EFFECT ON THE HEMOGRAM AND TISSUES OF PERMANENTLY INSTALLED CATHETERS IN BLOOD VESSELS OF RUMINANTS

by

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TABLE OF CONTENTS

INTRODUCTION

Many plastic materials have been used in medical research and as surgical prostheses. Because of the physical or chemical properties of these plastics, they are especially adapted to specific uses. Some plastics, such as Teflon or Dacron, are sufficiently inert so that they produce very little chemical reaction when used as artificial body parts and indwelling catheters. The physical properties, however, differ from those of tissue and some inflammation results. Many investigations have indicated the difficulty of maintaining patency of vascular catheters due to thrombophlebitis.

It was the purpose of this experiment to develop a technique for catheterization of the arterio-venous system of ruminants by which blood samples could be taken as desired over a prolonged period from unanesthetized animals. It was desirable, also, that the technique would have little effect on blood and tissues.

The information gained from this experiment could be applied in studies pertaining to absorption from the gastro-intestinal tract. Many determinations of rumen content have been made but little is known about the amount of absorption during a period of time. Analysis of portal blood combined with blood flow rates would furnish this information.

REVIEW OF LITERATURE

Early Vascular Appliances

The idea of introducing a tube into the lumen of a vessel is not original. Carrel (5) mentioned that by 1912 paraffined glass tubes were in use in many physiological laboratories for securing an artificial circulation of blood. The same method has been applied successfully by Brewer to transfusions of blood in human beings. Carrel indicated that some years prior to his article of 1912, Abbe tried to establish a permanent anastomosis between the cut ends of an artery by means of a glass tube. Abbe presented a cat in which he had cut the abdominal aorta and had united the ends of the vessel by a glass tube. In other cases, coagulation of the blood took place after a short time. It appeared that even if a foreign body could be placed temporarily in a vessel without accident, thrombosis would occur sooner or later.

Nevertheless, Carrel (5) thought it was conceivable that thrombosis was preventable and that the conditions under which a foreign body could be used for repairing a vessel might be ascertained. He removed a part of the anterior wall of the abdominal aorta of a dog, and replaced it by a piece of rubber tubing covered with vasoline. Fifteen months after the operation, the circulation was still normal and the vessel was intact inside and outside of the rubber.

Other of Carrel's experiments were with glass and aluminum tubes which were placed in the lumen of the descending aorta.

Aluminum corroded and was not suitable, while glass caused thrombus formation or for other reasons was generally unsatisfactory.

In about 1907, Carrel (6) placed human arteries in grafts of the abdominal aorta of dogs with some success, as the dogs lived several years and were finally killed by conditions other than circulatory disturbances. The transplanted segment had about the same caliber as that of the aorta, but its wall was composed of connective tissue with no evidence of muscular or elastic tissue. Apparently, the foreign tissue protein of the inserted vessel was removed by the usual process of replacement by the connective tissue of the host animal.

London (28) developed an original technique for obtaining portal blood. This was a two-stage operation where the first stage consisted of painting the portal vein with iodine solution, and immobilizing it by suturing it to the side of the adjacent part of the inferior vena cava. Two weeks later the second stage was performed, namely suturing the cannula to the painted area on the portal vein and then wrapping it with an elongated mass of omentum. The cannula was brought out between the lower ribs through a stab wound. The cannula was made from a 16-gauge lumbar puncture needle. Dent (12) modified the London technique to a one-stage procedure by omitting the iodine painting and the immobilization of the portal vein.

The inability of other workers (Denton, 13 and Prigmore, 33) to pierce the vein consistently by this procedure prompted studies of other surgical techniques. Prigmore used brass T tubes and later modified them to a brass tube with a screw-in stem which could be attached to plastic and brought to the outside. Brass screen was fitted into the ends of the T tubes to prevent clots from closing the tube. This was unsatisfactory, and Prigmore started using plastic tubing as catheters.

Denton (13) was unable consistently to procure blood samples from the portal vein of dogs by means of a modified London cannula technique. He, therefore, tried other techniques and found that vinylite plastic tubing could be inserted directly into the portal vein through a 12-gauge hypodermic needle. The tube was then anchored to the connective tissue around the vein, and the other end exteriorized. The tubing was coated with silicone before being placed in the vein, since this procedure appeared to reduce the tendency for the blood to clot in the tubing. Denton was able to keep the cannula working for a period of six to eight weeks. At this time a clot would form on the end of the tubing and, although the dog was in a healthy condition and saline could be infused through the tubing, he was unable to withdraw blood.

Materials Employed as Vascular Appliances

Harrison (20) made a study comparing the tissue reactions to various plastic materials. One-centimeter squares of woven Nylon, Orlon, Teflon, Ivalon, and Dacron were implanted in the subcutaneous tissue of the abdominal wall of dogs. The Ivalon was held in place with surgical silk and the others by sutures of their own material. The silk sutures were the most irritating of any of the materials. Gross and histological observations were made with the degree of tissue reaction in the following order: Nylon, Dacron, Ivalon, Orlon, and Teflon. Chemical stability also followed the same order, with Nylon losing 83 per cent of its tensile strength after six months of implantation. Teflon was non-reactive and did not lose strength.

Finely-ground Nylon, Celluloid, Lucite, and Teflon were placed in the abdominal cavities of dogs by Leveen (26). The degrees of inflammatory changes noted were in the following order: Celluloid, Lucite, Nylon, and Teflon. Protein material adsorbed to the surface of irritating material and marked inflammation followed.

Harrison (20) indicated that the reaction to a foreign material depends upon its chemical composition, physical properties, and the area of surface coming into contact with surrounding tissue. The degree of breakdown when subjected to body fluids and the surface tension properties are important. Plasticisers and colors used in the manufacture of plastics migrate into the surrounding environment and may produce intense tissue reactions.

Since the demonstration in 1952 by Voorhees et al. (41) that synthetic fabric prostheses could function satisfactorily in the arterial system, there has been an increasing interest in synthetic replacements. After a period of trial and error, several types of commercially available synthetic replacements have emerged.

Methods of Vascular Catheterization

A major step toward the widespread use of vascular catheters was made when Forssman (18), in 1929, catheterized the right auricle on himself after exposure of a vein of the arm by a surgeon. From 1930 to 1939 this technique of catheterizing the right heart was widely used in Europe for injecting contrast substances in order to visualize the right chambers of the heart and the pulmonary vascular tree.

Cournand and Ranges (10) modified the Forssman technique when they used a No. 8 flexible, X-ray opaque, varnished silk catheter with two holes, one at the rounded tip and another about 1 cm. from the tip. The catheter was inserted through a 10-gauge Lindeman type needle and was continually flushed by 15 drops of saline per minute. The passage of the catheter through the vein was accomplished while the patient was on a fluoroscopic table.

Vascular catheters have been widely used in clinical and experimental medicine. The variety of materials used and the applications for vascular catheterization are numerous. Myers (32) introduced vinylite plastic into the veins of human arms through a 17-gauge needle. The tubing was anchored to the arms with adhesive tape. Some of the tubes were kept open for as long as 39 days.

Zimmerman (43), while working with dogs, passed a plastic tube through a 15-gauge needle. The tube extended into the vein 4-5 cm. In 11 dogs the tubes remained in the external jugular veins for four to five weeks without serious developments. Thrombosis formed around the tube in some of these dogs.

A different method of using a needle for insertions was employed by Dotter (14). Polyethylene tubing of large bore was inserted into the veins by stretching the tube, thus causing it to fit tightly around the needle. The needle was guided into the vein, and the tube then passed over the needle and into the vein. By inserting the needle several inches from the end of the tube and then withdrawing the needle from the tube after veinipuncture, a long tube may be used. The long tube can then be used to enter the heart for radiographic techniques.

A similar method of sliding a tube over a needle was employed by Cope (9). His method required a length of polyethylene catheter and a needle somewhat longer than the catheter but of a diameter 1 to 2 gauges smaller than the internal diameter of the catheter tubing. With the needle lying within the catheter, one end of the catheter was narrowed by stretching until it fit very snugly over the needle shaft. Skin incisions preceded the installation of the catheter.

Cresson (11) inserted polyethylene tubing of .023 inches diameter into the portal veins of dogs. He used a retroperitoneal approach to the portal vein. The catheter was inserted into the portal vein through a 16-gauge needle. The needle was then removed and the vein tightened around the catheter by means of a purse string suture.

A method was described by Jungblut (24) for the introduction of a polyvinyl catheter into the portal vein of dogs. The catheter was introduced into one of the mesenteric veins and pushed through the vena mesenterica communis until the portal vein was reached. The catheter was anchored to the mesentery internally and was fixed to the skin exteriorly. Lewis (27) used this same method for the introduction of polyethylene into the portal vein of sheep.

Polyvinyl chloride tubing was inserted into the external jugular vein of dairy cows by Ralston (35). The polyvinyl tube was 2.2 mm. outside diameter and was passed into the jugular vein through a 10- or 12-gauge needle. The length of time a catheter remained open varied from 2.1 hours to 14.5 days. Ralston indicated that bleeding caused very little disturbance in the cows.

Jackson (22) catheterized the hepatic vein, portal vein, renal vein, and renal artery of sheep with polyethylene tubing. The tubing was passed through the body wall by threading the outer end through a 14-gauge needle. The vein was penetrated with the point of a suture needle and the beveled end of a catheter was passed through the opening. The small puncture allowed the catheter to fit snugly with very little hemorrhage. The venous end of the catheter was anchored near the vein by means of silk sutures. Jackson indicated that blood samples may be obtained as long as seven days after surgery.

Meschia (31) threaded polyvinyl catheters through a small easily accessible tributary into the uterine arteries and uterine veins of sheep. He maintained patency in arterial catheters for nine months and for three months in venous catheters. Apparently he had little difficulty from clotting, but thrombosis was induced where the endothetial lining near the tip of the catheter was damaged. His success may be attributed to the increase in the diameter of vessels during pregnancy and/or the varying physiologic state during the estrous cycle.

Warren (42) obtained blood directly from the hepatic vein of man by passing a uretheral catheter into an arm vein and guiding the end through the auricle of the heart and out into the inferior vena cava. Fluoroscopic guidance was used to guide the tip of the catheter.

Venous Catheters as Research Aids

Blood flow rate is frequently studied by means of vascular catheters. Conrad (8) studied gastrosplenic blood flow by means of erythrocytes tagged with P^{32} which were injected into the celiac artery and collected from the gastrosplenic vein which drains the stomach and spleen. The mean rate of gastric blood flow was calculated to be 764 ml. per minute for 100 pounds of body weight.

Bensadoun (4), Fegler (17), and others have studied gastric blood flow rate of sheep by passing a temperature sensing unit into the portal vein and determining the temperature change of blood due to injecting a known quantity of cold fluid into a tributary.

Frieden (19) made a study of cardiac output, using a comparison of blood obtained from the pulmonary artery and from the femoral artery by means of a catheter. Respiratory and volume flow studies were made by Cournand (10) from blood removed from the right auricle of man.

A study was made by Huckabee (21) on the effect of altitude upon uterine blood flow rate of pregnant sheep. Blood samples were obtained through small catheters placed in the uterine vessels. Meschia (31) studied carbon dioxide levels of uterine blood of sheep by means of vascular catheters.

Rhode (37) made use of polyvinyl chloride and polyethylene catheters for prolonged metabolic studies. The catheters were threaded into the jugular vein until the tip was in the superior vena cava. Nutrient solutions were fed continuously through the tube for as long as 141 days. Isotonic solutions were satisfactory but hypertonic solutions precipitated in the continuous feed mechanism and caused equipment difficulty.

Transflex plastic tubing was inserted into the jugular vein through a 12-gauge needle and used for supportive treatment following Caesarean section of a bovine by Tharp (40).

Siliconed polyethylene tubing was used by Rappaport (36) to administer prolonged continuous intravenous infusions to 38 human patients. The main complication was a local induration of the vein which, in 12 cases, gave rise to a phlebitis of the arm without systemic reaction. The cannula was kept open by constant flow into the vein.

Lewis (27) studied absorption of ammonia from the rumen of sheep by use of polyethylene catheters inserted into the portal vein. These experiments showed that in conscious sheep, ammonia was absorbed from the rumen into the portal system and thus confirmed the original observations of McDonald (30) which were accomplished with anesthetized sheep.

A study was made by Schambye (39) of fatty acids and other constituents in the portal blood of sheep during the digestive cycle. The differences between carotid and portal blood in respect to volatile fatty acids and glucose were determined. Blood samples were taken from various portions of the gastrointestinal tract of anesthetized sheep by McAnally (29) and analyzed for volatile fatty acids. Venous blood from the rumen and reticulum was the highest in volatile fatty acids. Barcroft (3) used a similar experiment to show that fatty acids were absorbed chiefly from the rumen and reticulum of sheep. Somewhat less absorption occurred from the abomasum. Barcroft also found some volatile fatty acids to be absorbed from the caecum and colon of pigs, the caecum of rabbits, and from the colon of horses.

Annison (2) studied the absorption of various fatty acids from the rumen of sheep and their effect on ketone body production. Administration of butyric acid resulted initially in high ketone body levels in portal blood, followed by a steady rise of concentration in extrahepatic peripheral blood.

Ralston (35) and Kleiber (25) used radioactive P^{32} to study phosphorus metabolism of dairy cows. It was found necessary to develop a suitable injection and bleeding technique which would fulfill the following requirements: (a) It must permit complete injection of the radioactive material for hazard control; (b) it must provide for accurate and rapid injection of the radioactive material, followed immediately with the withdrawal of blood samples; (c) it must allow for bleeding at frequent intervals for long periods; and (d) it must minimize the disturbance and apprehension of the cow. Polyvinyl chloride catheters inserted into the external jugular veins met the above requirements and made the work possible.

Vascular catheters have been proven very useful in diagnostic and experimental medicine but unfortunately are not without serious inherent dangers. Most workers agree with Rust (38), Anderson (1), and Cresson (11) that thrombus formation is a distinct danger. It has even been suggested that vascular catheters could be useful in experimental production of thrombi for research purposes. Cresson (11) found that, although adequate specimens of blood were aspirated for prolonged periods in 37.5 per cent of his experiments, some degree of reaction to the polyethylene tubing was noted in every case where the portal vein was examined. The first reaction to the polyethylene tubing was the formation of a thrombus surrounding the intraluminal portion of the tubing. Later, fibrous connective tissue was laid down over the tube with puckering of the adjacent intimal lining. These events took place in from two to four weeks after the tubing was inserted into the lumen of the vein. He indicated that in one instance the tubing was completely excluded from the vein lumen by fibrous tissue at three weeks.

MATERIALS AND METHODS

Materials and Surgical Procedures

Preliminary experiments for the development of catheterization technique were conducted on dogs in which Teflon tubing of outside diameter .068 inches was surgically implanted as an arterio-venous shunt. One end of a Teflon tube was inserted proximally into the femoral vein,and the other end was inserted proximally into the femoral artery. It was thought that the drop in blood pressure from arterial to venous pressure would keep a constant flow of blood passing through the tube, thus keeping the tube open. Substantial bandaging or a surgical procedure which buried the entire tube below the skin was found necessary to prevent the dogs from molesting the tubes.

Dairy breed bull calves, approximately one month of age, were employed for the major portion of the experiment. Teflon tubes of .068 inch outside diameter, polyvinyl tubes of .088 inch outside diameter, and polyethylene tubes of .067 and .110 inch outside diameter were inserted into the veins of these calves.

Several different methods of installing the catheters in the calves were used. The method employed for catheterizing small veins was to cleanse surgically the operative area, and locally infiltrate with procaine solution. An incision of three-fourths inch was made over the vein, and the vein was isolated from the surrounding tissue. A catgut ligature was placed around the distal portion of the isolated vein. A second catgut ligature was placed around the proximal portion of the exposed vein, but left loose for passage of the tube. A small opening was made in the vein through which the plastic tubing was passed into the vein and threaded proximally for the desired distance. After proper location of the tube in the vein, the upper catgut ligature was pulled tight around the vein and tube, thus making a seal around the tube. Skin closure was accomplished by means of interrupted sutures. A blunt hypodermic needle was then inserted into the external tip of the tube and both the tube and the needle anchored to the skin by means of sutures (Plate I, Fig. 1). Heparin, 3 mg. per cc., was injected through the needle and tube. The needle was then closed with a removable plug. The exposed tube and needle were dusted lightly with urea sulfonamide powder, and a protective bandage was applied to the area.

Another method was employed for inserting the plastic tubes into large veins such as the jugular vein. The area was surgically cleansed and anesthetized by procaine infiltration. The skin and subcutaneous tissue were incised for three-fourths inch to a depth which nearly exposed the vein. A 10-gauge needle was

EXPLANATION OF PLATE I

Fig. 1. Left rear leg of calf No. 6 with cannula inserted upward into the femoral vein, illustrating the needle and plug in place.

Fig. 2. Right side of head and neck of calf No. 6 with cannula inserted in the jugular vein, illustrating the needle and plug in place.

PLATE I

Fig. 1

Fig. 2

then inserted into the vein, and the plastic tube was threaded through the lumen of the needle. The needle was withdrawn, leaving the end of the tube inserted into the vein. The tube was located the desired distance into the lumen of the vein, and the skin closed with interrupted sutures. A blunt needle was then inserted into the external end of the tube (Plate I, Fig. 2). The tube and needle were anchored to the skin, flushed with heparin solution, and closed with a removable plug.

A second method which was employed for inserting plastic tubes into large veins required a length of plastic tubing and a needle somewhat longer than the catheter, but of a diameter one or two gauges smaller than the internal diameter of the catheter tubing. With the 18-gauge needle lying within the catheter, one end of the polyethylene tubing, .067 inch outside diameter, was narrowed by stretching until it fit very snugly over the needle shaft. A skin incision preceded the installation of the catheter as the plastic tubing would tear rather than pass through the intact skin with the needle. The external end of the catheter was then closed and anchored as in the above method.

A suspension containing 400,000 units procaine penicillin and .5 gm. dihydrostreptomycin was injected intramuscularly daily for the first four postoperative days in all the calves. Blood samples were drawn from the catheters at 24-hour intervals. Following blood samplings, the catheters were flushed with heparin solution, 3 mg. per cc., and the protective bandages were replaced. The ration fed the calves consisted of commercial milk replacer and a commercial grain mixture. Alfalfa hay was available after the calves were two weeks of age. The calves had free access to block salt and fresh water.

Experimental Procedure

It was originally thought that the inability to draw blood from the catheters for prolonged periods of time was due to closure of the tube. In order to test this belief, polyethylene was melted over the end of a piece of piano wire to form a plug that would just pass through the lumen of Teflon tubing. By careful measurement of the tube and wire, it was possible to locate the polyethylene plug at the desired position inside the Teflon tubing and remove it as desired (Plate II, Fig. 1). The Teflon catheter was then installed in the vein, and the plug passed through the catheter until one-eighth inch of plug extended beyond the end of the tube. The plug was removed at 24-hour intervals for blood sampling and flushing with heparin. This technique was abandoned because it did not prolong the period of patency.

Many workers have observed that injections could be made through vascular catheters after blood sampling was no longer possible. This was thought to be due to thrombus formation in the lumen of the vein, which served as a valve when negative pressure was applied for blood sampling. It was, therefore, thought that blood might be drawn if the thrombus could be bypassed. Small polyethylene tubes of .038 inch outside diameter were passed

EXPLANATION OF PLATE II

Fig. 1. Polyethylene plug on piano wire of length to place the end of plug 1/8 inch past the end of Teflon tube. The upper cannula with polyethylene plug and wire in place illustrates the end of the plug extending beyond the end of the cannula.

Fig. 2. Calf No. 6 one week after cannulas were placed in jugular and femoral veins. Cannulation had little effect on the health of the calf.

PLATE II

Fig. 1

Fig. 2

through the vascular catheter and into the vein beyond the end of the catheter. The use of a piano wire stylet inside the small polyethylene tube was a very useful aid for the passage of the small tube through the catheter and thrombus.

Varizyme, a commercially available enzyme complex consisting of streptokinase, 33,000 units; streptodornase, 8,300 units; and human plasminogen;* were injected at 24-hour intervals through the catheters in calves Nos. 0 and 3. The tubes were left filled with the enzyme complex from one infusion until the next. Heparin was not flushed through these catheters during the time of enzyme treatment.

Blood samples were taken from the jugular vein of the calves at 24-hour intervals for laboratory examination. Clotting time was determined by means of glass capillary tubes which were filled with the fresh blood. Small portions of the capillary tube were broken off at half-minute intervals until signs of clot formation were evident. Hematocrit determinations were made from oxalated jugular blood by means of capillary tubes, which were then centrifuged at 12,500 r.p.m. for five minutes. A reading of the packed cell volume was then made directly from the capillary tube. Differential white blood cell counts were made from oxalated jugular blood which was stained with a modified Wright's stain.**

At the termination of the experiments, a portion of the vein with the catheter in place was removed for histological

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* Compliments of American Cyanamid Company, New York, N.Y.

** C. L. stain, obtained from C. W. Alban Co., 1675 West Grand, St. Louis, Missouri.

study. Cross sections of 10 micron thickness were made through the vein and tube. Useful slides were difficult to produce as the plastic tubing became loosened and floated free during the staining process.

RESULTS

Results of Catheterization

A review of the properties of the various plastics indicated that Teflon was very inert chemically. For this reason it appeared that Teflon would not hasten blood clotting and might be desirable catheter tubing. Teflon was, therefore, used in the preliminary work with dogs and later with calves.

Teflon tubing, employed as arterio-venous shunts from the femoral artery to the femoral vein of dogs, caused a slight transient edema of the foot distal to the shunt in a few cases but was not associated with any significant systemic effect. The tubes could not be maintained in a patent condition, however, because of the stiffness of the Teflon tubing, which bent sharply and stopped the flow of blood. This procedure in dogs was not attempted with softer tubing materials which would not easily kink. It is possible, therefore, that more pliable plastic tubing would be more satisfactory.

The results obtained with Teflon tubing inserted into the femoral and the jugular veins of calves were not considered satisfactory as blood could not be drawn from the catheter for sufficiently long periods, as shown in Table 1. Polyethylene

Table 1. Plastic tubing material employed, and period of patency.

Tube material	Calf number	Tubing size inches	Place of insertion	Length of time open to negative pressure (days)	Length of time open to passage of small tube (days)
Teflon	2	.068	Left jugular	1	3
Teflon	3*	.068	Left jugular	2	5
Teflon	0*	.068	Right femoral	1	3
Teflon	0*	.068	Left femoral	1	3
Average time, open Teflon				1.25	3.5
Polyethylene	1	.067	Left femoral	3	4
Polyethylene	1	.067	Right femoral	2	10
Polyethylene	2	.067	Right jugular	2	4
Polyethylene	3*	.067	Right jugular	2	4
Polyethylene	5	.110	Right femoral	1	5**
Average time, open polyethylene				2.0	5.5
Polyvinyl	1	.088	Left jugular	3	8
Polyvinyl	1	.088	Right jugular	6	18
Polyvinyl	4	.088	Right jugular	6	16
Polyvinyl	4	.088	Left femoral	3	14
Polyvinyl	5	.088	Right jugular	4	5**
Polyvinyl	5	.088	Left femoral	2	5**
Polyvinyl	6	.088	Left femoral	1	23
Polyvinyl	6	.088	Right jugular	8	22
Average time, open polyvinyl				4.1	16.8

* Calves Nos. 0 and 3 received Varizyme.

** Calf died on fifth day with all tubes open.

plugs inserted to the internal extremity of the Teflon tubing by means of the attached piano wire (Plate II, Fig. 1) assured that the catheter was open throughout its entire length, but again the results were considered unsatisfactory. This would indicate that the lumen of the catheter does not close when flushed with heparin solution at 24-hour intervals. The source of trouble, therefore, must be on the inside of the vein but on the outside of the catheter.

Polyethylene tubing gave somewhat better results (Table 1), but the period of patency was again relatively short so that the results were considered unsatisfactory for practical application. Since the more pliable polyethylene allowed blood sampling for longer periods than Teflon, it was desirable to try other tubing materials which were more pliable than polyethylene.

Polyvinyl tubing was, therefore, obtained and used as venous catheter material, and proved to be the most satisfactory of the three catheter materials employed (Table 1). Polyvinyl was relatively soft and could be bent or twisted with relatively little tendency to collapse and close the lumen. Apparently, there was some chemical reaction between the tissue and polyvinyl since the tubing became white in color and somewhat larger after prolonged presence in a vein. This did not, however, cause apparent systemic reaction in calves during the duration of this experiment, as evidenced by Plate II, Fig. 2. The presence of any of the plastics used in the veins apparently was not associated with systemic reaction. A slight inflammation of no consequence occurred in the area adjacent to the tube.

Prigmore (33) indicated that injections could be made through the catheters for longer periods than blood could be drawn from the catheter. In the present experiment, heparin solution could be injected easily into a vein through a catheter from which blood could not be withdrawn by applying negative pressure to the catheter. Thrombus formation around the catheter possibly served as a valve which closed the end of the catheter when blood sampling was attempted.

The period during which blood samples could be obtained was at least doubled by repeatedly passing a small diameter polyethylene tube through the lumen of the catheter and into the vein beyond the possible thrombus formation. The data of Table 1 would suggest an even greater advantage in the polyvinyl catheters, partially due to improved technique, since polyvinyl was the last material used.

The infusion of the streptokinase, human plasminogen, and streptodornase complex through the catheters apparently was without effect on the calves. The period of patency of the catheters evidently was not prolonged by the use of these fibrinolytic agents.

Results of Blood and Tissue Study

Blood samples were obtained at 24-hour intervals from the jugular vein. Blood from the indwelling catheters was not used because of the probability that the heparin present in the tube would alter clotting time. The results of the clotting time determinations indicated wide variations without definite pattern. No explanation for the differences of clotting time was determined; however, variations of temperature could have been a factor. The average clotting time for the calves was 7.8 minutes, which would compare with 6.5 minutes for ox blood, as listed by Dukes (15).

Hematocrit readings were made at 24-hour intervals, using freshly drawn oxalated jugular blood. The average hematocrit reading for adult dairy cows, as determined by Jones et al. (23), was 35.08 per cent as compared to the 30.4 per cent of this experiment (Table 2). This difference possibly could be attributed to the younger age of the calves employed for this experiment. The packed cell volume tends to increase as the animal matures.

The results of the differential white blood cell counts were inconclusive in so far as the vascular catheter work was concerned. Some of the calves developed scours during the experiment and one calf could not be fed normally. For this reason little significance should be placed on the results of the differential count. The average readings for the calves were: lymphocytes, 68.7 per cent; neutrophils, 30.1 per cent; basophils, 0.1 per cent; eosinophils, 0.3 per cent; and monocytes, 0.8 per cent. Coffin (7) has given the differential white cell count of cattle blood as: lymphocytes, 63 per cent; neutrophils, 23 per cent; basophils, 1 per cent; eosinophils, 8 per cent; and monocytes, 6 per cent. The hematological work is summarized in Table 2.

It became evident early in the experiment that thrombus formation was a limiting factor in the use of vascular catheters. Gross and microscopic studies were made of the veins following the completion of the experiment. The apparent cause for the

Table 2. Clotting time, hematocrit, and differential white blood cell count of calves.

Calf No.	No. of readings	Hemato-crit	Clotting time	Differential white blood cell count					
				Lympho-cytes	Neutro-phils	Eosino-phils	Baso-phils	Mono-cytes	
1	20	25.0	5.0	55	21	0	0	0	Low
		37.35	13.25	77	42	2	1	3	High
		30.9	8.7	68.2	30.7	0.2		0.9	Average
2	4	34.8	5.5	65	27	0	0	0	Low
		36.2	6.0	72	36	1	0	1	High
		35.5	5.8	67.8	31.5	0.2	0	0.5	Average
3	6	29.7	4.75	65	18	0	0	0	Low
		33.6	10.5	79	35	1	1	3	High
		31.3	7.2	70.1	28.3	0.2	0.2	1.2	Average
4	12	24.0	4.5	57	20	0	0	0	Low
		31.6	14.5	80	42	2	1	2	High
		28.0	9.0	71.4	27.5	0.3	0.2	0.6	Average
5	2	37.5	8.5	65	32	0	0	1	Low
		43.2	8.5	67	33	1	0	1	High
		40.3	8.5	66	32.5	0.5	0	1	Average
Average		33.2	7.8	68.7	30.1	0.3	0.1	0.8	

inability to draw blood for prolonged periods was thrombophlebitis. A photomicrograph of a portion of a cross section of the femoral vein with Teflon tubing in place (Plate III) was representative of the tissue sections which were studied. The endothelium of the vein was roughened and showed attachment of the thrombus.

DISCUSSION

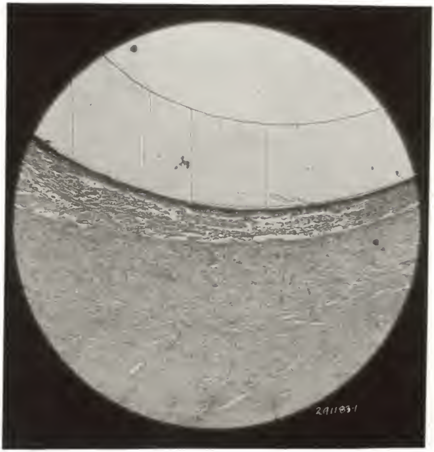
Vascular catheters have been employed as an adjunct to research work but their usefulness has been limited by the short period of patency. Teflon, polyethylene, and polyvinyl tubings were employed in this experiment as venous catheters in an attempt to develop a technique by which blood could be drawn for prolonged periods. Several methods of introducing the tubing into veins were employed. The method of introduction apparently was not a limiting factor in the period of usefulness of a venous catheter. Surgical trauma to the vessel during the catheterization operation appeared to have a direct effect upon the period in which blood could be drawn. Of the three tubing materials employed in this experiment, the polyvinyl which was the most pliable plastic, stayed open the longest, while Teflon which was the most rigid tubing used, was the least satisfactory.

Other workers have indicated that thrombus formation was the limiting factor in the usefulness of vascular catheters. In a discussion of the mechanism of thrombosis, Quick (34) wrote:

EXPLANATION OF PLATE III

Section of portion of femoral vein with Teflon tube in place eight days postoperative. The space between the Teflon tubing and the vein has filled with thrombus which was attached to the inside of the vein.

PLATE III



> It can be stated with a fair degree of certainty that no coagulation occurs in the intact vascular system provided there is no break in the endothelial lining. If this statement is accepted, it naturally follows that a change somewhere in the endothelium layer of the venous system is the first and essential step in the train of events which eventually may culminate in a massive thrombus.

The insertion of any material into the lumen of a vein would, therefore, be a predisposing factor for thrombus formation. The more rigid tubing would have a tendency to cause more abrasion to the endothelium of the vein than a more pliable tubing which would bend readily as the animal moved. The subsequent thrombus formation would, therefore, be expected to be more severe with Teflon tubing than with polyvinyl tubing, as was evidenced by this experiment. Table 1 indicates there was a relationship between the stiffness of the tubes and the period of patency.

The gross and histological studies of the veins with indwelling catheters also showed the presence of thrombus formation. A photomicrograph of a cross section of the femoral vein with Teflon catheter in place (Plate III), shows a large area of attachment between the thrombus and endothelium of the vein. This would indicate that the endothelium of the vein had been irritated and the process of thrombophlebitis was initiated.

It became obvious early in the experiment that injections could be made through a cannula for longer periods than blood samples could be obtained. It was thought that early thrombus formation could act as a valve and stop the return of blood from the cannula. Quick (34) made a schematic drawing of the mechanism of thrombus formation (Plate IV), which illustrates the probable sequence of events inside the veins. This shows the thrombus to extend downstream from the point of attachment and would explain how the free end could pull over the tube when negative pressure is applied, and thus prevent blood sampling. Plate V illustrates the probable sequence in the development of thrombosis following catheterization.

The use of polyethylene plugs in the lumen of Teflon catheters did not prolong the blood sampling period, but it indicated that the reason for failure was not blood clots in the lumen of the catheter. The idea that thrombus formation was the limiting factor was further proven by the use of the polyethylene plug and attached wire. The wire further stiffened the Teflon tube and thus tended to make the thrombophlebitis more severe.

Infusing solutions of streptokinase, streptodornase, and human plasminogen through the catheter did not prove beneficial in limiting thrombus formation. Further use of fibrinolyzing enzymes should, however, be considered for future experiments. Heparin was employed to keep the lumen of the catheter open but was not given in sufficiently large quantities for systemic anticoagulant action. Dicumarol was not used in this experiment but would warrant consideration.

The blood work conducted during the course of the experiment was inconclusive because of the small number and the condition of the calves during the test period. An interesting addition to the blood work would have been a thrombocyte count. Some

EXPLANATION OF PLATE IV

The probable sequence in the formation of a venous thrombus.*

Fig. 1. Injury of vessel wall, adherence of platelets to local area of trauma.

Fig. 2. Disintegration of platelets. Formation of a reticulated fibrin clot enmeshing intact platelets.

Fig. 3. Retraction of primary clot, thereby expressing serum rich in nascent thrombin.

Fig. 4. Formation of a new fibrin coagulum attached to the primary thrombus.

Fig. 5. Growth of thrombus by successive formation of new clots, and repeated elaboration of nascent thrombin by clot retraction.

* Quick - The Physiology and Pathology of Hematosis, page 85.

PLATE IV

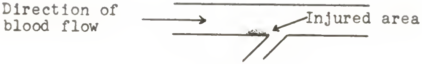


Fig. 1

Fig. 2

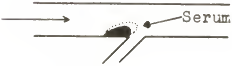


Fig. 3

Fig. 4

Fig. 5

EXPLANATION OF PLATE V

The probable sequence in the formation of a venous thrombus with indwelling catheter.

Fig. 1. Injured areas of vessel wall caused by mechanical irritation from the indwelling catheter.

Fig. 2. Platelets adhere to the injured areas and disintegrate. The fibrin clot then forms and enmeshes more intact platelets.

Fig. 3. The fibrin clot retracts and expresses thrombin which aids the further thrombus development. The relatively large area of attachment of the thrombus prevents fragments from dislodging and passing down the vein as emboli.

Fig. 4. The thrombus develops downstream from the area of injury and could act as a flutter valve over the end of an indwelling catheter.

Fig. 5. Thrombus growth occurs by repeated clot formation.

PLATE V

Fig. 1

Fig. 2

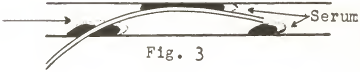


Fig. 3

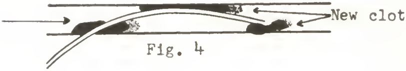


Fig. 4

Fig. 5

researchers who have studied thrombosis in humans have associated an increase of thrombocytes in the circulating blood with the development of thrombosis.

If catheterization is to be used as a means of long-term blood sampling, methods must be found to delay thrombus formation and to draw blood even though some thrombus formation has already developed. Progress has been made in this direction on two fronts. First, it has been shown that thrombus formation occurred less rapidly when soft, flexible tubing of chemically inert material was used as the catheter. Secondly, the insertion of a small tube stiffened by a piano wire stylet through the lumen of the catheter provided a means of breaking through the thrombus and securing blood samples for a considerably longer time than was otherwise possible. Further improvements in surgical technic, catheter material, and use of desirable anticoagulants should result in still greater extensions of functional collecting time.

SUMMARY

Teflon, polyethylene, and polyvinyl tubings of small diameter were employed as venous catheters in dairy breed bull calves in an attempt to develop a technique whereby blood samples could be drawn for prolonged periods. Polyvinyl tubing proved to be the most satisfactory tubing tested.

Gross and histological studies of the veins with catheters in place indicated the cause for the inability to draw blood for

prolonged periods to be due to thrombophlebitis. The more rigid tubing materials apparently irritated the endothelium of the vein more than the softer polyvinyl, and in this way caused a more rapid and severe thrombophlebitis. The developing thrombus apparently acted as a valve which prevented the drawing of blood before closure of the tube was complete enough to prevent injections through the cannula. Polyethylene tubing of small diameter could be passed through the lumen of the cannula and blood drawn from the vein beyond the thrombus after blood sampling was otherwise impossible.

The greatest problem to overcome in the use of venous catheters appears to be thrombophlebitis. This could best be prevented by gentle surgical installation of a catheter material which was soft, pliable, and chemically inert. In this way the irritation to the endothelium of the vein would be reduced to a minimum, which would reduce the tendency for thrombus formation.

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EFFECT ON THE HEMOGRAM AND TISSUES OF PERMANENTLY INSTALLED CATHETERS IN BLOOD VESSELS OF RUMINANTS

by

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Experiments were conducted to develop a technique for vascular catheterization of ruminants. Dairy breed bull calves approximately one month of age were employed for the experiment. Teflon, polyethylene, and polyvinyl plastic tubes were inserted into the femoral and jugular veins in an attempt to determine a method whereby blood could be drawn for prolonged periods.

Polyvinyl, which was the softest tubing material employed, proved to be the most satisfactory. Blood samples could be taken for the shortest time from the Teflon tubing, which was the most rigid plastic employed in the experiment.

A study was made of the gross and histological changes in the veins with the indwelling catheters. Thrombophlebitis was the apparent cause for the catheters to become nonfunctional. The development of thrombophlebitis apparently was due to irritation of the endothelium of the vein as a result of the surgical installation and subsequent abrasion by the catheter. Varizyme did not prove successful as a fibrinolyzing agent to prevent the development of thrombus formation.

It was noted early in the experiment that injections could be made through a catheter for a longer period of time than blood samples could be drawn. This apparently was due to the valve-like action of the developing thrombus in the lumen of the vein. The length of time during which blood could be drawn was at least doubled by passing a small gauge polyethylene tube through the lumen of the catheter and extending it past the thrombus.

The experimental results indicated that the period of patency was related to the rigidity of the catheter material. The catheter material employed, therefore, has much to do with the expected period of patency. The ideal plastic tubing should be as pliable as possible without collapsing, and as nearly chemically inert as is consistent with pliability.