

THE FALLOPIAN TUBE OF THE SHEEP

I. CANNULATION OF THE FALLOPIAN TUBE

By B. J. RESTALL*

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Summary

Successful long-term cannulation of the fallopian tube was achieved by the use of silicone rubber cannulae. Success was aided by removal of the causes of irritation to the animal and care in the prevention of bacterial contamination. The technique described produced no change in the morphology of the fallopian tube.

A suitable fluid-collecting system for studying the secretion rate of the fallopian tube has been described.

I. INTRODUCTION

At the present time the role of the fluids from the female genital tract in reproduction is receiving increasing attention. Investigators have used techniques for fluid collection which included ligation of portions of the genital tract (e.g. Blandau, Jensen, and Rumery 1958), flushing portions of the genital tract (Heap 1962), stripping tracts after slaughter (Olds and Van Demark 1957), and cannulation of portion of the tract in viable animals (e.g. Clewe and Mastroianni 1960). This latter technique appears to be the most desirable and this paper presents the results of a series of attempts to cannulate the fallopian tubes of sheep.

II. MATERIALS AND METHODS

(a) Animals

Mature Merino ewes in which one oestrous cycle at least had been detected prior to operation were used.

(b) Operative Procedure

Operations were carried out under strict aseptic conditions. The ewes were anaesthetized with an intravenous injection of a 5% solution of sodium thiopentone and the anaesthesia maintained by intermittent injections as required. The uterus, oviducts, and ovaries were exposed through a mid-ventral incision anterior to the udder and to one side of the mid-line. The reproductive organs were displayed on sterile gauze pads and the fallopian tubes were located. A cannula was inserted into the ovarian end of each tube to a depth of 1 cm and held in place by two atraumatic fine silk sutures. The uterine ends of the tubes, just above the utero-tubal junction, were doubly ligated with fine silk sutures and transected. Care was taken not to interfere with any blood vessels.

*School of Wool Technology, University of New South Wales; present address: Department of Veterinary Physiology, University of Sydney.

The cannulae were exteriorized by one of two methods. In the first method, subcutaneous channels, one on each side, were made with a probe from the mid-ventral incision out to the flank area. The cannulae were passed along these channels and exteriorized at the flank. In the second method, the cannulae were passed from the abdominal cavity to the exterior through stab wounds in the flank. Before being closed the incisions were treated with an antibiotic solution and post-operatively the animals were given two injections 1 week apart of 1.2 million units of a broad-spectrum antibiotic. The incisions were closed in the usual manner.

(c) Collection of Tubal Fluid

The tubal fluid-collection system consisted of a transparent polythene tube (int. diam. 5.0 mm) graduated in 0.1 ml units and stoppered at one end by an absorbent cotton plug impregnated with an antibiotic. The cannulae were connected to the open end of the tube by a plastic syringe connector. This collecting system was attached to the animal's side in a curved position by either:

- (1) suturing it directly to the skin;
- (2) attaching it to a coat worn by the animal; or
- (3) attaching the polythene tube to a rubber backing and attaching the rubber to the wool with a contact adhesive (Plate 1, Fig. 1).

(d) Operations

Four groups of animals were operated on as follows:

- Group 1: Six ewes were cannulated with nylon intravenous catheters (1.65 mm outer diam.) emerging via subcutaneous channels as previously described. The ewes were kept unrestrained and no fluid collectors were attached.
- Group 2: Ten ewes were cannulated as in group 1. The ewes were restrained in cages and the fluid collectors were either sutured to the skin or attached to coats worn by the animal.
- Group 3: Ten ewes had nylon cannulae emerging through stab wounds in the flank. The animals were restrained in cages and the fluid collectors attached with a contact adhesive.
- Group 4: Twenty ewes were cannulated as in group 3 with the exception that silicone rubber cannulae (int. diam 1.0 mm) were used. The ewes were given 1.2 million units of an antibiotic at 14-day intervals and the absorbent cotton plugs in the fluid collectors were sprayed daily with an aerosol antibiotic solution.

(e) Histological Examination

Two entire ewes from group 4 were used for histological examination. On slaughter the entire reproductive tract was placed in buffered formol saline fixative. Sections of fallopian tube were taken from points midway between the junction of

the ampulla and isthmus and the infundibulum and midway between the ampulla-isthmic junction and the transected end of the fallopian tube. The tissues were dehydrated, embedded in paraffin, sectioned at a thickness of 8μ , and stained with haemotoxylin and eosin. The stained sections were examined microscopically for morphological changes.

III. RESULTS

(a) Cannulation

The results are summarized in Table 1. Only the primary cause of failure has been considered when listing the number failing. Some failures showed a combination of causes, and kinking of the nylon catheters was more common than is indicated. However, this was not often a prime cause of failure.

Because the group 1 animals chewed off their cannulae, the group 2 animals were caged. However, the group 2 cannulations were not generally successful as the animals, irritated by the cannulae in the skin tunnels, removed or kinked cannulae by rubbing against their cages. This mechanical breakdown occurred in 50% of these ewes, but despite this a few successful cannulations remained patent for up to 57 days.

With group 3 ewes, 18 out of 20 cannulae remained patent for periods varying from 40 to 81 days. Patency was interrupted after varying periods due to kinking, blood clots, tissue growth into the end of the cannulae, and local infections of the oviduct (all infected animals were immediately slaughtered).

In group 4 ewes, one blocked cannula was found to terminate in a bend in the fallopian tube and was plugged with tissue. Two other cannulae became blocked with cell detritus and leucocytes. This problem was overcome in other ewes by gently stripping the exposed part of the rubber cannulae. All other cannulations in group 4 ewes remained patent until the animals were killed between 40 and 54 days. The ewes were free from infection and irritation and made no attempt to interfere with the cannulae or collectors even though they could easily have done so.

The post-mortem appearance of the reproductive tract in ewes from groups 1, 2, and 3 was characterized by extensive adhesions of all parts of the tract to the omentum, and tissue growth. The fallopian tubes appeared normal when dissected free. In contrast the group 4 ewes were almost entirely free of adhesions or tissue growth. Very slight reactions occurred around the silk sutures and the silicone rubber catheters were surrounded by a thin tissue envelope. The tubes appeared normal and in these animals the morphological appearance of the mucosa of the tubes was normal below the point of cannulation (Plate 1, Fig. 2). Where the cannula was apposed to the fallopian tube, the mucosal folds showed much flattening and tissue growth was evident in the vicinity of the holding sutures.

IV. DISCUSSION

Methods previously used for fluid collection have a number of disadvantages: ligation is unsuitable for long-term studies of secretion rates as fluid pressure inhibits secretion; flushings of the tract are unsuitable as no measures of concentration of the components are obtained. Stripping may cause damage to the epithelia and

TABLE 1
SUMMARY OF FALLOPIAN TUBE CANNULATIONS SHOWING PRIMARY CAUSES OF FAILURE

Group No.	No. of Ewes	Type of Cannulae	Mode of Emergence of Cannulae	Mode of Attachment of Collection Apparatus	No. of Fallopian Tubes Cannulated	No. Patent	Average Time Patent (days)	Causes of Failure (No. failing in parenthesis)
1	6	Nylon	Through skin tunnels	None attached	12	0	0	Removed by animal (12)
2	10	Nylon	As for group 1	Sutured to skin or attached to coats	20	5	45.2	Removed by mechanical actions of animals (10), blockage with blood clots (4), kinking in cannulae (3), infection (3)
3	10	Nylon	Through flank puncture	Attached to wool with contact adhesive	20	18	57.4	Infection (4), kinking in cannulae (2), blood clots (1), tissue growth into cannulae (11)
4	20	Silicone rubber	As for group 3	As for group 3	40	39	44.6	Animals slaughtered between 40 and 54 days. Three oviducts blocked by epithelium or cell detritus and leucocytes. All others patent until slaughter

contamination of the luminal fluid with cellular components. Abattoir material is unsuitable as post-mortem ionic shifts are extremely rapid and estimation of some components will be in error. Cannulation, while imposing artificial conditions in the fallopian tube, offers the best opportunity for long-term studies of the tubal fluids in viable animals.

Cannulation of the fallopian tube has been carried out in the rabbit (Bishop 1957; Clewe and Mastroianni 1960; Hamner and Williams 1963), the monkey (Mastroianni, Shah, and Abdul-Karim 1961), and the sheep (Black, Duby, and Riesen 1963). While the operative techniques are all similar, there is little information given on the percentage of success, morphology of the tube following cannulation, and associated problems. The criteria of successful cannulation would appear to be:

- (1) Long-term patency in a high proportion of cases.
- (2) Absence of bacterial contamination.
- (3) Absence of tissue reaction to the cannula material.
- (4) Absence of interference by the animal with any part of the system.
- (5) Presence of normal tubal tissue below the cannulae.

In these studies it was found that the above criteria were affected by the type of material used in the cannulae, their mode of exteriorization, the attachment of the collecting system, and attention to bacterial contamination. Nylon cannulae used in groups 1, 2, and 3 produced tissue reaction and adhesions, kinked, and, in some cases, contained blood clots indicating injury to the fallopian tube. On the other hand, silicone rubber cannulae were non-reactive and, being flexible, did not kink.

Irritation was caused by the use of skin tunnels for exteriorization, coats for the attachment of collectors, and skin sutures holding collectors. Caging the animals prevented oral interference but the animals rubbed the irritated portions against the sides of the cages causing damage and failure of the system. The use of flank punctures for exteriorization and contact adhesive for attaching collectors removed all irritation and the ewes showed no interest in the apparatus and could be kept unrestrained.

Bacterial contamination is a problem with any collecting system vented to the air. By daily treatment of the cotton plug in the air vent with an antibiotic, bacterial contamination was kept minimal. No infections were seen in group 4 ewes and no gross leucocytic invasions were seen in sections of the tubal mucosa. The only problem with the group 4 ewes was a tendency for the narrow cannulae to block with cell detritus and leucocytes. This was readily overcome as previously described. Microscopic examination of the tubal sections showed that no alteration occurred in the mucosa below the cannula and the tubes appeared normal and functional (Plate 1, Fig. 2).

Using the technique described for group 4 ewes, cannulations can be expected to remain patent, without bacterial contamination, for at least 54 days, the maximum time these ewes were allowed to live. The collecting system used here is cheap, easily constructed and cleaned, and is suitable for the quantitative study of secretion rates. If a paraffin layer is introduced into the polythene tube it allows the collection of fluids under anaerobic conditions. It is flexible, presents no stress to the animal, and can be evacuated by needle aspiration.

This technique of cannulation and fluid collection should be extremely useful in long-term studies of the physiology and biochemistry of fallopian tube fluid.

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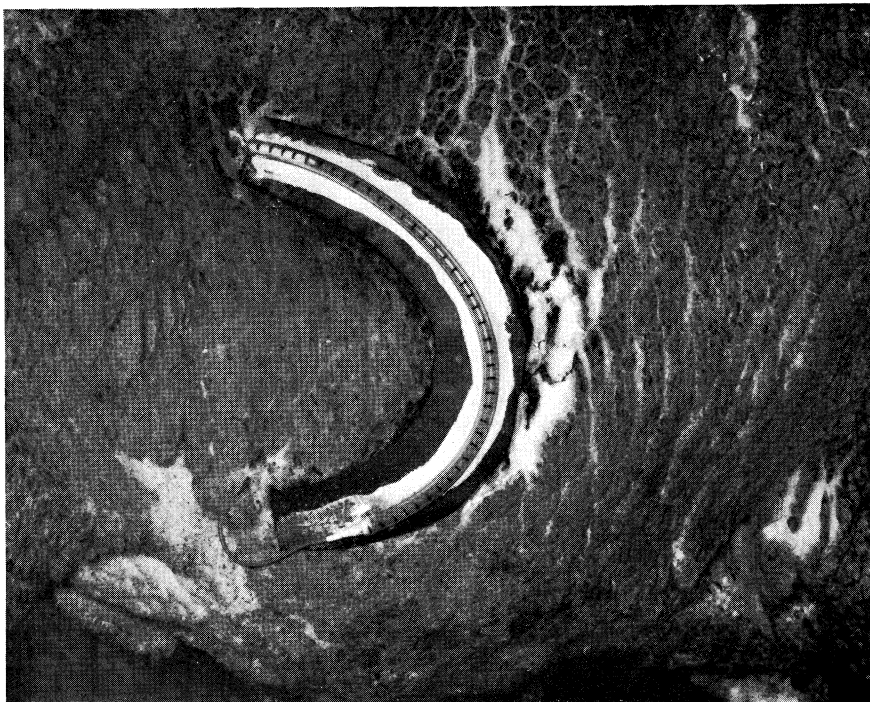


Fig. 1.—Fluid-collecting system attached to a sheep. The polythene collector is attached to the wool by means of a contact adhesive.

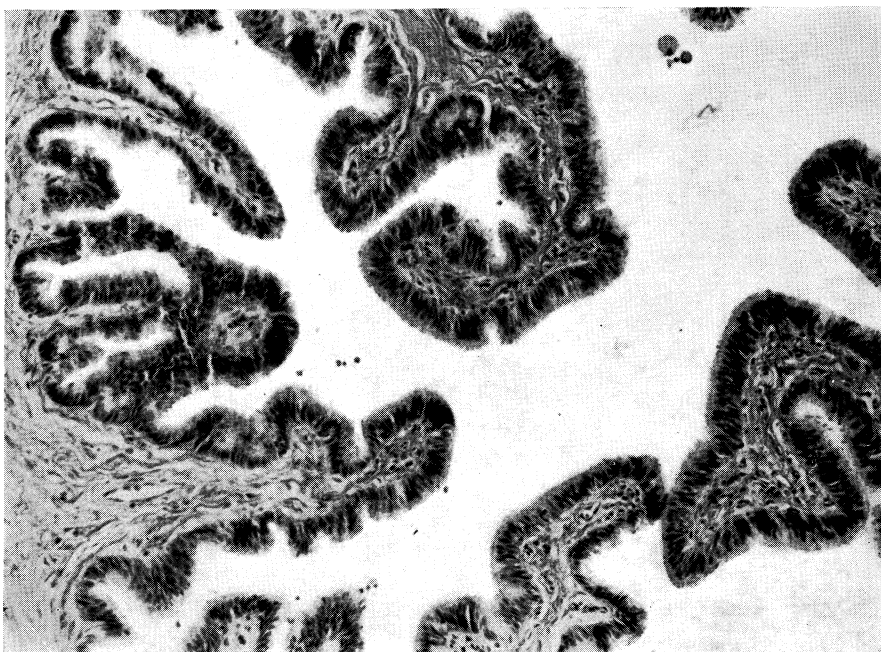


Fig. 2.—Normal epithelium from a fallopian tube with a cannula.

