arrangement provided for a gradient of pressure which allowed for a continuous uninterrupted flow into the intervillous space. The fetal spiral arrangement suggests a similar functional adaptation which can provide for maximal fetal vascular exchange.

Based upon studies of hydrostatic conditions and maternal blood flow in rabbit uteri during pregnancy, Reynolds has elaborated a concept of uterine accommodation. In the last third of pregnancy the period of uterine growth is supplanted by a period of uterine stretching which follows upon an abrupt conversion of the conceptus from spherical to cylindrical form.[10-13] Ramsey has indicated that a transition is to be observed in the maternal vessels of the endometrium of the pregnant rhesus monkey beyond the 52nd gestational day. The coils of the arteries are paid out, corresponding to the period of uterine stretching.[14] The finding of partially uncoiled paid out spirals of placental fetal vessels in mature term placentae is consistent with these observations. The placental vessels must be subjected to the same hydrostatic conditions and mechanical stretching.

Summary. The spiral nature of the primary villus stem vessels has been described for the first time. The underlying mechanism suggests a trophic response to steroid hormones, comparable to the effect produced on vessels of the uterus and ovary. This spiral pattern provides a gradient of pressure which may slow fetal circulation through the placenta and allow for a more thorough exchange.

We are greatly indebted to Dr. S. R. M. Reynolds and Mr. Chester Reather of the Carnegie Institution of Washington, Baltimore, Md., for much valuable assistance. Dr. Reynolds has confirmed some of our observations and offered many helpful suggestions. Mr. Reather has provided us with excellent photographic material.

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11 Reynolds, S. R. M., *Am. J. Physiol.*, 1947, **148**, 77.

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17297. Effects of Intravenous Injection in Dogs of Staphylokinase and Dog Serum Fibrinolysin.*

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Utilizing methods described in previous reports,[1] we have studied the *in vivo* effects of staphylokinase and dog serum lysin (fibrinolysin, plasmin, tryptase) in dogs. Normal dog serum contains the inactive precursor, *prolysin*, of an active fibrinolytic enzyme, *lysin*, and substance(s), *antilysin*, capable of inhibiting this active lysin† Staphylokinase,[2,3] a material obtained from staphylococcal culture filtrates, is capable, *in vitro*, of activating dog prolysin to lysin. It seemed of some interest to determine the effects of this material injected intravenously, as well as those of a potent fibrinolytic enzyme solution, prepared from dog serum by fractionation at 25%

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† This terminology is used for brevity.

2 Gerheim, E. B., Ferguson, J. H., Travis, B. L., Johnston, C. L., and Boyles, P. W., PROC. SOC. EXP. BIOL. AND MED., 1948, **68**, 246.

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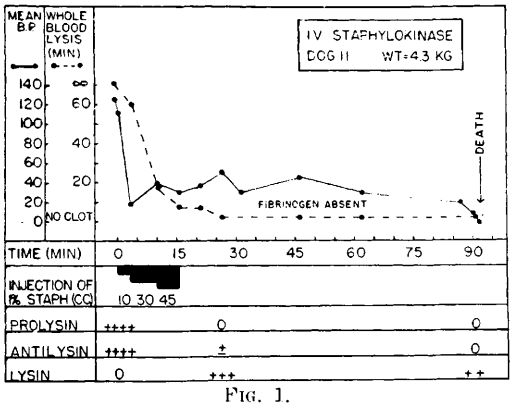

FIG. 1.

saturation with alcohol, treatment with chloroform, and aging for 6 weeks at 0°C. Due to the large amounts of these materials calculated as necessary to reproduce *in vivo* the established *in vitro* effects, we have studied only 2 dogs.

Injection of staphylokinase. 85 cc of a filtered 1% solution of partially purified staphylokinase was injected over a period of 16 minutes into a small fasting mongrel dog previously anesthetized with nembutal.

Fig. 1 summarizes the results observed in this dog. Arterial pressure fell precipitously following the injection of only a few cc. The injection was slowly continued and arterial pressure rose slightly, remaining between 30 and 60 mm Hg until the terminal fall. No whole blood lysis was observed in a pre-injection sample. After 10 cc of staphylokinase had been administered the whole blood lysis time was 60 minutes. As the injection continued, lysis times became shorter until at 10 minutes after the end of the injection lysis could not be measured as this blood did not clot. No significant changes were observed in whole blood coagulation times until this and subsequent specimens which did not clot even on addition of potent thrombin suggesting complete absence of fibrinogen probably caused by *in vivo* fibrinogenolysis.

Three serum samples were studied and results reported in Table I. The control specimen showed no active lysin, normal content of prolysin, as tested by *in vitro* activation with added staphylokinase, and normal antilysin titer. Serum obtained from blood taken 10 minutes after the injection showed marked lysin activity, no demonstrable prolysin (as indicated by the fact that the *in vitro* staphylokinase activation prolonged rather than shortened the lysis time), and markedly diminished antilysin. Immediately before death (90 min.) the serum still showed rather marked lysin content, no prolysin, and no appreciable antilysin. Prolysin times longer than lysin times were due to spontaneous loss of lysin which occurred during the 20 minute incubation at 37°C required for complete activation of prolysin by staphylokinase. In whole blood and serum samples obtained 10 minutes post injection and subsequently, a marked lipemia was noted. Microscopically, the globules were seen to stain readily with Sudan IV.

Injection of dog serum lysin. In vitro studies on the dog serum lysin showed it to be an extremely potent material containing approximately 4000 lysin units per cc. In order to allow sufficient lysin to combine with the calculated amount of circulating antilysin and leave a lysin excess detectable in the serum, we administered 1,000,000 units (250 cc of lysin solution) over a period of 11 minutes. Additional *in vitro* studies showed that this preparation of dog serum lysin contained a small amount of thrombin.

In an effort to minimize possible overloading of the heart due to the large volume of fluid introduced, we removed blood samples totalling 100 cc during the injection period.

Fig. 2 summarizes the observed findings in this 4.2 kg animal. Arterial pressure fell only gradually during and after the injection.

TABLE I.
Serum Lysin, Prolysin and Antilysin after Injection of Staphylokinase.

	Lysin[1]	Prolysin[2]	Antilysin[3]
Control	∞	3½ min.	2460 units
10 min.	8 min.	13½ "	40 "
90 "	13 "	21 "	10 "

[1] Lysis time of standard clot containing 0.5 cc serum.

[2] Lysis time of standard clot containing 0.5 cc serum after activation with staphylokinase.

[3] Calculated from lysis time of standard clot containing 0.5 cc serum previously incubated with 0.5 cc standard lysin.

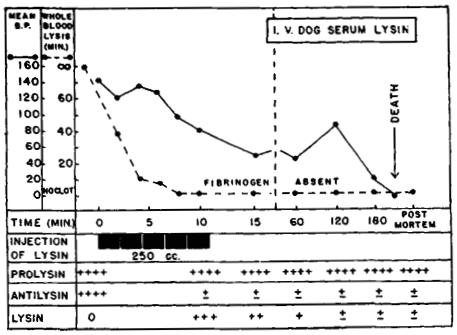

FIG. 2.

Pulse rate was noted to rise markedly during the injection. Whole blood coagulation and lysis times before, during, and following the injection are shown in Table II. The clotting times observed in the infusion period were significantly shortened at first, but after 200 cc of lysin had been introduced no clot was formed even on addition of thrombin, indicating complete fibrinogenolysis. Clot lysis times became progressively shorter until incoagulable blood was obtained.

TABLE II.
Whole Blood Coagulation Times and Lysis Times after Injection of Dog Serum Lysin.

	Clotting times (min.)	Lysis times (min.)
Control	7′	∞
Inj. after 50 cc	1′	39
" " 100 "	1′	8½
" " 150 "	4′*	6
" " 200 "	No clot	—
" " 250 "	" "	—
Min. after inj.		
5′	No clot	—
↓	↓	
180		—

* Never solid.

TABLE III.
Serum Lysin, Prolysin and Antilysin after Administration of Dog Serum Lysin.

	Lysin[1]	Prolysin[1]	Antilysin[1]
Control	∞	3¼ min.	2300 units
After 150 cc	18 min.	3½ "	300 "
" 200 "	17½ "	3½ "	290 "
" 250 "	16 "	3¼ "	230 "
Min. after inj.			
5	24 min.	3½ min.	180 units
15	42 "	3½ "	320 "
30	90 "	3½ "	180 "
60	3-12 hr	3½ "	206 "
120	3-12 "	3½ "	252 "
180	3-12 "	3½ "	320 "
Postmortem	3-12 "	3½ "	246 "

[1] See footnotes Table I.

Table III presents the serum findings in this dog. Control serum showed no active lysin, normal prolysin and normal antilysin. During and following the injection, moderate amounts of lysin appeared and gradually diminished to a trace by one hour. Prolysin was apparently not affected as no significant changes were observed on *in vitro* activation with staphylokinase. Antilysin fell dramatically but, in spite of the presence of demonstrable active lysin, the antilysin titer never reached zero. The experiment was terminated after 3½ hours. Gross postmortem examination was performed. Blood was fluid in the heart and all large vessels examined. The heart was contracted, and, on opening, showed widespread hemorrhagic areas over the endocardium, particularly of the left ventricle. The lungs were congested and showed hemorrhagic areas in both lower lobes. Small ecchymoses were scattered over the serosal surface of the stomach. Liver and spleen appeared grossly normal. No lipemia was noted in this dog.

Discussion. As might be expected from known *in vitro* reactions, both staphylokinase and serum lysin, when injected, resulted in the appearance of active lysin in the withdrawn serum. Evidence of *in vivo* proteolytic activity was seen in the appearance of incoagulable blood in which no fibrinogen could be detected. The mechanisms by which these results were obtained are obviously different. Staphylokinase activated the animal's own prolysin to lysin as indicated by the fact that the prolysin titer decreased as active lysin appeared in the serum. The antilysin titer also fell, indicating that enough active lysin was formed to destroy or combine with the circulating antilysin and still leave an excess of enzyme detectable in the serum. The injection of dog serum lysin, on the other hand, apparently did not affect the circulating prolysin, but the quantity administered was sufficient to neutralize available antilysin leaving excess lysin.

No antilysin titers of zero were obtained. This is probably due to interpretation of the results produced by this method. We are unable to obtain a serum control free from antilysin and, therefore, use simply a borate buffer control. We know that lysin is a proteolytic enzyme not specific for fibrin or fibrinogen as it also attacks casein, gelatin, etc. Other proteins in serum may exert minor nonspecific inhibitory effects by competing with the test substrate, fibrin.

Staphylokinase exerted a marked vasodepressor effect which may be associated with some other toxic material in the culture filtrate. Serum lysin showed lesser vasodepressor effects. Guest *et al.*[4] found marked vasodepressor effects following injection of their fibrinolysin[5] which was prepared, by a different method, from bovine serum. These authors found no changes in blood coagulation and did not report any lytic effects. It should be pointed out, however, that our experiment employed a much larger quantity of enzyme.

Some comment should be made on the marked shortening of coagulation time, from 7 minutes to one minute, immediately following administration of the first 50 cc of dog serum lysin. Numerous experiments, in which repeated arterial sampling from a siliconed cannula has been employed to study blood coagulation times in similarly anesthetized dogs, have frequently shown wide variations between 10 and 3½ minutes, with a tendency to become shorter in spite of repeated saline irrigations of the cannula and discarding of the first few cc of blood at each sampling. For this reason we did not feel that the slight shortenings of clotting-time observed after injection of staphylokinase were significant. On the other hand, the marked drop in clotting-time following injection of serum lysin cannot be dismissed as an experimental error. This is possibly due to the effects of small amounts of thrombin, which were present as a contaminant of this preparation of dog serum lysin.

Conclusions. 1. Staphylokinase, injected intravenously into a dog, caused appearance of active serum lysin, accompanied by a fall in prolysin and antilysin. The blood became incoagulable presumably due to lysis of the circulating fibrinogen. A precipitous fall in blood pressure was also observed.

2. Dog serum lysin, injected intravenously into a dog, also resulted in the appearance of active serum lysin, fall in antilysin, but no appreciable change in prolysin. Incoagulable blood, devoid of fibrinogen, was again obtained.

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17298. Lethal Effect of Triethylene Glycol Vapor on Air-Borne Mumps Virus and Newcastle Disease Virus.*

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Previous laboratory investigations by Robertson and his co-workers,[1,2] and by Henle and Zellat[3] demonstrated that both propylene and triethylene glycol vapor were lethal for air-borne Influenza A virus. Subsequently, Rosebury and his associates[4] showed that triethylene glycol vapor was effective against air-borne meningopneumonitis and psittacosis

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† National Institutes of Health Postdoctorate Research Fellow.

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