VIII. "On the Influence of Carbonic Acid and Oxygen upon the Coagulability of the Blood *in Vivo*." By A. E. WRIGHT, M.D. (Dubl.), Professor of Pathology, Army Medical School, Netley. Communicated by AUGUSTUS D. WALLER, M.D., F.R.S. Received February 8, 1894.

I have, in the course of previous researches on blood coagulation,* had occasion to suggest that the phenomena with which I was dealing might be explained in a very simple manner by assuming that carbonic acid gas exercised a favourable influence on the occurrence of blood coagulation. The present research consists of an examination of the hypothesis in question.

The method of experimentation employed consisted in determining the alterations of blood coagulability which occurred in animals when alterations were effected in the respiratory gases with which they were supplied.

Details of the Method of Experimentation employed.

The animals experimented upon were dogs and rabbits. The animals were in all cases tracheotomised under the influence of ether (rabbits) or of a mixture of ether and chloroform (dogs). In the case of the dogs, the animals were kept under the influence of the chloroform and ether during the whole course of the experiment. In the case of the rabbits, the repeated inhalations of carbonic acid and other gaseous mixtures served to keep up the anæsthesia. The tracheotomy tubes were connected up with a T-tube; one limb of the T-tube was fitted with a Speck's intestinal valve (made of rabbit-gut), and allowed of free expiration into the outer air. The other limb of the T-tube was connected up at pleasure with reservoirs (4,000 c.c. capacity) of pure gases or gaseous mixtures standing over water. The water was carefully kept at the same level inside and outside of the reservoirs during the whole course of an experiment. A convenient check upon this was afforded by the regular opening and closing of the intestinal valve.

The blood for the coagulability estimations was obtained from the ear. In the case of the rabbits, the blood was invariably drawn off from the central artery of the ear. Only rabbits with full ear arteries were employed in the experiments.

The blood coagulability determinations were made by the method of capillary coagulation tubes recently† described by me. The method

* 'Journ. of Physiol.,' vol. 12, No. 2; 'Roy. Irish Acad. Proc.,' 3rd Series, vol. 2, No. 2; 'Roy. Soc. Proc.,' February, 1893.

† 'Brit. Med. Journ.,' July 29th, 1893, and February 3rd, 1894.

differs from the method previously described by Vierordt* in the following particulars:—1. A series of capillary tubes, of equal calibre, is employed instead of the single capillary tube employed by Vierordt. 2. Coagulation time is determined by blowing down the capillary tubes, one after another, at regularly increasing intervals until a tube is found to have become blocked by clot. In Vierordt's method the occurrence and duration of coagulation is judged of by passing a chemically cleansed white horse-hair down the capillary tube, and observing the deposition of coagulum upon its surface.

In all cases I employed a column of blood of 5 cm. in length, and received it into tubes† which had a diameter of approximately 0·25 mm.

The following precautions were observed in order to ensure accuracy of results:—1. The coagulation tubes were washed out before use successively with distilled water, absolute alcohol, and ether. 2. They were then warmed in an incubator to a temperature of 37° C. 3. A fresh drop of blood was employed for filling each tube. 4. The column of blood was aspirated some little distance up the tubes to prevent desiccation occurring at the orifice. 5. In testing for coagulation the blood was blown out on to a piece of white filter paper in order to ensure the detection of the first traces of clot.

The gases which were experimented with were the following:—Atmospheric air, oxygen, hydrogen, carbonic acid, and a mixture of (approximately) 20 per cent. of oxygen with 80 per cent. of carbonic acid. I also examined the effect of clamping the trachea.

Effect of an Increase of Carbonic Acid.

In order to elicit the effect of an increase of carbonic acid upon coagulability, I caused the animals to inspire out of a reservoir containing a mixture of 1 part (approximately) of oxygen with 4 parts of carbonic acid. This mixture of gases presents the obvious advantage of supplying carbonic acid in association with the normal quantum of oxygen. Determinations of blood coagulability were made when the animals were breathing this mixture of gases, and the results were compared with the "coagulation times" which were elicited immediately before when the animals were breathing atmospheric air. Thirty experiments were made. Out of these twenty-seven showed a marked increase‡ of coagulability while the animal was breathing the mixture of carbonic acid and oxygen. In two experiments coagulation time was unaltered, and only in one experiment was a slight diminution of coagulability observed.

* 'Archiv für Heilkunde,' 1878.

† These tubes were supplied by Mr. A. E. Dean, Jun., 73, Hatton Garden E.C.

‡ This increase of coagulability is well shown in the first ten of the protocols appended to this paper.

In the three experiments last mentioned the coagulability of the blood was already at a maximum when the animal was breathing atmospheric air.

It is to be noted that the blood which was drawn off while the animal was breathing the carbonic acid and oxygen was arterial in colour in all the experiments which have been summarised above. The increase of coagulability must therefore be ascribed to the increase of carbonic acid in the blood, and not to any defect of oxygenation.

It has thus been demonstrated that the increase of carbonic acid in the blood does exert a favourable influence on coagulation.* Carbonic acid is therefore in all probability what I assumed† it to be, *i.e.*, a *vera causa* in the determination of intravascular coagulation to particular vascular areas.

Effect of a Diminution of Carbonic Acid.

This question was studied by examining the condition of coagulability in animals when an atmosphere rich in carbonic acid was replaced by (*a*) ordinary air, or (*b*) by oxygen.

a. Results of experiments in which an atmosphere of carbonic acid was replaced by ordinary air.—The result‡ of such a replacement of carbonic acid and oxygen by atmospheric air is a decrease of coagulability to the original norm.

b. Results of experiments in which an atmosphere of carbonic acid and oxygen is replaced by an atmosphere of unmixed oxygen.—The substitution of unmixed oxygen for the mixture of carbonic acid and oxygen is invariably followed by a decrease of coagulability. The diminution may be due to a specific effect of an atmosphere of unmixed oxygen. On the other hand it may with much greater probability be referred to the diminution of carbonic acid in the blood, for the rate of respiration is always extraordinarily accelerated (to 160 respirations per minute and upwards) by the inspiration of oxygen. This view is also suggested by the analogy of the experiments in which air is substituted for the carbonic acid mixture. It is further supported by the fact that the diminution of coagulability is apparently proportionate to the amount of carbonic acid which is present in the blood. The diminution is, for instance, well marked

* I have found this statement to hold true also in the case of human blood. The inhalation of an atmosphere which is rich in CO_2 causes an increased coagulability in my own blood. I have obtained a similar increase of coagulability (associated with an arrest of hæmorrhage) in a case of severe bleeding in hæmophilia. I have also obtained an increased coagulability by inhalation of CO_2 in the case of three members of another hæmophiliac family.—21/2/94.

† 'Journ. of Physiol.,' vol. 12; 'Roy. Irish Acad. Proc.,' 3rd Series, vol. 2, No. 2.

‡ This result is well shown on the protocols of rabbit 161 and dog 2.

when the blood is rich in carbonic acid (*e.g.*, in protocols of rabbits 165, 163 and 135), while there is practically no diminution of coagulability when the blood has been adequately ventilated by respiration in ordinary air (*vide* second oxygen inhalation in protocol of rabbit 155).

It evidently results from both these series of experiments that the diminution of carbonic acid in the blood which was assumed by me to afford a clue to the diminished coagulability of peptone* and of blood which has circulated through the lungs and heart alone is in reality capable of exercising a well marked retarding influence upon coagulation.

Effect of a Diminution of Oxygen.

It is extremely difficult to determine with precision what effect the withdrawal of oxygen exercises upon the coagulability of the blood. The difficulty consists in the complication of the phenomena which are due to the withdrawal of oxygen by other phenomena which are due to an increase of carbonic acid in the blood. To elucidate the matter, we evidently require methods which allow of at least a partial dissociation of the effects of the two gases. Such methods should aim at (*a*) a limitation of the amount of carbonic acid produced in the system after the oxygen is withdrawn; (*b*) the elimination of the carbonic acid which is produced; (*c*) a minimising of the effect of the carbonic acid increase. These objects can be partially realised by the two following methods:—

1. Inhalation of an atmosphere of indifferent gas (*e.g.*, hydrogen) while provision is made for free expiration into the external air.

This method provides to some extent for the elimination of the carbonic acid which is produced after the withdrawal of the oxygen. On the other hand, the method does not provide against the accumulation of carbonic acid which must occur during the dyspnœic standstill of respiration.

2. Substitution of an atmosphere of unmixed carbonic acid for an atmosphere of carbonic acid and oxygen.

This method presents two advantages: (*a*) it limits the production of carbonic acid in the system, inasmuch as the withdrawal of oxygen, when made under these particular circumstances, no longer evokes any dyspnœic spasms; (*b*) it minimises the effect of any increased carbonic acid tension inasmuch as such increase takes place in a blood which is already almost saturated with carbonic acid.

* 'Roy. Irish Acad. Proc.,' 3rd Series, vol. 2, No. 2, 1891; 'Roy. Soc. Proc.,' February, 1893.

1. *Results of Experiments in which Oxygen was withdrawn by the Substitution of Hydrogen for Atmospheric Air.*

I have employed this method in 29 experiments. In 15 of these experiments a diminution of coagulability was observed to result from the inhalation of hydrogen. In 14 other experiments an increase of coagulability was noted. With respect to the latter results, the following points are to be noted:—(*a*) The increase of coagulability was invariably confined within very moderate limits;* (*b*) in 2† out of the 14 experiments expiration was found to have been obstructed by an accidental compression of the tracheal tube.

The results of these experiments are patently ambiguous. On the one hand we have a bare majority of experiments or (if we subtract the experiments in which expiration was accidentally obstructed) a majority of only 15 to 12 experiments in favour of the result that the inspiration of hydrogen conditions a diminution, and not an increase, of blood coagulability. On the other hand, it is evident that there is nothing in these experiments when taken by themselves to justify a conclusion as to whether it is the decrease or the increase of coagulability which is to be regarded as the effect of the withdrawal of the oxygen. In such a case the only available method of interpretation consists in subducting from the aggregate of the observed phenomena such phenomena as we know by previous inductions to be the result of disturbing factors which cannot be eliminated from the experiments. The accumulation of carbonic acid in the blood, which occurs when the inevitable dyspnœic standstill of respiration takes place, or when (as in rabbit 176 and dog 4) expiration is accidentally obstructed, is just such a disturbing factor, and the effects of this disturbing factor must, in accordance with our previous inductions, manifest themselves in an increased blood coagulability. We may, therefore, legitimately assign to this cause all the phenomena of increased blood coagulability which came under observation in the hydrogen experiments. The residue of the observed phenomena, in other words the diminution of blood coagulability, then emerges as the effect of the absence of oxygen from the inspired air.

If the above train of argument is valid, we must conclude that the diminution of the oxygen of the blood conditions a diminution of coagulability.

* Coagulation-time was never found reduced below 1 minute 30 seconds (*vide* protocol rabbit 176). Coagulation-times of less than 1 minute are frequent (*vide* protocols *passim*) during inspiration of carbonic acid and oxygen.

† *Vide* protocols of rabbit 176 and of dog 4.

2. *Results of Experiments in which Oxygen was withdrawn by a Substitution of Unmixed Carbonic Acid for a Mixture of Carbonic Acid and Oxygen.*

This substitution of carbonic acid for the mixture of carbonic acid and oxygen tends to effectuate itself spontaneously by the slowing down and ultimate standstill of respiratory movements which supervene when an animal is continuously supplied with an atmosphere surcharged with carbonic acid. A defect of oxygenation was allowed to supervene in this manner in five* experiments. In all these cases a diminution of coagulability was observed.

Similar experiments were performed by the actual substitution of unmixed carbonic acid for an atmosphere of carbonic acid and oxygen. In the four† experiments which were performed a diminution of coagulability was invariably observed.

The diminution of coagulability which is observed by either variant of this method may be interpreted either (*a*) as an effect of an excess of carbonic acid in the blood, or (*b*) as an effect of the withdrawal of oxygen. Against the former interpretation of the facts the following considerations may be urged: (1) the tension of carbonic acid in the blood must already have been very high when the substitution of gases was effected; (2) with the then obtaining very slow respiratory movements the respiratory interchange in the lungs must have been at a minimum. It is, therefore, unlikely that any appreciable increase of carbonic acid tension can have effectuated itself in the blood in the interval during which the lungs were filled with unmixed carbonic acid.

If this reasoning is valid, we must evidently interpret the diminution of coagulability which came under observation in all these experiments as a direct result of the withdrawal of the oxygen.

It need hardly be pointed out that this interpretation would harmonise with the interpretation which has just been placed upon the hydrogen experiments.

Effect of a Restoration of Oxygen to Blood rendered Anoxyhæmic by the Inspiration of Hydrogen.

It may be premised that we have here, as in the case of the hydrogen experiments, to disentangle the effects of a duplicated series of phenomena, (*a*) the giving off of any excess of carbonic acid which has accumulated in the blood, and (*b*) the restoration of oxygen to the blood. The effect of (*a*) would, in accordance with our previous‡ experiments, be a diminution of coagulability. On the

* Three of these experiments will be found on the protocols of rabbit 175, dog 3, and rabbit 171.

† Two of these experiments will be found on the protocols of rabbit 175 and rabbit 171.

‡ *Vide supra*, Experiments on Effect of Diminution of CO_2.

other hand, if the interpretation which we have placed upon the results of our experiments on the effect of a diminution of oxygen is correct, we should expect an increase of coagulability to accompany the restoration of oxygen to the blood.

The following is a summary of the results of the experiments which were directed to the determination of this point.

In a total of fourteen experiments, restoration of atmospheric air was in ten instances* found to result in an increase of coagulability. In the four remaining instances a decrease of coagulability was noted. It is, however, significant that in two of these instances the diminution of coagulability which was observed lay well within the limits of error of the method of determination, while in the remaining two instances† the diminution of coagulability was only a rebound from a condition of increased blood coagulability which was brought about by an accidental obstruction to the expiration of carbonic acid.

We may, therefore, conclude that the restoration of oxygen *to anoxyhæmic blood* conditions an increase of coagulability.

Comparison of the Results obtained above with the Results obtained by other Observers.

In recent times the question of the influence exerted by the blood gases on coagulation has been investigated among others by Vierordt,‡ Hasebroek,§ and Bonne,‖ and also by Mathieu and Urbain.¶ The two first of these observers employed Vierordt's method of coagulability determinations, and both observers performed their experiments chiefly upon themselves. In none of their experiments does any attempt appear to have been made to dissociate the effect of changes in the quantity of carbonic acid in the blood from the effect of simultaneous changes in the quantity of oxygen in the blood. On the contrary, the phenomena which came under observation appear to have been referred to either one or other of these causes according to the particular bias of either observer. Thus Vierordt ascribes the increased coagulability which he detected in the stagnating blood of his ligatured finger to an increase in the CO_2 tension. On the other hand, Hasebroek, who reinvestigated this point, interprets the increased coagulability which is observed after a brief application of a ligature to the finger as an effect of a diminution of oxygen

* Examples of such increase of coagulability are given on protocols of rabbit 175, dog 4, rabbit 176, and rabbit 178.

† *Vide* third coagulation-determination on protocol of dog 4 and penultimate coagulation-determination on protocol of rabbit 176.

‡ *Loc. cit.*

§ 'Zeit. f. Biol.,' 1882.

‖ 'Ueber das Fibrin-Ferment,' Würzburg, 1889.

¶ 'Comptes Rendus,' 1874, vol. 2, pp. 665 *et seq.*, and 698 *et seq.*

in the blood, while he ascribed the diminished coagulability which is observed after a lengthened application of the ligature to an excess of the carbonic acid. In like manner this observer ascribes (*a*) the increased coagulability which he obtained after holding his breath for twenty seconds to an increase of CO_2 in his blood, (*b*) the diminished coagulability he obtained after holding his breath for forty-five seconds to an excess of carbonic acid, (*c*) the diminished coagulability of venous, as compared with arterial, blood to the same cause, and (*d*) the diminished coagulability of his blood after rapid respiration to an excess of oxygen. There is nothing in the experiments to justify any of these inferences.

Exactly the same objections can be urged against Bonne's experiments. It will suffice to point out that Bonne obtained a diminution of coagulability in a bare majority of experiments in pigeons in which asphyxia was produced by the inspiration of carbonic acid, and that he interprets this diminution of coagulability as an effect of the excess of carbonic acid tension, while the anoxyhæmia to which the animals succumbed is entirely left out of sight as a possible factor in the causation.

Lastly, the work of Mathieu and Urbain comes up for notice. These observers conjointly investigated the effect of carbonic acid upon blood coagulability, and came to the conclusion that carbonic acid was a very important, if not indeed the all-important, agent in the production of blood coagulation. This conclusion was based upon the following observations: (*a*) Blood coagulation is accompanied by a giving off of something like 50 per cent. of the carbonic acid originally present in the blood; (*b*) an artificial increase of the body temperature goes hand in hand with a diminution of the carbonic acid and with an increase in the oxygen in the blood, and this artificial increase of the body temperature results in a diminished coagulability; (*c*) the blood from the renal vein, which resembles in its gaseous composition the blood of the superheated organism, is characterised by a similar diminished coagulability; (*d*) an artificial reduction of the body temperature goes hand in hand with an increase of carbonic acid, which is quantitatively comparable to the increase which is produced by asphyxiating an animal by CO_2. This increase of carbonic acid in the blood under the influence of cold goes hand in hand with an increased blood coagulability; (*e*) when blood is prevented from clotting by the addition of a few drops of ammonia (the ammonia is assumed to retard coagulation by binding the free carbonic acid), and when a new formation of carbonic acid is prevented by eliminating the oxygen from the blood by a stream of CO, the blood is found to have lost its spontaneous coagulability. Such blood becomes coagulable when a stream of CO_2 is passed through it; (*f*) strong solutions of neutral salts have a large absorbing power for

free carbonic acid. The power which these solutions have of inhibiting blood coagulation is inferred to be associated with this property; (*g*) thrombosis of the pulmonary vessels occurs in dogs when they are caused to breathe atmospheric air in which the whole nitrogen has been replaced by CO_2; (*h*) after burns, the venous blood is found to contain a great excess of carbonic acid. The coagulability of such blood is abnormally high.

It will be observed that the experiments which have been the subject-matter of the present communication entirely confirm the conclusions which had already been arrived at by entirely different methods by Mathieu and Urbain. I have not in any systematic manner controlled the observations upon which the conclusions of these observers were based. I have, however, had many incidental opportunities of confirming their observations with respect to the alterations of blood coagulability which are conditioned by raising or cooling the general body temperature. On the other hand, I have not, in the very few cases I have examined for it, observed the occurrence of pulmonary thrombosis as an effect of a simple rise of carbonic acid tension in the blood, but I have, in one striking experiment, seen a rabbit whose blood coagulability had been increased by the administration of calcium chloride die instantaneously from universal intravascular coagulation, when it was supplied with an atmosphere which was surcharged with carbonic acid.

Appendix. Selected Protocols.

The appended protocols are to be read from left to right, and then back in a zig-zag manner, following the dotted lines from right to left. In accordance with the fact that the method of coagulability determinations which was employed is an approximal and not an absolute one, two data are given for each coagulability determination. These data (longest interval during which the blood was observed to remain in a tube unclotted, and shortest interval which the blood was found to require for complete occlusion of a tube) are entered in separate columns. Where only a single entry appears on the protocols this is indicative of a lacuna in the observations. Thus, when an entry appears in the first column only (as, for instance, in the case of the second coagulability determination on the last protocol on the list), it is to be understood that the last of the coagulation tubes which were appropriated to the particular coagulability determination was found liquid when tested after the interval noted in the protocol. Similarly, when an entry appears in the second column only, as, for instance, in the second determination on the first protocol on the list, it is to be understood that all the tubes were found completely clotted, although the testing of the tubes was not deferred beyond the fifty seconds noted on the protocol.

Animal employed.	Interval between first inhalation of gas and filling of first coagulation tube.	Atmospheric air.		Interval between first inhalation of gas and filling of first coagulation tube.	Hydrogen.		Interval between first inhalation of gas and filling of first coagulation tube.	Oxygen.		Interval between first inhalation of gas and filling of first coagulation tube.	Carbonic acid 80 per cent. Oxygen 20 per cent.		Interval between first inhalation of gas and filling of first coagulation tube.	Carbonic acid.		Remarks.
		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than	
	m.	m. s.	m. s.				m.	m. s.	m. s.	m.	m. s.	m. s.	m.	m. s.	m.	
Rabbit 125.		2′ 30″	2′ 40″							4′	—	50″				
	25′	1′ 30″	2′				4′	1′ 35″	2′ 5″							
	interval of 12 min.									2′	50″	55″				
Rabbit 158.		1′	1′ 15″				—	2′	2′							
	interval of 3 min.									2′ 4′	1′ 15″ 50″	1′ 30″ 1′				
Rabbit 161.		3′ 40″	3′ 45″							6′ 10′	2′ 35″ 35″	1′				
	2′ 10′	1′ 15″ 2′ 30″	1′ 15″ 2′ 45″										1′	2′ 50″	3′	

Animal employed.	Interval between first inhalation of gas and filling of first coagulation tube.	Atmospheric air.		Interval between first inhalation of gas and filling of first coagulation tube.	Hydrogen.		Interval between first inhalation of gas and filling of first coagulation tube.	Oxygen.		Interval between first inhalation of gas and filling of first coagulation tube.	Carbonic acid 80 per cent. Oxygen 20 per cent.		Interval between first inhalation of gas and filling of first coagulation tube.	Carbonic acid.		Remarks.
		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than	
	m.	m. s.	m. s.	m.	m. s.	m. s.	m.	m. s.	m. s.	m.	m. s.	m. s.	m.	m.	m. s.	
Rab-bit 165.		3′ 30″	3′ 30″							2′	2′ 45″	3′				Intestinal valve does not close air-tight, and admits some atmospheric air at each inspiration.
	inter	al of 10 min.								3′	2′	2′ 30″				
										15′	2′	2′				
										25′	50″	1′				
							4′	1′ 40″	1′ 45″							
							10′	3′ 30″	4′							
Rab-bit 175.		5′ 30″	5′ 50″	50″	4′ 30″	4′ 30″										
	8′	2′ 45″	2′ 45″													
	15	3′	3′ 20″													
	26′	4′	4′ 30″													
										3′	—	1′				
										9′	2′	2′	3′	4′	4′ 30″	

Animal employed.	Interval between first inhalation of gas and filling of first coagulation tube.	Atmospheric air. Coagulation time longer than	Atmospheric air. Coagulation time shorter than	Interval between first inhalation of gas and filling of first coagulation tube.	Hydrogen. Coagulation time longer than	Hydrogen. Coagulation time shorter than	Interval between first inhalation of gas and filling of first coagulation tube.	Oxygen. Coagulation time longer than	Oxygen. Coagulation time shorter than	Interval between first inhalation of gas and filling of first coagulation tube.	Carbonic acid 80 per cent. Oxygen 20 per cent. Coagulation time longer than	Carbonic acid 80 per cent. Oxygen 20 per cent. Coagulation time shorter than	Interval between first inhalation of gas and filling of first coagulation tube.	Carbonic acid. Coagulation time longer than	Carbonic acid. Coagulation time shorter than	Remarks.
		m. s.	m. s.	s.	m. s.	m. s.	m.	m. s.	m s.	m.	m. s.	m. s.				
Rabbit 163.		2′ 30″	2′ 35″							1′ 5′	2′ 15″ 1′	1′ 15″				
							2′	2′ 40″	2′ 45″							
	interval of 2 min.			45″	1′ 45″	1′ 55″										
	interval of 5 min.									2′	1′ 10″	1′ 15″				
	interval of 1 min.			45″	2′	2′ 15″										
Rabbit 149.		2′ 45″	2′ 45″							1′ 5′	1′ 45″ —	2′ 1′ 15″				
Rabbit 135.		2′	2′ 15″				—	4′ 15″	5′							
	interval of 25 min.									—	1′ 25″	1 40″				
							—	4′	4′							

Animal employed.	Interval between first inhalation of gas and filling of first coagulation tube.	Atmospheric air. Coagulation time longer than	Atmospheric air. Coagulation time shorter than	Interval between first inhalation of gas and filling of first coagulation tube.	Hydrogen. Coagulation time longer than	Hydrogen. Coagulation time shorter than	Interval between first inhalation of gas and filling of first coagulation tube.	Oxygen. Coagulation time longer than	Oxygen. Coagulation time shorter than	Interval between first inhalation of gas and filling of first coagulation tube.	Carbonic acid 80 per cent. Oxygen 20 per cent. Coagulation time longer than	Carbonic acid 80 per cent. Oxygen 20 per cent. Coagulation time shorter than	Interval between first inhalation of gas and filling of first coagulation tube.	Carbonic acid. Coagulation time longer than	Carbonic acid. Coagulation time shorter than	Remarks.
	m.	m. s.	m. s.	m. s.	m. s.	m. s.				m.	m. s.	m. s.				
Dog 1.		10′ 40″	10′45″							2′	6′ 25″	6′ 30″				In this and in the next two experiments the animal was supplied with a mixture of CO_2 and air.
	7′	6′ 30″	6′ 45″							4′	2′ 50″	2′ 50″				
Dog 2.		6′ 30″	6′ 35″							4′	4′ 53″	4′ 56″				See remark to last experiment.
	interval of 6 min.									6′	3′ 25″	3′ 30″				
	12′	6′ 30″	6′ 40″	1′ 30″	5′ 30″	5′ 45″										
Dog 3.		8′ 30″	9′ 15″	1′ 30″	4′	5′ 55″										
	8′	4′ 30″	5′ 30″							4′	1′ 40″	1′ 45″				*Vide* remark to experiment with Dog 1.
										6′	2′	2′ 15″				
	10′	—	1′ 20″													
	22′	5′ 20″	5′ 20″													

Animal employed.	Interval between first inhalation of gas and filling of first coagulation tube.	Atmospheric air.		Interval between first inhalation of gas and filling of first coagulation tube.	Hydrogen.		Interval between first inhalation of gas and filling of first coagulation tube.	Oxygen.		Interval between first inhalation of gas and filling of first coagulation tube.	Carbonic acid 80 per cent. Oxygen 20 per cent.		Interval between first inhalation of gas and filling of first coagulation tube.	Carbonic acid.		Remarks.
		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than	
Dog 4.	m.	m.	m. s.	m. s.	m. s.	m. s.										
		7′ 20″	7′ 30″	1′ 30″	2′ 30″	2′ 50″										Tracheal tube accidentally compressed during first hydrogen inhalation.
	7′	6′ 30″	7′ 30″	1′	7′ 15″	8′ 5″										
	10′	4′ 30″	3′ 30″													
	16′	4′ 10″	4′													
Rabbit 176.		1′ 55″	1′ 55″	40″	2′ 15″	2′ 30″										Tracheal tube accidentally compressed during second hydrogen inhalation. Blood employed in penultimate coagulability determination was still very venous. Blood of last coagulability determination was obtained from right heart after death.
	5′	1′ 57″	2′	45″	1′ 30″	1′ 30″										
	5′	3′ 15″	3′ 15″	3′	4′ 45″	4′ 45″										

Animal employed.	Interval between first inhalation of gas and filling of first coagulation tube.	Atmospheric air.		Interval between first inhalation of gas and filling of first coagulation tube.	Hydrogen.		Interval between first inhalation of gas and filling of first coagulation tube.	Oxygen.		Interval between first inhalation of gas and filling of first coagulation tube.	Carbonic acid 80 per cent. Oxygen 20 per cent.		Interval between first inhalation of gas and filling of first coagulation tube.	Carbonic acid.		Remarks.
		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than	
Rabbit 171.	m.	m. s. 3′ 15″	m. s. 3′ 15″	s.	m. s.	m. s.	m.	m. s.	m. s.	m. s. 8′ 15′	m. s. 2′ 45″ 3′	m. s. 3′ 3′ 45″		m. s. 4′ 25″	m. s. 4′ 30″	Blood employed in the penultimate coagulability determination was semi-venous in character.
Rabbit 155		2′ 30″	2′ 40″				2′	—	1′ 55″							
	interval of 8 min.									2′ 30″	—	50″				
	15′	1′ 30″	2′				3′	1′ 45″	2′							
	interval of 12 min.									2′	50″	55″				
	interval of 30 min.			50″	2′ 30″	2′ 45″										

Animal employed.	Interval between first inhalation of gas and filling of first coagulation tube.	Atmospheric air.		Interval between first inhalation of gas and filling of first coagulation tube.	Hydrogen.		Interval between first inhalation of gas and filling of first coagulation tube.	Oxygen.		Interval between first inhalation of gas and filling of first coagulation tube.	Carbonic acid 80 per cent. Oxygen 20 per cent.		Interval between first inhalation of gas and filling of first coagulation tube.	Carbonic acid acid.		Remarks.
		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than		Coagulation time longer than	Coagulation time shorter than	
	m.	m. s.	m. s.	s.	m. s.	m. s.	m.	m. s.	m.	m.	m. s.	m. s.				
Rabbit 178.		6′ 15″	6′ 30″	40″	6′ 30″	6′ 30″										
	6′	2′ 30″	2′ 30″	40″	5′	5′ 15″										
	5′	2′ 35″	2′ 45″	40″	3′ 55″	3′ 55″										
	5′	3′ 50″	4′ 15″													
Rabbit 172										Oxygen decreasing from 100 per cent. CO_2 increasing from 0 per cent.						Expiration in this experiment took place into the reservoir (4000 c.c.) of oxygen. After the second coagulability determination was made, a stream of CO_2 was passed into the reservoir so as gradually to replace the oxygen by carbonic acid. The last determination was made with blood obtained from the heart immediately after death.
							3′	3′ 45″	4′							
							11′	5′	—							
										10′	3′ 15″	3′ 54″				
										20′	3 30″	3′ 36″				
										35′	40″	50″				
										40′	2′ 15″	2′ 30″				
										55′	4′ 15″	4′ 55″				
											Death.					
											4′ 55″	4′ 55″				