks and Karn on British boys, as regards the actual values of the pressures recorded and the extent of the rise between the ages of 5 and 14 years. At the former age the American observers found systolic averages of 92 mm. and 93 mm. respectively; at the latter age 106 mm. and 110 mm. On the other hand Stocks and Karn report a lower average, 85 mm., at age 5 and a higher level, 115 mm. at age 14—a rise of 30 mm. which is nearly twice that found by the other observers. The accelerated rise during puberty and adolescence between the ages of 13 and 17 has been found by Stocks and Karn to amount to 16 mm.

Woley (134) dealing with systolic pressures in 1000 apparently healthy subjects, found an average level in males of all ages of 127.5 mm., in females of 120 mm., and a rise from 122 mm. in the age group 15 to 30 years to 132 mm. in the 50 to 60 age group. He distinguished a high pressure group with an average pressure of 141 mm. at the ages 15 to 30 to 149 mm. at 50 to 60, and a low pressure group rising from an average of 103 mm. in the 15 to 30 group to 115 mm. in the 50 to 60 category. At the intermediate ages averages of intermediate value were obtained, a gradual rise occurring with increasing age and a corresponding rise in high and low averages. He regarded a pressure of 144 mm. in the 50 to 60 age group as being definitely acceptable for

insurance. Women at all ages were 8 mm. below the male average, with the same ratio of increase at similar ages.

In Symonds' (122) report of 150,419 men successive age groups are presented from a 15 to 19 year group up to one of 60 years and over; also build groups (Medico-Actuarial Investigation I, 1912, 120) based on the average weight for each inch of height in men at the age of 37. Systolic pressure alone is studied. Age, weight and pressure are shown to increase together. Differences appear of 11 to 12 mm. between the youngest and the oldest in each build group, and of 10 mm. between the very light and the very heavy groups; even at the ages of 60 and over the difference was much the same. In the whole series the pressure averages range from 121.2 mm. in the youngest (15 to 19) group to 135.2 mm. in the oldest (60 and over) group. Mackenzie (87) reporting on 18,637 men, gives a range from 119 mm. to 137 mm. in similar age groups. Rogers and Hunter's (113) 62,000 rise from 120 mm. at 15 to 19 to 134 mm. at 55 to 59; the results of Fisher (46) and Goepp (56) are very similar though the age grouping differs slightly. As regards the pressures in women, Symonds' 12,000 with age grouping similar to the men show values ranging from 119.2 to 135.5—a much closer approximation to male pressures than has been found by most observers who have commonly reported pressures in women, at least in the first half of life, as 8 to 10 mm. lower than in men.

In a recent valuable study Stocks and Karn (120) present continuous evidence of the pressure behaviour through the ages of puberty and adolescence, from the ages of 5 to 40 years. They submit curves and tables for the correction of pressure readings for age, weight (affecting systolic pressure) height (affecting diastolic pressure) and pulse rate. They find that there is a positive correlation of systolic pressure with muscular strength apart from physical development and age. By their method of correlation with pulse rate they believe that an approximate correction of the well-known disturbing effects of psychological factors, such as nervousness, can be made. With regard to this conclusion of Stocks and Karn it is to be remarked that excitement, emotion, etc., influence blood pressure by acting on the vasomotor centre as well as on the heart, and that the relation between the two actions is by no means constant; hence the pulse rate cannot be relied on to give accurate indications of the degree of pressure alteration developed, though a correction for pulse rate no doubt diminishes the amount of the error. The range of apparent variability of the blood pressure in healthy persons is substantially diminished by such cor-

rections as the preceding, though not removed, since other and more obscure factors remain. Stocks and Karn's figures show a remarkably small increase of pressure with advancing age—from an average of 131 mm. at 20 years to 134 mm. in the group of 40 years and over (average age, 49).

It is warrantable to conclude from the concurrent evidence of the extended statistical evidence now available that the idea of an extensive progressive rise of systolic pressure in healthy persons as life goes on is an erroneous one. It is clear that the rising pressure of childhood undergoes accleration about puberty and attains what is approximately the adult level somewhere in the 17 to 20 period. There is some evidence of a slight subsequent lowering—in the early years of adult life. Apart from this, the pressure remains almost steady till the age of about 40, after which a more definite rise progresses. But the rise, though quite a definite one, is more limited in amount than is commonly assumed; the total rise shown in the statistics, due to the combined influences of age and increasing weight is on an average under 15 mm. The pulse pressure follows a course pretty similar to that of systolic pressure under the influence of age. The available evidence also bears weighty testimony to the relative constancy of systolic pressures at different ages, when large numbers are dealt with and the necessary allowances and corrections are made. It is clear, in view of the foregoing averages, that the occurrence of exceptionally high pressures in healthy persons must be relatively rare; otherwise the averages would be much higher. The very moderate level of the averages for middle and advanced life is all the more noteworthy in view of the fact that the real ordinary pressures are likely to be overestimated rather than under-estimated, under the influence of nervous excitement, etc. But while the averages for large numbers are relatively constant, the fact of notable variation in healthy individuals remains, the pressures in such persons being apparently set at levels different from the ordinary—from causes that cannot at present be adequately defined. The importance of hereditary influences has been emphasised by numerous observers, e.g., Oliver (104), Dana (34), Alvarez, Warfield (130) and others.

With regard to the very high systolic pressures, S.200 to 250, sometimes (though rarely) met with in apparently healthy vigorous men at such ages as 50, 55, etc., the mechanism of such pressures is well worthy of careful investigation—with respect to the peripheral resistance, capillary and venous pressures, blood volume, cardiac output,

etc.; such would probably yield valuable information as to circulatory conditions. Various observers have noted that in a considerable number of high pressure cases there are no definite symptoms and no evidence of disability; Kulbs (77) found so high a proportion as 20 per cent in this category in a series of 172 males and 116 females with pressures at or above 170 mm. As regards diastolic pressures the American life insurance data are open to criticism from the adoption of the end of the 4th phase as the diastolic index, the latter having been experimentally proved to coincide with the beginning of the 4th phase —in the dog by Warfield (129) and in the sheep by MacWilliam, Melvin and Murray (91). It is certain that serious error may occur in this way especially in young subjects where the duration of the 4th phase may in some cases be long. Thus Melvin and Murray, by careful examination in quiet surroundings using a sensitive Oliver auditory tambour, found in 14 young men out of a total of 59 a prolonged 4th phase, ranging between 24 and 55 mm. and averaging 38 mm. The lower limit of the sound was sometimes found to be as low as 10, 14, 20 or 22 mm. armlet pressure in healthy subjects with normal systolic pressures and complete absence of any collapsing character in the pulse, etc.; obviously these figures could not possibly represent the actual diastolic pressures.

Of course such very low readings of the lower limit of the sound are exceptional. Many observers have noted a 4th phase of shorter duration, e.g. Warfield up to 20 mm., Weysse and Lutz (131) not above 25 mm., Tixier usually 20 to 30 mm. at ages of 20 to 30 years and in some abnormal subjects 20 to 40 mm., etc. Others have reported figures 5 to 8 mm. (Goodman and Howell, Barach and Marks, Mackenzie, Smith and others). In middle-aged and elderly subjects the experience of the writer is that the sound rarely persists in any important degree (not more than a few millimeters) and consequently the lower limit of sound in these subjects approximately indicates the diastolic pressure—in contrast to the serious discrepancy which may occur in young persons.

In the insurance statistics referred to the relatively large numbers in the younger groups of subjects (19 to 25 and 25 to 30) would naturally tend to give scope for possible errors in this direction. But such errors would be in the direction of underestimating the actual diastolic pressure; on the other hand, the diastolic readings given in the insurance series referred to are by no means low.

There is sometimes a tendency to undervalue precision of blood-

pressure measurement and to regard differences of 10 to 20 mm. in blood-pressure readings as being of small moment in view of the larger variations that may occur from time to time with apparently little significance. But the importance of such differences varies greatly in relation to their position in the scale of pressures. When they start near the "normal" lower limits of systolic and diastolic pressures, differences of 10 to 20 mm. may mean much, e.g., between 100 and 80 or 90 systolic, or between 60 and 40 or 50 diastolic, such are of much significance as compared with similar amounts at higher levels.

Relations of systolic, diastolic and pulse pressures. The 3:2:1 ratio commonly cited as applicable to these pressures is subject to very considerable variations without coming into the category of the abnormal. The validity of the ratio is mostly evident with certain normal pressures, e.g., S. 120, D. 80, P. P. 40, the pulse pressure being one-half of the diastolic and one-third of the systolic. It does not hold good in such low pressures as may sometimes be found in healthy persons, e.g., S. 105, D. 60; where the P. P. is three-fourths, instead of one-half, of the diastolic and much nearer one-half than one-third of the systolic. Again with such a high diastolic as 120 mm. (muscular effort, etc.) a S. 180 and P. P. 60 are apt to be under what actually occur, the high diastolic tending to give a relatively higher S. and P. P. on account of the tense condition of the arterial walls; the discharge from an efficient L. V. causes a disproportionately large rise of systolic pressure—apart from the influence of an increased discharge per beat occurring in a distended or enlarged heart. Such effects of diminished distensibility of the arterial system at high diastolic pressures may also be paralleled by loss of elasticity and stiffening of the arterial walls from degenerative changes, apart from the presence of a high diastolic pressure. These factors being operative at different ages, it is evident that while the average (absolute) values of pulse pressure in large numbers of persons vary with age in the manner already stated, the actual amounts in individuals may be greatly affected, apart from the influence of age, in the ways just stated. Pulse pressures of deficient amounts associated with a high diastolic level are naturally of evil significance as indicating cardiac inefficiency, provided the low P. P. is not accounted for by acceleration of the pulse rate.

Blood pressure in muscular exercise. While a rise of blood pressure has long been known to be associated with muscular exertion, the great majority of the measurements have been made after the period of exertion has ended and have for the most part dealt only with sys-

tolic pressure. The methods adopted by Bowen (19) (systolic pressures only) and by Lowsley (86) (systolic and diastolic by the Erlanger sphygmomanometer) enabled the course of the pressure changes to be followed throughout the period of exercise (stationary bicycle). They found a rapid rise at first, reaching a maximum in a number of minutes, (Bowen, 5 to 10 minutes, Lowsley, 5 to 25 minutes) then slowly declining and after the end of the exercise sinking to normal or subnormal levels.

McCurdy (96) measured systolic pressure during brief maximal effort (heavy lifts involving thoracic fixation, etc.) lasting about 5 seconds, and found an average rise from 111 mm. before the effort to 180 mm. during and 110 after.

Measurements made "immediately" or at certain periods afterwards are not valid guides to the actual height of the pressure during the exercise, though such have been used by some observers in comparing the blood-pressure response to effort in different types of subjects. Thus some observers have taken estimations half a minute after the termination of exercise as a standard. Cotton, Lewis and Rapport (28) making repeated measurements found a fall to, or nearly to, normal in 10 seconds then a rise reaching a maximum in 20 to 60 seconds, there is then a gradual fall, reaching the resting level in from 1 to $4\frac{1}{2}$ minutes after the end of the exercise (20 pound dumb-bells). Similarly Chailly-Bert and Langlois (27) recorded a fall in 5 seconds after the cessation of exercise to about normal, followed by a subsequent rise. Graupner (58) and Barringer (9) had previously described a secondary rise after the end of the period of exertion with a Zuntz ergometer and with dumb-bells respectively. These observations mainly deal with systolic pressures. The interpretation put upon their results by Cotton, Lewis and Rapport is that, assuming the veins to be depleted during the period of exertion, these veins fill up with blood when the muscular action ceases and so cut down the return of blood to the heart and the arterial pressure, until the veins have refilled and the inflow into the heart is restored, leading to the subsequent rise in presence of the continuance of the factors, other than the pumping action of the muscles, operative during the exercise.

This interpretation has been disputed by Bainbridge (6) who regards the veins as being full during the period of exertion, and the fall of arterial pressure to be due to the cessation of the pumping action of the muscles inducing a momentary stasis of blood in the capillaries, involving a temporary diminution of the venous return to the heart.

It may be remarked that in neither of these explanations is it definitely stated whether the whole of the venous system is regarded as depleted or full—according to one view or the other—or whether the state of *a*, the large venous trunks in the thorax and abdomen, or *b*, the veins among or near the muscles in the trunk and limbs are specially in question. Rapport (108) noted variations in the duration of the secondary rise after moderate and great efforts respectively.

Observations by C. Reid (110) in this Laboratory show that the rate, character and extent of the pressure changes after the end of the exercise vary much in different individuals and in the same individual under different conditions.

The maximum height attained by the subsequent rise of pressure, when such occurs, varies much and bears no precise or constant relation to the maximum height during the period of exertion, though under some conditions it approximates or corresponds to that maximum.

After a type of exercise where the raised pressure during the exercise shows a simple decline afterwards, without a subsequent rise, the rate of the decline varies considerably, and measurements taken at some fixed point of time (e.g., $\frac{1}{2}$ minute) are not to be relied on. Again, in those forms of exercise where a subsequent rise does present itself, occurring in varying degree after a preliminary fall, it is obvious that much will depend on the exact point in the series of changes at which the estimation is made. Measurements at half a minute after the cessation of exertion will naturally give very different results according as a subsequent rise develops or a simple progressive decline occurs; even in the latter type the finding of equal readings at the half minute interval in two different individuals or in the same individual at different times and under different conditions does not prove that the maximum pressures attained during the exercise were equal in the two instances. It is to be emphasised that measurements during the period of exertion constitute the only valid evidence as to the actual rise of pressure. It is not surprising that many discordant results have been recorded by different observers dealing with exercises of different types and duration or even with comparable exercises, when the estimations are made after the end of the period of exertion. Quite small rises (e.g., 16 mm.) have been reported after short spells of severe exertion involving dyspnea with doubling of pulse rate, etc., when the actual pressure during the exercise has really been greatly raised.

Whatever significance may be attached to such estimations for some purposes, it is clear that they are not reliable for determining the

height of the blood pressure response to exertion. The extent and course of this response varies much in different types and degrees of muscular activity—whether the latter be 1, strong or maximal effort with fixation of the thoracic walls, etc., bringing in the factors concerned in Valsalva's experiment; 2, exercises of endurance as in walking, long distance running, cycling, etc.; 3, execution of difficult, though not necessarily strong, movements involving much mental concentration; 4, static contraction of muscles.

A direct relation of the blood pressure rise (associated with exertion) to the amount of work, rather than to its rate, has been affirmed. This is applicable in a general way to certain types of exercise where the mental factor remains tolerably constant, but it is not applicable for comparison between different types involving variable degrees of mental concentration, emotional accompaniments, etc.; in these very different amounts of blood pressure change may be associated with the performance of equivalent amounts of muscular work.

Blood pressures in sleep. That there is a lowered blood pressure during sleep has been found by various observers, often amounting to 15 to 30 mm. at the end of two hours' sleep, then gradually rising toward the time of waking. Such falls of general arterial pressure obviously mean only a relatively limited reduction in the brain vessels —the hydrostatic factor being largely taken off the head vessels in the recumbent posture. Greater reductions have been noted in persons with high pressures in the daytime, e.g., 44 mm. by Brooks and Carroll (22) in hypertonic subjects.

Muller (100) found the systolic pressure to be down to 94 mm. in men and 88 mm. in women during sleep, after a small dose of veronal. In persons with moderate day pressures Blume (15) recorded falls of 15 mm. and 21 mm. in men and women respectively while in subjects with high day pressures the lowering averaged 31 mm. and 39 mm. These observers describe a remarkable constancy of pressures during sleep (rarely more than 5 mm. variation in sleep), even in high pressure cases, in contrast to the great variability seen in the waking pressures. Katsch and Pansdorf (72) while confirming the fall of systolic pressure in sleep—parallel to the depth of the sleep—found that the diastolic pressure sinks little if at all, but on the contrary often rises during the deepest sleep, so that the pulse pressure is diminished. In essential hypertension they observed an abnormal range of systolic lowering; in other hypertensions little or no lowering.

The present writer (88) finds that there are two entirely different

conditions in question in sleep—1, sound sleep with lowering of pressure, 2, disturbed sleep, dreaming, etc., which may be attended by remarkable elevations of pressure, e.g., systolic pressure raised from 125 to 182 mm., or from 130 to 200, etc.; diastolic pressure raised from 75 to 105 mm., etc. These changes were much greater than were induced in the same individuals by moderate exertion (cycling, walking, stair climbing, etc.) straining abdominal efforts, dose of atropin to remove vagus control over the heart, mental excitement, etc. In view of the rapid development of such changes in sleep, especially in dreams of motor effort, nightmares, etc., it is evident that a formidable strain—harmless in the young and healthy person—may thus be thrown on the weak points of the circulatory system, whether these be cardiac with susceptibility to anginal attacks or to ventricular fibrillation and sudden death, or arterial with risk of hemorrhages, cerebral (especially in the recumbent posture), gastro-intestinal or pulmonary. The conception of sleep as a period of quiescence and recuperation has thus to be qualified by the contingency of disturbed sleep with active calls on the nervous system, the heart and the blood-vessels. The mechanism of the rise of pressure in disturbed sleep differs in some respects from that present in ordinary muscular exertion, since in the former the pumping action of working muscles, greatly augmenting the venous return to the heart, is absent. The above-mentioned disturbances may occur during disturbed sleep when there is after awaking no recollection of definite dreaming.

High blood pressure. Notwithstanding the very large amount of attention that the subject has received the causation and mechanism of persistently elevated blood pressure, whether in the form of simple or essential hypertension (the hyperpiesis of Clifford Allbutt (3)) or in association with kidney lesions, remain unexplained. While there is general agreement as to the existence of excessive pressures apart from any recognisable renal lesions and in the absence of any sign of functional inadequacy as tested by the modern methods for estimating renal efficiency, it is also clear from the evidence available that the significance of hypertension is greatly influenced by the co-existence of renal inadequacy, the latter giving a sinister aspect to the condition and seriously altering the prognosis. While there has long been a strong presumption from the clinical side that, 1, toxic substances, probably protein derivatives, are at work, whether *a*, absorbed from the alimentary canal (pressor amines, etc.), or *b*, products of microbic infection, or *c*, abnormal metabolism; or 2, that endocrine derangements

may be concerned (e.g., in hypertension associated with the menopause, etc.) the search for such pressor agents has failed to elucidate the problem, proving almost barren of results. When kidney involvement is also present there are the further undecided possibilities of 3, defective elimination, and 4, the genesis of pressor agents by the damaged renal tissues.

Mosenthal (98) concluded that high or low protein diet does not increase or lower high blood pressures; similarly Newburgh (102) and Squier and Newburgh (118) found high protein feeding ineffective, though acting as a kidney irritant. On the other hand the observations of H. J. Starling (119), bearing on tuberculous cases, indicate a definite elevation of pressure under the continued influence of an abundant meat diet. An important point is raised by the finding of Foster (48) that a reduction of blood pressure under the influence of a continued low protein diet may take two months to develop; this suggests that some negative conclusions with high or low protein diets may possibly be due to periods of insufficient duration being studied. Orr and Innes (105) observed a decided lowering of pressure after the drinking of large quantities of water; they suggested a washing out of metabolites as a probable cause of this effect.

On the other hand Strouse and Kelman (121), examining cases of raised pressure associated with various degrees of renal damage, found that high protein diet caused no rise of blood pressure and that diminution of the protein intake in cases of definite nephritis, while lowering the non-protein N of the blood, did not lower the pressure. Sudden variations of systolic pressure sometimes amounting to 60 mm. were often seen, attributed to emotional causes acting directly on the vasomotor centre; these variations were not affected by alterations in protein intake.

Salt has been surmised to have some relation to high blood pressure and this hypothesis has influenced treatment, as in Allen's regimen with a salt intake cut down to 0.5 gram per diem. The recent work of O'Hare and Walker (103) lends no support to such a view. No relation was found to hold between the blood pressure and the chlorides of blood and plasma, and no effect on the systolic and diastolic levels was seen during wide variations in the amounts (0.5 to 4 grams) of salt taken in high pressure cases without nephritis. Further in subacute nephritis with edema and maximum salt retention comparatively low pressures were often recorded.

Cholesterin has also been suspected, especially by some French

(Chaffard and his school) and Russian investigators. Cantieri's (25) results oppose this idea; he found no relation, in acute or chronic nephritis, between the blood pressure and the cholesterin content of the blood, which in a series of arterio-sclerotic cases was rather below the normal content; also administration of cholesterin does not raise the blood pressure.

Dixon and Halliburton (37) ascertained that the pressor effect of cholesterin given by intravascular injection is negligible.

As regards urea, though high blood percentages of this substance and high blood pressure are often found together, the relation is very variable and it is evident that it is not a causal one. The same statement holds good with regard to the viscosity of the blood, though a group of high pressure cases associated with polycythemia has been recognised.

The search for pressor bodies of endocrine origin (though possibly present in toxemia in pregnancy, etc.) as a cause of persistent high blood pressure has so far proved futile, and the same is to be said with regard to the conceivable possibility of a lack of depressor substances as an operative influence. A similar remark applies to the question of retained pressor bodies when a rise of pressure follows reduction of the kidney tissue below a certain limit, e.g., to one-third, as studied by Passler and Heincke (106), Janeway (70) (with Carrel) and others. It is a remarkable fact that no adequate explanation is available as to how suppression of kidney function kills.

In the presence of structural kidney damage the question of altered function becomes added to that of diminution of functional area. Extracts of kidney have been found by various observers to have pressor effects—Tigerstedt and Bergman's (125) "renin"—and the throwing off of some such pressor agent from disintegrating renal tissue in diseased conditions has been suggested (for some cases of hypertension) by Batty Shaw (12); it has not been found practicable to establish the presence of such agents in the circulation. On the other hand there is the possibility that the whole condition (hyperpiesia) in which persistent high blood pressure is present may be due to toxic agents in the general circulation, secondarily affecting the kidneys and thus leading to an aggravation of the morbid effects.

The latest pressor substance suggested is guanidine, studied by Major and Stephenson (92). These observers observed powerful effects, doubling or tripling of arterial pressure in a few minutes, from intravenous or intramuscular injection of guanidine salts in dogs,—

effects opposed by $CaCl_2$, KCl or NH_4Cl given intravenously. In experimental uranium nephritis (dogs) a marked and persistent diminution in the excretion of guanidine bases was found. In a number of patients with high blood pressures—essential hypertension or with chronic nephritis—a decreased output of guanidine was observed, as compared with the normal daily average of 100 mgm. in normal persons and in patients with normal blood pressure and temperature. It is suggested that kidneys only slightly damaged, e.g., with small vessel sclerosis, might have difficulty in excreting guanidine while other substances might pass and the renal defect fail to be rendered evident by the usual tests till the change has progressed further. With regard to this question evidence is desirable as to the blood pressure in the condition of tetany (e.g., from parathyroid defect) where guanidine has been detected in the blood by Noel Paton (107) and his fellow workers. It would also be of interest to find whether any types of high blood pressure cases are favourably influenced by administration of parathyroid and calcium salts.

The normal relation of blood pressure to the body weight in the healthy state has been shown to be a definite one, as illustrated by the insurance statistics of Symonds and others and by the series of observations of Faber and others, increments of about 10 mm., etc., being found by Symonds in individuals of heavier build at all ages, while the young subjects of Faber showed differences of 6 mm. according to their build. While this appreciable difference holds good in healthy persons the effects of obesity are much more pronounced and have been emphasised by various observers. Among recent investigations, Aubertin's (2) 70 obese subjects (average age 60) showed high pressures in the great majority, only 7 being at or under 150 mm. while 24 non-obese controls of similar age averaged 149 mm. While greater degrees of obesity were associated with higher pressures, arterial sclerosis and chronic nephritis were not found to be the effective connection between obesity and high pressure. Apoplexy and sudden death are evidently related to the high pressures rather than the associations or effects of obesity acting in other ways. It is noteworthy however that Symonds states that fat elderly subjects in good condition and acceptable for insurance commonly have systolic pressures below 140 mm. on an average.

Relation of high pressures to the regulating mechanisms. Marey's Law. Under the circulatory conditions of normal life this law is one that is more honoured in the breach than in the observance. An in-

verse relation of heart rate and arterial pressure only occurs in certain conditions, such as are not usually present. It does not occur in the great majority of normal elevations of blood pressure, e.g., in muscular exercise with its raised pressure and quickened heart, nor in the similar conjunction seen in emotional excitement, nor in sleep where both pressure and heart rate are lowered, nor in some forms of circulatory depression accompanied by a slowed heart and a reduced blood pressure.

A more warrantable statement, much more limited in scope than the so-called law of inverse relation, is that when the blood pressure in the head is raised by an increase of the peripheral resistance in the circulation or by local causes acting on the head (hydrostatic factor, etc.), such pressure tends to increase the controlling power of the vagus centre, provided no other influence plays upon that centre in the direction of reducing its activity—as occurs during motor effort, emotional stress, etc. Conversely a lowered pressure in the head involves diminished activity of the vagus centre unless this is opposed, as may happen, by some concomitant influence tending to stimulate the centre.

It is evident that if persistent high blood pressure is due, as is commonly assumed, to excessive peripheral resistance there must be some agency in action which counteracts the working of Marey's law—since, as is well known, the heart is not slowed even in presence of exceedingly high arterial pressures. Thus in Mannaberg's (93) observations on 241 cases of high pressure, 55 per cent had normal pulse rates, while 43 per cent showed tachycardia and 3 per cent bradycardia; the tachycardias were chiefly in women and probably related to endocrine disturbances (thyroid, etc.). The mechanism of this is unknown. There is no evidence to show why the usual slowing influence of high pressure is not exercised—through direct influence on the vagus centre; and also reflexly through high pressure in the heart and distention of the aortic walls, if such a mechanism exists—as affirmed by Eyster and Hooker (41) for the normal animal, though this view is not supported by the recent work of Anrep and Starling (5) with cross-circulation experiments.

While it is known that high venous pressure acting on the right heart reduces vagus control and accelerates the heart, as Bainbridge found by increasing the volume of the blood, there is no ground for regarding this as a means of abrogating the slowing effect of an excessively high arterial pressure due to abnormally great peripheral

resistance. For when the latter is excessive, e.g., during compression of the aorta at the level of the diaphragm, the right heart (as well as the left) becomes largely distended and the venous pressure very high. But slowing of the cardiac rhythm, due to the arterial pressure, persists in spite of the elevated venous pressure; the arterial pressure dominates the situation, so far as the heart rate is concerned.

As regards the direct relation of blood pressure to the normal functioning of the vasomotor centre, Anrep and Starling have obtained important evidence by a method of cross-circulation. They caused the head of an animal to receive its whole blood supply from a heart-lung preparation while the body of the animal retained its normal blood supply from its own heart; this enabled them to study the direct effects of changes of blood pressure in the head on the medullary centres. They found that a rise of blood pressure in the head actively and almost immediately (after a latency measured in fractions of a second) depresses the activity of the vasomotor centre, causing a fall of blood pressure in the body generally. Changes of pressure in the head induce reverse changes in the body; these are not transitory but last for a long time, generally till the pressure in the brain again changes. Such reversed changes in head and body, first observed by Francois-Franck (49), have been studied by Hedon (60), Tournade, Chabrol and Marchand (127), Foa (47) and others. They have usually been attributed to changes in the heart action through the vagus centre, but such a mode of action is excluded in Anrep and Starling's experiments. It is obvious that a mechanism of this sort must militate strongly against the maintenance of an excessive pressure in the intact circulation.

In view of many facts it is clear that in persistent high pressure in man the condition is not simply one of increased vascular constriction, whether determined by undue activity of the vasomotor centre or by chemical agents acting directly on the walls of the vessels. Simple vascular constriction, raising the general pressure and the pressure in the head would bring into operation various normal regulating mechanisms such as —1, increased control of the heart through direct action of the pressure on the vagus centre together with 2, a direct synergetic inhibiting influence on the vasomotor centre; 3, a reflex depressing influence on the vasomotor centre through (vagus) depressor fibres arising in the aorta and heart, and possibly 4, an alteration of a pressor reflex influence ascending from the terminations of the vagi—a reflex advocated long ago by Pavlov and recently by McDowall.

It is evident that, whatever chemical agencies may be operative in other ways, in persistent high blood pressures there is a marked interference with regulating nervous mechanisms, rendering them ineffective in keeping down the pressure to anything like the normal levels.

The question of a compensatory influence of raised blood pressure. Allbutt regarded high pressure as an attempt of the organism to maintain the equilibrium of the circulation. May the rise of pressure be in some sense compensatory to drive more blood through a vital organ that needs it, e.g., heart muscle or brain or kidney? In the last named the high pressure might conceivably be related to the efforts of the kidneys to excrete concentrated urine, salts or waste products when in excess or when the renal mechanism is inadequate. Possibilities in this direction are suggested by the known existence of the sensitive mechanism by which a defective blood supply to the head promptly sets up a rise in aortic pressure through synergetic changes of increased activity of the vasomotor centre and diminished activity of the vagus centre. A compensatory reaction might conceivably develop in connection with other important organs where the blood supply may be defective from narrowing of arterial channels or diminution in the number of capillaries, or where functioning of the tissue—relatively defective from other causes—might be improved by a higher capillary pressure. A compensatory relation was suggested by Bier with reference to the kidney and later by others. The existence of a compensatory function may be investigated by artificially lowering the pressures (by vaso-dilators, etc.) in order to find whether functional impairment or disturbances, renal, cardiac, or respiratory result from a reduction of the pressure from an elevated level, which, under the conditions present in these cases, had been favourable to efficiency. A recent investigation on such lines by C. Reid (111) does not lend support to the idea of a compensatory relationship as regards renal efficiency, tested by modern methods, blood urea and non-protein nitrogen being estimated and MacLean's urea concentration test, etc. being employed; the raised pressures present, associated with a variety of kidney conditions, were lowered by nitrites, venesection, etc.

As regards the effects of high pressures in causing elongation and tortuosity of arteries, it is obvious that such may result from more than one cause. 1. Impairment of the power of the arterial wall to resist distention may do this, even in the absence of abnormally and persistently high pressures, from the frequent or continued existence of a relaxed condition of the arterial muscle, especially in arteries with

poor support like the temporal, where elongation and tortuosity may develop in an apparently healthy vessel. The present writer (89) has shown that the elongation of an artery by internal pressure is enormously great in a relaxed as compared with a tonically contracted; further, as the process of elongation needs time to develop, the continuous (diastolic) pressure is more effective than the transient systolic rises. Abnormal conditions of the arterial wall may of course diminish its resistance to distention.

2. Apart from such impairment of resistance persistently high pressures tend to elongate the artery and to loosen or pull it away from its normal attachments along its normally straight course, as easily recognised in the case of the brachial, especially in a thin arm, where the vessel is felt as a tube running an elongated and devious course in bold curves down the arm—especially prominent a little above the elbow. The absence of such conditions in the presence of high arterial pressure of unknown duration affords presumptive evidence that the high pressure is not of long standing.

It may be taken as established that high blood pressure readings, when carefully taken, represent approximately correct measurements of the actual intra-arterial pressures as a rule. It is only in a small minority of abnormal cases of thickened arteries with excessive tonic contraction, etc., that serious discrepancy may occur, sclerotic conditions without muscular contraction having no important influence. Digital compression for 3 or 4 minutes or massage of the artery are useful in removing abnormal resistance and have the advantage of not causing congestion of the limb which may arise from repeated compressions by the armlet—with very disturbing results, especially in some susceptible cases, giving erroneous auscultatory indications or actual changes of arterial pressure, etc.

The pronounced effects of mental stress, excitement and worry in producing and maintaining high blood pressure emphasise the significance of the nervous system whether exercised directly through cardiovascular innervation or more indirectly through endocrine or metabolic alterations. The frequent variation of the pressure from day to day or even at shorter periods opposes the idea of structural causation involving increased peripheral resistance and tells against the presence of permanent chemical agencies acting on the vessels directly. The constancy of a lowered pressure during sleep reported in some high pressure cases points in the same direction (Muller, Blume, Katsch and Pansdorf).

Again the strikingly exaggerated pressure changes which may rapidly occur in response to nervous disturbances, emotional causes, etc., as noted by numerous observers in many cases of high blood pressure, bear testimony to the presence of disturbed innervation involving defective regulation as an important factor in the condition. Thus causes of slight elevations of pressure in the normal state may have abnormally great effects in causing rapid and extensive variations in many subjects of high pressure.

Low blood pressure. The mechanism of the acute condition of excessively low pressures seen in circulatory shock, etc.—due to the altered capacity factor dependent on capillary relaxation and later on diminished blood volume—has been elucidated by various investigations, especially by the work of Cannon (24) on traumatic shock and that of Dale with Laidlaw (32) and Richards (33) on the action of the histamine and histamine-like bodies. Similarly the pressure falls in acute infections like cholera, etc., are rendered intelligible. But in persistent low pressures attendant on exhausting diseases or occurring without obvious cause (essential hypotension) the available data are, as in the case of persistent high pressure, inadequate for a satisfactory explanation of the mechanism involved—whether a defective peripheral resistance or defective cardiac output depending, apart from cardiac enfeeblement, or lessened return of blood to the heart as a result of undue expansion of the capacity of the vascular system from capillary or venous relaxation, contraction of venules, diminished volume of blood in circulation, etc. It is also unknown how far such conditions are mediated through the nervous system and how far due to the direct influence of chemical agents—depressor bodies, lack of pressor substances, etc.

As to what constitutes "low pressure" the level below which a pressure is to be regarded as low or abnormal is not sharply defined and no doubt varies considerably, as in the case of high pressure, for the individual and the conditions present. Roughly anything decidedly below 100 systolic or 60 diastolic may be suspected of being "subnormal." Some athletes in good training have such pressures as systolic 105 and diastolic 65.

Subnormal blood pressures naturally exercise a generally depressing influence on the active tissues and tend to establish a vicious circle. There is significance in the observation of Markwalder and Starling (95) that for the mammalian heart (in the heart-lung preparation) an average (innominate artery) pressure of at least 90 mm. Hg is neces-

sary for the due vigour of the cardiac muscle; otherwise the coronary circulation is apt to be insufficient. *Excessively* low pressure injures the nutrition of the heart, favouring enlargement, etc. Muhlberg (99) is quoted by Friedlander to the effect that low pressure after the age of 50, unassociated with any organic lesion to account for it, constitutes the best criterion of life beyond the normal expectancy; it is also stated by Friedlander (50) from Fisher's figures that of 3,389 persons (ages 16 to 60) with systolic pressures of 100 mm. or less there was only 35 per cent of the expected mortality. Symonds (122) considers low pressures after the age of 40 desirable; he also reports that the lowest mortality was found in those subjects who were 15 per cent below the average weight. Pressures not *too* low seem to favour longevity.

The general relation of low blood pressure to tuberculosis has been the subject of several inquiries in recent years. Marfan and Vannieuwenhuyse (94) (700 cases) while finding systolic pressure lowered—more so as the disease was serious or getting worse—do not regard low pressure as excluding improvement or recovery. Normal or raised systolic pressure they regard as a good prognostic. Diastolic pressure falls only in the last stage. These workers emphasise the importance of repeated examinations. De Bloeme (36) in 500 cases by the auscultatory method affirmed the existence of an important group at 100 to 110 mm. S. where the seriousness of the condition was more recognisable by the blood pressure than by other methods, while cases at 80 to 100 mm. were recognisable by ordinary diagnostic means. There seemed to be a general relation between the higher pressures and better conditions of the patients. The most favourable cases of both sexes were between 110 and 150 mm. There was apparently an association between low pressures and the tendency to relapse or the occurrence of relapse, even after the local and general symptoms had subsided; blood pressure rose with improvement.

While regarding blood pressure as being below normal in 60 per cent of early cases, Naucler (101) sometimes found normal or higher pressures in early cases and concluded that low pressure is not a reliable sign of early phthisis. The low pressures seemed to depend more on the severity than on the extent of the disease.

R. J. Cyriac (31) recorded differences in the systolic pressure readings in the two arms in a number of tuberculous cases as E. F. Cyriac (30) did in association with some traumatic conditions.

That excessive smoking can lower blood pressure has long been known and comes into question when systolic pressures at or below 100 mm. are observed.

Pressures in aortic regurgitation. Since the remarkable arm-leg systolic pressure difference was recognised in cases of aortic regurgitation by Hill, Flack and Holtzman (64) in 1909, numerous observations have been made and differences of varying degrees of magnitude have been recorded, one of 200 mm. by Rolleston (114)—leg pressure 350, arm 150—while the differences have as a rule been much smaller. Similar phenomena have been observed in some other conditions—violent muscular exertion in healthy persons, some cases with arterial sclerosis, and in exophthalmic goitre. In these conditions question naturally arises as to which reading represents the "blood-pressure." The mechanism involved has proved difficult of elucidation. L. Hill suggested a different "conductance" in the leg arteries, transmitting the large systolic wave more effectively than in the arm. As the diastolic pressure is virtually if not absolutely similar in arm and leg it is evident that the systolic difference is a phenomenon of wave motion.

There are indications that both *a*, cardiac and *b*, vascular conditions are usually concerned in the mechanism of the arm-leg difference in pressure. That a cardiac factor plays a part is suggested by the clinical evidence to the effect that in man the differential pressure is slight or absent in recent aortic lesions, and is chiefly found in cases of compensated aortic regurgitation with their enlarged heart, large and powerful systolic wave and unusually extensive pulse pressure—conditions also present in greater or less degree in other instances where the arm-leg difference has also been recorded, e.g., exophthalmic goitre, some arterio-sclerotic cases, violent muscular exertion, etc.

In toxic exophthalmic goitre many of the circulatory conditions resembling those associated with aortic regurgitation (large heart, exaggerated pulse pressure, etc.) may be strikingly present. Taussig (123) has observed an arm-leg difference of 37 mm. Hg; the condition has also been described by Harris (59). In a case of arterio-venous aneurysm Lewis and Drury (83) have found that many similar circulatory features were temporarily abolished during artificial closure of the arterio-venous communication, but not the differential pressure, which they attribute to vascular conditions which had become established.

As regards experimental animals there is a conflict of evidence between the results of Bazett who described the immediate appearance of a differential pressure after an aortic valve lesion, and those of Leschke (84) who did not find a differential pressure at this stage.

The important investigation recently published by Bazett (13)

deals with schema results, animal experiments and clinical observations. He concludes that the differential pressure is essentially due to the transference of kinetic energy in a fluid in rapid motion into stress when the flow meets resistance, the relative degrees of slowing thus induced in different vessels and the relative masses of blood concerned being important, while the condition of the arterial wall (suggested by L. Hill) probably plays a part as may also the "breaker formation" of Bramwell and A. V. Hill (20), though not essential. The higher leg pressures are accounted for on these lines, local arterio-sclerotic changes being capable of exaggerating the phenomenon. Larger differences in arm-leg pressure were found with contracted arterioles—involving greater slowing and greater transformation of kinetic energy. It is suggested that the effects obtained by L. Hill and Rowland (65) with warm baths (equalisation of pressures) are explicable in this way as well as on the hypothesis of altered conditions in the arterial walls. The aortic arm-leg difference is thus regarded as a great exaggeration of the normal carotid-femoral difference described by various observers —essentially the water hammer action of Corrigan's "rushing current" in aortic regurgitation.

In animal experiments a reversed differential pressure was sometimes seen, i.e., a pressure higher in the upper than in the lower limb, in association with a forcible heart action with regurgitation present and an apparently low peripheral resistance. No evidence seems to be available of the existence of such a condition clinically.

Capillary pressure. In the attempts to gauge the capillary pressure the various methods and the different criteria applied have produced a discordant and somewhat bewildering assortment of results representing "capillary pressure" as anything between 25 to 50 mm. H_2O and 70 mm. Hg. There are at least four modes of observation that have been used.

1. Blanching methods (skin of finger or hand). Following Von Kries' (75) idea there have been applications by numerous observers, using however different criteria, e.g., the first production of visible paling, used by Basler (11) and by White (132) or complete blanching (v. Basch (10)) or the pressure at which the skin again begins to flush (Recklinghausen (109) and others); much confusion has resulted. Von Basch's figures were 25 to 30 mm. in healthy subjects; he concluded that capillary pressure can vary independently of arterial pressure; Recklinghausen's value was 52.2 mm. Basler with his ochrometer found normal capillary pressures at about 7 mm. Hg, but Landerer

(79) by the same method reported pressures of 17 to 25 mm. Briscoe (21) using Hooker's capsule to cause paling estimated the normal pressure at 23.5 cm. H_2O. Hill and McQueen (66) taking the returning flush as their criterion obtained values of about 10 mm. Hg.

The interpretations put upon the results of blanching experiments naturally depend on the different views held as to the causation of the colour of the skin. There is thus much diversity of opinion as to what is really being measured. While v. Kries, v. Basch, v. Recklinghausen and Basler took the paling of the skin to be due to compression of the capillaries, Lombard (85) and Danzer and Hooker (35) regard the emptying of one or more of the venous plexuses in the dermis as the main cause; they find that paling is not necessarily accompanied by cessation of flow in the capillaries. White (testing the influence of heat, etc.) also concludes that the paling of the skin from external pressure is not an indication of capillary pressure. He obtained values of 4 to 19.5 cm. H_2O. Hill interprets the 10 mm. value (obtained as stated) as indicating arteriolar, not capillary, pressure, *plus* the resistance of the epidermis which has to be deducted. When a capillary area is compressed the internal pressure is regarded as banking up to arteriolar pressure. Such an arteriolar pressure value as 10 mm. Hg is about half the amount reported for capillary pressures by many other observers. The real capillary pressure Hill (62) estimates at something like 20 to 50 mm. H_2O. Observing transparent parts of mammals and frogs by Roy and Graham Brown's (115) method he found such a compressing pressure to cause momentary checking of the blood flow and took this as the true index—as distinguished from a compression stopping the flow, which might require 350 mm. H_2O.

2. Pressure required to cause obliteration of capillaries under the microscope. Lombard (85), who introduced the use of a drop of oil on the skin to permit of direct examination of the capillaries, found very different pressures necessary to obliterate the vessels of different orders. Stated in millimeters H_2O, the following values were obtained —subcapillary venous plexus, 135 to 205; superficial venous branches 205 to 270, most compressible capillaries, 245 to 300, middle-size capillaries, 475 to 545, most resistant capillaries and arterioles, 815 to 950. Methods founded on Lombard's plan have been used by Krauss (74), Basler (11) (capillary tonometer) and Kylin (78). The last named recorded normal pressures of 8.5 to 14 mm. Hg and abnormal ones up to 40 or 50 mm. in glomerulo-nephritis and scarlet fever. Difficulty arises from the unequal compressibility of different capillaries in the field of observation.

3. Pressure required to cause stasis of corpuscular flow under the microscope—by Danzer and Hooker's (35) micro-capillary tonometer, a different criterion from those used in the preceding methods. They found values of 18 to 26.5 mm. Hg averaging 22 mm. Boas and Frant (17) using the same method reported normal capillary pressures at 18 to 22 mm. Hg, rarely above 30 mm.; high pressure cases reading usually between 30 to 60 mm. In essential hypertonus capillary pressure was found to be normal, i.e., below 30 mm. Boas and Mufson (18) found a much higher mortality in a high capillary pressure group (5 deaths in 28). Their post-mortem findings (a small number of cases) did not support Kylin's hypothesis of an association of high capillary pressure with glomerular nephritis.

4. Piercing capillaries with a very fine capillary glass needle (containing saline at a measured pressure) under the microscope to measure the pressure in a capillary loop—by Carrier and Rehberg (26). The values 45 to 75 mm. H_2O in two subjects at 7 cm. below the clavicle—reported by this method—unfortunately unsuited for clinical application—are relatively low and lend support to L. Hill's repeatedly stated view as to the lowness of capillary pressure. The venous pressure was parallel to the capillary pressure.

Supposing that the pressure in the minute vessels of the skin can be accurately measured, there remains the question of the application of such results to the conditions of the general circulation. Pressures in the finger and hand are naturally influenced profoundly by the local conditions of arterial tone as affected by vasomotor influences, heat, cold, exercise, sleep, etc., with the result that the digital pressure may rise while the brachial pressure falls, or vice versa. It is hardly necessary to recall the frequently opposed conditions in the skin and splanchnic areas, as with muscular exercise, asphyxia, adrenalin, etc., also the different incidence of the vaso-dilator effects of acetyl-choline—as described by Reid Hunt (69)—marked in the skin, slight in the liver and intestine, very slight in the voluntary muscles. Again the strong constriction of the renal vessels, which is presumably the cause of the anuria known to occur during short spells of violent muscular exercise, is associated with increased pressure in the skin. The habitually cold or habitually warm hands of different persons, naturally involve wide variations in the pressure relations of the minute cutaneous vessels.

There are thus no means of ascertaining the relations between pressure measurements in the skin and the pressures existing in other parts, internal organs, etc., and how they stand with regard to the

average pressure in the capillary field as a whole, made up as it is of a great and varying distribution of capillary pressure values in multitudinous districts. It is obviously not permissible to speak of capillary pressure in the same sense as arterial pressure, the latter being a definite measurement virtually the same in all the large arteries throughout the body, while capillary pressures vary widely and in different senses in numerous districts under physiological conditions. Still if there is definite association of high readings of pressure in the minute skin vessels in some category of high blood-pressure cases and not in others this—even if not representative of the capillary system as a whole—is obviously a matter of much interest calling for further investigation as to its mechanism and significance. Boas and Mufson report close correspondence in the capillary readings from the same individuals taken many months apart—with some exceptions for which explanations are offered. As to the relations of capillary and venous pressures there is some conflicting evidence. While it has generally been accepted that capillary pressures run much more nearly parallel with venous than with arterial pressures, Boas and Doonieff (16) in a recent investigation (using a needle in a vein connected with a manometer) find that a rise in venous pressure up to 39 cm. H_2O may have no effect on capillary pressure—evidence that the high capillary pressure which may occur in hypertension is not accounted for by high venous pressure. On the other hand, Danzer and Hooker found venous compression to cause increased capillary pressure; Carrier and Rehberg observed a parallelism between venous and capillary pressures. Von Basch and Kraus had formerly emphasised the close relationship between these pressures.

The peripheral resistance. Recent work on the capillary system and the very varied conditions that may obtain in it have re-opened the question as to what the peripheral resistance in the circulation is constituted by. Is the arterial resistance largely supplemented by resistance in the capillary field and possibly also, as suggested by Hooker's (68) work, in the venules, and capable of being altered in an important degree by variations in these as well as in the small arteries?

Favouring the commonly accepted view that the chief resistance is in the small arteries is the greater internal friction depending on the relatively rapid rate of flow in the arteries as compared with the slow flow in the capillaries under ordinary conditions; also such evidence as is available to show that the loss of pressure in passing through the capillaries is relatively small, the great fall from the arterial pressures

ordinarily measured having occurred before the capillary region with its apparently low pressures is reached.

On the other hand there is to be considered the active and strong contractility of capillaries as shown by Krogh (76) and others; Lewis (82) estimated their contractile power as being capable of expelling fluid against a pressure of 50 to 60 mm. Hg, and when contracted, of resisting the entry of fluid up to 90 to 100 mm. Hg. Excessive constriction or closing of an unusually large proportion of the capillary tubes, with diminution of the sectional area of the available capillary bed, must necessarily affect the resistance offered to the outflow from the arterial tree, as well as influencing the capacity of the vascular system, especially in view of the fact that in small capillaries the red corpuscles actually rub against the walls of the tube; it is not simply a matter of internal friction between the layers of the moving blood as in the arteries.

Conditions that might reduce the disparity between the sectional area of the arterial and capillary fields (e.g., closure of many capillaries, arterial dilatation etc.) would naturally tend to enhance in some measure the resistance presented in the capillaries. Again, Lombard estimated the fall of pressure between the small arteries and the veins at 40 to 50 mm. Hg which would postulate a resistance in the capillaries nearly as great as that in the small arteries. But the recent evidence favours low values of capillary pressure, involving a great fall from the pressure in the larger arteries (e.g., 120 mm.) before the capillary field is reached with pressures estimated at one-sixth or one-tenth or even much less—indicating the situation of the main resistance in the circulatory system as being in the small arteries and arterioles.

On the other hand if higher estimates of capillary pressure are correct, especially such as have been reported in some diseased conditions, with a large decline from capillary to venous pressure it is evident that a considerable part of the peripheral resistance must be located in the region of the capillaries and venules. With regard to the possible influence of constriction of the venules, such might obviously have important effects.

Venous pressure. Venous pressures, easily measured in the veins of the arm or hand by the method of Hooker (67), have been found by that observer to be usually between 10 and 20 mm. H_2O, progressively increasing with age from 8 cm. in early youth to 25 cm. in old age. Eyster and Middleton (42) report pressures rarely above 11 cm. normally; Briscoe about the same, 11.4 cm. White (132), using a

method of instantaneous instead of gradual application of external pressure to the vein, recorded lower values, often 4 to 6 cm., sometimes as high as 12.5 cm. Venous pressure has been noted as being raised in nephritic hypertension, in contrast to simple arterio-sclerosis, by Villaret (128) and his associates. But this conclusion is opposed by the results of Leconte and Yacoël (80). Like the pressures in the minute vessels of the skin the venous pressures in a limb are liable to be much influenced by local conditions. And it gives no actual measure of the venous pressure at which the filling of the right heart takes place—the "effective pressure" of Yandell Henderson and Barringer (61)—the difference between the intra-auricular and intra-thoracic pressures. As the latter pressure is variable, this, as Wiggers (133) points out, raises a serious difficulty as regards the application of limb venous pressure measurements to the study of circulatory conditions.

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DIASTATIC ACTIVITY IN BLOOD AND URINE.

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As views on the utility of the estimation of diastase in urine and blood are divergent, an investigation into the diastase activity of equal quantities of whole blood and urine was undertaken. The diastatic activity of a specimen of urine is estimated in terms of the amount of starch which, incubated at 37° C. with a definite volume of the urine, will be changed in 30 minutes, the disappearance of the starch being indicated by the failure of the mixture of starch and urine to give a blue colour or a violet tint with iodine. For the estimation of the diastatic activity of blood, it is necessary to estimate the amount of sugar present in a given amount of blood, to incubate a given amount of blood with a given amount of starch at 37° C. for 30 minutes, and to estimate the amount of reducing sugar which has been formed by the diastase in the blood. The diastatic activity of the blood is given in terms of reducing sugar, and in this way a comparison can be made between the diastatic activities of equal volumes of blood and urine, although the actual concentration of the diastase by the kidneys would not be available.

Technique.

Urine.—In order to obtain the diastatic activity of equal volumes of blood and urine, a slight modification of the method described by Dodds (1922) was used.

A series of ten small test-tubes was employed usually, the length of the tube being about 3 inches, and capacity 4 c.c. A quantity of buffered urine was prepared by mixing 5 c.c. of the urine with 20 c.c. of a mixture of Sörensen's solutions. The buffer solution was obtained by mixing 15 c.c. of a solution containing 11·876 grm. $Na_2HPO_42H_20$ in 1000 c.c. distilled water, and 85 c.c. of a solution containing 9·078 grm. of KH_2PO_4 in 1000 c.c. distilled water. These solutions were kept in paraffin-coated glass-stoppered bottles. To each of the small ten test-tubes was added 1 c.c. of the buffered urine, and to the series of ten tubes (1–10) were added respectively 2 c.c., 1·8 c.c., 1·6 c.c., 1·4 c.c., 1·2 c.c., 1·0 c.c., 0·8 c.c., 0·6 c.c., 0·4 c.c., 0·2 c.c. of a 0·2 per cent. solution of Lintner's soluble starch made up in 0·9 per cent. NaCl solution. The total volume in each tube was made up to 3 c.c. by the addition of distilled water. The tubes were shaken immediately, and placed in a water-bath in an incubator for 30 minutes at 37° C. The tubes were then removed

from the incubator, and their contents poured into a series of larger test-tubes about three parts full of cold tap-water. One or two drops of a N/10 solution of iodine were added to the series of tubes (1–10) until a tube was obtained where the blue or violet tint was not perceptible. The amount of diastase in this tube was sufficient to digest all the starch present. The diastatic activity of the urine was found empirically by dividing the number of c.c. of the starch solution digested by the amount of urine in c.c., the number obtained often being called Wohlgemuth units. With the above technique the range of diastatic activities would be 20, 18, 16, 14, 12, 10, 8, 6, 4, 2—an even series of numbers not given by simple pipettings in previous methods. The range of diastatic activities could be easily increased by the use of stronger solutions of starch. For example, the use of a 0·4 per cent. solution of starch would give a range of diastatic activities from 40, 38, 36, . . . 24, 22, but the necessity for this was not common when the specimen of urine for examination was passed in the second two-hourly interval after the first meal of the day. This period was chosen because it appeared to be a convenient time to examine both the blood and urine, and because most of the diuresis due to the intake of fluid with the meal had ceased.

It was shown by Michaelis and Peckstein (1914) that the pH at which diastase was most active varied with the salt content of the medium. Stafford and Addis (1924) pointed out that in the technique of Dodds (*loc. cit.*) and Sladden (1922) there seemed to be a danger of a disturbing variation in the chloride and phosphate concentration respectively. This difficulty has been got over by the above modification, in which there was always in each tube a sufficiency of chloride and phosphate, and in which the urine with the enzyme was diluted equally in all the tubes, although the concentration of the substrate varied. This, however, would not prevent a reasonably accurate study of the diastatic activity of different urines.

In the present investigation the above modification was adopted, and was used before the publication of the second and third papers referred to in the preceding paragraph. Sladden (*loc. cit.*) considered that the addition of phosphate buffer solutions tended to obscure the final readings. But in the present inquiry no difficulty in reading the end-point was observed if small tubes (4 c.c.) were used for incubation, and if these were emptied into larger tubes (20–30 c.c.) containing cold water before the addition of iodine.

Blood.—For the examination of the diastase in the blood, the method described by G. Matthew Fyfe (1923) was employed with one or two small modifications.

Into one of two 100 c.c. Erlenmeyer flasks, 1·8 c.c. of Sörensen's buffer solutions and 1 c.c. of 0·1 per cent. solution of Lintner's soluble starch in 0·9 per cent. NaCl are pipetted, and into the other 23·8 c.c. of a 15 per cent. solution of sodium sulphate acidified by the addition of glacial acetic acid to the extent of 0·1 per cent. Into each flask is introduced 0·2 c.c. blood by means of two special pipettes which are thoroughly rinsed out in the clear fluid. The flask containing starch is placed in a water-bath in an incubator for 30 minutes at 37° C. At the end of this period 21 c.c. of the acid sodium sulphate solution are added immediately. The amount of sugar is estimated in both flasks by MacLean's method (1919).

The amount of sugar formed by 0·2 c.c. blood from 1 c.c. of a 0·1 per cent. solution of starch expressed in milligrammes is taken as an index of the diastatic activity of the blood. The number obtained multiplied by 100 to displace the decimal point is used as the number indicating the diastatic activity of the blood.

VARIATIONS IN THE TWENTY-FOUR-HOURLY SPECIMENS OF URINE.

With regard to 24-hourly specimens of urine, considerable variations were found from day to day under apparently constant conditions, the variations tending to be greater in the case of those individuals whose urine had a high diastatic activity.

Table I gives the variations (in Wohlgemuth units) obtained in a number of healthy individuals, specimens of the total urine passed in the 24 hours being examined.

TABLE I.

J. R—	5·4, 4·5, 6, 6, 3, 3·5, 3·8.
D. M—	9·1, 6, 10, 10, 10, 9.
C. G—	7·5, 9·7, 11, 8.
G. M—	20·8, 16·3, 16, 12·5, 15·4, 11·8, 13·2, 13·6, 10.
F. M—	7·2, 8·8, 6·5, 5·2, 5·7, 7·2, 5·8, 7·0, 5·5.
R—	19, 19, 20, 18·2, 15·4, 22·2, 22, 12·5, 22, 21, 16·7, 18, 12·5, 19, 20, 12·8.

The diastatic activity was not always directly related to the specific gravity or inversely to the average hourly rate of the secretion of urine, although it was found that the 24-hourly specimens with a higher specific gravity tended to have a higher diastatic value, while specimens with a lower rate of secretion tended to have a higher diastatic activity.

The total diastatic activity for the 24 hours was taken as the amount of urine in c.c. multiplied by the diastatic index stated in Wohlgemuth's units. This was found to give widely different numbers in different individuals. But by taking a large number of observations on the same individual over a considerable period, it was found that the total diastatic activity gave, in the majority of cases, a number which was fairly constant. For example:

In 9 readings on one individual, 6 gave a total diastatic content between 26,000–30,000.

In 9 readings on a second individual, 6 gave a total diastatic content between 12,000–16,000.

In 10 readings on a third individual, 8 gave a total diastatic content between 17,000–19,500.

In 15 readings on a fourth individual, 12 gave a total diastatic content between 13,000–16,000.

The remaining results of the individual cases were either above or below the respective numbers, the highest number obtained being as much as 50 per cent. above the lowest in the same case.

With regard to the individual specimens passed throughout the 24 hours,

considerable variations in the diastatic activity stated in Wohlgemuth units were met with. For example, variations 5–18, 9–22, 2–10, 2–6, 4–8, 12–24, etc., were obtained in different individuals.

The fewer the specimens, the less were the variations of the diastatic activity of the different specimens.

Conditions of polyuria induced by exitement, cold, drinking large quantities of fluids gave more striking variations. In one case it was noted that during excitement the diastatic activity of the urine fell from 20 to 1 in the course of two or three hours.

Stocks (1915–16) stated that it appeared that the concentration of diastase was at a maximum just after breakfast (8 a.m.), and then decreased gradually with a secondary rise after dinner.

Table II gives some observations made by the present writer on hospital patients who had breakfast at 7 a.m., dinner at 12 noon, tea at 4 p.m., supper at 7 p.m.

TABLE II.

	8.30 a.m.	11 a.m.	1.30 p.m.	5 p.m.	8 p.m.	5 a.m.
F. M—	8	5	5	14	5	12·5
G. M—	16·6	25	10	25	20	16·6
C. G—(1)	8	5	8	12·5	12·5	12·5
,, (2)	12·5	8	10	10	12·5	14·3
D. M—	8	6	—	10	10	14·3

The highest concentration of diastase in hospital patients who were not suffering from renal disease, and who were confined to bed, was obtained in the specimen of urine passed at 5 p.m. or in the overnight specimen passed at 5 a.m.

The observations in Table III were made on healthy young adults whose ordinary daily routine was not altered in any way.

TABLE III.—*Breakfast 8 a.m.*

Subject A.		Subject B.	
Diastatic activity of urine. (Wohlgemuth units.)	Time.	Diastatic activity of urine. (Wohlgemuth units.)	Time.
16·7	8.45 a.m.	14·3	8.45 a.m.
22·2	10.5 ,,	22·2	9.45 ,,
20	11.40 ,,	20	1.25 p.m.
18·2	1.10 p.m.	Meal.	
Meal.		15·4	3.45 ,,
16·7	4.40 ,,	12·5	4.30 ,,
20	6.15 ,,	20	5.40 ,,
Meal.		Meal.	
14·2	8.0 ,,	18·2	10.30 ,,
15·4	9.5 ,,	25	8.0 a.m.
18·2	12 midnight		
22·2	8.20 a.m.		

In neither of the above observations did the urine which was passed immediately after breakfast have the highest concentration of diastase. It would appear that the urine, which is secreted overnight, and which in healthy individuals is secreted at the slowest rate, has the highest diastatic activity, provided that factors producing polyuria were excluded. Specimens of urine were obtained before and immediately after breakfast, the first specimen being passed about 8 a.m. and the second about 8.45 a.m. to 9 a.m. The first specimen included the urine secreted overnight (Table IV).

TABLE IV.

Urine passed at 8 a.m. before breakfast.		Urine passed at 8.45 a.m. after breakfast.	
Diastatic activity.	Urine per hour in c.c.	Diastatic activity.	Urine per hour in c.c.
18·2	22	16·7	25
22·2	17	16·7	40
25	17	14·3	27
25	18	16·7	27
22·2	24	16·7	35
25	23	20	27
22·2	21	18·2	30

The lower diastatic activity of the second specimen evidently depends largely on the increased rate of secretion as compared with the relatively slow rate of secretion of night urine. On the other hand, it was found occasionally that the overnight urine had not the highest diastatic activity of specimens passed during the 24 hours' period. This can be seen on reference to Tables II and III.

The diastatic activity of the individual specimens passed during the 24 hours varied inversely as a rule with the average hourly rate of secretion. Exceptions, however, were rather frequent.

Table V gives the type of results obtained on a day during which no food or fluid was taken from 8 a.m. until evening.

TABLE V.

Time.	Amount in c.c.	Time after food in hours.	Diastatic activity. (Wohlgemuth units.)	Rate of secretion per hour in c.c.	Average hourly total diastatic activity.
7.30 a.m.	330	—	16	33	528
7.45 „	18	—	20	36	720
8.0 „	(breakfast)				
10.30 „	160	2½	16	58	932
11.35 „	102	3½	12	94	1132
1.5 p.m.	131	5	10	87	883
2.10 „	73	6¼	10	67	674
4.40 „	83	8¾	16	33	532
5.20 „	18	9¼	18	27	486

VARIATIONS IN DIASTATIC ACTIVITY OF BLOOD.

Wohlgemuth's method for diastase in the urine would appear not to be sufficiently delicate for serum, owing possibly to the concentration of diastase in the urine being higher than in the serum. The tint of the sera masks the delicacy of the colour reaction which occurs on the addition of iodine. Variations were found in the diastatic activity of whole blood when examined by Fyfe's method, and they would appear to be related to the ingestion of food.

Table VI shows the the variations obtained in two normal cases selected at random from a number of estimations made on several normal cases.

TABLE VI.

Case A.		Case B.	
Time.	Blood diastase.	Time.	Blood diastase.
7.30 a.m.	10·2	8 a.m.	6
Meal at 8 a.m.		Meal at 8.15 a.m.	
10.15 a.m.	11·9	9.15 a.m.	5
11.30 ,,	6·8	10.45 ,,	2·9
1.30 p.m.	6·3	1.0 p.m.	6·5
4.30 ,,	9·9	3.0 ,,	6·4
		5.0 ,,	5

Stafford and Addis (*loc. cit.*) foresaw the possibilities of variations in the diastatic activity of plasma, but they gave no details. Cammidge and Howard (1923) noted variations in the diastase content of the blood of a rabbit.

DIASTATIC ACTIVITIES OF URINE AND BLOOD, AND THE DIASTATIC CONCENTRATION FACTOR.

The diastatic activity of a specimen of urine has been shown to be—

$$\frac{\text{Number of c.c. of 0·2 per cent. solution of starch converted by 0·2 c.c. urine}}{\text{0·2 c.c. urine}} = \frac{\text{number of milligrammes of starch converted}}{\text{0·2 c.c. urine}}$$

$$i.\,e.\ \text{U.D.} = \frac{\text{number of milligrammes of starch}}{\text{0·2.}}$$

The diastatic activity of the blood (B.D.) has been taken as the number of milligrammes of sugar × 100 formed by the diastase in 0·2 c.c. blood from the substrate, viz. starch.

The diastatic concentration factor (D.C.F.) for the kidneys which are being examined may be taken as the power of the kidneys to concentrate diastase from the blood. If, for example, a specimen of urine is obtained for the second two-hourly period after the first meal of the day, in order to get the mean value of the blood diastase during this period, it would be necessary to carry out blood-diastase estimations at the beginning and end of this period or

alternatively at the middle of this period. As it is impossible to estimate actually the amount of diastase in the urine and blood, the ratio of the amount of starch converted by 0·2 c.c. urine to the amount of sugar formed by 0·2 c.c. blood has been taken as the diastatic concentration factor.

$$\text{D.C.F.} = \frac{\text{number of milligrammes of starch converted by 0·2 c.c. urine}}{\text{number of milligrammes of sugar formed by 0·2 c.c. blood}}$$

$$= \frac{\text{U.D.} \times 0{\cdot}2}{\frac{\text{B.D.}}{100}}$$

$$= \frac{\text{U.D.} \times 0{\cdot}2}{\text{B.D.}} \times 100.$$

The diastatic concentration factor was found to vary considerably throughout the 24 hours. The results in Table VII from two subjects—typical of a series of at least half-a-dozen—show the sort of variations obtained in a number of individuals.

TABLE VII.—*Breakfast* 8 *a.m.*

	Time.	B.D.	U.D.	D.C.F.
Case A:	7.30 a.m.	10·2	16	30 +
	10.15 „	11·9	16	29
	11.30 „	6·8	12	25
	1.30 p.m.	6·3	10	30
	4.30 „	9·9	16	40
Case B:	8.0 a.m.	6	6	20 approximately.
	9.15 „	5	—	—
	10.45 „	2·9	8	36
	1.0 p.m.	6·5	6	25
	3.0 „	6·4	8	25
	5.0 „	5	8	28

From the point of view of convenience it was decided to carry out a number of investigations on healthy adults, and to examine the diastatic activity of the blood at two hours and four hours or alternatively at three hours after the first meal of the day, and to examine the diastatic activity of the urine secreted during the second two-hourly period after the same meal.

Table VIII gives the diastatic concentration figures obtained in young adults and in children of fifteen and under by the above method.

A factor which must be considered in cases giving low urinary diastatic figures, apart from those due to polyuria, is that of the blood-diastase figure.

Case 6 has a low U.D. and moderately low B.D., but the lowness of the D.C.F. is due to the presence of a certain amount of diuresis due to excitement. Case 14, on the other hand, shows low figures for the U.D. and B.D., but the D.C.F. in the absence of polyuria is within normal limits.

It will be noted that if due allowance is made for variations due to polyuria, the figure obtained from these normal individuals examined for the diastatic concentration factor lies between 15 and 40.

TABLE VIII.

	Sex.	Age.	U.D.	B.D.	D.C.F.	Amount of urine during two hours.	
1.	M.	31	16	9·4	29	110 c.c.	
2.	M.	25	12	8·1	29	65 „	
3.	M.	25	6	4·7	25	175 „	(usually passes large quantity).
4.	M.	29	12	6·4	37	115 „	
5.	M.	9	6	8	15	80 „	sp. gr. 1020.
6.	M.	5	2	5	8	155 „	„ 1010; polyuria.
7.	M.	9	6	8	15	125 „	„ 1014.
8.	M.	11	8	7	23	90 „	„ 1021.
9.	M.	10	2	8	5	275 „	„ 1010; polyuria.
10.	M.	10	2	5	8	240 „	„ 1010; „
11.	F.	11	8	8	20	55 „	
12.	F.	7½	8	8	20	35 „	
13.	F.	6½	6	5	24	130 „	„ 1018.
14.	M.	5¾	4	3	27	75 „	„ 1022.
15.	M.	13	6	10	12	110 „	
16.	M.	13	4	10	8	160 „	— polyuria.
17.	M.	14	8	11	15	70 „	
18.	M.	13	6	9	14	70 „	

SUMMARY OF RESULTS.

Healthy Subjects.

I. *Diurnal variations in the urine.*—The urine secreted during the night has generally a higher diastatic activity than that secreted during the day. The diastatic activity of the urine secreted during the day varies inversely, as a rule, with the rate of excretion, and directly to a certain extent with the specific gravity. The total diastatic activity of the urine (amount in c.c. × diastatic index) for the 24 hours gives numbers only approximately constant in the same individual. The total diastatic activity of specimens of urine examined hourly after a meal is highest in the period of the second to the fifth hour. Thereafter the value tends to fall.

II. *Variations in the blood.*—The blood diastase when examined by Fyfe's method exhibits well-marked variations apparently related to the ingestion of food.

The level is lower in the period from the third or fourth hour to the seventh or eighth hour after a meal than it is after fasting or shortly after a meal.

III. *Variations in the diastatic concentration factor.*—In the same individual this varies throughout the day, being highest, as a rule, in the urine secreted overnight or in urine secreted during the fasting condition.

IV. *The figures obtained for the period 2–4 hours after a meal in healthy individuals:* (1) *Blood diastase.*—In the majority the figure obtained lies between 6 and 10, with outside limits of from 3 (exceptionally) to 11 or 12.

(2) *Urine diastase.*—In the absence of polyuria the diastatic index in the majority of healthy individuals lies between 6 and 14.

(3) *Diastatic concentration factor.*—The normal figure obtained lies in the majority of cases between 20 and 30 with outside limits of about 15 to 40.

Nephritic Subjects, etc.

Numerous cases of nephritis mostly, of diabetes, arteriosclerosis, prostatic enlargement, etc., were examined in the same way as the preceding normal cases with a view to seeing whether there was any variation from the normal diastatic concentration factor in renal disease.

The results are shown in Table IX.

Blood diastase.—High levels were found in a number of kidney conditions such as acute nephritis, uræmia, cardio-renal cases with failing heart. In addition, high levels were also found in 2 out of 5 diabetic cases and in 2 out of 4 cases of prostatic enlargement with retention of urine.

Urine diastase.—High levels were found in acute nephritis and in cases of cardio-renal disease showing failure of urinary secretion. Low levels were found in many cases of chronic kidney disease and of prostatic enlargement with retention.

Diastatic concentration factor.—While the figures obtained in acute nephritis were high, low figures were obtained in cases of chronic renal disease, enlarged prostate and diabetes.

Prostatic Cases.

Table X shows the results obtained in four cases of prostatic enlargement with retention of urine.

TABLE X.

Case.	Age.	Blood urea.	Urine urea.	B.D.	U.D.	D.C.F.	Remarks.
38	65	90	—	11·4	2	3·5	General condition fair.
39	85	150	1·35%	3	<2	Low	General condition poor; chronic uræmic state.
40	79	60	1·2%	7	<2	„	Pus in urine; clearing; general condition improving.
41	63	90	1·6%	16	6	7	Stricture and enlarged prostate; blood and pus in urine; tongue dry; general condition poor.

Wide variations were observed in the blood-diastase figure. The urine diastase figures were low with one exception. All the cases, however, gave a low diastatic concentration factor, as the above-mentioned exception (Case 41) had a high blood-diastase figure.

Urea Concentration and Diastatic Concentration Factor.

In the cases shown in Table XI the urea concentration test was performed and a comparison was made with the diastatic concentration factor.

TABLE IX.

Case.	Breakfast. a.m.	Time. a.m.	B.D.	Time. a.m.	B.D.	Urine amount in c.c. 6.0–8.0 a.m.	U.D.	Urine amount in c.c. 8.0–10.0 a.m.	U.D.	D.C.F.	Remarks.
1 M. 26:											
(1) 8 . 8 . 23	6.30	8.30	12	10.35	5·5	—	—	—	—	—	Albumen 12 %. Œdema.
(2) 10 . 8 . 23	6.30	8.30	5	10.50	7·6	35	18	110	18	57	Albumen 12 %. Improved under urea and high protein diet.
(3) 4 . 9 . 23	6.0	8.15	8·4	10.30	10	120	12	150	10	26	Albumen 7 %.
(4) 17 . 9 . 23	7.40	10.40	15	—	—	—	—	—	—	—	Albumen 6 %. Recovery good—one year later.
2 M. 55 . .	7.0	9.5	7	11.5	3·5	—	—	100	8	30	Albumen, trace; deposit phosphates.
3 M. 51 . .	6.0	8.0	10·2	10.20	9·1	—	—	—	14	29	No albumen; previous history of albuminuria.
4 F. 22 . .	6.0	8.10	13·2	10.15	16·1	100	18	150	12	16·5	Eclampsia. Albumen + granular casts. Recovery.
5 F. 34 . .	6.0	8.15	11·9	10.40	7·9	300	2	100	12	24	Albumen, small quantity; occasional cast. Health good.
6 F. 55 . .	6.0	8.15	6·2	10.30	6	45	8	90	12	38	Albumen, small amount; granular casts; few blood-cells.
7 F. 30 . .	6.0	8.15	7·8	10.30	9·2	105	4	90	4	9·4	Puerperal septicæmia. Albumen + blood-cells; occasional cast. Recovery.
8 M. 50 . .	6.0	8.20	10·5	10.10	7·3	45	10	165	12	27	Albumen, trace; occasional granular and cellular cast.
9 F. 30 . .	6.0	8.0	4·5	10.5	8·2	—	—	75	8	25	Eclampsia. Albumen, small amount; few blood-cells; hyaline and granular casts.
10 F. 68 . .	6.0	8.0	12·8	10.15	11·8	180	—	40	18	29	Occasional casts; numerous blood-cells. (Auricular fibrillation.)
11 M. 60 . .	6.0	8.10	10	10.10	9	240	8	150	8	17	Albumen + + numerous casts and R.B.Cs.; arterio-venous aneurysm forearm. Wassermann R. + +.
12 F. 60 . .	6.0	8.15	7	10.30	9·4	—	—	105	16	39	Albumen, small amount; occasional R.B.C.; bronchitis.
13 M. 63 . .	6.0	8.15	17·3	10.0	12·9	120	14	240	10	13·3	Aortic case. Albumen, trace. Died two days later, suddenly.
14 F. 73 . .	6.0	8.10	10	10.25	14·3	180	10	165	4	5	Albumen, trace. Auricular fibrillation and C.S. breathing. Died shortly after.
15 M. 57 . .	6.0	8.0	17·8	10.15	10	18	16	—	16	23	Albumen 2–4 %; R.B.Cs. and casts. Œdema present.

16	M. 73: (1) 11 . 9 . 23	7.0	8.55	4	11.0	2	—	—	240	10	67	Albumen, small amount. Malignant disease of liver and lungs. Died 14 . 9 . 23.
	(2) 13 . 9 . 23	7.0	8.55	18	—	—	—	—	—	—	—	—
17	F. 40 . .	7.30 (fluids)	8.40	11·5	11.0	11·8	—	—	270	2	3·5	Hæmaturia of several weeks. Large amount of blood. Albumen; cells.
18	M. 52 . .	6.30 12.30 p.m. (50 gm. lævulose)	12.20 p.m.	4	1.10 p.m. 2.5 p.m. 3.50 p.m.	11·5 11·8 14·5	—	—	—	—	—	Sugar percentage: ·109, ·104, ·109, ·102.
19	M. 45 . .	8.30 (50 gm. lævulose)	9.30 a.m.	6·5	10.40 a.m. 11.40 a.m. 12.40 p.m.	5 3·6 3·6	—	—	—	—	—	Sugar percentage: ·128, ·162, ·137, ·131.
20	F. 55 . .	6.0 . (Quaker oats)	8.15	7	10.20 a.m.	7·6	60	6	180	4	11	Albumen, trace; occasional cast. Percentage sugar in blood: ·312 and ·236.
21	F. 56 . .	7.30 (ord. diet)	9.10	8·8	11.40	4·4	—	—	—	6	18	Urine 12 % sugar; percentage in blood: ·38, ·46, ·468.
22	M. 38 . .	—	—	—	—	22	—	—	—	12	11	Blood-sugar 0·2 %; urine sugar ½ %.
23	M. 30 . .	—	—	—	—	15	—	—	—	6·6	9	Blood-sugar 0·12 %; urine sugar ½ %.
24	M. 29 . .	—	—	—	—	4	—	—	—	10	50	Blood-sugar 0·11 %; urine sugar ½ %.
26	F. 11 . .	—	—	—	10.0	7	—	—	9–11 a.m. —	<2	3	Sp. gr. 1011; 135 c.c. Very few blood-cells; occasional hyaline and granular casts.
27	F. 13 . .	—	—	—	10.0	9·2	—	—	—	4·5	10	Sp. gr. 1028; 65 c.c. Numerous R.B.Cs.; occasional granular casts.
28	M. 10 . .	—	—	—	10.0	14	—	—	—	2	3	Sp. gr. 1015; 175 c.c. Numerous R.B.Cs.; occasional granular casts.
29	M. 18 . .	—	—	—	10.0	12	—	—	—	12	20	Sp. gr. 1030; 80 c.c. Some R.B.Cs.; very numerous casts. Recovering from uræmia.
30	M. 15: (1) 8 . 12 . 23	—	—	—	10.0	6	—	—	—	20	66	Very scanty urine with a large quantity of blood. Œdema present.
	(2) 20 . 12 . 23	—	—	—	„	15	—	—	—	12	16	Sp. gr. 1022; 35 c.c. R.B.Cs. Numerous casts of all kinds. Small quantity of blood.
31	M. 46 . .	—	—	—	„	8	—	—	—	<2	<5	Arterio-sclerosis; retinal hæmorrhages.
32	F. 42 . .	—	—	—	„	4	—	—	—	3	15	High blood-pressure; small amount of sugar in urine.
33	M. 68 . .	—	—	—	„	5 5	—	—	—	6	22	Auricular fibrillation and arterio-sclerosis.
34	F. 50 . .	—	—	—	„	20	—	—	—	3	3	Arterio-sclerosis.
35	M. 50 . .	—	—	—	„	8	—	—	—	7	18	Chronic interstitial nephritis.
36	F. 47 . .	—	—	—	„	10	—	—	—	8	16	Hyperpiesis.
37	F. 40 . .	—	—	—	„	9	—	—	—	20	44	Acute nephritis with œdema.

TABLE XI.

Age.	Sex.	Urea concentration.	D.C.F.	Remarks.
30	M.	2·4%	24	Healthy.
25	M.	2·5%	22	,,
46	M.	1·25%	5	Arterio-sclerosis with retinal hæmorrhages.
42	F.	2·4%	15	Hyperpiesis.
63	M.	3·0%	22	Auricular fibrillation.
47	F.	0·75%	3	Chronic interstitial nephritis.
50	M.	2·0%	18	,, ,,
47	F.	3%–3·5%	16	Hyperpiesis.
40	F.	4·3%	44	Acute nephritis.
85	M.	1·3%	5	Enlargement of prostate.
79	M.	1·2%	5	,, ,,
63	M.	1·6%	7	,, ,,
81	M.	3·05%	25	,, ,,

The figure obtained in healthy individuals for the diastatic concentration factor lies between 15 and 40. When the figures in the two columns for the urea concentration and the diastatic concentration factor are compared they are found to be in general agreement, normal values of one corresponding to normal percentages of the other, and low values of one corresponding to low percentages of the other.

Stafford and Addis (*loc. cit.*) compare the rate of excretion of diastase with the power of the kidney to concentrate urea, and state as a remarkable fact that emphasis has usually been laid on the concentration of diastase, and not on the rate of excretion of diastase. They obtain the rate of excretion or the total hourly diastatic activity by multiplying the amount of urine in c.c. by the urinary diastatic activity (Wohlgemuth units), and dividing by the time in hours which the kidney took to secrete the specimen considered. In MacLean's urea concentration test stress is laid on the power of the kidney to concentrate urea. In the present investigation stress is laid on the power of the kidney to concentrate diastase from the blood, and not on the rate of excretion as shown by the total hourly diastatic activity.

It is possible to have (*cf.* Case 13, Table IX) a normal urinary diastatic activity and total hourly diastatic activity along with a high blood diastatic activity, thus producing a diastatic concentration factor lower than normal. Further, the hourly rate of excretion is found to vary so widely in different normal individuals that the application of the hourly rate of excretion of diastase to abnormal cases would not appear to be justifiable.

In addition, in the present investigation the diastatic concentration factor is compared in many cases with the urea concentration as obtained by MacLean's test, and in this way conclusions are avoided that depend on a comparison between the urea concentration and the hourly rate of excretion of diastase, which is very variable even in normal subjects.

CONCLUSIONS.

It is advisable to estimate the blood diastatic activity in all cases where the urine diastatic activity is being examined, and especially in those cases which give a urinary diastase figure towards the lower limits of normality.

It would appear that the diastatic concentration factor would serve as an additional confirmatory test to MacLean's urea concentration test, as the figures obtained for both tests in healthy and pathological cases were in general agreement.

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THE EFFECT ON RENAL EFFICIENCY OF LOWERING THE BLOOD-PRESSURE IN CASES OF HIGH BLOOD-PRESSURE[1]

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The following investigation was undertaken to ascertain the effects, especially as regards the efficiency of the kidneys, of lowering the hypertension present in cases which did and others that did not show evidence of renal disease. The plan adopted was to make the following examinations on each case before, during, and after periods of administration of vasodilator drugs; the amounts of urea and non-protein nitrogen in the blood, the power of the kidney to concentrate urea after a dose of 15 grm. urea (MacLean's test (9)), the total amount of urea and the total volume of urine excreted, records of blood-pressure, pulse, respiration, and general condition of the subject being made also.

A current conception of elevated blood-pressure is that, while attended by drawbacks in the way of increased heart work and stress on the arterial walls, it is in large measure a compensatory process in the organism. This view has gained wide acceptance, and many observers have emphasized the inadvisability of indiscriminate lowering of the pressure by such artificial means as the use of vasodilating drugs. Cases have been cited of deterioration in general condition being associated with lowering of high pressures, and, on the other hand, improvement in health being attended, not by lowering, but by some further rise of an already elevated pressure. The high-pressure levels in such cases are regarded as not being excessive in the circumstances, but rather as optimal, or at least not markedly superoptimal under the conditions present in the body at the time. When no symptoms are in evidence artificial reduction of the pressure is held to be inadmissible. Even when symptoms are present it is conceivable that a necessary compensatory action may be exercised in some respects, though the high pressure may involve disturbances in other respects.

In view of the separation of a new antipressor principle from hepatic extracts in comparative purity by James, Laughton, and Macallum (7), and of the prospect of this method of lowering the raised blood-pressure being given

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an extended clinical trial, the importance of data dealing with the efficiency of the kidney under conditions of lowered blood-pressure is obvious—with regard to the differentiation of cases in which reduction of high pressure may be permissible or desirable or the reverse. Some results of the clinical use of the hepatic extracts have been described by Macdonald (8), by Major (13), and by Major and Stephenson (15). Major (14) reported that in two cases of hypertension the excretion of guanidine was not diminished, but rather increased, during a period (several days) of blood-pressure lowering by hepatic extracts. Gruber, Shackelford, and Ecklund (4) found that, when high arterial pressure was lowered by pheno-barbital, no harmful effect was produced on the excretion of phenolsulphonephthalein.

The latter, however, is a foreign substance, and might be thrown out by the kidney independently of any but very extensive changes in blood-pressure, so that, while the above investigation agrees, so far as phenolsulphonephthalein is concerned, with the findings of the present investigation as regards urea, the evidence obtained in the former inquiry is not necessarily valid as an argument against the compensatory theory.

Possible Compensatory Mechanisms.

It is evident that elevated blood-pressure might be a compensatory adjustment in the way of driving more blood through some vital organ, e. g. brain, heart-muscle, or kidney: such might be needed where there is inadequacy of blood-flow depending on alteration in its vascular channels, arterial or capillary, or when, even apart from such alteration, a higher capillary pressure and more rapid blood-flow would be beneficial in enhancing the functioning of an organ—deficient from structural or other causes.

There is the familiar instance of the mechanism by which an interference with the normal blood-supply to the head (e. g. cerebral compression, experimental closure of the carotids) promptly calls forth a rise of aortic pressure with an obviously compensatory significance through excitation of the vasomotor centre, causing constriction in the splanchnic and other areas, and diminution of the activity of the vagus centre leading to increased action of the cardiac pump. The recent experimental work of Anrep and Starling (1) by cross-circulation experiments shows the converse action of increased blood-pressure in the head in depressing the vasomotor centre, in addition to the well-known influence of such pressure in stimulating the vagus centre and slowing the heart.

L. Hill (6) wrote in 1900, 'The vasomotor centre is not only excited reflexly, but responds to every change in the circulation through the spinal bulb. A rise of pressure in the cerebral arteries provokes a fall of aortic tension; conversely, a fall of pressure in the cerebral arteries provokes a rise. In other words, cerebral anaemia, however produced, excites the centre and increases vascular tone, while cerebral hyperaemia decreases vascular tone.'

In cases of high blood-pressure Starling (17) attaches much importance

to a stimulating influence on the vasomotor centre resulting from a defective blood-supply to that centre. The remarkable variability of the pressure from day to day or hour to hour in some high-pressure cases has to be kept in mind in relation to such a view.

It is obviously possible that with regard to other vital organs, as in the case of the brain, there may be vascular adjustments of a compensatory character involving a rise of aortic pressure.

In the case of the kidney a rise of general blood-pressure might have a compensatory value in aiding the excretion of concentrated urine, salts, abnormal substances, or excess of acid or other waste products; or, again, when the materials to be excreted are not abnormal or excessive in amount, but the functioning of the organ is defective from structural change or other causes. The improvement might be associated with increase in the flow of water or determined in other ways.

Bier (2) first suggested that hypertension with the arteriosclerotic or atherosclerotic kidney is best regarded as a compensatory effort of the organism, to be interfered with only when danger threatens either of cardiac failure or of cerebral haemorrhage. According to this view, by diminishing hypertension, a danger more or less imminent would be replaced by the certain danger derived from an upset of the kidney efficiency, maintained only at an efficient level by the raised blood-pressure.

Relations of Blood-pressures and Renal Efficiency.

The existing data bearing on the frequent coexistence of high blood-pressures and defective kidney efficiency do not afford grounds for determining the relations between the former and the latter. Examination of the relations between the heights of the blood-pressures and the existence and degree of ascertained defects in urinary excretion (urea, &c.) as studied in different individuals is obviously inadequate, since the degree of kidney damage which may be present in the different subjects constitutes a factor of unknown value. This factor may obviously determine various relations between the levels of blood-pressures present and the degrees of defect in urinary excretion. If it is assumed for the moment that high blood-pressure (as many believe) can favour kidney efficiency, the fact remains that there might be very different degrees of defective excretion in presence of equally high blood-pressures, and, on the other hand, that excretion might be relatively good in association with comparatively low blood-pressures—the existence of varying (unknown) amounts of kidney damage constituting the deciding factor in the different subjects examined.

To test the relationship of high blood-pressures and renal efficiency it is clearly necessary to make observations on the same individual in whom, with given kidney conditions, lowering of the blood-pressure is purposely induced in order to ascertain what alteration, if any, in renal efficiency occurs in association with the alteration in the blood-pressure, the response of the kidneys

to a definite test (urea concentration) being ascertained, while the blood urea and the non-protein nitrogen are also examined.

As regards the known relation of blood-pressure to the excretion of water, Herringham (5) states that, broadly speaking, blood-pressure and amount of urine vary together, though not from day to day in individual cases; in disease the quantity of urinary water does not vary so directly with blood-pressure as might be expected. The urine may diminish while the pressure is steady, or the urine may remain steady while the pressure falls. Such variations are not accounted for by fresh access of local inflammation in the kidney, &c.; they are ascribed to local vascular changes.

Deviations from the general relationship between height of general blood-pressure and volume of urine are readily intelligible in view of what is known of the occurrence of special alterations in the calibre of the renal vessels from nervous or chemical influences, apart from or in addition to variations in aortic pressure, as well as the effects of changes in the composition of the blood (hydraemia, presence of diuretic constituents, &c.), such being capable of affecting the water excretion without parallel change in aortic pressure. But in view of the general relationship between blood-pressure and urinary volume it is, of course, to be expected that the administration of nitrites should have decided effects.

Mason (16) has recently found that sodium nitrite alters the urinary volume sufficiently and frequently enough to warrant its withdrawal during a water test for renal efficiency; the effects on blood and urinary nitrogen were not described.

There is no evidence of nitrites influencing kidney function otherwise than through the vascular changes induced. It is evident that dilatation of the renal vessels and the usual fall of general blood-pressure under nitrites act in different directions on the flow of urine, the former tending to give increased transudation or filtration, and the latter to diminish the excretion of water. Upon the relative predominance of one or other of these two influences the urinary result will naturally depend.

The Method employed in the Study of the Renal Efficiency of Cases with High Blood-pressure and of the same Cases under the Influence of Vasodilators.

On the first day of examination, breakfast was taken about 5 a.m. No food or drink was allowed after this until after completion of the urea concentration test on that day.

About 9.30 a.m. to 10 a.m. at least 6 c.c. of blood were removed by puncture of one of the veins over the anterior aspect of the elbow, and received into a sterile test-tube containing a small quantity of powdered neutral potassium oxalate. The blood was used for the estimation of the urea and non-protein nitrogen.

Immediately after the vein puncture, the bladder was emptied, and 15 grm. of urea in 100 c.c. of water administered.

Specimens of urine were obtained, with the exception of one or two cases, at

one hour, two hours, three hours after the administration of the 15 grm. urea. The quantity, urea concentration, specific gravity, presence of albumin, examination of the centrifugalized deposit, were noted. The patient was then allowed to resume his normal diet, and in the course of the afternoon, between 2 p.m. and 3 p.m., the exhibition of liquor trinitrini ℧ij three-hourly for the succeeding twenty-four hours was commenced.

In one series of cases referred to in Table VI, erythrol tetranitrate was used as a vasodilator; the general effects are seen to be similar. On the second day of examination, i. e. about eighteen hours after the administration of the first dose of trinitrinum, the same examinations of the blood and urine were carried out at approximately the same time and with the same routine.

After the completion of the second urea test no further trinitrinum was given. On the third day, the blood and urine were examined again as before.

Blood-pressure readings, both systolic and diastolic, pulse-rates, and respiration-rates were observed each day during the forenoon in practically every case. Full clinical notes were also made. No difficulty was experienced in getting blood from the same vein on successive days, so that the other arm was always used for blood-pressure readings.

In Case 1 no liquor trinitrini was employed, as a venesection was decided on by the medical officer in charge of the case. No untoward effects due to the liquor trinitrini were observed except in one or two cases. One case (No. 2) complained of flushes and palpitation, while another case (No. 7) did not show a lowered blood-pressure until the liquor trinitrini had been administered in two-minim doses three-hourly for forty-eight hours, and three-minim doses four-hourly for the succeeding twenty-four hours. This caused the patient to vomit and to suffer from headache, palpitation, &c., while his blood-pressure fell considerably on the third day of the administration of the liquor trinitrini.

One or two cases showed a slight increase in the urea and non-protein nitrogen of the blood while under the influence of the vasodilator. In order to make certain that this increase in the blood urea was not due to diminished power of the kidney, while the blood-pressure was lowered, to excrete the increased amount in the blood due to the giving of the initial dose of urea on the preceding day, the above routine was slightly altered as follows in a series of cases:

1st day: Routine examination of blood, urine, &c.

2nd day: Administration of the trinitrinum begun.

3rd day: Routine examination of blood, urine, &c. Trinitrinum stopped after completion of the urea concentration test.

4th day: Nil.

5th day: Routine examination of blood, urine, &c.

In all cases patients were kept in bed throughout the three- or five-day periods of examination to obviate as far as possible the influence of variations in external temperature, &c., on diuresis.

Table I.

Date.	Sex and Age.	Blood-pressure. Syst.	Blood-pressure. Diast.	Pulse-pressure.	Syst. B.-P. Fall.	Pulse-rate.	Resp.	Blood Urea.	Non-pro. Nit.	Urea Concentration. 1st Hr.	Urea Concentration. 2nd Hr.	Urea Concentration. 3rd Hr.	Quantity in c.c. 1st Hr.	Quantity in c.c. 2nd Hr.	Quantity in c.c. 3rd Hr.	Total.	Amt. Urea in grm. 1st Hr.	Amt. Urea in grm. 2nd Hr.	Amt. Urea in grm. 3rd Hr.	Total.
(1)																				
14.8.24	M. 53	220	120–30	90–100	—	100	20	47	42	1·3	1·7	2·1	235	165	145	545	3·1	2·9	3·0	9·0
15.8.24		160	100	60	60	90	18	43	31	2·3	2·8	3·1	140	125	50	315	3·2	3·5	1·5	8·2
16.8.24		210	132	78	—	98	20	52	42	1·7	2·0	2·4	110	150	135	395	1·9	3·1	3·3	8·3
(2)																				
15.8.24	F. 70	216	110	106	—	68	18	36	—	1·4	2·2		190	170		360	2·7	3·3		6·0
16.8.24		184	90	94	32	72	20	34	22	1·8	3·0		210	90		300	3·7	2·7		6·4
17.8.24		184–6	94–8	90	30	76	20	32	21	1·5	2·3		180	125		305	2·7	2·9		5·6
18.8.24		204	100	104	—	70	18	32	—	1·2	2·3		190	—		—	2·5	—		—
(3)																				
18.8.24	M. 72	180	72	108	—	58	—	28	34	2·1	2·9	3·7	150	105	55	310	3·2	3·0	2·1	8·3
20.8.24		144	60	84	36	64	—	25	31	3·1	3·7	3·4	95	60	55	210	2·9	2·2	1·9	7·0
21.8.24		164	70	94	—	60	—	25	31	2·5	3·0	3·3	130	80	55	265	3·3	2·4	1·9	7·6
(4)																				
18.8.24	M. 75	204	106	100	—	58	30	73	43	1·6	2·0	2·3	250	70	40	360	4·0	1·4	0·9	6·3
20.8.24		190	100	90	14	62	30	110*	77*	3·7	3·5	3·5	110	55	55	220	4·1	1·9	1·9	7·9
21.8.24		—	—	—	—	—	—	77	53	3·8	3·7	3·8	145	60	66	271	5·5	2·2	2·5	10·2
(5)																				
22.8.24	M. 72	184	70-6	110	—	58	18	64	39	1·8	2·0	2·4	205	195	65	465	3·7	4·0	1·6	9·3
23.8.24		160	56–60	100	24	62	20	68	41	2·4	2·7	2·8	180	80	45	305	4·3	2·2	1·3	7·8
24.8.24		180	68–70	110	—	55	18	67	40	4+	4+	4+	230	85	85	400	—	—	—	16+
10.9.24		—	—	—	—	—	—	—	—	1·6	1·4	1·8	224	222	94	540	3·6	3·1	1·7	8·4

* Urea given shortly before vein puncture.

Remarks.—(1) Chr. nephritis and dilatation of heart. Venesection 37 oz. 14.8.24 : Urine alb. tr. R. B. C.'s : gran. casts.
(2) Hemiplegia (R.) duration 4/12 yrs. Urine alb. tr. R. B. C.'s : +occasional gran. casts.
(3) Arteriosclerosis. Urine nil.
(4) Arteries very sclerosed. Urine nil.
(5) Arteriosclerosis : loss of memory. Urine nil.

(6)																			
22.8.24	M. 78	170	105	65	—	80	20	94	61	2·6	2·7	3·1	50	55	40	145	1·3	1·5	1·2	4·0
23.8.24		150	90–5	55–60	20	73	18	89	59	2·7	3·1	3·3	80	40	45	165	2·2	1·2	1·5	4·9
24.8.24		164	100	64	—	78	20	89	59	2·7	2·9	3·1	60	70	55	185	1·6	2·0	1·7	5·3
(7)																				
22.8.24	M. 57	220	125	95	—	76	16	133	66	1·1	1·2	1·4	235	50	100	385	2·6	0·6	1·4	4·6
23.8.24		—	—	—	—	—	—	138	77	—	—	—	—	—	—	—	—	—	—	—
24.8.24		230–40	125–30	110	—	—	—	130	67	1·5	1·6	1·7	255	140	130	525	3·9	2·2	2·2	8·3
26.8.24		170	110	60	50	93	20	153	82	2·1	2·2	2·3	300	25	35	360	6·3	0·5	0·8	7·6
28.8.24		216	105	110	—	72	16	150–70	78	1·8	1·8	1·8	160	115	30	305	2·9	2·0	0·5	5·4
(8)																				
25.8.24	M. 58	142	56	86	—	102	C.-S.*	138	92	3·1	3·3	3·3	135	75	65	275	4·2	2·5	2·1	8·8
26.8.24		130	64–6	65	12	92	33	177	102	3·1	3·1	3·1	140	70	60	270	4·3	2·1	1·9	8·3
27.8.24		144	60–2	80	—	85–90	C.-S.*	210	98	3·0	2·9	—	220	70	—	—	6·6	2·0	—	—
(9)																				
25.8.24	M. 54	184	96–8	88	—	63	20	63	48	1·5	2·2	1·8	180	80	85	345	2·7	1·8	1·5	6·0
26.8.24		150–2	90–2	60	30	62	22	59	52	1·7	2·7	2·4	210	85	40	335	3·6	2·3	1·0	6·9
27.8.24		170–4	94–6	80	—	54	18	60	46	1·6	2·0	1·9	245	105	115	465	3·8	2·1	2·2	8·1
(10)																				
25.8.24	M. 81	240	125	115	—	60	—	68	55	1·1	1·5	1·8	300	160	100	560	3·3	2·4	1·8	7·5
26.8.24		172–6	110–2	60	68	63	—	68	56	2·7	3·8	4·0	115	50	40	205	3·1	1·9	1·6	6·6
27.8.24		220–5	120–5	100	—	60	—	60	53	2·1	2·3	2·4	130	155	130	415	2·8	3·7	3·1	9·6
(11)																				
29.8.24	M. 26	160–2	105–10	55	—	70	12	44	38	1·7	1·5	1·9	90	155	105	350	1·5	2·4	2·0	5·9
31.8.24		130	90	40	30	75	17	40	38	1·2	1·6	2·1	245	150	115	510	3·1	4·1	2·4	9·6
2.9.24		136	80–5	50	—	75	13	40	42	2·0	2·1	2·6	135	95	62	292	2·7	2·0	1·6	6·3

* C.-S. = Cheyne-Stokes.

Remarks.—(6) Arteriosclerosis: arthritis deformans. Urine nil.
(7) Chr. nephritis: rheumatoid arthritis. Lead poisoning (25 yrs. ago). Urine alb. tr.: occasional R.B.C.'s. Sickness, vomiting, &c., during night of 25/26.8.24.
(8) Myocarditis. Urine clear. Died 27.8.24.
(9) Arteriosclerosis with Stokes-Adams' syndrome (?). Urine clear.
(10) Myocarditis with rheumatoid arthritis. Urine clear.
(11) Acute nephritis (5 days). Urine alb. +. Blood cells and casts present.

TABLE I (*continued*).

Date.	Sex and Age.	Blood-pressure.		Pulse-pressure.	Syst. B.-P. Fall.	Pulse-rate.	Resp.	Blood Urea.	Non-pro. Nit.	Urea Concentration.			Quantity in c.c.			Total.	Amt. Urea in grm.			Total.
		Syst.	Diast.							1st Hr.	2nd Hr.	3rd Hr.	1st Hr.	2nd Hr.	3rd Hr.		1st Hr.	2nd Hr.	3rd Hr.	
(12)																				
29.8.24	M. 21	150	75–80	75	—	70	18	41	35	1·6	2·1	2·3	150	75	85	310	2·3	1·6	2·0	5·9
31.8.24		130	70	60	20	70	16	40	35	2·0	2·8	2·9	225	65	65	355	4·6	1·8	1·9	8·3
2.9.24		150–4	75–85	70	—	63	15	43	38	2·0	2·7	2·9	237	47	43	327	4·6	1·3	1·3	7·2
(13)																				
30.8.24	F. 60	280–90	150-5	130	—	94	26	58	43	1·4	2·1	2·5	120	150	100	370	1·7	3·1	2·5	7·3
1.9.24		230–50	130–40	100	50	86	24	—	—	2·4	2·7	2·9	125	82	62	269	3·0	2·2	1·8	7·0
3.9.24		275–300	130–45	130	—	83	20	—	—	2·6	2·6	3·0	81	90	70	241	2·1	2·4	2·1	6·6
(14)																				
29.8.24	F. 40	130	75	55	—	83	20	48	38	2·9	4·0	4·2	125	75	60	260	3·7	3·0	2·5	9·2
31.8.24		114–20	80	40	15	90	20	46	38	1·9	2·7	3·5	210	80	75	365	4·1	2·2	2·7	9·0
2.9.24		130	80–5	50	—	84	19	45	42	1·9	2·4	3·1	200	95	65	360	3·8	2·3	2·0	8·1
(15)																				
30.8.24	F. 63	194–204	90	110	—	65	16	49	39	2·1	2·7	2·6	90	60	50	200	1·9	1·6	1·3	4·8
1.9.24		170	80–5	90	30	64	16	49	39	2·0	2·7	2·9	86	75	40	201	1·7	2·0	1·2	4·9
3.9.24		190+	80–5	110	—	54	14	49	41	1·9	2·7	2·8	90	67	40	197	1·7	1·8	1·1	4·6
(16)																				
30.8.24	M. 71	180	80	100	—	65	16	41	38	2·3	2·4	2·4	170	60	95	325	3·9	1·4	2·3	7·6
1.9.24		155	80	75	25	80	18	42	41	2·1	2·5	3·0	155	87	60	302	3·3	2·2	1·8	7·3
3.9.24		176	80–5	90	—	72	16	39	38	2·2	2·8	2·7	135	80	45	260	3·0	2·2	1·2	6·4
(17)																				
4.9.24	M. 61	140	58–65	80	—	72	14	60	50	2·1	0·9	1·7	76	195	93	364	1·6	1·7	1·6	4·9
6.9.24		120	70-5	50	20	65	15	54	48	1·2	1·4	1·6	141	140	84	365	1·7	2·6	1·4	5·7
8.9.24		130	70	60	—	60	12	54	48	2·2	2·3	2·3	70	90	57	217	1·5	2·0	1·3	4·8

Remarks.—(12) Nephritis 4 months: no oedema at present. Urine alb. + : R. B. C.'s and occasional gran. casts.
(13) Hemiplegia (R.) 5 months: very excitable. Urine alb. tr.: occasional R. B. C.'s. Blood and gran. casts.
(14) Aneurysm of ascending aorta (with erosion of sternum). Urine clear.
(15) Angina pectoris. Urine clear.
(16) Arteriosclerosis: myocarditis. Urine clear.
(17) Urine clear.

(18)																			
4.9.24	M. 50	160–70	85–90	80	—	80	—	45	49	2·0	1·7	2·1	25	146	72	243	0·5	2·5	1·5	4·5
6.9.24		180	100	80	+20	68	—	45	48	2·9	2·8	2·3	110	112	57	279	3·2	3·1	1·3	7·7
8.9.24		160–4	90	70	—	76	—	42	46	2·6	2·5	2·3	108	84	40	232	2·8	2·1	0·9	5·8
(19)																			
4.9.24	M. 47	140–50	80–90	60	—	72	18	60	42	2·8	3·2	3·0	160	80	75	315	4·4	2·6	2·3	9·3
6.9.24		130	90	40	15	68	18	55	42	—	3·1		—	83		—	—	2·5		—
8.9.24		140	80–5	60	—	70	15	56	43	2·2	2·9	2·2	96	116	192	404	2·1	3·3	4·2	9·6
(20)																			
4.9.24	F. 60	160–80	95	80	—	72	16	49	39	1·1	2·2	2·4	235	46	98	379	2·5	1·0	2·4	5·9
6.9.24		134–8	90	45	35	76	17	55	42	1·6	2·8	3·4	140	83	51	274	2·3	2·3	1·7	6·3
8.9.24		140–50	80–5	60	—	68	14–15	52	41	1·5	2·8	3·2	160	81	40	281	2·3	2·2	1·3	5·8
(21)																			
7.9.24	M. 53	220–10	125–20	90	—	75	15	60	49	1·9		2·1	235		140	375	4·6		2·9	7·5
		210–200	125–30																	
9.9.24		170–80	120–5	50	40	70	15	54	46	2·4		3·1	108		120	228	2·6		3·7	6·3
11.9.24		206	124	85	—	68	14	53	44	2·1	2·0	2·0	200	124	100	424	4·1	2·5	2·0	8·6

Remarks.—(18) Hemiplegia (L.).
(19) Hemiplegia (L.) 14 months ago. Urine clear.
(20) Urine clear.
(21) Cerebral apoplexy. Urine alb. tr.: occasional R. B. C.'s.

Methods: (1) *Urea.* The urease method of Van Slyke as modified by MacLean (10) was employed.

(2) *Non-protein nitrogen.* For this estimation the adaptation of Folin's method described by MacLean (11) was used.

(3) *Percentage of urea in the urine.* The hypobromite method was used, the volume of nitrogen evolved being measured within one minute after shaking, and the equivalent percentage of urea read off from tables compiled by means of estimations carried out on various specimens of urine by the urease method.

(4) *Systolic and diastolic blood-pressure.* The auscultatory method was used throughout. The patient was kept in the semi-recumbent position, and the same upper arm was employed for the compression armlet in all observations, an Oliver auditory tambour being used. The systolic blood-pressure index was always checked by simultaneous employment of the tactile method, recommended as a routine method a number of years ago by MacWilliam and Melvin (12). The diastolic pressure was taken as usual as the beginning of the fourth phase. In no cases were first readings relied on; the readings were repeated in each case several times during the course of half an hour till a constant level was obtained as shown by both auditory and tactile indices. During this time, variations in the pulse and respiration were noted. Each reading was made quickly, so as to avoid prolonged compression by the armlet, undue congestion of the arm, &c. The pressure was estimated twice in each three-hourly period, once before the middle of each period and another in its latter part. Such a distribution of the estimations tends to reduce possible disturbance of values due to any variations of pressure that may occur within the period, and give a nearer approach to the average level of pressure. Substantial lowering of pressure shows a general parallelism with the reduction in the volume of urine excreted during the same period.

The chief results obtained are stated in Table I on pp. 416–19, dealing with pathological cases of high blood-pressure, some with and others without kidney lesions, cardiovascular changes of various kinds being usually present as noted in the table. The patients were mostly in middle or advanced life—sixteen males and five females. Their general condition varied much.

The dietetic conditions were similar in almost all cases—the usual infirmary full diet. The urea test was completed and blood samples taken in the morning, no food or drink having been taken for at least five to six hours previously.

Preliminary Conclusions from the Results obtained in the Pathological Cases in Table I under the Influence of the Vasodilator Drug.

1. *Effects on the blood urea and non-protein nitrogen.* The blood urea figure and the non-protein nitrogen figure expressed in milligrams per 100 c.c. blood were not affected to any extent except in Cases 4, 7, and 8, in which a decided rise occurred. The reasons for the rise in Cases 4 and 8 are quite definite. In

Case 4 the observation was vitiated by the urea being given some minutes before the blood sample was taken, while Case 8 was a terminal one dying from aortic heart disease. In Case 7 the vasodilator drug was pushed to the point of intolerance (vomiting, &c., being induced) as the blood-pressure was affected with difficulty. This case received liquor trinitrini ℥ij three-hourly for two days, followed by ℥iij three-hourly for one day. It may be noted that in Case 8, during the period of comparatively slight lowering of systolic pressure under the influence of trinitrinum, the general condition was obviously improved, the patient much more comfortable, the pulse slower, and the Cheyne-Stokes' respiration abolished —to recur on the day following the discontinuance of the vasodilator drug when the pressure had again risen.

2. *Effect on the power of the kidney to concentrate urea.* All the cases, with one exception, showed no impairment of the power to concentrate urea. This Case—14—showed a 66 per cent. rise in the first hour in the total amount of urine excreted. With regard to the other cases, the power of the kidney to concentrate urea was increased commonly 25 per cent. up to 75 per cent. above the original value.

3. *Effect on the total quantity of the urine excreted during the three hours following the administration of 15 grm. urea.* The majority of the cases showed a decrease in the total amount of urine excreted in the three hours. The percentage decrease amounted in many cases to 20–40 per cent., with one case showing a reduction of over 50 per cent. Notable exceptions to the decrease were Cases 7, 11, and 12—the last two being recent cases of acute nephritis. Cases 11 (acute nephritis of five days' duration) and 7 showed a 40 per cent. increase in the amount of urine. The increased amount was observed principally during the first hour (Cases 11, 12, 14), and during the first and second hours (Case 7).

4. *Effect on the total urea excreted during the three hours' urea test.* The majority of the cases showed little or no substantial change. No case showed any marked cutting down of the total amount. On the other hand, Cases 7, 11, 12, 18 showed percentage increases of from 35 to 80. The increase in each case was associated with an increase in the total amount of urine. Two cases (4, 20) showed an increased excretion of urea with a decrease in the total amount of urine.

5. *The relation of the increase in the urea concentration to the fall in blood-pressure* is brought out in the following table:

TABLE II.

Case No.	Fall in Systolic Pressure. (mm. of mercury.)	Fall in Diastolic Pressure. (mm. of mercury.)	Fall in Pulse-pressure. (mm. of mercury.)	Percentage Increase in Urea Concentration.
1	60	20-30	40	65
2	32	20	12	27
3	36	12	24	28
4	14	6	8	75
5	24	15	9	35
6	20	10-15	5-10	15
7	50-70	15-20	35-50	38-83
8	12	+10	2	−6
9	34	8	26	23
10	70	15	55	153
11	30	15-20	10-15	7
12	20	5-10	10-15	33
13	40	15-25	15-25	29
15	30	5-10	20-25	0
16	25	0	25	4
20	30-40	5	25-35	27
21	40-50	5-10	35-40	48

Cases with a large systolic fall (Cases 1, 7, 10, 21, especially) showed the biggest percentage increase in the urea concentration.

6. *Relation of the height of the systolic blood-pressure to the blood urea content.* No definite relationship is observable between the amount of urea in the blood and the height of the blood-pressure.

TABLE III.

Relation of Changes in Urine Volume to Pulse-pressure Changes.

Case.	Change in Pulse-pressure. (mm. of mercury.)	Change in Vol. of Urine in c.c. (3-hourly interval after 15 grm. urea.)
1	−40	−230
2	−12	−60
3	−10-24	−98
4	−8	−140
5	−9	−160
6	−5-10	+20
7	0 to +20	−140
	−35	−25
8	−2	−5
9	−20	−10
10	−55	−355
11	−10-15	+240
12	−10-15	+45
13	−15-25	−101
14	−15	+10
15	−5-10	+1
16	−15-25	−23
17	−20	+1
18	0 to 5	+36
20	−25-35	−105
21	−35-40	−147

Cases 7 and 18 showed apparently some increase in the urea concentration, total urea, and volume of urine, while the blood-pressure was not lowered. The cases showing the greatest decrease in pulse-pressure tended to exhibit the greatest decrease in the volume of the urine.

Two factors enter into the determination of the increase in the total urea excreted in cases showing an increase: (*a*) increased concentration of urea; (*b*) increased volume of urine. These factors may operate singly or in combination. The increase in the total urea is due to:

(1) Increased concentration of urea in the urine.
Cases 4, 9, 12, 20.

(2) Increased volume.
Case 11 (acute nephritis).

(3) Both factors.
Cases 6, 7, 18.

The slight decrease in the total urea in Cases 3, 5, 10, 21, is associated with a great diminution in the volume of urine excreted.

Two healthy young adults were examined under the same routine observed in the preceding pathological cases. The results are contained in Table IV on pp. 424–5.

Case 22 showed, under the influence of the vasodilator, increased excretion of urine (33 per cent.), increased total urea (22 per cent.), and slight decrease in the urea concentration percentage of the urine. The increased excretion of urea would thus be accounted for by the increased excretion of urine. On the other hand, Case 23 showed a slight diminution in the amount of urine excreted, no substantial change in the total amount of urea, and no diminution in the urea concentration percentage. The variation in the effects on urinary volume in these two cases is in accordance with Cushny's statement (3) that occasionally a slight increase in the urinary volume may be observed, at other times a decrease. These effects are evidently due to the changes in the calibre of the renal vessels. A small quantity may widen them when they are too contracted to allow of the maximal secretion, while on the other hand, if the normal calibre is the optimal, a nitrite may lessen the secretion by lowering the general blood-pressure. When large quantities lower the pressure greatly, they inevitably lead to a lessened secretion or anuria.

In order to exclude the possibility of a retention of blood urea in the early stages of the administration of the vasodilator drug (leading to the increased percentage of urea in the urine), the blood urea was examined about four hours after the drug had been given in the two normal subjects and in the following group of pathological cases dealt with in Table VI.

Cases 22 and 23.

30.10.24. *Liq. trinitrini* ☊*ij 3-hourly for 24 hours.*

	Case 22.	Case 23.			
	Blood Urea.	Blood Urea.	Time.		
31.10.24	25	31	10 a.m.		
1.11.24	22	34	10 a.m.		
3.11.24	24	32	10 a.m.	15 grm.	Urea after 10 a.m.
			Liq. trinitrini ☊*ij at 3 p.m. and 6 p.m.*		
	29	41	7 p.m.		

TABLE IV.

Observations on Two Healthy Adults.

	Systolic.	Diastolic.	Pulse-rate.	Blood Urea.	Amount in c.c.	% Urea.	Total Amount in c.c.	Total Amount of Urea (grm.).
Case 22.								
30.10.24	132	74-6	80	—	90	2·62		2·358
	122	74-8			97	2·52		2·444
					92	2·52		2·318
							279	7·120 (2·55 %)
			Liq. trinitrini begun 3.30 p.m. and continued 3-hourly.					
4.50 p.m.	140-6	80	90					
8 p.m.	140-2	76	92					
			Liq. trinitrini Ⅹ*ij given at noon.*					
31.10.24	120-6	80	—	25	128	2·30		2·944
	118				116	2·48		2·877
					120	2·41		2·892
							364	8·713 (2·31 %)
1.11.24	118-20	70	78	22	84	3·11		2·612
					88	3·17		2·790
					102	2·56		2·611
							274	8·013 (2·92 %)
3.11.24	130-2	70-4	78	24	106	2·16		2·290
	124-6	74-6			131	2·31		3·026
					74	2·81		2·079
							311	7·395 (2·38 %)
			Liq. trinitrini Ⅹ*ij given 3 p.m. and 7.15 p.m.*					
	132-40	70	96	29				

Case 23.								
30.10.24	128–122	80–74	66	—	185	1·75		3·238
					98	2·52		2·470
					72	2·52		1·814
							355	7·522 (2·12 %)
	Liq. trinitrini (X)ij given 3.30 p.m. and approximately every 3 hours until noon 31.10.24.							
			Liq. trinitrini (X)ij, 7.45 p.m.					
8.30 p.m.	108–10	78–80	72					
			Liq. trinitrini (X)ij, 12 noon.					
31.10.24	108–10	80–4	72	31	163	2·07		3·373
12.45 p.m.					75	2·97		2·227
					60	3·28		1·968
							298	7·568 (2·54 %)
1.11.24	102–5	70	69–70	34	130	2·41		3·133
					76	3·03		2·303
					64	3·12		1·997
							270	7·433 (2·57 %)
3.11.24	114	78–80	68–70	32	150	1·97		2·955
					72	3·16		2·275
					60	3·33		1·998
							282	7·228 (2·56 %)
			Liq. trinitrini (X)ij given 3.30 p.m. and 7.15 p.m.					
7.20 p.m.	128	74	80	41				
7.45 p.m.	(from 114–70)							
8.30 p.m.	96	60	60					

As a urea concentration test had been carried out nine hours previously, the small increases in the blood urea at 7 p.m. on 3.11.24 might have been due to the excess of urea in the blood not having been completely excreted. Accordingly the blood urea was estimated on days on which no urea concentration test was carried out and on which the vasodilator drug was administered:

Blood Urea in mg.

Case 22.	11 a.m.	7.30 p.m.	Case 23.	11 a.m.	7.30 p.m.
5.11.24	26	25		25	30
6.11.24	*Liq. trinitrini (X)ij at 8 a.m. 3-hourly for 12 hours.*				
,,	27	25		30	33
7.11.24	24	21·5		27	31

It would appear therefore possible to exclude as a cause of the increased percentage of urea in the urine the possibility of a retention of blood urea in the early stages of the administration of the vasodilator drug in healthy subjects. The excess of urea in the blood resulting from a dose of 15 grm. urea does not appear to be excreted completely in nine hours in the case of the healthy subjects considered above, although the residual amount is small.

The above observations with regard to the blood urea content in the early stages of the vasodilator administration were repeated on six subjects with raised blood-pressure. There was no evidence of an early retention of blood urea, which might conceivably have been a factor in the increased percentage of urea in the urine.

The total amount of urea excreted daily was examined in two healthy cases and in two high blood-pressure cases before and during the administration of vasodilator drug, no urea being given.

TABLE V.

	Normal Day.			Day with Vasodilator Drug.		
	Total Urinary Urea in grm. (1)	Amount Urine in c.c. (2)	Blood Urea in mg. (3)	Total Urinary Urea in grm. (1)	Amount Urine in c.c. (2)	Blood Urea in mg. (3)
Case 22 (healthy)	12·1	906 (12 hours)	26–25	18·0	1173	27–25
Case 23 (healthy)	12·3	453 (12 hours)	25–30	12·6	400	30–33
Case 24 (high pressure)	23·6	1770	83	26·5	2169	85
Case 25 (high pressure)	8·7	379	38	20·4	680	33

	Day after.			Four Days after.		
	Total Urinary Urea in grm. (1)	Amount Urine in c.c. (2)	Blood Urea in mg. (3)	Total Urinary Urea in grm. (1)	Amount Urine in c.c. (2)	Blood Urea in mg. (3)
Case 22 (healthy)	15·2	1246	24–22	14·7	837	—
Case 23 (healthy)	13·9	700	27–31	13·7	720	—
Case 24 (high pressure)	27·8	2745	82			
Case 25 (high pressure)	19·2	727	33			

The amount of urea in the blood was not substantially changed, and the amount excreted in the urine was not diminished in any of the above cases.

The following table, in addition to giving the blood-urea, urinary volume, urea concentration test result, range of the urea percentage, and total urea of the individual specimens of urine in Cases 24 and 25, gives the results observed in four additional high blood-pressure cases where the vasodilator erythrol tetranitrate was given for longer periods up to seven days without any administration of urea. The object of this is to show the effects of the vasodilator apart from any disturbance caused by the artificial introduction of urea into the circulation in the application of the urea test.

TABLE VI.

	Blood-pressures.	Amount Urine in c.c. (24 Hours).	Total Urea in grm.	% Urea in Individual Specimens.	Blood Urea in mg.	Urea Concentration Test %.
Case 24. M. 46	240–160	1770	23·6	1·06–1·63	83	1·25
	220–150	2169	26·5	1·03–1·52	85	
		2475	27·8	0·92–1·42	82	
Case 25. F. 46	220–110	379	8·7	1·3–2·8	38	2·7
	210–105	680	20·4	2·9–3·2	33	
		727	19·2	2·51–2·7	33	
Case 26. M. 63	180–60	760 (25 hours)	24·7 (25 hours)	2·9–3·3	83	3·0
		1234	32·5	2·1–3·4		
		1025	29·7	2·7–3·1		
		1097	26·6	1·4–3·05	44	
Case 27. F. 47	148–88	773	6·5	0·81–0·93	50	0·75
		806	8·3	1·01–1·06		
		811	7·0	0·77–0·96		
		1200	7·8	0·6–0·85	50	
Case 28. M. 50	203–130	1672	15·3	0·8–1·24	43	—
		1192	17·5	0·76–2·31		
		1380	21·1	0·56–2·25		
		1625	19·2	0·4–1·81		
		2257	20·1	0·65–1·65		
		980	15·4	1·03–1·84		
	170–118	1180	18·1	0·94–2·06	43	
Case 29. F. 47	205–105	300 (10 hours)	7·5 (10 hours)	2·5	59	3–3·5
		545	16·2	2·56–3·11		
		530	13·0	2·0–2·7		
		313	6·5	1·9–2·15*		
		745	10·4	1·4–1·2		
		843	13·4	1·4–1·7		
		889	15·6	1·4–1·8	52	

* E. N. stopped on account of headache.

Remarks.—Case 24. Liq. trin. ℥ij 3-hourly; showed early intolerance to erythrol tetranitrate.
Case 25. Liq. trin. ℥ij 3-hourly.
Case 26. Erythrol tetranitrate gr. i 4-hourly.
Case 27. Erythrol tetranitrate gr. i 4-hourly.
Case 28. Erythrol tetranitrate gr. i t.d.s.
Case 29. Erythrol tetranitrate gr. i 4-hourly.

Cases 24 and 29 showed an early intolerance to the vasodilator drug, and the latter in addition showed a very marked reduction in the amount of urine and urea excreted.

With regard to the general effects of lowering blood-pressure, it was exceptional to get any evidence of disturbance in the condition of the patient, except in the cases where the vasodilator drug was used in the larger doses over a longer period. Occasionally slight palpitation and headache were complained of in one or two cases. There was no complaint of giddiness or faintness, and no noticeable change in the colour of the face was observed. The chief complaints in the cases exhibiting intolerance to the drug were of pain and throbbing in the head. Pulse and respiration generally were but little affected (as shown by the recorded figures in Table I), being, as a rule, increased very slightly in frequency. Oedema, which was present in two or three cases, was not increased by the use of the vasodilator drug. One case (acute nephritis), which showed a slight amount of oedema, exhibited a marked increase in the amount of urine and urea excreted, and a fall in the blood urea and blood-pressure during the time that the drug was being used; there was disappearance of the oedema. Another case of acute nephritis with a very large amount of oedema was not affected adversely. The amount of urine increased to some extent, although the drug was employed for a period extending over a week. This is suggestive with regard to the question of salt retention, since the latter is readily indicated by evidences of oedema.

Although the effects of a single dose of liquor trinitrini on the systemic blood-pressure are diminished in a hour or so and pass off according to different observers in periods varying up to two and a half hours, the decrease in the amount of urine secreted throughout the three hours after the administration of 15 grm. urea in cases which had been previously under the influence of repeated doses of nitrite, compared with the amount obtained on days on which the test was carried out without the administration of nitrite, suggests that the effects of repeated doses of nitrites on the kidney outlast those of a single dose on the systemic blood-pressure. Erythrol tetranitrate, used in a number of cases, has a more prolonged action. As a result of the above-mentioned decrease in the amount of urine leading to an artificial increase in the specific gravity and urea percentage of the urine, vasodilator drugs should not be given during the application of MacLean's test, although cases giving urea percentages much under 2 are unlikely to show specimens of urine above this percentage even during the administration of the vasodilator drug. In connexion with this an interesting point comes up. For example, in the application of MacLean's test to Cases 1, 7, 10, 17, 21, under normal conditions, the second hourly specimen of urine gives urea percentages of 1·7, 1·2, 1·5, 0·9, 2·1, respectively. During the period of lowered pressure the corresponding figures obtained were 2·8, 2·2, 3·8, 1·4, 3·1. The question arises naturally whether the efficiency of the kidney is indicated by the higher figures, or whether kidneys which under normal conditions give a low urea percentage with MacLean's test can be differentiated further as regards their response to the test under lowered pressure.

The percentage of urea obtained from the highest of the three-hourly specimens of urine after the application of MacLean's test to an individual is

not necessarily the maximum for the kidneys of that individual, since some of the individual specimens of urine obtained (apart from administration of urea) throughout the twenty-four hours contain in many cases as high percentages of urea as those obtained in the test specimens, sometimes even higher, as shown in Table VI, Cases 24, 26, 27. It would appear, therefore, that useful guidance to the power of the kidney to concentrate urea can sometimes be obtained by ascertaining the percentages of urea in individual specimens of urine passed at different periods throughout the twenty-four hours.

Conclusions.

1. In the healthy subjects the diuresis which usually follows the administration of 15 grm. urea may or may not be cut down by drugs of the nitrite series in the doses stated, and the power of the kidney under the above conditions to concentrate urea is not impaired. The blood urea and non-protein nitrogen are not increased.

2. In high-pressure cases the diuresis which usually follows the administration of 15 grm. urea is usually cut down by drugs of the nitrite series in the doses stated.

3. The total excretion of urea following the administration of 15 grm. urea is usually not diminished by the administration of nitrites in doses sufficient to cause a considerable lowering of the high blood-pressures present (falls of 20–60 mm.).

4. The power of the kidney to concentrate urea after the exhibition of 15 grm. urea is not impaired, inasmuch as urine of higher urea concentration is still excreted during the period of lowered pressure, e.g. 2·8, 3·0, 3·5, as compared with 1·7, 2·2, 2·0 respectively, when the test is applied before the lowering of the pressure. It remains to be seen whether (apart from the evidence afforded by the unimpaired total urea excretion) high urea concentration values, e.g. 3·5, 3·8, during the period of lowered pressure are significant with regard to reduction of pressure being warrantable, so far as the kidney is concerned.

5. If vasodilator drugs are given in pharmacopoeial doses as distinguished from the larger doses referred to above, the functions of the kidney as regards the excretion of water, the excretion of urea, and the power to concentrate urea are not diminished.

6. The urea and non-protein nitrogen content of the blood is not increased by the administration of vasodilator drugs over periods ranging from twenty-four hours to more than one week. The increased urea concentration in the urine is evidently not dependent on an increased percentage in the blood.

7. If the larger doses are maintained over a longer period, symptoms of intolerance to the drug supervene long before the stage of suppression of urine. Symptoms of intolerance may arise in different cases where the power of the kidney to concentrate urea is either (1) above 2 per cent., or (2) well below 2 per cent.

8. The excretion of urea was not interfered with by a large fall of blood-pressure in a high-pressure case by venesection.

9. The virtual maintenance of the total excretion of urea during the period of lowered blood-pressure in cases of hyperpiesia indicates that the mechanism of hyperpiesis is not to be regarded as compensatory, at least so far as the excretion of urea and non-protein nitrogen is concerned—a conclusion in accord with the results of some clinical observations as regards phenolsulphonephthalein excretion reported by Gruber (4), and guanidine excretion by Major (7).

10. No definite relationship is observable between the amount of urea in the blood and the height of the blood-pressure.

11. Nitrites should not be administered either prior to or during the application of MacLean's test.

12. Apart from the application of the urea concentration test, useful evidence as to the urea-concentrating power of the kidney may often be obtained from the examination of individual specimens of urine over the twenty-four hours, since in some of such specimens there may be a percentage of urea as high as, or even higher than is shown by MacLean's test.

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