FREE ILEAL AUTOGRAFT AS A SUBSTITUTE FOR THE THORACIC OESOPHAGUS

An experimental study in adult dogs and puppies.

PROEFSCHRIFT

TER VERKRIJGING VAN DE GRAAD VAN DOCTOR IN DE GENEESKUNDE
AAN DE ERASMUS UNIVERSITEIT ROTTERDAM
OP GEZAG VAN DE RECTOR MAGNIFICUS
PROF. DR. J. SPERNA WEILAND
EN VOLGENS BESLUIT VAN HET COLLEGE VAN DEKANEN.
DE OPENBARE VERDEDIGING ZAL PLAATSVINDEN OP
WOENSDAG 28 MEI 1980 DES NAMIDDAGS
TE 4.15 UUR

door

Jan Dirk Meeuwis

Geboren te Bussum

grafische verzorging:
davids decor alblasserdam

PROMOTOR : PROF. DR. J.C. MOLENAAR
CO-REFERENTEN: PROF. DR. D.L. WESTBROEK
PROF. DR. H. VAN HOUTEN

To my parents.

Japie Sarina Josien Margreet Lex Marijke Emma Joseph Borus Bobbie Franka Frida Bora Bonitos Cornelia Erna Toon Vincent Jonas Remco Kuno Dewi-Sinta Juno Athene Deirdre Meutje France Karel Hein Annebelle Rozemarijn.

Und einmal,
eines Tages,
musst du fragen:
Was hast du mit der Zeit getan,
mit der Zeit deines Lebens?
Wo sind deine Freunde,
deine Jugend, wo?
Wo ist deine Liebe,
die dich einst gehindert hat
am Schlafen.

Bertrand Alfred Egger.

CONTENTS

CHAPTER 1 INTRODUCTION 11

1.1. Clinical approach to oesophageal reconstruction . 11
1.2. Rationale of the study 12
1.3. Objectives of the study 14

CHAPTER 2 MATERIAL AND METHODS 15

2.1. Laboratory animals 15
2.2. Pre-operative treatment 15
2.3. Anaesthesiological management 15
2.4. Surgical technique 16
2.5. Immediate post-operative management 23
2.6. Follow-up studies 23
2.6.1. Nutrition and bodyweight 24
2.6.2. Cinematography 24
2.6.3. Oesophagoscopy 24
2.6.4. Angiography 25
2.6.5. Morbid anatomy 26

CHAPTER 3 RESULTS 27

3.1. Operative results and mortality 27
3.2. Follow-up studies 34
3.2.1. Nutrition and bodyweight 34
3.2.2. Complications 37
3.2.3. Cinematography 38
3.2.4. Oesophagoscopy 43
3.2.5. Angiography 46
3.2.6. Morbid anatomy 48

CHAPTER 4 DISCUSSION AND CONCLUSIONS 53

4.1. The intestinal segment 53

4.2. Preservation of the transplant 54
4.3. Denervation and lymphatic drainage of the transplant 54
4.4. Selection of recipient vessels 55
4.5. Revascularization 55
4.6. Assessment of the vitality of the transplant 56
4.7. Vascularization of the transplant 56
4.8. The oesophagus-bowel anastomosis 57
4.9. Deglutition - Functional analysis of the oesophagoplasty 57
4.10. Morbid anatomy 58
4.11. Nutrition and growth 59
4.12. Dog no. 5 59
4.13. Conclusions 60

SUMMARY 61
SAMENVATTING 63
REFERENCES 65
AKNOWLEDGEMENTS 69
CURRICULUM VITAE 71

1 INTRODUCTION

1.1. Clinical approach to oesophageal reconstruction

Reconstruction of the oesophagus for both malignant and benign disease is a matter of controversy. Uncertainty exists about the best method of restoring gastro-intestinal continuity.

In malignant disease the discussion remains restricted to personal preferences and technical details of the procedure. It concerns patients who have a poor life expectancy. Therefore re-establishment of the continuity is the primary concern. This objective can be effected satisfactorily using colon (Belsey 1965) or stomach (Collis 1972), the substitutes of choice. In benign disease, however, the normal expectancy of life following reconstruction demands not just a satisfactory but the best possible solution, especially where children are concerned. And yet the same debatable methods of reconstruction are used, with colon or stomach the preferred substitutes, and the jejunum as an alternative (Merendino and Dillard 1955).

Each substitute has its disadvantages. Stomach substitutes may give rise to intrathoracic distress, regurgitation, oesophagitis, stricture and dumping syndrome (Hanna et al. 1967). Colonic substitutes may cause complaints varying from fullness in the neck, palpitations, foul breath and regurgitations to dumping syndrome (Grimes 1960). Functionally the colon has a rather delayed passage and retains food in the haustral pockets (Clark et al. 1976), sometimes intensified by a siphon-shaped position and herniation of the bowel in the thoracic cavity (Louhimo et al. 1969). Peptic ulcer in the colon and stenosis of the colon-gastrostomy are also reported (Malcolm 1968). The objections to the use of the jejunum are largely anatomical, short vascular arcades limit the available length (Grimes 1967).

Comparative function studies (motor activity, transit time, food retention, regurgitation and plasticity) of visceral oesophageal substitutes, however, favour the small bowel (Hanna et al. 1967, Marshall 1972, Rodgers et al. 1978). Oesophagoplasties using skin require multiple stage procedures and are complicated by fistulas and stricture formation.

The fact that no single surgical procedure is universally accepted indicates that none of the procedures brings the aimed for solution. The aforementioned problems justify an ongoing search for new paths.

New methods have already been developed and tested with laboratory animals and on a very small scale these methods have been applied to human patients. Circular myotomy of the oesophagus has been introduced to bridge a gap of several cms (Livaditis et al. 1972). In oesophageal atresia elongation of the segments by bougienage (Woolley et al. 1969) and inducing a fistula along a thread with dilatation of the resultant fibrous canal have been reported to restore the continuity (Rehbein and Schweder 1971). These three methods by which substitution is avoided have not always been clinically successful and more often than not they end in strictures and bad functional results needing prolonged treatment. Prostheses of various materials and allografts are tested experimentally and sometimes even clinically: plastic tubes, Marlex mesh, silastic, teflon, silicone rubber and aorta or lyophilized dura as allografts. None of these substitutes have proved satisfactory since the prostheses dit not adhere to the tissue nor did they remain patent and all allografts ended up as strictures.

With the advent of reliable techniques in vascular surgery autotransplantation of a segment of the intestinal tract has become feasible. Using this technique the small bowel, functionally the most suitable substitute but limited in its use for vascular reason, has become more applicable. Long vulnerable vascular pedicles can be avoided. Since the first experimental and clinical report on restoring the cervical oesophagus with a revascularized jejunal graft (Seidenberg et al. 1959) this method has become a practical reality (Grage and Quick 1978). Small segments of jejunum, colon (Nakayama 1967) and stomach (Hiebert and Cummings 1961) were transplanted to repair defects in the cervical oesophagus.

1.2. Rationale of the study

The urge for providing a better, more physiological method of reconstructing the thoracic oesophagus is very actively felt in paediatric surgery. For in paediatric surgery small defects in the thoracic oesophagus e.g. oesophageal atresia, peptic or caustic strictures, are bypassed with colon or stomach with considerable morbidity as a result (Otherson et al. 1967, Rehbein and Schweder 1971). In these cases large or small segments of viable oesophagus are resected or bypassed, often inclusive of the lower oesophageal sphincter.

In oesophagoplasty for benign disease the aim of the surgical reconstruction should be threefold:

to save as much oesophagus as possible;
to preserve the sphincter;
to provide the best functional substitute.

Some efforts have been made to meet these objectives. Waterston (1964) introduced a technique that preserved the lower oesophageal sphincter,

using a pedicled colonsegment. This technique however, brings along the morbidity inherent in colonic substitutes. Pichlmaier (1973) interposed a pedicled segment of jejunum in the lower part of the oesophagus preserving the lower oesophageal sphincter. This method still has the disadvantage of a vulnerable vascular pedicle and is therefore limited to the lower part of the oesophagus.

From the progress in autografting intestinal segments in the cervical region a logical sequel is to perform an analogous operation in the thoracic cavity.

Only 3 reports of experimental research concerning thoracic oesophageal replacement with transplanted intestinal segments have been found in the literature.

Meier (1968) reported a study with 43 dogs. He transplanted a segment of small bowel in the thoracic cavity, using the internal mammary artery or an intercostal artery and the hemiazygos or azygos vein. One dog survived for 6 months and died of cachexia. The other 42 dogs died within 2 weeks after surgery.

Kozuschek et al. (1972) performed two series of experiments. In the first series he used a segment of autologous large bowel with the inferior mesenteric artery with a patch of the aorta and inferior mesenteric vein serving as vascular pedicle. The vessels were anastomosed to the descending aorta and the azygos vein. The 8 dogs that were operated upon died all within 2 days after surgery. In the second series a segment of small bowel was harvested from a donor dog. The superior mesenteric artery and vein that would serve as nutrient vessels were transsected just where they branched off from the aorta and the caval vein. The operation was completed in the same way as in the first experiment. This allogenic transplantation was performed on 20 dogs. Eleven dogs succumbed within 5 days while the other 9 dogs were sacrificed 5 or 6 days after surgery before acute graft rejection could occur.

Dragojevic et al. (1975) transplanted a segment of autologous small bowel to the thoracic cavity using the aorta and azygos vein as vascular supply. Of the 20 dogs operated upon, 12 dogs died within 6 days due to necrosis of the transplant and the other 8 dogs were sacrificed 8 days after surgery. His main concern was the monitoring of the vitality of the transplant in the post-operative period. The recording of the electromyogram of the transplant proved to be a reliable method.

In this way several unsatisfactory attempts have been made to apply intestinal autografting to the reconstruction of the thoracic oesophagus. The results were disappointing and a follow-up study was impossible due to poor survival. The vascular technique proved to be the major difficulty. Similar studies have not been performed on small, young and rapidly growing individuals.

A further study of the potential of a surgical technique for bridging the thoracic oesophagus seemed justified and might prove useful. The aim should be to preserve the lower oesophageal sphincter while using the small bowel as a substitute. The technique should be applicable to small, young and rapidly growing individuals and a follow-up study was mandatory.

1.3. Objectives of the study

The aim of this experimental research in dogs was threefold.

- *The first objective was to develop a reliable technique to bridge a defect in the thoracic oesophagus with an autologous ileal transplant.*

- *The second objective was to apply the developed technique to puppies.*

- *The third objective was to study the function and morphology of such an oesophagoplasty in adult dogs as well as in puppies during a period of rapid growth. A follow-up study of one year was planned for each animal consisting of clinical observation, macroscopic and microscopic examination of the interposed ileal segment, swallow cinematography and angiography of the transplant.*

The research was carried out in two series of experiments; the first series involved young adult dogs and the second series dealt with puppies. The experiments were carried out in sequence. The methods used in both series did not differ essentially. However, we did modify our technique to some extent in the second series as a result of experience gained in the first series and also based on a pilot study carried out with puppies before starting the second series. Because of these modifications the following chapters and paragraphs will occasionally be divided into two parts, A and B. Part A will describe the technique applicable to both series, whereas part B will only describe the modifications introduced in the second series conducted with puppies. In this way unnecessary duplication is avoided.

2 MATERIAL AND METHODS

2.1. Laboratory animals

A. First series

In the first experimental series the laboratory animals consisted of 20 beagles, obtained from "Het centraal proefdieren-bedrijf", Austerlitz, the Netherlands, chosen at random. Their weight at operation ranged from 7.9 - 15.5 kgs with an average of 11.6 kgs. Their age was 13, 14 or 15 months.

B. Second series.

In the second experimental series the laboratory animals consisted of 11 mongrel puppies from two different litters, with 7 puppies from one litter and 4 from the other one. The puppies were obtained from local dog handlers; bodyweights of the respective parents were unknown. Five and six weeks after birth they were brought into the laboratory kennels. At surgery the puppies weighed from 8.0 - 10.0 kgs, with an average weight of 9.3 kgs and in age they varied from 8 - 10 weeks.

2.2. Pre-operative treatment

A. For pre-operative reduction of the gut flora Neomycin® (Lundbeck) was given orally, 1 g daily, for two days before surgery. No solids or liquids were given for 16 hours pre-operatively. As pre-medication atropine ½ mg i.m., and Thalamonal® (Janssen), 2 mls i.m., were given 30 minutes before anaesthesia. 2500 U of heparin were admistered subcutaneously. The abdomen and the right side of the chest were shaved.

B. In addition the puppies were routinely dewormed with piperazine (Intervet) during their pre-operative stay at the laboratory.

2.3. Anaesthesiological management

Anaesthesia was inducted with Pentothal® (Abbott) i.v. Following endotracheal intubation the animal was ventilated with a Bennet respirator with O_2, N_2O and Ethrane® (Abbott). Whenever necessary Fentanyl® (Janssen) and Pavulon® (Organon Teknika) were given.

During the operation antibiotics were started, 1 million units of penicillin and 500 mgs of streptomycin daily i.m. During surgery 100 mls of Rheomacrodex 10%® (Organon Teknika) was given i.v.

2.4. Surgical technique

A. The animal was placed upon the operation table on its left side and the lower part of the body was turned so as to lie with the belly upwards. After desinfecting the skin with 70% ethyl alcohol, sterile dressings were placed in such a way that the abdomen and the right side of the thorax were equally accessible. The operation was performed in five stages:

Preparation of the transplant;
Preparation of the recipient vessels and oesophagus;
Perfusion and revascularization of the transplant;
Resection of the oesophagus and interposition of the transplant;
Wound closure.

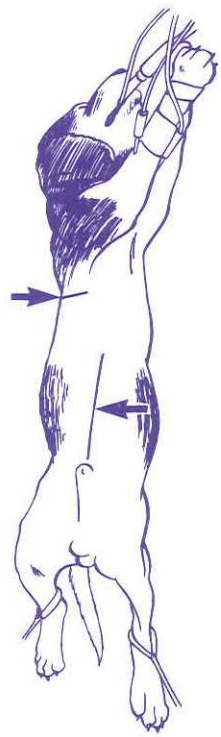


Figure 1: Position of the dog on the operation table. Arrows indicate the incision lines.

Preparation of the transplant.

The abdomen was opened through a median incision. The abdominal organs were inspected. The total length of the small bowel was measured from the ligament of Treitz to the ileocolic orifice. A segment of the distal ileum supplied by a good ileal artery was chosen to serve as transplant. The mesenteric lymph nodes lying along the vessels in the mesentery were removed for the most part. The vessels supplying the ileal segment that had been selected were mobilized to a point where the diameter of the superior mesenteric artery was large enough to permit vascular anastomosis. The mesenteric vein was always larger than the mesenteric artery. Since the ileal arteries in the dog form only one poorly developed arcade close to the ileum, it was always necessary to dissect more of the ileum than needed for the resultant transplant. The ileal segment was dissected from the bowel leaving its vascular pedicle intact. A specimen of the ileum was taken for histological examination. The oral side of the transplant was marked with a long silk suture. The continuity of the bowel was restored with an end-to-end anastomosis using a one layered continous 3.0 chromic catgut suture. The mesentery was approximated using interrupted 3.0 chromic catgut sutures.

Preparation of the recipient vessels and oesophagus.

The thoracic cavity was entered through a right-sided thoracotomy in the 4th intercostal space. The right surface of the mediastinum was approached transpleurally. The internal mammary artery and the superior vena cava were selected as recipient vessels. As in man the internal mammary artery in the dog branches off from the first portion of the subclavian artery and descends into the chest along the lateral margin of the sternum. The internal mammary artery in the dog reaches the sternum at the level of the 4th intercostal space. The artery passes through the thoracic cavity and lies in a double fold of the pleura. The internal mammary artery was dissected from the pleura. A small vascular clamp was placed upon the proximal part of the artery. After ligation close to the sternal side, the artery was transsected just proximal to the ligature. The artery was then irrigated with heparinized saline solution 0,9%. Any superfluous adventitia from the anastomotic site was removed. With the internal mammary artery mobilized in this way enough length was obtained to reach the azygos vein easily by rotating the artery to the posterior mediastinum. Subsequently the oesophagus was dissected from the mediastinum. The mediastinal pleura was opened lengthwise. The azygos vein was ligated and transsected. It was then easy to free the oesophagus from the loose connective tissue. Preparation of the superior vena cava consisted of incising the mediastinal pleura just behind the vein to allow sideways clamping with a vascular clamp.

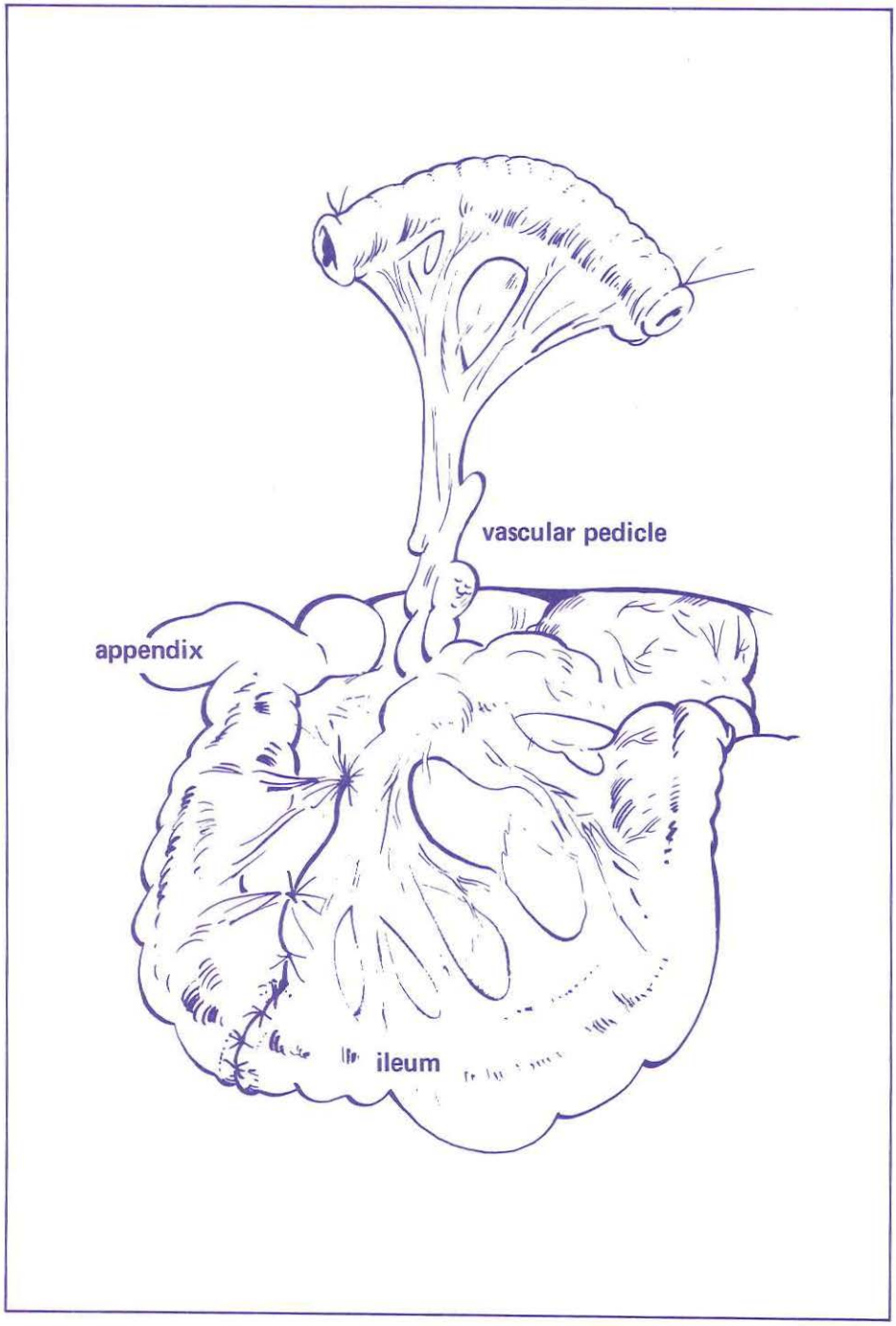


Figure 2: The selected ilealsegment is prepared for transplantation. The continuity of the bowel is restored.

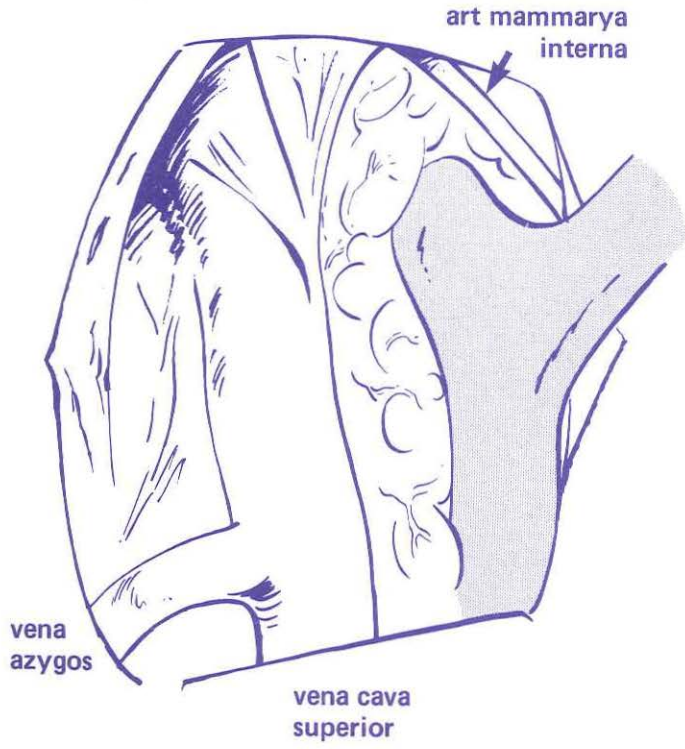


Figure 3: View of the anatomical situation after right-sided thoracotomy in the 4th intercostal space.

Perfusion and revascularization of the transplant.

The artery of the transplant was ligated proximally. The distal part was cannulated and the vessels of the transplant were perfused with a solution of 15 mgs heparin and 10 mgs procaine in 500 mls saline 0.9% at a temperature of 4° C. The vein was ligated and transsected. The transplant was removed from the abdomen. No specific perfusion flow was maintained but a constant perfusion pressure of 100 cms was exerted until macroscopically clear, blood free effluent was obtained.

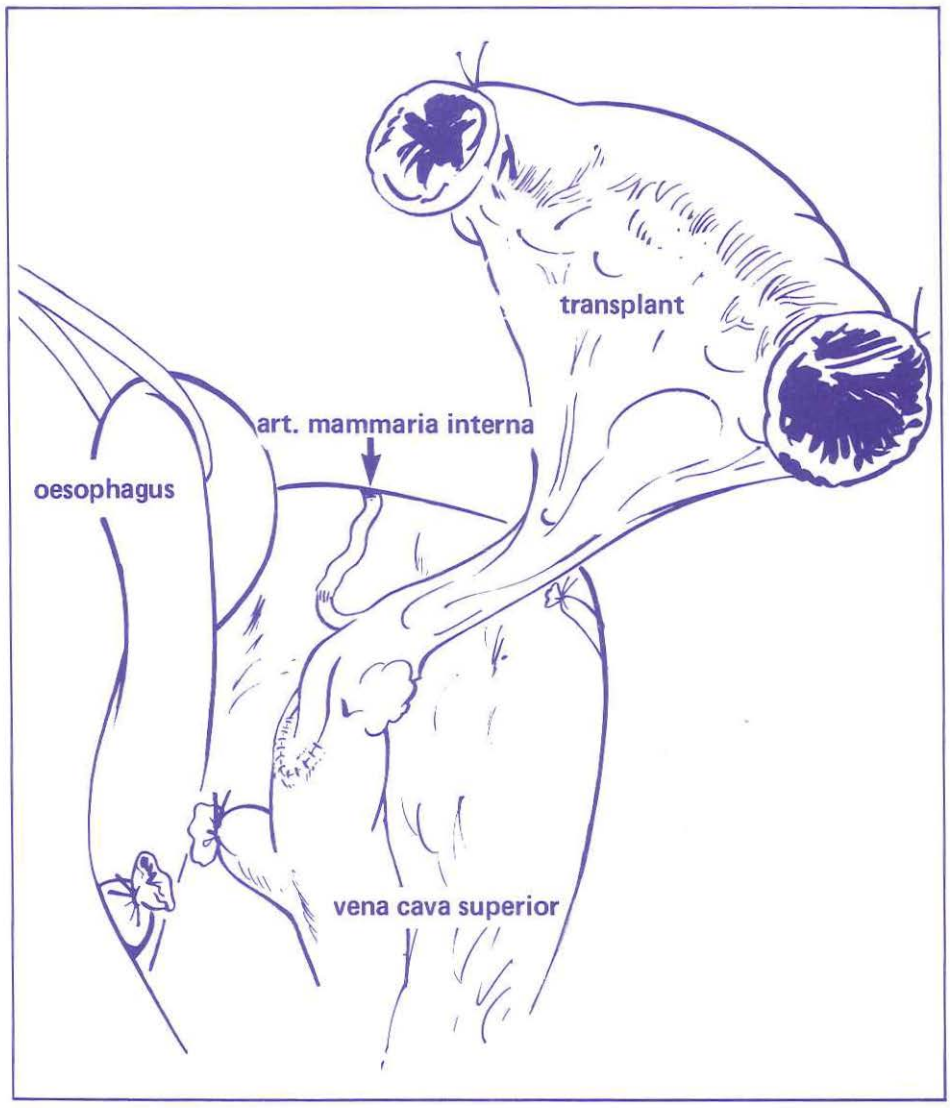


Figure 4: The transplant is revascularized in the thoracic cavity.

The transplant was brought to the thoracic cavity for revascularization. First the mesenteric vein was anastomosed. After clamping the vena cava sideways a small oval part of the wall of the vena cava was excisted with a pair of scissors. The veins were approximated with two guide sutures and then anastomosed end-to-side with a continuous over-and-over suture technique using 7.0 silk. Anastomosis was aided by the ongoing perfusion of the transplant which separated the thin walls of the veins. The perfusion was stopped during the last two stitches. Then the mesenteric artery was anastomosed. The artery was trimmed back because of the damage done by the perfusion cannula. Any loose perivascular tissue was removed. The internal mammary artery and the mesenteric artery were then approximated with two guide sutures and anastomosed end-to-end with a continuous over-and-over suture technique using 7.0 silk. Both vascular anastomoses were performed without the use of magnifying glasses or operating microscope. After releasing the clamp from the superior vena cava and the internal mammary artery the transplant was recirculated. The ischaemic time was noted. The ischaemic time was defined as the time between ligation of the mesenteric artery in the abdomen and recirculation of the transplant. The vitality of the transplant was confirmed by observing colour and the peristalsis of the bowel and the pulsation of the vessels. Immediately after revascularization a specimen of the ileum was taken for histological examination.

Resection of the oesophagus and interposition of the transplant.

Next a segment of the thoracic oesophagus at the level of the tracheal bifurcation was resected. The transplant was shortened to a suitable length. The normal anatomical position of the mesenteric vessels did not permit suturing the vessels in such a way that the transplant could be interposed isoperistaltically. Consequently the transplant was interposed anti-peristaltically to prevent kinking of the vessels. The proximal and distal oesophagus-bowel anastomosis was performed using a two-layered telescopic technique (Haight 1969). The first layer was made with a continuous suture using 3.0 chromic catgut sewing the mucosal layer of the oesophagus to the full thickness of the ileal wall. The second layer was made with interrupted sutures using 3.0 silk, stretching the muscularis of the oesophagus telescopically over the first layer and attaching this to the serosa of the ileum. For identification on X-ray examination each oesophagus-bowel anastomosis was marked with a haemoclip.

Wound closure.

After inspection of the abdomen the abdominal wall was closed in two layers. The thoracic cavity was washed thoroughly with warm saline solution 0.9%. Tetracyclin 275 mgs was deposited in the thorax. A drain was inserted

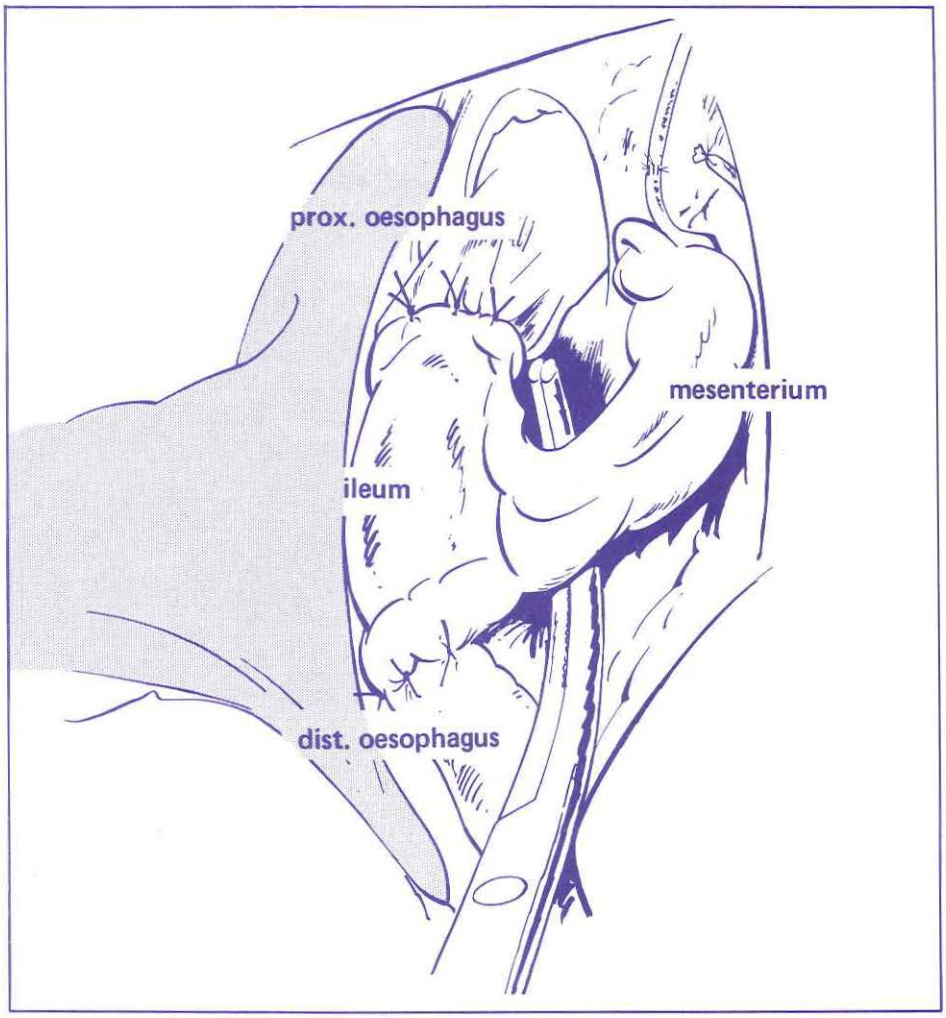


Figure 5: Interposition of the transplant in the oesophagus has been completed. Note the vascular pedicle lying on the clamp.

through a separately tunneled stab wound and fixed to the skin. The thoracic wall was closed in layers.

B. In the puppies only 7.0 nylon suture material was used for the vascular anastomoses. All vascular anastomoses were performed with the aid of 3x magnifying glasses.
The arterial anastomosis was effected using interrupted sutures while the venous anastomosis was sutured in a continuous technique.
In the puppies the artery and vein in the mesenteric pedicle were separated for about 3 cms. This allowed for an iso-peristaltic interposition instead of an

anti-peristaltic interposition without the risk of vascular complications such as kinking or torsion.
Interposing the transplant between the two oesophageal ends great care was taken to interpose the same length of the removed oesophageal segment. The transplant was always positioned accurately in the bed of the oesophagus behind the trachea. This was done by approximating the mediastinal pleura with one silk suture.

2.5. Immediate postoperative management.

A. The first postoperative day another 100 mls of Rhemacrodex® (Organon Teknika) was given intravenously. For 3 weeks postoperatively 2500 U of heparin daily was administered subcutaneously. The antibiotics started during surgery were continued for the next 5 days.
The thoracic cavity was drained with intermittent suction and the thoracic drain was removed on the first postoperative day.
The animals were given 1 - 1½ litres fluids parenterally, through hypodermoclysis, glucose 10% and saline 0.9%, for one week. During this week the dogs did not get any water orally and to inhibit the secretion of saliva they got ¼ mg atropine 3 times daily, intramuscularly.

B. The puppies, who where considered to have a more unstable circulation, were given an extra 200 mls glucose 10% and 100 mls plasma intravenously 6 hours postoperatively. No atropine was given to the puppies postoperatively.

2.6. Follow-up studies.

The dogs were followed for one year. Various examinations were carried out regularly. The follow-up studies covered:

Nutrition and bodyweight;
Cinematography;
Oesophagoscopy;
Angiography;
Morbid anatomy.

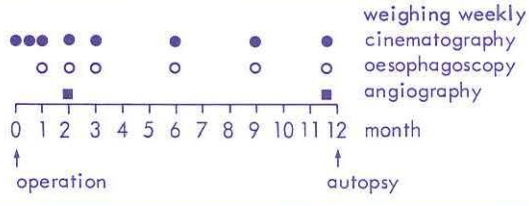


Figure 6: The follow-up programma the dogs were subjected to.

2.6.1. Nutrition and bodyweight.

One week after surgery the adult dogs were given water orally and put on a liquid diet (glucose and Protifar®, Nutricia, Zoetermeer). Subsequently the diet changed to semi-solids (Pelsifood®, Trouw Veevoeder Fabrieken, Putten) and as soon as possible to a normal diet (Canex® dogfood, Hope Farms, Woerden). The puppies of the first litter (nos. 1-6) were given water orally from the 5th postoperative day and the puppies of the second litter (nos. 7-11) were allowed to drink water from the first postoperative day.
During the follow-up year the eating habits of each dog (liquid, semi-solid or solid oral intake and vomiting) were noted. The bodyweight was checked once a week before eating and a curve representing the bodyweight was constructed.

2.6.2. Cinematography.

During the follow-up year 8 swallow cinematographies were made of each animal, after the 1st, 3rd, 5th, 9th, 13th, 26th, 39th, and 51st week. Except for 5 examinations that were made with fluoroscopy only for technical reasons, all examinations were recorded on X-ray films taken at 16 frames per second. Gastrografin® (Schering) was used as contrast medium.
On the day of examination food was omitted prior to the cinematography. The dog was fixed in a harness and then left hanging with its side against a vertically placed X-ray screen. In this way the film was taken with the dog in a normal position eliminating the influence of gravity on the deglutition. With a syringe and a plastic tube a bolus of contrast medium of about 20 mls was injected in the back of the mouth. If necessary an additional bolus of contrast medium was given. The cinematography was continued until a representation of the oesophagus and transplant had been obtained. The film was later projected at variable speed and analyzed.

The analysis consisted of two parts. The first part concerned the morphological description of the structures, or rather the position and diameter of the ileal segment and the proximal and distal oesophagus, and finally the anastomosis. The second part involved the functional description, viz the motor activity, the passage of contrast medium and the plasticity. The plasticity was defined as the ability of a viscus to collapse when empty and to distend with the introduction of material.

2.6.3. Oesophagoscopy.

During the follow-up year 6 oesophagoscopies were planned for each dog and puppy in the course of the 4th, 9th, 13th, 26th, 39th, and 51st week. The oesophagoscopies were performed using a flexible fiberscope OLYMPUS, type EFPA (diameter 6 mms, length 90 cms) with cold light supply Olympus

type CLE 3. The dogs and puppies received a general anaesthesia and were intubated with a cuffed tube. Then the dog was placed on its back upon the table. A stiff oesophagoscope (inner diameter 12 mms, length 25 cms) was placed in the mouth, the end entering the oesophagus. This protected the flexible 'scope that was guided through, against the teeth of the dog and facilitated the introduction into the oesophagus. Before introducing the flexible 'scope, the oesophagus was sucked out with a soft plastic tube to remove the ever present saliva.

During the oesophagoscopy attention was paid to the following points:

introduction of the 'scope;
proximal oesophagus;
oesophagus-bowel anastomosis;
mucous membrane of the transplant;
segmental contraction;
passage of the 'scope;
position of the transplant;
the distal oesophagus.

Finally a biopsy of the mucous membrane of the transplant was taken for histological examination.

2.6.4. Angiography.

In vivo

The vascularization of the transplant was checked after 2 and again after 11½ months by selective angiography of the internal mammary artery. For reasons mentioned in paragraph 4.7. the internal mammary artery was embolized in two cases (puppy no. 10 and 11) after the angiography at 11½ months with Spongostan® (Ferrosan, Denmark).

The dogs and puppies got general anaesthesia and were placed in the right lateral position. The femoral artery was dissected in the groin and an angiographic catheterTorcon® (torque control braided catheter, type Cobra BP5-C2, diameter 1,91 mms) was inserted by puncture technique. The catheter was pushed up under fluoroscopic control and selectively inserted in the right internal mammary artery. As contrast Conray® 60 (Byk Nederland BV, Zwanenberg N.H.) was used. The X-ray picture was made, flushing the contrast through the catheter by hand.

The objective was to visualize the whole vascularity of the transplant; the arterial, capillary and if possible the venous phase.

Postmortem

At autopsy every effort was made to remove the transplant inclusive of the

supplying vessels and the adjacent oesophagus out of the corpse without damage. The arterial side was cannulated and flushed with a barium suspension Micropaque® (Nicholas BV, Amsterdam) at a constant pressure of 140 mmHg. When the contrast was flushed through the whole transplant an X-ray picture was made. As a matter of course the two puppies that had undergone embolization of the internal mammary artery (nos. 10 and 11) were excluded.

2.6.5. Morbid anatomy.

Biopsies

Each animal had 9 biopsies of the ileal transplant taken during the follow-up year. During surgery two full thickness biopsies were taken, one before ischaemia and one after recirculation. Mucosal biopsies were taken during each of the 6 oesophagoscopies. At autopsy again a full thickness biopsy was taken.

Autopsy

All animals were examined postmortally. The long term survivors were sacrificed one year after surgery. They were given barbiturates intravenously and subsequently bled to death. The abdominal and thoracic cavities were opened and inspected. In the puppies the length of the small bowel was also measured. The ileal transplant together with the adjacent proximal and distal oesophagus was dissected from the surrounding tissues. During the dissection attention was paid to the length and the position of the transplant, the diameter of the proximal and distal oesophagus and the vessels of the transplant. The internal mammary artery was cannulated and perfused. The transplant was flushed with a barium suspension to take X-ray pictures (see angiography 2.6.4.). After radiography the transplant was opened lengthwise. The oesophagus-bowel anastomoses and the mucous membranes were examined. A full thickness biopsy was taken.

Histological technique

Immediately after obtaining the biopsies the tissue was fixed in a buffered 10% formalin solution. Paraffin sections were made and stained with hematoxylin, azofloxin and saffron (HAS-staining).

3 RESULTS

3.1. Operative results and mortality.

A. adult dogs

The surgical data and results of the 20 adult dogs are set out in table 1.

OPERATIVE COURSE

Small bowel

The length of the total small bowel varied from 270 to 375 cms with an average length of 325 cms. The length of the dissected ileum averaged 82 cms, from 45 to 125 cms, which amounts to an average of 25% of the entire small bowel.

Vascular anastomosis

The diameter of the mesenteric artery selected for anastomosis was always 3 mms. This was always comparable to the size of the recipient artery. The diameter of the superior vena cava was approximately 20 mms, so side clamping posed no difficulties. No technical problems were encountered in anastomosing the venous side. The arterial anastomosis rendered no difficulties in ten out of 20 cases. In nine cases one extra suture had to be applied after recirculation, whereas in one case (dog no. 13) three extra sutures were needed. Both the arterial and the venous anastomosis could be performed without tension.

Ischaemic time

The ischaemic time varied from 43 to 66 minutes, 54 minutes on average.

Vitality of transplant

In seventeen out of twenty cases the transplant regained a normal colour rapidly after declamping. Vigorous pulsations were observed up to the bowel wall and vivid peristalsis returned after some instances. In the other three

Table 1: The surgical data and results of the 20 adult dogs.

Dog no.	resected small bowel (cms)	(%)	length of interposition (cms)	length of oesophagus resected (cms)	ischaemic time	observation time	complication
1	75	24.2	5	4	56	4 days	vascular thrombosis
2	80	25.8	4	3	43	1 year	
3	85	28.6	4.5	3.5	51	1 year	
4	125	38.4	5	3	58	12 hours	shock
5	80	22.8	4	2.5	53	1 year	transplant resorbed/ stricture
6	90	26.1	4	2	53	4 days	vascular thrombosis
7	80	24.2	4	2.5	48	3 days	vascular thrombosis
8	90	24.0	4	2	50	4 days	vascular thrombosis
9	80	24.2	5	3	66	1 year	
10	90	25.0	4	3	50	1 year	
11	90	29.0	5	3	57	1 year	
12	90	29.0	4	3	65	1 year	
13	90	31.6	5	3	63	5 days	vascular thrombosis
14	100	26.6	5	3	52	1 year	
15	70	22.2	5	3	60	31 days	vascular thrombosis
16	75	27.7	5	3	57	1 year	
17	75	23.8	5	4	58	1 year	
18	45	14.0	5	4	53	1 year	
19	75	20.0	5	4	45	1 year	
20	55	15.9	6	5	44	1 year	
average	82	25.0	4.6	3.1	54		

cases (dog no. 4, 6 and 13) colour was regained slowly, while only minimal pulsations could be detected. But peristalsis did return after several minutes, albeit less pronounced. Incision of the mesentery at one end of the ileal loop revealed blood oozing from the mesenteric capillaries. In all cases extensive oedema arose in the transplant and the bowel became hyperaemic.
All the transplants were interposed even though in three cases the vitality was doubtful.

Oesophagus

The oesophagus was mobilized over a length of 6 to 8 cms. The length of the resected oesophagus segment fluctuated from 2 to 5 cms, with an

average of 3.1 cms, for no definite reason. The resultant gap in the oesophagus was bigger due to retraction of both ends of the oesophagus. As a consequence, the segment of the transplant used to fill the gap was longer than the resected oesophageal segment to avoid any traction on the sutures. The length of transplant interposed antiperistaltically varied from 4 to 6 cms, an average of 4.6 cms; on average 1.5 cms longer than oesophagus segments resected.
After the interposition the ileum was lying slackly between the proximal and distal part of the oesophagus, protruding to some extent into the right thoracic cavity.

Operation time

The surgical procedure took 4¼ - 5¾ hours, an average of 4½ hours.

Bloodloss

The loss of blood during surgery varied from 100 to 350 mls, an average of 190 mls. Bloodtransfusion was only given in dogs nos. 5, 13 and 15 (blood-loss 300, 350 and 250 mls respectively). The other seventeen dogs did not get any bloodtransfusion.

POSTOPERATIVE COURSE

Thorax drainage

The thorax drainage amounted to 0 - 450 mls of haemorrhagic fluid, an average of 140 mls. The drain was removed after 16 - 20 hours in all cases.

Woundhealing

Except for dogs nos. 5 and 17, all wounds healed without complications. Dog no. 5 got a fistula to a suture of the ribs that healed after the suture had been removed. Dog no. 17 developed an abscess in the thoracotomy wound that healed after drainage.

Survival

Of the 20 dogs operated upon 13 dogs survived and came in the follow-up programme.

Mortality

The mortality numbers 7 deaths out of 20 dogs. The deaths were due to vascular thrombosis in five cases, obstruction of venous outflow in one case and hypovolaemic shock in another case.

Dog no. 1 died on the 4th postoperative day. The operation was performed without difficulty. One extra suture was placed on the arterial anastomosis. The bloodloss amounted to 250 mls and the operation took 4½ hours. At autopsy the entire transplant had become necrotic. The arterial and venous anastomosis were thrombosed.

Dog no. 4 died within 20 hours after surgery. During surgery a technical problem was encountered concerning the superior mesenteric artery that described a rather curled course. This caused a faulty ligation of the superior mesenteric artery. A new ileal segment with a new vascular pedicle was chosen and prepared. Consequently a total of 125 cms of small bowel was dissected. After recirculation the transplant recovered slowly to its normal colour and pulsations were weak. The bloodloss was estimated at 250 mls. No bloodtransfusion was given. The operation took 5¾ hours. At autopsy the arterial and venous anastomosis were patent. No haemorrhage was found. It was concluded that death was caused by a hypovolaemic shock.

Dog no. 7 died on the 3rd postoperative day. The operation was uneventful. The drainage of the thorax amounted to 450 mls. This was a very high loss compared to the other dogs. At autopsy the right thoracic cavity was filled with haemorrhagic fluid. The entire transplant had become haemorrhagic and necrotic. The arterial and venous anastomosis was patent. It was concluded that the necrosis of the transplant was caused by venous obstruction.

Dog no. 6 died on the 4th postoperative day. During surgery the transplant regained its normal colour slowly after the vascular clamps were released. Pulsation were not felt during the first 30 minutes. Thereafter pulsations could be felt. Incision of the mesentery revealed blood merely oozing from the vessels of the transplant. Therefore the vitality of the transplant was doubtful. Nevertheless the operation was completed. At autopsy the entire transplant had become necrotic. The arterial side of the vascular pedicle was thrombosed.

Dog no. 8 died on the 4th postoperative day. Until death nothing remarkable had occurred. The operation and the postoperative course were uneventful. At autopsy the transplant was found to be necrotic due to vascular thrombosis.

Dog no. 13 died on the 5th postoperative day. During surgery after recirculation three extra sutures were made on the arterial anastomosis to stop the bleeding. The recirculation proceeded sluggishly, pulsations were felt only after several minutes. At autopsy the arterial and venous anastomosis were thrombosed and the transplant had become necrotic.

Dog no. 15 died on the 31st postoperative day. The operation was uneventful. The bloodloss amounted to 250 mls. During the first postoperative hours a period of hypovolaemic shock had been diagnosed. This was corrected

with a bloodtransfusion of 250 mls. One week after surgery on the cinematogram a narrow canal was seen between the two ends of the oesophagus. Oral intake started normally in the second week, worsened during the 3rd and 4th postoperative week. At oesophagoscopy a short strictured canal of granulation tissue was seen. The transplant could not be seen. Several days later the dog succumbed. At autopsy no traces of the transplant could be found. An empyema was present in the right thoracic cavity.

B. puppies

The surgical data and results of the 11 puppies are set out in table 2.

OPERATIVE COURSE

Small bowel

The length of the total small bowel varied from 240 to 370 cms with an average length of 300 cms. The length of the dissected ileum averaged 55 cms, from 30 to 85 cms, which amounts to an average of 18,5% of the entire small bowel. Measuring and preparing the bowel it was noted that the total length of the small bowel was comparable to that of the adult dogs. However, the caliber of the bowel and its vascularity was proportional to the puppy and thus smaller than adult dogs.

Vascular anastomosis

The diameter of the mesenteric artery selected for anastomosis was 1½ - 2 mms. This was slightly smaller than the diameter of the recipient artery, 2 - 2½ mms. No difficulties were encountered when performing the venous anastomoses except in the case of puppy no. 3, where the stitchholes kept oozing. The oozing stopped using Spongostan® (Ferrosan). The arterial anastomoses were performed without encountering technical problems. In seven cases one extra suture was made after releasing the clamps. Both the arterial and the venous anastomosis was performed without tension.

Ischaemic time

The ischaemic time varied from 45 to 60 minutes, an average of 51 minutes.

Vitality of transplant

After declamping normal colour of the transplant returned rapidly in all eleven cases. Vigorous pulsation and the resumption of peristalsis were noted. Consequently in all cases the vitality of the transplant was considered good and all the transplants were interposed.

Table 2: The surgical data and results of the 11 puppies.

Puppy no.	resected small bowel		length of interposition	length of oesophagus resected	ischaemic time	observation time	complication
	(cms)	(%)	(cms)	(cms)			
1	40	13.1	5	5	50	17 days	vascular thrombosis
2	30	9.5	5.5	5.5	50	1 year	
3	30	10.7	5	5	50	18 days	vascular thrombosis
4	45	16.0	6	6	59	12 hours	shock
5	45	18.7	5	5	60	1 year	
6	55	20.0	5	5	54	5 days	vascular thrombosis
7	50	20.0	5	5	50	1 year	
8	75	20.8	6	6	47	1 year	
9	75	20.2	6	6	45	19 weeks	complete functional obstruction
10	80	23.5	6	6	48	1 year	
11	85	29.8	6	6	52	1 year	
average	55	18.5	5.5	5.5	51		

Oesophagus

The oesophagus was mobilized for a length of 7 to 8 cms. With a view to standarization, approximately the same length of oesophagus was resected in each case, 5 to 6 cms. The same length of ileum segment was interposed isoperistaltically, 5 to 6 cms, causing some tension on the suturelines. But in this way the ileum segment was lying beautifully straight between both ends of the oesophagus and properly in the oesophagusbed.

Operation time

The surgical procedure took 4 - 4½ hours. By the time the second series was started the operation had become a standard procedure which explains the constancy of the duration.

Bloodloss

The bloodloss during the operation varied from 100 to 250 mls, 130 mls on average. This bloodloss was replaced by a bloodtransfusion in all cases.

POSTOPERATIVE COURSE

Thorax drainage

The thorax drainage amounted to 95 - 225 mls of haemorrhagic fluid, an average of 110 mls. The drain was removed 16 hours after surgery in all cases.

Woundhealing

In the puppies woundhealing proceeded smoothly.

Survival

Of the 11 puppies operated upon 7 puppies survived and entered the follow-up programme.

Mortality

The mortality numbers 4 deaths out of 11 puppies. The deaths were due to delayed vascular thrombosis in three cases and hypovolaemic shock in one case.

Puppy no. 1 died 17 days after surgery. The operation was uneventful. One week after surgery the puppy had lost weight (from 9.8 down to 8.5 kgs) and had become apathetic. The swallow cinematogram at one week revealed a narrow canal. There was passage of contrast and no leakage was seen. Four days later the puppy started vomiting, no feeds could pass anymore. The puppy deteriorated and was sacrificed. At autopsy the entire transplant had become necrotic due to vascular thrombosis.

Puppy no. 3 died 18 days after surgery. In this case the venous anastomosis kept oozing during surgery. Oozing was stopped using Spongostan® (Ferrosan). In the first postoperative week the puppy lost one kg in bodyweight and became apathetic. Subsequently the course was identical to the course of puppy no. 1. At autopsy the transplant was found to be necrotic due to vascular thrombosis.

Puppy no. 4 died within 12 hours after surgery. The operation was uneventful. The estimated bloodloss was replaced. During the first post-operative hours the puppy suffered a hypovolaemic shock that was not treated and resulted in death. At autopsy no reason could be found for the shock. The transplant and its vascular pedicle were intact.

Puppy no. 6 died on the 5th postoperative day. The operation and the post-operative course were uneventful. The puppy died unexpectedly. The autopsy revealed necrosis of the entire transplant due to vascular thrombosis.

3.2. Follow-up studies.

3.2.1. Nutrition and bodyweight.

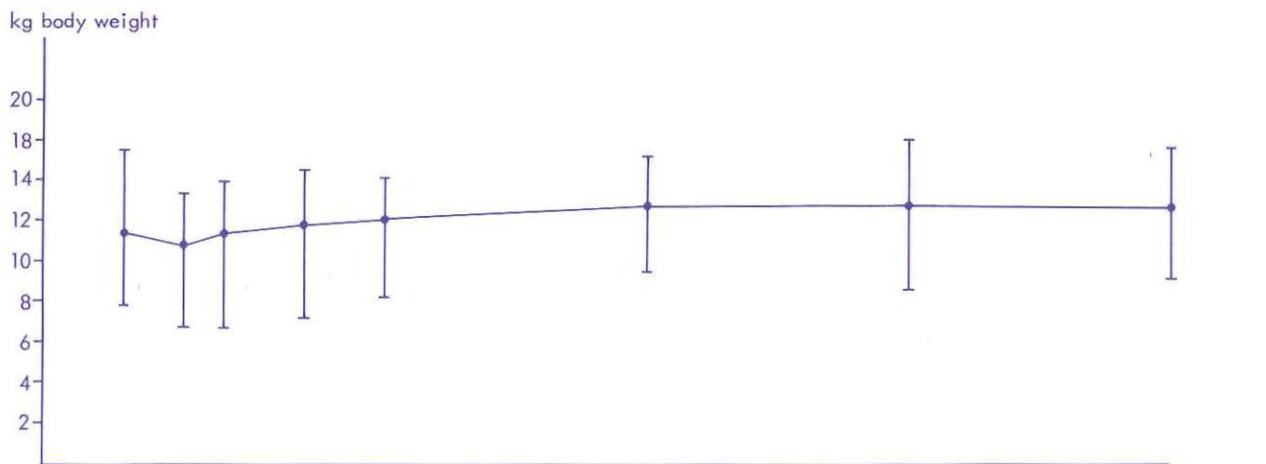
A. adult dogs

Of the 13 surviving dogs, one dog (no. 5) had a complicated course. This dog is described in the paragraph complications (3.2.2.). The other surviving dogs did well. All lost some weight during the first week. After one week they were started on water and subsequently on semi-solids. As a result their general condition improved rapidly. Solid feeds were added 5-6 weeks after surgery even though all of them vomited almost daily. The feeds were regurgitated while feeding. This was due to their habit of eating greedily and swallowing large chunks of food that could not pass that quickly through the transplant. This sometimes resulted in eating the same food twice. During the follow-up year five dogs ceased to vomit and were eventually put on an entirely solid diet. The other seven dogs continued to vomit although less frequently (about twice a week) and were kept on a combination of semi-solids and solids. Following this regime the dogs regained their pre-operative weight about one month after surgery. And during the remainder of the observation time they gained weight slightly (see fig. 7). Until they were sacrificed after one year these twelve dogs remained healthy.

B. puppies

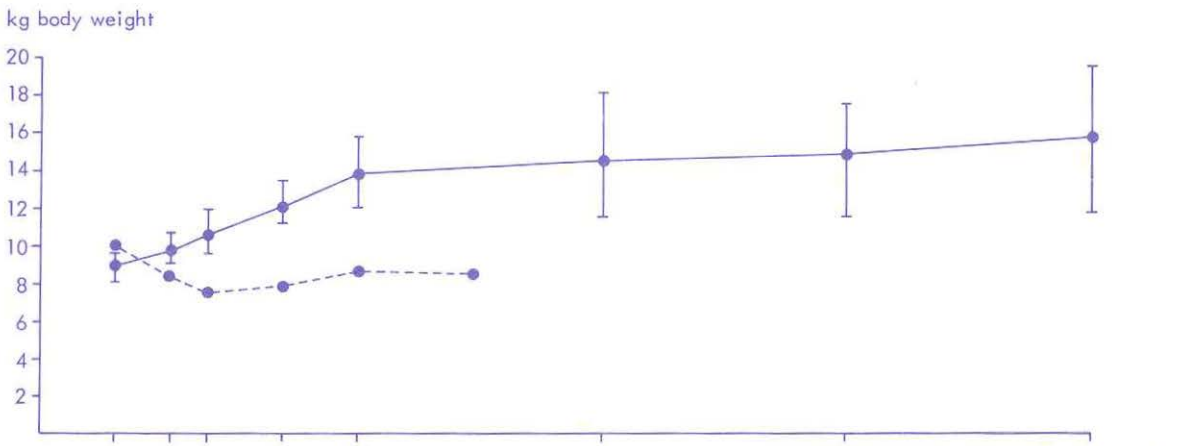
There were seven surviving puppies. Two of these puppies (nos. 2 and 5) belonged to the first part of this series, consisting of six puppies that were given fluids through hypodermoclysis only during the first postoperative week. It was noted that these puppies became apathetic and lost more than 1 kg of their bodyweight during this week. Four of these six puppies died. Therefore it was decided that the last five puppies of this series were allowed to drink water from the first postoperative day. None of these five puppies lost more than ½ kg bodyweight and they remained lively throughout the first week. All of these five puppies survived.
One week after surgery all seven puppies were started on liquids and a few days later on semi-solids. After three months five of these seven puppies were given solids in addition to the semi-solids even though they vomited regularly. The vomiting decreased in frequency during the observation period but never ceased completely. The other two puppies had dietary problems right from the start. They vomited more than the other puppies. One of them (puppy no. 9) was sacrificed eventually after 19 weeks (see complications 3.2.2.). The other one never managed a solid diet and was kept on a semi-solid diet vomiting regularly.

Figure 7: Combined weight curve of the 13 surviving dogs, inclusive of dog no. 5 with the oesophageal stricture.



	+4,0	+2,6	+2,6	+2,7	+2,0	+2,4	+3,3	+2,9
average	11,5	10,8	11,4	11,8	12,1	12,7	12,7	12,6
weight	-3,6	-4,0	-4,6	-4,6	-3,9	-3,2	-4,1	-3,5

Figure 8: Growthgraph of seven puppies. Combined curve of six puppies. Puppy no. 9 never reached his pre-operative weight and was sacrificed after 19 weeks.



	+0,7	+0,9	+1,3	+1,4	+2,0	+3,7	+2,8	+3,8
average weight	8,9	9,7	10,6	12,1	13,8	14,5	14,8	15,7
	-0,9	-0,7	-1,0	-0,9	-1,8	-3,0	-3,3	-4,0
weight gain %		9%	18,9%	35,5%	54,4%	62,2%	65,5%	75,5%

The six puppies that were observed during the entire follow-up period gained weight rapidly during the first three months amounting to 54.4%, from 8.9 kgs to 13.8 kgs. For the remaining period they showed a slight weight gain, resulting in an entire weight gain of 75,5% after one year (see fig. 8). All six puppies grew into well-nourished adult dogs.

3.2.2. Complications

Dog no. 5. The operation was uneventful although the loss of blood was estimated at 300 mls. This loss was replaced by fluids and plasma. In the immediate postoparative period the dog was found to be in deep shock. This shock was treated sucessfully with blood and fluids. In the first postoperative week he suffered the same weightloss as the other dogs. His condition did not differ from that of the other dogs. But in the swallow cinematogram after one week leakage of contrast was seen and a cavity filled with contrast at the place where the transplant should have been. So he was not put on oral intake but he received intravenous hyperalimentation. A second cinematogram after two weeks revealed that the leakage of contrast amounted to only a small deposit of contrast. Two days later an oesophagoscopy was performed. No signs of the transplant were seen. Only granulation tissue and a small cavity were seen. The oesophagoscope could not pass on to the distal oesophagus. The dog was put on water and semi-solids with success. However he started to vomit with an ever increasing frequency. Four weeks after surgery he could no longer take any foods orally. A tight stricture had developed and was dilated with catheters. During the consecutive two months this stricture was dilated another three times. Thereafter the stricture did not tighten anymore, and kept a diameter of 6mms and a length of about 1 cm. During the last 9 month of the follow-up period his condition did not differ from that of the other adult dogs, as far as diet and frequency of vomiting was concerned. He was kept on a combination of semi-solids and solids, vomiting about twice a week. Eventually he gained bodyweight from 10.8 kgs at operation to 13.5 kgs when he was sacrificed.

Puppy no. 9 gave dietary problems right from the start just like puppy no. 11. They vomited more than the other puppies, but with puppy no. 9 it got worse. Three months after surgery puppy no. 9 could not eat anything. All food and liquids were regurgitated and he became cachectic. The proximal oesophagus was enormously dilated and the motor activity of the proximal oesophagus was totally absent but the transplant showed no abnormality during oesophagoscopy (see 3.2.4.). No stricture was to be seen. A rethoracotomy was performed. The transplant with its supplying vessels was completely intact. The proximal oesophagus was enormously dilated and was folded over the proximal oesophagus-bowel anastomosis. This fold was dissected from the transplant. Then a Y-V plasty was performed in the

proximal oesophagus-bowel anastomosis to enlarge the entrance and the dilated oesophagus was plicated lengthwise. He recovered from this rethoracotomy without complications. However, functionally the operation did not have any success. The desired improvement did not show up. Eventually after 19 weeks it was decided to sacrifice the puppy. This puppy never regained his pre-operative weight (see fig. 8).

3.2.3. Cinematography

In this paragraph the deglutition of the animals operated upon is described on the basis of 157 X-ray examinations. These findings concern the 12 adult dogs and the 7 puppies with an intact transplant. Adult dog no. 5 that had a severe stricture after total necrosis and resorption of the transplant is described separately. The results are set out in table 3.

Morphology

The configuration of the oesophagus-bowel anastomosis, proximal and distal, showed an identically peculiar picture in all animals. The anastomotic side of the ileal segment always protruded into the lumen. Directly distal and proximal to the transplant the oesophagus itself never demonstrated its usual propensity for collapse. A stricture of the oesophagus-bowel anastomosis was never observed. The picture of the rest of the ileal segment remained consistent. The inside diameter was never found to be reduced or dilated and mucosal folds were present in varying numbers. The position of the transplant remained the same from the first cinematography. For the adult dogs this meant a more or less curved position in 9 cases and in 3 cases a straight position (fig. 9) while in the puppy group a straight position was seen 6 times and a curved one only once (fig. 10). The diameter of the proximal oesophagus was always slightly enlarged in the adult dog group and once markedly enlarged. This dilatation occurred within the first week and did not increase any further. In the puppy group the dilatation of the proximal oesophagus increased even more and by the 5th or 9th week it had become marked or enormous. As a result the ileal "wall" seemed to protrude even more into the dilated lumen of the proximal oesophagus. Dilatation of the distal oesophagus was never found.

Functional analysis

Motor activity of the ileal segment could not be detected on the films without reasonable doubt. This was due to several factors. The length of the ileal transplant was short. There was always a swift passage of contrast through the transplant and retention of contrast within the transplant never occurred whereas the normal motor activity of the small bowel is rather slow.

Table 3: Morphological and functional findings on the cinematograms. The degree of motor dysfunction of the proximal oesophagus is defined in the text (page 41).

Dog no.:	2	3	9	10	11	12	14	16	17	18	19	20	5
Morphology													
- curve in transplant	+	+	+	+	++	++	++	++	++	0	0	0	
- dilatation of proximal oesophagus	+	+	+	+	+	+	+	+	++	+	+	+	+
Functional analysis													
- motor dysfunction prox. oesophagus	1	2	1	1	1	1	2	2	2	1	1	1	2
- yo-yo phenomenon	++	+++	++	++	++	++	+++	+++	+++	++	++	++	+++
- delay of passage	+	++	+	+	+	+	++	++	++	+	+	+	++

Puppy no.:	2	5	7	8	9	10	11
Morphology							
- curve in transplant	0	0	0	+	0	0	0
- dilatation of proximal oesophagus	++	++	++	++	+++	+	+++
Functional analysis							
- motor dysfunction prox. oesophagus	3	2	2	3	4	2	3
- yo-yo phenomenon	++	+++	+++	++	0	+++	++
- delay of passage	+++	+++	++	+++	++++	++	+++

0 : absent
\+ : slight
++ : marked
+++ : severe/enormous
++++ : blocked passage

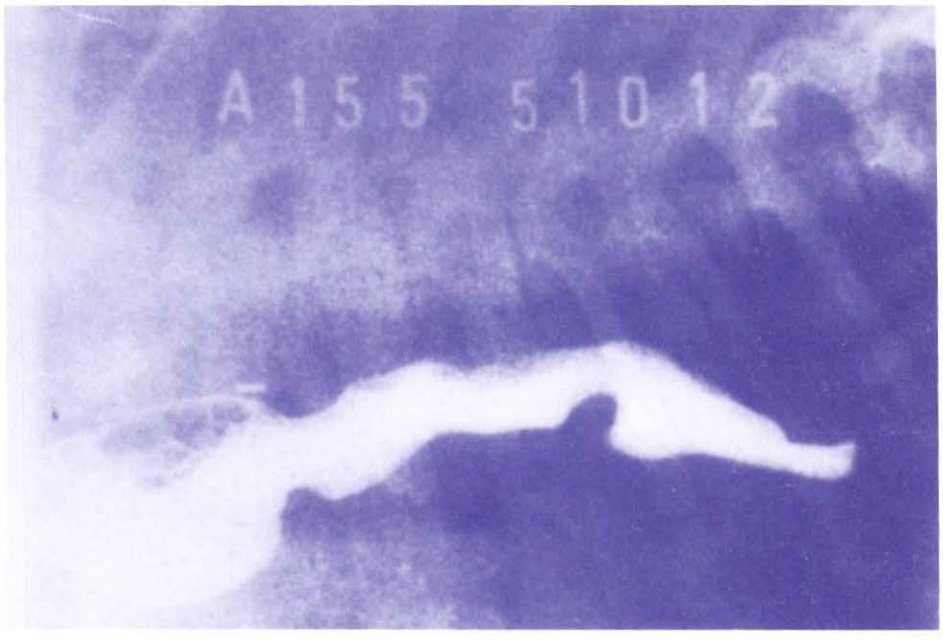


Figure 9: The configuration of the oesophageal-bowel anastomosis, showed an identically peculiar picture in all animals. Straight position of the transplant in the case of adult dog. no. 12.

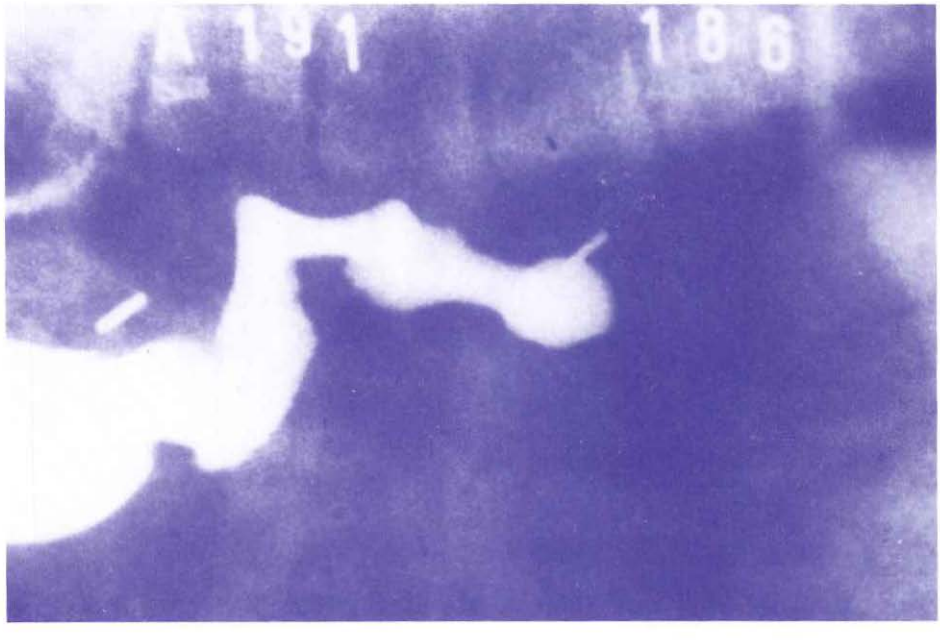


Figure 10: Curved position of the transplant in the case of puppy no. 8.

The plasticity of the transplant remained normal for small bowel. With a forceful contraction of the proximal oesophagus the ileal segment always resumed its maximum diameter and collapsed after emptying.

The motor activity of the proximal oesophagus was disturbed in all animals. The extent of this motor dysfunction was different in each dog. It was noted that the degree of severity remained more or less unchanged during the 8 consecutive examinations.

This motor dysfunction was classed in four degrees of severity. The first degree concerned those cases that alternated incomplete peristaltic contraction, stopping several centimeters before the oesophagus-bowel anastomosis, with complete peristaltic contractions, proceeding until the oesophagus-bowel anastomosis. The second degree concerned dogs that showed frequent incomplete peristaltic contractions only. The dogs with a third degree motor dysfunction were characterized by sporadic incomplete peristaltic contractions. In case motor activity could not be registered at all a fourth degree of motor dysfunction became applicable.

An animal with a first degree motor dysfunction had reached that stage by the first cinematography. For the animals with a more serious degree this took 5 weeks to develop. Subsequently the degree of motor dysfunction remained the same throughout the observation period, except for puppy no. 9 that showed a fourth degree motor dysfunction after 13 weeks (total lack of motor activity). In general the motor dysfunction in the puppy group was more serious than in the adult dog group (see table 3).

The incomplete and bad propulsive contractions of the proximal oesophagus caused a retention of contrast in the dilated proximal oesophagus that rocked back and forth. This hitching of contrast was called the yo-yo phenomenon. The yo-yo phenomenon was always closely related to the degree of motor dysfunction, the amount of contrast present and the degree of dilatation. The plasticity of the proximal oesophagus had diminished. The proximal oesophagus never collapsed completely but remained open. In each case the distal oesophagus showed peristaltic waves of a normal character, starting when filled with contrast from the distal oesophagus-bowel anastomosis and proceeding to the stomach. A clear relationship between peristalsis proximal and distal to the transplant was not observed. The plasticity remained that of a normal oesophagus and dilatation could not be noted.

The passage of contrast from the mouth to the stomach was always delayed in comparison with normal deglutition. The bolus of contrast moved forward, yo-yo-ed before the oesophagus-bowel anastomosis, passed swiftly through the transplant after a good contraction of the proximal oesophagus and moved to the stomach propelled by a normal peristaltic wave. If there was retention of contrast this was always in the dilated proximal oesophagus. The total passage time therefore depended closely on

the function of the proximal oesophagus and was either slightly, markedly or seriously delayed. In some cases a bolus of solid food had remained in the proximal oesophagus making it even more difficult for the contrast to pass through.

In the case of puppy no. 9 the total lack of motor activity of the proximal oesophagus after 13 weeks resulted in stagnation of contrast fluids in the enormously dilated proximal oesophagus. Only passive passage could be achieved in this case by holding the puppy upright thus permitting gravity to exert its force onto the contrast (fig. 11). This puppy was operated upon once again and sacrificed after 19 weeks (see complications 3.2.2.)

In dog no. 5 total necrosis and resorption of the transplant occurred and a severe oesophageal stricture developed. Extravasation of contrast was seen on cinematography after the first week. The place where the transplant should have been showed a cavity filled with contrast. After some time contrast leaked into the stomach. Motor activity was not observed. On subsequent films a short tight stricture had developed. The proximal oesophagus became slightly dilated and second degree motor dysfunction was noted combined with a serious yo-yo phenomenon. The passage of contrast became markedly delayed. From the 9th week on the picture remained stable up to sacrifice after one year.

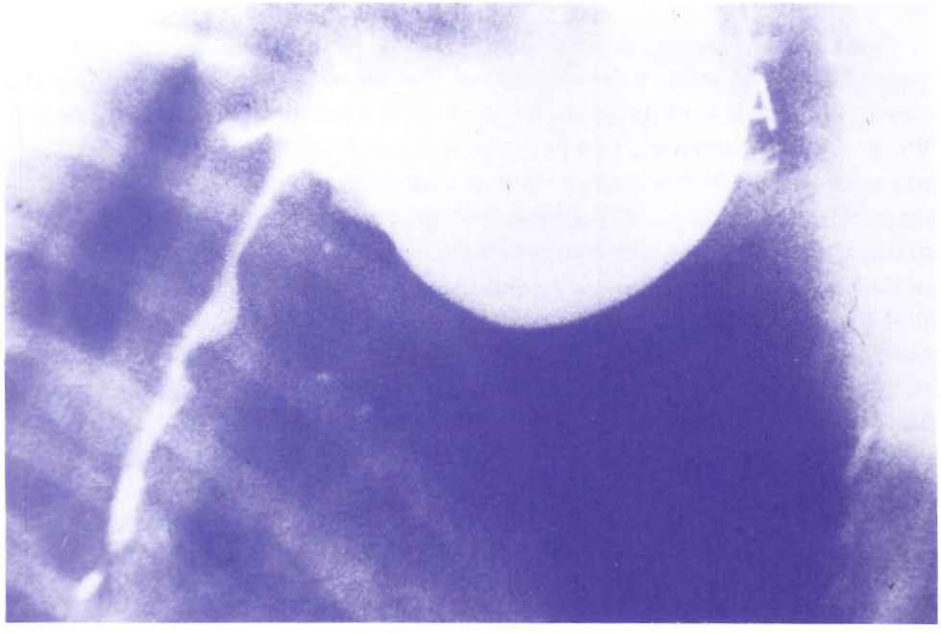


Figure 11: Enormous dilatation of the proximal oesophagus of puppy no. 9. Passive passage of contrast medium by holding the puppy upright.

3.2.4. Oesophagoscopy

To inspect the inside of the transplant interposed in the oesophageal defect and to obtain biopsies of the mucosal layer 107 oesophagoscopies were performed during the follow-up period. For technical reasons 4 oesophagoscopies had to be cancelled. For oesophagoscopies performed on dog no. 5 see complications (3.2.2.).

A. adult dogs

Introduction of the 'scope

In each instance the introduction of the 'scope into the oesophagus progressed smoothly. The transplant was always immediately visible and looked like a mucous stoma protruding into the lumen of the oesophagus. This was at a distance of 40 to 50 cms from the anterior teeth.

Proximal oesophagus

No macroscopical changes of the proximal oesophageal wall were observed. The lumen, just before the transplant was always rather wide and slightly dilated.

Mucous membrane of the transplant

The mucous membrane of the ileum was always smooth, glossy and pink coloured. Damage of the epithelium was never found. Only during the first oesophagoscopy after 4 weeks the mucous membrane bled easily when touched. Afterwards this was never noted again.

Oesophagus-bowel anastomosis

The proximal oesophagus-bowel anastomosis was always clearly visible. There was a sharp demarcation line between the white oesophageal epithelium and the pink mucous membrane of the ileum. The anastomotic line seemed to be stretched out around the mucous stoma of the ileum. This anastomotic line was never shortened or strictured due to fibrosis. In 4 instances a silk suture was seen to protrude through the bowel wall at the anastomotic side. The distal oesophagus-bowel anastomosis could not be visualized due to optic limitations and the anatomical position but no problems ever occurred in connection with this distal anastomosis.

Segmental contractions

At each oesophaguscopy slow or brisk autonomous segmental contractions of the ileal segment could be observed. Whether these

contractions were propulsive in character and thus peristaltic could not be determined. These contractions were often activated by touching the transplant with the 'scope.

Passage of the 'scope (see table 4)

An attempt was always made to pass the transplant with the 'scope. In 8 cases this proceeded smoothly because the lumen of the transplant was entirely patent, enabling an uninterrupted view of the distal oesophagus. In all other cases it was necessary to push and separate the wall of the ileum by inflation to get into the transplant. And it was rather difficult to open the ileal segment. After that it was almost always possible to pass through the whole transplant. In 4 dogs passage was blocked although in 3 dogs only temporarily. This blockage was always due to a curved position of the transplant which prevented the 'scope to pass through. When the 'scope was withdrawn from the transplant the ileum immediately collapsed making a renewed passage equally difficult.
The overall impression of the successive oesophagoscopies was that in all dogs the passage was improving continuously.

Table 4: Passage of the oesophagoscope through the transplant during each oesophagoscopy.

Weeks:	4	9	13	26	39	51
Dog no.						
2	++	++	++	++	—	++
3	+	++	++	++	—	++
9	++	++	++	++	++	++
10	0	0	0	+	++	++
11	0	0	0	++	++	++
12	0	0	0	++	++	+++
14	+++	++	++	++	++	+++
16	0	0	0	0	0	0
17	++	++	++	++	++	++
18	++	++	++	++	++	+++
19	+++	++	++	—	++	+++
20	++	++	++	—	+++	+++
Puppy no.						
2	++	++	++	++	++	++
5	++	++	++	+++	++	+++
7	++	++	+++	++	+++	+++
8	++	+++	++	+++	+++	+++
9	++	++	++			
10	++	+++	+++	+++	+++	+++
11	+++	++	+++	+++	+++	+++

+++ : easy passage with 'scope, lumen patent
++ : passage possible with'scope, separating the wall
\+ : passage possible only after several attempts
0 : no passage with 'scope

Position of the transplant

The transplant did not form a straight line with the oesophagus. To enter the transplant it was always necessary to bend the 'scope to the right side of the dog and to pass through the transplant the 'scope had to be bent to the other side during the passage. In four dogs this manoeuvre could not always be completed during oesophagoscopy due to the curvature of the transplant.

Distal oesophagus

The distal oesophagus never presented pathology.

B. puppies

Introduction of the 'scope

The introduction of the 'scope was carried out in the same way as in the adult dogs. During the follow-up the distance of the transplant to the anterior teeth increased from 40 to 60 cms.

Proximal oesophagus

In the puppies the lumen of the proximal oesophagus was also dilated to some extent and in two cases (puppy no. 9 and no. 11) this dilatation became extreme as noted in the subsequent oesophagoscopies. A large sac was formed containing rests of saliva.

Passage ot the 'scope (see table 4)

More often than not the lumen of the transplant was entirely patent. Therefore the 'scope could always pass quite easily through the transplant to the distal oesophagus. It must be stressed that this also applied to puppies no. 9 and no. 11 that vomited frequently, needed special dietary care and showed such a divergent picture on cinematography.

Position of the transplant

The transplant always formed a straight line with the oesophagus without any angle or curve.
Concerning the mucous membrane of the transplant, the oesophagus-bowel anastomosis, the segmental contractions and the distal oesophagus, the same findings were noted as for the adult dogs.

3.2.5. Angiography

In vivo

Altogether 37 angiographies were made in the adult dog group and the puppy group combined, 19 early angiographies 2 months after surgery and 18 late angiographies 11½ months after surgery (puppy no. 9 was sacrificed after 19 weeks). In 33 cases a picture of the transplant was obtained, 31 times by selective cannulation of the internal mammary artery and 2 times by a general angiography of the right subclavian artery. In 4 cases the angiography failed to give a picture of the transplant. This concerned the early and the late angiography of dog no. 2 and the early angiographies of dogs nos. 16 and 17. The X-ray picture obtained at the successful angiographies did not differ essentially from each other and likewise the early and late angiography produced identical X-rays. In all 33 cases the arterial and capillary phase and in 31 cases also the venous phase was represented. The arterial anastomoses were always visible as a short narrowed spot in the artery. The course of the mesenterical artery was curled. In the capillary phase the whole transplant became visible (see fig. 12). In the venous phase a large mesenteric vein was seen alongside the mesenteric artery and in most cases a discharge of contrast to the superior vena cava

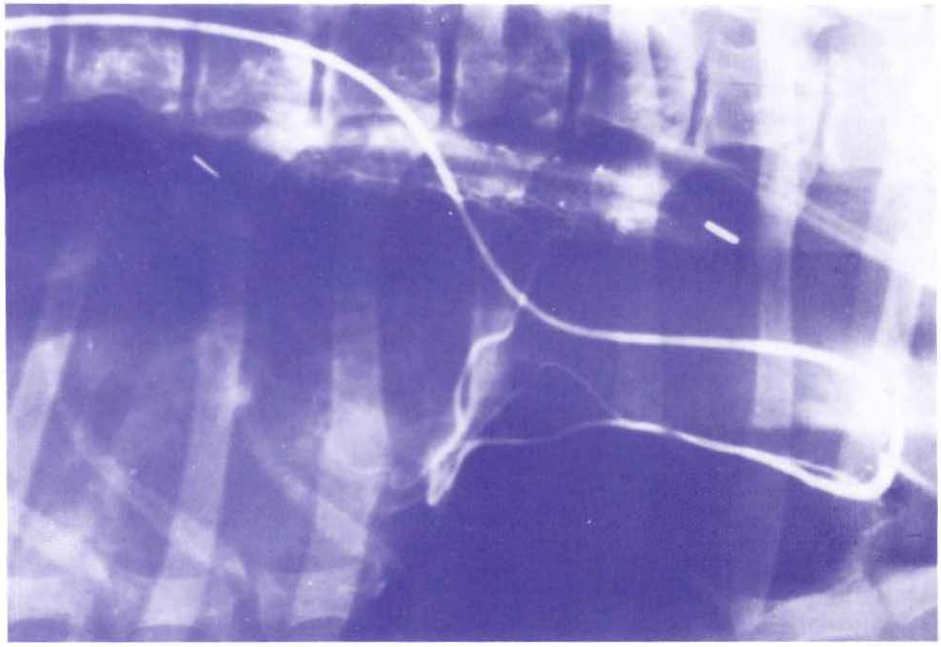


Figure 12: In vivo angiography, showing the arterial and capillary phase.

could also be seen. In the case of puppies nos. 10 and 11 the internal mammary artery had been embolized, after the patency of the arterial anastomoses had been confirmed. Subsequently the complete embolization was confirmed.

Postmortem

At autopsy 17 transplants were considered for postmortem angiography. In 14 cases the postmortem angiography was successful and in 3 case it failed due to damaged mesenteric vessels during the dissection (dog no. 2 and 14, and puppy no. 5).

The transplants that were examined in this way showed a consistent picture. The arteries, the capillaries and sometimes the vein of the transplant all filled with contrast. In all cases filling of the vessels of the adjacent oesophagus was seen due to an extensive transanastomotic collateral circulation at the oesophagus-intestine transition (see fig. 13). At histological examination of the biopsies the contrast was found within the small vessels in the villi.

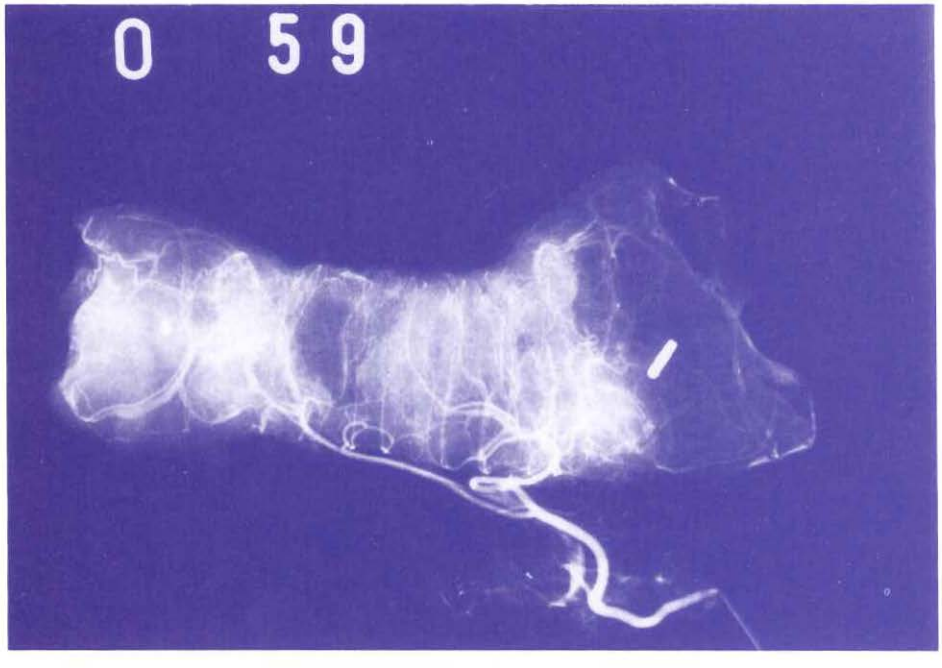


Figure 13: Postmortum angiography showing an extensive transanastomotic collateral circulation at the oesophagus-intestine transition.

3.2.6. Morbid anatomy

HISTOLOGICAL FINDINGS

In total 190 biopsies of the ileal transplants were obtained and reviewed. The findings on all biopsies taken during oesophagoscopy are reviewed at one and the same time. The reason being threefold. Firstly, the quality of the biopsies was often poor. Secondly, no essential differences could be found in the biopsies taken at different intervals. And thirdly, reviewing the biopsies chronologically could easily give occasion to unsubstantiated interpretations and conclusions.

Small bowel before ischaemia

All biopsies before ischaemia revealed a normal crypt-villus ratio of ± 1 : 3. The villi were finger-shaped and had contracted. Numerous mitoses were present in the crypts. Goblet cells were dispersed over the mucous membrane in a normal pattern. A normal amount of mononuclear infiltration and lymph nodules were seen in the lamina propria. The muscularis mucosae and the tunica muscularis were normal (see fig. 14)

Small bowel after recirculation

In all cases oedema had developed in the short interval of ischaemia. This oedema was located mainly in a strip of the lamina propria right under the villi. The amount of mononuclear cells was slightly increased. The villi were somewhat broadened and the lymph vessels were dilated. The same amount of mitoses were present in the crypt before and after ischaemia. Abnormalities of the muscularis mucosae, the tunica muscularis and the bloodvessels were not observed (see fig. 15).

Biopsies taken during oesophagoscopy

The crypt-villus ratio could not be determined from the material. In all biopsies an increase of goblet cells in the mucosal layer and of collagenous connective tissue in the lamina propria was seen in comparison with the normal small bowel. Infiltration of mononuclear cells seemed less than in the normal small bowel (see fig. 16)

Biopsies taken at autopsy

In all animals, dogs and puppies (including puppy no. 9), the biopsies presented the same picture. The crypt-villus ratio was 1 : 1½ or 1 : 2, whereas the mucous membrane was covered by cuboid to flattened epithelium. Damage or ulceration of the mucous membrane was never found. The

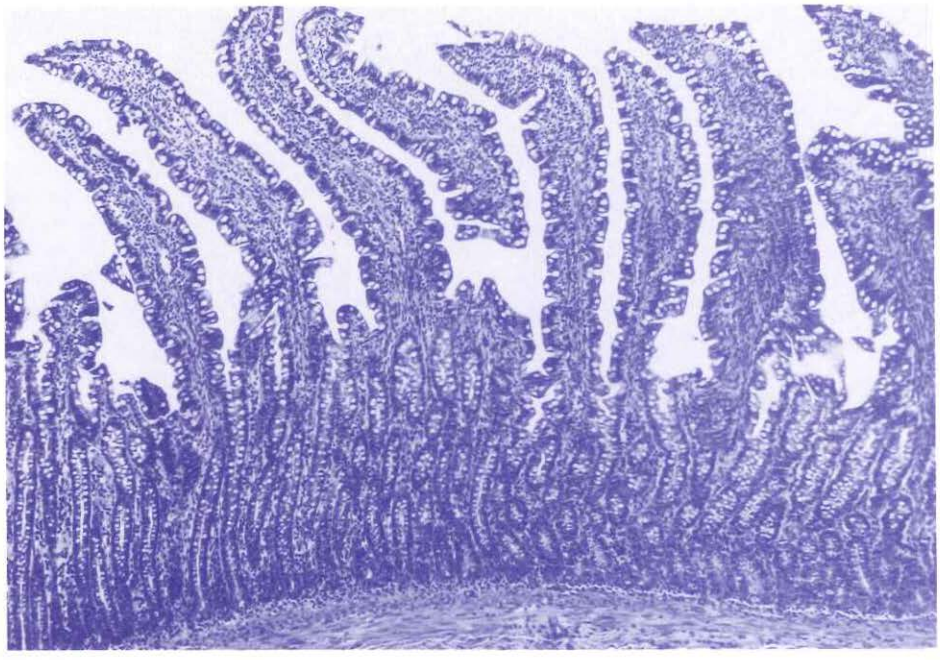


Figure 14: Normal histology of the small bowel before ischaemia.

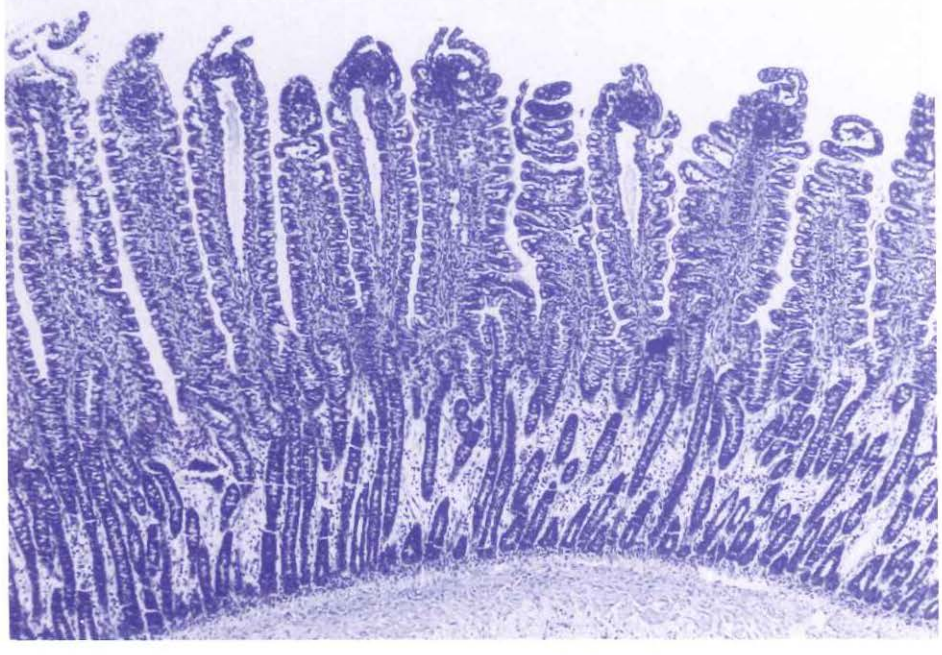


Figure 15: Oedema of the lamina propria of the small bowel after ischaemia.

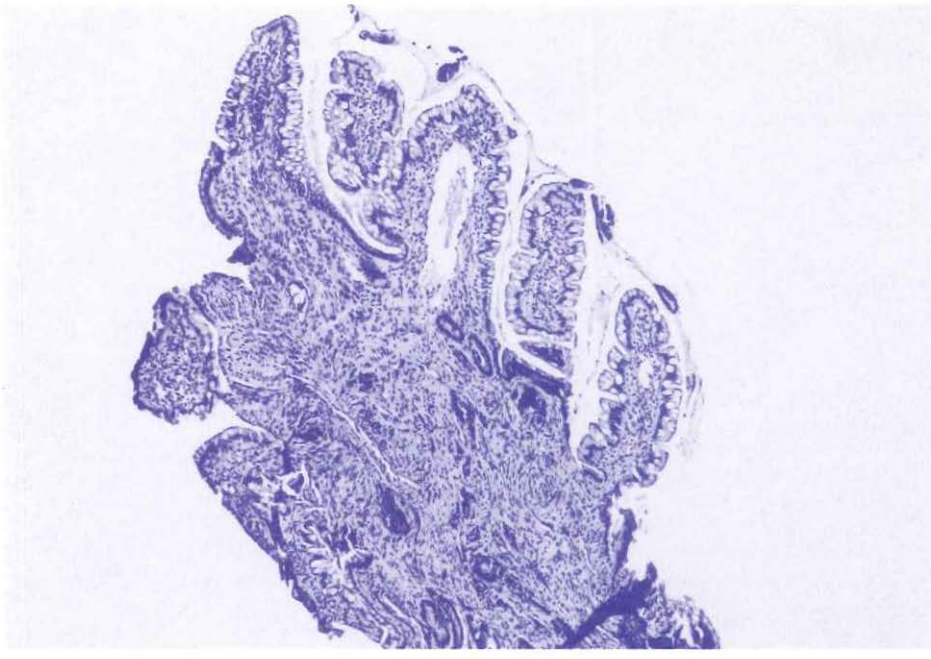


Figure 16: Biopsy taken during oesophagoscopy.

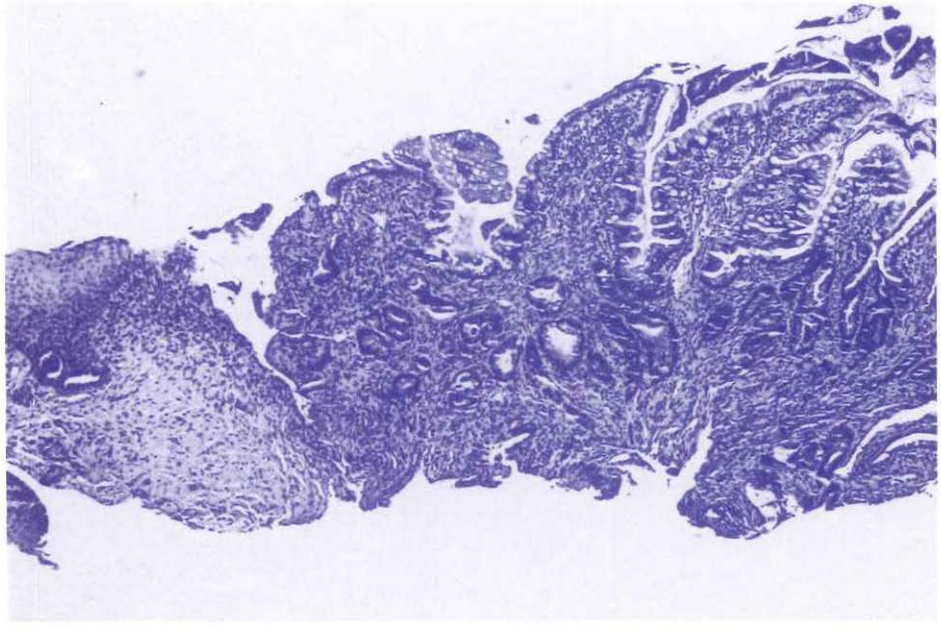


Figure 17: Biopsy take at autopsy. Fibrosis through the whole lamina propria; sharp transition line between the oesophageal and small bowel epithelium.

amount of mitoses in the crypts and the number of goblet cells had obviously increased. Microscopically a sharp transition line was also noted between the oesophageal epithelium and the small bowel epithelium. Fibrosis was seen through the whole lamina propria. This fibrosis was most severe in the strip of the lamina propria where oedema occurred after the period of ischaemia. Infiltration of mononuclear cells in the lamina propria had clearly decreased and was totally absent in some cases. The lymph vessels remained somewhat wider than in the normal bowel. The muscularis mucosae, the tunica muscularis and the bloodvessels of the transplant demonstrated no divergence from the normal small bowel (see fig. 17).

AUTOPSY

The findings in the animals that succumbed due to the operation as such are already described in paragraph 3.1. Thirteen adult dogs, inclusive of dog no. 5 (with total resorption of the transplant), and six puppies were sacrificed one year after surgery. Puppy no. 9 had to be sacrificed 19 weeks after surgery due to dietary problems. This puppy and dog no. 5 are described separately at the end of this paragraph.

In the abdomen some membranous adhesions were found in all animals. The end-to-end ileal anastomoses had healed nicely without stricture formation. The length of small bowel of the adult dogs was not measured. The total length of the small bowel of the puppies varied from 200 to 300 cms and compared well with the original length of small bowel left in the abdomen during the operation.

In the right thoracic cavity some membranous adhesions were also found. The right lung could always be dissected easily from the transplant. In 5 out of 12 adult dogs with an intact transplant, practically the entire transplant was found outside the mediastinum. The transplant, forming a curvature in the thoracic cavity, was lying against the ribs. In 4 cases the transplant formed only a small curvature, protruding slightly into the thoracic cavity and in 3 cases the transplant was situated the mediastinum. In all 6 puppies the transplant was found to form a straight line with the oesophagus in the mediastinum.

Macroscopically the transplant of the two puppies (nos. 10 and 11) that underwent embolization of the internal mammary artery did not differ from the other transplants. No signs of necrosis were seen.

In the adult dogs as well as in the puppies the length of the transplant at autopsy was exactly the same as the length of transplant interposed during surgery. Fibrous tissue outside the transplant was hardly ever seen. The proximal oesophagus was always slightly dilated in the adult dogs. In the puppies the dilatation of the proximal oesophagus was marked in five cases

and enormous in one case (no. 11). The distal oesophagus was always normal, not dilated.

In all animals the transplant together with the adjacent oesophagus was dissected in one piece. In 5 cases the internal mammary artery was not cannulated and the transplant was not flushed with barium suspension for radiography, in 3 cases (dogs nos. 2 and 14 and puppy no. 5) due to damaged mesenterial vessels during dissection and in 2 cases (puppies nos. 10 and 11) due to the embolization of the internal mammary artery. At autopsy the internal mammary artery of these two cases had completely thrombosed. In all other 14 cases, 10 adult dogs and 4 puppies, the transplant was flushed with barium suspension and X-ray pictures were made (see angiography). Inspection of the superior caval vein from the inside revealed a patent venous anastomosis in all 19 animals that were sacrificed after one year.

Finally each transplant was opened lengthwise. Strictures due to fibrous tissue at the anastomotic sites were never seen. Macroscopically the mucosal layer of the transplant and the oesophagus was normal without defects or ulcerations.

Dog no. 5 showed a short (about 1 cm), stiff and narrow (mms.) stenosis of fibrous tissue in the oesophagus. The oesophagus proximal to the stenosis was slightly dilated whereas the oesophagus distal to the stenosis was normal. No remnants of the transplant were found.

Puppy no. 9 showed a similar picture as puppy no. 11. An enormous dilatation of the proximal oesophagus was present. The transplant was found to be straight in line with the oesophagus and had retained the same length that was previously interposed. The vessels were flushed with barium suspension and an X-ray picture was taken. In the proximal oesophagus-bowel anastomosis of puppy no. 9 the Y-V plasty, made during rethoracotomy, could clearly be recognized. There was no stenosis at the anastomotic sites. The mucous membranes of the transplant and the oesophagus were intact.

4 DISCUSSION AND CONCLUSION

4.1. The intestinal segment.

In our experiment we copied the technique of harvesting a segment of distal ileum as developed by Pull ter Gunne (1975) in our laboratories. The advantage of this technique is that one can simply dissect the superior mesenteric artery closer to its origin to obtain an artery with a bigger diamater should this be necessary for the vascular anastomosis, without harm to the rest of the small bowel. A great disadvantage connected with this technique is caused by the absence of good vascular arcades along the small bowel in dogs. Extra length of small bowel have to be resected while only a short segment is transplanted. Pull ter Gunne resected 30-50% of the total small bowel length, 38.5% on average. Refinement of microvascular surgical techniques enabled us to reduce this percentage in our series to 25% and 18,5% in the adult dog group and puppy group respectively. The diameter of the vessels anastomosed in our experiments were, however, of the same size as one of the first 3 or 4 jejunal arteries.

All other investigators in this field of experimental surgery, Seidenburg (1959), Peters et al. (1970), Green and Som (1966), Ancona and Frasson (1978), Germain et al. (1979), used a segment of jejunum supplied by a single jejunal artery instead of an ileal segment. The length of small bowel resected using this technique was even less than in our experiments, although they used dogs at least twice as heavy as our adult dogs.

Our choice of a small bowel segment as oesophageal substitute was based on comparative functional studies in humans concerning transposed intestinal loops (Hanna et al. 1967, Marshall 1972, Rodgers et al. 1978) that proved the small bowel superior to large bowel or stomach. However, Germain et al. (1979) demonstrated in their comparative study in dogs using jejunal, colonic and tubed stomach autografts that the colonic segment was the oesophageal substitute of choice. This was probably due to the inherent anatomy and physiology of the various parts of the intestinal tract (diameter, plasticity, the absence of haustration in the large bowel) and the short segment used. Ancona et al. (1979) reported on 15 patients that underwent autotransplantation of short intestinal segments (10 colon, 5 jejunum) to the cervical region. He favoured the colonic segment as a substitute in short

oesophageal defects because of its diameter. His follow-up period however was rather short.

We conclude that the choice of a segment of small or large bowel to replace the oesophagus depends on the species involved, the length of oesophagus to be substituted and on the specific circumstances of the individual case.

4.2. Preservation of the transplant.

Each transplantation involves a period of ischaemia. Lillehei et al. (1959) were the first to report on the tolerance of ischaemia of the bowel. In their experiments on adult dogs they showed that the intestine can tolerate a complete circulatory stop lasting over a period of two hours without resultant damage, after intravascular perfusion and provided the intestine is cooled down to room temperature (25° - 28° C). Cooling to 5° C lengthens the time of ischaemic tolerance to a maximum of 5 hours.

McGill et al. (1979) argued that intravascular perfusion risks damage to the microvasculature. They showed the efficacy of surface cooling (6° - 12° C) for short time preservation (90 minutes) of intestinal segments. Germain et al. (1979) transplanted intestinal segments without cooling or perfusion with success.

The need for perfusing the free bowel transfer prior to the vascular anastomoses is a matter of controversy. Harashina and Buncke (1975) showed that perfusion is not necessary under conditions where the period of ischaemia in a free tissue transfer or replantation has not been prolonged (90 minutes of less). After prolonged periods of ischaemia, changes occur in the peripheral vascularity which act as an obstruction to blood reflow. This occurrence has been termed the "no-reflow phenomenon" (May et al. 1978).

Since ischaemic trauma to the transferred tissue should be kept to a minimum, a combination of cooling and perfusion of the transplant was used in our study. Surface cooling alone is not possible in the thoracic cavity where one has to work in a cramped space. Exclusion of cooling and perfusion is dangerous because if normothermic ischaemia becomes excessive the "no-reflow phenomenon" occurs. In this study the ischaemic damage to the transplant was minimal (see 3.2.6.), partly because ischaemic time did not exceed 66 minutes (see 3.1.) An added advantage of the prolonged perfusion was the fact that the thin walls of the vein were separated, aiding the anastomosis of the veins.

4.3. Denervation and lymphatic drainage of the transplant.

Since the transplanted ileal segment was to serve only as a functional conduit, the presence or absence of lympathic drainage and denervation were not of great importance. Furthermore it has been proved that as a rule

regeneration of lymphatic drainage after autografting of small bowel does occur in dogs (Goott et al. 1960 and Kocandrle et al. 1966). During the follow-up no problems were encountered concerning denervation and lymphatic drainage of the transplant. The transplant retained its own autonomous peristalsis (see 3.2.4.).

4.4. Selection of recipient vessels.

The internal mammary artery has been used for various vascular surgical purposes. The most familiar application is in coronary artery surgery. Longmire (1947) used the internal mammary artery as auxiliary blood supply in antethoracic oesophageal reconstruction using a pedicled colonic segment. And Meier (1968) made use of the internal mammary artery as recipient artery for the intrathoracic intestinal autograft. However, during his experiments he abandoned this artery and turned to an intercostal artery which he eventually preferred because of the supposed lower risk of torsion and tension.

Our preference for the internal mammary artery was threefold. Firstly this artery has a constant anatomical course and a constantly good caliber throughout its course. Secondly the artery can be dissected easily over a considerable length. Thirdly it can therefore be applied for revascularization purposes throughout the thoracic cavity.

As recipient vein the superior vena cava was chosen. Apart from the vena azygos it was the only vein that was considered for anastomosis because of the diameter of the mesenteric vein (4-5 mms). The vena azygos was ligated to permit access to the oesophagus in the posterior mediastinum and was therefore less applicable. Using the internal mammary artery and superior vena cava as recipient vessels both anastomoses were lying on the same side of the bed of the oesophagus. This made for easy handling of the transplant after recirculation without a risk of torsion.

4.5. Revascularization.

In microvascular surgery great emphasis is put upon a meticulous surgical technique. The most common cause of thrombosis is a technical error in the actual suturing of the vessel. With a meticulous surgical technique, expertise and proper equipment (magnifyers) nearly 100% patency rates can be expected with vessels with a diameter of as little as 1.0 mm (Hayhurst and O'Brian 1975). Anticoagulant drugs have not proven to have any effect in protecting vascular anastomoses from thrombosis in nontraumatized vessels (Engrave et al. 1975). Nevertheless, the majority of microsurgeons rely on some kind of prevention of thrombosis. In this study low molecular dextran and low dose heparin was used for prevention of thrombosis. The effect of these drugs, however, was not studied.

In our experiments revascularization was achieved in all cases (20 adult dogs and 11 puppies) during surgery. Early thrombosis (within 20 minutes) was not diagnosed in our series although in 3 cases the vitality of the transplant was doubtful. In 12 out of 20 adult dogs and 7 out of 11 puppies the transplant remained unimpaired throughout the experiment.

Postoperative vascular thrombosis proved the major complication (7 out of 20 adult dogs and 3 out of 11 puppies). We felt that there were at least two reasons for this complication occuring mainly at the beginning of the first series. Firstly, an initial lack of experience was the most important source of failure. Secondly, due to the fact that in the first series no bloodtransfusion could be given, additional sutures at the site of the arterial anastomosis, were made too soon after recirculation in order to minimize blood loss. Therefore in the second series bloodtransfusion was provided for and the technique of vascular surgery was changed. Interrupted sutures with 7.0 nylon and magnifying glasses were introduced. However, vascular thrombosis was still our major complication in the first part of the second series (puppies nos. 1-6). It was believed that dehydration of these puppies during the first postoperative week was the main cause. In the second part of the second series (puppies nos. 7-11) the puppies were allowed to drink water from the first postoperative day and no thrombosis occurred in these puppies.

4.6. Assessment of the vitality of the transplant.

The return of colour and peristalsis of the transplant and the pulsation in the vessels were used as parameters for the vitality of the transplant. These parameters are rather debatable. Perception of the colour of the transplant and the pulsation in the vessels with palpation is a subjective observation. What is more, pulsations can be rigorous while bloodflow is minimal or absent. Peristalsis does also exist in intestinal segments subjected to ischaemia.

Since vascular thrombosis and subsequent necrosis of the transplant is directly associated with death in this kind of surgery, a more objective parameter should be recommended. Electromagnetic or Doppler-flow measurements during surgery (Cooperman et al. 1979) and electro-myography (Dragojevic 1975) or regular endoscopic inspections after surgery are suggested.

4.7. Vascularization of the transplant.

Angiography of the transplant in the early and late phase of the follow-up period revealed that the vascularization of the transplant did not change during this follow-up period (see 3.2.5.). We only failed to obtain a picture of the vascularization of the transplant from dog no. 2 in the early and late phase as well as at autopsy. An explanation for the failure of the in vivo

angiographies cannot be given. The postmortal angiography could not be made due to the damaged mesenterial vessels. Yet at autopsy the transplant and vessels of dog no. 2 were no different from those of the other animals. So in this case it is unjustified to presume that the vessels were thrombosed in the early phase and the transplant was saved by collateral circulation.

However, during the course of the experiments it became clear from the postmortal angiographies that an extensive collateral circulation had developed at the site of the oesophagus-bowel anastomosis. This finding prompted us to demonstrate the validity of the collateral circulation. During selective angiography 2 weeks before autopsy showing the vascularization of the transplant to be intact, the internal mammary artery of puppies no. 10 and 11 were embolized. At autopsy the transplant revealed no signs of necrosis. This shows that some time after the oesophagus-bowel anastomoses have healed, an intact vascular pedicle is no longer essential at least where short segments of intestine (up to 6 cms) are concerned. In this connection we postulate that the anxiety of Okmian (1976) concerning the patency of the vessels during rapid growth in young individuals in analogous experiments is unfounded.

4.8. The oesophagus-bowel anastomosis.

In the anastomosis of oesophagus to small bowel the two layered telescopic suture technique of Haight (1969) was followed as in earlier experiments in our laboratory (Pull ter Gunne, 1975). Leakage, the occurrence of fistulae or stenosis at the site of the anastomosis were never noted. This was seen as the advantage of this technique. Althought great care must be taken not to stretch the muscularis of the oesophagus telescopically over the anastomosis to an excessive extent. This may create a kind of valve which may have happened in puppies nos. 9 and 11, rendering good passage impossible (3.2.3.).

4.9. Deglutition - Functional analysis of the oesophagoplasty.

Since survival after reconstruction of the thoracic oesophagus with free intestinal autografts in the studies cited from literature was poor (Meier 1968, Kozuschek et al. 1972, Dragojevic et al. 1975), a functional analysis of such a thoracic oesophagoplasty is unknown. Even a comprehensive functional analysis of such an oesophagoplasty in the cervical region has not been published. All investigators said that the cervical oesophagoplasty functioned well or satisfactorily. Apart from that, comparison between the cervical and thoracic oesophagoplasty with short autografts of intestine will be practically impossible. For in the cervical oesophagoplasty the length of normal oesophagus proximal to the transplant is only a few centimeters long and exposed to almost convulsive contractions of the pharyngeal

constrictors during deglutition. In contrast where the thoracic oesophagoplasty is concerned at least 10 cms of oesophagus is present proximal to the transplant. And in this part of the oesophagus a bolus in normally propelled by slow involuntary contractions of the oesophageal circular fibers. Thus, there is a basic difference in physiology of the two different positions of the transplant.

Deglutition of our animals was analyzed by cinematography (3.2.3.). Motor dysfunction of varying degrees and a decrease in plasticity appeared in the proximal oesophagus while the ileal segment and distal oesophagus kept its respective characteristics. The passage of contrast was delayed to some extent in each case. In the case of puppy no. 9 total stagnation of contrast had developed. Dog no. 5 showed a functional disorder due to an oesophageal stricture. It was observed that after an initial period of 5 weeks the findings did not change essentially during the rest of the follow-up year. During each oesophagoscopy autonomous segmental contractions were observed in the ileal segment albeit of no functional significance.

From the analysis of the swallow cinematographies it was concluded that the ileal segment acted merely as a passive conduit and formed a functional obstruction due to its unchanged inherent plasticity being totally different from the oesophagus. This resulted in a functional disorder of the proximal oesophagus and a delay of passage in comparison with normal deglutition.

The assumption that the antiperistaltically interposed transplant and its curved position in the first series with adult dogs was detrimental to good passage led to the alterations in the second series with puppies. In that series the transplant was interposed isoperistaltically and in a straight line with the oesophagus. However, these alterations did not have the desired effect on the passage. On the contrary, the overall functional results of the puppy group were worse than those of the adult dog group. Finally, where deglutition was concerned the dog with the oesophageal stricture did not differ essentially from the dogs with an intact transplant.

4.10 Morbid anatomy.

The macroscopical and microscopical morbid anatomy of the transplant and the oesophagus was followed intensively with oesophagoscopy, cinematography, histological examination and finally at autopsy. In each animal the ileal segment and the distal oesophagus remained morphologically unchanged whereas the proximal oesophagus had dilated to some extent.

A stricture was never found in connection with an intact transplant. The mucous membrane of the ileal segment as well as the proximal and the distal oesophagus remained unchanged macroscopically. Microscopically as well, the ileal wall kept its own characteristics. Passage of the oesophagoscope

through the transplant was possible except for those few cases where the position of the transplant prevented passage. Remarkably enough this also applied to the puppies nos. 9 and 11, that showed such a divergent picture on cinematography and in nutrition. The position of the transplant, which never altered, was always straight in the puppies, better in line with the oesophagus than in the adult dogs.

From these results of the follow-up programme it is concluded that the findings remained constant during the period of observation. No essential change could be observed in the consecutive examinations. The ileal segment remained unchanged. The adult dog group showed no essential divergence from the puppy dog group. The same conclusions were reported by other investigators (Pull ter Gunne 1975, Germain et al. 1979).

4.11. Nutrition and growth.

All the adult dogs could eat adequately, were able to take solids, kept their pre-operative weight or gained some weight and were clinically in good shape. Five of the six puppies that were followed for one year showed the same picture. In the other one, special dietary measures were resorted to. All six puppies grew into well- nourished adult dogs. Whether the transplant was straight or curved, interposed isoperistaltically or antiperistaltically, it made no difference clinically.

The growthrate and weightgain of the puppies is considered normal for such dogs during the first year of life. However, the quality of the interposed ileal segment seemed to have made little difference. It is more likely that the decisive factor was an incredibly powerful urge to live.

During the rapid growth of puppies in the follow-up period, the distance of the transplant to the anterior teeth increased 20 cms on average. So the length of the proximal oesophagus had increased. But autopsy of the puppies revealed that the interposed ileal segment kept the same length during this period of growth. Furthermore it was amazing that in the abdomen the length of the small bowel of the puppies at autopsy did not differ from the length of the small bowel left in place during operation. So the small bowel in the abdomen did not grow in length either.

These observations may prove valuable. The fact that the small bowel segments interposed in the oesophagus do not grow in length during the growth of the individual may preclude curvature of the interposition.

4.12. Dog no. 5.

In this dog the transplant was completely resorbed, due to thrombosis and necrosis. A possible explanation for the thrombosis may be the deep shock in the immediate postoperative period. It was amazing that the oesophageal defect and connecting leakage did not lead to death. Nevertheless, the dog

was kept in the follow-up programme in order to compare this dog functionally with the other dogs with an intact transplant.

As described in complications (3.2.2.) and cinematography (3.2.3.) dog no. 5 did not differ clinically from the other dogs, nor was there a divergent development of motor dysfunction in the proximal oesophagus. Conversely it can be stated that the 12 adult dogs with an intact transplant functionally did no better than a dog with an oesophageal stricture with a diameter of 6 mms and a length of about 1 cm.

4.13. Conclusions.

All three objectives of this study as formulated in chapter 1.3., have been accomplished.

1. A method has been developed to bridge a defect of the thoracic oesophagus with a free ileal autograft and this method can be applied to puppies without essential modifications. The operation will be successful when a meticulous microvascular technique is applied.
2. It proved possible to bridge a defect in the thoracic oesophagus, using a free ileal autograft without a subsequent disruption of normal growth and development in young adult dogs and puppies. The interposed ileal segment acted independently and functionally it was merely a passive conduit and it could not be distinguished from a short stiff dilated oesophageal stricture. Antiperistaltically or isoperistaltically interposed short ileal segments (up to 6 cms) gave the same functional results, as was the case with straight or curved interposed ileal segments. Morphologically the transplant kept its own characteristics. No increase in the length of the transplant was noted during the follow-up period of the adult dog or the puppy group.
3. Embolization of the vascular pedicle of the transplant has shown that the supply of blood through its own vascular pedicle is no longer essential some time after transplantation and when short intestinal segments (up to 6 cms) are used.
4. As a result of these experiments it seems justified to explore the possibilities of bridging a thoracic oesophageal defect in man using a free small bowel autograft. This premise is strengthened by the fact that a small bowel segment will conceivably function better in humans than in dogs due to inherent physiological and anatomical characteristics.

SUMMARY

In the first chapter of this thesis a short resume is given of the present state of affairs in connection with reconstruction of the oesophagus. The latest development concerns the transplantation of a free revascularized autologous bowel segment in the cervical region as an oesophageal substitute. In the last decade this method of reconstruction has become accepted in addition to existing techniques. The objectives of this study are discussed in the latter half of chapter I. The morbidity secondary to the existing technique of reconstruction using stomach or large bowel substitutes complicates the treatment of benign oesophageal disease in childhood to a serious extent.

Compared with publications concerning the transplantation of an intestinal segment to the cervical region very little has been reported on the substitution of the thoracic oesophagus using a revascularized autologous bowel segment. The poor experimental results reported in the literature prompted us to investigate the potential of a free intestinal autograft to replace the oesophagus in the thoracic region in laboratory animals. In view of the specifically paediatric surgical nature of the problems involved, this method had to be applied in small, rapidly growing individuals. To evaluate the function of such an oesophagoplasty a very intensive follow-up was essential.

The second chapter provides the data concerning dogs used, surgical techniques and methods.

The results of this experimental research are described in chapter three. The most dominant complication was vascular thrombosis of the transplant. The transplanted ileal segment, about 6 cms in length, remained unchanged during the entire follow-up period, microscopically as well as macroscopically. The transplant was merely a passive conduit in the oesophagus and formed a functional obstruction to the proximal oesophagus. This led to a more or less serious functional disorder in the proximal oesophagus. During the follow-up period it was shown, by means of embolization of the vascular pedicle of the transplant, that this pedicle is no longer essential for the supply of blood some time after transplantation.

The fourth chapter provides a critical review of the surgical technique and methods applied and the results achieved. It is concluded that it is feasible to replace a segment of the thoracic oesophagus with a free revascularized autologous ileal segment in adult dogs as well as in puppies. Although the functional results were by no means perfect, the dogs could feed adequately and there was no deterrent for the puppies to grow into adult dogs. The results of this experimental research seem to warrant investigation of the feasibility of applying this method of oesophageal reconstruction to human patients.

SAMENVATTING

In het eerste hoofdstuk van dit proefschrift wordt in het kort de huidige stand van zaken besproken met betrekking tot de reconstructie van de slokdarm. De jongste aanwinst op dit gebied is de transplantatie van een vrij gerevasculariseerd autoloog darmsegment naar de hals ter reconstructie van de spijsweg. Deze reconstructieve methode heeft zich in het laatste decennium een volwaardige plaats verworven naast de reeds bestaande technieken. In het tweede gedeelte van hoofdstuk I worden de beweegredenen die tot dit onderzoek hebben geleid, besproken. De morbiditeit na het gebruik van de bestaande reconstructieve technieken met transpositie van maag of dikke darm compliceert in ernstige mate de behandeling van goedaardige aandoeningen van de slokdarm op de kinderleeftijd.

Naar analogie van de transplantatie van een darmsegment naar de hals worden in de literatuur slechts spaarzame gegevens aangetroffen betreffende de vervanging van de thoracale slokdarm met behulp van een vrij gerevasculariseerd autoloog darmsegment. De slechte experimentele resultaten uit de literatuur zijn er de oorzaak van de mogelijkheden van een darmtransplantatie naar de borstholte ter vervanging van de slokdarm nogmaals te onderzoeken bij proefdieren. Gezien de specifieke vraagstelling vanuit de kinderchirurgie moet deze methode ook toepasbaar zijn bij kleine, snel groeiende individuen. Om de functie van een dergelijke slokdarmplastiek te kunnen beoordelen is een intensieve na-controle noodzakelijk.

In het tweede hoofdstuk zijn de gegevens betreffende de gebruikte honden, de chirurgische techniek en de gevolgde methoden beschreven.

De resultaten van deze experimentele studie zijn vastgelegd in het derde hoofdstuk. Thrombosering van de vaten van het transplantaat is de belangrijkste complicatie. Het ongeveer 6 cm lange getransplanteerde ileumsegment bleef gedurende het gehele na-onderzoek, zowel macroscopisch als microscopisch onveranderd. Het transplantaat gedroeg zich uitsluitend als een passieve geleidingsbuis in de slokdarm en vormde een functionele obstructie voor het proximaal gelegen deel van de slokdarm. Dit leidde tot meer of minder ernstige motorische stoornissen in de proximaal gelegen slokdarm. Tijdens het na-onderzoek werd door middel van embolisatie van de voedende mesen-

teriale arterie van het transplantaat aangetoond dat deze arterie te enigertijd na de transplantatie niet langer noodzakelijk is als bron van bloedvoorziening.

In het vierde hoofdstuk worden de chirurgische techniek, de gevolgde methoden en de bereikte resultaten kritisch besproken. Geconcludeerd wordt dat het mogelijk is een gedeelte van de thoracale slokdarm te vervangen door een vrij gerevasculariseerd autoloog ileumsegment, zowel bij volwassen honden als bij puppies. Hoewel de functionele resultaten te wensen overlieten, konden de honden op adekwate wijze voedsel tot zich nemen en konden de puppies uitgroeien tot volwassen honden. Als resultaat van dit onderzoek lijkt het gerechtvaardigd de mogelijkheden te onderzoeken om de beschreven techniek van slokdarmreconstructie bij patiënten toe te passen.

REFERENCES

ANCONA, E., PERACCHIA, A., FRASSON, P. . Microsurgical techniques in the reconstruction by free bowel autotransplantation of the cervical oesophagus. Intern. J. Microsurg. 1 : 13, 1979.

ANCONA, E., FRASSON, P. Autotransplantation of intestinal segments for the replacement of cervical oesophagus. Eur. Surg. Res. 10: suppl. 1, 74, 1978.

BELSEY, R.. Reconstruction of the esophagus with left colon. J. Thorac. Cardiov. Surg. 49 : 33, 1965.

CLARK, J., MORALDI, A., MOOSSA, A.R., HALL, A.W., DE MEESTER, T.R., SKINNER, D.B. . Functional evaluation of the interposed colon as an esophageal substitute. Ann. Surg. 183 : 93, 1976.

COLLIS, J.L. . The long-term clinical state after resection with gastro-oesophagostomy. In Smith, R.A., Smith, R.E. (eds): Surgery of the oesophagus The Coventry Conference. London, Butterworth, 1972.

COOPERMAN, M., MARTIN, E.W., KEITH, L.M., CAREY, L.C. . Use of doppler ultrasound in intestinal surgery. Am. J. Surg. 138 : 856, 1979.

DRAGOJEVIC. D., HETZER, R., CORTERIER, H. Experimenteller Teilersatz der Speiseröhre mit freiem Dünndarm- Transplantat. Langenb. Arch. Chir., Suppl. Chir. Forum 1975.

ENGRAVE, L.H., BENJAMIN, C.I., CRANDALL, H., PERRY, J.F. Experimental effects of heparin or magnesium sulfate on the patency of microvascular anastomoses. Plast. Reconstr. Surg. 55 : 618, 1975.

GERMAIN, M., GREMILLET, C., LESEC, G., et al. Cervical oesophagoplasty with free grafts revascularized by microsuture. A comparative study. Eur. Surg. Res. 11: suppl. 2, 106, 1979.

MCGILL, C.W., TAYLOR, B.H., FLYNN, M.B., ACLAND, R.D., FLINT, L.M. Effects of cooling and intraluminally administered antiseptics on surgically induced ischemia of the intestine in dogs. Surg. Gynec. Obstet. 149 : 377, 1979.

GOOTT, B., LILLEHEI, R.C., MILLER, F.A. Mesenteric lymphatic regeneration after autografts of small bowel in dogs. Surgery 48 : 571, 1960.

GRAGE, T.B., QUICK, C.A. The use of revascularized ileocolic autografts for primary repair after pharyngolaryngoesophagectomy. Am. J. Surg. 136 : 477, 1978.

GREEN, G.E., SOM, M.L. Free grafting and revascularization of intestine.1 Replacement of the cervical esophagus. Surgery 60 : 1012, 1966.

GRIMES, O.F. Replacements of the esophagus. Am. J. Surg. 100 : 278, 1960.

GRIMES, O.F. Surgical reconstruction of the diseased esophagus. Part I. Interposition of the jejunum. Surgery 61 : 325, 1967.

HAIGHT, C. The esophagus. In Mustard, W.T., et al. (eds): Pediatric Surgery (2nd ed. Chicago) Year Book Medical Publishers, Inc. 1969, Vol. I, p. 357.

HANNA, E.A., HARRISON, A.W., DERRICK, J.R. Long- term results of visceral esophageal substitutes. Ann. Thorac. Surg. 3 : 111, 1967.

HANNA, E.A., HARRISON, A.W., DERRICK, J.R. Comperative function of visceral esophageal substitutes by cinefluoroscopy. Ann. Thorac. Surg. 3 : 173, 1967.

HARASHINA, T., BUNCKE, H.J. Study of washout solutions for microvascular replantation and transplantation. Plast. and Reconstr. Surg. 56 : 542, 1975.

HAYHURST, J.W., O'BRIEN, B.M. An experimental study of microvascular technique, patency rates and related factors. Br. J. Plast. Surg. 28 : 128, 1975

HIEBERT, C.A. CUMMINGS, G.O. Successful replacement of the cervical esophagus by transplantation and revascularization of a free graft of gastric antrum. Ann. Surgery 154 : 103, 1961.

KOCANDRLE, V., HARTTUIN, E., PROHASKA, J.V. Regeneration of the lymphatics after autotransplantation and homotransplantation of the entire small intestine. Surg. Gynec. Obstet. 122, 587, 1966.

KOZUSCHEK, W., AIGNER, P.W., LINDECKEN, K.D., BAYERL, W. Experimenteller Oesophagus- Totalersatz durch freie homologe und autologe Darmtransplantate. Langenb. Arch. Chir. 331 : 119, 1972.

LILLEHEI, R.C., GOOTT, B., MILLER, F.A. The physiological response of the small bowel of the dog to ischaemia, including prolonged in vitro preservation of the bowel with successful replacement and survival. Ann. Surg. 150 : 543, 1959.

LIVADITIS, A., RADBERG, L., ODENSJÖ, C. Esophageal end-to-end anastomosis. Reduction of anastomotic tension by circular myotomy. Scand. J. Thor. Cardiov. Surg. 6 : 206, 1972.

LONGMIRE, W., A modification of the Roux's technique for antethoracic esophageal reconstruction. Anastomosis of the mesenteric and internal mammary blood vessels. Surgery 22 : 94, 1947.

LOUHIMO, J., PASILA, M., VISAKOPRI, J.K. Late gastroïntestinal complications in patients with colonic replacement of the esophagus. J. Pediat. Surg. 4 : 663, 1969

MALCOLM, J.A. Occurrence of peptic ulcer in colon used for esophageal replacement J. Thorac. Cardiov. Surg. 55 : 763, 1968.

MARSHALL, R. The results of laboratory and cine- radiographic investigations after oesophageal resection. In Smith, R.A., Smith, R.E. (eds): Surgery of the oesophagus/ The Coventry Conference. London, Butterworth, 1972.

MAY, J.W., CHAIT, L.A., O'BRIEN, B.M., et al. The no-reflow phenomenon in experimental free flaps. Plast. and Reconstr. Surg. 61 : 256, 1978.

MEIER, W. Experimenteller Oesophagus-Totalersatz durch freies autologes Dünndarmtransplantat. Wiederbelebung und Organersatz. Band 5 : 108, 1968

MERENDINO, K.A., DILLARD, D.H. The concept of sphincter substitution by an interposed jejunal segment for anatomic and physiologic abnormalities at the esophagogastric junction. Ann. Surg. 142 : 486, 1955.

NAKAYAMA, K. New reconstructive method after radical removal of carcinoma of cervical esophagus. Free autografts of intestine. Asion Med. J. 10 : 373, 1967.

OKMIAN, L. Reconstruction of the esophagus using autotransplanted small intestine. Z. Kinderchir. 18 : 25, 1976.

OTHERSON, H.B., CHARLESTON, S.C., CLATWORTHY, H.W. Functional evaluation of esophageal replacement in children. J. Thorac. Cardiov. Surg. 53 : 55, 1967.

PETERS, C.R., MCKEE, D.M., BERRY, B.E. Pharyngoesophageal reconstruction with revascularized jejunal transplants. Am. J. Surg. 121 : 675, 1971.

PICHLMAIER, H. The pathophysiology of the esophagus and its replacement. In Sorensen, H.R. et al. (eds): The function of the esophagus. Odense University Press, 1973.

PULL TER GUNNE, H.P. Vervanging van de cervicale oesophagus door een gerevasculariseerd ileumsegment. Een experimenteel onderzoek bij honden. Thesis of University of Rotterdam, 1975.

REHBEIN, F., SCHWEDER, N. Reconstruction of the esophagus without colon transplantation in cases of atresia. J. Pediat. Surgery 6 : 746, 1971.

RODGERS, B.M., TALBERT, J.L., MOAZAM, F., FELMAN, A.H. Functional and metabolic evaluation of colon replacement of the esophagus in children. J. Pediatr. Surg. 13 : 35, 1978.

SEIDENBERG, B., ROSENAK, S.S., HURWITT, E.S., SOM, M.L. Immediate reconstruction of the cervival esophagus by a revascularized isolated jejunal segment. Ann. Surg. 149 : 162, 1959.

WATERSTON, D.J. Colon replacement of esophagus (intrathoracic). Surg. Clin. N. Amer. 44 : 1441, 1964.

WOOLLEY, M.M., LEIX, F., JOHNSTON, P.W., HAYS, D.M. Esophageal atresia types A and B; upper pouch elongation and delayed anatomic reconstruction. J. Pediat. Surg. 4 : 148, 1969.

ACKNOWLEDGEMENTS

The experiments took place at the Laboratory for Experimental Surgery, Erasmus University, Rotterdam during my fellowship at the Department of Paediatric Surgery of the Sophia Children's Hospital, Rotterdam, and my surgical training at the Department of Surgery of the University Hospital "Dijkzigt", Rotterdam.

It was probably Prof. Dr. D.L. Westbroek who first conceived the basic idea to reconstruct the thoracic oesophagus in the manner described in this thesis. His faith and active participation in this project are acknowledged. Prof. Dr. J.C. Molenaar breathed new life into the basic idea. I am most grateful for the inspiring supervision in his very personal way. The critical reading of the manuscript by Prof. Dr. H. van Houten is greatly appreciated. The dogs were anaesthesized skillfully by Mr. E.C.C. Colly, Mr. J. Poortman and Mr. E. Ridderhof.

Mrs. J. Dortland-de Kam and Mrs. M.J. Lagerman assisted int he surgical procedures.

Excellent pre- and post-operative care of the dogs was provided by Messrs J. Kasbergen, R.W.J. Meyer, R. Spruit and R. Briegoos.

Mr. R.W.J. Meyer prepared the histological sections.

The cinematograms and angiograms were made by Mr. J.F. Fassotte and Mrs. B.M. Fassotte-van Leeuwen.

All cinematograms were reviewed in co-operation with Dr. M. Meradji, radiologist. The description of cinematograms has gained much in value by his expertise.

Dr. A. Douwes, paediatricion, taught me to use the fiberscope for the endoscopic examinations. The endoscopic equipment was always cared for by the personnel of the outpatient department of the Sophia Children's Hospital (head Mrs. J.C. Koch).

Dr. F.J.W. ten Kate, pathologist, is thanked for his help in evaluating the histology.

The figures were provided by the "Audiovisuele Dienst" of the Erasmus University.

I wish to thank Mrs. A.M.C. Goslinga-Ribbink for her assistance in editing the English style of this thesis.

Mr. E.J.W. Davids carried out the grafical outline.

CURRICULUM VITAE

De schrijver van dit proefschrift werd op 20 april 1949 geboren te Bussum. Hij behaalde zijn eindexamen Gymnasium ß aan het Openbaar Dalton Lyceum te Voorburg in 1967. In hetzelfde jaar werd de medische studie aangevangen aan de Rijksuniversiteit te Leiden. In 1975 werd het artsexamen afgelegd. Na het vervullen van de militaire dienstplicht was hij gedurende 9 maanden werkzaam als arts-assistent op de afdeling kinderchirurgie van het Sophia Kinderziekenhuis te Rotterdam. Sinds mei 1977 is hij in opleiding tot chirurg in het Academisch Ziekenhuis Dijkzigt te Rotterdam.