THE ROLE OF PLANNED RELOOK LAPAROTOMY IN THE MANAGEMENT OF SEVERE INTRA-ABDOMINAL INFECTION IN AN EXPERIMENTAL RAT MODEL

N G BROWNING

The Role of Planned Relook Laparotomy in the Management of Severe Intra-abdominal Infection in an Experimental Rat Model

Neil Browning

A Research Report submitted to the Faculty of Medicine of the University of the Witwatersrand, Johannesburg in partial fulfilment of the requirements for the degree of Master of Medicine in the discipline of Surgery.

1991

ABSTRACT

Severe intra-abdominal infection (IAI) carries a high mortality. Methods of treatment such as radical peritoneal debridement, continuous post-operative lavage and local intra-peritoneal instillation of anti-biotics have not improved survival.

To date the "on demand" relaparotomy is the approach used by most surgeons but even this method has produced disappointing clinical results.

This study tested the efficacy of planned re-look laparotomy in a controlled situation using an experimental rat model. The study involved introducing a controlled septic challenge into the peritoneal cavity of known volumes of human faeces. The test dose of this septic challenge was determined in the first phase of the study. The test dose was found to be 0.4 ml of innoculum which produced a mortality rate of 80%. The second phase of the study involved 6 groups of rats receiving (a) irradiated faces (control group); (b) innoculum and immediate antibiotics; (c) innoculum and antibiotic 5 hours later; (d) innoculum and then washout of the peritoneal cavity with saline 5 hours later; (e) innoculum then washout of the peritoneal cavity and antibiotic 5 hours later (operate and observe group) and (f) innoculum, washout of the peritoneal cavity and antibiotic 5 hours later and then a relook laparotomy at 24 hours. (Planned relook laparotomy group).

Survivors of the first four groups of rats were euthanased at the end of seven days and the survivors of the last two groups after two months. The variables measured were survival, body mass and the number of intra-abdominal abscesses formed.

The results showed that an "operate and observe" policy gave the best overall results in terms of survival, catabolic response to infection (body mass) and number of intra-abdominal abscesses. Planned relook laparotomy policy gave results which were not statistically significantly different from this group, and thus relook laparatomy does not confer any additional benefit.

The group treated with antibiotics had the lowest mortality rate and the least loss of body mass, but developed intra-abdominal abeceeses.

The highest mortality rate occurred in the group receiving washout only. However the washout did reduce the number of intra-abdominal abscesses formed, and the amount of body mass lost.

I conclude that planned relook laparotomy is of little clinical benefit, at least in this animal model.

DECLARATION

I hereby declare that this research report is my own work. It is being submitted in partial fulfilment for the degree of Master of Medicine in the discipline of Surgery at the University of the Witwatersrand, Johannesburg. It has not been submitted before for any degree or examination in any other Univereity.

ETHICAL APPROVAL

This study was approved by the Animal Ethics Committee of the Univereity of the Witwatersrand (AEC 89/48/5).

.....

N. Browning

Johannesburg, 1991

ACKNOWLEDGEMENTS

My sincere thanks and appreciation:-

To Professor G. Mitchell and Professor H. Lawson for their advice and encouragement as joint supervisors of this research report.

To Professor H. Koornhof of the Department of Microbiology, SAIMR for his advice concerning the antibiotic used in this experiment and the monitoring of important bacterial species in the innoculum.

To technical staff of the Department of Microbiology, SAIMR who prepared the capsules of innoculum and performed the monitoring of bacterial species as well as other technical aspects of the microbiological part of this experiment.

To the staff of the Department of Medical Biostatistics of the University of Manchester for their advice and assistance with the statistical aspects of this experiment.

To the staff of the M.R.C. Department of Statistics for their advice with regard to the statistical aspects of this study.

To the technical staff of the Department of Surgery of the University of the Witwatersrand for their assistance with the surgical aspects of this study.

To Dr Steven Maeder and the staff of the Central Animal Service of the University of the Witwatersrand for their assistance in looking after the rats post-operatively, their assistance with autopsies and advice as to when the end points laid down for this study, had been reached.

MRC/University for financial support.

To the Department of Surgery of the University of the Witwatersrand Medical School for funds granted to make this study possible.

TABLE OF CONTENTS

LIST OF TABLES

Page

LIST OF FIGURES

Page

CHAPTER 1

INTRODUCTION AND REVIEW OF THE LITERATURE

1.1 INTRODUCTION

Abdominal surgery has seen many recent advances in surgical techniques, more stringent indications for surgery, better identification of subgroups requiring surgery, better anaesthesia and antibiotics and better post-operative care. Yet the management of severe intra-abdominal infection (IAI) does not seem to have kept pace with these advances. Current surgical treatment of the problem remains disappointing and its associated mortality rate is unacceptably high[1] [2]. This dissatisfaction has stimulated abdominal surgeons to seek more effective modalities of treatment. Among these newer modalities is the planned re-look laparotomy, a procedure which has not been analysed systematically. The aim of this study was to evaluate the efficacy of this modality of treatment in the management of severe intra-abdominal infection in a controlled laboratory environment using an experimental rat model.

1.2 REVIEW OF THE LITERATURE

Intra-abdominal infection (I.A.I.) in humans occurs as a result of the release of endogenous gastro-intestinal flora into the peritoneal cavity. Infection may follow perforation of a part of the gastro-intestinal tract, for example a perforated peptic ulcer or perforated colonic diverticulum, perforation after blunt or penetrating trauma and anastomotic leaks after surgery. It can also occur with primary intra-abdominal infection such as pancreatic sepsis.

The spectrum of clinical presentation of I.A.I is large. At one end of the spectrum, a healthy individual with a localised perforation of the appendix; at the other end, intra-abdominal infection overwhelms the patient's ability to contain it, resulting in overwhelming, diffuse and often lethal peritonitis. It is this latter group which persists and does not localize, despite conventional supportive and operative measures, that poses the real threat to the patient and the real challenge to the surgeon. Patient survival depends on the struggle between the host's peritoneal and systemic defences on the one hand and the nature, volume and virulence of the contamination on the other. If intra-abdominal sepsis progresses multiple organ failure manifests itself and with it the chance of survival decreases[8].

1.2 REVIEW OF THE LITERATURE

Intra-abdominal infection (I.A.I.) in humans occurs as a result of the release of endogenous gastro-intestinal flora into the peritoneal cavity. Infection may follow perforation of a part of the gastro-intestinal tract, for example a perforated peptic ulcer or perforated colonic diverticulum, perforation after blunt or penetrating trauma and anastomotic leaks after surgery. It can also occur with primary intra-abdominal infection such as pancreatic sepsis.

The spectrum of clinical presentation of I.A.I is large. At one end of the spectrum, a healthy individual with a localised perforation of the appendix; at the other end, intra-abdominal infection overwhelms the patient's ability to contain it, resulting in overwhelming, diffuse and often lethal peritonitis. It is this latter group which persists and does not localize, despite conventional supportive and operative measures, that poses the real threat to the patient and the real challenge to the surgeon. Patient survival depends on the struggle between the host's peritoneal and systemic defences on the one hand and the nature, volume and virulence of the contamination on the other. If intra-abdominal sepsis progresses multiple organ failure manifests itself and with it the chance of survival decreases[1].

Three organ failure persisting for longer than three days carries a 93% mortality[4].

Recognition of the nature and severity of this problem has resulted in a number of modalities of treatment being tried.

Earlier promising methods like radical peritoneal lavage[6][7] have not really helped in the treatment of severe IAI. Local intra-peritoneal installation of various anti-biotics and anti-septic solutions also have not improved patient survival[8].

The "on demand" re-look laparotomy is the preferred management policy practised by many surgeons when confronted with severe IAI. The principle criticism of this method is that it relies on the appearance of clinical signs of residual intra-abdominal infection and of the development of multiple organ failure[9] as the principle indicators for further surgical intervention. This policy has also been disappointing as a method of treatment[10][11].

One of the key elements in treating severe intra-abdominal infection is early diagnosis. Pitcher and Musker[12] recognized this and identified factors responsible for delay and hence increased mortality. These were:-

- a prolonged period of observation

- fruitless attempts to try and confirm the presence of severe IAI using ancillary studies
- the opinion that the patient was too sick for surgical intervention.

To try and overcome this problem of delay the concept of staged planned re-look laparotomy was introduced[13]. The idea behind this seems logical and attractive - why wa't for severe intra-abdominal infection to manifest itse'f locally or systemically? Rather operate before multiple organ failure begins! In this method the septic abdomen is re-explored every 48-72 hours until it is macroscopically clean, pus loculations are removed, slough debrided and residual sources of soiling dealt with as deemed necessary by the surgeon at the time. Together with planned relook laparotomy the concept of open management of the infected abdomen has developed, whereby the abdomen is not closed between re-look laparotomies. The interest and hope generated by these new modalities of treatment is reflected in increasing numbers of reports [13-27], editorials[20,29] and review articles[2,30-32].

Yet the surgeon remains uncertain about the merits of this newer modality of treatment and a number of questions need to be answered before it can be accepted. These are:-

a) Does a policy of re-look laparotomy improve survival compared to conventional treatment?

b) Does relook laparotomy reduce the catabolic response to infection?

c) Does relook laparotomy decrease the number of intra-abdominal abscesses?

The difficulty in answering these questions lies in three areas.

Firstly, no controlled randomized trials have been performed. Secondly, published series suffer from their retrospective natures, lack of precise definition of selection criteria, imprecise or different definitions of the severity of sepsis and inadequate patient stratification[15,27]. Published studies deal with patients suffering from a wide spectrum of IAI and receiving multi-faceted and complex treatments. Moreover, the range of disease processes in which relook laparotomy has been used is large. Staged planned re-look laparotomy has been used for first time peritonitis e.g. perforated peptic ulcers or appendicitis as well as post-operative and pancreatic sepsis[16,18,19,21,25,26,33]. When a treatment modality is used for such a wide spectrum of disease it is very difficult to decide which types of IAI are bested suited for this modality of treatment since the results are all pooled. It is well known that diffuse faecal peritonitis, post-operative peritonitis due to anastomotic dehiscence

and pancreatic sepsis are associated with the highest mortality[34] compared to first time peritonitis[1].

Thirdly, since planned re-look laparotomy is now used in conjunction with "open management" of the septic abdomen it is very difficult, if not impossible, to separate these two modalities in order to gauge their respective merits[16,18,19,21,25,26,33], - although they are synergistic rather than antagonistic. One of the studies which has attempted to seperate these types of treatment is the study by Schein[27] who submitted twenty-two patients to planned re-look laparotomy for severe IAI. Of these twenty-two, thirteen had the abdominal wall closed between laparotomies. In nine patients the abdominal wall was left open and covered with Marlex Mesh. Eleven of the first group (85%) survived while only four of the second group (44%) survived. A flaw in this study, however, was that patients in the second group were sicker and thus could be expected to have a lower survival than patients in the first group.

The value and place of re-look laparotomy will depend therefore, on how well the questions asked earlier in this chapter can be answered. The most important question is whether relook laparotomy improves survival. Data from human studies are equivocal. Firstly there are very few studies published which used planned re-look laparotomy alone as the only component of these newer methods of

treatment. Secondly the results are variable. Pennickx[13] published a retrospective study of forty-two patients with severe IAI in which he compared a conventional "on demand" re-look laparotomy group and a staged planned re-look laparotomy group. Both groups included patients with primary and secondary infection. The mortality of the "on demand" group was 73% as against 36% for the staged planned re-look laparotomy group. He reported a negative re-look laparotomy rate of 33% in this latter group. Interestingly, fewer patients with three or more organ failure died in the planned staged re-look laparotomy group (42%) than in the "on demand" group (80%). This study seemed to support planned relook laparotomy, but looking critically at the paper it is not obvious what the criteria were for submitting patients to staged planned re-laparotomies. While the two groups seemed well matched in certain respects, information was lacking in critical areas, namely, whether the groups were comparable in terms of the cause of sepsis, the severity of contamination and the time delay before treatment. These factors affect the prognosis.

In another study reported by Teichman[14], sixty-one patients with severe IAI resulting from post-operative anastomotic leaks and from spontaneous perforations, were submitted to planned re-look laparotomies. The mortality rate was found to be 22.9%, and no differences were found between those with post-operative IAI and IAI resulting

treatment. Secondly the results are variable. Pennickx[13] published a retrospective study of forty-two patients with severe IAI in which he compared a conventional "on demand" re-look laparotomy group and a staged planned re-look laparotomy group. Both groups included patients with primary and secondary infection. The mortality of the "on demand" group was 73% as against 36% for the staged planned re-look laparotomy group. He reported a negative re-look laparotomy rate of 33% in this latter group. Interestingly, fewer patients with three or more organ failure died in the planned staged re-look laparotomy group (42%) than in the "on demand" group (80%). This study seemed to support planned relook laparotomy, but looking critically at the paper it is not obvious what the criteria were for submitting patients to staged planned re-laparotomies. While the two groups seemed well matched in certain respects, information was lacking in critical areas, namely, whether the groups were comparable in terms of the cause of sepsis, the severity of contamination and the time delay before treatment. These factors affect the prognosis.

In another study reported by Teichman[14], sixty-one patients with severe IAI resulting from post-operative anastomotic leaks and from spontaneous perforations, were submitted to planned re-look laparotomies. The mortality rate was found to be 22.9%, and no differences were found between those with post-operative IAI and IAI resulting

from spontaneous perforation. This study however had no control group.

A better study than Teichman's is that of Andrus et al[15]. Andrus' study was not randomized but consisted of seventy-seven patients with severe IAI from spontaneous and post-operative intestinal perforation both of the small and large bowel, as well as sepsis from other sources. Patients were divided into a "control" group of forty-three patients who had no planned re-look laparotomy following their initial abdominal exploration and a second group of thirty-four patients all of whom had a planned re-look laparotomy. Patients were closely matched for age, sex, aetiology and source of sepsis and for acute physiological score (which correlates closely with mortality). This study showed that

- there was no difference in survival between the two groups irrespective of the severity of the acute physiological score.
- the effect of planned re-look laparotomy on survival in elderly patients was not significantly better than the results in patients more than seventy years old, operated on once and then observed.
- patients with post-operative IAI who had planned re-look laparotomies had a 38% survival as compared to 45% survival rate in a comparable group who did not have planned re-look operations. Thus a policy of planned re-look laparotomy did not appear to improve

overall survival. The significance of this finding is that patients with post-operative IAI have been recognized as a high risk group and as such may be a group to benefit from a policy of planned re-look laparotomy.

- there was no significant difference between the two groups when upper gastro-intestinal, lower gastro-intestinal and non gastro-intestinal sources of intra-abdominal infection were compared in those having and not having planned re-look laparotomies. This fact is again surprising as patients with faecal peritonitis of lower gastro-intestinal origin are also felt to be a high risk group and may be a group to benefit from planned re-look laparotomy.

Andrus's study can be criticised because the "control" group was not a real control group but a choice of the surgeon and because of the relatively high number of re-laparotomies in the control group which may have masked the effects of the planned re-laparotomy group.
Frequent re-operations also carry the problem of repeated anaesthetics, progressive oedema and maceration of the anterior abdominal wall. There is also the danger of causing fistulae as oedematous and adhesive bowel is repeatedly mobilized.

In virtually all other published series (see Table I) planned re-look laparotomy has been used in conjunction

with open management of the septic abdomen. These are complementary rather than opposing methods. Merely leaving the abdomen open is ineffective. One study[2] showed a drop in mortality from 66% when the abdomen was left open post-operatively, to 30% when systematic, regular, re-operations were done. Open management of the septic abdomen nevertheless does have some attractive and practical advantages.

It:-

a) facilitates the drainage of intra-abdominal and infective potentiating agents like necrotic material.

b) causes less pulmonary compromise because no attempt is made to return distended loops of oedematous bowel to the peritoneal cavity in order to close the abdominal wall.

c) does prevent oedema and maceration of the abdominal wall associated with multiple closures.

But it does have potential co..lications lik id loss, fistulae, evisceration of abdominal con , and large incisional hernias.

When staged planned re-look laparotomies and open management are used together then it becomes very difficult to evaluate the staged re-look laparotomy component of the treatment.

with open management of the septic abdomen. These are complementary rather than opposing methods. Merely leaving the abdomen open is ineffective. One study[2] showed a drop in mortality from 66% when the abdomen was left open post-operatively, to 30% when systematic, regular, re-operations were done. Open management of the septic abdomen nevertheless does have some attractive and practical advantages.

It:-

a) facilitates the drainage of intra-abdominal and infective potentiating agents like necrotic material.

b) causes less pulmonary compromise because no attempt is made to return distended loops of oedematous bowel to the peritoneal cavity in order to close the abdominal wall.

c) does prevent oedema and maceration of the abdominal wall associated with multiple closures.

But it does have potential complications like fluid loss, fistulae, evisceration of abdominal contents and large incisional hernias.

When staged planned re-look laparotomies and open management are used together then it becomes very difficult to evaluate the staged re-look laparotomy component of the treatment.

Table 1 shows the morality rate from published series when open management was used in conjunction with planned re-look laparotomy (*) or with "on demand" re-look laparotomy (+). It is noticeable that mortality rates vary greatly and overall do not show a significant survival difference for a planned re-look laparotomy policy as opposed to an "on demand' re-look laparotomy policy, taking into account that open management was used in both groups. If the mortalities for the two groups are averaged out, they are 28,1% for the "on demand" group (121 patients) and 30,7% for the planned group (105 patients). Also noticeable is the wide variety of conditions for which the procedures were used. This fact underlines again the difficulty in trying to evaluate these treatment modalities.

In summary the literature does not offer any conclusive evidence to suggest a real benefit or a policy of re-look laparotomy in t(ms of improved mortality. Although Pennickx's data[13] suggest that planned re-look laparotomy is better than on demand re-look laparotomy, Andrus's paper[15] shows no such benefit. Teichman's series[14], although it has a relatively low mortality rate, has no control group for comparison. All these flaws make it difficult to draw firm conclusions.
Similarly, as shown in Table 1, if one tries to separate the series into open management plus planned re-look laparotomy

TABLE I

AUTHOR	NUMBER OF PATIENTS	INDICATION	MORTALITY
+ Duff & Moffat[17]	18	I.A.S. (1° & 2°) & P.S.	38%
+ Goris[20]	26	I.A.S. (2°) & P.S.	50%
+ Wouters[23]	20	I.A.S. (1° & 2°)	20%
+ Broome et al[21]	30	I.A.S. (1° & 2°) & P.S.	47%
+ Maetani and Trobe[18]	13	I.A.S. (1° & 2°)	7%
* Anderson et al[19]	20	I.A.S. (1° & 2°) & P.S.	60%
* Pemberton et al[25]	17	P.S.	18%
* Hedderich et al[33]	10	I.A.S. (1° & 2°) & P.S.	20%
* Wertheimer & Norris[26]	10	P.S.	20%
* Mughal et al[22]	18	I.A.S. (2°) & P.S.	28%

KEY:

I.A.S. = Intra-abdominal sepsis due to leakage of hollow visceral contents

P.S. = Pancreatic sepsis

* = Open management and planned relook laparotomy

\+ = Open management and demand relook laparotomy

1° = Primary leakage of hollow visceral contents

2° = Leakage of hollow visceral contents secondarly to anastomotic break down from previous surgery or septic complication from previous surgical intervention.

versus open management plus "on demand" re-look laparotomy, then by eliminating the common factor of open management one would hope to be left with two comparable groups. Unfortunately the mortality figures vary so widely and the spectrum of IAI being treated is so wide that little can be concluded from these data either.

In addition to the questions which are the subject of this report, some other questions need to be asked. These are:

a) What categories of severe IAI should a policy of planned relook laparotomy be used for?
b) Can planned relook laparotomy reverse multiple organ failure?
c) Does planned relook laparotomy justify the complications and negative re-exploration rate?

It is not unreasonable to assume that even if staged re-look laparotomy does not have a role in the whole spectrum of severe IAI, that there would be subgroups in this spectrum that would benefit from this more aggressive form of therapy. The most logical subgroups would be those that carry the highest risk in terms of mortality. Both Schein[36] and Mughal[22] have suggested that these would be severe IAI due to faecal peritonitis, breakdown of surgical anastomoses and pancreatic sepsis which carries a mortality of over 50%.

In the group of post-operative IAI there seems to be no good evidence that a policy of re-look laparotomy with or without open management has had any dramatic impact on prognosis. Schein[9] found a mortality of 55% in his own series in this subgroup of patients, most of them dying from multiple organ failure despite routine re-laparotomies and prolonged staye in the Intensive Care Unit. It may be that these radical mechanical methods to deal with severe intra-abdominal infection are either too late or inappropriate. Certainly, in Schein's series[9] the mortality in this group was far higher in patients that were referred from outside hospitals where there had been a delay in diagnosis and treatment (67%) than in patients that developed intra-abdominal infection in his hospital (25%). This tends to support the concept that early definitive drainage of intra-abdominal infection is critical for survival.

The average mortality for patients suffering from post-operative intra-abdominal infection and treated by planned re-look laparotomy with or without open management is 41% (20-62%)[9]. Mortality rates for the same group treated by conventional methods is between 30-75%[9]. Thus re-look laparotomy does not seem to have reduced mortality in this subgroup of patients and its role must be in doubt.

Infected pancreatic necrosis seems ideally suited for this form of treatment since it allows frequent inspection, debridement and removal of pancreatic and peripancreatic slough.

Bradley reported a mortality rate of 14%[24] using frequent re-explorations for this form of intra-abdominal infection. Good results have been reported by other groups[25,26] using this method. But Schein[3] and others[36,37] using th me modality of treatment have not been able to replica these results.

Decreased mortality rates have also been reported by groups using debridement and closed catheter drainage[38,39,40] and debridement and post-operative lavage[41,42].

In a collected series of 173 patients suffering from pancreatic infection and treated by these newer aggressive methods Schein[9] found a mortality rate of 26%. Similar mortality rates (22%) were also achieved by closed management modalities[38-42]. Thus it is not possible to say that frequent relook laparotomy and open management has been shown to be the superior treatment modality for intra-abdominal infection due to pancreatic sepsis.

The third high risk group is that of faecal peritonitis. The highest mortality is found in non-localized forms of faecal peritonitis following colonic perforation. One of the problems in trying to assess published reports is the

lack of definition of what constitutes generalized or localized peritonitis and what constitutes purulent or faecal peritonitis. The inappropriate inclusion of patients with localized or seropurulent peritonitis and abscesses in studies of diffuse faecal peritonitis means that mortality rates are reduced because these patients have a more favourable prognosis[43].

In a collected series of 262 patients with generalized faecal and purulent peritonitis secondary to diverticular disease, who were treated by conventional resection (without anastomosis) - the mortality was 12.2%[43].

In another series the mortality rate following sigmoid resection or exteriorisation for faecal peritonitis was 21%[44]. Bakkar[45] reported a 37% mortality for perforated diverticular disease of undescribed severity treated by Hartmann's procedure. On the other hand Walsh[46] reported a mortality rate of (11%) in patients with faecal peritonitis treated with open management and planned relaparotomies. Schein[9] reported a mortality rate of 14% in a similar group of patients treated in the same way. Although Schein[9] believes that a policy of planned relook laparotomy in patients with diffuse faecal peritonitis is beneficial I feel that this may be unnecessarily aggressive since it has been shown that a similar mortality rate can be achieved with conventional methods[43].

The second question to be answered is whether a policy of planned relook laparotomy is able to address the problem of multiple organ failure.

Intra-abdominal sepsis is the most common cause of multiple organ failure[47]. In Schein's series[9] multiple organ failure was the cause of death in 87% of mortalities despite routine relook laparotomies until death. This experience is shared by Norton[47] who showed that in most patients organ failure persists despite eradication of intra-abdominal infection. This does not seem to be influenced by the number, location or bacteriological characteristics of the abscesses or by the number of re-explorations. Surgeons who have some experience with intra-abdominal infection are only too familiar with the picture of patients with macroscopically clean abdomens, well controlled gastro-intestinal fistulae and on appropriate anti-biotics and nutritional support, initially seeming to do well only to die from progressive organ failure initiated and sustained by immunological failure, opportunistic gut infection and the I.C.U. environment[48]. The current explanation of this problem is that it is caused by a breakdown of gut barrier function[49]. The gut is a source of pathogens and in critically ill surgical patients bacteria cross the gut mucosa to reach the portal venous and systemic circulation. Factors that pre-dispose to the translocation of bacteria from the gut include malnutrition (nutrition is important for the physical

integrity of gut mucosa), prolonged parenteral nutrition (which causes gut mucosal atrophy and decreased IgA secretion to the gut), sepsis (which promotes bacterial translocation) and intra-abdominal pathology (eg. intra-abdominal abscesses are colonized by gut bacterial. These factors are all active in patients with severe IAI. So there is scant, if any evidence that adequate drainage and control of intra-abdominal infection can reverse established multiple organ failure.

Thirdly, does the policy of planned relook laparotomy justify complications and the negative re-exploration rate?

Gastro-intestinal fistulae are a problem and in one series occurred in 70% of patients and carried a mortality of 51%[9]. They seem to occur mainly in patients suffering from post-operative intra-abdominal infection and pancreatic sepsis who are treated by open management. The causes of the fistulae are exposure injury, dehiscence of suture lines or the trauma of re-explorations. However, they can be dealt with by way of proximal diverting enterostomies to control leakage into the peritoneal cavity. Schein[9] feels that complications from open management like evisceration, fluid loss and spontaneous fistulae from exposed bowl can be virtually eliminated using Marlex Mesh to cover the defect in the anterior abdominal wall.

A policy of planned relook laparotomy will always entail a certain percentage of negative re-laparotomies. This may be as low a 4%[9] to as high as 33%[13]. The experience of the surgeon is important in this regard. As he grows more experienced he learns what type of intra-abdominal injection to leave to the patients own defence mechanisms[1].

Thus, a close look at the literature reporting the use of planned relook laparotomy with or without open management of the septic abdomen shows a wide variety of results. At one end of the spectrum is a group of relatively fit patients with a less severe form of IAI in whom relook laparotomy is probably not necessary as they would do as well with conventional treatment. They contribute to the favourable results reported by some workers[16,18,23]. At the other end of the scale are sick patients with established and irreversible multiple organ failure. In these cases even the most aggressive surgical modalities fail and the mortality rate increases. Somewhere between these two extremes may lie a group that would benefit from these newer treatment modalities mentioned, but to date this group has not been clearly identified.

My study is an attempt to assess the efficacy of planned staged relook laparotomy in a controlled situation using an experimental rat model. By eliminating variables such as the cause of the intra-abdominal sepsis, the degrees

of peritoneal soiling, and the timing and nature of treatment. I hope to evaluate this method of treatment in a more scientific way. This approach to resolving the controversy has not been previously reported in the literature.

CHAPTER 2

MATERIALS AND METHODS

The model used in this experiment involved producing severe IAI in rats, of the faecal peritonitis type. This model was created by introducing gelatine capsules containing a faecal-barium sulphate mixture into the peritoneal cavity of the rats. The model specifically aimed at eliminating the many variables that have made clinical reports so difficult to analyse. Thus the sex of the rats, their weight range, the type of septic challenge used, the volume and bacteriology of the infective innoculum and the types of treatment were all standardized. This ensured that the results could not be influenced by variations in methodology. In addition, the study was conducted in such a way that the methodology only varied by one factor as the experiment progressed from one group of rats to the next. By keeping all other variables the same, the effect of the single variation could be more accurately assessed.

1. The Animal Model

Numerous animal models have been used to try and produce IAI that mimics human IAI. In selecting an animal model I felt that the following criteria needed to be fulfilled:

i) The animals used needed to be easily available in large numbers, cheap to house and feed, genetically identical, on the same diet and be specific pathogen free.

ii) The type of septic challenge had to mimic the human situation as closely as possible.

iii) The pathogenesis of the septic challenge had to be the same as that occurring in the human i.e. contamination, inflammation, peritonitis and then abscess formation.

iv) The volume and bacteriological content of the infective challenge had to be the same for all animals or so nearly so that it did not influence the results.

As far as the first requirement was concerned, small rodents fulfilled most of these criteria. Dogs have not been used to study abdominal sepsis and the literature on them is scanty. Rabbits are not ideally suited as a model for peritonitis as they have a high content of cellulose digesting bacteria not found in the human intestine. In addition rabbits have a relatively low intra-intestinal pH compared to humans. Guinea-pigs are seldom available free from specific pathogens[50].

2. The Septic Challenge

Several methods have been used to try and produce an adequate animal model for severe IAI.

2.1 Endotoxin Model

Many animals including rats have been used in endotoxin models. Endotoxins are released from gram-negative organisms and are an important component of sepsis. However there is concern about the clinical relevance and significance of endotoxin injection into animals[51] and I considered it totally unsuitable since it does not mimic the human situation of IAI. Moreover, endotoxins have been used to study septic shock rather than severe intra-abdominal infection and abscess formation.

2.2 Injections of live organisms with or without an adjuvant into the peritoneal cavity

It is generally felt that to produce a model of intra-abdominal sepsis mimicking the human situation that the following bacteria should be present - E. Coli, Enterococci, Bacteroides and Clostridia species[52-57]. Furthermore, there is evidence that intra-abdominal abscess formation is related to synergy between the anaerobes and facultative bacteria[57]. The aerobic bacteria play an important role in producing acute

peritonitis with its associated bacteraemia and high mortality. The anaerobes play a dominant role in abscess formation[54]. Some workers have injected single organisms like E. Coli[50,59] or Bacteriodes fragilis[60] intraperitoneally or with an adjuvant like haemoglobin to increase toxicity[61].

But these models are poor because they do not contain the appropriate number or combination of bacteria needed to mimic the human situation as discussed above. It is also felt that this type of model represents endotoxic shock, and is not a model of peritonitis and intra-abdominal abscess formation[51].

2.3 Implantation of faeces into the peritoneal cavity;

A number of different methods have been employed in this type of model. These include implantation of irradiated sterile faeces with precisely measured quantities of E. Coli and Bacteriodes[62] into the peritoneal cavity; implanting gelatine capsules containing the pooled large bowel contents of sacrificed rats[52,53,54,58,63] into the peritoneal cavity and using human faeces rather than animal faeces in gelatine capsule implants into the peritoneal cavity[65].

All three of these faecal models mimic the human situation of faecal peritonitis. There is initially acute peritonitis, E. Coli bacteraemia and high mortality.

Animals surviving the acute peritonitis stage then go on to develop intra-abdominal abscesses in which anaerobic bacteria are the predominant organisms.

I used the first method (sterile faeces plus added bacteria) in a pilot study but found it unsatisfactory as anaerobes did not seem to be able to survive the technical process involved. It was thus abandoned.

The second method is reported to work well[52,53,54,55,63] but I felt that it was a wasteful misuse of animals. It also requires dietary changes since it has been found that meat fed rats whose faeces are used for the inoculum have a lower mortality than rats fed on grain[52]. Two objections have been raised to using pooled rat faeces. Firstly, the contamination is uncontrolled because the dose and species of bacteria are impossible to quantify and thus each animal gets a different dose of bacterial inoculum. Secondly, many species of animals are resistant to their own faecal material[50].

Both these objections have been overcome by Nichols[55] who used a single sample of human faeces. Nichols froze the sample at -60°C once it had been prepared and showed that over a three week period that the fall off of the main bacterial species was insignificant.

2.4 Ischaemic organs, Caecal ligation and Bowel Perforation Models;

The ischaemic organ model consists of devascularization of a segment of intestine which is allowed to slough thus mimicking strangulation or transmural necrosis and infection.

The caecal ligation model consists of a non obstructing ligature tied around the caecum plus interruption of the blood supply of the caecum. This results in leakage of caecal contents from the necrotic caecum[64,65]. Some have found this model unsatisfactory[51]. In order to improve leakage of faeces from the caecum, needle perforations have been employed with caecal ligations[66].
A major problem with the caecal ligation model is that there is no Bacteroides fragilis in the caecum of grain-fed rats so that although the model produces acute peritonitis it does not produce abscesses in surviving animals. Thus Bacteriodes needs to be added to the contaminant of the devascularized caecum[65].

Models of colonic or small bowel laceration and leakage of intestinal contents (to cause peritonitis), have also been used[51]. These models of organ ischaemia, caecal ligation and perforation and bowel laceration have two main flaws. Firstly, the degree of infection is totally uncontrollable. The severity of peritonitis depends on

the amount of bowel necrosis or laceration, which is difficult to standardize, the rate of development of peritonitis and the bacterial contents of the bowel at the time of injury. Also bacteria may vary in species and number between animals so the infective challenge may differ from animal to animal. Secondly, one of the main principles in the policy of planned relook laparotomy is that the source of sepsis must be controlled. In the human this is usually done with a proximal stoma to divert bowel contents or by exteriorization of the site of leakage of bowel contents. In rodents, like rats, this is technically difficult and impractical. Thus one would be left with an uncontrollable source of peritoneal sepsis which would make the evaluation of this treatment modality impossible.

The model I chose was that of Nichols[66] in which a known volume of human faeces (mixed with Barium Sulphate) contained in a gelatine capsule was introduced into the peritoneal cavity of the rat. I felt that this model was the most convenient one and met the criteria mentioned earlier (see 3.3 above).

3. Experimental Design

3.1 Animals:

95 female Sprague-Dawley spf albino rats with an average weight of 201 ± 18.5 gms were used in this study. Post-operatively rats were caged individually and maintained on rat chow and water ad libitum during the post operative period.

3.2 Inoculum:

Inoculum was prepared according to the method of Nichols[55]. Human faeces was freshly collected and transferred to an anaerobic glove box. The faeces were mixed to obtain a uniform mixture and diluted 1:1 with a pre-reduced peptone: yeast extract; glucose broth. This was filtered through gauze to remove solid material. The filtered mixture was then mixed with barium sulphate (10% W/V) as described by Weinstein[52]. Aliquots of this mixture were placed in sealed containers and frozen at -60°C until used. Aerobic and anaerobic cultures were taken from the inoculum at each time of use.

Barium sulphate was added to the faeces as it is known to increase the toxicity and tissue reaction of experimentally produced faecal peritonitis[52]. Also rats have very active and efficient phagocytes in the

peritoneal cavity and barium sulphate inhibits these phagocytes[53].

3.3 Bacteriology of Inoculum:

Bacteriological analysis of the faecal inoculum was done initially to get a baseline colony count of the major bacteria. Each time further aliquots of the inoculum were used for a new group of rats one container was used to perform bacteriological analysis to ensure that the freezing, storage and thawing had not had a major effect on the numbers of the predominant bacteriological species.

All the preparation of innoculum and its bacteriological assessment and monitoring was performed by the Department of Microbiology, S.A.I.M.R.

The literature suggests that the important bacteria needed to produce a model of intra-abdominal infection which has acute peritonitis and then abscess formation and thus mimics faecal peritonitis in the human are E. Coli, Enterococci, Bacteroides fragilis, Fusobacterium species and Clostridia species[52-58]. Since this study was not a bacteriological one, I felt that it was unnecessary to identify and monitor the full range of faecal flora in the stool specimen used. It was thus decided that the organisms that would be monitored would be the key ones i.e. Coliforms (E. Coli), Enterococci, Clostridia species and Bacteroides.

Two primary faecal samples were provided to make up the inoculum for this experiment. The first sample was made up as described and then sterilized with gamma radiation. Sterility was documented with the appropriate anaerobic and aerobic cultures. This inoculum was used for the control group. The second sample was made up as described and this sample provided all the inoculum for the other groups of rats. The quantitative bacteriological analysis of the monitored flora in this sample over th period of time of the experiment is listed in table II.

TABLE II

ORGANISM	COLONY COUNTS PER GRAM OF FAECES ON DATES USED				
	14.7.89	21.7.89	25.7.89	4.8.89	16.8.89
Enterococci	$23x10^5$	$12x10^5$	$12x10^5$	$11x10^5$	$11x10^5$
Coliforms	$8x10^5$	$3x10^5$	$2x10^5$	$2x10^5$	$4x10^5$
Clostridia Sp.	$5x10^5$	$1x10^4$	$2x10^4$	$8x10^4$	$11x10^4$
Bacteroides	$1220x10^6$	$1080x10^6$	$1340x10^6$	$1280x10^6$	$980x10^6$

The changes in the colony counts of the bacteria over the period of use of the inoculum were not significant enough to make any material difference to the septic challenge to the rats in the various groups tested. These results are also confirmed by the work of Nichols[55]. The variations in colony counts especially the last counts of Coliforms and Clostridia Sp. can be due to technical factors such as: clumping of bacteria in aliquots taken

for the colony counts; a number of bacteria lying together in culture medium and only forming a single colony; errors in counting bacteria are increased when their numbers in aliquots are multiplied to express the numbers of bacteria in a standard measure of faeces.

3.4 Implantation of Inoculum;

When ready for implantation the inoculum was removed from the freezer and thawed. The required measured amount was transferred to a size 1 gelatin capsule which was then closed and put inside a sized 0 gelatin capsule. A single gelatin capsule was found to be unsuitable as it became soft and leaky very quickly after placing the inoculum mixture inside it. Each double capsule was then placed in its own sealed anaerobic bottle. It was at this time that an aliquot of the inoculum was taken for bacteriological analysis.

The rats were anaesthetized as follows. Where only a single operation had to be performed the anaesthetic agent was 0.2mℓ/kg body mass of Innovar-Vet. Where a second operation had to be performed on the same day the anaesthetic agent given was Halothane (2-4%) (by inhalation) for the first operation and Innovar-Vet for the second.

For implantation of the gelatin capsule containing the inoculum the rat was anaesthetized, the anterior abdominal wall shaved, cleaned with Hibitane solution and a small 1cm midline vertical incision was made. The peritoneal cavity was opened and the capsule introduced. The incision was closed with a single layer of continuous 2/0 silk. This operative procedure lasted 2-3 minutes. When a further laparotomy had to performed, the rat was anaesthetized as described. After cleaning the abdominal wall with Hibitane solution, the previous sutures were removed and the mid-line abdominal incision was extended upwards and downward until it was about 4cm in length. This gave adequate access to the peritoneal cavity. In groups receiving a peritoneal washout and intra-peritoneal antibiotic, the peritoneal cavity was sucked out to remove obvious faecal/barium contaminant. Then 20mℓ of sterile saline was introduced with a syringe into the peritoneal cavity to wash it out and this was also sucked out. The peritoneal cavity was than closed with continuous 2/0 silk suture. When the wound had been closed about three-quarters of its extent the intra-peritoneal antibiotic was installed via a syringe. This was not sucked out but was left in the peritoneal cavity and the remaining quarter of the abdominal incision was closed.

The antibiotic used was Cefoxitin (Mefoxin). Based on the average weight of 200 grams per rat each rat received 50mgs of Cefoxitin intra-peritoneally dissolved in 2mls of saline. This dose was well within the therapeutic range, and the saline provided fluid.

In choosing the antibiotic and its mode of administration in this experiment, the aim was to use a simple yet effective antibiotic regime. In making this decision I wanted a single antibiotic that could give acceptable gram-positive and gram-negative cover. It was felt the Cefoxitin (Mefoxin) best met these requirements. The decision to give it intra-peritoneally was based on the work of Lally[67] who using a similar rat model found that rats receiving a single lavage of intra-peritoneal Cefoxitin survived as well as rats receiving a full nine day course of parenteral Gentamycin and Clindamycin.

3.5 Monitoring of Animals;

Rats were monitored clinically and by daily weighing. Those that looked ill i.e. listless, lethargic, not feeding and who had lost >15% of their pre-operative body mass were euthanased on ethical and humanitarian grounds.

Mortalities occurring within 6 hours of capsule implantation were considered to be due to improper anaesthesia or operative bleeding as revealed at postmortem. These rats were not included in the study. All other animals were autopsied after death or were sacrificed and autopsied at 7 days post-operatively except for the last two groups where survivors lived live for 61 and 68 days respectively and were then euthanased and autopsied. At autopsy the presence and nature of free intra-peritoneal fluid was noted as well as other gross abnormalities. The presence and number of intra-peritoneal abscesses was also noted. Aerobic and anaerobic cultures of these abscesses were taken randomly from the groups.

3.6 Finding the ideal dose of inoculum to establish the animal model:

It is obviously crucial to find the most suitable dose of faecal-barium inoculum. For this reason phase one of the experiment was devoted to this purpose.

The aim was to find a dose of inoculum which would kill 70-80% of the rats over a 1 to 2 day period post-operatively. It was felt that this would most closely mimic the human situation, it would allow sufficient time for therapeutic intervention and it would enable the various therapeutic modalities to be evaluated. If the inoculum was too weak, all rats would survive and it

would be very difficult to differentiate between the groups of rats, especially as mortality was one of the factors that was monitored. Similarly, if the inoculum was too strong then the rats would die rapidly of overwhelming septicaemia and septic shock. This fact had already been demonstrated in a pilot study quoted earlier in this text.

The most suitable dose of inoculum was found by submitting four groups of rats to progressively increasing volumes of faecal-barium innoculum (0.1, 0,2, 0.3 and 0.4 mℓ) and measuring the response of these groups to this intra-abdominal septic challenge in terms of mortality, body mass changes and intra-abdominal abecess formation (found at autopsy).

3.7 Control group:

Both surgery and anaesthesia are insults that carry their own mortality and morbidity. It has also been shown that in mice that the presence of barium-sulphate in the peritoneal cavity can cause death[68]. I therefore considered it essential to evaluate the effect of surgery and anaesthesia on a group of rats to ensure that these procedures were not causing mortality by themselves. In order to evaluate the effects of these two factors a control group of rats was submitted to an anaesthetic and implantation of a gelatine capsule containing a faecal-barium mixture that had been irradiated and was thus

sterile. This control group therefore differed from the other groups only by the absence of a septic challenge.

3.6 Design of the main experiment:

The experiment was divided into two phases. The first phase was devoted to finding the ideal dose of innoculum so as to establish an acceptable and reliable animal model for the second phase of the experiment (see 3.6). The second phase of the experiment involved not only evaluating the merit of staged planned relook laparotomy as against an "operate and observe" policy but also attempted to evaluate the efficacy of the various components of these treatment modalities - namely the use of antibiotics and mechanical washout.

Thus the second phase was designed with six groups of rats. The first group was the control group (see 3.7). The second and third groups evaluated the use of antibiotic alone given at the time of the septic challenge and five hours later. The fourth group was designed to assess the value of mechanical washout alone. The fifth group evaluated the combination of anti-biotic and mechanical washout. The last group was one in which staged planned relook laparotomy was performed.

Thus overall the experiment was divided into 10 groups of rats. These were:

Phase 1: Established the ideal inoculum dose

Group 1: 0.1 ml of inoculum

2: 0.2 "

3: 0.3 "

4: 0.4 "

Phase 2: Control and Experimental Groups

Group I Irradiated faeces = Control group

II 0.4 ml inoculum and intra-peritoneal antibiotic at the same time

III 0.4 ml inoculum then intra-peritoneal antibiotic 5 hrs later

IV 0.4 ml inoculum then washout of peritoneal cavity with saline 5 hrs later

V 0.4 ml inoculum then washout of peritoneal cavity with saline and intra-peritoneal antibiotic 5 hrs later. No further treatment was done on this group. Once the peritoneal cavity had been washed out and the antibiotic given, it was just observed. This group will be referred to as the "operate and observe" group.

VI 0.4 ml inoculum then washout and antibiotic 5 hrs later (as in Group V) and then washout of the peritoneal cavity with saline at 24 hrs (relook

laparotomy). This group will be referred to as the "relook laparotomy" group.

The rationale behind the use of groups I-VI in Phase 2 merits some discussion. In the clinical context any surgical treatment of severe intra-abdominal infection whether it be on a "on demand" or planned relook basis - both antibiotics and washout of the abdominal cavity with saline will normally be used. It was thus felt necessary to evaluate these two modalities as separate entities and to try and assess their impact individually on mortality, weight, body mass changes and abscess formation. The timing of giving antibiotics was also evaluated (immediately vs after five hours). In practice antibiotic treatment is seldom started immediately as a patient usually has an unpredictable intra-abdominal crisis (e.g. a perforated diverticulum) and over a period of time develops faecal peritonitis. The surgeon usually sees the patient at some point in time after this acute crisis has started and only then is antibiotic treatment commenced. The giving of antibiotic immediately may have some application where antibiotics are given prophylactically for gastro-intestinal surgery, but this is usually only given for 3-4 doses.

Once the value of antibiotic and washout had been evaluated individually they were used together to assess their combined efficacy. This represented the situation

of "operate and observe" group. This "operate and observe" group is closest to the "on demand" relaparotomy that is used in the clinical human context. The main difference is that in the "on demand" context, the surgeon only re-operates on the patient if there are indications of ongoing intra-abdominal sepsis. In this study the "operate and observe" group was only operated on once and then observed even if there were clinical indications of active ongoing intra-abdominal sepsis (eg. weight loss).

The "operate and observe" group was then compared to a planned relook laparotomy group where the only difference between the two groups was the performance of a second laparotomy. I felt that this progression represented the most effective way of evaluating not only the value of planned relook laparotomy but also its components.

4. Statistics:

Statistical analysis was performed on the data collected. The main variables measured were mortality rate (% of group dying), changes in body mass of the rats and the number of intra-peritoneal abscesses found at autopsy. Differences in mortality and intra-abdominal abscess formation between groups were assessed using confidence intervals and the differences in changes in body mass between groups were analysed using Students t-test, a two

factor analysis of variants and Tukeys critical range test.

5. Ethical Approval

All experiments reported were approved by the Animal Ethics Committee of the University of the Witwatersrand (AEC 89/48/3).

The end point constraints placed on this experiment were

a) 7 days post inoculation (Groups 1-4 and I-IV)
b) 2 months post inoculation (Groups V and VI)
c) A loss of >15% of initial body mass
d) clinical signs of distress - e.g. lethargy and lack of grooming.

Any animal meeting any one of these criteria was withdrawn from the experiment, euthanased and autopsied. Euthanasia was performed using inhalation of 100% carbon dioxide as approved by the Animal Ethics Committee.

CHAPTER 3

ESTABLISHING THE ANIMAL MODEL

Establishment of an animal model constituted phase one of the experiment and was aimed specifically at finding the ideal dose of inoculum and establishing a reliable rat model.

In finding the ideal inoculum dose the requirements were that this dose should

- cause severe intra-abdominal infection of slow onset.
- cause a high mortality (70-80%).
- allow sufficient time for treatment to be instituted.

To find the most satisfactory dose of inoculum four doses were selected. The faecal-barium sulphate mixture was implanted intra-peritoneally into 4 groups of 10 rats each as follows:

10 rats received 0,1 mℓ of inoculum
10 rats received 0,2 mℓ of inoculum
10 rats received 0,3 mℓ of inoculum
10 rats received 0,4 mℓ of inoculum

CHAPTER 3

ESTABLISHING THE ANIMAL MODEL

Establishment of an animal model constituted phase one of the experiment and was aimed specifically at finding the ideal dose of inoculum and establishing a reliable rat model.

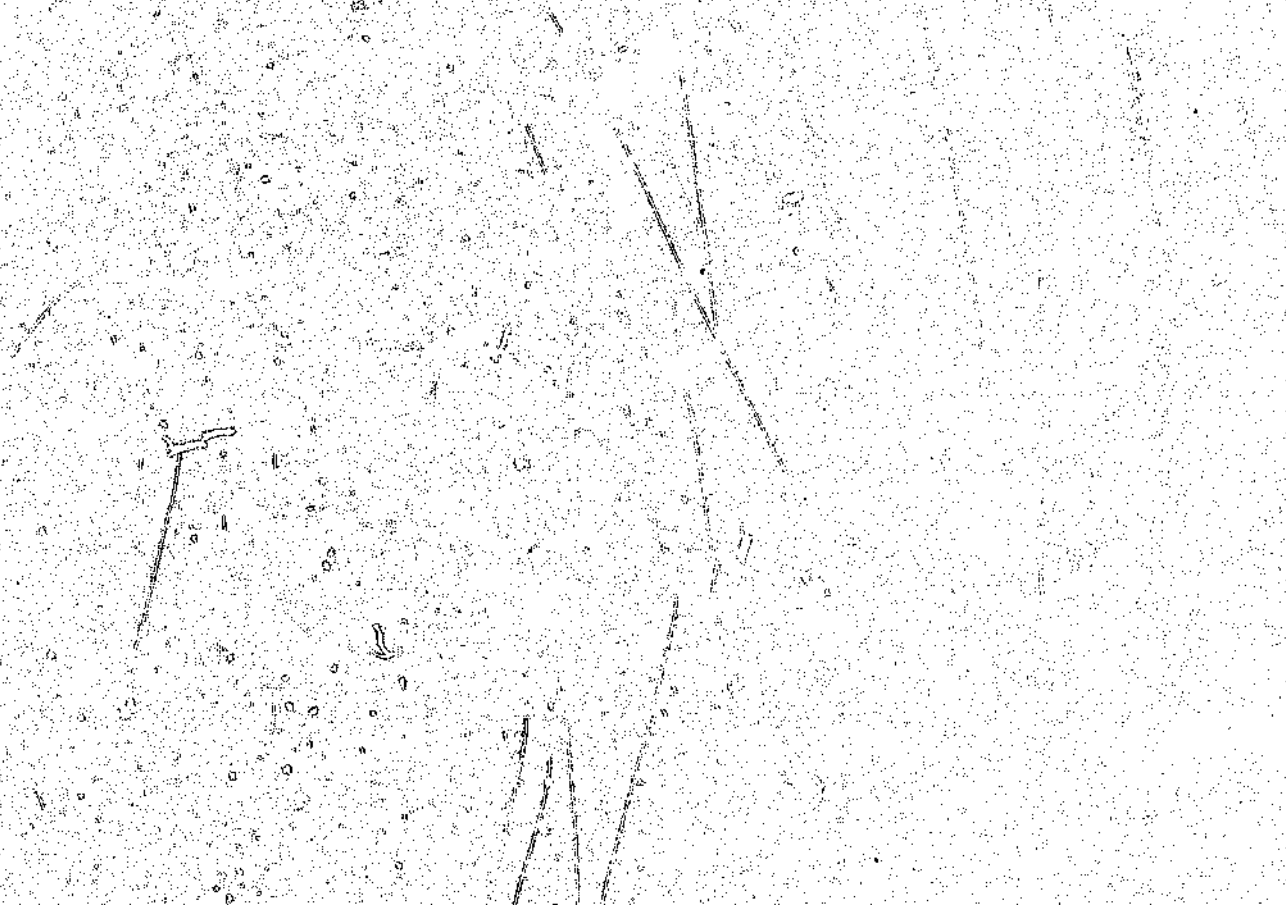
In finding the ideal inoculum dose the requirements were that this dose should

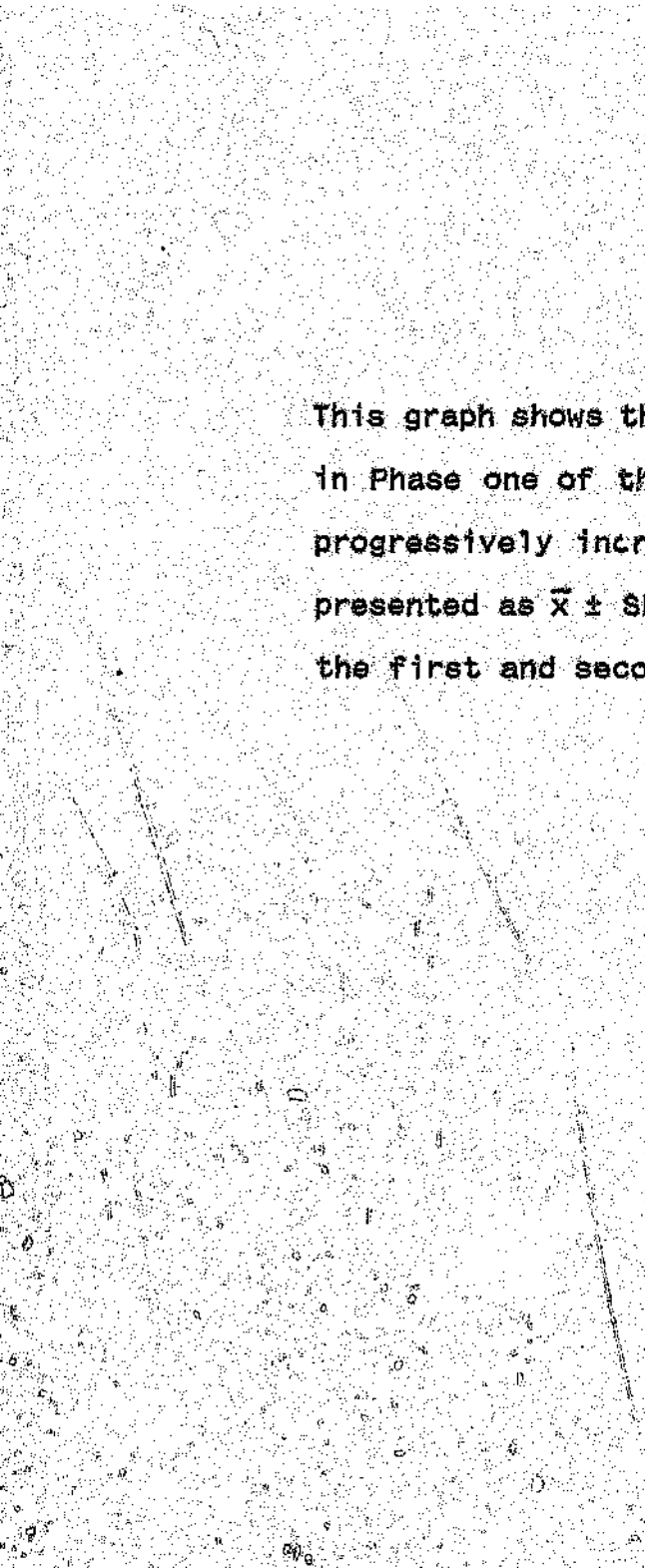
- cause severe intra-abdominal infection of slow onset.
- cause a high mortality (70-80%).
- allow sufficient time for treatment to be instituted.

To find the most satisfactory dose of inoculum four doses were selected. The faecal-barium sulphate mixture was implanted intra-peritoneally into 4 groups of 10 rats each as follows:

10 rats received 0,1 mℓ of inoculum
10 rats received 0,2 mℓ of inoculum
10 rats received 0,3 mℓ of inoculum
10 rats received 0,4 mℓ of inoculum

Figure 1

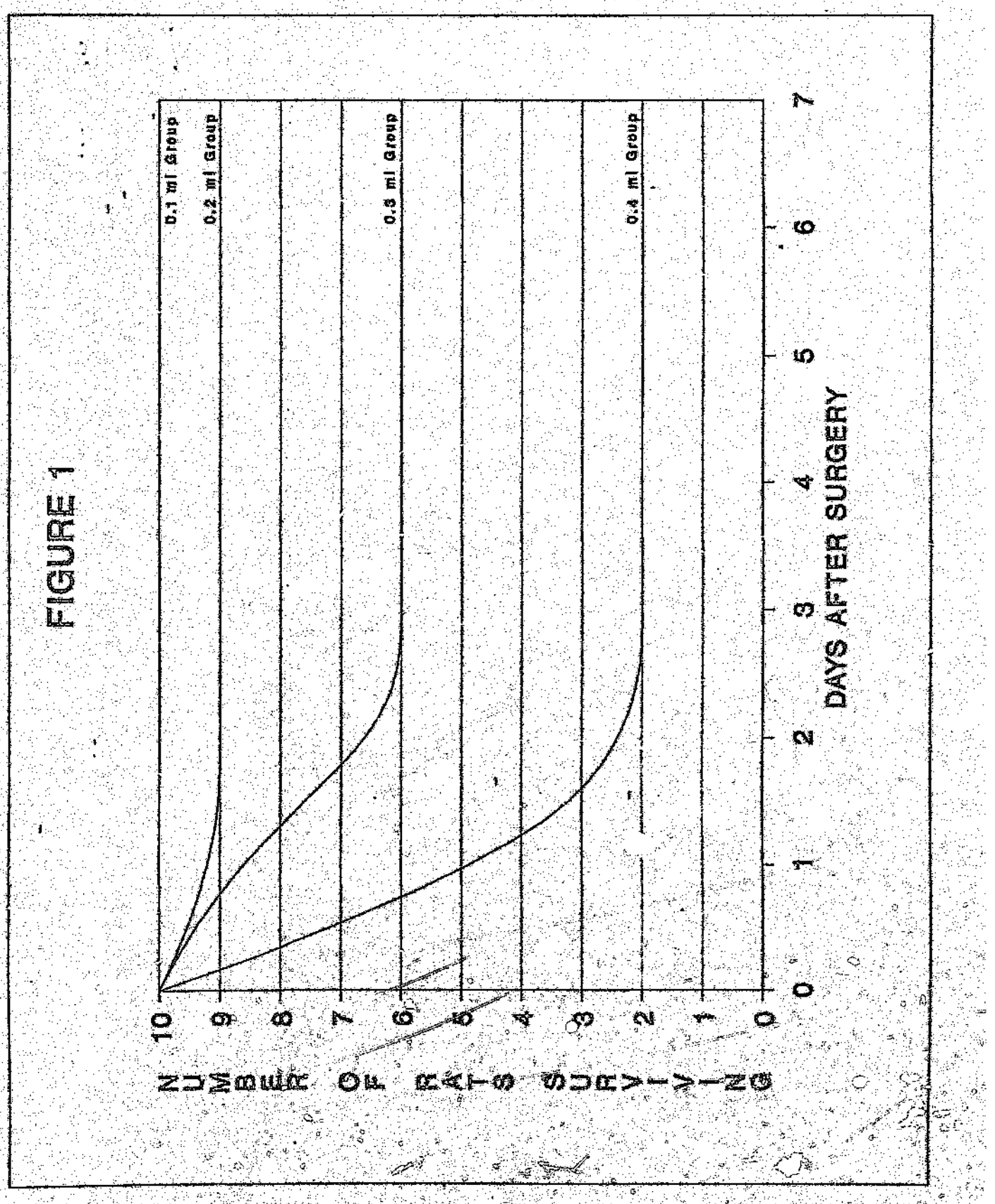




This graph shows the survival of the four groups of rats in Phase one of this experiment. Each group received a progressively increasing dose of inoculum. Results are presented as $\bar{x} \pm SE$. Note that most mortalities occur on the first and second post operative days.

val of the four groups of rats
riment. Each group received a
dose of inoculum. Results are
that most mortalities occur on
operative days.

FIGURE 1



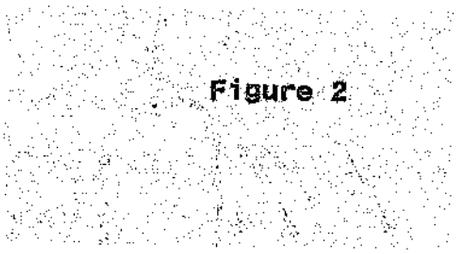


Figure 2

This graph shows changes in average body mass of the 4 groups of rats receiving graded doses of inoculum (0.1 mℓ - 0.4 mℓ). Results are presented as $\bar{X} \pm SE$. Note that body mass - reached its lowest level on the first post operative day, then increased reaching a plateau phase between days 4-6 and finally increasing again.

...ss in average body mass of the 4
...g graded doses of inoculum (0.1 ml
... presented as $\bar{x} \pm SE$. Note that
...s lowest level on the first post
...creased reaching a plateau phase
...inally increasing again.

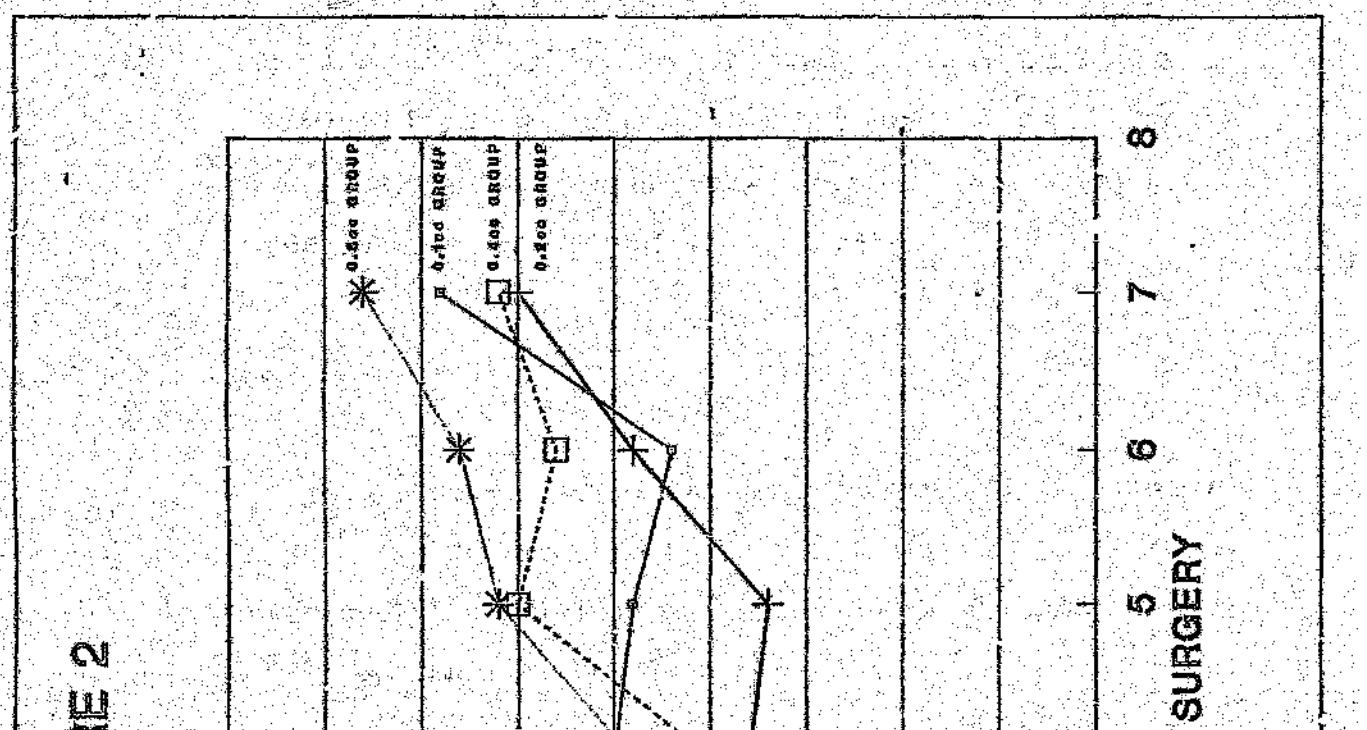


FIGURE 2

The response of the rats to the septic challenge was monitored clinically, by changes in body mass and by autopsy findings (the number of intra-abdominal abscesses) either when they died or when they were euthanased at the end of the experiment.

RESULTS:

1. Group receiving 0,1 mℓ of inoculum (0,1 mℓ group)

The results for this group are shown on figures 1 and 2 and in appendix 1.

There was no mortality in this group. All rats showed a decrease in body mass below the pre-operative values for the first two days post-operatively. The average body mass for the group reached its lowest point on the first day after surgery and was back to the pre-operative value by the third post-operative day. The average loss in body mass for the group for the first and second post-operative days was not statistically significant ($p>0.05$: t-test). By the 7th post-operative day 80% of the group had exceeded their pre-operative body mass. The average gain in body mass for the group on the 7th post-operative day was 15 g as compared to their pre-operative body mass. This was statistically significant ($p<0.05$: t-test).

At autopsy all rats showed an intra-abdominal collection of pus where the small bowel had become adherent to the inside of the incision site on the anterior abdominal wall. The rest of the abdomen in all rats was clear. The lungs were macroscopically normal.

2. Group receiving 0.2 mℓ of inoculum (0.2 mℓ group)

The results of the group are shown on figures 1 and 2 and in appendix 1.

There was a 10% mortality in this group. The rat that died did so on the first post-operative day. The autopsy on this rat showed brown intra-peritoneal fluid and congested lungs. There was no intra-abdominal abscess formation. These features were suggestive of death due to septicaemia and D.I.C. (Disseminated Intravascular Coagulopathy).

As with the previous group (0.1 mℓ) this group showed a decrease in average body mass below the pre-operative value for the first three days post-operatively. The lowest average body mass was reached on the first post-operative day. By the 4th post-operative day the average body mass exceeded the baseline weight i.e. the pre-operative body mass (as compared to the 3rd post-operative day in the 0.1cc group). Loss of body mass for the 1st, 2nd and 3rd post-operative days was not

statistically significant ($p>0.05$; t-test). It appeared therefore that an increase in dose of inoculum delayed the recovery of a gain in body mass. However 88% of the survivors were above the pre-operative body mass by the 7th post-operative day. This was better than the 0.1 mℓ group (80%). The average gain in body mass was 16 g which was statistically significant ($p<0.05$; t-test).

Autopsy studies on the survivors of this group all showed abscesses at the site of adhesion of the small bowel to the inside of the surgical incision on the anterior abdominal wall. These abscesses were all well localized. There was no free intra-abdominal sepsis and macroscopically the lungs were normal.

3. Group receiving 0.3 mℓ of inoculum (0.3 mℓ group)

The results for this group are shown on graphs 1 and 2 and in appendix 1.

Four rats died in this group giving a mortality of 40%. One of these rats died on the first post-operative day and three on the second post-operative day. At autopsy the rat that died on the first post-operative day showed an exudative faecal peritonitis and free barium in the peritoneal cavity. The lungs were congested and there was free fluid in the pleural cavity. There were no localized intra-abdominal abscesses seen. These signs suggested

death due to septicemia and D.I.C. The three rats that died on the second post-operative day all showed free faeces and exudate in the peritoneal cavity but not as much as in the rat dying on the first post-operative day.

The average body mass of the group showed a decrease below the pre-operative value for the first and second post-operative days. These decreases in weight were not statistically significant ($p>0.05$; t-test). By the third post-operative day the average body mass of survivors exceeded their average pre-operative body mass. The increase in dose of inoculum did not, therefore, seem to delay the recovery in body mass. By the seventh post-operative day all survivors were above their pre-operative body mass and the average body mass of the group was 31 g greater than the average pre-operative body mass. This was statistically significant ($p<0.05$; t-test).

Autopsy on survivors of this group performed on the eighth post-operative day showed intra-abdominal abscesses where the small bowel had become adherent to the inside of the surgical incision. Two rats had intra-omental and other abscesses. All these were well localized. No rats showed free intra-abdominal pus. The lungs were macroscopically normal.

4. Group receiving 0.4cc of innoculum (0.4 ml group)

The results of this group are shown on figures 1 and 2 and in appendix 1.

Eight rats died in this group giving an 80% mortality. Six of these rats died on the first post-operative and two on the second post-operative day. The six rats dying on the first post-operative day all showed the same findings at autopsy - purulent/faecal peritonitis with free exudate and barium in the peritoneal cavity. The lungs were congested and free fluid was present in the pleural cavity. There were no abscesses present. These findings were consistent with death due to septicaemia secondary to peritonitis.

The two rats dying on the second post-operative day showed free brown fluid in the peritoneal cavity but no abscess formation. There was mild lung congestion.

The average body mass of the group decreased below the pre-operative value for 4 days post-operatively reaching its lowest level on the first post-operative day. These losses in body mass were not statistically significant. The average body mass of the survivors exceeded its pre-operative value by the 5th post-operative day. This represented a longer delay in the recovery period of the body mass compared to the previous groups where this recovery had occurred by the third or fourth post-

operative day. By the seventh post-operative day the two survivors in the group exceeded the pre-operative body mass by an average of 17 g. So the increased inoculum dose in this group had a very significant effect on mortality (80%) and also prolonged the recovery period of average body mass of survivors.

The two survivors were euthanased on the eighth post-operative day. Autopsy showed an intra-abdominal abscess at the site where the small bowel had become adherent to the inside of the surgical incision. The lungs macroscopically were norm

TABLE III

SURVIVAL DATA OR THE FOUR GROUPS RECEIVING GRADED DOSES OF INOCULUM.

Group	Number in Stydy	Number who Survived	% Survival	95% confidence interval
0,1mℓ	10	10	100%	69 to 100%
0,2mℓ	10	9	90%	56 to 99,7%
0,3mℓ	10	6	60%	26 to 88%
0,4mℓ	10	2	20%	2 to 56%

Statistical analysis of the survival data for these four groups (See table III) showed that the 0,4mℓ group had a significantly lower percentage of survivors than the 0,1mℓ, 0,2mℓ and 0,3mℓ groups. The 0,3mℓ group also had a significantly lower survival rate that the 0,1mℓ group.

Figure 1 shows that most of the deaths occurred within the first two days of inoculation, and the rats appeared to die of overwhelming sepsis. Those that survived this acute septicaemic phase seemed to localize their intra-abdominal sepsis and at the time of autopsy were all gaining weight (See Appendix 1).

The aim of this phase was to produce a model of intra-abdominal infection with a relatively slow onset and high mortality. From the results (Table III) it can be seen that the 0,3 and 0.4 mℓ inoculum dose most closely met the requirements, producing a 40-80% mortality with deaths occurring on the first and second post-operative days. Because the 0,4 mℓ innoculum dose best fulfilled the criteria mentioned and because of the potential that treatment modalities could show increased survival and altered post-mortem findings if successful, the 0,4 mℓ dose of inoculum was used for the second phase of the experiment.

CHAPTER 4

EVALUATION OF VARIOUS TREATMENT MODALITIES INCLUDING STAGED PLANNED RE-LOOK LAPAROTOMY

Phase two of the experiment involved implanting the ideal dose of inoculum derived from phase one into six groups of rats (see Chapter 3). These groups were then submitted to different treatment modalities including staged planned re-look laparotomy. All groups consisted of 10 rats except group 1 which consisted of 5 rats. The groups were as follows:

Group I: Control Group: Received 0.4 mℓ inoculum of irradiated faeces and barium sulphate. This group was then observed.

Group II: Received 0,4 mℓ of inoculum plus 50mg (2mℓ) of Cefoxitin introduced into the peritoneal cavity at the same time as the inoculum. This group was then observed.

Group III: Received 0,4 mℓ of inoculum and then 50mg (2mℓ) of Cefoxitin introduced into the peritoneal cavity 5 hours later. This group was then observed.

Group IV: Received 0,4 mℓ of inoculum and then a washout of the peritoneal cavity with 20 mℓ of saline 5 hours later. This group was then observed.

Group V: Received 0,4 mℓ of inoculum and then a washout of the peritoneal cavity with 20 mℓ of saline plus 50mg (2ml) of Cefoxitin intra-peritoneally 5 hours later. This group was then observed.
This group is the "operate and observe" group.

Group VI: Received 0,4 mℓ of inoculum and then a washout of the peritoneal cavity with 20 mℓ of saline plus 50mg (2ml) of Cefoxitin intra-peritoneally 5 hours later. A planned re-look laparotomy was then performed 24 hours later and the peritoneal cavity washed out with 20 mℓ of saline. This group was the obeserved.
This group is the "relook laparotomy" group.

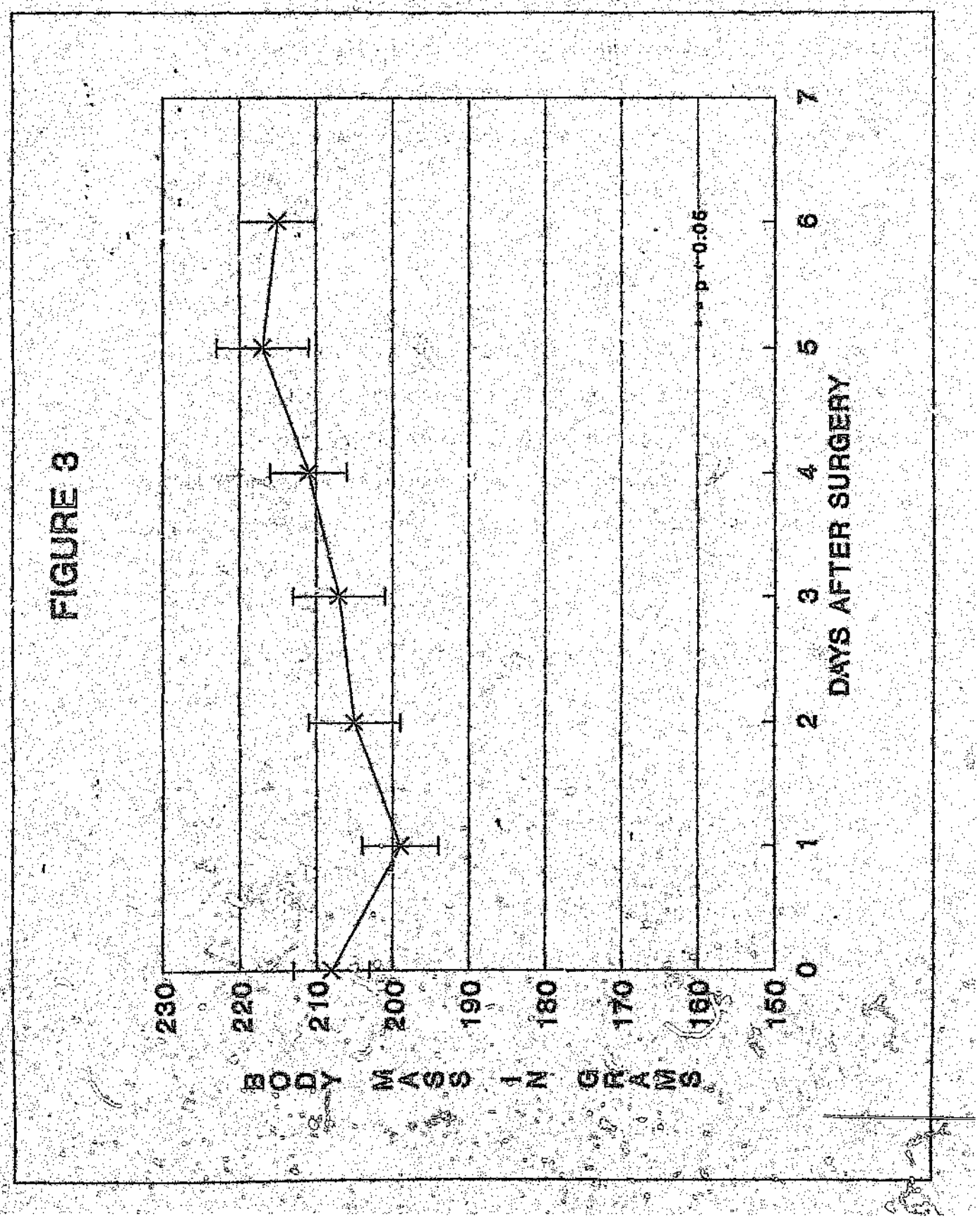
These rats were monitored in the same way as previously stated. The end point of the experiment for groups I-IV was that all survivors were euthanased and then autopsied seven days post-operatively. Those dying before this end-

Figure 3

This graph shows the changes in average body mass of 5 rats receiving irradiated barium-faecal inoculum. Results are pr sented as $\bar{X}$ ± S.E.

Note the decrease in average body mass which reaches its lowest level on the first post operative day and then increases continuously to day five.

anges in average body mass of 5
d barium-faecal inoculum. Results
:.
rage body mass which reaches its
st post operative day and then
o day five.



point were autopsied after death. Survivors in groups V and VI were allowed to live for approximately two months in order to determine more accurately any differences in mortality, body mass change and intra-abdominal abscess formation.

Results:

Group I: This group received the irradiated faecal-barium sulphate mixture, and acted as a control group to assess the effect of anaesthesia, opening of the peritoneal cavity and the implantation of a gelatine capsule containing sterile faecal-barium sulphate mixture.

The results of this group are shown in figure 3 and in appendix 1.

There was no mortality in this group. The average body mass of the group showed a decreased below the pre-operative value for three days post-operatively. The lowest body mass was reached on the first post-operative day - this was not statistically significant ($p>0.05$; t-test). The other decreases in body mass were also not statistically significant. The average body mass of the group recovered to just above its pre-operative value on the fourth post-operative day. At the time of being sacrificed all rats in this group were above their pre-

operative body mass and the group was on average seven gms (3.3%) above its pre-operative body mass. This was not statistically significant ($p>0.05$; t-test).

At autopsy one rat showed an intra-abdominal collection of barium. The peritoneal cavities of the rest of the group were clear.

Group II: This group received 0,4 ml of inoculum plus 50mg. of Cefoxitin (Mefoxin) intra-peritoneally at the same time as the innoculum.

The results of this group are shown on Figure 4 and in appendix 1.

There was no mortality in this group. The average body mass of the group showed a decrease below the pre-operative body mass for six days post-operatively. The lowest value was reached on the second post-operative day. Only the drop in average body mass on the 2nd post-operative day was statistically significant ($p<0.05$; t-test).

By the seventh post-operative day 60% of the group were above their pre-operative body mass and average body mass of the group was five gms (2.6%) above the pre-operative value. This was not statistically significant ($p>0.05$; t-test).

Figure 4

This graph shows the changes in average body mass of 10 rats receiving 0.4 mℓ of inoculum + 50 mg cefoxitin at the same time.

Results are presented as $\bar{X} \pm$ S.E.

Note loss of average body mass over first and second post-operative days followed by a plateau period of little body mass change and then a significant increase in average body mass.

e changes in average body mass of 10
mg of inoculum + 50 mg cefoxitin at

ed as $\bar{x} \pm$ S.E.

ge body mass over first and second
s followed by a plateau period of
ange and then a significant increase
ie.

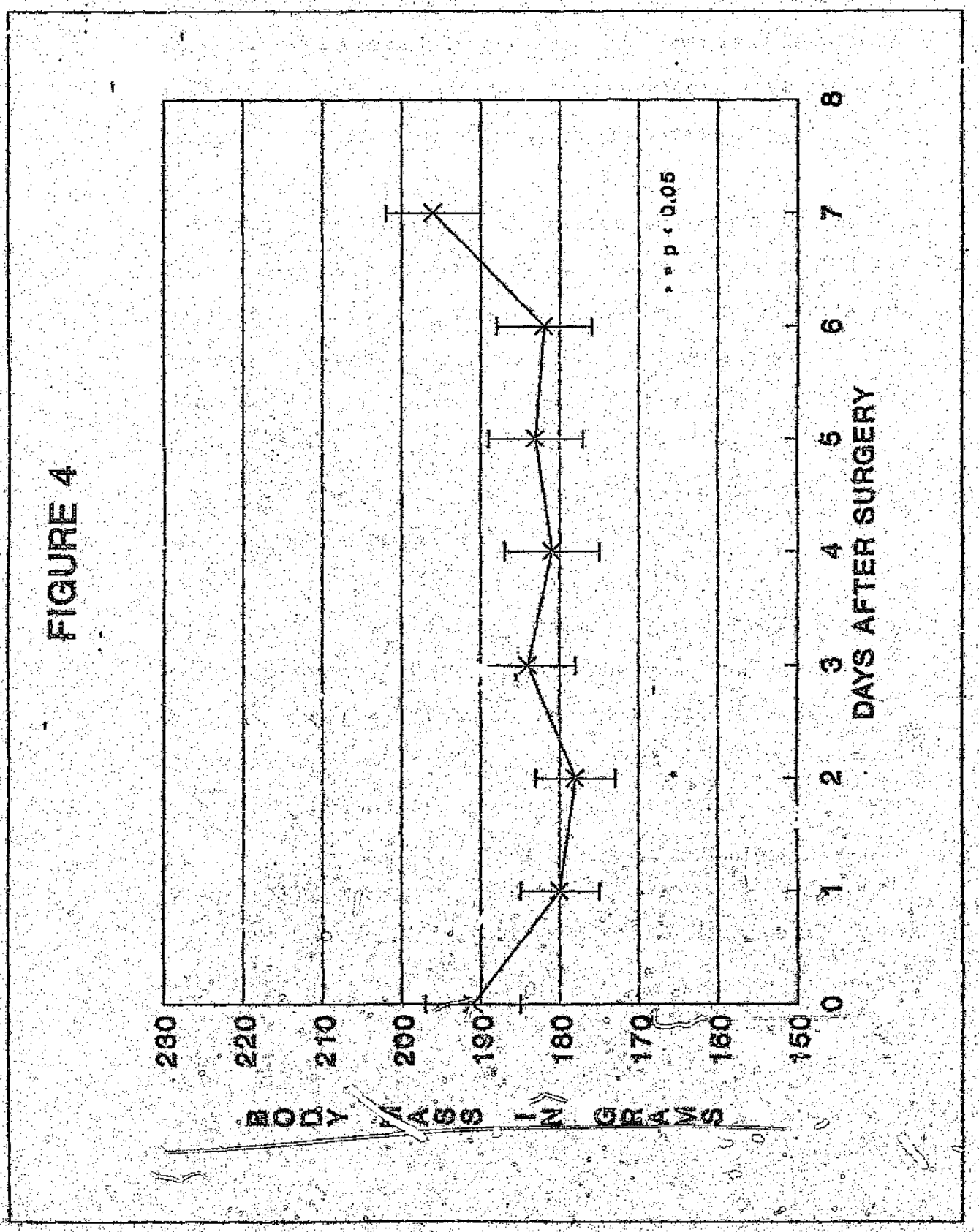


FIGURE 4

At autopsy nine rats had localized intra-abdominal abscesses: two rats had four abscesses;two rats had three abscesses; one rat had two abscesses;four rats had one abscess each; and one rat had no abscesses.

The main site of abscess formation was where the small bowel had become adherent to the inside of the abdominal wall at the site of surgical incision. Other sites of abscess formation were the postero-lateral abdominal wall and the surface of the liver. All abscesses were well localised. There was no free pus or intra-peritoneal fluid. The lungs were macroscopically normal and there was no free intra-pleural fluid.

Thus giving antibiotic immediately, significantly improved survival (100% versus 20% in 0.4 mℓ group) but did not speed up the recovery in average body mass or decrease the number of abscesses formed (as compared to the 0,4 mℓ group).

Group III: This group received 0,4 mℓ of inoculum and then 50mg of Cefoxitin (Mefoxin) intra-peritoneally five houre later.

The results of this group are shown on Figure 5 and in appendix 1.

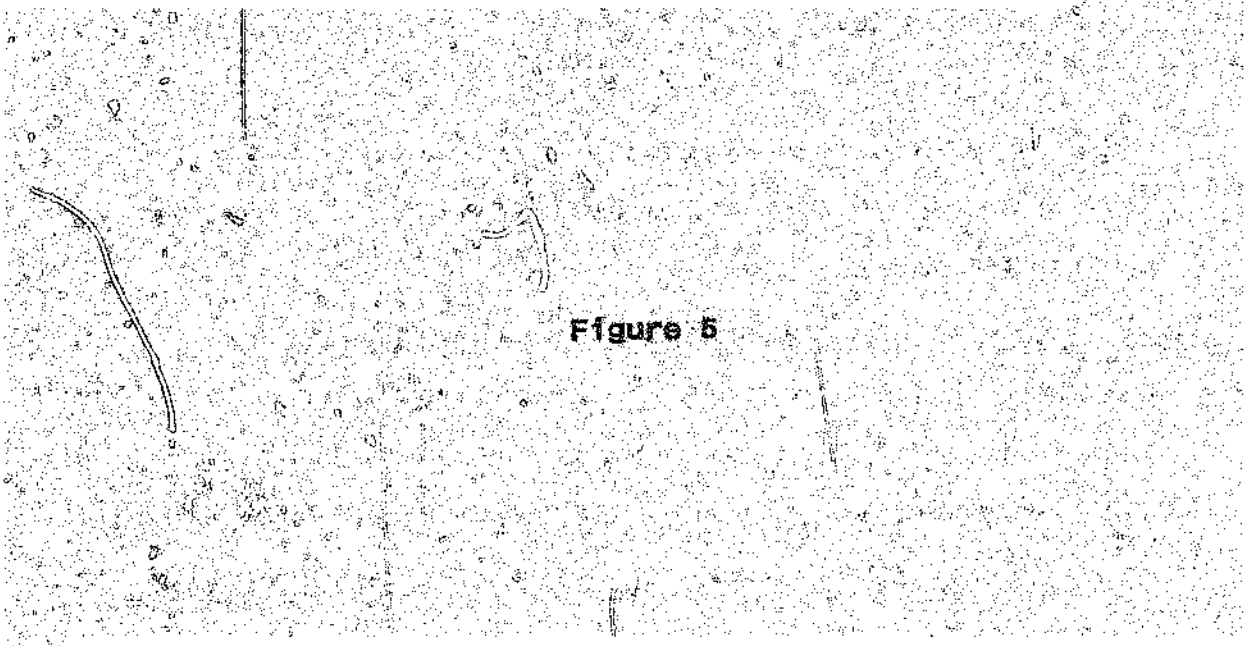


Figure 5

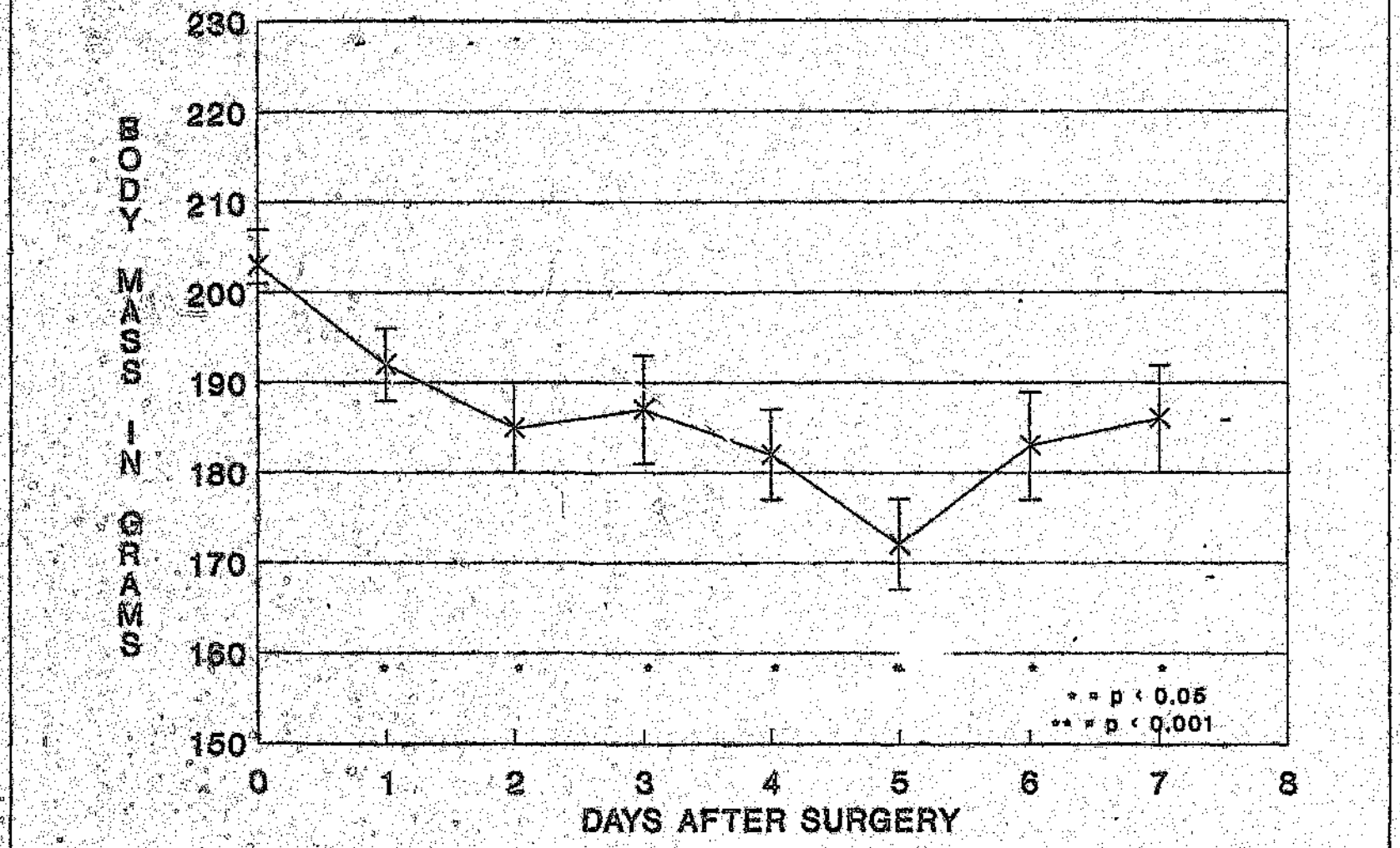
This graph shows changes in average body mass of 10 rats receiving 0.4 mℓ of inoculum then 50 mg cefoxitin five hours later.

Results are presented as $\bar{x} \pm$ S.E.

Note that there is a continuous downward trend in average body mass with its lowest value on the fifth post-operative day. The average body mass never reaches its pre-operative value.

s in average body mass of 10 rats
oculum then 50 mg cefoxitin five
as $\bar{x} \pm$ S.E.
ntinuous $\sim$-upward trend in average
owest value on the fifth post-
rage body mass never reaches its

FIGURE 5



The mortality rate of this group was 20%. One rat died on the first post-operative day. Autopsy showed free fluid and faeces in the peritoneal cavity but no abscesses. The lungs were congested and there was free fluid in the pleural cavity. These features were consistent with death due to overwhelming septicaemia and D.I.C. A second rat was humanely killed on the fifth post-operative day after a drop in body mass from 208 gm to 155 gm. Clinically this rat was lethargic and unwell. In this animal at autopsy there were three areas containing organized pus within the peritoneal cavity but no well localized abscesses as seen in previous groups. The lungs were macroscopically normal.

The average body mass of the group decreased below the pre-operative value for all seven days post-operatively reaching its lowest value on the fifth post-operative day. These decreases in body mass were all statistically significant ($p<0.05$; <0.05; <0.05; <0.05; <0.001; <0.05; <0.05 respectively: t-test). From the sixth post-operative day the average body mass of survivors began to increase but by the seventh post-operative day when the survivors where sacrificed, none had exceeded their pre-operative body mass and the average body mass of the group was still sufficiently below the pre-operative average body mass (17g or 8.3%) to make it statistically significant ($p<0.05$; t-test). Compared to the immediate antibiotic (Group II), these rats therefore fared worse

in that mortality was increased, the loss in average body mass was greater and recovery time of average body mass was more prolonged.

Autopsy of these eight survivors showed that:-

- 1 rat had a large abscess on the inside of the anterior abdominal wall and numerous smaller ones at other sites.

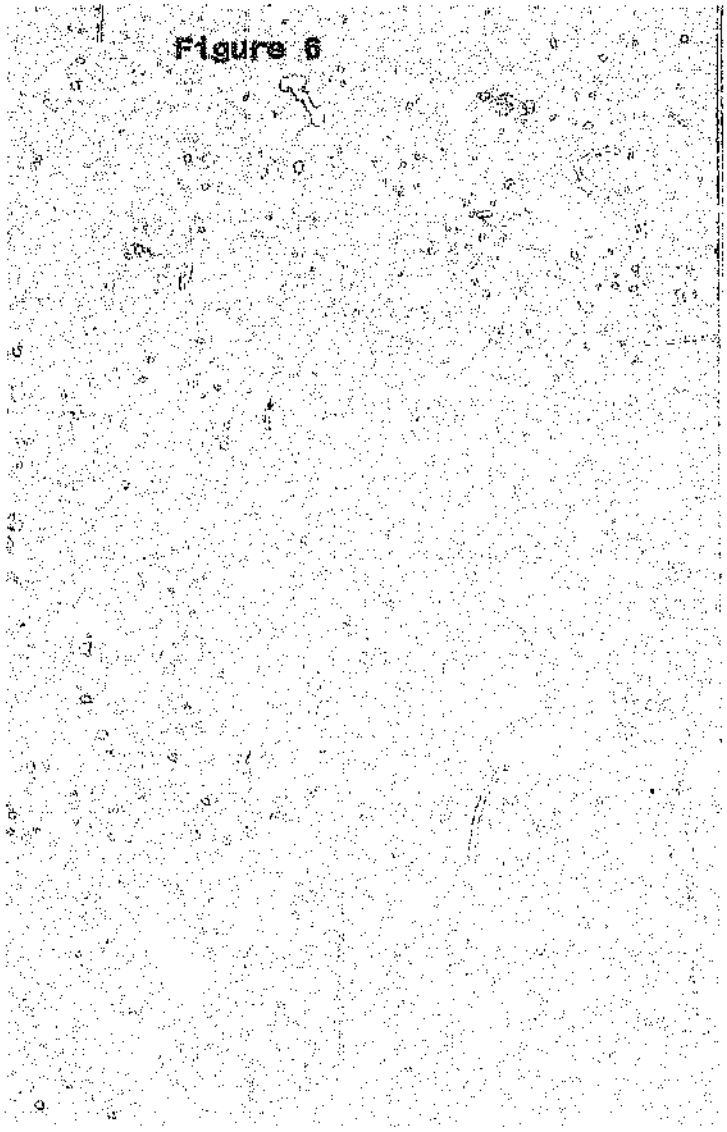
- 2 rats had two abscesses each. One on the inside of the anterior abdominal wall at the site of the incision and one on the antero-lateral aspect of the abdominal wall.

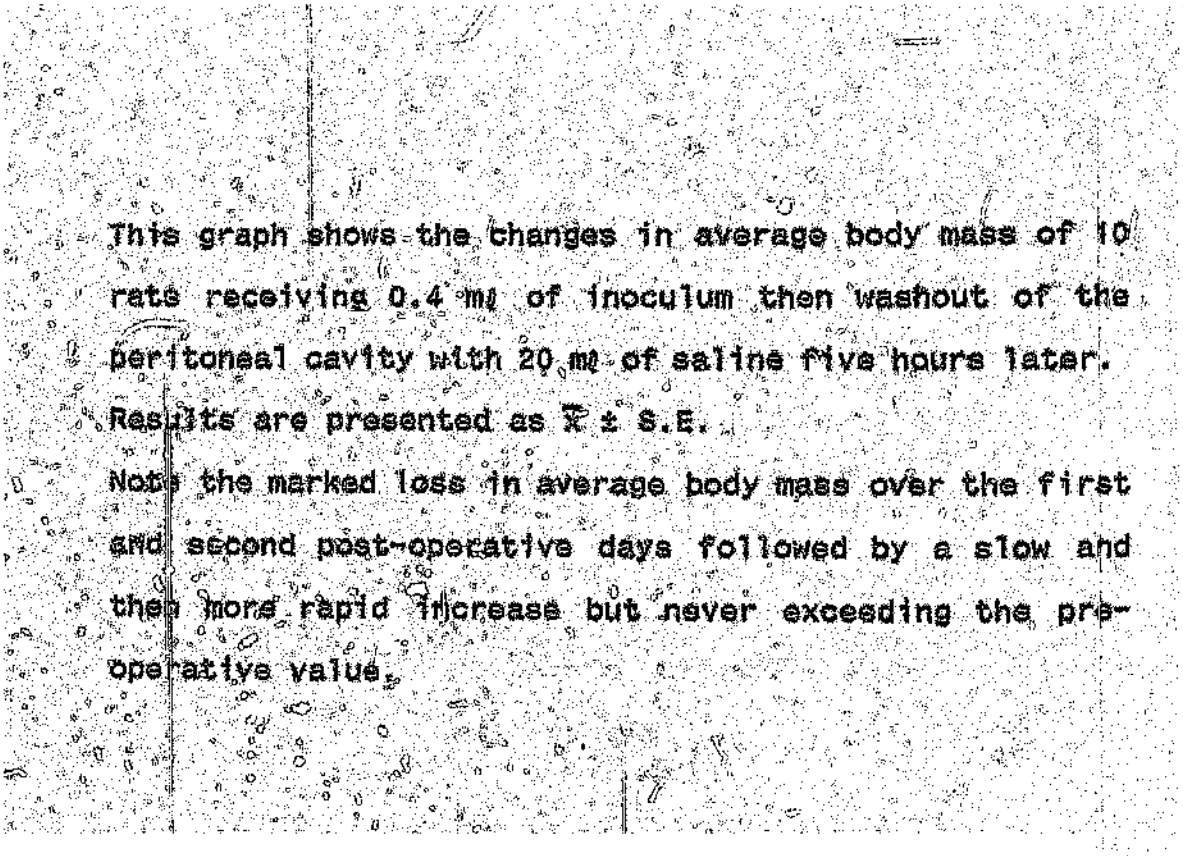
- 2 rats had an intra-abdominal abscess at the site of the incision plus an omental abscess.

- 3 rats had one abscess each at the site of the incision on the inside of the anterior abdominal wall.

None of these rats had free intra-abdominal fluid and their lungs were macroscopically normal.

Figure 6



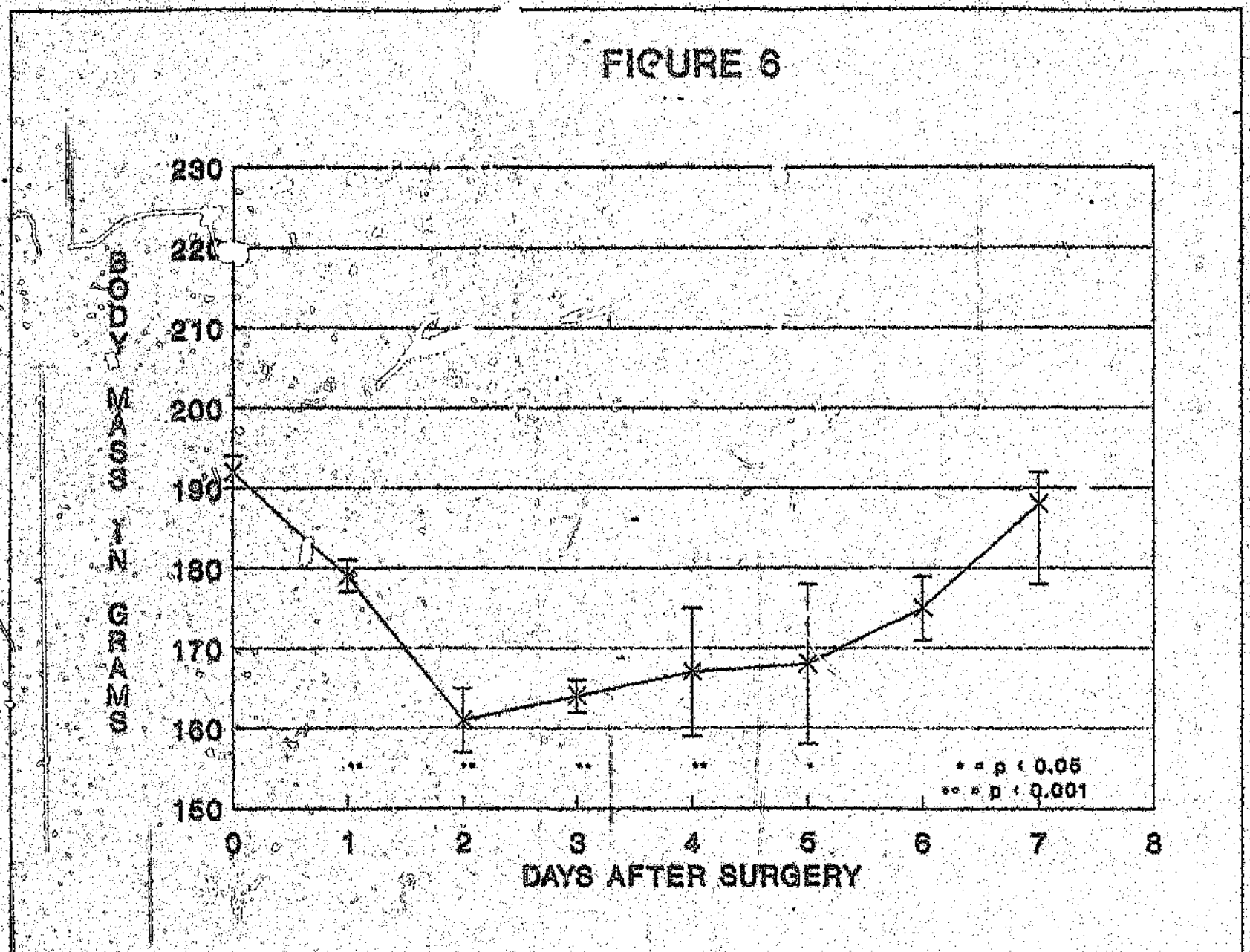


This graph shows the changes in average body mass of 10 rats receiving 0.4 ml of inoculum then washout of the peritoneal cavity with 20 ml of saline five hours later. Results are presented as $\bar{X} \pm$ S.E.

Note the marked loss in average body mass over the first and second post-operative days followed by a slow and then more rapid increase but never exceeding the pre-operative value.

changes in average body mass of 10
s of inoculum then washout of the
h 20 ml of saline five hours later.
l as $\bar{X} \pm$ S.E.
in average body mass over the first
ative days followed by a slow and
ease but never exceeding the pre-

FIGURE 6



Colony counts taken randomly from the intra-abdominal abscesses of a number of these rats showed the following:

Coliforms +++ (>50 colonies)
Bacteroides +++ (>50 colonies)
Enterococci + (0 - 5 colonies)
Clostridial species -

Delaying the administration of the antibiotic therefore appeared to increase the mortality, and increase catabolism in that the period of loss of body mass was prolonged and the recovery time of gain in body mass was delayed.

Group IV: This group received 0,4 ml of inoculum and then an intra-abdominal washout with 20 ml of sterile saline five hours later.

The results of this group are shown on figure 6 and in appendix 1.

This group had an overall mortality of 80%. Of these, five rats died on the first post-operative day, two died on the second post-operative day and one died on the fifth post-operative day. At autopsy, rats dying on the first and second post-operative day showed a small amount of faeces in the peritoneal cavity, no pus and no free fluid. The lungs were congested and there was free fluid

in the pleural cavity. Lung histology showed marked congestion with areas of intra-alveolar haemorrhage consistent with either D.I.C. or severe left heart failure. This suggested that death was due to overwhelming septicaemia. One rat in this group was found to have a perforated bowel and was thus excluded from the study. The rat that died on the fifth post-operative day had shown marked weight loss over this period and at autopsy there was free pus in the peritoneal cavity. Thus localisation of its intra-abdominal sepsis had not taken place.

The analysis of the trend in the average body mass in this group is not as accurate as previous groups because of the high mortality in the early post-operative period. The average body mass of the group dropped below the pre-operative value for all seven days post-operatively, reaching its lowest value on the second post-operative day. By the third post-operative day the average body mass showed a slight gain. The changes in average body mass over the first four post-operative days were statistically significant ($p<0.001$; <0.001; <0.001; <0.05 respectively: t-test). By the seventh post-operative day one of the two survivors had exceeded its pre-operative body mass however the average body mass of the two survivors was below that of the average pre-operative level (four gms or 2%).

At autopsy, the two survivors both showed an abscess on the inside of the interior abdominal wall where the abdominal incision had been made and where the small bowel was adherent to this area. One rat had six additional smaller abscesses in the mesentery of the small bowel and the other had four such abscesses. These were sent for histology which confirmed that they were micro-abscesses. Swabs from the abscesses found in these rats were sent for colony counts. The results were as follows:

Coliforms +++ (>50 colonies = abundant growth)
Enterococci +++(" ")
Bacteroides +++ (" ")
Clostridia species + (0-5 colonies = scanty growth)

This suggests that washout allowed more vigorous bacterial growth than did antibiotic treatment (Gp III).

Thus using washout only with no antibiotic resulted in a significantly higher mortality rate than Gp II and Gp III. The body mass changes were not significantly different from the previous group (Gp III) which received delayed antibiotic only in that it showed a marked decreaase in body mass and took longer to recover body mass than did the group receiving immediate antibiotic (Gp II).

The results from three groups (II, III and IV) are summarised in Tables IV to VII.

TABLE IV

		% SURVIVAL		
GROUP	NUMBER IN STUDY	NUMBER SURVIVING	% SURVIVAL	95% CONFIDENCE IN INTERVALS
II	10	10	100%	69 TO 100%
III	10	8	80%	44 TO 98%
IV	9†	2	22%	3 TO 60%

† = ONE RAT EXCLUDED FROM STUDY (SEE TEXT)

TABLE V

		% OF SURVIVORS WITH ABSCESSES.		
GROUP	NUMBER OF SURVIVORS	NUMBER WITH ABSCESSES	% ABSCESSES	95% CONFIDENCE INTERVALS
II	10	9	90%	56 TO 99,7%
III	8	8	100%	63 TO 100%
IV	2	2	100%	16 TO 100%

TABLE VI

COMPARATIVE BODY MASS (in grams) PRE-OPERATIVELY AND ON DAY 1, 3 AND 7 (POST-OPERATIVELY)

GROUP	PRE-OPERATIVE	DAY 1	DAY 3	DAY 7
II	191±17.9	180±16.3 (-11)	184±19.4 (-7)	196±19.3 (+5)
III	203±12	192±11.9 (-11)	187±17.2 (-16)	186±18.1 (-17)
IV	192±7	179±6.3 (-13)	164±4 (-28)	188±6.3 (-4)

TABLE VII

MEDIAN NUMBER OF INTRA-ABDOMINAL ABSCESSES IN SURVIVORS

GROUPS	NUMBER OF SURVIVORS	MINIMUM NUMBER OF ABSCESSES	MAXIMUM NUMBER ABSCESSES	MEDIAN NUMBER OF ABSCESSES
II	10	0	4	1½
III	8	1	MULTIPLE	2
IV	2	5	7	6

In terms of overall survival (see Table IV) Group IV showed a significantly lower percentage of survivors than did II and III. There were no significant differences

between the three groups in respect of the presence or number of abscesses. The small number of survivors in Group IV makes the higher median number of abscesses far less significant.

Group V: This group received 0.4 mℓ of inoculum and then five hours later an intra-abdominal washout with 20 mℓ of sterile saline was performed and 50 mg of Cefoxitin (Mefoxin) was placed intra-peritoneally.

The results of this group are shown on figure 7 and in appendix 1 and 2.

In order to better compare groups V and VI it was decided that survival and body mass changes would be compared over the first seven post-operative days and then at the end of two months.

This group had a 20% mortality. One of these rats died on the third post-operative day. At autopsy this rat showed diffuse fibro-purulent peritonitis with pus covering all abdominal organs. Multiple adhesions were present as well. The lungs were congested and the thoracic cavity contained some free fluid. This rat had lost thirty-four gms of body mass over this period. These findings are consistent with overwhelming sepsis and secondary to diffuse peritonitis. The second rat was euthanased on the sixth post-operative day due to a marked weight loss

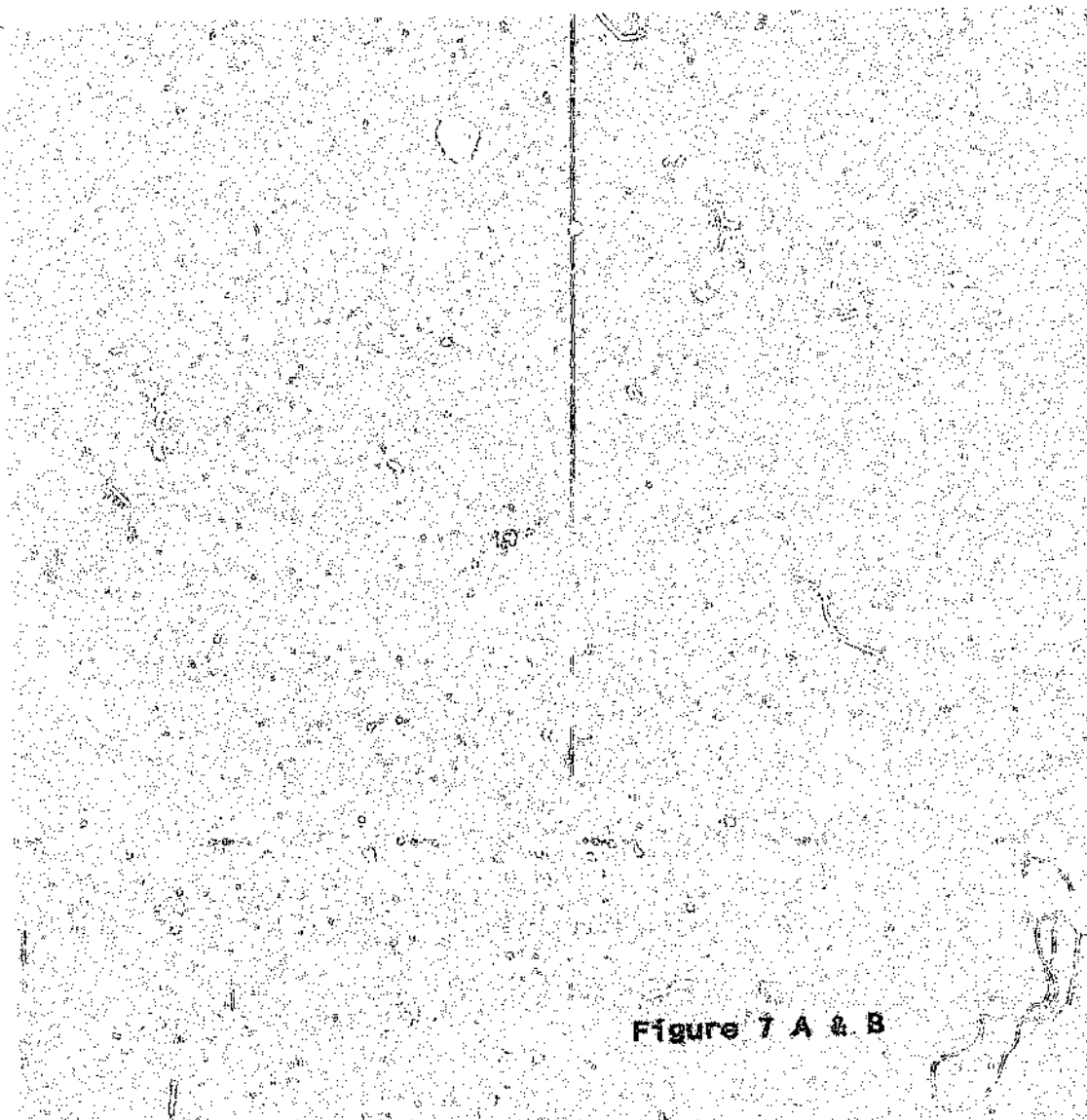


Figure 7 A & B

These graphs show the change in average body mass of 10 rats receiving 0,4 mℓ of inoculum then five hours later a washout with 20 ml of saline and 50 mg of cefoxitin. Results are presented as $\bar{x} \pm$ S.E.

Figure 7A shows the change in body mass of the group over the first seven post-operative days.

Figure 7B shows the change in body mass of the group of sixty one days.

Note the drop in average body mass reaches its lowest level on the 2nd post-operative day. This is then followed by a steady increase.

ange in average body mass of 10
inoculum then five hours later
saline and 50 mg of cefoxitin.
$\bar{x}$ ± S.E.
e in body mass of the group over
ative days.
e in body mass of the group of

body mass reaches its lowest
operative day. This is then
ease.

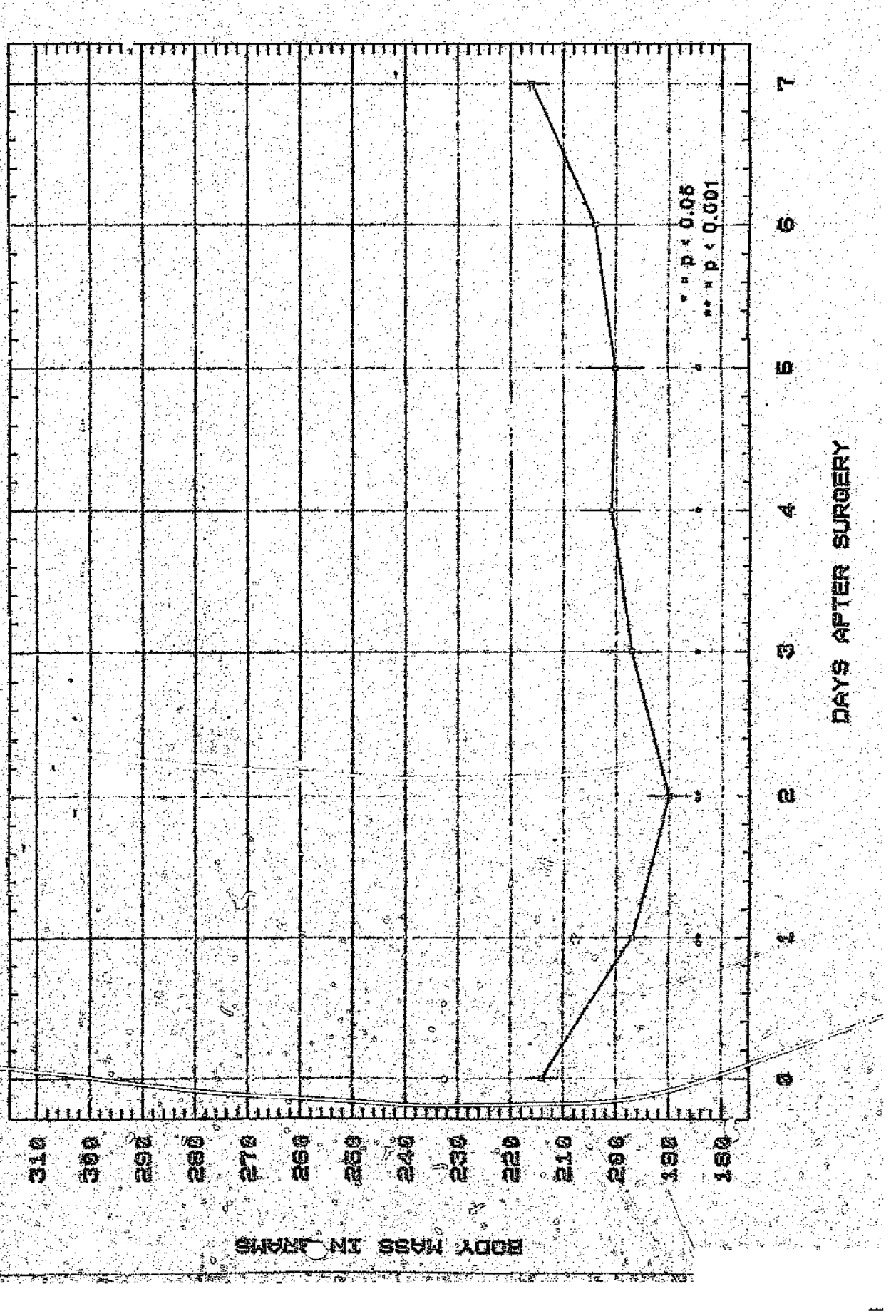
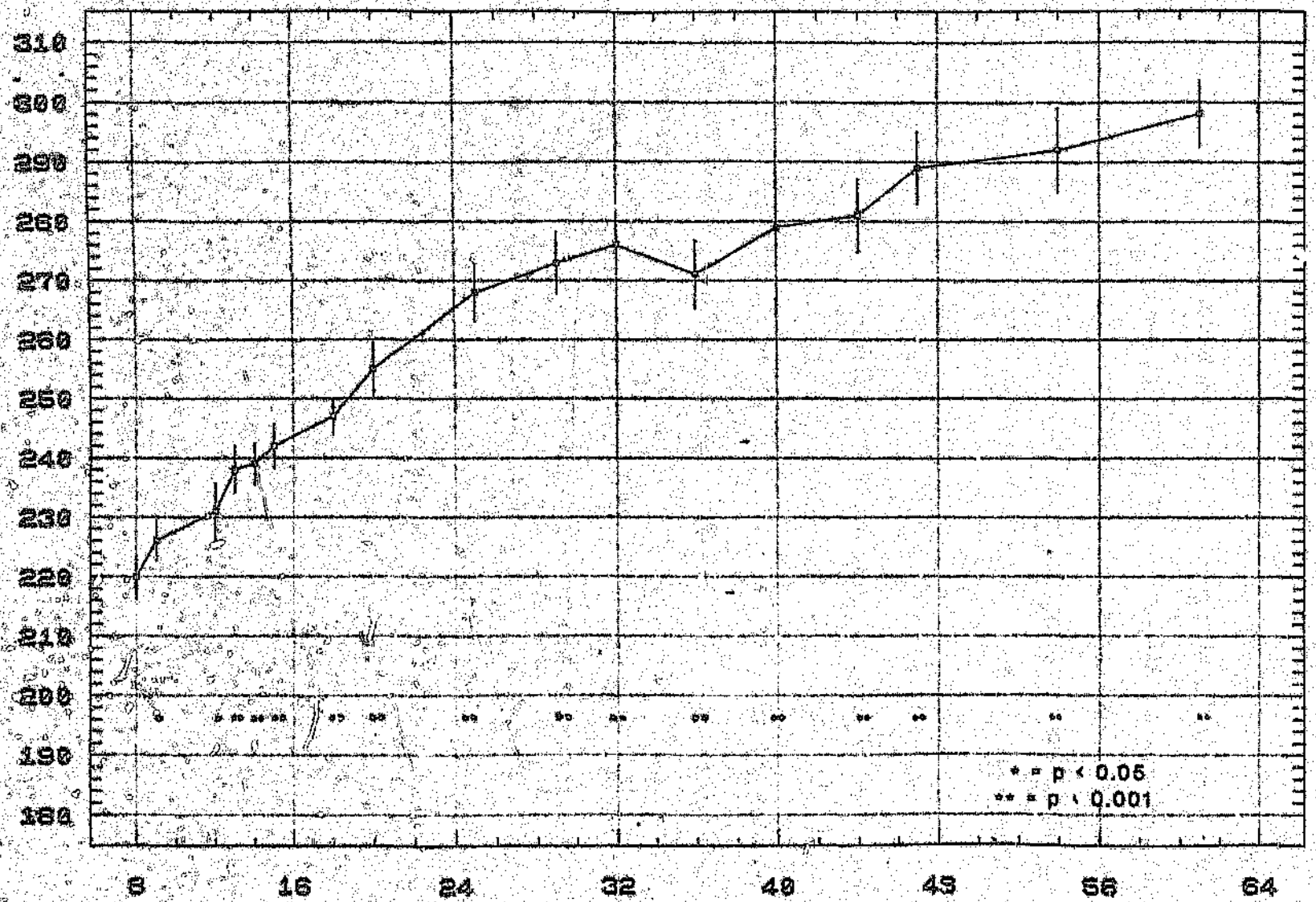


FIGURE 7a

FIGURE 7b


BODY MASS IN GRAMS
310
300
290
280
270
260
250
240
230
220
210
200
190
180
8
16
24
32
40
48
56
64
* = p < 0.05
** = p < 0.001

(fifty-five gms). At autopsy there were intra-abdominal adhesions and multiple small abscesses (± 10) among loops of bowel and on the inside of the abdominal wall. The lungs were macroscopically normal. It was clear that this rat had not managed to localize and overcome the intra-abdominal sepsis. Although this rat had overcome the initial period of the septic challenge and may have survived if left alone, it was euthanased because it had reached previously established end points (see Chapter 2).

The average body mass of the group showed a decrease below the pre-operative value for the first six days post-operatively, reaching its lowest value on the second post-operative day. These losses in body mass were statistically significant for the first five post-operative days ($p<0.001$; <0.001; <0.05; <0.05; <0.05 repectively: t-test). From the third post-operative day the average body mass started increasing and exceeded its pre-operative value for the first time by the seventh post-operative day (by two g or 0.9%). This value was not statistically significant ($p<0.05$; t-test). The average body mass of the survivors in the group then increased steadily and by the end of the sixty first post-operative day was eighty four gms or 39% greater than the pre-operative body mass. All body mass measurements made after the seventh post-operative day to the sixty first post-operative day showed values that were statistically

significantly greater than the pre-operative value ($p<0.001$; t-test).

The survivors of this group (8 rats) were euthenased on the sixty first post-operative day. At this time they were all in good condition. At autopsy no abscesses were found intra-abdominally. Occasional barium sulphate granulomas were noted and there were adhesions of the small bowel to the site of surgical incision on the inside of the anterior abdominal wall.

The combined use of antibiotic and washout had, therefore, a marked improvement on survival compared to the group where washout only was used. There was also less intra-abdominal abscess formation compared to the groups were antibiotic only was used. The combined use of antibiotic and washout also increased the rate of recovery of body mass (by the seventh post-operative day) as compared to the delayed use of antibiotic and the use of washout only (loss in body mass had not recovered by the seventh post-operative day in both these groups).

Group VI: This group received 0,4 mℓ of inoculum intra-peritoneally; five hours later the abdomen was washed out with 20 ml of sterile saline and then 50 mg of Cefoxitin (Mefoxin) was given intra-peritoneally. 24 hours later a planned re-look laparotomy was performed.

The results of the group are shown in figure 8 and in appendix 1 and 2.

This group had a 10% mortality. The rat that died did so on the first post-operative day after the re-look laparotomy. At autopsy the lungs were normal and the abdomen showed flimsy fibrinous adhesions but there was no free pus and no abscess formation.

This rat thus showed no signs of acute overwhelming septicaemia or chronic intra-abdominal sepsis. It was thus assumed that this death was related to the stress of two anaesthetics, and it emphasises a problem of a policy of planned re-look laparotomy i.e. submitting patients to multiple anaesthetics.

The average body mass of the group showed a decrease below the pre-operative value for the first five post-operative days (after the day of initial surgery) reaching it lowest value on the second post-operative day. Thereafter the average body mass of the group started increasing but only exceeded the average pre-operative body mass on the sixth post-operative day (after the initial surgery) by two g or 0.93%. The decrease in average body mass was statistically significant for the 2nd post-operative day only (after

Figure 8 A & B

These graphs show the changes in average body mass in 10 rats receiving 0.4 mℓ of inoculum then a washout of the peritoneal cavity with 20 mℓ of saline + 50 mg of cefoxitin five hours later

Twenty four hours later a planned relook laparotomy was done and the peritoneal cavity washed out with 20 mℓ of saline.

Results are presented as $\bar{X} \pm$ S.E.

Figure 8A shows the changes in body mass of the group over the first seven post-operative days.

Figure 8B shows the changes in body mass of the group over sixty eight days.

changes in average body mass in 10
of inoculum then a washout of the
ith 20 mℓ of saline + 50 mg of
later

ter a planned relook laparotomy was
al cavity washed out with 20 mℓ of

as $\bar{X} \pm$ S.E.

changes in body mass of the group
post-operative days.

changes in body mass of the group

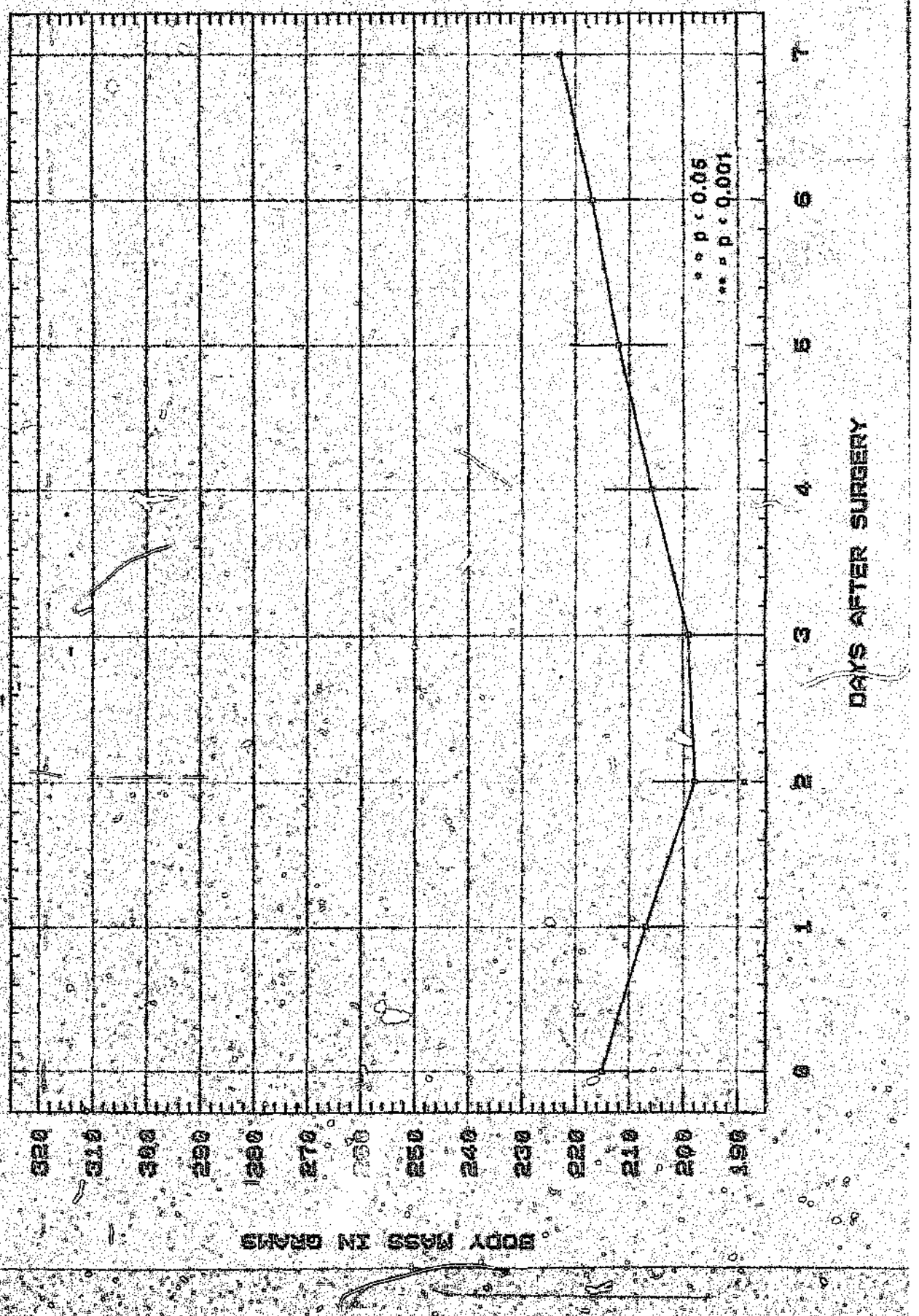


FIGURE 8a

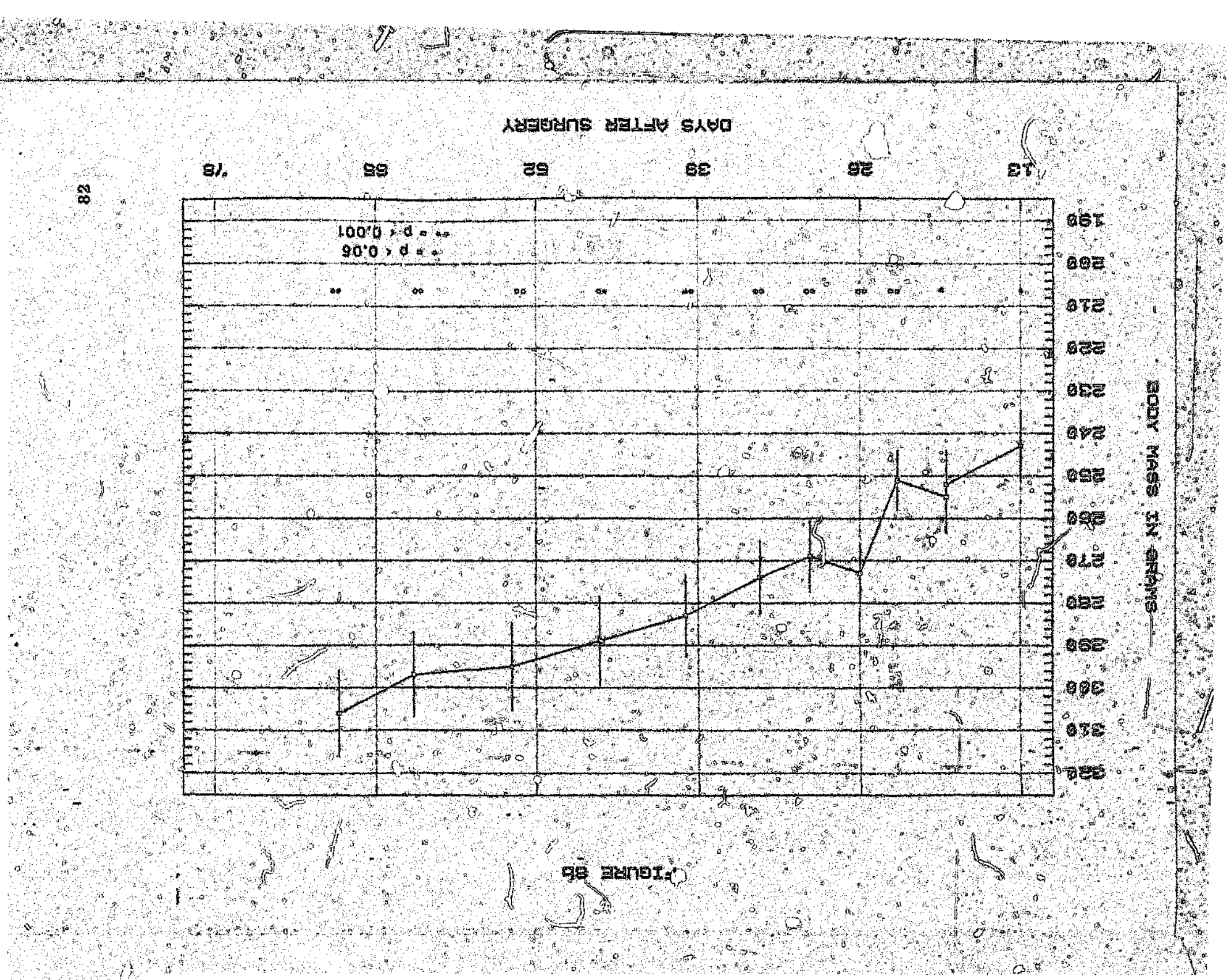

DAYS AFTER SURGERY
13
26
39
52
65
78
91
BODY MASS IN GRAMS
190
200
210
220
230
240
250
260
270
280
290
300
310
320
* = p < 0.05
** = p < 0.001

FIGURE 8b

initial surgery) ($p<0.05$; t-test). From the sixth post-operative day onwards to the sixty eighth post-operative day the average body mass of the group increased progressively and at the time of euthanasia the average body mass of the group was ninety one gms or 42% higher than the pre-operative body mass. This increase was statistically significant ($p<0.001$; t-test).

The planned re-look laparotomy did not retard the recovery in the average body mass of the group, in fact body mass recovered a day quicker compared to group V. In both these groups (V and VI) the lowest body weight was reached on the second post-operative day (after initial surgery). This decrease in average body mass was 11% in group V and 8% in group VI, again showing that the planned re-look laparotomy had virtually no effect on the average body mass of the group.

Survivors of this group were euthanased on the sixty eighth post-operative day. Autopsy showed that all animals had mild fibrinous adhesions between bowel and other intr-abdominal organs. However one rat did have a large 0.5 ml mesenteric abscess filled with caseous pus. This was somewhat unexpected as one would have expected the planned re-look laparotomy to have prevented this.

Groups V and VI were compared as their treatment differed only in respect of a re-look laparotomy performed in Group VI, and the comparison is shown in Table VII, IX and X.

TABLE VIII OVERALL SURVIVAL

GROUPS	NUMBER IN GROUP	NUMBER OF SURVIVORS	% SURVIVORS	DIFFERENCE	95% CONFIDENCE INTERVALS
V	10	8	80%	10	-21 TO 40%
VI	10	9	90%		

From Table VIII it can be seen that there is a difference between the two groups of only 10% in terms of overall survival and this was not statistically significant.

TABLE IX % OF SURVIVORS WITH INTRA-ABDOMINAL ABSCESSES

GROUPS	NUMBER OF SURVIVORS	NUMBER OF SURVIVORS WITH ABSCESSES	% OF SURVIVORS WITH ABSCESSES	DIFFERENCE	95% CONFIDENCE INTERVALS
V	8	0	0%	11%	-9 TO 32%
VI	9	1	11%		

Table IX shows that there was an 11% difference between the two groups in terms of abscesses formation in surviving rats. This was found not to be statistically significant.

TABLE X COMPARATIVE BODY MASSES (in grams) PRE-OPERATIVELY AND ON DAY 1, 3, 7 AND 61/62 (POST-OPERATIVELY)

GROUP	PRE-OPERATIVE	DAY 1 POST-OPERATIVE	DAY 3	DAY 7	DAY 61/62
V	214±9	197±9 (-17)	197±16 (-17)	216±9 (+2)	298±15 (+84)
VI	215 ±25	207±24 (-8)	199±23 (-16)	223±24 (+8)	297±30 (+82)

A two factor repeated measures analysis of variants was used to determine whether the two groups experienced significant body mass changes over time and whether these body mass changes differed between the groups. Specific differences in body mass were assessed using Tukey's critical range test. Since the days following surgery on which measurements were made differed for the two groups the analysis was carried out on: 1) the twelve days on which body mass was measured on both groups 2) the seventeen days on which body mass was measured within one day apart in both groups.

In addition since there may be bias introduced by the animals who died during this study the analysis was carried out on: 1) all twenty animals and 2) the seventeeen survivors only.

Thus the analysis was carried out on the four combinations of data described above. Results of all four analyses (see appendix 2) showed no significant differences between the groups in their body mass change over time. Both groups showed a significant body mass loss followed by a significant body mass gain (See appendix 2 and 2 and Figures 7 and 8).

TABLE XI. CRITICAL CHANGES IN BODY MASS OVER THE FIRST 7 POST OPERATIVE DAYS

	Group II	Group III	Group IV	Group V	Group VI
Lowest body mass reached	2nd post operative day -13g or 6.8% (p<0.05)	2nd post operative day -18g or 8.8% (p<0.05)	2nd post operative day -31g or 16% (p<0.001)	2nd post operative day -24 g or 11% (p<0.001)	2nd post operative day -17g or 1.9% (p<0.05)
Gain in body mass started	3rd post operative day (p>0.05)	3rd post operative day (p<0.05)	3rd post operative day (p<0.001)	3rd post operative day (p<0.05)	3rd post operative day (p>0.05)
Rate of gain/ loss	0.71 g/day	2.4 g/day	0.57 g/day	0.28 g/day	1.1 g/day
Reached/ Exceeded Pre operative body mass	By 7th post operative day	Not by 7th post operative day	Not by 7th post operative day	By 7th post operative day	By 6th post operative day
Body mass at 7th post operative day relative to pre operative body mass	+6 g or 2.6% (p>0.05)	-17 g or 8.3% (p<0.05)	-4 g or 2%	+ 2 g or 0.9% (p>0.05)	+8 g or 3.72% (p>0.05)

CHAPTER 5

DISCUSSION AND CONCLUSIONS

5.1 DISCUSSION

This study set out to establish a model of IAI and to assess the efficacy of various treatments and combinations of treatments on survival rate, intra-abdominal abscess formation and health (as measured by changes in body mass of the rats).

As regards the establishment of the model of IAI, the animal model that was chosen proved to be very satisfactory. Rats were easily available, a model of intra-abdominal infection in the form of faecal peritonitis was reliable and easily reproducible. It also successfully produced the acute (faecal peritonitis) and chronic stages (abscess formation) of IAI.

As previously discussed the high risk types of IAI are felt to be those due to pancreatic sepsis, anastomotic dehiscence following surgery and faecal peritonitis. In choosing the model to be used I felt it necessary to reproduce one of these types preferably the one that was easiest to do. The faecal peritonitis model best fitted the desired requirements. Faecal peritonitis is also the one form of IAI that Schein[1] felt might possibly benefit

from a policy of planned re-look laparotomy. In addition the other two forms of IAI would have been very difficult to reproduce in the rats. There is no literature known to me that describes an animal model of IAI due to pancreatic sepsis and it is difficult to envisage how, even if it could be done, the amount of sepsis could be controlled so that each rat would be exposed to the same septic challenge. The model of IAI due to anastomotic dehiscence is also impractical in rats. In the human where severe intra-abdominal infection is encountered, because of a disrupted colorectal anastomosis for example, most surgeons would convert this to a diverting colostomy to control the source of sepsis. This type of procedure would not be possible in a rat model.

My results show that the insertion of 0.4 ml of human faecal barium sulphate inoculum produced a high mortality (80%) and relatively slow onset type of intra-abdominal infection, thus allowing adequate time and scope to assess the efficacy of the various treatment modalities on the measured parameters (mortality, body mass and abscess formation).

My study clearly underlines the importance of antibiotics in the treatment of severe intra-abdominal infection. The use of antibiotics has its most important effect on survival rate. There is a marked increase in survival when antibiotic is used at the time of the septic

challenge (100% survival) as compared to its use after a time delay of five hours (80% survival) and when it is not used (22% group IV and 20% in 0.4cc inoculum group).

The main role of antibiotics seems to be in preventing overwhelming sepsis in the early period of the septic challenge. Those rats not receiving antibiotics die from overwhelming sepsis within the first day or two after surgery (Table III - 0.4 ml group and Table IV - group IV). Those receiving antibiotics seem to be able to survive the initial period and if they do so they have a good long term prognosis. The earlier the antibiotic is administered the better the survival (Table IV - group II and III and Table VIII - group V and VI) as it appears to modify or reduce the severity of sepsis arising from the peritoneal cavity.

The use of antibiotics does not however seem to protect against the development of intra-abdominal abscesses (see Table V groups II and III). Moreover, whether antibiotics are given at the time of soiling or five hours later does not appear to prevent the development of abscess formation (see Table V group II versus group III) or lower the median number of abscesses that develop (see Table VI group II - 1½; group III - 2 abscesses). Antibiotic does seem to have an affect on the body mass. Comparing the group not receiving antibiotic (group IV), the groups receiving delayed antibiotics (group III, V

and VI) and the group receiving immediate antibiotics (group II), all reach their lowest average body mass on the second post-operative day (see Table XI). However the largest percentage weight loss was in the group not receiving antibiotics (group IV - 16%). By the seventh post-operative day all groups had exceeded their average pre-operative body mass except for the group not receiving antibiotic (group IV) and the group receiving delayed antibiotic but no washout (group III). So it appears that giving antibiotic may limit the amount of body mass that is lost and may hasten the recovery period from the loss in body mass.

In contrast to the literature, this study suggests that there is a role for mechanical washout of the peritoneal cavity in the treatment of the septic abdomen.

The data shows that mechanical irrigation of the peritoneal cavity has little or no effect on improving survival. The lowest survival rate in the second phase of the study occurred in the group that had a mechanical washout only 22% (see Table IV group IV). This survival rate was significantly lower than in the group receiving antibiotics and no washout (see group II and III - Table IV) and lower than in those groups receiving antibiotic and washout (see group V and VI - Table VIII).

However the use of mechanical washout seems to reduce the incidence of the development of intra-abdominal abscesses. In the groups which had antibiotics but no

washout (group II and III - Table V) virtually all survivors had intra-abdominal abscesses whereas in groups which had antibiotics and washout only one of the survivors of both these groups had an intra-abdominal abscess (group V and VI - Table IX). Two important effects of I.A.I. are early mortality (survival) from septicaemia and intra abdominal abscess formation. The apparent contradiction that mechanical washout does not improve the former but does the latter can be explained by the fact that mechanical washout was used 6 hours after the septic innoculum was administered. This allowed overwhelming septicaemia to develop which resulted in the high mortality (low survival) (group IV). Only antibiotic can favourably affect this aspect of intra-abdominal sepsis. When antibotic and washout were used together (group V + VI), the antibiotic countered the septicaemia and allowed the mechanical washout to decrease the intra-abdominal abscess formation.

My results also suggest that washout had a beneficial effect on metabolic processes. If the variation in body mass of the group receiving a delayed dose of antibiotic only (group III) is compared to that in the group receiving a delayed dose of antibiotic plus a mechanical washout (group V) then it is noticeable that both these groups reach their lowest average body mass on the second post-operative day and the percentage loss of body mass at this point was very similar (8.8% versus 11%). Both groups started their weight gain on the third post-operative day, however the group not receiving the mechanical washout (group III) did not reach it pre-operative body mass by the seventh post-operative day, having on average a body mass some seventeen g lower at this time, a mass which was significantly lower. The group receiving the delayed dose of antibiotic plus a mechanical washout exceeded its pre-operative body mass by two g on the seventh post-operative day.

In the clinical context, many surgeons believe that washout is beneficial but there is surprisingly little solid scientific evidence despite the publications on the subject. The controversy can be summed up in two

conflicting statements. In 1974 Maingot declared in his textbook "irrigation of the peritoneal cavity for cleaning purposes is, in my opinion, never justified even in the presence of gross faecal contamination[69]. Another leading surgical textbook advises that the control of faecal contamination "should be accompanied by copious irrigation of the peritoneal cavity"[70].

Many of the experimental studies on the subject of the efficacy of mechanical intra-abdominal peritoneal lavage can be criticized[71].

There are only two clinical trials reported on washouts. One[72] concerned the use of saline intra-peritoneal lavage in elective colorectal surgery. It showed that the peritoneal washout did decrease the bacterial counts in the abdominal cavity but the rates of peritoneal abscesses (26%) and septicaemia (13%) were very high and did not correlate with the bacterial counts. This study did not use prophylactic antibiotics. The other study[73] was a prospective randomized study which compared mechanical intra-operative lavage with simple suction of the peritoneal contamination, in two groups of patients. Both received prophylactic antibiotics. There was no difference in septic complications or mortality rates between the two groups.

The most important question this study tried to answer was whether planned re-look laparotomy (as represented by group VI) had a role to play in the treatment of severe IAI. An attempt to answer this was made by comparing it to an "operate and watch" group as represented by group V).

In terms of survival there was a difference of 10% between groups V and VI and this was found not be statistically significant. This finding may be criticized on the basis that the numbers were too small. However, there are a number of factors to consider before making this conclusion.

Firstly, as this was an exploratory study in which the results were not predictable, it was felt from a statistical point of view that the numbers were adequate to showed up marked differences between groups. Secondly, Table III and IV show an almost identical survival rate (20% vereus 22%) between 0,4 mℓ and group IV receiving 0,4 mℓ of inoculum but no antibiotic. This result showed that the model was reproducible. This conclusion suggested that the difference of 10% in survival rates between groups V and VI was more accurate than the confidence intervals suggested.

Despite these points, it is reasonable to conclude that survival rate in this study was not increased by planned relook laparotomy.

A policy of re-look laparotomy also did not reduce the number of intra-abdominal abscesses. The 11% difference between groups V and VI in terms of abscess formation was found to be statistically significant (see Table IX).

Evaluation of the body mass profiles showed very little difference between the "operate and observe" group (group V) and the re-look laparotomy group (group VI). Both groups reached their lowest average body mass on the second post-operative day; both started a gain in the average body mass on the third post-operative day. The re-look laparotomy group exceeded its average pre-operative body mass a day earlier than the "operate and observe" group and on the seventh post-operative day the average body mass of this group was a little higher than in the "operate and observe" group. However at the time of euthanasia, both groups exceeded their average pre-operative body mass by about the same amount (42% versus 38%). The statistical analysis carried out showed no significant difference between these two groups in terms of the body mass change over time (see appendix 2).

5.2 CONCLUSION

In summary I conclude that this study has established a successful, reliable and reproducible animal model of IAI which can be used to assess treatment modalities. Of the treatment modalities assessed against the criteria of body mass changes, survival rate, and the development of

intra-abdominal abscesses, the best treatment of IAI is prevention by giving antibiotics at the time of surgery. In developed IAI the best treatment appears to be an "operate and observe" policy (Group V). This treatment means giving antibiotic at the time of surgery, and using a mechanical washout followed by a period of observation. This treatment results in the same mortality as using antibiotics only at the time of surgery or using a planned re-look laparotomy policy. It also gives a much lower mortality than using mechanical washout only. The "operate and observe" policy also gives better results in terms of intra-abdominal abscess formation compared to giving antibiotic only (irrespective of when the antibiotic is given) and gives the same result as the re-look laparotomy policy. In addition, in terms of body mass changes (catabolic response), the "operate and observe" policy gives a much better result compared to using washout and no antibiotic or using antibiotic only at the time of surgery, and gives the same result as a policy of planned re-look laparotomy.

Using mechanical washout alone to deal with intra-abdominal infection gives the worst result in terms of mortality and its affect on health (i.e. body mass). It is thus the worst option available.

Planned re-look laparotomy therefore gives similar results to the "operate and observe" policy and confers no additional benefit. As a possible treatment for IAI it appears, therefore, to have no special advantages, at least in the animal model used in this experiment.

REFERENCES

1. Bohnen J., Boulanger M., Meakins J.L., et al. Prognosis in Generalised Peritonitis: Relation to Cause and Risk Factors. Arch. Surg. 1983; 118: 285-290.

2. Maddaus M.A, and Simmons R.L. Leave the Abdomen Open for Peritonitis: Yes, No, Maybe? Adv. Surg. 1987; 21: 1-18.

3. Baue A.E. Recovery from Multiple Organ Failure. Am. J. Surg. 1985; 149: 420-421.

4. Knaus W.A., Draper E.A., Wagner D.P., Zimmerman J.E. Prognosis in Acute Organ System Failure. Ann. Surg. 1985; 202: 685-693.

5. Polk H.C., Fry D.E. Radical Peritoneal Debridement for Established Peritonitis: The Results of a Prospective Randomized Clinical Trail. Ann. Surg. 1980; 192: 350-355.

6. Stephen M., Lowenthal J. Continuing Peritoneal Lavage in High Risk Peritonitis. Surgery 1979; 85: 603-606.

7. Hallerback B., Anderson C., Englund N. et al. A prospective randomized study of continuous peritoneal lavage post-operatively in the treatment of purulent peritonitis. Surg. Gynaecol. Obstet. 1986; 163: 436.

8. Schein M., Saadia R., and Decker G. Intra Operative Peritoneal Lavage. Surg. Gynecol. Obstet. 1988; 166: 187-195.

9. Schein M. Planned Re-operations and Open Management in Critical Intra-abdominal Infections. A Prospective Personal Experience in 52 cases. Unpublished.

10. Harbrecht P.J., Garrison R.N., Fry D.E. Early Surgery Relaparotomy. Arch. Surg. 1984; 119: 369-374.

11. Victor L.T.C., Farraris M.C. Exploratory Laparotomy for Potential Sepsis in Patients with Multiple Organ Failure. Arch. Surg. 1983; 118: 1130-1133.

12. Pitcher W.D., Musher D.M. Critical Importance of Early Diagnosis and Treatment of Intra-Abdominal Infection. Arch. Surg. 1982; 117: 328-333.

13. Penninckx F.M., Kerremans R.P., and Lauwers P.M. Planned Relaparotomies in the Surgical Treatment of Severe Generalised Peritonitis from Intestinal Origin. World J. Surg. 1983; 7: 762-766.

14. Teichmann W., Wittman D.H., and Andreone P.A. Scheduled Reoperations for Diffuse Peritonitis. Arch. Surg. 1986; 121: 147-152.

15. Andrus C., Doering M., Herrman V.M., and Kaminski D.L. Planned Reoperation for Generalised Intra-Abdominal Infection. Am. J. Surg. 1986; 152: 682-686.

16. Steinberg D. On Leaving the Peritoneal Cavity Open in Acute Generalised Suppurative Peritonitis. Am. J. Surg. 1979; 137: 216-220.

17. Duff J.H. and Moffat J. Abdominal Sepsis Managed by Leaving Abdomen Open. Surgery 1981; 90: 774-778.

18. Maetani S. and Trobe T. Open Peritoneal Draining as Effective Treatment of Advanced Pesitonitis. Surgery 1981; 90: 804-809.

19. Anderson E.D., Mandelbaum D.M., Ellison E.C., Carey L.C., and Cooperman N. Open Packing of the Peritoneal Cavity in Generalised Bacterial Peritonitis. Am. J. Surg. 1983; 145: 131-135.

20. Goris, J.A. (Ogilvie's Method Applied to Infected Wound Disruption. Arch. Surg. 1980; 115: 1103-1107.

21. Broome, A., Hansson L., Lundgren F., and Smedberg S. Open Treatment of Abdominal Septic Catastrophes. World J. Surg. 1983; 792-796

22. Mughal M.M., Bancewicz J. and Irving M.H. Laparostomy: A Technique for the management of Intractable Intra-Abdominal Sepsis. Br. J. Surg. 1986; 73: 253-256.

23. Wouters D.W., Krom R.A.F., Slooff M.J.H., Kootstra G. and Kuijjer P.J. The Use of Marlex Mesh in Patients with Generalised Peritonitis and Multiple Organ System Failure. Surg. Gynecol. Obstet. 1983; 156: 609-614.

24. Bradley E.L and Fulenwider T. Open Treatment of Pancreatic Abscesses. Surg. Gynecol Obstet 1984; 159: 509-513.

25. Pemberton J.H., Decker J.M. Dozois R.R., Nagorney D.M., Ilstrup D., and Remine W.H. Controlled Open Lesser Sac Drainage for Pancreatic Abscesses. Ann. Surg. 1986; 203: 600-604.

26. Wertheimer M.D and Norris S. Surgical Management of Necrotizing Pancreatitis. Arch. Surg. 1986; 121: 484-487.

27. Schein M., Saadia R., Freinkel Z., and Decker G.A.G. Aggressive treatment of severe diffuse peritonitis: a prospective study. Br. J. Surg 1988; 75: 173-176.

28. Krukowski Z.H. Post-operative Abdominal Sepsis. Br. J. Surg. 1988; 75: 1153-1154.

29. Richardson J.D. and Polk H.C. Newer Adjunctive Treatments for Peritonitis. Surgery 1981; 90: 917-918.

30. Rogers P.N., Wright I.H., Ledingham I.McA. Review: Critical Abdominal Sepsis. J.R. Coll. Surg. Edinb. 1989; 32: 1-8.

31. Rogers P.N. and Wright I.H. Post-operatively Intra-abdominal Sepsis. Br. J. Surg. 1987; 74: 973-975.

32. Schein M., Saadia R. and Decker G.A.G. The Open Management of the Septic Abdomen. Surg. Gynecol. Obstet 1986; 163: 587-592.

33. Hedderich G.S., Wexler M.J., Mc Lean A.P.H., and Meakins J.L. The Septic Abdomen: Open Management with Marlex Mesh with a Zipper. Surgery 1986; 99: 399-407.

34. Ranson J.H.C. and Spencer F.C. Prevention, Diagnosis and Treatment of Pancreatic Abscess. Surgery 1977; 82: 99-106.

35. Schein M. Planned or "On Demand" Relaparotomy for Severe Intra-Abdominal Infection. S.A. J. Surg. 1987; 25: 108-109.

36. Chan S.T.F., Essufali S.T. Extended Indications for Polyprophylene Mesh Closure of the Abdominal Wall. Br. J. Surg. 1986; 73: 3-6.

37. Mughal M.M., Bancewicz J., Irving M.H. The Surgical Management of Pancreatic Abscess. Ann. Roy. Coll. Surg. Eng. 1987; 69: 64-69.

38. Larvin M., Chamers A.G., Robinson P.J., McMahon M.J. Debridement and Closed Cavity Irrigation for the Treatment of Pancreatic Sepsis. Br. J. Surg. 1989; 70: 465-471.

39. Malangoni M.A., Richardson J.D., Shallcross J.C., Seiler J.G., and Polk H.C. Factors Contributing to Fatal Outcome after Treatment of Pancreatic Abscess. Am. Surg. 1986; 203: 603-613.

40. Warshaw A.L., and Jin G. Improved Survival in 41 patients with Pancreatic Abscess. Am. Surg. 1985; 202: 408-417.

41. Berger H.G., Buchler M. et al. Necrosectomy and Post-operative Local Lavage in Patients with necrotizing pancreatitis. Results of a Prospective Clinical Trial. WJS 1938; 12: 255-262.

42. Nicholson M.L., Mortenson N.J., and Espiner H.J. Pancreatic abscess. Results of Prolonged Irrigation of Pancreatic Bed after Surgery. Br. J. Surg. 1988; 75: 88-91.

43. Krukowski Z.H., Matheson N.A. Emergency Surgery for Diverticular Disease Complicated by Generalized and Faecal Peritonitis. Br. J. Surg. 1984; 71: 921-927.

44. Nagorney D.M., Adson M.A., and Pemberton J.H. Sigmoid Diverticulitis with Perforation and Generalized Peritonitis. Dis. Colon. Rectum. 1985; 28: 71-75.

45. Bakker F.C., Hoitsma H.F.W. and Den Otter G. The Hartmann Procedure. Br. J. Surg. 1982; 69: 580-582.

46. Walsh G.L., Chiasson P. et al. The Open Abdomen. Surg. Clin. N.A. 1988; 68: 25-40.

47. Norton L.W. Does Drainage of Intra-abdominal pus reverse Multiple Organ Failure. Am. J. Surg. 1985; 149: 347-350.

48. Fry D.E. Multiple Organ Failure. Surg. Clin. N. Amer. 1988; 68: 107-122.

49. Schein M., Saadia R., MacFarlane C., and Boffard K. Gut Barrier Function and the Surgeon. Br. J. Surg. 1990; 77: 487-492.

50. Browne M.K. and Leslie G.B. Animal Models of Peritonitis. Surg. Gynecol. Obstet. 1976; 143: 738-740.

51. Wichterman K.A., Baue A.E., and Chaudry I.H. Sepsis and Septic Shock - A Review of Laboratory Models and a Proposal. J. Surg. Res. 1980; 29: 189-201.

52. Weinstein W.M., Onderdonk A.B., Bartlett J.G., and Borbach S.L. Experimental Intra-Abdominal Abscesses in Rats: Development of an Experimental Model. Infect. Immun. 1974; 10: 1250-1255.

53. Bartlett J.G., Onderdonk A.B., Louie T., Kasper D.L., and Gorbach S.L. A Review: Lessons from an Animal Model of Intra-Abdominal Sepsis. Arch. Surg. 1978; 113: 853-857.

54. Onderdonk A.B., Weinstein W.M., Sullivan N.M., Bartlett J.G. and Gorbach S.L. Experimental Intra-Abdominal Abscesses in Rats: Quantitative Bacteriology of Infected Animals. Infect. Immun. 1974; 10: 1256-1259.

55. Nichols R.E., Smith J.W., and Balthazer E.R. Peritonitis and Intra-Abdominal Abscess: An Experimental Model for the Evaluation of Human Disease. J. Surg. Res. 1978; 25: 129-134.

56. Weinstein W.M., Onderdonk A.W., Bartlett J.G., Louie T.J., and Gorbach S.L. Anti Microbial Therapy of Experimental Intra-Abdominal Sepsis. J. Infect. Dis. 1975; 132: 282-286.

57. Onderdonk A.B., Bartlett J.G., and Louie T. et al. Microbial Synergy in Experimental Intra-Abdominal Abscess. Infect. Immun. 1976; 13: 22-26.

58. Rokke O., Revhang A., and Gieriksky K.C. PMN Activity and Endotoxin Kinetics in Peritonitis Induced after Moderate Trauma. Acta. Clin. Scand. 1989; 155: 497-502.

59. Short B.L., Gardiner W.M. et al. Rat Intra-peritoneal Sepsis. Circ. Shock 1983; 10: 351-359.

60. Cheadle W.G., Brown G.L. et al. Effects of Muramyl Dipeptide and Clinamycin in a Murine Abdominal Abscess Model. J. Surg. Res. 1986; 41: 319-325.

61. Steepman H.K., Higgs J.W., Hayes D.I.C., and Hamit H.F. Value of Antibiotics, Corticosteroids and Peritoneal Lavage in the Treatment of Experimental Peritonitis. Surgery 1969; 66: 1060-1066.

62. McCanville J.H., Snyder M.J. et al. Model of Intra-abdominal Abecess in Mice. Infect. Immun. 1981; 31: 507-509.

63. Onderdonk A.B., Kasper D.L. et al. Experimental Animal Models of Anaerobic Infections. Rev. Infect. Dis. 1979; 1. 291-301.

64. Narat J.K., Cipolla A.F. et al. Intra-peritoneal drip in the presence of Abdominal Infection. Am. J. Surg. 1950; 439-442.

65. Fry D.E., Garrison R.N., et al. An Experimental Model of Intra-Abdominal Abscess in the Rat. Adv. in Shock Res. 1982; 7: 7-11.

66. Dickerson R.N., Fried R.C. et al. Effect of Propranolol on Nitrogen and Energy Metabolism in Sepsis. J. Surg. Res. 1990; 48: 38-41.

67. Lally K.P., Trettin J.C., and Torma J. Adjunctive Antibiotic Lavage in Experimental Peritonitis. Surg. Gynecol. Obstet. 1983; 156: 605-608.

68. Kay S., and Choy S.H. Results of Intra-Peritoneal Injection of Barium Sulphate Contrast Medium. A.M.A. Arch. of Path. 388-393.

69. Maingot R. Abdominal Operations Vol 11 6th Edition N.Y. Appleton Century Crafts 1974; 1416.

70. Colin I., and Nance F.C. Intestinal Antisepsis and Peritonitis from Perforations. In: Sabiston D.C. ed Davies-Christopher Textbook of Surgery. Philadelphia: W.B. Saunders 1981; 1076.

71. Schein M., and Saadia R. To Wash or not to Wash? Intra-operative Peritoneal Lavage in the Contaminated Peritoneal Cavity. SAJS 1989; 27: 22-23.

72. Mihervim S., Bentley S., Youngs D., et al. Prophylactic Saline Peritoneal Lavage in Elective Colorectal Operations. Dis. Colon. Rectus. 1980; 23: 392-394.

73. Hunt J.L. Generalized peritonitis: To Irrigate or not to Irrigate the Peritoneal Cavity. Arch. Surg. 1982; 117: 209-212.

74. Musiker R. Style Guide for Theses and Dissertations. University of the Witwatersrand Library. Occasional Publications 1980.

APPENDIX 1

BODY MASS OF ANIMALS

TO FIND BEST DOSE OF FAECAL INNOCULUM

- 40 Rats Divided into 4 Groups of 10 Each and Given Graduated Doses of Innoculum.

0.1 ml Innoculum Group.

KEY: $D^{\dagger}$ = Rat Died.

Rat	Day of Surgery	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Rat 1	198 Grams	193	200	213	218	216	218	231
Rat 2	170 Grams	167	168	174	182	187	191	201
Rat 3	211 Grams	204	206	204	209	212	215	227
Rat 4	210 Grams	200	205	219	224	228	229	241
Rat 5	180 Grams	175	183	187	190	187	186	196
Rat 6	202 Grams	197	196	186	193	191	195	204
Rat 7	215 Grams	210	216	222	228	226	224	233
Rat 8	185 Grams	169	179	180	182	183	186	197
Rat 9	190 Grams	180	187	192	198	193	187	187
Rat 10	229 Grams	223	226	222	221	220	224	226
Mass + SD	199±17.97	191± 18.59	197± 17.57	200± 18.05	205± 17.60	204± 17.7	202± 17.8	214± 19.1
SE	5.5	5.6	5.5	5.7	5.5	5.6	5.6	6.0
t value		1.24	0.45	0.15	1	0.9	1.16	2.5
p value		>0.05	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05

0.2cc Innoculum Group

Rat	Day of Surgery	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Rat 1	178 Grams	172	177	161	154	157	162	162
Rat 2	186 Grams	174	180	184	195	200	212	216
Rat 3	187 Grams	183	-	-	-	-	-	-
Rat 4	194 Grams	188	186	185	196	192	200	206
Rat 5	216 Grams	208	214	310	213	216	222	235
Rat 6	193 Grams	192	192	192	198	199	199	210
Rat 7	200 Grams	199	197	204	206	205	212	216
Rat 8	186 Grams	175	162	204	211	212	216	219
Rat 9	175 Grams	186	166	171	179	176	179	190
Rat 10	222 Grams	218	217	226	232	232	233	244
Mean + SD	194±15.26	187± 15.4	188± 19.24	193± 20	198± 22	197± 22	204± 21.9	210± 24
SE	4.8	5.1	6.4	6.6	7.3	7.3	7.3	8.0
t value		1.25	1.02	0.21	0.61	0.59	1.29	2.0
p value		>0.05	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05

0.3cc Innoculum Group

Rat	Day of Surgery	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8
Rat 1	192 Grams	191	190	-	-	-	-	-	-
Rat 2	182 Grams	180	175	183	187	194	195	198	204
Rat 3	198 Grams	192	194	202	204	206	209	213	218
Rat 4	164 Grams	160	-	-	-	-	-	-	-
Rat 5	191 Grams	186	183	-	-	-	-	-	-
Rat 6	180 Grams	178	175	179	186	188	192	197	202
Rat 7	224 Grams	207	205	211	215	221	223	231	241
Rat 8	190 Grams	196	200	206	209	222	224	231	243
Rat 9	226 Grams	216	209	216	225	235	237	238	247
Rat 10	193 Grams	203	203						
Mean + SD	194±19	190 ±16.0	192 ±12.9	199 ±15.1	204 ±15.4	211 ±18.1	213 ±17.8	218 ±17.9	225 ±20.39
SE	6.0	5.0	4.3	6.16	6.28	7.3	7.2	7.3	8.28
t value		0.62	1.11	0.94	0.69	1.6	1.8	2.46	3.1
p value		>0.05	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05

0.4cc Innoculum Group

Rat	Day of Surgery	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8
Rat 1	235 Grams	230	-	-	-	-	-	-	-
Rat 2	188 Grams	191	187	185	185	194	191	195	201
Rat 3	218 Grams	213	209	209	215	227	225	228	237
Rat 4	231 Grams	227	222	-	-	-	-	-	-
Rat 5	184 Grams	182	-	-	-	-	-	-	-
Rat 6	175 Grams	174	-	-	-	-	-	-	-
Rat 7	229 Grams	218	-	-	-	-	-	-	-
Rat 8	194 Grams	164	-	-	-	-	-	-	-
Rat 9	172 Grams	185	183	-	-	-	-	-	-
Rat 10	199 Grams	191	-	-	-	-	-	-	-
Mean + SD	202.6±23.8	197± 22.48	200± 18.4	197± 16.9	200± 21.2	210± 23.3	208± 24.04	211± 23.3	219± 25.4
SE	7.5	6.9	9.2	11	14.9	16.4	16.9	16.4	17.6
t value		0.68	0.21						
p value		>0.05	>0.05	-	-	-	-	-	-

Control Group Irradiated faeces barium mixture (Sterile solution)

Body mass in grams

Rat	Day of Surgery	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6
1	192 Grams	185	120	190	195	199	210
2	207 Grams	190	196	200	208	211	210
3	220 Grams	215	224	224	224	235	230
4	211 Grams	202	208	213	214	223	219
5	216 Grams	205	210	212	217	220	221
Mean + SD	208±11.1	199± 12	205.6± 13.2	207± 13.08	211± 10.9	217± 13.48	215± 12.1
SE	4.96	5.36	5.9	5.8	4.8	5.9	5.4
t value		1.16	0.47	0.1	0.6	1.5	1.3
p value		>0.05	>0.05	>0.05	>0.05	>0.05	>0.05

GROUP II. 0,4 ml OF INNOCULUM (FAECAL SUSPENSION) PLACED INTO PERITONEAL CAVITY + 50 MILLIGRAMS OF CEFOXITIN AT THE SAME TIME.

BODY MASS IN GRAMS

Rat	Day of Surgery	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Rat 1	204 Grams	190	184	179	1[illegible]	181	180	200
Rat 2	220 Grams	204	208	218	217	217	217	230
Rat 3	196 Grams	182	185	193	188	196	196	209
Rat 4	190 Grams	181	180	175	172	174	176	193
Rat 5	152 Grams	143	142	148	145	148	150	162
Rat 6	194 Grams	183	186	194	191	193	189	210
Rat 7	202 Grams	190	181	203	197	190	190	200
Rat 8	178 Grams	167	165	170	160	166	166	175
Rat 9	190 Grams	186	185	192	193	199	196	199
Rat 10	184 Grams	174	172	177	173	176	172	182
Mean + SD	191±17.9	180± 16.3	178± 17.0	184.9 ±19.4	181.1 ±20.4	183± 19.4	182.8 ±18,64	196 ±19.3
SE	5.6	5.1	5.3	6.1	6.4	6.1	5.8	6.10
t value		2.1	2.3	1.0	1.5	1.18	1.24	0.98
p value		>0.05	<0.05	>0.05	>0.05	>0.05	>0.05	>0.05

P.M.:

2 Rats - 4 Abscesses
2 Rats - 3 Abscesses
1 Rat - 2 Abscesses
4 Rats - 1 Abscesses
1 Rat - 0 Abscesses

} Sacrificed on day 7.

GROUP III. 0.4 ml OF INNOCULUM PLACED INTO PERITONEAL CAVITY THEN 50 MILLIGRAMS OF CEFOXITIN GIVEN INTRA-PERITONEALLY 5 HOURS LATER.

BODY MASS IN GRAMS

Rat	Day of Surgery	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Rat 1	182 Grams	172	167	170	171	154	162	163
Rat 2	222 Grams	206	214	217	209	200	203	212
Rat 3	208 Grams	201	190	176	163	155	-	-
Rat 4	200 Grams	189	175	176	177	176	183	187
Rat 5	186 Grams	177	172	176	181	168	182	179
Rat 6	220 Grams	209	200	213	204	193	215	215
Rat 7	204 Grams	193	188	194	182	170	177	178
Rat 8	198 Grams	185	181	184	184	174	174	180
Rat 9	206 Grams	194	-	-	-	-	-	-
Rat 10	204 Grams	197	181	177	170	160	170	175
Mean + SD	203± 12.69	192 ±11.9	185 ±14.8	187 ±17.2	182.3 ±15.2	172.1 ±15.87	183.25 ±17.53	186.1 ±18.1
SE	4	3.7	4.8	5.7	5.0	5.2	6.1	6.3
t value		2.8	3.6	2.7	4.0	5.8	3.0	2.5
p value		<0.05	<0.05	<0.05	<0.05	<0.001	<0.05	<0.05

P.M.:

1 Rat - Multiple Micro-Abscesses
4 Rats - 2 Abscesses
3 Rats - 1 Abscess

} Survivors - sacrificed on Day 7

GROUP IV. 0,4 ml OF INNOCULUM PLACED INTO PERITONEAL CAVITY THEN WASHOUT OF PERITONEAL CAVITY WITH 20cc OF SALINE 5 HOURS LATER.

BODY MASS IN GRAMS

Rat	Day of Surgery	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Rat 1	192 Grams	183	-	-	-	-	-	-
Rat 2	190 Grams	178	-	-	-	-	-	-
Rat 3	186 Grams	178	-	-	-	-	-	-
Rat 4	188 Grams	175	171	-	-	-	-	-
Rat 5	185 Grams	168	156	168	179	183	184	193
Rat 6	195 Grams	186	-P.M.- Hole in Bowel therefore discarded from trial					
Rat 7	198 Grams	185	-	-	-	-	-	-
Rat 8	190 Grams	178	149	160	169	172	175	184
Rat 9	192 Grams	173	169	-	-	-	-	-
Rat 10	210 Grams	188	161	165	153	149	-	-
Mean + SD	192.6±7.26	179.2 ±6.3	161.2 ±9.12	164.3 ±4.04	167 ±13.1	168 ±17.3	175.5 ±6.36	188.5 ±6.36
SE	2.2	1.9	4.1	2.3	7.7	10.1	3.74	3.74
t value		7.0	7.7	13.3	3.6	2.6	-	-
p value		<0.001	<0.001	<0.001	<0.05	>0.05	-	-

P.M.:

1 Rat - 7 Abscesses
1 Rat - 5 Abscesses

} The two survivors No 5 & 8 sacrificed on Day 7

GROUP V. 0.4 ml OF INNOCULUM INTO PERITONEAL CAVITY THEN 5 HOURS LATER WASHOUT OF PERITONEAL CAVITY WITH 20ml OF SALINE + 50 MILLIGRAMS OF CEFOXITIN INTRA-PERITONEALLY.

RAT NO	0 DATE OF SURGERY	1 5/8/89	2 6/8/89	3 7/8/89	4 8/8/89	5 9/8/89	6 10/8/89
1	224	205	205	216	218	213	216
2	220	202	199	208	204	210	219
3	207	192	201	208	187	190	202
4	205	190	176	171	DIED OF SEPTIC SHOCK + PERITONITIS, NO ABSCESSES.		
5	219	205	195	204	212	204	214
6	200	179	185	178	188	192	196
7	223	206	205	218	225	228	222
8	218	200	193	203	209	209	220
9	204	188	179	189	196	200	201
10	226	208	190	178	174	186	150*
Mean ± SD	214.6 ±9.57	197.5 ±9.66	190 ±13.42	197.3 ±16.92	201.4 ±16.4	200.7 ±16.6	204.4 ±[illegible].4
SE	3.0	3.0	4.2	5.3	5.4	5.2	7.4
t value		5.7	5.6	3.2	2.6	2.8	1.51
p value		<0.001	<0.001	<0.05	<0.05	<0.05	>0.05

GROUP V, 0.4 ml OF INNOCULUM INTO PERITONEAL CAVITY THEN 5 HOURS LATER WASHOUT OF PERITONEAL CAVITY WITH 20 ml OF SALINE + 50 MILLIGRAMS OF CEFOXITIN INTRA-PERITONEALLY (CONTINUED I)

RAT NO	7 11/8/89	8 12/8/89	9 13/8/89	12 15/8/89	13 16/8/89	14 17/8/89	15 18/8/89
1	224	225	230	242	248	247	249
2	222	229	232	246	245	246	252
3	208	208	219	222	226	235	236
4	DIED OF SEPTIC SHOCK + PERITONITIS, NO ABSCESSES.						
5	220	227	236	238	248	251	250
6	204	205	209	208	218	222	222
7	228	228	233	238	246	248	251
8	223	229	236	240	245	239	242
9	206	209	217	219	229	230	234
10	R.M. : 10 SMALL ABSCESSES IN ABDOMINAL CAVITY. EUTHANASED.						
Mean ± SD	216 ±9.34	220 ±10.62	226 ±10.12	231 ±13.5	238 ±11.87	239 ±9.92	242 ±10.65
SE	3.3	3.7	3.5	4.7	4.1	3.5	3.7
t value	0.75	1.5	3.46	3.6	5.85	7.1	7.4
p value	>0.05	>0.05	<0.05	<0.05	<0.001	<0.001	<0.001

GROUP V. 0.4 mℓ OF INNOCULUM INTO PERITONEAL CAVITY THEN 6 HOURS LATER WASHOUT OF PERITONEAL CAVITY WITH 20 mℓ OF SALINE + 50 MILLIGRAMS OF CEFOXITIN INTRA-PERITONEALLY. (CONTINUED II)

RAT NO	18 21/8/89	20 23/8/89	25 28/8/89	29 1/9/89	32 4/9/89	36 7/9/89	40 11/9/89
11	259	274	284	288	295	280	291
12	255	265	273	275	281	275	284
13	236	243	251	256	262	258	264
14	DIED OF SEPTIC SHOCK + PERITONITIS. NO ABSCESSES.						
15	259	263	285	298	288	299	304
16	234	236	248	253	253	241	258
17	256	265	274	275	280	275	276
18	237	253	261	275	276	271	281
19	242	247	268	270	274	272	276
20	P.M.: 10 SMALL ABSCESSES IN ABDOMINAL CAVITY. EUTHANASED.						
Mean ± SD	247 ±11.0	255 ±13.05	268 ±13.86	273.7 ±14.9	276 ±13.49	271.2 ±16.5	279.2 ±15.54
SE	3.18	4.6	4.8	5.2	4.7	5.8	5.1
t value	8.47	8.9	10.9	11.4	13.1	9.8	12.7
p value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

GROUP V. 0.4 ml OF INNOCULUM INTO PERITONEAL CAVITY THEN 5 HOURS LATER WASHOUT OF PERITONEAL CAVITY WITH 20 ml OF SALINE + 50 MILLIGRAMS OF CEFOXITIN INTRA-PERITONEALLY. (CONTINUED III)

RAT NO	44 15/9/89	47 18/9/89	54 25/9/89	61 2/10/89
1	296	298	297	309
2	268	297	294	300
3	271	280	281	291
4	DIED OF SEPTIC SHOCK + PERITONITIS. NO ABSCESSES.			
5	315	322	331	327
6	260	261	261	271
7	275	287	288	299
8	280	284	287	297
9	267	289	293	297
10	P.M.: 10 SMALL ABSCESSES IN ABDOMINAL CAVITY. EUTHANASED.			
Mean ± SD	281.5 ±17.75	295.7 ±17.41	292 ±20.6	298 ±15.7
SE	6.25	6.4	7.1	5.5
t value	10.94	12.3	11.0	15.35
p value	<0.001	<0.001	<0.001	<0.001

GROUP VI. 0.4 ml OF INNOCULUM INTRA-PERITONEALLY THEN 5 HOURS LATER WASHOUT WITH 20ml OF SALINE AND 50 MILLIGRAMS OF CEFOXITIN INTRA-PERITONEALLY FOLLOWED 24 HOURS LATER BY PLANNED "RELOOK" EXPLORATION OF ABDOMINAL CAVITY (LAPAROTOMY) AND WASHOUT WITH 20 ml OF SALINE.

RAT NO	0 DATE OF SURGERY	1 17/8/89	2 18/8/89	3 19/8/89	4 20/8/89	5 21/8/89	6 22/8/89
1	199	192	184	187	190	196	208
2	220	210	200	- ANAESTHETIC DEATH			
3	206	196	188	183	189	192	195
4	210	201	193	196	188	196	200
5	196	191	188	186	189	203	212
6	217	208	206	217	221	229	238
7	210	202	185	193	204	207	212
8	199	189	180	183	191	190	196
9	218	210	201	202	215	221	221
10	284	273	264	256	267	274	278
Mean ± SD	215.8 ±25.3	207 ±24.4	198.9 ±24.3	199.7 ±23.9	206 ±26.0	212 ±25.7	217.6 ±26.36
SE	8.0	7.2	7.6	7.9	8.6	8.9	8.7
t value		1.19	1.2	1.98	1.09	0.38	0.25
p value		>0.05	<0.05	>0.05	>0.05	>0.05	>0.05

GROUP VI. 0.4 mℓ OF INNOCULUM INTRA-PERITONEALLY THEN 5 HOURS LATER WASHOUT WITH 20mℓ OF SALINE AND 50 MILLIGRAMS OF CEFOXITIN INTRA-PERITONEALLY FOLLOWED 24 HOURS LATER BY PLANNED "RELOOK" EXPLORATION OF ABDOMINAL CAVITY (LAPAROTOMY) AND WASHOUT WITH 20 mℓ OF SALINE. (CONTINUED I).

RAT NO	7 23/8/89	13 29/8/89	16 1/9/89	19 4/9/89	23 8/9/89	26 11/9/89	30 15/9/89
1	206	229	248	241	241	253	254
2	ANAESTHETIC DEATH	-	-	-	-	-	-
3	196	210	218	225	222	283	232
4	210	280	240	248	241	256	260
5	220	230	235	248	239	248	255
6	247	270	278	289	289	302	304
7	222	244	257	248	251	270	275
8	210	228	235	242	241	260	255
9	226	258	265	263	265	285	282
10	277	290	298	304	277	304	310
Mean ± SD	223.7 ±24.6	243.2 ±25.02	252 ±24.5	255.8 ±25.3	251.1 ±21.2	273.1 ±21.0	269.6 ±25.4
SE	8.2	8.4	8.12	8.4	7.0	7.0	8.4
t value	1.0	3.3	4.5	4.8	5.9	8.2	6.4
p value	>0.05	<0.05	<0.05	<0.05	<0.001	<0.001	<0.001

GROUP VI. 0.4 ml OF INNOCULUM INTRA-PERITONEALLY THEN 5 HOURS LATER WASHOUT WITH 20 ml OF SALINE AND 50 MILLIGRAMS OF CEFOXITIN INTRA-PERITONEALLY FOLLOWED 24 HOURS LATER BY PLANNED "RELOOK" EXPLORATION OF ABDOMINAL CAVITY (LAPAROTOMY) AND WASHOUT WITH 20 ml OF SALINE. (CONTINUED II).

	34	40	47	54	62	68
RAT NO	19/9/89	25/9/89	2/10/89	9/10/89	17/10/89	23/10/89
1	263	267	264	272	282	209
2	ANAESTHETIC DEA					
3	239	247	10	259	260	261
4	258	270	276	278	282	287
5	254	254	257	263	269	284
6	310	316	338	333	352	356
7	277	285	296	303	352	356
8	263	271	286	282	287	298
9	285	294	302	306	315	326
10	318	332	336	349	331	347
Mean	274 ±26.1	283 ±29.1	289.4 ±31.9	295 ±31.0	297.7 ±30.0	306 ±31.2
SE	8.1	9.7	10.6	10.3	10.0	10.14
t value	6.7	7.0	6.9	7.7	8.2	8.7
p value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

SURVIVORS SACRIFICED 23/10/89 - 1 RAT HAD ABSCESS. REST ALL CLEAR.

APPENDIX 2

STATISTICS

1. COMPARISON OF 'RELOOK LAPAROTOMY' VS 'OPERATE AND OBSERVE' FOR THE 12 DAYS IN WHICH THE 2 GROUPS COULD BE MATCHED EXACTLY: FOR ALL 20 ANIMALS

Mean Values of Weights:

	0	1	2	3	4	5	6	7	12	40	47	54
(n=10) Laparotomy	216	207	199	200	206	212	218	224	243	283	289	295
(n=10) No Laparotomy	214	198	191	197	201	201	201	217	232	279	290	295

ANOVA.

SOURCE	ss	df	ms	F
Between subs	97 253	19		
Group	3 627	1	3 627	0.7
Residual	93 626	18	5 201	
Within subs	267 732	198		
Day	250 996	11	22 818	256*
Group. Day	1 075	11	98	1.1
Residual	15 661	176	89	
Total	364 985	217		

(i) Both groups of animals showed a significant weight change* over the time period [F(11,176) = 256: p=0.0001]. A significant weight lose over the first few days was followed by a significant weight gain.

However, there were no significant difference between the two groups in their weight change [F(11,176) = 1.1: p=0.36].

(ii) Within each group, Tuckey's critical range test showed that difference in weight of greater than 16g between days were statistically significant at the 5% level.

2. COMPARISONS OF 'RELOOK LAPAROTOMY' VS 'OPERATE AND OBSERVE' FOR THE 12 MATCHED DAYS FOR THE SURVIVORS ONLY (17 ANIMALS).

Mean Weights:

	DAY											
	0	1	2	3	4	5	6	7	12	40	47	54
(n=9) Laparotomy	215	207	199	200	206	212	218	224	243	283	289	296
(n=8) No Laparotomy	214	197	193	203	205	205	211	217	232	279	290	293

ANOVA.

SOURCE	ss	df	ms	F
Between subs	74 514	16		
Group	945	1	945	0.2
Residual	73 569	15	4 905	
Within subs	262 841	187		
Day	250 518	11	227748	328*
Group. Day	867	11	79	1.1
Residual	11 456	165	69.4	
Total	337 354	203		

(i) Both groups of animals showed a significant weight change over the time period. [F(11.165) = 328; p 10.0001]. A significant weight loos over the first few days was followed by a significant weight gain. However, there were no significant differences between the two groups in their weight changes [F(11,165) = 1.1: p = 0.36].

(ii) Within each group, Tukey's critical range test showed that differences in weight of greater than 14g between days were statistically significant at the 5% level.

3. COMPARISONS BETWEEN THE GROUPS FOR 17 'CLOSELY MATCHED' DAYS FOR ALL 20 ANIMALS

Mean Weights:

Day	Laparotomy	No Laparotomy		Laparotomy	No Laparotomy
Day 0	216	215	15/16	253	242
Day 1	207	198	18/19	256	247
Day 2	199	191	25/26	268	268
Day 3	200	197	26/30	270	274
Day 4	206	201	40	283	279
Day 5	212	201	47	289	290
Day 6	218	204	54	295	293
Day 7	224	217	61/62	298	299
Day 12	243	232			

ANOVA.

SOURCE	ss	df	ms	F
Between subs	139 486	19		
Group	4 044	1	4 044	0.5
Residual	135 442	18	7 525	
Within subs	382 464	283		
Day	359 969	16	22 498	273*
Group. Day	1 823	16	114	1.4
Residual	20 672	251	82.4	
Total	521 950	302		

(i) Both groups of animals showed a significant weight change over the time period [$F(16,251) = 273 : p<0.0001$]. A significant loss over the first few days was followed by a significant weight gain. However, there were no significant differences between the two groups in their weight changes [$F(16,251) = 1.4; p = 0.14$]

(ii) Within each group, Tukey's critical range test showed that differences in weight of greater than 16g between days were statistically significant at the 5% level.

4. COMPARISONS BETWEEN THE GROUPS FOR 17 'CLOSELY MATCHED' DAYS FOR THE SURVIVORS ONLY (17 ANIMALS).

Mean Weights:

Day	Laparotomy	No Laparotomy		Laparotomy	No Laparotomy
Day 0	215	214	15/16	253	242
Day 1	207	197	18/19	256	247
Day 2	199	193	25/26	268	268
Day 3	200	203	26/30	270	274
Day 4	206	205	40	283	279
Day 5	212	205	47	289	0
Day 6	218	211	54	295	293
Day 7	224	217	61/62	298	299
Day 12	243	232			

ANOVA.

SOURCE	ss	df	ms	F
Between subs	103 087	16		
Group	1 081	1	1 081	0.2
Residual	102 006	15	6 800	
Within subs	377 574	272		
Day	359 492	16	22 468	327*
Group. Day	1 615	16	101	1.5
Residual	16 467	240	68.6	
Total	480 660	288		

(i) Both groups of animals showed a significant weight change over the time period [F(16,240) = 327; p<0.0001]. A significant loss over the first few days was followed by a significant weight gain. However, there were no significant differences between the two groups in their weight changes [F(16,240) = 1.5; p = 0.10]

(ii) Within each group, Tukey's critical range test showed that differences in weight of greater than 14g between days were statistically significant at the 5% level.