EXPERIMENTAL AND CLINICAL
STUDIES OF THE SPINE OF
THE DOG.

BY

GEORGE DEREK BURRIDGE
B.Sc.(Edin), M.R.C.V.S.
CENTENARY POST-GRADUATE FELLOW,
ROYAL (LICK) VETERINARY COLLEGE
EDINBURGH.
TABLE OF CONTENTS.

PART ONE.

Puncture of the Cisterna Magna in the Dog.

PART TWO.

Experimental and Clinical Investigations into the use of Iodized Oil as a radiopaque medium in the examination of the Spinal Subarachnoid Space of the Dog.
PART ONE.

Puncture of the Cisterna Magna in the Dog.

Introduction.

Experimental and other investigations.

Section A. The Cisterna Magna of the Dog.
   (1) General Anatomy.
   (2) Depth.
   (3) Factors influencing the depth.

Section B. The Surgical Anatomy of the Atlanto-occipital Space, the Dorsal Atlanto-occipital Membrane and the overlying musculature.
   (1) The A-O space.
   (2) The A-O membrane.
   (3) The tissues of the neck overlying the cistern area.

Section C. Surface markings.

Section D. The Puncture.
   (1) Preparation of the Dog.
   (2) Needles.
   (3) Control.
   (4) The operation.
   (6) Failures.
   (7) Accidents.

Section E. The Effects of Puncture of the Cisterna Magna.
   (3) Studies of the cerebrospinal fluid Gp.2.Dogs.
      (a) Red Blood Cells in the c.s.f.
      (b) White Cells in the c.s.f.

Section F. Some Characters of the Normal Cerebrospinal Fluid of the Dog
   (1) Amount obtainable by Cistern Puncture.
   (2) Rate of flow during the operation.
   (3) Colour.
   (4) Coagulability.
   (5) Cell Content.

Section G. Clinical Applications.

Section H. Summary and Conclusions.

References.
Introduction.

Puncture of the Cisterna Magna in the lower animals has long been the routine method of reaching the sub-arachnoid space, for the withdrawal and/or study of the cerebrospinal fluid.

According to Becht (16) Magendie was the first to practice cistern puncture in the dog, an operation termed, in experimental work, the "fistula method". The procedure has been briefly described by other laboratory workers. Dixon and Halliburton (1) in 1913, referred to their technique as practised on the dog and goat. The animals were anaesthetised (chloroform, followed by urethane and morphine) and a mid-line incision made through the skin backwards from the occipital protuberance. A trocar and canula was then passed through the muscles of the neck into the cistern, the head being flexed and the trocar directed to a point midway between the eyes. Fraser and Peat (15) used a canula introduced into the cistern to study the flow of cerebrospinal fluid. Ether was used as a general anaesthetic; the dorsal atlanto-occipital membrane was exposed, a window excised, and the canula sutured in position. The same authors employed a similar technique the following year (II). Wegeforth, Ayer, and Essick (6), 1919, discuss the technique in the cat, and review the results of 1186 punctures in this animal. Weed and Hughson (9) refer to cistern puncture in the dog under general anaesthesia. Edwards (19) has described the puncture in the horse.
It was felt that such descriptions of technique as were available were inadequate for the routine practice of the operation. A method entailing general anaesthesia or a previous operation to reach the cistern was undesirable, and experiments were undertaken to test the practicability of cistern puncture as a clinical procedure.

SECTION A.

THE CISTERNA MAGNA OF THE DOG.

I. Anatomy. The cerebral arachnoid does not conform closely to the irregularities of the brain-surface. At the summit of the gyri the arachnoid is closely applied to the pia, but bridges over the sulci and so becomes separated from the pia by spaces of variable dimensions. In certain situations these spaces are quite large and are then termed cisterns. One of these, the cisterna magna, is formed in the angle between the cerebellum and the upper surface of the medulla (figures 1, 2, 3). Typically, the sub-arachnoid space is crossed by numerous filiform trabeculae arising from the arachnoid and attached to the pia. In the cisternae (c magnae etc.) the trabeculae are replaced by less numerous septae. The cisterna magna is roughly pyramidal in shape (figure 2). The base, situated anteriorly, is oblique, and formed by the lower posterior face of the cerebellum (vermis). The blunt apex is directed backwards and represents a direct continuation with the spinal sub-arachnoid space. The roof is formed by that part of the occipital bone comprising the upper part of the foramen magnum and by the dorsal atlanto-occipital membrane. The floor
is formed by the medulla oblongata and the first part of the spinal cord proper. The cistern communicates freely posteriorly with the spinal sub-arachnoid space (figure 3). Three communicating foramina are situated anteriorly; the paired and laterally situated Foramina of Lluschka and the single median Foramen of Magendie. The latter establishes communication with the fourth ventricle, and according to Frasier and Peat (21) is visible in young dogs if the dorsal atlanto-occipital membrane be excised.

2. Depth.
The vertical distances from the mid-point of the upper border of the foramen magnum to the floor (i.e. medulla-cord junction) of the cistern (figure 2, numbers 3 and 4), and from the level of the anterior border of the atlas arch (figure 2, numbers 1 and 2), were recorded in a series of sixty dogs of different breed and age by the following methods:

(a) Dissection of carcases of freshly killed dogs. The carcases were those of dogs destroyed by electrocution at the Stray Dogs' Home, Edinburgh. Examinations were made as soon as possible after death. The head was flexed at right angles, the muscles of the neck over the atlanto-occipital junction were reflected, the dorsal atlanto-occipital membrane exposed and dissected from its attachments and from the subjacent adherent dura. A window was then cut in the dura and the arachnoid to open into the cistern. The measurements referred to were then made with a graduated needle fitted with a sliding collar.
(b) Carcasses as above were placed in a refrigerator, some with the head flexed at right angles and some in the normal position; after freezing, longitudinal and transverse sections were made from which the depths of the cistern were ascertained.

(c) Examination of the cistern in the living dog was carried out in three instances. Each animal received one grain morphine sulphate hypodermically and a general anaesthetic (A.C.E. mixture). The dorsal atlanto-occipital membrane was exposed by a mid-line incision immediately behind the occipital protuberance. The membrane was excised with the associated dura and the arachnoid. The usual measurements of the cistern were then made. (I).

(I) Note. Other observations, referred to in detail later, were made in these three experiments. When the dorsal atlanto-occipital membrane was first exposed, all bleeding was controlled, and with the membrane quite free from blood, a spinal needle was passed into the cistern and the fluid withdrawn for examination (see p.34). The needle was withdrawn, and the leakage through the perforation studied (see p.10). With the cistern exposed, movements of the medulla in the living animal were observed (see p.6) and also the effect on the depth of holding the head in different positions.
The following table was compiled from the above data:

<table>
<thead>
<tr>
<th>Type</th>
<th>Depth F. Magnum</th>
<th>Depth Border Atlas</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adult Airedale similar</td>
<td>0.5 - 0.6 cms.</td>
<td>0.4 - 0.5 cms.</td>
</tr>
<tr>
<td>Adult Fox Terrier similar</td>
<td>0.3 cms.</td>
<td>0.25 cms.</td>
</tr>
<tr>
<td>Puppies about 12 weeks</td>
<td>0.2 cms. &amp; less</td>
<td>0.2 cms.</td>
</tr>
</tbody>
</table>

These measurements have been made with the head flexed, as cistern puncture is best carried out with the patient in this position.

3. Factors influencing the depth of the cistern.

(a) Movements of the medulla during life. In the three experiments referred to (p. 4) the movements of the medulla and anterior portion of the cord were watched. Under the conditions of the experiment the medulla and cord were observed to be subjected to a slight throbbing movement, synchronizing with the heart beat. The movement was of such a character and so slight as to make no practical difference to the depth of the cistern.

(b) Movements of the head. The effect of movements of the head on the depth was studied by the three methods used for measuring the depth (p. 3 and 4). Lateral movements made no difference. Flexion of the head caused a slight decrease in depth. This is due to two factors.

(a) The disposition of the atlanto-occipital joint is such that when flexion takes place, the upper border of the foramen magnum moves forward and slightly downwards. Thus the atlanto-
occipital membrane, and the dura which is attached to this anteriorly, is also carried slightly ventrally, and their distance from the floor of the cistern correspondingly reduced.

(b) The dens of the axis projecting forward under the cord tends to elevate the latter as flexion of the atlanto-occipital articulation takes place. The reduction in depth by flexion of the head is small enough to unimportant compared with the advantages gained by holding the head in that position during puncture.

SECTION B.


(1) The atlanto-occipital space. The shape and dimensions of the space were studied in different breeds of dogs and with the head in variable positions.

The space is elliptical in shape, having two borders, one anterior and the other posterior (figure 5). The latter is formed by the anterior border of the dorsal arch of the atlas. Its shape and relations appear to be constant in all breeds. The anterior border is formed by the lateral parts (Sisson) of the occipital bone and shew important variations in shape in different types of dogs (figure 4). Typically, the posterior face of the occipital bone descends de-
clivitously from the occipital crest and then turns horizontally between the two articular condyles to form a so-called "occipital shelf", the edge of which is the anterior boundary of the atlanto-occipital space. Variations in the shape of this shelf have been noted.

In the first type the shelf, well formed, projects backwards to overhang the foramen magnum. The posterior face of the cerebellum is situated well forward from the border (figure 4, skull number 2).

In the second type the shelf is cut out in the mid-line in the form of a notch. The notch usually invades the shelf half-way to the level where the bone ascends to the occipital crest. The cerebellum (vermes) is still protected by bone.

In the third type the shelf is practically absent, and, in addition, the mid-line notch is relatively larger (figure 4, skull number 1). In such cases the posterior face of the cerebellum is exposed, and looking through the foramen magnum, lies unprotected by bone and immediately related to the anterior part of the atlanto-occipital membrane. In puncturing this type the needle must be so directed as to pass as near to the atlas as possible. If puncture be made in a more forward position the cerebellum may be impinged. This deficient shelf is found generally in the short-nosed breeds, viz:—

Pekingese, Pugs, French Bulldogs.

In cases of doubt, X-ray photos will demonstrate the border clearly (see figure 6).

Flexion of the head increases the antero-posterior measurement of the atlanto-occipital space. The following list indicates some of the measurements obtained post-mortem.
<table>
<thead>
<tr>
<th>Breed</th>
<th>Head normal position</th>
<th>Head flexed at 90°</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Average, mid-line, in cm.</td>
<td>90° in cm.</td>
</tr>
<tr>
<td>Aberdeen Terrier</td>
<td>0.5</td>
<td>1.0</td>
</tr>
<tr>
<td>Airedale</td>
<td>1.0</td>
<td>1.6</td>
</tr>
<tr>
<td>Alsatian</td>
<td>0.6</td>
<td>1.4</td>
</tr>
<tr>
<td>Bull Dog</td>
<td>0.4</td>
<td>0.8</td>
</tr>
<tr>
<td>Bull Terrier</td>
<td>0.5</td>
<td>1.1</td>
</tr>
<tr>
<td>Cairn Terrier</td>
<td>0.6</td>
<td>1.2</td>
</tr>
<tr>
<td>Fox Terrier</td>
<td>0.8</td>
<td>1.4</td>
</tr>
<tr>
<td>Greyhound</td>
<td>1.1</td>
<td>1.6</td>
</tr>
<tr>
<td>Kerry Blue</td>
<td>0.8</td>
<td>1.1</td>
</tr>
<tr>
<td>Mastiff</td>
<td>1.2</td>
<td>1.6</td>
</tr>
<tr>
<td>Pomeranian</td>
<td>0.9</td>
<td>1.4</td>
</tr>
<tr>
<td>Sealyham</td>
<td>0.8</td>
<td>1.25</td>
</tr>
<tr>
<td>Spaniel</td>
<td>0.6</td>
<td>1.3</td>
</tr>
<tr>
<td>Yorkshire Terrier</td>
<td>1.0</td>
<td>1.4</td>
</tr>
</tbody>
</table>

The small measurements in the case of the Bulldog and Bull Terrier are due to the marked development of the occipital shelf with no central notch, associated doubtless with muscle development in these breeds. A Yorkshire Terrier on the other hand with a small shelf and mid-line notch has measurements larger than the Bull Terrier.

From the above observations it is evident that cistern puncture should be carried out with the head flexed. Such a position (1) gives a wider bony space through which to pass, (2) is safer from the point of view of injury to the cerebellum in certain breeds, and (3)
does not materially lessen the depth of the cistern itself.

2. The dorsal atlanto-occipital membrane presents several features of interest in relation to puncture.

(a). It is adherent to the subjacent dura; this bond is strengthened in that both the membrane and the dura are anchored to the bony boundary of the foramen magnum. The two can be dissected apart, but it is unlikely that a needle having pierced the membrane proper would push the dura before it after the fashion of a gloved finger as can happen in other situations for spinal puncture.

(b). Small vessels permeate the membrane along the line of attachment to the atlas. Blood contamination of the cerebrospinal fluid occurs if during puncture the needle is passed near the bony boundaries of the space (see table 2, Dog II). This contamination appears to be due to rupture of these vessels.

(c). The elasticity of the membrane is important from the point of view of post-puncture leakage. There is evidence to suggest that such leakage occurs. In the experiments described on p. 4 a puncture was made through the exposed atlanto-occipital membrane with a standard sized needle, the head being flexed and the membrane, therefore, tense. When the needle was withdrawn, the cerebrospinal fluid spurted from the hole in small jets, synchronising with the pulsations of the fluid within. When the head was placed in the normal position, visible leak-
age ceased. This was observed in each of the three dogs operated upon. Frasier and Peat (21) when injecting methylene-blue into the cistern observed that leakage of the dye occasionally occurred. It did not occur if before the injection an equal amount of cerebrospinal fluid was withdrawn. Weed and Hughson (9) investigating the cerebrospinal fluid pressure in dogs occasionally met unusually low pressures (below 90 m.m. Ringers solution). This was only found when the occipital ligament had been punctured more than once. The writers state: "the explanation of this finding is apparently to be related to the escape of cerebrospinal fluid through the initial puncture hole of the ligament into the neck".

Proof is wanting however that leakage occurs after puncture performed in the routine manner. As will be mentioned (p. II) the rectus capitis dorsalis minor muscle is closely associated with the membrane to the contractility of which it doubtless adds, aiding in the closure of the needle hole. It is however recommended that after puncture the head be brought back to the normal position at once so that leakage if any is minimal; also any factor likely to increase cerebrospinal fluid pressure should be avoided (e.g. excitement).

2. The tissues of the neck overlying the cistern area (i.e. that part of the neck immediately behind the occipital protuberance) are divisible into skin, subcutaneous fascia and muscles. THE THICK SKIN is very loose and hence a needle inserted through
it can be moved to different positions without withdrawal; this is useful when attempting a second puncture in a different plane.

THE MUSCLES can be divided into five layers. All are paired lying one on either side the mid-line with the exception of the deepest situated, the rectus capitis dorsalis minor. From without inwards the layers are:

(a) The auricular muscles (Long Levator, middle levator, and long abductor) together with the aponeurosis of the cleido-cervical (paired).
(b) Splenius (paired).
(c) Biventer (paired).
(d) Rectus capitis dorsalis major (paired).
(e) Rectus capitis dorsalis minor (single). This muscle is inserted partially into the membrane and closely adherent to it.

A needle passed in the mid-plane of the neck passes along the fascial plane which divides these pairs of muscles. This fascial division is constituted as follows:—The origins of the long levator and long abductor of the ear together with the anterior parts of cleido-cervical aponeurosis and insertion of the splenius are intimately related in the mid-line to form a thin fibrous cord analogous to the abdominal linea alba. The division between the remaining pairs is fascia. Finally the rectus minor, as before mentioned, lies as a single muscle over the mid-part of the atlanto-occipital membrane. The ligamentum nuchae does not reach to this level. Thus a needle transverses first a resistant cord, secondly a loose fascial division and thirdly a single
muscle before reaching the atlanto-occipital membrane. 

Nerves trunks in the region are not numerous and none lie in the direct path of the needle. The most closely situated are the dorsal branches of the second cervical nerve. These pierce the cleido-cervical aponeurosis on either side the mid-line to reach the external ear but in no part of their course are near enough to be injured.

Blood vessels are scanty. A few reach the mid-line, between the auricular muscles from the posterior auricular vessels. The most important are two veins which lie, one on either side, between the dura and the atlanto-occipital membrane and pass forward along the lateral boundaries of the atlanto-occipital space. They are the direct continuation of the longitudinal venous sinuses of the spinal canal proper. In this region the sinuses leave the floor of the canal, incline upwards on either side of the cord until at the level of the atlanto-occipital space they are situated dorso-lateral to the cord (see figure 13). Their size renders them liable to puncture if the needle passes a little to either side of the mid-line. For instance in a mastiff on post-mortem each vein had a diameter of 0.5 cm. This is doubtless a maximum, but even in small breeds their size is considerable. The two veins are more widely separated anteriorly so that injury is less likely if the needle is passed close to the occipital bone (see discussion on Blood in the Fluid p. 20).
SECTION C.

SURFACE MARKINGS.

The surface landmarks given below will define the position of the atlanto-occipital space in relation to the surface of the animal.

A cistern puncture is invariably carried out with the head flexed at right angles; the surface markings are accordingly described with the head flexed and the patient in a position described on page 15 under "Control" (Section D, 3).

Note. Figures 5-II have been mounted to correspond to the position of the patient when viewed by an operator standing as described in the text; (i.e. behind the dog with its head to the right hand).

Identify the external occipital protuberance through which the line C passes in figures 5-II; draw the imaginary line A (figures 5-II); this cuts the wings of the atlas at their most prominent part (see figures 5, 6, 8, II). The line B (figures 6-II) lies exactly halfway between A and C (see figures 5, 6, 8, 9, II). A transverse perpendicular plane at the level of the line B passes through the atlanto-occipital space.

If a needle be introduced at the point where B cuts the midline D E and is passed inwards at right angles to the dorsal line of the neck, it will traverse the atlanto-occipital space and enter the cisterna magna. (See X-ray photograph. Figure II: also Figure 6).

SECTION D.

THE PUNCTURE.

This procedure will be dealt with under the following headings:
Preparation of the dog. An area of skin from the occipital crest backward for some inches is shaved, preferably on the day previous to operation. Food should be withheld for some hours. Half to one grain of morphia sulph. given hypodermically half an hour before operation is useful but not essential.

Puncture Needles. The cistern puncture needle for the dog is similar in construction to the ordinary lumbar puncture needle for the human. It is made of stainless metal and whilst being slightly flexible must be rigid enough to be sighted and propelled in any selected direction through the soft tissues of the neck; or the comparatively tough skin in that region. The needle is fitted with a stillette which locks itself by means of a small projection which is received into a corresponding socket in the head of the needle. Five grooves, each one centimetre apart are lightly engraved around the stem of the needle; the first being placed one centimetre from the point. These are of help to the operator in estimating the depth at which the cistern lies. A ring of solder is fitted 6 cms from the tip to serve as an additional guide. The graduations should not be cut too deeply or the needle may fracture when used. The writer has latterly utilized needles on which the markings are engraved only halfway around the barrel, and in such a position that they are in view to the operator when the needle is in position for operation. The head of the needle should be made to fit a
standard type syringe or needle adapter. A needle for use with dogs of the average sized or larger breeds has the following dimensions:

Length with stillette in position \(10.5\) cm.

External diameter of the barrel \(1.5\) mm.

Internal diameter of the barrel \(1.0\) mm.

For the smaller breeds and puppies a shorter needle with a proportionately smaller diameter is recommended. The needle is sterilised in boiling water, after which a little absolute alcohol and then ether are drawn through to remove the water. After use it is treated with distilled water, absolute alcohol and ether, and then dried. The needle may be sterilised in olive oil (13), which while boiling at a much higher temperature than water does not blunt the point.

(3) Control. The proper control of the patient is essential to safety in operating and success in obtaining good samples of cerebrospinal fluid. Two assistants are necessary. The dog is placed on a table on its right side with the back towards the operator and the head to his right. One assistant, standing behind the dog, leans over and holds the hind legs in one hand and the fore in the other at the same time keeping the animal's back in a straight line along the edge of the table. The second assistant standing on the opposite side of the table, places the palm of the right hand flatly behind the neck to check sudden backward movements and to keep the mobile skin in position. The left hand grasps the "snout" the thumb gripping into the inter-mandibular space. With this hand the head can
be flexed and maintained at right angles in two directions, viz. at right angles to the long axis of the neck and right angles to the horizontal table. It is important to adopt and maintain this position. A faulty alignment of the head and neck is the most frequent source of failure to puncture quickly and successfully. If the patient be a long-necked type a sandbag "sausage" may be placed under the neck to support this horizontally.

Note. In the very short-nosed breeds, such as the Pekingese, it is not possible to hold the head without danger of sudden movement. Such animals are best operated on with a general anaesthetic, or basal narcotic such as Avertin.

(4) The Operation. Sterile cloths are arranged to leave the head uncovered; this helps to "sight" the needle more accurately. The shaved skin is treated with spirit and then with ether. A few cubic centimetres of local anaesthetic (novocaine) are injected subcutaneously in the mid-line and also deeply into the neck muscles along the estimated track of the needle. A short needle should be used for injection so that, should the patient make a sudden movement, there is no risk of puncturing the cord or medulla. The operator stands in such a position that the shaved operation area is facing him, and the dog's head is pointing to his right. The figures 5-II, relating to the puncture proper, have all been mounted to correspond to the position of the dog when viewed by an operator standing as described above.

The point on the wing of the atlas cut by the line A (see figures 5-II) is now palpated through the cloths with the index finger of the left hand. The external occipital protuberance is
defined with a finder of the right hand and the needle, held in the same hand, passed through the skin at the point estimated as described on page 13 under surface markings. The loose skin and fascia allows the needle to be moved a little in any direction to correct the position. With its head against the ball of the thumb the needle is pushed steadily towards the atlanto-occipital space to reach which it must pass inwards at right angles to the dorsal line of the neck and along the fascial division of the neck muscles. At a depth of about 3 cm. the needle passes into the cistern. This is detected by a sudden cessation of resistance. The sensation of entering a space is simultaneously imparted to the hand of the operator. The trocar should be immediately withdrawn; the flow of clear cerebrospinal fluid from the needle indicates a successful puncture. Rotating the needle slightly or clearing the lumen with the stillette may give an increased flow. The depth to which a needle must be passed to reach the cistern varies with the breed, age and condition of the individual, and accurate estimation can only be made with experience.

The following figures form a rough working basis.

A. Large Breeds. (Airedale, Alsatian, Bull Terrier) 3.5 cm
B. Medium-sized Breed (Fox Terrier) 2.8-3.5"
C. Toy Dogs and young puppies

(17)

(5) After-treatment. Allow the head to return to normal position as soon as the puncture is completed. This reduces the risk of leakage (see above). Similarly any factor that raises c.s.f. pressure should be avoided. The skin puncture should be
sealed with collodion.

Failure to enter the cistern may be due to:

1. Incorrect position of the head and neck of the patient. This is the commonest cause of failure and must be corrected as indicated.

2. Impingement of bone (see figure 12). If bone be encountered it will be either the occipital shelf or the atlas arch (see figures 5, 6, 8, 11); it is easy to decide between the two by examining the direction of the needle. To correct withdraw the needle from the muscles and re-direct.

Accidents. During the experiments described and others concerning lipiodol injection, together with the hospital cases referred to, 94 punctures have been performed on 20 dogs of varying breed. The following mishaps have occurred:

(a) Breaking of needle. This happened once. The needle had been passed into the neck but no puncture affected. When pushed in a little further it passed near to a bony margin. The dog felt some pain and moved suddenly. The needle snapped off at the level of one of the centimetre markings, the distal segment remaining embedded near the occipital bone. Such an eventuality can be guarded against by effective control of dog and the use of stout needles, the centimetre markings on which are not too deeply cut and extend only halfway round the barrel. The needle should not be forced in the neighbourhood of bony edges, since this caused pain.

(b) Impingement of medulla (?). In two instances, when the flow of cerebrospinal fluid was interrupted and the needle was in-
serted a little further into the cistern, the dog evinced sudden acute pain; the needle was withdrawn at once and the animal showed no further symptoms. It was assumed that the pia mater overlying the floor of the cistern had been touched.

SECTION E.

THE EFFECTS OF THE PUNCTURE.

The effects of puncture have been studied in experimental dogs and later in patients subjected to cistern puncture for diagnosis. Seven experimental dogs were used. All were clinically healthy and young adult cross-breeds of the airedale type. They were divided into groups I and 2, viz: I, 2, 3, 4 and II, 12, 13.

(I) Clinical observations Gp I. Dogs in the first group were punctured every fourth day until seven successive punctures had been performed on each animal. Temperatures were taken twice daily and general health (appetite, faces, urine etc.) noted. Puncture appeared to have no effect whatever on the dogs.

(2) Clinical observations Gp 2. The dogs in the second group (numbers II, 12, 13) were examined similarly. The temperatures of the dogs in this group are plotted on the accompanying charts No.5, 6 and 7.

(3) Studies of the Cerebrospinal Fluid, Gp 2.

Cell Counts.

The fluid was received into sterile chemically clean and dry centrifuge tubes; the wool stoppers were wrapped in "Kraft paper" to obviate small fibres of
wool reaching the sample.

A white cell count was made as soon as possible owing to tendency for white cells to disappear from a fluid on standing. A Fuchs-Rosenthal chamber was used, the c.s.f. being diluted 1 in 10 with the following fluid:

- Ethyl violet 40 mgm.
- Glacial acetic acid 6 cc.
- Distilled water 30 c.c.

The whole chamber was counted (3.2 c.mm.). Red blood cells when present were counted in c.c.mm. of the undiluted c.s.f. in the same chamber. Samples containing a high blood content (e.g., over 2,500 R.B.Cs per c.mm.) were counted in the Thoma-Zeiss Chamber.

To estimate the effect of several punctures, the white cell count of the cerebrospinal fluid was studied and the relevant data is set out in tables 2, 3, and 4. The columns from left to right indicate:

1. Date of puncture; 2. Interval; i.e. of days between each puncture; 3. Red blood cells found in fluid sample; 4. Correction figure column (see below); 5. The total white cell count; 6. The actual white cell count after correction; (see below). 7. Remarks.

Before reviewing the White Cell count it is necessary to discuss the occurrence of Red Cells in the samples of fluid.

(3a) RED BLOOD CELLS (R.B.Cs) IN THE CEREBROSPINAL FLUID.

Twenty-four punctures were performed on the three dogs 11, 12, 13. Of the samples thus obtained

(a) One was free from R.B.Cs.
(b) Eight contained less than 10 R.B.Cs per c.mm.
(c) Nineteen contained less than 500 R.B.Cs. per c.mm.
(d) Twenty contained less than 1,000 R.B.Cs per c.mm.

Of the remaining four samples, the first contained
17,000, the second 17,760, and the third 1,700. No count was made of the fourth sample. (Dog 12; on 17-3-28).

The R.B.C.s were invariably crenated even when examined within a few minutes of puncture. If the needle or the tube in which the cerebrospinal fluid was collected were wet, the crenation was more marked; hence collecting tubes were always dry sterilised.

The writer regards red blood cells as foreign to normal cerebrospinal fluid of the dog, and believes that they are introduced as contaminants during puncture, for the following reasons.

(I) In the three experiments described on page 4 it was mentioned that after the atlanto-occipital membrane had been exposed, all bleeding was controlled and then a spinal needle was passed into the S-A space; samples of cerebrospinal fluid were obtained and counts made. No red cells were encountered in 6.4 c.mm. of each sample. Under these conditions the possibility of extraneous contamination was reduced to a minimum, and resulted in a fluid entirely free from red cells.

In addition six samples amongst those taken in the routine manner by the writer, have been found to be blood free. These blood free samples were obtained as follows:

(a) Dog No: II 24.2.28.
(b) Dog No: 6 3.10.28. This dog was airedale cross used for lipiodol injections. (see Part Two of this paper).
(c) Dog No: 6 3.10.28 Ditto.
(d) Dog No: 14 21.3.28 A terrier cross used for lipiodol injections. (see Part Two of this paper).
(e) Airedale in clinic of London Veterinary College (Dec: 1928).
Sustained punctured wound occipital region; cerebrospinal fluid withdrawn for diagnosis.

(f) Pekingsese in clinic of London Veterinary College (Jan: 1929) shewing inco-ordination; cerebrospinal fluid withdrawn for diagnosis.

(2) The red cell content as demonstrated in fluids obtained by the technique described, varies very considerably. In cases where a puncture was not performed with facility, the resultant sample often contained a great number of red cells (Dog 13 on 3.3.28, Dog 14 on 16.3.31; and Dog II on 25.3.28). The converse held good that a sample shewing high R.B.C. count had invariably been obtained with difficulty. As the technique became perfected later in the experiments, the red cell counts were constantly low.

(3) If the needle during puncture passed close to one of the bony boundaries of the atlanto-occipital space, the sample obtained frequently had a high red cell content. As mentioned in Section B.2, the ramification of the small A-O sinuses has previously been described, numerous small vessels ramify in the membrane near its bony attachments. The blood contamination which has been observed when the needle passes close to the bony boundaries of the A-O space is probably due to puncture of these vessels.

(4) If, during an attempt at puncture, pure blood issued from the needle in a slow stream and the needle was withdrawn and the puncture then performed with a new needle, the sample often shewed high red cell counts. It is assumed on anatomical evidence that such cases represent puncture of the venous sinuses mentioned previously.
as lying on either side the atlanto-occipital space. Puncture of such vessels must doubtless lead to considerable local extravasation of blood resulting in the subsequent contamination of a second needle or sample.

(5) It has been noticed occasionally that the first part of a sample of cerebrospinal fluid was visibly blood stained or blood streaked; the second part less so, or microscopically free. This suggests that blood has been carried into the needle lumen by the stillette and swept out with the first rush of fluid.

The red blood cells in the cerebrospinal fluid of the normal dog are therefore probably contaminants. The detection of these cells is best done microscopically. A sample of fluid which is crystal clear to the naked eye may on microscopic examination be found to contain several red cells per c.mm. A fluid moderately contaminated with blood has a yellow brown colouration, and can be fairly accurately matched against weak aqueous solutions of potassium bichromate of varying dilutions. Such colour standards are useful for rough comparison.

Since it has been assumed that the red cells are accidentally introduced from the blood, it must be assumed that the white cells of the blood stream can also contaminate the sample but in a proportionately smaller number. The blood of the dog has 5–7 million and 12,600 (average 6) red blood corpuscles per c.c. white corpuscles. (Ellenberger and Trautmann, 1921). From these figures one can compile a table indicating the number of white cells presumably introduced.
into cerebrospinal fluid samples with a given number of red cells.

Such a table (Table I) has been used to calculate the number of white cells to be subtracted from the total white cell count on the assumption that some of the latter must have been accidentally introduced with the reds, if so. For instance in the case of Dog II (Table 2 Page 28) on 15.3.28, the sample contained 429 R.B.C's per c.mm. From Table I, the number of white cells probably carried in with these reds is one, which is accordingly deducted from the total white cell count of 47.5, leaving an actual white cell content of 46.5. The white cell count of all samples containing more than 230 red cells has been similarly readjusted. In two instances this "proportion system" has proved incorrect, viz:-

On 25.3.28, Dog II yielded a sample with 7,000 reds (counted Thoma Zeiss) and 12.8 whites per c.mm. The number of whites assumed to have been carried in with the reds is approximately 32. Subtracting this figure from the white count leaves a negative count of 19.4. This occurred in the case of Dog II punctured on 16.3.31, when 7,600 red cells per c.mm. were counted. A proportionate deduction from the total whites (23.2) left a negative value of about 1.8 for white cells in the fluid. These two examples concerned fluids highly contaminated following bad punctures.

(3b) THE WHITE CELL COUNTS OF THE CEREBROSPINAL FLUID of the dog successive subjected to cistern punctures are indicated in columns 4 and 6 (tables No: 2,3,4, and graphs I,2,3). In column 4 the total white count is set out, and in column 6 the corrected count after allowing for the white cells introduced by blood contamination referred to
The figures for Dog II on 25.3.28, and for Dog II' on 16.3.31 have not been considered because of the very high blood contamination which occurred. As mentioned before in these two instances the necessary correction resulted in a negative count.

From the figures it is seen that in each dog a rise occurred in the white cell count. The rise occurred suddenly, followed by a gradual decrease. (see graphs 1, 2, 3). In Dog II, the initial increase up to 46.5 occurred after four punctures had been performed in 17 days (average interval 4.2 days). In Dog I2 the increase was to 188 after 6 punctures performed in 19 days (average interval 3.3 days). Dog I3 showed an increase to 70.2 after 2 punctures performed in 7 days (average interval 3.5 days).

The average result for the three experimental animals may be stated as follows: -

A cell rise to 104 occurred after 4 punctures performed at intervals of 3.5 days.

A bacteriological examination of the cerebrospinal fluid of Dog II was made on 16.3.28, one day after the rise to 46.5. Similarly on 17.3.28, with Dog I2 when a rise to 185.5 was noted. The investigation comprised examination of films of centrifuged deposit and attempted cultures from the deposit on the following media.

Agar, serum +, glucose +, broth, glucose broth, blood-agar broth. The films and cultures (72 hours) were negative. Also when a cell rise was encountered, washings were taken from the needles after the routine cleanings and examined in the counting chamber for res-
idual cells which could have influenced subsequent counts. The washings were always cell free.

The experiments are not numerous enough to draw conclusions as to the cause or mechanism of the white cell increase following serial puncture. The time interval between punctures, the size of the needle and the facility of the operator may be shown to play a part. What can be stated is that the cerebrospinal fluid of three dogs showed an increase in white count during serial cisternal puncture under the experimental conditions described. The cell increase may occur after two punctures but in two of three experimental dogs appeared later. After reaching a high figure the cell count tended to fall, especially if no puncture were performed for 8-10 days. The animals appeared normal and temperatures (see charts) and appetite remained unaltered throughout.
TABLE I.

Correction Table for White Cell Count in Cerebrospinal Fluid (dog) when blood contamination is present.

Dog's Blood Count R.B.Cs. 5-7 million; whites 12,600
(Ellenberger & Trautmann) 450 R.B.Cs correspond to 1 white cell.

<table>
<thead>
<tr>
<th>Number R.B.Cs.</th>
<th>Correction for White Cell Count</th>
</tr>
</thead>
<tbody>
<tr>
<td>230 - 460</td>
<td>1</td>
</tr>
<tr>
<td>460 - 690</td>
<td>1.5</td>
</tr>
<tr>
<td>690 - 920</td>
<td>2</td>
</tr>
<tr>
<td>920 - 1150</td>
<td>2.5</td>
</tr>
<tr>
<td>1150 - 1380</td>
<td>3</td>
</tr>
<tr>
<td>1380 - 1610</td>
<td>3.5</td>
</tr>
<tr>
<td>1610 - 1840</td>
<td>4</td>
</tr>
<tr>
<td>1840 - 2070</td>
<td>4.5</td>
</tr>
<tr>
<td>2070 - 2300</td>
<td>5</td>
</tr>
<tr>
<td>2300 - 2530</td>
<td>5.5</td>
</tr>
<tr>
<td>2530 - 2760</td>
<td>6</td>
</tr>
<tr>
<td>2760 - 2990</td>
<td>6.5</td>
</tr>
<tr>
<td>2990 - 3220</td>
<td>7</td>
</tr>
<tr>
<td>3220 - 3450</td>
<td>7.5</td>
</tr>
<tr>
<td>3450 - 3680</td>
<td>8</td>
</tr>
<tr>
<td>3680 - 3910</td>
<td>8.5</td>
</tr>
</tbody>
</table>
TABLE 2.

The Cell Counts of the C.S.F. of Dog II.
(see also Graph: No I.)

<table>
<thead>
<tr>
<th>DATE</th>
<th>INTERVAL (days)</th>
<th>RED BLOOD CELLS</th>
<th>TOTAL WHITES</th>
<th>CORRECTION</th>
<th>ACTUAL WHITES</th>
<th>REMARKS.</th>
</tr>
</thead>
<tbody>
<tr>
<td>24.2.28</td>
<td>0</td>
<td>0</td>
<td>1.7</td>
<td>-</td>
<td>1.7 per cm/m</td>
<td></td>
</tr>
<tr>
<td>27.2.28</td>
<td>3</td>
<td>3.6</td>
<td>2.5</td>
<td>-</td>
<td>2.5</td>
<td></td>
</tr>
<tr>
<td>1.3.28</td>
<td>3</td>
<td>3.1</td>
<td>2.1</td>
<td>-</td>
<td>2.1</td>
<td></td>
</tr>
<tr>
<td>8.3.28</td>
<td>7</td>
<td>62.0</td>
<td>7.3</td>
<td>-</td>
<td>7.3</td>
<td>Several attempts made before c.s.f. was obtained.</td>
</tr>
<tr>
<td>12.3.28</td>
<td>4</td>
<td>71.0</td>
<td>2.0</td>
<td>-</td>
<td>2.0</td>
<td>Needle passed near bone. Note R.B.C. count.</td>
</tr>
<tr>
<td>15.3.28</td>
<td>3</td>
<td>429.0</td>
<td>47.5</td>
<td>1</td>
<td>46.5</td>
<td>c.s.f. obtained on 1st attempt.</td>
</tr>
<tr>
<td>16.3.28</td>
<td>1</td>
<td>165.0</td>
<td>21.5</td>
<td>-</td>
<td>21.5</td>
<td>Bacteriological exam; of this sample.</td>
</tr>
<tr>
<td>25.3.28</td>
<td>9</td>
<td>17,000.0</td>
<td>12.8</td>
<td>(32) (- 19.2)</td>
<td>4 attempts before before c.s.f. obtained.</td>
<td></td>
</tr>
</tbody>
</table>
### TABLE 3.

The Cell Counts of the C.S.F. of Dog 12.
(see also Graph: No I.)

<table>
<thead>
<tr>
<th>DATE</th>
<th>INTERVAL (in days)</th>
<th>RED BLOOD CELLS</th>
<th>TOTAL WHITES</th>
<th>CORRECTION</th>
<th>ACTUAL WHITES</th>
<th>REMARKS</th>
</tr>
</thead>
<tbody>
<tr>
<td>27.2.28</td>
<td>-</td>
<td>1</td>
<td>2.1</td>
<td>-</td>
<td>2.1</td>
<td></td>
</tr>
<tr>
<td>2.3.28</td>
<td>4</td>
<td>5.9</td>
<td>1.6</td>
<td>-</td>
<td>1.6</td>
<td>6.4 cm/m counted for whites.</td>
</tr>
<tr>
<td>6.3.28</td>
<td>4</td>
<td>0.9</td>
<td>2.9</td>
<td>-</td>
<td>2.9</td>
<td>Ditto.</td>
</tr>
<tr>
<td>9.3.28</td>
<td>3</td>
<td>72</td>
<td>1.9</td>
<td>-</td>
<td>1.9</td>
<td>Ditto.</td>
</tr>
<tr>
<td>13.3.28</td>
<td>4</td>
<td>0.6</td>
<td>1.7</td>
<td>-</td>
<td>1.7</td>
<td>Ditto.</td>
</tr>
<tr>
<td>16.3.28</td>
<td>3</td>
<td>11,760</td>
<td>21.4</td>
<td>23.2 (- 1.8)</td>
<td>First three attempts gave pure blood; fourth attempt bloody fluid.</td>
<td></td>
</tr>
<tr>
<td>17.3.28</td>
<td>1</td>
<td>185.5</td>
<td></td>
<td>-</td>
<td>185.5</td>
<td></td>
</tr>
<tr>
<td>17.3.28</td>
<td>No counts made.</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Bacteriological examination.</td>
</tr>
<tr>
<td>25.3.28</td>
<td>6</td>
<td>38</td>
<td>4</td>
<td>-</td>
<td>4</td>
<td></td>
</tr>
</tbody>
</table>
TABLE 4:
(see also Graph: No 3.)

<table>
<thead>
<tr>
<th>DATE</th>
<th>INTERVAL (in days)</th>
<th>RED BLOOD CELLS</th>
<th>TOTAL WHITES</th>
<th>CORRECTION</th>
<th>ACTUAL WHITES</th>
<th>REMARKS</th>
</tr>
</thead>
<tbody>
<tr>
<td>29.2.28</td>
<td>-</td>
<td>1</td>
<td>4</td>
<td>-</td>
<td>4</td>
<td></td>
</tr>
<tr>
<td>3.3.28</td>
<td>3</td>
<td>1,700.0</td>
<td>8.6</td>
<td>4</td>
<td>4.6</td>
<td></td>
</tr>
<tr>
<td>7.3.28</td>
<td>4</td>
<td>409.0</td>
<td>71.2</td>
<td>1</td>
<td>70.2</td>
<td></td>
</tr>
<tr>
<td>7.3.28</td>
<td>5 hrs</td>
<td>409.0</td>
<td>61.1</td>
<td>-</td>
<td>60.1</td>
<td>Bacteriological exam;</td>
</tr>
<tr>
<td>10.3.28</td>
<td>3</td>
<td>132.0</td>
<td>19.1</td>
<td>-</td>
<td>19.1</td>
<td></td>
</tr>
<tr>
<td>14.3.28</td>
<td>4</td>
<td>18.0</td>
<td>7.2</td>
<td>-</td>
<td>7.2</td>
<td></td>
</tr>
<tr>
<td>24.3.28</td>
<td>10</td>
<td>319.0</td>
<td>2.0</td>
<td>-</td>
<td>2.0</td>
<td>Three attempts before successful.</td>
</tr>
</tbody>
</table>
SECTION F.

SOME CHARACTERS OF THE NORMAL CEREBROSPINAL FLUID, OF THE DOG.

From the data available a few facts may be stated about the normal cerebrospinal fluid of the dog. These are founded on observations from the first punctures on Dogs II, I2 and I3 since these animals were then virtually normal dogs, and also from certain dogs in the hospital of the London Veterinary College. These were punctured of course, for routine diagnosis and were as follows:-
(1) Airedale, adult, I.XI.28. Sustained deep punctured wound of parietal region two days previously; cerebrospinal fluid examined with reference to meningitis. The cell count and the subsequent history was negative for meningitis. Referred to as Dog X.

(2) Puppy, I.X.29. Fox Terrier. Abscess of right ear with unusual symptoms of dizziness. Punctured to eliminate possibility of perforation meningitis. Cell count normal (2.6) and patient recovered under usual treatment. Referred to as Dog Y.

(3) Terrier. 21.XII.28. Punctured for cerebrospinal fluid examination after enucleation of eyeball ruptured and torn during dog-fight. Cell count and subsequent history point to no extension of sepsis. Referred to as Dog Z.

(I) Amount. The six dogs referred to above yielded the following amounts of cerebrospinal fluid by cistern puncture.

   (a) No.11. 24.2.28; 5.5 ccs;   (b) No.12. 27.2.28; 5.5 ccs.
   (c) No.13. 29.2.28; 4 ccs;   (d) Dog X. I.II.28; 6 ccs;   (e) Dog Y.
   I.I0.29; 4 ccs;   (f) Dog Z. 21.II.28; 5 ccs.

The average for these is 5 ccs. In the writer's experience large breeds (airedale, alsatians etc.) yield proportionately more than the small breeds. In dogs which, as patients, have been punctured and could not be included in the above list as strict "normals", amounts varying from 1.5 ccs in the adult pekingese to 9 ccs in the alsatian have been recorded. Elevation of the hind quarters or the head when the flow of cerebrospinal fluid has practically ceased will cause an additional flow of about 0.5 ccs.

(2) Rate of Flow. The rate of flow will of course vary with the type
of needle or canula used: needles of the same calibre were used throughout this work, so that the figures can be compared. The observations are set out as follows:

Dog II. 3 ccs. 30 secs; 4.5 ccs. 60 secs; total 5.5 ccs.
90 secs; fluid came very slowly in drops.

Dog I2. Similar to II.

Dog I3. 3.2 ccs. 30 secs; 3.6 ccs. 60 secs; 3.8 ccs. 90 secs; total 4.4 ccs. 120 secs.

Dog X. Total. 6 ccs. in 90 secs.

Dog Y. No record.

Dog Z. Total 5 ccs. in 120 secs.

The rate of flow accelerated by whining or struggling. These bring about their action through their effect on respiration. The efficient injection of local anaesthetic is therefore important.

Note. When the rate of flow is reduced to a few drops per minute, the fluid will be seen to oscillate within the needle. These movements are chiefly related to movements of respiration, and also to the heartbeat. Inspiration causes a decrease, and expiration an increase in pressure and hence oscillation of level in needle (20).

Many writers have made extended observations with electrical apparatus upon the rate of flow of cerebrospinal fluid from the dog by cistern puncture. Dixon and Halliburton (I) say, "in all instances the dogs were anaesthetised first with chloroform and then urethane and morphine were injected subcutaneously, the amount of urethane used being usually one grain per kilo body weight. No further volatile anaesthetic was required to retain complete anaesthesia throughout the experiment". The writers say, after describing cistern puncture by operation "as soon as the fluid is free to flow through the canula, it at first comes out with a gush; 2 or 3 ccs.
being passed in the first minute, but the rate of flow rapidly diminishes for the next two or three minutes and then more slowly. The drops occur at longer and longer intervals, but the flow continues in the slow manner for many hours. A few measurements were made of the amount which could be collected during the course of an experiment"...

"In the dog, the figures ran in successive 10 minutes as follows: 6, 1.7, 1.5, 0.8, 0.5, 0.6, 1.0, 0.7, 0.4, 0.6, 0.8, 0.8, 0.5, 0.4, 0.4, 0.3 C.C. The rises every now and then in the experiment were due to the administration of volatile anaesthetics. Ether and chloroform administration always produce an acceleration of the rate of flow and they were administered here not for anaesthetic purposes, but to make certain that the canula was clear." These figures are of interest, referring as they do to the dog, but are complicated by the fact that they were obtained under the variable effects of general and other anaesthetics.

3. Colour. Cerebrospinal fluid is a crystal clear fluid; contaminated with blood, e.g. from puncture, it is brown-yellow or in extreme cases pinkish. Nevertheless, crystal clear fluids may on examination be found to contain many red blood cells. A fluid containing many white cells has a whitish opalescence.


5. Cell Content. (a) White Cells. The number of cells per c.mm in the six dogs mentioned above was as follows:

No: 11, 1.7; No: 12, 2.1; No: 13, 4; X, 5.0; Y, 2.6;
Z, 3.2. The average is thus 2.1 white cells per c.mm.

(b) Red Cells. The presence of red cells has been discussed. In these particular samples they were absent on three occasions (dogs II, Y and Z); No. 12 had one, No 13, one; and X. 2 cells per c.mm giving an average of .66 cells per c.mm. for the six animals. The red cells examined were crenated.

Note. Though the white cells were usually of the small lymphocyte type, no further differentiation has been attempted as it was felt that a more precise method such as the ALZHEIMER technique would be necessary for such work. (See references 21,23,24).

SECTION G.

CLINICAL APPLICATIONS.

Cistern puncture constitutes a means of obtaining fairly large samples of cerebrospinal fluid for the diagnosis or investigation of disease processes associated with the central nervous system.

It forms a route for the injection of sera or medicaments into the sub-arachnoid space. Since many drugs given by the mouth or via the blood stream do not reach the cerebrospinal fluid, their action may be found to be enhanced when injected direct. The same may be found to apply to certain sera.
Experimental work upon, and the technique of, Puncture of the Cisterna Magna in the Dog, has been described. The Depth of the Cisterna Magna was investigated. With the head in position for operation, i.e., flexed at right angles, the depth at the Foramen Magnum was found to be 0.5 cm. in large breeds, and 0.3 cm. in the smaller breeds. In puppies it was about 0.2 cm. Factors likely to influence the depth were investigated such as pulsation of the medulla during life or passive movement of the head. None was found to modify the depth to any extent.

The Surgical Anatomy of the parts has been dealt with. The degree of development of the so-called "Occipital Shelf" was found to be of importance with relation to puncture and dogs have been divided into 3 types with regard to the development of this shelf.

The Surface Markings were defined and enable the operator to determine the relation of the 1-0 space to the body surface.

The Procedure for Puncture of the Cisterna Magna has been described. The technique is simple and is readily applicable in Clinical work. Though puncture is potentially dangerous a knowledge of the regional anatomy supplemented by practice on the dead animal reduces risks to a minimum.

The Effects of the Puncture have been studied. Clinically the patient appears unaffected. Several punctures performed at comparatively short intervals (e.g. 3 or 4 days) induced an in-
crease in the white cell content of the c.s.f. In the experimental animals it was shown that on the average the cell count increased to $10^4$ after four punctures had been performed at intervals of 3-5 days.

From the experimental data certain characters of the normal cerebrospinal fluid of the dog have been described. The amount obtainable by cistern puncture varied with the breed and individual, but was on the average 4-5 c.c. The rate of flow, colour and coagulability were also investigated. The average white cell count was shown to be 2.1 cells per c.mm. Red Blood cells were regarded as contaminant, but it was found very difficult to procure samples blood-free. Microscopical examination is the best test for the presence or absence of red blood cells in any sample.

The Clinical applications of Cistern Puncture are reviewed. Puncture is chiefly important as a means of obtaining comparatively large amounts of cerebrospinal fluid for purposes of diagnosis and/or for the injection of drugs or sera into the sub-arachnoid space.
EXPLANATION OF GRAPHS 1, 2 AND 3.

In each graph the figures on the upright represent the number of cells per c.mm. The figures along the horizontal refer to the day on which cistern puncture was performed, e.g. in the case of Dog II punctures were performed on the first, fourth, eighth, eleventh day from the commencement of the experiment.

Graph 1. The white cell counts of Dog II when subjected to a series of cistern punctures. B. indicates that a bacteriological exam. of the c.s.f. was made.

Graph 2. The white cell count of Dog II when subjected to a series of cistern punctures. The counts until the fifteenth day remained around the normal. The sample taken on the eighteenth day was rejected since the high blood content rendered the count negative (see table 2, 16-3-28). On the nineteenth day the count had risen to 185.5. On the same day a sample was taken for bacteriological exam. (B). On the twentyseventh day the count had returned to four. Since the fluctuation in the count between the fifteenth day and the first puncture on the nineteenth and between the latter and that on the twentyseventh day were not known the graph has been traced with dotted lines only.

Graph 3. The white cell counts of Dog II when subjected to several cistern punctures on the seventh day of the experiment two punctures were performed the second sample being used for bacteriological exam.

The results of the several bacteriological examinations are referred to in the text.
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PART TWO.

EXPERIMENTAL AND CLINICAL INVESTIGATIONS INTO THE USE OF IODIZED OIL AS A RADIOPAQUE MEDIUM IN THE EXAMINATION OF THE SPINAL SUBARACHNOID SPACE OF THE DOG.
PART TWO.

EXPERIMENTAL AND CLINICAL INVESTIGATIONS INTO THE USE OF IODIZED OIL AS A RADIOPAQUE MEDIUM IN THE EXAMINATION OF THE SPINAL SUBARACHNOID SPACE OF THE DOG.

SECTION A.

The characters and uses of Lipiodol Descendant (Lafay) and other iodized oils with particular reference to experimental and clinical work in animals.

(1). Physical properties of Lipiodol Descendant (Lafay).
(2). Chemical characters.
(3). Antiseptic properties.
(4). The several types of Iodized oils.
(5). The uses of Lipiodol and other iodized oils in the human subject, with notes on experimental and clinical work in animals.
(6). Persistence of Lipiodol in the body tissues.
(7). Elimination.
(8). Iodism following Lipiodol injection.
(9). Contra-indications to the use of Lipiodol.
(10). The reaction of tissues to Lipiodol.
(11). The relation between the withdrawal of cerebrospinal fluid, and a subsequent subarachnoid injection of Lipiodol.

SECTION B.

Experimental injection of Lipiodol Descendant (Lafay) into the subarachnoid space of the dog.

(1). Methods.
(2). Experimental injection of oil into the oisterna magna of five dogs, with observations on the meningeal reaction; and changes in the position and radiopacity of the oil.

(3). General conclusions with special reference to:
   (a), time of descent of oil into the subarachnoid cul-de-sac;
   (b), the conformation of the cul-de-sac;
   (c), adhesions occurring in the course of a descent;
   (d), movements of the oil after injection;
   (e), the effects of injection.

SECTION D
Lipiodol descendent and the demonstration of certain spinal lesions in the dog.

SECTION D
Clinical applications of the subarachnoid injection of lipiodol in the dog.

APPENDIX I
Certain features of the spine of the dog, the recognition of which is important in the reading of photographs of lipiodol injections.

SUMMARY.
CONCLUSIONS.
REFERENCES.
ACKNOWLEDGMENTS.
PART TWO.

SECTION A.

THE CHARACTERS AND USES OF LIPIODOL DESCENDANT (LAFAY) AND OTHER IODIZED OILS WITH PARTICULAR REFERENCE TO EXPERIMENTAL AND CLINICAL WORK IN ANIMALS.

In research work relating to human disease information derived from animal experimentation is of comparative value and that accruing from observations on man of direct value. In investigation into diseases of animals the reverse is true. Accordingly it has been thought appropriate to incorporate a necessarily brief review of the literature on the subject of iodized oils and their use in human work, setting out alongside any information available from animal experiments.


Lipiodol is a clear amber-coloured oil a little thicker than olive oil, with a specific gravity of 1.550. A drop sinks in water as quickly as a small stone (7), but floats if spread over the surface. The writer studied the behaviour of the oil when dropped into a test tube of fresh, warm cerebrospinal fluid. Formed drops of oil ejected from a lipiodol syringe (see later), sank at once to the bottom. Subsequent drops fused with the original if brought into close contact.
at the foot of the tube. This is of importance when injecting the oil into the cisterna magna of animals and will be mentioned later. Pirie (7) made the following experiment. "If a glass tube is made to taper to a fine point and filled with water to represent a bronchial static bronchicle full of pus or secretion and lipiodol is poured down it, the drug sinks through the water until it reaches the lumen of about 3 mm. and then stops with a convex lower margin". Further experiments on the movements of lipiodol were carried out by Contre-moulins (15) who took a long tube of irregular lumen, here narrow, here wide and filled with isotonic saline. He noted the propulsive effect of percussion on the descent of the drops and the counter effect of air bubbles.

Lipiodol will not easily displace pus in, say, a lung cavity, but tends to float on the surface. (28). Lipiodol in common with other iodised oils is opaque to X-rays. This is due to the high atomic weight of the contained iodine.

(2). Chemical Characters.

Lipiodol descendant is insoluble in water or alcohol. It is one of the so-called iodised oils being an additive compound of poppy seed oil and iodine, each cubic centimetre containing 0.54 gm. iodine (40% by weight). The iodine cannot be detected by the usual tests, but is released by:

(a) Exposure to light or air; a brown colouration indicates the liberation of iodine (3, 9, 11, 18), and oil exhibiting this change should not be used since the oxidation
products present are irritant to certain tissues.

(b) Heat; if the oil be heated iodine is liberated with the formation of copious purple fumes (6,19) when chemical tests for iodine are positive.

(c) The body tissues, iodine being excreted by several paths.

(d) The alkaline carbonates of the saliva and intestinal secretions. Stomach acids do not release iodine.

(3). The Antiseptic Properties of Lipiodol.

Neuswanger (17) smeared agar plates with lipiodol and then with B. coli, Streptococcus haemolyticus and Staphylococcus aureus. After incubation he noted organisms growing in close proximity to the globules of oil which appeared to exert no inhibitory effect. He exposed the oil, however, to air for months, but failed to note evidence of decomposition or the development of colonies of bacteria or fungi. He concluded that iodised oil does not possess bactericidal or bacteriostatic properties. Archibald and Brown (12) mixed lipiodol with sputum from a case of bronchiectasis. The mixture was kept at room temperature for 48 hours and then streaked on various culture media and incubated. A luxuriant growth was obtained including pseudo-diptheroides, pneumo-cocci, staphloccoci, and micrococcos catarrhalis. A second experiment of a different nature was performed. The writers conclude. "Therefore we believe that no trust can be placed in any presumed antiseptic action of lipiodol".
(4). Types of Iodised Oils.

Whilst the experiments referred to later were carried out with Lipiodol Descendant (Lafay) containing 0.54 gm. iodine per C.c. (40% by weight) references to many other iodised oils have been encountered and it was thought appropriate to tabulate them as under:

(a) Lipiodol Lafay, containing 0.45 gm. iodine per C.c., or 33% by weight of iodine.

(b) Lipiodol Ascendant (Light Lipiodol, Lipiodol Montant, New Lipiodol), evolved by Sicard, is made with olive oil and contains 11% of iodine. The specific gravity is less than that of cerebrospinal fluid. (5,70).

(c) Iodumbrin (Danish = jodumbrin), produced in Denmark. Contains 0.54 gm. iodine per C.c. i.e. 40% by weight (21); more fluid than lipiodol with equal local tolerance and X-ray opacity (22,71).

(d) Iodipin (German = Jodipin) (1,12,13,17,22,23,72,73,74) is a German preparation of sesame oil. There are two types; one contains iodine to the amount of 40% by weight and the second 10% by weight. Ayer and Wixter (22) employed 40% Iodipin and refer to "our experience with the French preparation of Iodipin" implying that this iodised oil is also of French origin.

(e) Belfield and Holnick (24) elaborated a combination of Cod Liver Oil and Iodol and Cod Liver Oil and Thymol Iodide, a contrast medium being prepared by mixing these; the mixture is stated to be sufficiently fluid to fill the vast deferentia, absorbable and of fair opacity but less so than
collargol or lipiodol.

(f) Collargol is mentioned by Belfield (24).

(g) Neuswanger (17) quotes Winternitz (Deutsche Med. Wochenschr. 1897, xxiii, thorp: Beil 33), who describes the preparation of an iodised oil using animal fats.

(h) Iodoleine is mentioned by Neuswanger (17) as a French product of poppy seed oil and iodine (30% approx.). The same writer refers to Lipiodol as being a French product of carnation oil combined with iodine to 0.54 gm. per cc. This, probably a misprint, is intended to infer that iodoleine is the carnation oil compound.

(i) A 40% mixture of Lipiodol with Amylal is mentioned (5). This mixture is very fluid and used for injecting the vasa deferentia; a mixture of equal parts of Lipiodol and Amylal is recommended by the Lafay Laboratories for a similar use.

(j) A mixture of Lipiodol and sterile olive oil is recommended for injecting into the urethra (5).

(k) Dominal X is mentioned by Stahl and Müller (25) as having been used by Von Wartenburg in dogs.

(l) Iodothionic IO%(Bayer) is referred to by Stahl and Müller (25) as having been used by Von Wartenburg in dogs.

(5) The Uses of Lipiodol and other iodised oils in the human, with notes on experimental and clinical work in animals. Iodised oils have been in use therapeutically for twenty-five years (7) prior to the elaboration of the French Lipiodol by Lafay in 1900 (26, 27).
Lipiodol Lafay is mentioned as having been employed therapeutically in February 1921 (28), but was first employed as a radiopaque medium by Sicard and Forestier in October 1921. Their work carried out on animals (3), and continued as epidural injections in the human, was published in 1921 (29). In this article the first reference to subarachnoid injection is made the authors remarking "......for in the event of accidental puncture of the subarachnoid space the cerebrospinal fluid tolerates the lipiodol remarkably as we will shew in a future communication". This is followed in 1922 by observations on the use of lipiodol injected into the subarachnoid space. (30). In the same article and elsewhere (3) Sicard and Forestier say that they have introduced lipiodol into the lung and obtained pictures of the bronchi. Thus as is claimed by some (23,32) Sergent and Cottenot (May 11th, 1923) were not the first to use lipiodol endo-bronchially. Armand and Delille (33) followed with bronchial investigations in children. In 1925, Lafay (II) introduced an oil containing 0.45 gm. iodine which was used by Sicard and Laplane.

At this stage the uses of Lipiodol as an opaque medium were variously developed, as for example in the radiological investigation of serious cavities, uterus, fallopian tubes, sinuses and fistulas, stomach digestion, the pericardial sac and as an intravenous injection. The tissue-tolerance of lipiodol rendered it more or less useful in all these spheres.

Lipiodol, and iodised oils in general, have been used in the lower animals as follows:-

Experiments in the DOG were recorded in 1923 by Sicard and Forestier (34) who investigated the movements of lipiodol when injected into
the femoral vein, portal vein, femoral artery and the carotid artery.

In 1924 Stahl and Muller (25) remarked that in German literature there was only one reference to iodipin injection into the lumbar canal of animals (and man) this having been carried out by L. Hirsch. The same authors refer to the injection of Dominal X and Iodothional (Bayer) into the lumbar canal of the dog by Von Wartenburg. Neuwanger (17), in 1926, injected lipiodol into the ureters and renal pelvis of the dog and studied the effect of this. Later he injected various amounts of 40% iodised oil intravenously and tabulated the results and post-mortem findings. In 1927 Kennedy and Baird (35) reported the clinical use of lipiodol as a radiopaque medium for delineating the uterine cavity in the bitch. No reference to the injection of lipiodol into the Cisterna Magna of the dog was found.

In the CAT Ayer and Mixter (22), in 1924, conducted experiments with iodipin (poppy seed oil containing 0.54 gm. iodine per cc = 40% by weight) and similar oils. Amounts of 1 cc and 1.5 cc were injected into the Cisterna Magna replacing equal amounts of c.s.f. The results of their experiments are referred to later and are of considerable interest. In 1925 Forsdike (19) injected lipiodol into the Fallopian tubes and the peritoneal cavity of the cat examining the results post-mortem at periods of three days to six weeks after the experiment. Archibald and Brown (12) in 1927 reported experimental endo-bronchial injections in the cat.

With regard to work in the RABBIT Sicard and Forestier (19) in 1924 mention intra-pulmonary injections. Two years later Lindblom (43) records the results of sub-dural injection of lipiodol. Devé (38) in 1927 injected lipiodol into the rabbit to estimate the effects of
the oil on experimentally induced subcutaneous hydatid cysts (Coenurus serialis).

In the GUINEA-PIG Sicard and Forestier (18) in 1924 investigated experimental endo-bronchial injections.

Finally in the RAT, Foredike (19) in 1925 studied the effects of Lipiodol injections into the fallopian tubes and peritoneal cavity.

It is seen that in the lower animals the use of iodised oils generally and lipiodol in particular have, with one exception, been experimental whilst no record of subarachnoid injections in the dog from the clinical standpoint has been found.

(6). Persistence in the tissues.

Lipiodol persists for a variable time according to the tissue or body cavity in which it is located. Subcutaneous tissue retains it longer than muscle (118), the epidural cavity longer than the subarachnoid space; serous surfaces imprison it for a shorter interval whilst the bronchial tree gets rid of it most easily.

The following figures summarise the observations in man.

(a) Subarachnoid space; 2 years or longer (11, 25, 40, 41).

(b) Peritoneal cavity; 7-10 days (19); elsewhere stated (42) as persisting for months.

(c) Fallopian tubes; several months (42).

(d) Lungs; 65 days (7); seen in lung 3 months after injection (43); 2 months (44); total elimination in 2 weeks (44); 15 months but with average of a few weeks (12); elimination after a few days, "il n'en reste plus trace" (45).
Three references were found to persistence in animals. The presence of iodipin in the subarachnoid space of cats is described 9 weeks after injection apparently unchanged (22). A reference (6) to the reaction following subarachnoid injection into the rabbit concludes that the reaction subsides "even though lipiodol does persist for weeks".

Oil injected into the ureters of dogs was found to have passed out within twelve hours (17).

(7). Elimination.

The iodine of lipiodol located in any of the tissues of the human body is eliminated more or less in the urine as a dialysable compound and in all probability in the mineral state (39). Lipiodol in the lung is expelled chiefly by expectoration (8,12,44,46,47,48), but so long as there are lung shadows elimination takes place via the urine to the amount of several milligrammes per diem constituting the so-called "lipodierèse pulmonaire" (8,9,39,18,44). The excreted iodine in such cases is stated to be in the form of an iodide detectable by the violet colouration obtained with nitric acid or with chloroform. If lipiodol be taken by mouth iodine appears in the urine.

The elimination of lipiodol resident in the subarachnoid space will be dealt with in some detail as being important from the comparative aspect of experiments recorded later in this paper.

Roger (II) referring to elimination after subarachnoid injection says it is an "elimination très lente de l'iode par lipodierèse
iodine exists only in traces in the urine and cerebrospinal fluid (30) and is detected by the usual tests for iodides. This urinary elimination has been studied in detail (39). Two cubic centimetres of iodised oil equivalent to 1gm.30gm. of metallic iodine were injected into the cerebrospinal fluid and the amounts of iodine in the urine estimated for twenty successive days. A maximum of four milligrammes was recorded on the second day in case 1, and one milligramme on the fourth day in case 2 respectively. With regard to the mode of liberation of iodine it may be remarked that the maximum iodine content of the urine corresponds to the highest polymorphonuclear counts in the cerebrospinal fluid following the injection (39). Sicard therefore suggests that the iodine is excreted most rapidly when the leucocytes which have pervaded the cerebrospinal fluid, and are loaded with iodine, are again in the blood stream and have liberated the metalloid with which they are charged. Stahl and Müller (25) recovered lipiodol from the subarachnoid space at operation and under the microscope observed emulsification but no admixture of cells.

Thus the precise mechanism of absorption of lipiodol and its iodine from the subarachnoid space of man is not known, but its route of exit from the body has been settled.

It is probable that elimination of lipiodol from various situations when injected into lower animals is similar to that in man but references to this are not numerous. Lindblom (6) in experiments on rabbits found variable sized droplets of oil taken up by the pial cells which may be another mode of resorption. Ayer and Wixter (22) studying
subarachnoid injections of iodipin (40%) in the cat state that absorption is extremely slow. One animal 9 weeks after receiving an injection showed most of the oil in the subarachnoid space. Neuswanger (17) states that the oil was found to escape very readily after injection into the urinary tract of dogs. X-ray examination 12 hours after injection showed no evidence of residual oil in the renal pelvis or ureter. Archibald and Brown (12) studying the oils in the lung of the cat found droplets situated in various lung tissues including cartilage.

Finally it may be mentioned that Binet injected various oil subcutaneously in dogs, guinea-pigs and rabbits. He found that resorption is the result of an accession of leucocytes which cause encystment and digestion of the oil. This gives some support to Sicard's theory of absorption from the cerebrospinal fluid (v.s.).

(8). Iodism.

Iodism has been recorded following the injection of lipiodol in the human subject (1, 5, 8, 12, 18, 50, 51, 52). It may occur under two conditions.

(a). When the oil is present in a site where the iodine is rapidly released such as in intestinal tract, or in the lungs where, by coughing, the lipiodol is expelled and then swallowed.

(b). Where an individual susceptibility exists (18, 50, 51, 53).

It has been suggested in man (30) that the iodine tolerance of patients be tested before a lipiodol injection into certain sites by administration of 15 grains of Potassium Iodide three times in the
course of one day. No reference has been found to iodism following the injection of lipiodol into sites other than those mentioned and, from what has been said about elimination and persistence, symptoms are unlikely to occur.

No writers have referred to iodism in animals following the use of lipiodol.

(9). **Contra-indications.**

In the human lipiodol injections are chiefly contra-indicated into the lung of a patient suffering from pulmonary tuberculosis or from asthma (7,8,12,51,52); and into the uterus at or near the menstrual period (55). Since a urinary excretion of iodine occurs it is not to be recommended when contracted kidney is present (46).

In animals contra-indications have not been recorded and can therefore be deduced only by analogy.

(10). **The reaction of Tissues to Lipiodol.**

Though lipiodol is well tolerated by all tissues, it is not without irritant qualities. These have been studied with reference to lung tissue in man (12,32,53), the pleural cavity in man (56), urinary tract in the dog (17), blood vascular system of the dog (17), and the uterus, fallopian tubes and peritoneal cavity of woman, cat and rat (19) but the vast majority of investigations have concerned the effect of lipiodol on the meninges when injected into the subarachnoid space of man or the lower animals.

Generally speaking most writers are agreed that in the human subject subarachnoid injection gives rise to a transient reaction with
cell increase in the cerebrospinal fluid and some local pain (6,11,25,37,58,59,60,61,62,63,64). Wbau and Vella (quoted 13) say that an aseptic chemical meningitis is set up.

Opinion varies as to the effects after the oil has remained in position for some time. French authors (57) have written "L'huile iodée a toujours été parfaitement tolérée utilement". McClaire (63), however mentions an instance where after 5 months residence the lipiodol became encysted and had to be removed by laminectomy.

Laplane (quoted II) records post-mortem findings in five subjects who had undergone subarachnoid injection from three to eleven months previously. After three months the lipiodol was said to be imprisoned "par un léger voile d'arachnoidite amorphe", after eleven months residence the oil was found to be fixed by a fibrous wall sending bands into the globule tending to resorb the oil. Thus lipiodol in the subarachnoid space of man appears to give rise to an immediate mild and transient irritation. The effects of long residence vary but in a few recorded cases unfavourable reactions have been encountered.

The results of subarachnoid injections in the lower animals have been studied in the CAT by Ayer and Wixter (22) in 1924, and by Wixter (37) in 1925. Lindblom (6) investigated injections in the rabbit in 1926.

Ayer and Wixter (22) used Iodipin containing 43% Iodine by weight. It is to be noted that these authors refer to Iodipin as a French product. The oil was injected into the cisterna magna in quantities of 1.0 - 1.5 c.cm replacing equal amounts of c.s.f. The authors conclude that the irritability of the oil was obvious in all
of the six animals injected with the French oil. The cats were usually "groggy" for one, two or three days. As an index of the irritation of the meninges it may be noted that in one cat the cell count in the spinal fluid was 4,420 mostly polymorphs on the third day after oil injection. The day after injection one cat showed a fluid containing 2,700 cells. One animal went into convulsions immediately after injections and died. At necropsy there was no evidence that the needle had entered the brain.

Wixter (37) reports the injection of 1-2cc Lipiodol in the cat. This dose, he says, corresponds to 20-40cc in man but does not state the basis for this estimation. A marked cellular reaction occurred in the c.s.f. This reached its height in 24-48 hours when counts of 1,000 cells and moderate increase in the protein content of the c.s.f. were noted. This subsided gradually and by the tenth day the fluid was normal. There is no mention of the number of times cistern puncture was performed on these animals. Lindblom (6) injected Lipiodol subdurally (subarachnoidally?) in the rabbit, and made microscopical examinations at varying intervals after the injection. Rabbits in two or three weeks after the injection were distinctly un lively and took little food. The head was bent back and the ears laid along the body. One rabbit died in the position of opisthotonous. Histological examination of the rabbits showed that an acute leptomeningitis was set up evidenced by the infiltration of Leucocytes. This subsided two or three weeks after the injection.
a subsequent subarachnoid injection of lipiodol.

Many writers advise, in human work, that no subarachnoid injection of lipiodol be performed unless a certain period has elapsed if subarachnoid puncture has been previously carried out with or without withdrawal of cerebrospinal fluid (5, 11, 13, 40, 52, 65, 66). It is maintained that the loss in volume of cerebrospinal fluid occasioned by the puncture and post-puncture leakage may conceivably bring about a false arrest of lipiodol between the first and fourth thoracic vertebrae where the spinal canal is anatomically narrower (5). The period which should elapse between the injections varies with different writers from a few hours to 10 days, the majority advising 7 days. In some instances this refers to lumbar as opposed to cistern puncture. No indications have been found with reference to this in the lower animals, but since post-puncture leakage probably occurs after cistern puncture in the dog (see Part One of this paper) and the subarachnoid space is of comparatively small dimensions similar precautions should be adopted.
SECTION B.

THE EXPERIMENTAL INJECTION OF LIPIODOL (LAFAY) INTO THE SUBARACHNOID SPACE OF THE DOG.

From the above review of the general characters and uses of, and experiments concerning, lipiodol and other iodized oils it will be seen that no reference has been found to their use in demonstrating the patency of the subarachnoid space in the dog, nor are there any details, likely to be a safe guide in clinical work, of the reaction following a subarachnoid injection.

The following experiments were accordingly undertaken to ascertain:

(a) If iodized oil could be used in clinical work as a radio-opaque medium to test the patency of the subarachnoid space in the dog and if so to set up figures for the rate of movement and the type of shadows depicted in the normal dog.

(b) If any reaction followed the injection of the oil.

(c) The changes, if any, in the position or radiopacity of the oil after residence in the animal tissues.

I. METHODS.

The experiments consisted in the injection of a known quantity of Lipiodol Descendant (Lafay), containing 0.54 g. of Iodine per c.c. i.e. 40% Iodine by weight, into the subarachnoid space of the dog.
by cistern puncture according to the technique described in PART ONE of this paper. The dog was then held in an upright position and observations made by means of X-ray photography on the rate of movement of the oil, the characters of such adhesions as occurred and the position ultimately assumed by the oil. Cell counts of the cerebrospinal fluid were determined at intervals after the injection as a criterion of any reaction whilst changes in the position or radiopacity of the oil were recorded photographically, at varying intervals.

(a) EXPERIMENTAL DOGS.

The dogs selected were males and where possible, of a breed having a spine of average length. Details of the type and measurements of each dog are given in Table 5. A clinical and X-ray examination was made before accepting an animal for experiment. The clinical examination was the usual one for general health and included observation for one month, during which the management of the dog was personally supervised and records kept of appetite, behaviour at exercise and temperatures. The usual examination of faeces (parasite eggs) and urine were carried out. The X-ray examination comprised lateral and ventro-dorsal photographs of the spine and cranium to detect possible bony or other abnormalities.

Three of the animals were kept for several months after the injection (March-October 1928) and were then looked after by a trained kennel-man from whom monthly reports were received until the writer took over management again for the last fourteen days of the experiment.
(b) THE INJECTION AND PHOTOGRAPHIC RECORD OF DESCENT.

Food was withheld for 12 hours and an enema given one hour previous to the commencement of the experiment. A wrap of gamgee tissue was placed round the body and over this a canvas corset affixed. From the ventral and dorsal sides of the corset, straps ran in four directions to attach to the corners of a light rectangular board so that the dog could be suspended vertically.

The board consisted of a strong outer frame with a three-ply centre piece so that screening could be carried out, with the animal held to the board, in an upright position. Layers of wool were placed between the board and the dog so that he could rest comfortably. The legs also were held lightly by bandages to the respective corners of the board.

The lipiodol was obtained in aluminium ampoules, a fresh consignment being used for each experiment. Any oil exposed to the air and not used at the time was discarded. Before use the oil was warmed to a temperature of 101.5°F. by immersion of the ampoules in a water bath. A special syringe was used for the injection. This so-called Lipiodol Syringe has a calibrated glass barrel and a rubber tube with needle adapter on the nozzle. The plunger can be moved in either direction by a screw attachment which ensures a uniform pressure and rate of injection. The syringe, warmed by wrapping in hot saline swabs, was filled with the oil. The exclusion of air bubbles from the oil during this procedure was a matter of some difficulty. It was found to remove the plunger and fill the barrel by pouring slowly down the inner glass face. The plunger was then carefully replaced, the
syringe reversed and the large supernatant air mass expelled. The syringe was then laid in a bath of sterile saline at 101.5°F. The proper conduct of this stage is essential to a successful descent of the oil after injection. In preliminary trials it was proved that air bubbles in the oil, a cold syringe or oil at low temperature gave rise to false arrests of the oil in the S-A. space.

A cistern puncture was performed with the dog wearing the corset and lying on its side on the board but not fastened to it. The latter was inclined at about 15° to the horizontal so that the animal's head was at the higher level. If this were not done, oil tended to flow forwards at the time of injection into the anterior part of the cisterna magna and finally congregate in the cerebral arachnoid spaces. Care was taken to prevent the escape of cerebrospinal fluid between the tapping of the cistern and connection of the syringes. In most instances by covering the needle with the thumb not more than four or five drops were lost. The technique of puncture have been described in Part One.

The oil was injected quite slowly (about 1.00 in two minutes) so that internal pressures might adjust themselves. Immediately after injection the patient was rolled gently onto its back with the head extended and held in that position for some minutes. It is believed that in this position the oil droplets tended to fuse into a mass on the roof of the cistern and favour a more rapid descent later. This view is based on an observation referred to under Section A, I. At this stage the animal was secured comfortably on its back to the board which was then placed in an almost vertical position. With appropriate
positioning of the X-ray tube movement of the oil could be studied on the fluorescent screen in a darkened room, the rays passing easily through the three-ply wood. Photographs were taken at intervals by laying the board upon a sloping table so that the hind-quarters of the dog were a little lower than the head. This is important since the oil runs forward in certain parts of the spine if the dog be held horizontally (see text later). In this position the tube was affixed over the dog, cassettes slipped between the animal and the board and ventro-dorsal photographs obtained. Lateral views were not obtainable without releasing the dog and so were only taken at the conclusion of each experiment.

(a) THE STUDY OF THE TISSUE REACTION.

Samples of cerebrospinal fluid were obtained by cistern puncture at intervals after the injection. The fluid was collected and the cell count made by the methods described in Part One; Section (b). The observations are set out in tables 6, 7, 8, 9, 10. Each table has several columns which are arranged in an exactly similar manner to that adopted in Part One of this paper in the study of the effects of several cistern punctures.

(d) THE STUDY OF THE CHANGES IN POSITION AND RADIOPACITY OF THE OIL.
This consisted in procuring X-ray photographs of the oil at certain periods after injection.

(e) EXPERIMENTAL PROGRAMME.

The plan of work is indicated in Table 5. Five dogs numbered 5, 6, 7, 8, and 14 were used. The table indicates the breed and spinal
measurements of each dog, and the dates of the preliminary X-ray examination, lipiodol injection, subsequent X-ray examination and death respectively. The dates of cistern puncture carried out to ascertain the cell counts of the cerebrospinal fluid are set out in tables 6, 7, 8, 9, 10. The original X-ray negatives obtained during the experiments are in the possession of

Throughout the experimental observations the numbers of the negatives under discussion are indicated. The figures referring to the experiments are prints from selected original negatives.

Before setting out the results, some indication will be made of abbreviations adopted. The Subarachnoid Space and the Cerebrospinal Fluid are designated respectively the S-A space and the c.s.f. The subarachnoid space has been divided arbitrarily into "segments" according to the vertebra within the boundaries of which a particular part of the space lies. In the thoracic region for instance, there are thirteen such divisions or "segments"; the first division lying within the limits of the first thoracic vertebra, the second within the second and so on. At the same time the vertebrae themselves have been designated by numbers. Thus, T.3, L.5, Cc.2, would signify the third thoracic, the fifth lumbar, and the second coccygeal vertebra respectively. In the case of the sacrum the lettering S.1, S.2, S.3, indicates the first (anterior), second and third (posterior) bony segments, a fusion of which the bone morphologically represents. This arrangement facilitates the description of the negatives. A globule of oil resting in the subarachnoid space at the level of the middle part of the
third lumbar vertebra is referred to as "a globule situated at the middle of L.3."

Other abbreviations have been used. The C-T junction, T-L junction, L-S junction and S-Cc denote the region of the last cervical and first thoracic vertebra, the last thoracic and first lumbar vertebra, the last lumbar and sacrum and the sacro-occipital junction respectively. The inter-arcual space between the first lumbar and the sacrum is similarly designated the L-S space.

The negatives have been termed Ventro-dorsal (V-D) or Lateral (lat). In the ventro-dorsal photographs the rays have passed from the ventral abdominal wall to the spine and on to the cassette the tube, of course, being on the abdominal side.
TABLE 5.
Description of Dogs and the Experimental Programme.

<table>
<thead>
<tr>
<th>DESCRIPTION AND SPINAL MEASUREMENTS FROM OCCIPUT TO LUMBO-SACRAL JUNCTION</th>
<th>PRELIMINARY PHOTOS TAKEN OF SPINE</th>
<th>LIPIODOL INJECTION</th>
<th>FURTHER REMARKS</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>LABRADOR,♂ ADULT, CURLY-COATED. LENGTH 65 cm.</strong></td>
<td>2.1.28. under morphia</td>
<td>15.1.28.</td>
<td>Destroyed (morphia-chloroform) 1.2.28. photographs of lipiodol in situ taken 2 hrs. after death.</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td><strong>AIREDALE,♂ AGED 2 YEARS. LENGTH 60 cm.</strong></td>
<td>16.1.28. under morphia</td>
<td>22.1.28.</td>
<td>Photographs were obtained again on 24.3.28. and 4.10.28. Destroyed (morphia-chloroform) in October 1928.</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>TERRIER-CROSS,♂ ADULT. MIDDLE-AGE. LENGTH 45 cm.</strong></td>
<td>23.1.28.</td>
<td>29.1.28.</td>
<td>Photographs of lipiodol in situ 31.3.28. and 2.10.28. Destroyed (morphia &amp; chloroform) in October 1928.</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>AIREDALE,♂ ADULT. MIDDLE-AGE. LENGTH 58 cm.</strong></td>
<td>1.2.28. under morphia</td>
<td>5.2.28.</td>
<td>Photographs of lipiodol in situ 10.3.28. Under morphia &amp; chloroform. Respiration ceased at end of photography. Attempts at resuscitation failed.</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>KERRY-BLUE CROSS,♂ AGED 15 MONTHS (APPX.) LENGTH 55 cm.</strong></td>
<td>29.2.28.</td>
<td>3.3.28.</td>
<td>A second series of photographs with lipiodol in situ 2.10.28. Destroyed (morphia &amp; chloroform) October 1928.</td>
</tr>
</tbody>
</table>
**EXPERIMENT I.**

**DOG No. 6. Airedale, aged 2 years, length 60 cm.**

**THE CLINICAL EXAMINATION** was satisfactory.

**RADIOLOGICAL EXAMINATION** of the spine; stereoscopic photographs were obtained under morphia and chloroform numbered as follows:

- 50 (paired), 51 (paired), 52 (paired), 53 (paired), 53 and 54, 55 (paired), 56 (paired), 57 (paired).

No abnormalities were detected.

**THE INJECTION** was performed on 22.1.28, the descent being recorded on negatives numbered 58-77 inclusive.

<table>
<thead>
<tr>
<th>Time</th>
<th>Total Suspension Time (in minutes)</th>
</tr>
</thead>
<tbody>
<tr>
<td>11.45 a.m.</td>
<td>0</td>
</tr>
<tr>
<td>11.55 a.m.</td>
<td>0</td>
</tr>
<tr>
<td>12.25 p.m.</td>
<td>30</td>
</tr>
<tr>
<td>12.50 p.m.</td>
<td>30</td>
</tr>
<tr>
<td>1.50 p.m.</td>
<td>90</td>
</tr>
</tbody>
</table>

2.5 cc Lipiodol Descendant (Lafay) containing 0.54 g. of iodine per c.c. were injected by cistern puncture. The mass of oil, seen in figure 16 (negatives 58 & 59) extended roughly from within the F. magnum backwards to the anterior border of the arch of C.2 and below the cord along the whole length of C.2. The V-D. negative 59 illustrated clearly how the droplets at anterior end of the oil mass were merging.

Dog placed upright.

V-D. negatives 60, 61, 62, 63. The oil had passed down to the level of the L-S junction and occupied the canal mainly from there to the level of L.4. The lower tip of the shadow was blunt. Droplets of oil temporarily arrested were seen chiefly in the cervical region. They were globular in form and situated in all positions around the cord. The cisterna magna was free of oil with the exception of a few small dots within the rim of the F. magnum.

Dog placed upright.

A lumbosacral V-D. negative number 64 was obtained with the dog vertical, and a second, number 68, when
the animal was placed upon a sloping table. These
shewed the oil chiefly within L.5, L.6, and L.7. At
the level of the L-3 junction the cul-de-sac be-
came suddenly narrower and was continued as a fil-
iform prolongation within the sacrum to level of
junction of S.1 and S.2. bony segments.

The animal was now left for 30 minutes in a
HORIZONTAL position to ascertain if, in this posi-
tion, the oil would move towards the head again.

After 30 minutes photographs shewed (negatives
No. 66 and 67) that the oil has moved forwards along
the length of L.3, L.2, L.1 and T.13. It is there-
fore apparent that whenever photography is being
carried out the subject should be kept on a slight
incline with the fore-quarters and head at the high-
er level.

Adhesions at this stage were found in the cervi-
cal region chiefly at the level of C.6 and C.7.

3.15 p.m. 90.
Dog placed upright.

3.50 p.m. 125.
Negatives were obtained as follows:

(a) V-D. 69-73 inclusive.
(b) Lateral 74-77 inclusive.

At this stage the experiment was concluded.

A description of the cul-de-sac shadow and remaining adhesions will be
dealt with in detail.

(1) Cul-de-sac. (see figures 16 and 17). The cul-de-sac was filled
with oil and outlined from the level of L.4 backwards. Its shape was
roughly that of a pencil with the point directed towards the tail. Un-
til the middle of L.5 the diameter remained constant and then commenced
to diminish uniformly throughout L.6 and L.7, so that at the level of
of the posterior border of L.7 the shadow was only one-third the
breadth that it was at L.5. Passing across the L-S space the calibre
of the cul-de-sac diminished suddenly so that it continued into the
sacrum as a thread-like prolongation reaching to the level of the
junction of the middle and posterior thirds of the sacral bony canal.

The borders of the shadow were not regular; in negative 69, and
less so in 73, small filiform or globular projections, directed out¬
wards and backwards, occurred on the right side throughout L.5, L.6 and
L.7. Posteriorly they became more numerous. In L.5 there were two
on the right side and suggestions of corresponding projections on the
left side at the same level. The projections on a given side were
thrown into relief if an oblique photograph were taken. After some
study of the incidence and form of these shadows it has been conclud¬
ed that they represent insinuation of oil along the nerve roots. This
is suggested because the projections,

(a) tend to show a paired arrangement.
(b) are disposed uniformly.
(c) increase in number posteriorly.
(d) do not occur in photographs taken from the lateral aspect.
(Compare figures 16 and 17)
(e) Those of a given side are thrown into relief in an oblique
photograph. It may be recalled that the nerve roots in
this region leave the dura at its VENTRO-lateral aspect so
that an oblique view would tend to throw into relief their
points of emergence. This is illustrated in several neg¬
atives obtained during the experiment.

From the lateral aspect, in figure 17, the cul-de-sac had the
characters of a finely tapering cone. The diminution in calibre was
comparable to that described in the ventro-dorsal photographs. It
commenced about the level of the middle of L.5 and continued uniform¬
ally to the level of the L-S space. Here a sudden reduction in cali¬
bre occurred and the sac continued as a fine thread. The cul-de-sac
tended to follow the inside border of the curve in the bony canal which occurs at the L-S junction. Thus it lay in close relation to the roof of the canal and so to speak "cut across" the curve. The result was that whilst the dorsal border of the shadow followed the roof, the ventral border gradually receded from the floor until at the level of the L-S junction the whole of the sac shadow lay within the upper third of the spinal canal. This fact will be referred to later as having a bearing on so-called lumbar-punctures in the dog.

(2). The Adhesions. Droplets of oil were adhering as follows:

1.3. Globules about middle of body dorsal to cord. Larger masses at level of anterior border of body dorsal to cord.


T.1, C.6, C.5, C.4. Several small globular adhesions.

C.3. Three faint adhesions to the left side of the cord.

Axis C.2. At this level there were three globules. One small one over the cord at the level of the C.2 - C.3 junction, and a similar one ventral to cord half-way along body of C.2. A larger irregular blob lay in the mid-dorsal line at the level of a transverse plane passing through the anterior border of the vertebral arch. In this and later experiments the adhesion occurred quite regularly and has been termed the "axis adhesion" because of its constancy of position in relation to the axis. Figures 44 and 45 illustrate its appearance.

Within the skull one globule was detected to the right of the mid-line in the region of the cerebello-medullary angle.

A study of the photographs showed that during the course of the
experiment these adhesions had not remained stationary. Slight move-
ment and change of form was detected in all of them. In photographs
taken some months later their position had changed considerably, and
for these reasons they may be termed "temporary adhesions" as opposed
to the "false arrests" referred to in human work. The large adhesion
mentioned as lying dorsal to the cord at the level of the anterior
border of the axis arch remained stationary throughout the whole
suspension, but two months later had disappeared.

The Affects of the Injection.

A. CLINICALLY the dog remained unaffected. He was kept under observ-
vation until October 1928, a period of 6 months. The animal had
perfect co-ordination of movement and could leap on to and off
tables and over obstacles. Passive movements of the neck and spine
gave no pain; urine and faeces were passed normally. The appetite
was good and the animal always bright and interested. The rectal
temperature was recorded each day before feeding at 9 a.m. and 5 p.m.
A rise occurred in the three days following the puncture, but after-
wards fell to normal. Cistern puncture for c.s.f. examination was
being carried out at this time but since this procedure has been
shown to have no effect on the temperatures (see PART ONE of this
paper) the slight fever must be attributed to the Lipiodol injection.
The temperatures for three weeks succeeding the injection are in-
dicated on Chart 8.
## TABLE No. 6

Cell Count of the c.s.f. of Dog 6 after a Lipiodol Injection.

<table>
<thead>
<tr>
<th>Date</th>
<th>Interval between punctures (in days)</th>
<th>Red cells present per cubic millimetre</th>
<th>Whites introduced proportionately with R.B.C.s.</th>
<th>Total Whites per cubic millimetre</th>
<th>Actual Whites per cubic millimetre</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>22.1.28.</td>
<td>--</td>
<td>Lipiodol injected</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>22.1.28.</td>
<td>6 hours</td>
<td>570</td>
<td>1.5</td>
<td>14</td>
<td>12.5</td>
<td>Two polymorphs seen in 3.2 c/mm.</td>
</tr>
<tr>
<td>23.1.28.</td>
<td>1 day</td>
<td>3,845</td>
<td>8.5</td>
<td>912</td>
<td>903.5</td>
<td>Several polymorphs present. Punctured on 2nd attempt but needle passed near bony boundary.</td>
</tr>
<tr>
<td>24.1.28.</td>
<td>1</td>
<td>523</td>
<td>1.5</td>
<td>444</td>
<td>442.5</td>
<td>Repeated attempts to puncture before success; at one time pure blood</td>
</tr>
<tr>
<td>26.1.28.</td>
<td>2</td>
<td>3,570</td>
<td>8</td>
<td>126</td>
<td>118.0</td>
<td></td>
</tr>
<tr>
<td>27.1.28.</td>
<td>1</td>
<td>415</td>
<td>1</td>
<td>70.8</td>
<td>69.8</td>
<td></td>
</tr>
<tr>
<td>1.2.28.</td>
<td>5</td>
<td>66</td>
<td>-</td>
<td>340.75</td>
<td>340.75</td>
<td></td>
</tr>
<tr>
<td>4.2.28.</td>
<td>3</td>
<td>124</td>
<td>-</td>
<td>360.6</td>
<td>360.8</td>
<td></td>
</tr>
<tr>
<td>5.2.28.</td>
<td>4</td>
<td>73.7</td>
<td>-</td>
<td>98.7</td>
<td>98.7</td>
<td></td>
</tr>
<tr>
<td>10.2.28.</td>
<td>2</td>
<td>567</td>
<td>1.5</td>
<td>109.6</td>
<td>108.1</td>
<td></td>
</tr>
<tr>
<td>15.2.28.</td>
<td>5</td>
<td>174</td>
<td>-</td>
<td>74.2</td>
<td>74.2</td>
<td></td>
</tr>
<tr>
<td>18.2.28.</td>
<td>3</td>
<td>284</td>
<td>1</td>
<td>287</td>
<td>286</td>
<td></td>
</tr>
<tr>
<td>22.2.28.</td>
<td>4</td>
<td>21</td>
<td>-</td>
<td>55.9</td>
<td>55.9</td>
<td></td>
</tr>
<tr>
<td>1.3.28.</td>
<td>6</td>
<td>12.5</td>
<td>-</td>
<td>20.6</td>
<td>20.6</td>
<td></td>
</tr>
<tr>
<td>14.3.28.</td>
<td>13</td>
<td>0</td>
<td>-</td>
<td>13.7</td>
<td>3.7</td>
<td></td>
</tr>
<tr>
<td>3.10.28.</td>
<td>203</td>
<td>0</td>
<td>-</td>
<td>1.5</td>
<td>1.5</td>
<td></td>
</tr>
</tbody>
</table>

Note: The high red cell content when the puncture proved difficult or when the needle passed near to the bone. This has been discussed in PART ONE of this paper.
B. CHANGES IN THE CEREBROSPINAL FLUID.

The white cell count exhibited a sudden rise to 900 cells within 24 hours of the injection. As has been shewn, (see Part One) a cell rise can be induced by cistern puncture alone, but the increase here is much in excess of what puncture, per se, has been observed to cause. Hence the Lipiodol itself must be held responsible for the rapid primary increase and to a greater or lesser extent for the high cell count that persisted until 14.3.28. As will be noted the count was within normal limits when estimated on 3.10.28.

Subsequent movements of the Lipiodol.

X-ray photographs were obtained 8 weeks and 27 weeks after the injection.

EIGHT WEEKS after the injection (negatives 207, 208, 209, 210, 211, 213, 214, 215, 216), new features were found in the connection with oil within the cranium and the lumbar cul-de-sac.

IN THE CRANIAL CAVITY many globules of oil lay along the base of the brain in, and on either side of, the mid-line (figures 20 and 21) (negatives 211 and 215), the majority were globoid but some were linear in form. The oil appeared to have migrated from the cervical and cervico-thoracic region since the globules that were present here had disappeared with the exception of a few at the level of C.7 and T.1.

The appearance of THE CUL-DE-SAC is shown in figure 18. The general outline previously described was still present; the shadow was, however, less dense and had broken into lines and streaks and globular formations, especially anteriorly. Within the sacrum a
new disposition obtained. At the conclusion of the descent experiment it will be recalled that the cul-de-sac shadow entered the sacrum as a filiform prolongation reaching as far as the 3-2 = 3-3 junction. Several variably shaped globules had now appeared both to the left of and dorsal to the cul-de-sac proper. These were situated apparently in the bony canal itself. A few droplets could be traced in the mid-line as far back as the level of Co.2 (see figure 18). The projections located previously along the lateral borders of the cul-de-sac shadow were still recognizable at the level of L.5 and L.6; elsewhere they were absent.

TWENTY-EIGHT WEEKS after the injection the oil was seen to be disposed in much the same manner as it was after eight weeks residence (see figure 19 and negative 1004, 1005, 1006, 1007). The splitting up of the main mass of the oil in the cul-de-sac was suggested by streaks and breaks in the shadow and was more marked than in previous stages. A careful comparison between conditions at 8 weeks and 28 weeks was made. Definitely less oil appeared to be present after 28 weeks residence.

Summary. During the weeks following the injection the lipiodol showed firstly, tendency to migrate to the base of the brain, and secondly a new disposition in the form of globules lying outside the termination of the cul-de-sac as outlined in the original descent experiment. This condition was verified ——: 28 weeks after the injection.

Conclusions. Pag. 6.

(1) lipiodol after injection into the cisterna magna was observed
in the posterior limit of the cul-de-sac in 90 minutes. It probably reached this position in less than 90 minutes.

(2) Temporary adhesions occurred at all stages of the experiment chiefly in the cervical and anterior thoracic regions.

(3) A mild temporary reaction was set up evidenced by a high cell count in the c.s.f. Clinically the dog remained unaffected.

(4) The oil underwent movements in the animal after the injection collecting round the brain and extending in the sacral region.
EXPERIMENT 2.

DOG, Number 14, KERRY BLUE cross, aged 15 months (approximately)

Length 55 cm.

The CLINICAL EXAMINATION was satisfactory.

The RADIOLOGICAL EXAMINATION of the spine was carried out on 180
29.2.28. (negatives 175, 176, 178, 179, 181) and showed no abnormalities.

The LIPIODOL INJECTION was carried out on 3.3.28. and was recorded
on negatives numbered 182-187 and 189-197.

Observations were as follows:-

<table>
<thead>
<tr>
<th>Time</th>
<th>Total Suspension</th>
</tr>
</thead>
<tbody>
<tr>
<td>3.25</td>
<td>0.5 cc Lipiodol Descendant (Lafey) containing 0.54 g Iodine per 0.5 cc was injected into the cisterna magna. The mass of oil (negatives 182, 183) rested around the cord within the atlas and on the right side within the axis. A large globule was situated in the anterior part of the cisterna magna to the left side; a smaller globule rested on the floor of the foramen magnum. The mass shadow appeared to be made up of fused globules.</td>
</tr>
<tr>
<td>3.30</td>
<td>0. The dog was placed upright.</td>
</tr>
<tr>
<td>4.50</td>
<td>35. Negatives 184, 115, 186. The oil had descended to L.I. The general descent resembled that in Dog No. 6. The majority of the adhesions occurred in the cervical region and were globular in form. In the thoracic area they were less numerous and linear becoming globular again on approaching the lumbar region. The largest arrest was situated at the level of the anterior border of the axis. (Compare Dog 6).</td>
</tr>
<tr>
<td>4.10</td>
<td>35. Dog placed upright.</td>
</tr>
</tbody>
</table>
| 5.10 | 95. Negatives 187, 189, 190. The oil had now reached the level of the junction of the middle and posterior thirds of the sacrum, and was filling the cul-
de-sac. The cul-de-sac was outlined from the level of the anterior border of L.6 and from here backwards underwent a gradual and uniform diminution in calibre being more pointed and slender than in the case of Dog 6. A break in continuity of the shadow was seen on the left side at the level of the L-5 space. As the photograph was taken in an oblique direction the long axis of the sacrum was thrown to the left and the cul-de-sac shadow consequently carried to the same side.

The adhesions were mainly located within C.6 and C.7 and also L.3, L.4 and L.5. A few droops were demonstrable here and there in the thoracic region whilst the large adhesion in the axis remained in the same position but changed in shape.

Dog placed upright.

Negatives 191-197. The cul-de-sac shadow (see figures 22 and 23), commenced within L.6 where its calibre was about one sixth that of the body of the vertebra. It ran backwards tapering uniformly to a fine line at the sacro-coccygeal junction. Here a slender prolongation connected it with an elongated irregular mass lying within the posterior two-thirds of Ca.2 and the first part of Co.3. The lateral photograph (figure 8) showed that the expansion of the oil shadow lay along the roof of the coccygeal canal and was made up of two parts. The larger and more anterior was situated chiefly within the arch of Co.2; a small break occurred at the level of the articulation between Co.2 and Co.3 to be followed by the second part lying within the arch of Co.3. The lateral photographs showed that the cul-de-sac tended to follow the inner contour of the curves in the bony canal. As the spinal canal inclined dorsally at the L-5 junction the shadow lay close up against the roof of the canal. The reverse was the case at the sacro-coccygeal junction.

The adhesions were disposed chiefly in the cervical region. A few globules lay within C.6 and C.7 chiefly to the right of the mid-line; the thoracic and first two lumbar areas were clear. Globules occurred within L.3, L.4, and L.5, and were of variable size and shape. The lateral photographs (negatives numbered 195, 196 and 197) showed them to be situated in all planes. The adhesion at the level of the
axis arch was dorsal to the cord and subjacent to the superior atlanto-axial ligament (lig.interar- cualis). As in Dog 6, it was the only adhesion which could be regarded as a false arrest. In photographs taken six months later it had disappear- ed so that it must be classified as a temporary adhesion.

Effects of the Injection.

A. CLINICALLY the animal was unaltered. It was kept under observation for six months and nothing abnormal in behaviour, mentality or co-ordination was noticed. The temperatures were recorded and those for the two weeks succeeding the injection are shown on CHART 6. A slight rise (103 °F.) followed the injection. A subsequent rise to 103.6 °F. was followed by a permanent fall. During this time cistern punctures were performed as indicated on the chart, for the purpose of examining the c.s.f.

B. CHANGES IN THE CEREBROSPINAL FLUID. Cell counts were made as indicated in Table 7. The Table has been set out as in the case of Dog 5, with remarks on the rate of flow added.

The white cell count increased to 301 after 24 hours and 348 with in 48 hours which is again higher than has been induced by cistern puncture alone and must be ascribed to the effect of the Lipiodol. The cell reaction is less than in the other experimental animals and may be associated with the small amount of Lipiodol injected and the lesser number of adhesions that occurred during the experiment. The count of 6.6 nine days after the injection is within the normal limits laid down in PART One of the Paper; in October the count had fallen to 2.5.
cells per cubic millimetre.

The fluid drawn on 4.3.28. was faintly opalescent. This has been noted as indicative of an increased white cell content especially in a sample comparatively free from blood contamination.

**Subsequent movements of the Lipiodol.**

X-ray photographs were taken 220 days after the injection (negatives numbered 1008-1071).

The oil in the cul-de-sac showed the following changes (see figure 24 and negatives 1003 and 1009).

(a) It had run forward to the level of L.4; previously it lay chiefly within and behind L.6.

(b) Although the general outline of the sac was retained, the shadow was irregular in density, the oil mass appearing to have broken up.

(c) Globules of oil appeared within the third sacral segment, Co. I, Co. 3 and Co. 4. In the sacrum and Co. I the globules were scattered over a comparatively wide area and were well outside the boundaries of the cul-de-sac as outlined in the original lipiodol injection and shown in figures 22 and 23. Two larger drops at the Co. 2-Co. 3 junction appeared to represent the dilatations in the cul-de-sac shadow seen in the original experiment (figures 21 and 22). A few linear shaped drops lay in the mid-line as far back as the posterior end of Co. 4. These new deposits of oil are irregular in size and show no tendency to ordered arrangement or alignment.

(d) The adhesions originally noted in the anterior lumbar and
the posterior cervical regions had disappeared. The arrested globule at the level of the axis had also moved or undergone absorption.

Summary. The oil within the cul-de-sac shewed changes in the shadow-density; new depositions of oil occurred within the sacrum and coccygeal vertebrae. 

During the weeks subsequent to the experiment the adhesions in the posterior cervical and anterior lumbar region tended to disappear. It is not clear in this animal if this was a result of absorption or mechanical migration.

Conclusions Dog 14.

(1). The passage of 0.5 cc Lipiodol from the cisterna magna to the cul-de-sac occupied 95 minutes. Certain extensions in the cul-de-sac boundaries developed as far as the level of Cc.2 and Cc.3 in the succeeding 90 minutes.

(2). Temporary adhesions occurred at all stages of the experiment typically in the region of the cervico-thoracic junction and middle lumbar regions; an apparently permanent arrest occurred within the axis but some months later this had disappeared.

(3). Clinically the dog remained unaffected except for a temporary rise in temperature. Cell counts of the c.s.f. indicated a temporary reaction which subsided after about 10 days.

(4). The oil later underwent movement and/or absorption and appeared to migrate outside the cul-de-sac in the sacral and coccygeal regions. The significance of this will be discussed later.
TABLE 1
Cell Counts of the c.s.f. of Dog I.4 after a Lipiodol Injection.

<table>
<thead>
<tr>
<th>Date</th>
<th>Interval between Punctures (in days)</th>
<th>Red Cells present per cubic millimetre</th>
<th>Whites introduced proportionately with R.B.C.'s</th>
<th>Total Whites per cubic millimetre</th>
<th>Actual Whites per cubic millimetre</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>3.3.28</td>
<td>4½ hours</td>
<td>817</td>
<td>2</td>
<td>6</td>
<td>4</td>
<td>6.4 C.W. counted for whites</td>
</tr>
<tr>
<td>4.3.28</td>
<td>1 Day</td>
<td>169</td>
<td>0</td>
<td>301</td>
<td>301</td>
<td>A few polymorphs present. Fluid showed white cell opalescence.</td>
</tr>
<tr>
<td>5.3.28</td>
<td>1</td>
<td>129</td>
<td>0</td>
<td>348.5</td>
<td>348.5</td>
<td></td>
</tr>
<tr>
<td>8.3.28</td>
<td>3</td>
<td>146</td>
<td>0</td>
<td>42</td>
<td>42</td>
<td></td>
</tr>
<tr>
<td>21.3.28</td>
<td>9</td>
<td>0</td>
<td>0</td>
<td>27.1</td>
<td>27.1</td>
<td></td>
</tr>
<tr>
<td>2.10.28</td>
<td>195</td>
<td>0</td>
<td>0</td>
<td>2.5</td>
<td>2.5</td>
<td></td>
</tr>
</tbody>
</table>
EXPERIMENT 3.

DOG. No.7, Terrier-cross, about 4 years of age. Length 45 cms.

CLINICAL EXAMINATION was satisfactory.

THE RADI OLOGICAL EXAMINATION of the spine was made under morphia and chloroform on 23.1.28 (negatives numbered 78-85 inclusive). No abnormality was found, with the exception of a lumbar rib (figure 25). The lipiodol injection indicated, and a post-mortem examination confirmed, an enlargement of the intervertebral disc between the last lumbar vertebrae and the sacrum, the border of the disc projecting from the floor of the spinal canal; the cul-de-sac was bent dorsally as it traversed this eminence. This is shown in figure 20, in which the cul-de-sac has been outlined with oil.

This condition however had very little effect on the descent of the oil so that the times and records obtained in the case of this dog have been included in this series of observations on the normal dog.

THE LIPIODOL INJECTION was carried out on 29.1.28, and was recorded on negatives numbered 86-99 inclusive and 102-105 inclusive.

<table>
<thead>
<tr>
<th>Time</th>
<th>Total Time of Suspension (in minutes)</th>
</tr>
</thead>
<tbody>
<tr>
<td>12:10</td>
<td>I.5 cc. Lipiodol Descendant (Lafay) containing 0.54 g. iodine per cc. was injected into the cisterna magna.</td>
</tr>
</tbody>
</table>

The oil filled the cisterna magna and extended back along the spinal s-a space as far as C.2 (negatives 86 and 87). Since the patient lay on the right side at time of the injection much of the oil has gravitated to the right side. (Negative 87).
Dog placed upright.

Negatives numbered 88, 89, 90, 91. The oil had reached the level of the third sacral segment and was filling the cul-de-sac. At the level of the L-S junction the shadow shewed a diminution in opacity and an indentation along the right border. The usual globular adhesions were present within C.5, C.6, and C.7 throughout the last few thoracic vertebrae and backwards to the cul-de-sac were globules of varied size and shape; within L.4 and the posterior part of L.3 a sheet-like adhesion stretched across the width of the canal.

The characteristic adhesion in front of the axis was present. (compare dogs 6 and 14).

Dog placed upright.

Figure 26 and negatives numbered 92, 93, 94, 95. The oil had descended to the level of the S-0c junction. The cul-de-sac, in the form of a tapering cone, was filled from the level of the anterior border of L.6 tapering uniformly to the end of the sacrum. Each border of this cone was studied at irregular intervals with slight dilatations. Some, but not all occurred at the same transverse level and hence suggested as in Dog 6 the insinuation of oil along the nerve roots. The deficiency in outline on the right side (figure 28, x) and in general density of shadow at the level of the L-S junction was still present. The adhesions elsewhere were less numerous and were located as usual within the anterior part of the axis throughout the last few cervical and early thoracic and in the lumbar region generally. The sheet-like adhesion in L.4 had moved on to leave a few large globules.

Dog placed upright.

With the dog upright a V-D photograph of the L-S area was taken through the 3-ply wood (negative number 96). Definition was poor but the conformation of the sac could be picked out.

The dog was placed in a HORIZONTAL position for twenty minutes to estimate the degree of mechanical backflow of the oil after entry into the cul-de-sac.
The adhesions elsewhere were few in number and occurred chiefly in the posterior cervical region, with the exception of the comparatively large mass lying dorsal to the cord at the level of the dorsal-atlanto-axial membrane.

**Effects of the Injection.**

A. **CLINICALLY** the animal appeared unaffected. He was observed until October 1928, a period of 6 months. The temperatures shewed the usual slight rise (see Chart 10) on the day after the injection with transient increases on the fifth and tenth day.
B. CHANGES IN THE CEREBROSPINAL FLUID. Samples of the c.s.f. were taken by cistern puncture and cell counts (see Table 8) made. Forty-eight hours after the injection the white count was 828. This varied somewhat until 15.2.28, ten days after the injection, when the count rose to 4,000. The fluid was opalescent but did not escape under increased pressure. A second confirmatory sample counted the same day contained 4,060 white cells and this was utilized for a bacteriological examination (see below). The dog remained quite normal and the temperature and the appetite were satisfactory. The counts then fell steadily so that on 15.3.28, twenty-nine days later the sample contained only 347 whites. Finally in October 1928 two hundred days afterwards the count stood at 1.2 cells per c.mm.

The details of the bacteriological examination of the cerebrospinal fluid on 15.2.28 were as follows:-

(A) Culture: Agar slopes; Blood-agar-broth; Glucose agar.
   All negative after 72 hours incubation.

(B) Slides were made from centrifugalised deposit and stained with methylene blue, by Gram's method and with Haematoxylin and Eosin. No organisms were found.

This dog shewed a particularly high cell count and one which occurred rather late after the injection. The count of 4,000 was noted after five cistern punctures had been performed in seventeen days at an average interval of three and one-half days. In the experiments on cistern puncture it was shown that on the average a white cell count of 110 per cubic millimetre followed four punctures at intervals of three and one-half days, and the highest count recorded in all the
### Table 8

Cell Counts of the c.s.f. Dog 7, after Lipiodol Injection

<table>
<thead>
<tr>
<th>Date</th>
<th>Interval between punctures in days</th>
<th>Red Cells present</th>
<th>White cells introduced to R.B.C.’s</th>
<th>Total White Cells</th>
<th>Actual Whites</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>31.1.28</td>
<td>2</td>
<td>489</td>
<td>1.5</td>
<td>828</td>
<td>826.5</td>
<td>Several polymorphs present.</td>
</tr>
<tr>
<td>2.2.28</td>
<td>2</td>
<td>4,500</td>
<td>10</td>
<td>272</td>
<td>262</td>
<td></td>
</tr>
<tr>
<td>4.2.28</td>
<td>2</td>
<td>540</td>
<td>1.5</td>
<td>889</td>
<td>887.5</td>
<td>3.4 c.mm. counted for whites.</td>
</tr>
<tr>
<td>8.2.28</td>
<td>4</td>
<td>142</td>
<td>0</td>
<td>426</td>
<td>426</td>
<td>Several polymorphs present.</td>
</tr>
<tr>
<td>15.2.28</td>
<td>7 Count (a) 1000</td>
<td>0</td>
<td>4,000</td>
<td>4,000</td>
<td>4,000</td>
<td>Both fluids showed &quot;white cell opalescence&quot;. The whites had to be counted in a Thoma Ziehl chamber.</td>
</tr>
<tr>
<td></td>
<td>Count (b) 50</td>
<td>0</td>
<td>4,050</td>
<td>4,050</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Bacteriological examination made from sample B.

<table>
<thead>
<tr>
<th>Date</th>
<th>Interval between punctures in days</th>
<th>Red Cells present</th>
<th>White cells introduced to R.B.C.’s</th>
<th>Total White Cells</th>
<th>Actual Whites</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>17.2.28</td>
<td>2</td>
<td>105</td>
<td>0</td>
<td>1,735</td>
<td>1,735</td>
<td></td>
</tr>
<tr>
<td>22.2.28</td>
<td>5</td>
<td>159</td>
<td>0</td>
<td>919.4</td>
<td>919.4</td>
<td></td>
</tr>
<tr>
<td>1.3.28</td>
<td>0</td>
<td>500</td>
<td>1.5</td>
<td>397</td>
<td>395.5</td>
<td></td>
</tr>
<tr>
<td>15.3.28</td>
<td>14</td>
<td>12</td>
<td>0</td>
<td>347.4</td>
<td>347.4</td>
<td></td>
</tr>
<tr>
<td>4.10.28.202</td>
<td></td>
<td>10</td>
<td>0</td>
<td>1.2</td>
<td>1.2</td>
<td></td>
</tr>
</tbody>
</table>
trials was 188 after six punctures performed in nineteen days. The counts in Dog 7 are thus far in excess of those that could be referred to cistern puncture alone and must therefore be attributed to the effects of Lipiodol in the absence of any intercurrent infection or disease of which one could find no evidence. An explanation of the fluctuation of cell counts is attempted later.

Subsequent movements and changes of the Lipiodol.

Radiological examinations were carried out on 31.3.28, sixty-two days after the injection, and in October 1928, two hundred days later. SIXTY-TWO DAYS AFTER THE INJECTION (negatives numbered 220, 221, 223, 224, 225, 226, 227, 228, 229, 230), three main changes had occurred.

1. Migration of the oil forward into the cranium with disappearance of the oil in the more anterior of the cervical vertebral.

2. Breaking up and moving forward of the oil in the cul-de-sac.

3. Apparent escape of oil from the limits of the subarachnoid space.

1. Migration of the oil forward into the cranium.

Oil had moved forward so that it rested at the base of the brain about the level of the body of the sphenoid bone and around the midline (figures 20 and 21, negatives 227 and 228). Five large and many smaller globules could be discerned. Globules originally in the cervical region (negative 105) had disappeared leaving a residuum in C.6, C.7, T.1, and T.2.

2. Breaking up of the oil in the cul-de-sac.

The negatives 220, 226, 229, 230 (see figures 30 and 31) showed the oil in the cul-de-sac to extend in broken and irregular masses and
globules from the L.I.-L.2 junction backwards to Co.5. The main mass of the oil lay within L.6-L.7 and the sacrum. A glance at figures 30 and 31 shews that the globules are situated in all positions around the cord but the majority lie along the dorsal and ventral silhouette of the cord which is accordingly outlined with more definition in the lateral than the V-D photograph.

(3). Apparent escape of the oil from the S-A space.

The negatives 220,226,229 and 230 and figures 30,31 and 32, shewed that from the level of the L.6-L.7 junction backwards throughout L.7 and the sacrum and again in Co.3 and Co.5 globules and streaks of oil had appeared outside the limits of the cul-de-sac as defined in the final photographs of the descent experiment. (Figures 28 and 29). These extensions will be dealt with in detail.

(a). In the V-D photographs (see figures 31 and 32 also negatives 229 and 230) a large globule of oil (A,figure 31 and 32) lies in each of the posterior angles of the vertebral shadow, that is, about the level of the intervertebral foramen. On the right side the oil has come to lie outside the limits of the shadows of the vertebrae proper as a streak (A',figures 30,31 and 32) directed backwards and downwards just dorsal to the right transverse process of L.7. The lateral view (figure 30, negative 220) shews the intervertebral foramina to be filled with an oil shadow. (A) and the lateral extension can be picked out. (A').

(b). Throughout L.7 masses have escaped and lie in an area limit-
ed only by the boundaries of the bony vertebral canal. Towards the posterior limits of L.7 the oil tends to align itself towards each posterior angle of the body shadow and to emerge finally from the spine. On the left side a series of droplets have issued and are directed downwards and backwards through the region of the intervertebral foramen. (J, figures 31 and 32). On the right side a track foil (B, figures 30, 31 and 32) can be traced backwards, outwards and downwards below the right wing of the sacrum along the ventral margin of the sacro-iliac articulation and then within the wing of the ileum to the level of the commencement of the neck of the latter. In its course the oil track is joined by two other fingers of oil (O, figures 30, 31 and 32) which come outwards and backwards from the sacrum. Each of these extensions emerges from the bony spinal canal in the region of the first and second ventral foramina of the sacrum. Within the sacral canal the oil is scattered in globules and lines of various sizes. On the right side there is a tendency to converge towards the region of the three sacral foramina to be continued outside the sacrum as described above. On the left side a similar concentration of the oil is discernible but none has escaped beyond the limits of the shadow of the sacral canal proper (D, E, F, figures 31 and 32).

In the coccygeal region three small globules can be picked out. These are within Co.1, Co.3 and Co.5. A suggestion of a continuous thread of oil between the globule in Co.1 (O, figures 30, 31 and 32) and that in Co.3 (H) can be made out in the negative (number 230) but is not discernible in the print.

IN OCTOBER 1928. (figure 33, negatives 1,000, 1,001, 1,002, 1,003)
the oil in the lumbo-sacral region had undergone very little modification in position. Unfortunately only one photograph, a ventro-dorsal, was available (figure 33). The oil lying outside the bony canal could be identified almost globule for globule. That within the bony canal had shifted a little in position and shape. The globules which were located in Co.1, Co.3, and Co.5 were still in position. A few globules were present at the cervico-thoracic junction disposed similarly in shape and position to those present on the sixty-second day.

Summary. From a study of the negatives obtained on the sixty-second and two hundredth days after the lipiodol injection it must be accepted that the opaque medium has passed.

1. Outside the generally accepted limits of the S-A space.
2. In some instances escaped from the bony canal itself.

The questions which arise are:

(a) To where has the oil migrated.
(b) By what mechanism and route has it reached its new position.

The answer to these questions demands further experimentation. At this stage it may be conceded that in order to get outside the spinal canal the oil has either passed along inside the nerve roots and sheaths themselves or passed through the dura from the S-A space into the epidural space and passed out alongside the nerves. From the anatomical and pathological standpoint the first appears the more probable. Moreover outside the bony canal itself the oil appears to have arranged itself along definite tracts and the completed picture is reminiscent of a tracing of some of the roots of the sacral plexus, the oil following
closely the early course of the sciatic nerve and its radicles. (I).

It is difficult to conceive in terms of the pathology of absorption or resorption that so much oil could have passed through a membrane of the consistence of the dura mater other than by the route mentioned above. Rupture of the membranes with liberation of the oil into the epidural space, which could have conceivably occurred from the weight of the column of oil when the dog was upright, would surely have effected very profoundly the pressure and quantity of cerebrospinal fluid in the cisterna magna; during this period however, cistern puncture was being performed and normal quantities of fluid were being obtained at normal rate of flow.

On the other hand the extravasations of the oil both in this and other dogs are, in various instances, frequently much larger than any of the nerve roots known to be in this position. This is well illustrated by the oil within the sacrum of the Dog 7. The ventro-dorsal and lateral views (figures 30, 31, 32 and 33) provide several examples of masses of oil which exceed in size the nerve roots occurring in the CAUDA EQUINA of a dog of this size.

The position of the oil and route by which this was assumed are questions which must remain in doubt until the conditions are reproduced and further experiments on photography of the dissected nerve roots are undertaken.

(I). The sciatic nerve in the dog arises from the fifth, sixth and seventh lumbar nerves and the first sacral; Bradley.
Conclusions Dog 7.

(1). Lipiodol outlined the cul-de-sac after 30 minutes suspension. A complete filling was recorded in 60 minutes.

(2). Temporary adhesions of varied size and shape occurred at all stages of the experiment chiefly about the cervico-thoracic junction.

(3). Cell counts of the c.s.f. indicated a mild reaction. After an initial rise to 887 the cell count fell to 426 and then rose to 4,000. This figure is higher than usual. The count had returned to normal limits in October 1928.

(4). The oil underwent subsequent movements. That in the cervical region moved forward to the base of the brain; that in the cul-de-sac escaped beyond the limits of both the cul-de-sac and the bony canal; outside the latter it appeared to follow the track of the ventral branches of some of the spinal nerves.

(5). An enlargement of the intervertebral disc between L.7 and the sacrum was present, and is described later; its effect upon the disposition of the cul-de-sac and its shadow when filled with lipiodol was described and illustrated.
EXPERIMENT 4.

Dog No. 8, Airedale, adult, length 58 cms.

THE CLINICAL EXAMINATION was satisfactory.

THE RADIOLOGICAL EXAMINATION (negatives numbered 106-114 inclusive) of the spine under morphia and chloroform on 1.2.28. revealed a lesion of spondyleitis deformans associated with the articulation between L.3 and L.4 (negatives 106,110,111). The characters of the lesion are described later in Section 6.

Among the dogs used for trial injections of lipiodol before the commencement of this experiment was Dog 5, a labrador, the observations relating to which are given in detail later (Section 6). This animal had a similar lesion in the posterior thoracic region and the X-ray negatives with post-mortem findings supported the view that the lesion had no local effect on the dimensions of the S-A space (see page 11). On these grounds and as there was difficulty in finding experimental subjects with normal spines it was decided to carry out a descent experiment on Dog 8.

THE LIPOIODOL INJECTION, was carried out on 5.2.28 and was recorded on negatives numbered 122-142 (inclusive).

3. Total Time of Suspension.
(in minutes).

50 a.m. 0. I c.c. Lipiodol Descendant (Lafay) containing 0.54 g. of iodine per c.c. was injected by cistern puncture.
(negatives 122,123).

Dog placed upright.
12.40 p.m. 40. Negatives numbered 124, 125, 126, 127 taken with dog on the sloping table. The oil had reached the junction of L.1-L.2 and was disposed elsewhere as follows:
(a) One drop in the Cisterna Magna,
(b) the typical adhesion at level of anterior arch of axis was already present. Throughout the cervical series globules of oil were demonstrable especially within C.5, C.6, and C.7.
(c) In the early thoracic region adhesions were globular but later linear in shape.

12.55. 40. Dog placed upright.

I.55. 100. Negatives numbered 128, 129, 130, 131 taken with dog on sloping table. The oil had now reached the level of the anterior part of L.7. The adhesions were disposed as follows:
(a) Cervical Region. The arrest associated with the axis was present. Many globules within C.5, C.6 and C.7.
(b) Globular-shaped depositions within T.1, T.2, and T.3. Elsewhere in the thoracic part of the canal they were linear shaped.
(c) Lumbar Region. In L.1 one globule; in L.2 one globule. Throughout the posterior two-thirds of L.3 and the greater part of L.4 the oil has congregated as small globules which in places have fused to form are almost uniformly opaque shadow. This has principally occurred in the left half of the spinal canal of this region. Arrest of this type at this level has been noted previously (see Dog 7.) and cannot therefore be regarded as related to the spondylesitis lesion present at this level.

2.15. 100. Dog placed upright.

1.40. 185. Negatives numbered 132, 133, 134, and 135 were taken with the dog on a sloping table. The cul-de-sac was now outlined from the middle of L.7. (see figure 34). At the level of L.5 the shadow contracted suddenly to traverse the sacral canal as a filiform prolongation reaching to the level of the junction between Co.1 and Co.2. An irregularly shaped bulb of oil (A, figure 36) of moderate size was situated at the level of the middle third of the first coccygeal vertebra.
Elsewhere the adhesions were typical, being disposed in the axis, the cervico-thoracic junction and the middle of the lumbar region. None of the globules in the latter had remained stationary during the last 100 minutes.

3.45 p.m.  185.  Dog placed upright.

4.40.  240.  The experiment was concluded at this stage. Negatives of ventro-dorsal and lateral views were obtained (numbered 136 - 142 inclusive).

A modification had occurred in the cul-de-sac (see figure 35) in the form of a new dilatation of the thread of oil as it passed across the sacro-coccygeal junction. (b, figure 35). This oil expansion was a little smaller than the one (a) situated more posteriorly and lay directly in the mid-line.

The lateral photographs showed:

(a) that the "coccygeal dilatations" were dorsal in position in relation to the roof of the canal.

(b) The cul-de-sac was disposed along the inner curve of the bony canal at its lumbo-sacral bend. (compare dogs 6, 7 and 14).

(c) The majority of the arrested oil in the region of L.3 and L.4 was situated dorsal to the cord.

(d) The adhesions about the C-T junction were situated in all planes.

(e) The adhesion seen in the axis was located as in other dogs, dorsal to the cord immediately in front of the anterior border of the vertebral arch.

Summary Dog 8.

(1). A lesion of spondyleitis deformans was present at the level of the L.3-L.4 articulation. This was regarded as being of no significance in relation to the movements of lipiodol in the S-A space.

(2). The cul-de-sac was outlined after a lapse of time of between 100 and 185 minutes from the commencement of the experiment.
(3). The adhesions were characteristic in form and position and those in the lumbar region did not appear to be related to the bony lesion affecting the spine.

(4). The cul-de-sac shadow exemplified the presence of certain dilatations in the course of its terminal filament. These have been noted elsewhere (see Dog 14) and are discussed later.

**Effects of the Injection.**

A. **CLINICALLY** the dog was dull and had little appetite for two days after the injection. Passive movements of the neck and back caused dull pain. These symptoms cleared up quickly. The temperatures (Chart 8) did not rise after the injection. On 14.3.28. a rise to 103°F occurred but fell away rapidly.

B. **CHANGES IN THE C.S.F.** The white cell count of the c.s.f. (see table 9) rose to 955 in the first 26 hours, but dropped to 213 two days later. Fluctuations persisted until a final count of 40 was made on 6.3.28, thirty days after the injection. Thus the oil injection had been followed by a moderate but temporary increase in the cell count.

**Subsequent movements of the Lipiodol.**

Photographs were obtained on 10.3.31, thirtyfour days after the injection (negatives numbered 198, 199, 200, 201, 202, 203, 204, 205, 206).

Three changes had occurred which from observations made in the previous experiments had come to be regarded as characteristic.
### Table 9

**Cell Counts of the c.s.f. of Dog 8. after Lipiodol Injection.**

<table>
<thead>
<tr>
<th>Date</th>
<th>Interval between Punctures (in Days)</th>
<th>Red Cells present per cubic millimetre</th>
<th>Whites introduced % to R.B.C.'s.</th>
<th>Total Whites</th>
<th>Actual Whites</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>6.2.28</td>
<td>26(hours)</td>
<td>400</td>
<td>-</td>
<td>955</td>
<td>955</td>
<td></td>
</tr>
<tr>
<td>7.2.28</td>
<td>2</td>
<td>215</td>
<td>-</td>
<td>213.8</td>
<td>213.8</td>
<td></td>
</tr>
<tr>
<td>9.2.28</td>
<td>2</td>
<td>291</td>
<td>-</td>
<td>33</td>
<td>33</td>
<td></td>
</tr>
<tr>
<td>11.2.28</td>
<td>2</td>
<td>2,715</td>
<td>6.5</td>
<td>55</td>
<td>48.5</td>
<td>Reds counted Thoma Zeiss.</td>
</tr>
<tr>
<td>15.2.28</td>
<td>4</td>
<td>Attempted puncture. Unsuccessful.</td>
<td>-</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>18.2.28</td>
<td>4</td>
<td>346</td>
<td>-</td>
<td>104</td>
<td>104</td>
<td></td>
</tr>
<tr>
<td>22.2.28</td>
<td>4</td>
<td>206</td>
<td>-</td>
<td>512</td>
<td>512</td>
<td></td>
</tr>
<tr>
<td>6.3.28</td>
<td>15</td>
<td>-</td>
<td>-</td>
<td>40</td>
<td>40</td>
<td></td>
</tr>
</tbody>
</table>
(1) Breaking-up of the oil in the cul-de-sac. (See figure 36).

(2) The collection of oil along the ventral aspect of the cord in the region of the cervico-thoracic junction. (See figure 37).

(3) The migration of oil into the cranium around the base of the brain.

(1). Changes in the cul-de-sac. (figure 36).

The oil had moved out of the cul-de-sac and was represented by a ribbon-like collection of oil situated along the mid-ventral line within L.6 and L.7. (figure 36). This was doubtless the result of gravitation since, with the dog in the standing posture, the sacral canal and therefore the cul-de-sac within slopes downwards and forwards. None of the oil had moved outside the boundaries of the S-A space, as happened in other dogs. The dilatations (a and b) which had been noted along the track of the termination of the original cul-de-sac were still present and an additional one (e, figure 36) had appeared a little anterior to the first and situated slightly to the left side just within the sacrum.

(2). Gravitation of Oil towards C-T Junction. (figure 37).

Oil formerly represented by adhesions scattered in the posterior cervical and early thoracic series had gravitated to the floor of the subarachnoid space at its lowest parts as the cord passes around the cervico-thoracic curve. This has been observed in trial experiments where a good deal of oil remained as adhesions at the conclusion of and experiment.

(3). Globules of oil were demonstrable about the level of the sphenoid bone. There was more oil in the cervical region than had been
present at the conclusion of the original experiment apparently indicating the migration of oil from the lower cervical region forward to the cerebral subarachnoid space.

Conclusions Dog 8.

(1). The descent of the oil occupied a longer interval than in previous experiments. From the position and type of the adhesions it appeared that the spinal lesion known to be present had no effect upon the movements of the oil in the S-A space.

(2). Changes in the cell counts of the cerebrospinal fluid indicated an early, moderate and transient reaction.

(3). The systemic reaction was minimal.

(4). After 34 days residence in the tissues the oil was found to have exhibited three typical changes, but shewed no tendency to appear outside the boundaries of the cul-de-sac as in other experiments.
EXPERIMENT 5.

Dog No.5, curly-coated Labrador cross, O, adult, length 65 cms.

This animal was experimented upon early in the series when our methods of injection and suspension were imperfect; accordingly the clinical effects of the injection and of its effects on the c.s.f. cell count have been recorded, but the descent times have been rejected as unreliable. The dog shewed a lesion of spondyleitis deformans involving the articulation between the eleventh and twelfth thoracic vertebrae. It was thus possible to make some observations on the movements of oil along the 3-A space in the neighbourhood of the lesion. A description of the lesion and of the effects on the oil are dealt with later.

An injection of 1.5 cc Lipiodol Descendant containing 0.54 g. of iodine per c.c. was made into the cisterna magna on 15.I.28.

EFFECTS OF THE INJECTION.

A. CLINICALLY no change was noted in the dog. Co-ordination was perfect at all times. The temperatures were recorded on chart, ^12 They are somewhat irregular but only once rise above 103° F. and are hence within normal limits.

B. THE VARIATIONS IN THE CEREBROSPINAL FLUID COUNT have been tabulated in the usual manner. An initial rise to 56 in six hours and to 988 after 24 hours occurred. This reaction is much higher than can be accounted for by cistern puncture alone, and is accordingly attributable to the presence of the oil. The counts were followed until 28.I.28. when the animal was destroyed (morphia and chloroform).

The red blood cell content of the cerebrospinal fluid of Dog 5 was frequently high as difficulty was experienced in avoiding the bony
boundaries of the atlanto-occipital space. After death the occipital region was dissected. The occipital shelf was found to be very well developed and the atlanto-occipital space proportionately small. During puncture the needle therefore often passed near to bone which, as has been shewn in Part One, frequently results in the contamination of a fluid sample with red blood cells.

**Conclusion Doc 5.**

The data relating to the time and mode of descent of the oil within the S-A space have not been included in our observations since the experiment was carried out in order to perfect technique.

The effects of an injection of 1.5 cc of oil were studied. Clinically there were no observable results.

Cell counts of the cerebrospinal fluid indicated a transient reaction.
**TABLE 10.**

DOG 5.

Cell counts of the c.s.f. after Lipiodol injection into the Cisterna Magna.

<table>
<thead>
<tr>
<th>Date</th>
<th>Interval between punctures (in days)</th>
<th>R.B.C.'s.</th>
<th>White Cells introduced &amp; to R.B.C's.</th>
<th>Total Whites</th>
<th>Actual Whites</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>15.1.28</td>
<td>6 hours</td>
<td>11,700</td>
<td>50</td>
<td>106</td>
<td>56</td>
<td>Difficult puncture; fluid obtained only after several attempts.</td>
</tr>
<tr>
<td>16.1.28</td>
<td>1 day</td>
<td>3,600</td>
<td>8</td>
<td>988</td>
<td>980</td>
<td></td>
</tr>
<tr>
<td>17.1.28</td>
<td>1</td>
<td>191</td>
<td>-</td>
<td>260</td>
<td>260</td>
<td></td>
</tr>
<tr>
<td>19,1.28</td>
<td>2</td>
<td>56</td>
<td>-</td>
<td>176</td>
<td>176</td>
<td></td>
</tr>
<tr>
<td>21.1.28</td>
<td>2</td>
<td>15</td>
<td>-</td>
<td>262</td>
<td>262</td>
<td></td>
</tr>
<tr>
<td>25.1.28</td>
<td>4</td>
<td>102</td>
<td>-</td>
<td>44</td>
<td>44</td>
<td></td>
</tr>
<tr>
<td>28.1.28</td>
<td>3</td>
<td>1,250</td>
<td>3</td>
<td>402</td>
<td>399</td>
<td>Needle passed near to bone.</td>
</tr>
</tbody>
</table>
SECTION B. (CONTINUED).

3. GENERAL EXPERIMENTAL CONCLUSIONS.

The conclusions to be drawn from the experiments recorded above will be discussed under the headings of:

(a). Time of descent of the oil and factors influencing this.
(b). The conformation of the arachnoid cul-de-sac.
(c). Adhesions which occurred in the course of a descent.
(d). Movements and changes in the appearance of the oil during residence in the tissues.
(e). The effects of injection on the dog.

A. The time of descent from the Cisterna vacna to the lumbo-sacral cul-de-sac.

The figures for the four experimental dogs 6, 7, 8, and 14 are set out in Table II. For the purposes of comparison a descent has been adjudged complete when the cul-de-sac was filled sufficiently to form a symmetrically cone-like shadow terminating in a fine point within the sacrum.

<table>
<thead>
<tr>
<th>DOG</th>
<th>TIME OF DESCENT</th>
<th>AMOUNT OF LIPIODOL INJECTED</th>
</tr>
</thead>
<tbody>
<tr>
<td>6</td>
<td>90 minutes (negative 64)</td>
<td>2.5 cc.</td>
</tr>
<tr>
<td></td>
<td>Had reached the L-S junction in 30 minutes. Probably filled in 45 minutes. (negative 60).</td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>60 minutes (negative 92).</td>
<td>1.5 cc.</td>
</tr>
<tr>
<td></td>
<td>A considerable degree of filling at 30 mins;</td>
<td></td>
</tr>
</tbody>
</table>
The average time of descent was 108 minutes. Had the photographs been recorded at shorter intervals this figure would probably have been less. The shortest descent-time was 60 minutes and the longest 185 minutes. The latter occurred in Dog 8. This dog, as mentioned, was affected with spondyleitis deformans but it will be shown (see Appendix C) that this condition is unlikely to have exerted any effect on the descent. The period of descent in the dog is much greater than in the man, in whom it is usually estimated at 5-6 minutes (5,13,16,25,40,67,68).

Longer times of descent are mentioned in human work. McKerchie (67) writes, "Lipiodol gravis falls into the cul-de-sac in at most 10 hours", and Ironside (68) remarks that "since the oil travels slowly down the subarachnoid space the best pictures are obtained at least 24 hours after the injection".

The delayed descent in the dog is due, in the writer's opinion to several causes and these have been enumerated below in order of their importance.
A. The comparatively small dimensions of the $\text{SA}$ space.

B. Spinal Curves.

C. Breaking up of the oil by (1) the injection and (2) the movements of the patient.

D. Special shape of the cul-de-sac in some individuals.

A. The $\text{SA}$ space of the dog is actually and relatively smaller than that of man. Experiments have been mentioned (see Part Two, Section A) which show that the dimensions of a space influence the rate of movement of the oil, and can if small enough bring about an arrest (7). Reference to any of the series of photographs will confirm that arrest of a temporary nature occurred most frequently about the level of C.6-T.3 and L.2-L.4. This roughly corresponds to the positions of the intumescentia cervicalis and intumescentia lumbalis respectively and where the dimensions of an already small $\text{SA}$ space are still further reduced. It is not, therefore, unreasonable to assume that the delayed descent in the dog is, in part at least, purely a question of the physics of the passage of a viscous fluid along a space of small dimensions.

B. A change in the direction of the spinal canal such as occurs at the C-T junction is associated with arrest. Adhesions about this site were very frequent though it is admittedly difficult to assess the extent to which the intumescentia cervicalis or the spinal curve were responsible for the stoppage.

C. Breaking-up of the oil. It was observed both in preliminary trial injections and in the experiments that have been described (e.g. Dog 8) that if the oil split up into a great number of globules
either in the cisterna magna or along the course of the S-A space that a delayed descent resulted. The causes of this dividing up of the oil appeared to be twofold.

(I) the technique of injection (2) the struggling of the patient during suspension.

(I) When oil is forced from a lipiodol syringe through a needle of the size described it issues in the form of separate droplets.

It is desirable that the oil globules, after injection into the cisterna magna, should be allowed to fuse again into a single mass to get, so to speak, a "start" in its descent. It has been shown previously that the oil globules injected into a test tube of warm c.s.f. will fuse on coming into close contact at the foot of the tube. Accordingly, after an injection, a dog should be rolled gently onto its back and maintained there for some minutes. Under these conditions the globules of oil lying on the roof of the cistern will coalesce (see figure 15).

(2) We have noticed, in the animals that were restless when held upright, that the oil broke up into many small globules, particularly in the mobile parts of the spine (cervical and lumbar areas). This was regarded as a direct result of spinal movements. It should be prevented by making the patient as comfortable as possible on the woollen pads, and passing a catheter in the male if the time of suspension is prolonged.

P. The S-A cul-de-sac in certain individuals does not taper uniformly; about the level of the L-S junction it may contract suddenly to
continue as a thread-like prolongation (see Dog 6, and 8). In both instances the oil was arrested for some time at this level before entering the fine termination.

In conclusion it may be remarked that the amount of oil injected exerts no effect on the rapidity of descent. An injection of 0.5 c.c. in Dog 14 descended almost as rapidly as 2.5 c.c. in Dog 6 and more rapidly than 1.5 c.c. in Dog 8.

B. The Conformation of the Subarachnoid Cul-de-Sac.

The shape and relations of the so-called lumbo-sacral cul-de-sac or the blind posterior termination of the subarachnoid space have been demonstrated in four adult dogs (numbers 6, 7, 8 and 14). The radiological picture cast by this structure when filled with lipiodol is depicted in the following figures.

Dog 6. V-D view. Figure 16 (negative number 73). Lateral view. Figure 17 (negative number 74).

Dog 7. V-D view. Figures 26 and 28 (negative number 92) Lateral view. Figure 29 (negative number 103).

Dog 8. V-D view. Figures 34 and 35. (negative number 136) A lateral view is recorded on negative number 138 but owing to movement was not suitable for reproduction.

Dog 14. V-D view. Figure 22 (negative number 194). Lateral view. Figure 23 (negative number 195).

The characters of the shadow of the completely outlined cul-de-sac of each dog dealt with will now be described followed by observations on the evolution of this shadow at different stages of each experiment. A general comparison will then be drawn up.

The cul-de-sac shadow resembled an inverted cone reaching as far as the level of the second segment of the sacrum. The reduction in calibre commenced at the level of a transverse plane passing through L.5. and was not uniform. At the level of the anterior border of the sacral arch there was a sudden reduction in size of the shadow; behind this it persisted as no more than a filiform prolongation within the sacrum. A lateral view of the shadow (figure 17) shows a similar reduction in calibre at the same transverse level; whilst the upper border of the shadow remains closely related to the roof of the spinal canal throughout its length the lower border recedes progressively further away from the floor.

The borders of the shadow viewed laterally are smooth and continuous, but in the ventro-dorsal photographs they exhibit irregularities. In negative 69 and less so in negative 73 small filiform or tubercle shaped projections occur on the right side throughout L.5, L.6, and L.7. In L.5 there are two, with suggestions of similar projections on the left-side at the same level. Posteriorly they are more numerous. The projections of a given side are thrown into relief in an oblique photograph such as that in negatives 69 and 73. The assumption that these projections represent insinuation of the oil along the nerve roots is supported by the following facts:

(a). The nerve roots in this position leave the cul-de-sac along its ventro-lateral aspect so that an oblique view would throw into relief their points of emergence.

(b). The projections suggest a paired arrangement, pass out obliquely...
in a posterior direction; they are disposed regularly and increase in number posteriorly.

On the other hand none has been observed behind the level of the L-3 space where nerves are still issuing from the cul-de-sac; neither has the picture of twin projections on the same side at about the same level been seen as might be expected of spinal nerves which have two roots emerging at the same transversus level.


After thirty minutes suspension (negative 60) a uniform filling had occurred as far back as the level, where, as shown by later photographs, the rapid diminution of calibre was to take place. The incomplete sac terminated as two points. The laterally-placed projections thought to be associated with nerve roots were faintly suggested. After 90 minutes (negative 64) the outline of what proved to be the complete cul-de-sac had been obtained. Since the oil in this case exhibited a reluctance to enter the finer termination of the cul-de-sac and remained for a time arrested at the level of the L-3 space it may be remarked that should stoppage at this level be encountered in a clinical case a further suspension should be tried.

THE CUL-DE-SAC OF DOG 8.

The completed shadow of the sac (figure 35, negatives 132, 136 and 142) is reminiscent of that met with in Dog 6. It is roughly the shape of a radish, being of bulky globoid form within the posterior half of L-7, and then suddenly diminishing to a fine thread-like structure reaching through the sacrum to the S-0c junction. Here a bead-like
expansion of the oil has taken place to be followed in turn by a
further thread terminating within the body of Co. I as an irregularly
shaped dilatation. The lateral view on negative 142 shows the
usual characters of the sac, as described in the case of Dog 6; throughout
the whole of its length the upper border of the sac follows the
contour of the roof of the canal. The diminution in calibre chiefly
depends on the lower border progressively retreating from the floor.


The cul-de-sac shadow at an intermediate stage of development appeared
on only one photograph. When negative 128, after 100 minutes sus-
pension, was taken no filling had occurred. Eighty-five minutes later
the oil had outlined the sac in the form shown in figure 34. At this
stage it chiefly differs from the complete shadow described above in
that only the more posterior of the two terminal dilatations is present.
This is a little smaller than at a later stage and appears to be more
to the side of the centrally situated thread of oil. The two structures
are visibly connected and the thread is easily seen to project a little
beyond the dilatation almost into Co. 2.


The characters of the completely filled cul-de-sac are shown in
figures 22 and 23, and negatives 194 and 195. It is in the form of a
finely tapering inverted cone. The shadow commences half-way back along
L. 6 and hence the diminution in size which, as in Dog 6, starts within L. 5
has already commenced. From that level backwards there is a uniform
tapering to the middle of the sacrum, when through the course of S.3 a slight thickening occurs which is continued as an exceedingly fine thread to the middle third of Cc.2. Here the oil expands into an elongated irregular blob lying a little to the right of the mid-line and reaching backwards to finish as a tapering point within Cc.3. In the lateral view the shadow is that of a finely pencilled cone following the roof of the bony canal until reaching the curve at the sacro-coccygeal junction where it lies along the floor. In other words it "cuts" the curves of the bony canal. The bulbous expansion is seen to lie along the roof of the coccygeal canal commencing under the arch of Cc.2 and extending subjacent to the interarcual ligament to the level of Cc.1.

The filiform connection between the cul-de-sac proper and this coccygeal dilatation cannot be traced on the lateral photograph. (negative 195). The border of the shadow in the ventro-dorsal view shows localised irregularities. A convexity occurs on the left side at the level of the junction of the middle and posterior thirds of L.6. Within the anterior third of L.7 a similar condition is present on either side of the sac. An unpaired projection directed outwards and backwards occurs at the level of the posterior border of the 7th lumbar arch, and again as the shadow crosses the L-S space.

This photograph is slightly oblique and confirms in several particulars the observations made in the case of Dog 6, viz:—

(a). Occasional paired arrangement; (b). Projections thrown into relief chiefly on one side of an oblique photograph; (c). Projections are closer together and are more numerous posteriorally. None of these
projections was apparent behind the level of the first sacral bony segment.


Ninety-five minutes after the injection the oil had outlined the cul-de-sac in the form of a tapering column reaching to the middle of the sacrum. The tip however, appeared blunt and incomplete. After 180 minutes all the characters of the completed cul-de-sac were reproduced. The oil expansion had appeared within Co.2 and was extending to Co.3.

THE CUL-DE-SAC OF DOG 7.

The characters of the cul-de-sac of Dog 7 are discussed later in Section C. The shadows suggested, and the post-mortem confirmed, an abnormality that directly affected the shape of the sac and the shadow cast by it when filled with lipiodol.

From a consideration of the above data it is possible to lay down rulings as to the characters of the normal cul-de-sac of the dog for the guidance of the clinician.

The shape varies considerably but is roughly that of a cone with the tip directed posteriorally. The reduction in calibre which commences about the level of the fifth lumbar vertebrae is sometimes uniform and gradual. In other instances a sudden reduction is noted, the sac then continuing as a thread-like prolongation. This change in diameter usually occurs about the level of the L-S space. In certain instances a terminal thread exhibits isolated dilatations in the form of small variously shaped globules of oil. These are usually situated within
the coccygeal region and related to the roof of the canal. These expansions of oil may lie to either side of, or more rarely in, the midline. A connection with the cul-de-sac proper is usually demonstrable especially in ventro-dorsal photographs, since the whole structure is then more nearly in contact with the cassette and less subject to movement. (Page 109 A to be appended here).

The cul-de-sac shadow is symmetrically disposed in the mid-line in the ventro-dorsal photograph.

In a lateral view the sac is seen to follow the inner border of any curve in the spinal canal. Thus it is related to the roof of the bony canal throughout the lumbo-sacral bend and to the floor in the sacro-coccygeal deflection of the canal.

It is to be observed that if an oblique V-D photograph be taken the line of direction of the sacral part of the spinal canal, and hence the shadow of the terminal part of the cul-de-sac appears to be considerably deflected to one side of the mid-line. This is directly due to the angle at which the sacrum is set in relation to the lumbar vertebrae. The appearance is exemplified in figures 34 and 35 and negatives 104 and 132.

(0) Adhesions in the course of a descent.

Adhesions of the oil were met with in every case. The tendency of the oil to adhere to the meninges in animals has been observed in the cat by Mixter (37). In our experiments the majority occurred at:

(I) the C-T junction (i.e. at the level of C.6, C.7, T.1, T.2) in the form of globules situated at any point around the cord.
The dilatations noted in our experiments only occurred in cases where the animal was kept upright for at least 180 minutes. Dilatations were seen in the following dogs and the times of suspension are appended:

<table>
<thead>
<tr>
<th>Dog</th>
<th>Time at which Dilatations were noticed</th>
<th>Amount of Oil injected</th>
</tr>
</thead>
<tbody>
<tr>
<td>11.</td>
<td>180 mins.</td>
<td>0.5 c.c.</td>
</tr>
<tr>
<td>8.</td>
<td>(A) 185 mins. (B) Second Dilatation after 240 minutes,</td>
<td>1.0 c.c.</td>
</tr>
</tbody>
</table>

The amount of oil resting in the cul-de-sac does not appear to bear any relation to the development of these oil expansions. The author is unaware of any observations recording the appearance of these terminal dilatations in the shadow of the cul-de-sac of man or other animals. Their nature cannot be defined without further experiment.
(2) the mid-lumbar region L.2, L.3, L.4. Here adhesions were both globular and linear; (figure 34) occasionally sufficient oil fused to form a plaque or plate-like shadow.

An adhesion of rather regular occurrence and one which often remained in position from the early stages of an experiment, was located above the cord at the level of the anterior border of the axis. It occurred in Dogs 6, 7, 8 and 14 and is illustrated in figures 49 and 50.

Adhesions occurred of course elsewhere in the spine. If, in the cervical series, they were usually globular in form and of varying size. In the thoracic region they were linear and tended to arrange themselves on either side the cord, thus outlining it. Quite small globules, the size of a pin-head were sometimes seen.

It is apparent that adhesions principally occurred in tracts where there was greatest movement of the spine (i.e. cervical and lumbar regions). Spinal movements tended to break up the oil into globules and these having a greater total surface area than the original appear to have a greater tendency to adhere.

Intumescentia cervicalis and lumbalis cause an increase in the size of the cord and a corresponding decrease in the dimensions of the subarachnoid space at these levels and so far as the C-T junction is concerned, there is a change of direction of the bony canal. Accordingly it has been concluded that spinal movements, combined with a reduction in the dimensions of the subarachnoid space and/or change of direction in the canal are the main factors in inducing adhesions.
Movements of the oil after primary descent.

Movements of oil in the subarachnoid space of the cat has been observed by Ayer and Mixter (22). In their experiments iodipin and other iodized oils were injected into the cisterna magna of the cat in quantities of 1.0 cc and 1.5 cc. The authors in their conclusions state, "the oil is disseminated slowly throughout the spinal and cerebral subarachnoid spaces of the cat under normal conditions".

In our own experiments the following observations were made.

Three main changes took place in the position of the oil; the first, the appearance of globules around the base of the brain; the second, the collection of oil along the floor of the canal especially at its lowest point and the third, modifications of the oil originally filling the cul-de-sac.

(I) Migration of oil around the base of the brain.

The appearance presented is depicted in figures 20 and 21, (negatives 227 and 228). It was noted in dog 5 fifteen days after injection (negative 119), in dog 8 thirty-four days after injection (negatives 201 and 206), in dog 6 fifty-six days after injection (negatives 210, 211, 215); and dog 7, sixty-two days after injection (negatives 223, 224, 227, 228). The globules in all cases lie in an area overlying the body of the occipital bone, the body and wings of the sphenoid and sometimes, as in negative 210, as far forward as the ethmoid region.
If globules had moved backwards they would have added to the collection which gravitated to the cervico-thoracic junction, but in these instances the total amount of oil resting there appeared insufficient to account for this. The simultaneous appearance of oil in the subarachnoid spaces at the base of the brain is therefore attributed to a migration from the cervical region. The head of a dog when eating, drinking or sleeping is usually carried downwards and forwards so that arrival of the oil in the cranial cavity is not unexpected, and is probably the result of gravitational gravitation. The effect of the circulation of the c.s.f. may play a part in the movement of the oil, but further experiments would be necessary before an opinion could be formed.

(2). The collection of oil along the floor of the canal. This is the direct result of gravitation (figure 36, negative 200). It occurred typically in the region of the cervico-thoracic junction, where it was aided by the disposition of the bony canal, and in the anterior part of the lumbar region.

(3). Modification in the position and appearance of oil in the subarachnoid cul-de-sac.

The changes have been studied in several dogs at different intervals of time after injection, as follows:-

Dog 6. (a). After 8 weeks and (b) after 28 weeks.
" 7. (a). After 9 weeks and (b) after 30 weeks.
" 8. (a). After 6 weeks
The changes in each individual case have been analysed elsewhere. The migration of the oil to limits apparently outside those of the subarachnoid space proper has been noted in all the above dogs. The process was observed within five weeks of the injection (Dog 8) and at eight or nine weeks had advanced nearly as much as at twenty-eight and thirty-one weeks (dog 6, dog 7). It is not dependent for its appearance on the amount of oil injected since this varied; nor upon the presence of the terminal dilatations of the cul-de-sac which were noted during the experiments since these only occurred in Dog 3 and 14. The nature of this migration and its mechanism has been discussed, but whilst the weight of evidence points to its representing an insinuation of oil directly along the nerve sheaths, other reasons can be advanced against this.

Changes also took place in the shadow of the cul-de-sac proper. The uniformly dense unbroken shadow remaining at the conclusion of a descent experiment broke up into streaks, lines and globules. In most cases the original outline of the cul-de-sac was preserved, but in Dog 8 the oil gravitated out of the sacrum and rested, ribbon-like, along the floor of the lumbar region. The oil in the other cases however, tended to move forward, and the writer regards gravitational movements of this nature to be chiefly responsible for the breaking-up of the oil as opposed to any bio-chemical resorption.

E. The effect of the subarachnoid injection of Lipiodol was studied in the five dogs numbered 5, 6, 7, 8 and 14.

The general effect on the dog as estimated from its appearance, de-
meal, appetite and temperature appeared to be minimal. In some instances a slight elevation of temperature occurred immediately after the injection but quickly subsided. Sensitiveness to passive movement of the neck or back was observed in only one case, and then was temporary. The cell counts of the c.s.f., revealed a rapid increase with a slow but definite decrease. Apart from the initial rise which was due to the oil, the significance of subsequent counts is complicated by the fact that serial cistern puncture is known to induce fluctuations in the cerebrospinal fluid cell content (see Part one). On the average however the figures were too high to be imputed altogether to the puncture so that it may be said that the irritative effect of the oil, signalled by an initial increase in white cells in the cerebrospinal fluid, persisted for some weeks becoming gradually less. Occasionally a sudden increase in the count occurred. (see Table 8, Dog 7). On 8.2.28 ten days after the lipiodol injection the white cell count was 426. One week later it had risen to 4,000. Other instances of fluctuations can be found in the tabulated records.

These variations may depend upon the arrival of a globule of oil in a new situation as for instance during the migration towards the base of the brain. It can be postulated that the increase in cell count of cerebrospinal fluid following an injection of lipiodol is the result of meningeal irritation, and that when a meningeal surface is first brought into contact with lipiodol a primary and maximum reaction ensues. The membrane then appears to tolerate the oil better for the cell count falls rapidly both in man and dogs (v-s). In other words the meningeal areas in contact with lipiodol appear to develop a
local tolerance. Thus the movement of oil to a new situation and to a new part of the meningeal wall of the subarachnoid space should be followed by a local and comparatively marked reaction expressed as a cell increase in the cerebrospinal fluid. The variation in cell count in the days and weeks succeeding injection may depend upon certain migrations of oil which are known to have occurred at this time.

Lipiodol after long residence in the subarachnoid space would appear to have no effect so far as our investigations proceeded. The dogs which were kept for many weeks, remained outwardly normal and preserved perfect co-ordination. They walked or moved at fast paces without disability. The cell count of the c.s.f. returned to normal.

Unfortunately no post-mortem microscopical work was conducted on the spine, the cord or its membranes, as the experiments had to be brought to a conclusion.

A general post-mortem was conducted on each dog at the times indicated in Table 5. No abnormalities were found and the macroscopic appearance of the cord and membrane was normal.
SECTION C.

Lipiodol descendant and the demonstration of certain spinal affections of the Dog.

Amongst the dogs used in the above experiments, dog number 7 proved to have an enlargement of the intervertebral disc at the lumbo-sacral junction; numbers 5 and 6 were affected with spondyleitis deformans; an injection was also carried out in a clinical case of suspected spinal compression. It is thus possible to discuss the behaviour of lipiodol during descent in these cases.

Enlargement of the Intervertebral Disc at the L-3 Junction.

The experiment carried out on Dog 7 with observations on the subsequent movements of the lipiodol have been described previously. It will be recalled that preliminary photographs of the spine shewed no abnormality. 1.5 c.c. Lipiodol descendant were injected by cistern puncture and the descent carried out in the usual manner. Forty-five minutes after the commencement of the experiment the oil had commenced to outline the cul-de-sac and a defect in its outline was to be seen in the form of an indentation in the right border and a transverse zone of reduced opacity at the level of the L-3 junction. (Negative 89 R). After 95 minutes these characters were still demonstrable. (figure 26, negative 92). Later in the experiment the animal was placed in the horizontal position for twenty minutes. It had been shown that if this were practised the oil commenced to flow forward out of the cul-de-sac (see Experiment No.1, Dog 6). Photographs then shewed that the column of oil had broken into two parts (figure 27, negative 97). The oil in
front of the L-S junction, that is, in front of the defect in the original shadow, moved forward freely. The oil behind this level remained, so to speak, imprisoned in the cul-de-sac. This suggested the presence of an obstruction at this level. Photographs were taken after a further suspension of thirty minutes. The V-D views shewed the two parts of the oil to have become again continuous (figure 28, negative 102), but the characteristic defect still persisted. Viewed laterally the cul-de-sac "pencil" was seen to be subjected to a sudden bend (convexity dorsal) when crossing over the L-S articulation as if negotiating an eminence on the floor of the canal. (figure 29, negative 103).

In October 1928, two hundred days after the initial experiment, the dog was destroyed (morphia and chloroform). The roof of the lumbo-sacral portion of the spinal canal was removed with bone forceps, and the related spinal cord and its membranes reflected forwards. A projection in the form of a transverse eminence lay on the floor of the canal at the level of the articulation between the bodies of the seventh lumbar vertebra and the sacrum. The lesion was sectioned sagitally when it was seen to be essentially a hypertrophy of the intervertebral disc which had thrust itself up into the canal reducing its diameter at this level. No microscopical examination was made.

A similar condition has been recognized on the continent (Diagnose u Therapie der inneren Krankheiten des Fundes. Jacob, Stuttgart 1913). It is chiefly met with in dachshunds and then usually about the level of the T-L junction and leads to paralysis of hind limbs with incontinence of urine and faeces. In the instance described in Log 7, the lesion was located in a position well calculated to avoid immediate pressure
on the cord with attendant paralysis. The smaller dimensions of the spinal cord and its membranes and the comparative capaciousness of the epidural space at this level delayed or even obviated the development of complete spinal block. Thus a clinical and general X-ray examination failed to detect the lesion.

Conclusions.

(1). In one instance, displacement of the lumbo-sacral cul-de-sac by an elevation on the floor of the canal was demonstrated by filling the cul-de-sac with lipiodol.

(2). The lipiodol which flowed past the obstruction failed to flow back when the patient was placed in a horizontal position. This was an example of true arrest.

Lipiodol injection in a suspected case of spinal compression.

Subject. Terrier cross, J, aged 10 months.

History. 2.4.29. The animal had been picked up in the street seven days previously after having been apparently run over, since when it had been unable to use its hind limbs.

Examination. Total paralysis of hind quarters was present with incontinence of urine and feces; the spinous process of T.10 was displaced backwards and downwards. When tested by the pin-prick method an anaesthesia of the skin could be demonstrated as far forward as a transverse plane passing approximately through T.11. (Fig. 2.)

X-ray photographs showed a compression fracture of the body and arch of T.10, and a fracture of the third last rib on the left side. Lipiodol descendant 0.5 cc. was injected into cisterna magna and the
patient held upright in wool pads. Photographs were obtained after thirty minutes and fifty minutes respectively. After thirty minutes suspension (negative 237), the oil was arrested at the level of T.10, in a vertical column of uniform density. The posterior limit of the column was situated within T.10, and had an irregular outline. The anterior limit reached to the level of T.8 - T.7 junction. Globoid adhesions of oil rested within the T.1.

After fifty minutes (figure 48, negative 238), a little more oil had descended so that the cylinder reached forward within T.7. No change had occurred at the actual point of arrest. Characteristic adhesions were noticeable about the cervico-thoracic junction.

No lipiodol passed the obstruction or escaped into the thoracic cavity.

Conclusions.

(1). In one instance of spinal block, lipiodol confirmed the condition and demonstrated the site of the pressure.

(2). The time of descent to the mid-thoracic region where the block was located was less than 30 minutes. This compares favourably with the data recorded in the case of experimental dogs previously referred to.

Spondyleitis deformans and its relation to the movements of Lipiodol in the S-A space.

Case I. Dog No. 5. Curly-coated, Labrador crossbred; spinal measurements 65 cm. The spine was relatively long for a dog of its size.

Preliminary X-ray photographs were taken on 10.I.28 and shewed a
lesion at the junction of T.12 - T.13 (see figure 36 and negatives numbered 10 0,17,22,27).

It is proposed to describe (a) the radiographic appearances of the lesion (b) the behaviour of the lipiodol, (c) the post-mortem findings. (a). The Radiographic appearance of the lesion. Ventro-dorsal photographs (see negatives 17,27) showed a dense shadow projecting outwards and backwards from each posterior corner of the shadow of the body of T.12. On the one side the projection ran to the region of the anterior corner of the shadow of T.13, and the associated rib articulation, but did not fuse with those. On the other side the projection was more prominent and appeared to surround and involve the rib articulation and to fuse with the shadow of the vertebral body (negative 10 C, 27). The outlines of the intervertebral articulation were irregular and poorly defined. In the V-D photograph of the dissected lesion (figure 40, negative 121) these characters were confirmed; it was seen however that the costal articulation on the right side was only abutted upon, but not obliterated by the new tissue.

Lateral photographs depicted a thin spur of dense tissue springing from the ventral aspect and postero-ventral angle of the body shadow of T.12. It passed backwards in the form of a curved bridge, with convexity ventral, to the region of the antero-ventral angle of the body of T.13 with which however it did not fuse completely (negatives 10 C, and 22; also post-mortem negatives 117,121; figures 40 and 41). Figure 41 is a lateral photograph of the lesion with the muscles dissected away after death; the characters are depicted quite clearly.

The density of the new tissue is about that of the bone of a vertebral body.
(b). The behaviour of Lipiodol. The observations on the descent of lipiodol in this dog have not been annotated since they were not included among the experimental records cited above. They were rejected because our technique of injecting the oil had not been perfected. It was nevertheless possible to utilise the photographs obtained in order to estimate the effect, if any, that the lesion exerted upon the patency of the S-A space. The experimental details were as follows:

<table>
<thead>
<tr>
<th>Time</th>
<th>Total Time of Suspension</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.40</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>I.5 c.c of Lipiodol Lafay containing 0.54 g of iodine per c.c. were injected into the cisterna magna. Dog placed upright.</td>
</tr>
<tr>
<td>2.30</td>
<td>50 mins.</td>
</tr>
<tr>
<td></td>
<td>Negatives (V-D) numbers II,12,13,14.</td>
</tr>
<tr>
<td>2.40</td>
<td>50 &quot;</td>
</tr>
<tr>
<td></td>
<td>Dog placed upright.</td>
</tr>
<tr>
<td>3.15</td>
<td>85 &quot;</td>
</tr>
<tr>
<td></td>
<td>Negatives (V-D) numbers 15,16,17,18.</td>
</tr>
<tr>
<td>3.25</td>
<td>85 &quot;</td>
</tr>
<tr>
<td></td>
<td>Dog placed upright.</td>
</tr>
<tr>
<td>4.35</td>
<td>175 &quot;</td>
</tr>
<tr>
<td></td>
<td>Negatives (V-D) 19-25</td>
</tr>
<tr>
<td></td>
<td>Lateral 25A - 29.</td>
</tr>
</tbody>
</table>

A study of these negatives suggests no obstruction to the passage of oil at the level of the lesion; neither are adhesions noticeable in that position.

Seventeen days later the dog was destroyed and immediately afterwards a series of photographs were obtained (negatives II5,II6,II7,II8). Negative II7 and figure 38 illustrate the conditions at the level of the last few thoracic vertebrae. The lipiodol has gravitated to the floor of the S-A space and lies as a thin line silhouetting what is virtually the lower contour of the spinal cord and its membranes. As the radiopaque line passes over the articulation between T.I2 - T.13, there is no
suggestion of deflection or of encroachment upon the S-A space. This is well defined in the photographs of the dissected specimen (negative 121 figure 39). The muscles were dissected away and the articulated vertebrae in the neighbourhood of the lesion isolated by cutting across at the level of T.11 - I2 and I.2 - 3. The spinal cord and membranes were left in position in the isolated portion. The lipiodol still undisturbed chances to overlay the articulation involved and can be seen in the figure, but indicates no interruption in the continuity of the S-A space.

(c) Post-mortem findings. On dissection the affected joint was found to be immobile. The intervertebral joints before and behind were normally moveable. On sagittal section it was seen that there was a considerable laying down of dense fibrous (bony?) tissue around the articulations between the bodies and transverse processes of T.12 - T.13 and the associated costal joints. The contour of the bony canal at this point appeared unaltered.

CASE 2: Dog No:8. As the experimental evidence and post-mortem findings in Dog 5 suggested that spondyleitis deformans did not bring about a modification of the S-A space, Dog No:8 which exhibited a lesion between L.3 - L.4, was utilised to study lipiodol descent. The characters of the descent have been discussed so that it remains to describe the lesion as it appeared radiologically and on post-mortem.

Radiographic appearance. The lateral view (negative 106) showed a shadow arising from the posterior-ventral angle of the shadow of the body of L.3 and passing to the related angle, as a curved spur, convexity ventral
The new tissue and the body of L.3 at their line of junction were of equal radiopacity, but the shadow cast by the new tissue became gradually less dense as it passed backwards.

The ventro-dorsal views (negatives II0, III, I32) showed that a backwardly projecting spur of new tissue was springing from each lateral angle of the body shadow of L.3. The shadow of the new tissue was less dense on the left than on the right side where it also projected a little further back, though in neither case had a complete bridge been formed on to L.4. The density of the tissue was less than that of a vertebral body but greater than that of a transverse process.

Negative I32 was taken obliquely, so that the right wall of the spinal column was more exposed to the tube. The bridge of new tissue on this side was silhouetted therefore at a more ventral plane than in the direct ventro-dorsal negatives II0 and III. The shadow compared with that seen in a direct V-D was a little denser, and the attempt at bridging over more complete. The anterior part of the shadow was again the darker. It was concluded from these observations that a bridge of tissue of similar radiopacity to the vertebral body connected the posterior end of L.3, to the anterior end of L.4 in the mid-ventral line. The process had extended up each side of the body of L.3, the bridge dorsally, becoming progressively less complete and its tissue less dense.

In all the photographs the shadow of greatest density was thrown by tissue anchored to L.3; when the bridge was incomplete the new tissue was associated with L.3. Hence it may be assumed that the laying down of the new tissue had taken place from before backwards.

The X-ray appearance of the articular processes of L3 did and L.4 did not appear to be modified, neither did the outlines of the
spinal canal appear altered. X-ray photographs were taken in connection with the lipiodol descent 4 days and 38 days later. The lesion shewed no change (negatives numbered I22-I42 and I98-206).

Post-mortem findings. A post-mortem examination was made two days after the last photographs were obtained. The affected joint shewed reduced mobility. The new tissue formed a convexity over and around the ventral part of the articulation of the vertebral articulation and appeared in section to consist of dense fibrous tissue. The maximum thickening had occurred in the mid-ventral line. Neither the articular processes nor the intervertebral foramina were involved.

Conclusions.

(1). Lesions of spondyleitis deformans found in two dogs at the level of T.12 - T.13 and L.3 - L.4 respectively gave rise to no clinical symptoms.

(2). Experimental and other findings suggest that in neither instance was there pressure on the S-A space, or reduction in its size. The varying degree of co-ordination sometimes associated clinically with this condition probably depends upon some factor other than pressure on the cord proper.

(3). From observations on its X-ray density the development of the lesion appears to take place from before backwards and from below upwards; the process therefore should commence at a point in the mid-line towards the posterior part of a given vertebral body and would be best detected in its earliest stages by a lateral photograph.
(4). The immediate mechanical affects are reduction in, or abolition of, the mobility of the articulation involved; in advanced cases the joints between related articular processes and ribs are invested, and their mobility affected. Local pressure on structures passing through the associated intervertebral foramina appears to be a possibility, but was not found on post-mortem of the two cases dealt with.

(5). Both the affected animals were of poor conformation having relatively long spines.
SECTION D.

CLINICAL APPLICATIONS OF THE SUBARACHNOID INJECTION OF LIPIODOL.

It has been shown that lipiodol can be used as a radiopaque medium to investigate the patency of the S-A space in the dog and, as shown in one case, its injection therefore constitutes a method to detect spinal block. This is not easy of demonstration in the dog by the other methods in common use in man. The injection of a needle into the S-A space in the lumbar region is very difficult or impossible by simple puncture.

Such valuable tests as the Queckenstedt or the biological examination of the CSF in front of and behind a suspected block cannot therefore be utilized; again the definition of anaesthetic areas by pin-pricks is unsatisfactory except in extreme and otherwise obvious cases. A safe method of investigation of the S-A space in dogs can be therefore of some clinical importance. Whilst Lipiodol Descendant (Lafey) satisfies certain requirements in this direction, it has the following limitations and disadvantages.

(I). The rate of descent is slow. A patient must be maintained upright for considerable periods, and although carried out with care, this must tend to aggravate most spinal conditions, especially injuries. In the male, the bladder must be emptied by catheterization if the descent time be prolonged.

(I). The only interarcual space in the lumbar region of the dog which is sufficiently large to permit the passage of a needle, is the lumbo-sacral space. Reference to the dimensions and disposition of the S-A space at this level as shown by outlining it with lipiodol, (see figures 15, 16, 17, 22, 23, 28, 29, 34, 35) confirms the well recognized clinical fact that subarachnoid puncture at this level by the simple passage of a small needle is very difficult and at the best a matter of chance.
(2). The oil adheres readily at all positions along the S-A space and the diagnosis of partial or incomplete block particularly at certain points would be a matter of some difficulty. One criterion is that if the position of a patient be reversed the oil in the case of true arrest commences to flow back almost immediately, whilst the temporary adhesion remains unaffected for some time.

(3). The oil is not perfectly tolerated and in all our experimental animals it gave rise to a marked cell increase in the cerebrospinal fluid. This seems to be of little significance in the normal dog as it gives rise to no clinical symptoms and is of limited duration.

In man, Lipiodol has been observed to exert similar effects when injected into what proved to be a normal spine, but in cases where the oil came into contact with pathological tissues it has been observed to be definitely irritant. It is possible therefore that in the dog it may be shown to possess irritant properties when brought into contact with pathological tissues. Its use should be accordingly limited to such cases in which simple block from tumour growth (preferably extradural), fracture, compression, or haemorrhage is suggested after a preliminary clinical examination.

All preliminary X-ray photographs of a spine should of course be completed before the injection of the oil.

A second point of some clinical importance is that of the position of the cord and its membranes within the bony canal. Figure 36 obtained from Dog 8 illustrates the manner in which the cord inclines away from the floor of the canal when negotiating an upward bend of the bony spine. The same disposition has been mentioned as occurring
at the level of the lumbo-sacral curve and in a reverse manner around the sacro-occygeal curve. If in a naturally occurring bend of the bony spine the cord and its membranes are carried towards the inner face of the curve it is probable that this occurs, if the spinal canal be bent passively during an operation, as, for instance in placing an animal in position for cistern puncture or for epidural puncture through the lumbo-sacral space. Thus in the case of either of these procedures the bending of the spine with the object of opening up the atlanto-occipital or lumbo-sacral spaces respectively, has the added advantage of carrying the spinal cord in the occipital region, and the spinal cord and membrane in the lumbar region towards the floor of the canal. This increases the margin of safety of the operation.

APPENDIX I.

A CONSIDERATION OF CERTAIN FEATURES IN X-RAY PHOTOGRAPHS OF THE SPINE OF THE DOG.

In reading negatives of the spine of the dog it is helpful to bear in mind the characters of the shadows cast by certain associated or superimposed structures.

In the ventro-dorsal view the recognition of the shadows of the following structures is important.

(1). The os penis in the male; (2). the prepuce in the male; (3). the scrotum in the male; (4). faecal material in the colon and rectum;
(5). Gas in the intestinal tract; (6). the nipples in the female;
(7). the liver; (8). the heart; (9) the sternebrae; (10) spinous processes; (11) the tracheal tube.
In the lateral view the following occur:-

(I) The transverse processes of the lumbar vertebra; (2) rib articulations; (3) the scapular spines; (4) the external ear; (5) gas in the oesophagus.

(I). The os penis. The characters of the shadow of the os penis are seen in the following figures.

Figure 24, (O.P.0'P'); figure 26, (O.P.0'P'); figure 34 (O.P').

The appearance when directly super-imposed upon the vertebral shadows is depicted in figure 31 and figure 22, (O.P.,0! P!). A lateral view is exemplified in figure 30 (O.P.). Other examples are to be found in figures 18,27,32,33,34.

In V-D photographs the position of the shadow in relation to the spine naturally varies. It usually overlies the first four coccygeal vertebrae, the sacrum and last lumbar, but frequently lies to one side or other of the spinal column. It is generally seen as an elongated shadow with a light centre and two dense borders. A rounded swelling occurs about the junction of the middle and posterior thirds and is seen in figure 24. The shadow terminates posteriorly as a slightly expanded convexity (see figure 24). In the lateral photograph (see figure 30) the shadow has a wide posterior end tapering to a blunt and comparatively narrow anterior end. The light middle area of the shadow is bounded by upper and lower well-defined borders of which the upper is the more dense.

Recognition of the shadow depends upon its position which can be varied if the operator moves the penis before re-photographing and the characters of the elongated shadow with dense borders and an expanded
posterior end.

(2). The shadow of Prepuce in the V-D view can be studied in figures 18 (P.R.), 24, 27 (P.R.); 34 (P.R.) and 36 (P.R.).

The position in relation to the spine naturally varies in all directions, but is situated characteristically at the level of L.6 and L.5. (see figure 36).

The shadow is thumb-shaped with the rounded end anterior. If the preputial hairs have become soiled or matted with urinary concretion they may be picked out as intertwining linear shadows.

(3). The Scrotum occasionally casts a semi-circular shadow which lies at the level of the posterior part of the pubic symphysis. The shadow is dome-like with convexity backwards, and is depicted in several figures 15. (. . .), 17. (. . .), 18. (. . .).

(4). Faecal Material in the lower bowel is easy of recognition, generally shewing as a column of irregular density interpersed with dense centres (bone etc.). Its appearance in a lateral view is seen in 0, figure 29. It can be obviated by giving an enema before photographs are taken.

(5). Gas in the intestinal tract is not common but if morphia has been given or the dog has been held upright for some time, collections of gas occur. A lateral photograph will define its position, and confirm its nature.

(6). The nipples in the female cast a series of small semi-dense circular shadows on either side of the spine. Usually only those nipples behind the level of the liver shadow are reflected. The shadows lie well away from the spine except in oblique photographs.
The Heart and Liver Shadows are readily recognizable; the former as an ovoid obliquely positioned shadow in the thorax and the latter a dome-like transversely situated shadow about the level of the eighth thoracic (figure 41).

Sternebrae. Some of the eight sternebrae can occasionally be recognized as a series of dumb-bell shaped shadows. Their size varies with the breed, but in an airedale are quite as large as the largest lumbar transverse process and owing to their distance from the cassette their shadow is magnified in dimensions. They are generally seen bridging the space between the heart and liver shadows. (6, 6, figure 41).

Spinous processes. The shadows of these structures lie at intervals in the mid-line along the length of the spinal shadow proper; those of the thoracic are often viewed obliquely, their length making it difficult to keep the animal symmetrically on its back.

(a) Cervical Region (figure 42). The spine of the axis simulates an arrow with the point directed forwards; posteriorly it bifurcates, one arm running on to each articular process (figure 42, a, b, c) C.5, C.6 and C.7, carry an ovoid shadow with a dense thin periphery incomplete behind, after the manner of a laterally compressed horse-shoe. These shadows lie in the mid-line, and whilst that of C.5 is about the centre of the body shadow C.6 and C.7 are situated progressively nearer the anterior border. C.3 and C.4 carry a nodule, casting a hardly recognizable shadow.

(b) The Thoracic Spinous processes throw a shadow of similar form to that of the last cervical. They take up an increasingly posterior position in relation to the body shadow until about the third last; the direction of slope of the bony process now alters and the
shadow advances again until at the last thoracic it lies in the centre of the body shadow.

(c). The Spinous process of a lumbar vertebra appears as a laterally compressed ellipse (figure 43). The periphery in its anterior half is dense and thin; the posterior half is indefinite and incomplete; the size is uniform until about L.5 (A, figure 43); the shadow in L.6 and L.7 becomes more knife-like and is represented by two sharply defined lines diverging posteriorly at a very acute angle. (B and C, figure 43).

(d). The Sacral Crest. (figure 43). The shadow cast by this structure can simulate closely that of a streak of lipiodol in the cul-de-sac. Its appearance should therefore be studied. Just as the crest itself can be morphologically divided into three parts so the shadow is separable into anterior middle and posterior thirds. (D, D', D² figure 43).

The anterior part (D, figure 43) consists of a single narrow line of uniform density. It is pointed before and behind and then continued into the middle third (D' figure 29), which simulates a laterally compressed ellipse with a clean-cut periphery and a centre of reduced density. The posterior third (D², figure 43) is in the form of two tapering lines diverging slightly posteriorly and terminating a little in front of the S-Cc space.

(II). The column-shaped shadow of air in the trachea shows, on the V-D negative, sometimes to either side of, and other times imposed upon, the shadow of the cervical vertebrae. The cartilaginous rings seen in section down each border of the shadow are usually demonstrable
and are useful for identification.

In lateral views the radiological appearance of following structures should be recalled.

(I). The Lumbar transverse processes.

These processes sweep downwards and forwards from their base; the posterior ones (L.7,6,5) are much the larger reaching well over, and projecting below, the shadow of the vertebral body next in front. The processes on the more anterior vertebrae, being shorter, pass forward across the joint space of the intervertebral articulation next in front but do not project below the body shadow (figure 44).

The root of the process in the more posterior vertebrae (L.7,6,5) is represented as a thin line in the form of a horse-shoe with the heel directed backwards (C, figure 39 and 44). In the more anterior vertebrae the upper arm of the shoe gradually disappears, and the lower arm becomes flattened and elongated posteriorly. Moreover whilst the root of the process in L.7 is on a level with the shadow of the spinal canal the root of L.1 lies about half-way down the body shadow proper. Between these limits there are different degrees of this change in position.

Since the processes spring from both sides of a vertebra a duplicate series of super-imposed or nearly-related shadows can be made out on any one photograph.

(I3). The appearance of the Articulating Ribs is readily appreciated. (Figure 45).

(I4). The Scapulae. (see figure 46). The lower ends of the scapulae can naturally vary widely in position but usually lie near the
cervico-thoracic junction. Their general form is readily recognized, but the scapula spine should be noted as a dense rod at times represented by two nearly parallel dense contour lines separated by a lighter area; (A,A', figure 46), at other times as a single dense bar (B,B', figure 46), but in any case terminating distally as an expanded button (X,X', figure 46). This is situated at the level of the neck of the scapula and takes the form of a dense thin peripheral shadow with a light centre. If viewed obliquely the scapula spine throws a shadow varying in conformity with its anatomical relation to the scapula and hence easily identified.

It may be observed here, that in taking lateral photographs of the C-T junction it is wise to have one fore-limb drawn well forward and the other well back; thus the scapula shadows are not directly superimposed, and do not form a dense obstruction area.

(15). The External Ear. (figure 47). The long, pendulous, and sometimes hairy ears of certain breeds will cast a striated shadow of variable density. With a dog lying on its side for a lateral photograph this extraneous shadow will be thrown in the angle between the occipital protuberance and the line of the cervical vertebrae (A,A', figure 47). The ears of a patient should therefore be held, or bandaged, forward at the time of photographing.

(5). Gas in the Oesophagus. Traces of gas in the cervical part of the oesophagus appear from time to time on X-ray photographs. In the negative the gas registers itself, of course, as a dark area, and the appearance is exemplified in negatives 109, 197, 223 and 227.
In these instances the gas is usually represented as an elongated shadow lying about one-third of the distance from the upper margin of the trachea to the lower border of the shadow of the cervical vertebrae. The anterior end of the shadow is often blunt and situated at, or behind the level of the axis. Posteriorally the shadow tapers to a fine point reaching back to the level of the fourth cervical vertebra.
PART TWO.

SUMMARY.

Part Two is entitled "Experimental and Clinical Investigations into the use of Iodized Oil as a Radiopaque medium in the Examination of the Spinal Subarachnoid Space of the Dog". It is divided into four sections and an appendix. Thirty-six figures, 7 tables and five charts are included.

In Section A, a review is made of some of the literature on iodized oils and their use in man and the lower animals. Section B, is devoted to a description of original experiments, made by the writer on the injection of Lipiodol Descendant (Lafay) into the subarachnoid space of the dog. Section C, deals with further experimental and some clinical observations on the subarachnoid injection of Lipiodol in relation to the demonstration of certain diseases of the spine of the dog. In Section D, the clinical application of these experiments is discussed. In Appendix I a description is made of the several radiographic features of the spine of the dog, the recognition of which is important in the reading of photographs of Lipiodol injection.

SECTION A. The characters and uses of Lipiodol Descendant (Lafay) and other iodized oils with particular reference to experimental and clinical work in animals. This section was considered under the following headings:

1. The Physical properties of Lipiodol Descendant (Lafay).
2. The chemical characters of Lipiodol Descendant.
3. The antiseptic properties of Lipiodol Descendant.

4. Brief notes on the composition of twelve other types of iodized oils to which reference was found.

5. The uses of Lipiodol and other iodized oils in the human subject with notes on experimental and clinical work in animals. Reference to experimental intra-vascular, intra-spinal and intra-uretral injections were found in the dog. The only reference to the use of Lipiodol in veterinary clinical work was that by Baird and Kennedy (35) who injected the oil into the uterine cavity of a bitch suffering from cystic endometritis. Records were found of experimental work on the cat, rabbit, guinea-pig and rat.

No reference was found to the injection of iodized oil into the subarachnoid space of the dog.

6. The periods for which Lipiodol after injection persists in the different body tissues. Records are quoted relating to subarachnoid injections in the cat and rabbit and intra-uretral injection in the dog.

7. Elimination of Lipiodol after injection into the various body tissues and spaces.

8. Iodism following the injection of Lipiodol; no reference to this in animals has been found, though it is recorded in man.

9. Contra-indications to the use of Lipiodol.

10. The reaction of various tissues to Lipiodol and other iodized oils. This has been studied closely in man by many workers, particularly in the case of subarachnoid injection. In animals the effects of subarachnoid injection of Lipiodol and Iodipin in the cat and
Lipiodol in the rabbit have been studied. No articles have been found referring to similar injections in the dog.

II. The effects of subarachnoid puncture and/or withdrawal of c.s.f. upon a subsequent Lipiodol descent. Observations relating to this problem in man were summarized, and their application to work in the dog pointed out.

SECTION B. The experimental injection of Lipiodol Descendant (Lafay) into the subarachnoid space of the dog.

The experiments conducted by the writer consisted in the injection of varying amounts of Lipiodol Descendant (Lafay), containing 0.54 g. of iodine per c.c. into the Cisterna Magna of five dogs. The animal was then held in the upright position for a certain length of time and the migration of the oil into the sacro-lumbar cul-de-sac recorded on X-Ray photographs. After the experiment each animal was kept under observation. During this period c.s.f. was obtained by cistern puncture and the white cell counts ascertained; further X-Ray photographs were obtained of the oil in the tissues, the changes in its position and appearance being recorded. The programme of experimental work is set out in Table 5.

EXPERIMENT I. DOG 6.

2.5 c.c. Lipiodol were injected into the Cisterna Magna.

The oil filled the lumbar cul-de-sac in 90 minutes. Temporary adhesions occurred at all stages of the experiment along the S-A space chiefly in the cervical and anterior thoracic regions.
Clinically the dog remained unaffected.

The white cell count of the c.s.f. following the injection is set out in Table 6. Over 900 cells were present one day after the injection. Two-hundred and three days later the count had fallen to 1.5 cells per c.mm.

X-Ray photographs were obtained 3 weeks and 27 weeks after the injection. Two changes were observed. Several globules of oil had migrated forward into the cerebral subarachnoid spaces, whilst some of the oil in the cul-de-sac appeared to have escaped from this structure and was disposed in globules within the sacrum.

The following figures relate to experiment I.
Nos: 16, 17, 18 and 19.

EXPERIMENT 2. DOG 14.

0.5 c.c. of lipiodol was injected into the Cisterna Magna.

The oil reached the cul-de-sac in 95 minutes. Certain extensions and dilatations of the shadow of the sac occurred in the succeeding 90 minutes. Temporary adhesions occurred at all stages of the descent along the S-A space chiefly around the cervico-thoracic junction and middle lumbar regions. An apparently permanent arrest occurred within the axis. This termed the "Axis Adhesion" had disappeared some months later.

Clinically the dog was unaffected.

The white cell count of the c.s.f. recorded in Table 7, rose to 348 two days after the injection. It fell to 2.5 about two hundred days later.
K-Ray photographs were taken about two hundred days after the injection. Changes were chiefly restricted to the oil in the cul-de-sac, where the lipiodol had moved forward, broken into masses and streaked and as in Dog 6, some appeared to have escaped beyond the boundaries of the cul-de-sac and lay scattered within the sacrum and some of the coccygeal vertebrae.

The following figures illustrate Experiment 2.
Nos: 22, 23 and 24.

EXPERIMENT 3. DOG 7.

The experiment revealed a projection on the floor of the spinal canal at the level of the lumbo-sacral junction, and this was confirmed post-mortem. The descent was not affected and the data appertaining to this were therefore included in our normal series. The disposition of the cul-de-sac was however modified and its appearance is described separately in Section C. (see later).

1.5 c.c. of lipiodol were injected into the Cisterna Magna.

The oil entered the cul-de-sac 30 minutes later and filled this structure after a total suspension of 50 minutes.

Clinically the dog remained unaffected.

The variations in the cell count of the u.a.f. after the injection, are set out in Table 8. The count rose to 800 cells per c.mm. two days after the injection. After a slight decline a second rise to 4,000 occurred on the seventeenth day. Two hundred days later the count was 1,2.

X-Ray photographs were obtained sixty-two and two hundred days
after the injection. The oil in the cervical region had moved forward to the base of the brain. Unexpected changes had occurred in, and around the cul-de-sac. A great deal of oil lay scattered through the last two lumbar, the sacrum and the first coccygeal vertebrae apparently outside the boundaries of the cul-de-sac. In addition, some of the oil had passed through the intervertebral foramina, outside the spinal canal, where it appeared to follow the ventral branches of some of the spinal nerves particularly those of the sacro-lumbar plexus which go to form the sciatic nerve. (see figures 30, 31, 32).

The following figures illustrate Experiment 3.
Nos: 26, 27, 28, 29, 30, 31, 32, 33.

EXPERIMENT 4. DOG 8.

This dog was affected with spondyleitis deformans at the level of the articulation between the third and fourth lumbar vertebrae. In a dog similarly affected it had been shown that such a lesion appeared to exert no effect upon the movements of oil in the S-A space (see Section C). Dog 8, was accordingly accepted for experiment and the data are as follows:

I. c. c. of Lipiodol was injected into the Cisterna Magna.

The oil entered the cul-de-sac between 100 and 185 minutes after injection. The descent time is thus longer than was the case in dogs 6, 14 and 7. From the position and type of adhesions the writer is of the opinion that the spinal lesion known to be present had no effect upon the movements of oil in the S-A space; the cause of delayed descent was probably the breaking up of the oil into minute globules.
early in the experiment.

Clinically the dog was dull and had a reduced appetite. These symptoms persisted for two days after the injection.

The variations in the cell count of the c.s.f. are set out in Table 9. The count rose to 955 twenty-six hours after injection and fell gradually to 40 cells per c.m.m. thirty-four days later.

X-Ray photographs were obtained thirty-four days after the injection of the oil. The latter had not migrated outside the accepted boundaries of the cul-de-sac; some oil had appeared around the base of the brain.

The following figures illustrate the experiment:-
Nos: 34, 35, 36.

EXPERIMENT 5. DOG 5.

This dog was experimented upon when our technique was imperfect. The data relating to times of descent have therefore been rejected, and only the effects of the injection been considered. This animal was also affected with spondylitis deformans involving the twelfth and thirteenth thoracic vertebrae. The characters of the lesion are dealt with in Section C.

1.5 c.c. of lipiodol were injected into the Sesterna Magna.

Clinically no change was noted in the dog.

The variations in the cell count of the c.s.f. are recorded in Table 10. After six hours the count was 56 and after 24 hours had risen to 980.

The lesion of the spine appeared to have no effect on the movements of the oil along the S-A space.
THE GENERAL EXPERIMENTAL CONCLUSIONS were then dealt with under the following headings:

(a). Rate of the descent of the oil.  (b). The conformation of the arachnoid cul-de-sac.  (c). Adhesions.  (d). Movements and changes in the appearance of the oil during residence in the tissues.  (e). The effect of the injection on the dog.

These observations will be briefly reviewed.

2. The descent of the oil from the cisterna magna to the cul-de-sac occupied an average time of 108 minutes. The shortest period was sixty minutes and the longest 180 minutes. Compared with man the descent is very slow. The writer believes that the small dimensions of the S-A space in the dog are chiefly responsible for the delayed descent, but other factors are discussed.

b. The conformation of the arachnoid cul-de-sac in dogs 6, 7, 8 and 14 is depicted in figures 16, 17, 22, 23, 26, 28, 29, 34 and 35. When filled with lipiodol the cul-de-sac appears on the negative in the form of an inverted cone. The diminution in calibre commences within the fifth lumbar vertebra and the tip of the cone terminates usually within the sacrum. Individual variations were observed. In some instances the calibre of the sac would undergo sudden reduction, the structure then continuing as a thread-like prolongation within the sacrum. Such a diminution was usually observed about the level of the lumbosacral junction. In other instances the posterior part of the sac after diminishing to a fine point exhibited irregular dilatations in the form of droplets of oil situated usually to one side of the mid-line. These dilatations
commonly occurred within the coccygeal vertebrae and their connection with the anococcygeal canal—were usually demonstrable. (See figure 22).

2. Adhesions occurred at all stages of the descent. They were most common about the cervico-thoracic junction and the region of the second, third and fourth lumbar vertebrae. In such situations they were usually globoid in form whilst in the thoracic region, when present they were linear in shape. In most cases they moved during the course of an experiment and could not be looked upon as false arrests. A large mass which was usually to be found dorsal to the cord at the level of the anterior border of the arch of the axis was the exception. This adhesion occurred regularly, and remained stationary throughout the period of suspension. It disappeared in all cases during the first few weeks after injection.

3. After injection three main changes were noted. Firstly the appearance of globules around the base of the brain; secondly the collection of oil along the floor of the canal especially at its lowest points, and thirdly modifications in position and appearance of oil in the anococcygeal canal.

The migration to the cerebral subarachnoid spaces was noticed in the case of dog 8, thirty-four days after the experiment. The oil probably gravitates to this position as the result of the posture of a dog when lying down or eating.

The collection of oil along the floor of the canal, as at the cervico-thoracic junction, is probably a result of gravitation.
The modifications in the cul-de-sac were of some interest. In dogs 6, 7, 8 and 14, at varying periods after injection, globules of oil appeared outside the usual accepted limits of the cul-de-sac. The process in Dog 8 was observed within five weeks of the injection. In Dog 7, the oil not only spread throughout the bony canal, within the last two lumbar vertebrae, the sacrum and first coccygeal but appeared to pass out through the intervertebral foramina and follow the course of some of the spinal nerve branches. The X-Ray appearance of the tracks of oil was reminiscent of a tracing of some of the roots of the lumbosacral plexus of nerves. (See figures 30, 31, 32).

2. The effect of the injection upon the dogs was very slight. Only one dog (Number 8) showed discomfort after the injection. The c.s.f. showed a rapid but temporary increase in the white cell content in all cases. In all the dogs the count fell gradually, though irregularly and, in the case of patients kept for about two-hundred days, returned to normal. No microscopical examination of the spinal cord or its membranes or nerve roots was carried out. On post-mortem examination the macroscopic appearance of these structures was normal.

SECTION C. Lipiodol Descendant and the demonstration of certain spinal lesions in the Dog.

Lipiodol was injected in the manner described above into:-

(a). A dog in which the intervertebral disc between the last lumbar vertebra and the sacrum was enlarged.
(b). A dog which had sustained a compression fracture of a thoracic vertebra.

c. Two dogs affected with spondyleitis deformans.

m. Dog 7, one of the experimental group, was injected with Lipiodol Descendant in the usual way. The cul-de-sac filled readily with oil, but at the level of the lumbo-sacral junction there was an indentation in the border of the shadow and a transverse band of reduced opacity. (Figure 26).

The patient was placed in a horizontal position when oil in the lumbo-sacral region will usually pass forward again. Oil anterior to the defect in the shadow gravitated forward but that behind the defect remained imprisoned in the cul-de-sac. (Figure 27). The animal was placed upright again for some time, and a picture similar to figure 26 obtained (figure 28). A lateral photograph showed a deflection of the cul-de-sac at the level of the lumbo-sacral articulation where it was thrust upwards as if surmounting an eminence. (Figure 29). On the floor of the canal at post-mortem examination carried out some months later, an enlargement of articular disc was found which had expanded up into the floor of the spinal canal carrying the cul-de-sac towards the roof. A similar condition has been described by continental clinicians, but not at this site.

b. A dog which had been run over was examined. X-ray photographs showed a fracture of the tenth thoracic vertebra. All the clinical signs of spinal compression were present. Lipiodol Descendant was injected into the cisterna magna. The oil was arrested at the
level of the fracture and clearly demonstrated the nature of the injury. (Figure 48).

Lesions of spondyloitic deformity were found during the preliminary examination of two of the experimental dogs; numbers 5 and 6. In Dog 5 the lesion involved the articulation between the twelfth and thirteenth thoracic vertebrae. In Dog 6 the third and fourth lumbar vertebrae were involved.

In both cases the descent of Lipiodol was studied but the lesion exerted no effect upon the movements of the oil within the subarachnoid space. It is therefore probable that the inco-ordinat ion observed in some clinical cases exhibiting these lesions is not due to pressure on the cord. Neither Dog 5 nor Dog 6 however exhibited clinical symptoms.

From the X-ray photograph it was shown that the pathological process commenced on the anterior of the two vertebrae involved and in the mid-ventral line. The lesion is best detected by lateral photographs but, if advanced will be revealed on X-D examination.

SECTION D. Clinical applications of the Subarachnoid injection of Lipiodol in the dog.

The Queckenstedt Test and the bio-chemical examination of c.s.f. in front and behind the suspected spinal block, tests which have proved so helpful in the diagnosis of spinal compression in the human being, both involve the insertion of a needle into the subarachnoid space in the lumbar region. This is difficult and in many cases impracticable in the dog because of certain anatomical peculiarities. A method
which can be used to test the patency of the subarachnoid space in the dog is therefore of definite clinical importance. Lipiodol can be used as a radiopaque medium for this purpose, and as the experiments show, is quite safe in the normal dog. Since however, it is known to possess irritant qualities when injected into the human spine where some lesion is present it should only be used in selected instances.

Lipiodol has several disadvantages in the dog, chiefly of which is the slowness of its movements within the SA space.

Appendix One. Certain features of the spine of the dog, the recognition of which is important in the reading of photographs of lipiodol injections.

In X-Ray photographs of the dog's spine the shadows of certain structures are superimposed upon the spinal shadow proper. The recognition of these especially where they are absent in men (e.g. os penis) or have a characteristic appearance (the sacral crest) is important to the worker who has had no veterinary training. An appendix has been added which together with figures defines the appearance and relations of the shadows of such structures as the os penis, acratus, prepuce, external ear, lumbar transverse processes and the sacral crest. Figures 41-47 serve to illustrate these points.
CONCLUSIONS. PART TWO.

1. Lipiodol Descendant (Lafay) containing 0.54 g. of iodine per c.c. (hereinafter referred to as Lipiodol) is a radiopaque medium which may be safely injected into the subarachnoid space of the normal dog.

2. After injection into the disterna magna the oil will descend if the dog be held upright, into the subarachnoid cul-de-sac.

3. The average time of descent, in our experiments is 110 minutes.

4. The conformation of the subarachnoid cul-de-sac may be demonstrated radiologically by filling it with Lipiodol.

5. The cul-de-sac shows individual variations in shape; generally speaking it is in the form of a cone with the apex directed towards the tail. The reduction in calibre commences within the fifth lumbar vertebra and the tip of the cone lies within the sacral canal.

6. The injection has little noticeable effect upon the dog.

7. In one case loss of appetite and spinal pains occurred, but were of short duration.

8. The white cell count of the c.s.f. increases in the first twenty-four hours after the injection. The average count at this stage is 796 cells per c.mm.

9. Some months after the experiment the white cell count of the c.s.f. falls to within the normal limits defined in Part One of this paper.

10. The cell count does not fall uniformly after the initial increase.
At variable periods after the injection secondary rises occur to be followed again by a subsidence.

10. The highest secondary rise was one to 4,000 cells per c.mm. in Dog 7.

11. No clinical symptoms accompanied this secondary cell increase and bacteriological investigations of the c.s.f. were negative.

12. It is believed that these secondary increases in cell content depend upon the migration of globules of oil to new sites within the spinal or cerebral subarachnoid spaces.

13. Three dogs were observed for some six months after the injection. Their condition, appetite and spirits remained unaffected; they could jump over obstacles and run at fast paces; they exhibited no signs of inco-ordination or spinal irritation.

14. Lipiodol injected into the cisterna magna of the dog and allowed to descend into the arachnoid cul-de-sac exhibits the following changes in position or appearance in the first six months following the operation:—

A. Several globules migrate to the base of the brain; they rest in and on either side the mid-line within an area overlying the body of the occipital bone, the body and the wings of the sphenoid bone and in some instances extending as far forward as the ethmoid bone.

B. Oil gravitates to the floor (i.e. ventral aspect) of the subarachnoid space particularly in the region of the cervico-thoracic junction.

C. Oil within the cul-de-sac tends to move forward along the
floor of the subarachnoid space splitting into mases and globules.

D. Globules of oil appear outside the limits of the cul-de-sac and are situated apparently in the epidural space. In one instance oil passed outside the bony canal through the intervertebral foramina and appeared to follow the course of the radicles of the sciatic and other nerves as far as the region of the hip joint. The precise position of the oil in this case and the mechanisms and route by which this position was assumed cannot be defined from these experiments. It is believed that the process represents an insinuation of oil along the nerve roots.

15. Lipiodol was injected into the Cisterna Magna in a suspected case of thoracic spinal block in the dog. The presence and site of pressure were demonstrated clearly.

16. Lipiodol was injected into the subarachnoid space of a dog in which the intervertebral disc between the last lumbar vertebra and the sacrum was enlarged and projected from the floor of the spinal canal. The cul-de-sac was filled with oil; the manner in which the sac was deflected at this level was then demonstrable by X-Ray photography.

17. Spondyleitis deformans, examined in two dogs, exerted no effect upon the movements of lipiodol within the subarachnoid space.
REFERENCES. PART TWO.


20. Private communication from the Lafay Laboratories.


25. 


31. See 23.


36. See 22.

37. See 27.


40. See 15.


48. See 45.


55. See 4.

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See also. J. Bone and Joint Surg. 1926. 8. 348-353.


66. See Reference 10.


68. See 14.


75. See Reference 24.

"The writer is indebted to the Lafay Laboratories for the provision of the following References."


ACKNOWLEDGEMENTS.

This work was carried out during the tenure of the Centenary Post-Graduate Research Fellowship of the Royal (Dick) Veterinary College, Edinburgh.

The expenses were contributed to by the Committee of the Centenary Post-Graduate Research Fellowship of the Royal (Dick) Veterinary College, and the Endowment Committee of the Earl of Moray Endowment for the Promotion of Original Research, Edinburgh University. The writer is indebted to both these Committees for the considerable grants which were made.

The writer desires to thank:

Principal O. Charnock Bradley, Royal (Dick) Veterinary College, Edinburgh, for the kindly interest taken in, and the helpful advice concerning the investigations.

Professor D.P. M. Wilkie, and the technical staff of the Department of Surgical Research, University of Edinburgh, for help with an accommodation for the experimental work; without these facilities it would have been impossible to have instituted these investigations.

Professor W. S. Mitchell, and the staff of the Department of Surgery, Royal (Dick) Veterinary College, where most of the post-mortem examinations were made.

The Governing Committee of the Stray Dogs' Home Edinburgh, for permission to use the carcasses of dogs for anatomical investigations.
Dr Hope-Fowler, Medical Radiologist, Edinburgh, for considerable assistance in obtaining the preliminary X-Ray photographs of the experimental subjects.

Professor F.A.E. Crewe, Animal Breeding Research Department, University of Edinburgh, for providing accommodation for the experimental dogs during the periods of observation.

Professor F.T.G. Hobday, Royal Veterinary College, London, for kindly permitting the use of data referring to patients in his clinic.

Messrs Dengue and Company, Limited, Fitzroy Street, London, for supplying the Lipiodol (Lafay) used in the experiments free of charge.

Finally, Mr C.A. Grove, V.S.R, Technical Assistant to Dr Hope-Fowler, who gave so freely of his abilities and time; without his invaluable help it would have been impossible to have carried out the considerable amount of X-Ray photography included in the experiments in Part Two.