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GASTRIC CANCER
NATURAL HISTORY AND TREATMENT

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MD THESIS

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DECLARATION

I confirm that I am the sole author of this thesis which is a record of my personal research work. None of the subject matter has previously been submitted as a higher degree. All references cited have been consulted personally.

David Cunningham

CHAPTER ONE

INTRODUCTION

1.1 INTRODUCTION

"As medical means can do nothing more than relieve the symptoms of cancer of the stomach, and as surgery holds out the only hope of relief or cure - moreover as the earlier these cases are seen by the surgeon the greater is the probability of real good being done - it would seem to me that in every case where cancer of the stomach is suspected the question of surgical treatment should be considered at a much earlier period than is now the custom."

A.Mayo Robson, FRCS, 1900

This is an extract from a lecture delivered by A.Mayo Robson to the Royal college of Surgeons of England at the turn of this century (1). How relevant are these comments 9 decades later? In this thesis an attempt will be made to answer this question by examining the present natural history of gastric cancer and assessing the current results of surgery and chemotherapy. However, the emphasis will be on the role of chemotherapy in the treatment of gastric cancer because as a

medical oncologist this is the area where I can
make the most relevant contribution.

1.2 INCIDENCE AND AETIOLOGY

The most recent statistics from England and Wales show that 11,553 cases of stomach cancer were registered in 1983 (Table 1.1) (2). It is the third most common cause of cancer deaths in males and the fourth most common cause of cancer deaths in females (3). There is a marked male preponderance with a male to female ratio of 2:1 and it is more common among lower socioeconomic groups (2). During the past 50 years there has been a steady, unexplained decline in the number of cases affecting both sexes (4) but the crude 5 year survival has remained between 7-11% (5). In Scotland the number of cases of gastric cancer has also decreased during the past 50 years (6). The average incidence is similar to England and Wales but there are some areas where the incidence is up to 1.5 times the national average. These are generally the industrial areas such as Glasgow and Dundee with a lower incidence in the rural areas (6).

On a worldwide scale, the incidence of gastric cancer varies considerably (Table 1.2) (7). The incidence in Japan exceeds any other country and is more than 2.5 times that of the United Kingdom and 9 times that of the U.S.A..

Site Description	Number	%*
a) Males		
All sites (ICD 140-208)	100,645	100
Trachea, bronchus and lung	26,403	26
Skin other than melanoma	11,317	11
Prostate	9,127	9
Stomach	7,060	7
Bladder	6,683	7
Colon	6,488	6
Rectum, rectosigmoid junction and anus	5,171	5
Pancreas	2,881	3
Oesophagus	2,294	2
Kidney and other unspecified urinary organs	1,894	2
Others	21,327	21
b) Females		
All sites (ICD 140-208)	97,604	100
Breast	21,297	22
Skin other than melanoma	10,419	11
Trachea, bronchus and lung	9,475	10
Colon	8,124	8
Ovary and other uterine adnexa	4,521	5
Stomach	4,493	5
rectum, rectosigmoid junction and anus	4,329	4
Cervix uteri	3,875	4
Body of uterus	3,346	3
Pancreas	2,752	3
Other	24,973	26

*The sum of percentages for individual sites do not always add to 100 because of rounding.

Table 1.1 The ten most common malignant sites registered in England and Wales in 1983 with the percentage that each of the ten sites forms of all sites

Country	Males	Females
Japan	50.4	25.4
Chile	46.0	23.4
Hungary	33.4	15.8
Austria	26.1	13.6
Italy	23.4	11.2
Spain	21.4	11.1
Scotland	18.8	9.2
England and Wales	18.1	8.1
Mauritius	16.3	7.4
Sweden	13.4	7.0
France	13.1	6.0
Canada	10.5	4.8
USA	6.4	3.13
Nicaragua	0.1	0.4

Table 1.2 Age-adjusted death rates for cancer of
the stomach in the World

Japanese migrants to the U.S.A. have a lower incidence of stomach cancer but it remains 5 times that of the indigenous population (8). The marked inter-country variation in the incidence of gastric cancer, and the studies of migrant populations have implicated environmental factors in its aetiology. The most important of these is thought to be diet but other factors such as exposure to coal dust or asbestos are also relevant (9).

There have been a number of studies which have attempted to identify the dietary constituents which lead to an increased risk of gastric cancer (10-15). The results have been conflicting. For Japanese migrants to Hawaii the ingestion of salted and dried fish was found to be a strong risk factor (10) but this was not confirmed in a further study from Japan (11). Similarly in South Louisiana, smoked foods and home cured meat were associated with an increased risk of gastric cancer in the black population but not in the white (12). One recent study from Greece identified pasta beans and nuts (13) and another from Canada identified chocolate and carbohydrate as risk factors (14). In Columbia, which has one of the highest incidences of gastric cancer outside Japan, people from areas with a

high incidence of gastric cancer were found to eat fava beans in far larger amounts than equivalent groups from low incidence areas (15). However the only consistent finding from all of these studies was the protective effect of the ingestion of vegetables, particularly lettuce and citrus fruits (10-15). In Scotland, there have not been any studies of diet and gastric cancer but the Scots do eat up to 50% more processed foods than the English and this may be related to the higher incidence of the tumour in certain areas(6). Alcohol and cigarettes have no proven link with the development of gastric cancer (10-15).

On the basis of some of these epidemiological data, nitrites have been implicated in the aetiology of gastric cancer. Nitrites are used as a preservative and a colouring agent in a variety of fish and meat products especially home-cured meats, dried fish and sausages. Also, nitrates in the diet can be converted to nitrites by bacterial action in the food or in the stomach (16). Dietary nitrates are mainly derived from crop fertiliser. They are taken up by the crops, and may also contaminate fresh water supplies. Nitrates are also used in the process of recycling sewage. These factors have led to a general increase in the amount of dietary nitrate in the

past 20 years (17). Nitrites can also interact with Japanese soy sauce (18) and with fava beans to make them mutagenic (19).

One possible mechanism for the development of gastric cancer which incorporates these nitrite/nitrate data was suggested by Correa et al (20). In this hypothesis gastric atrophy played a key role by leading to a reduction in gastric acid secretion and permitting bacterial colonisation of the stomach; this would not be possible in an acid environment. These bacteria, it was postulated, could reduce dietary nitrate to nitrite and form N-nitroso compounds through the nitrosation of dietary amines. N-nitroso compounds are known to be carcinogenic in animals (21) and could therefore act on the atrophic gastric mucosa causing intestinal metaplasia, dysplasia and ultimately cancer.

It has been suggested that the decline of gastric cancer in the U.S.A. and the U.K. is linked to more widespread refrigeration of foodstuffs with a resultant reduction in the use of nitrites. In the USA during the last 50 years there has been a 75% reduction in the use of nitrites in meat to prevent botulism (22). However 2 studies from the United Kingdom failed to show a positive association between nitrate in the

drinking water (23) or in the saliva (24) and gastric cancer. Levels of salivary nitrate, which are taken to be a good marker of the dietary intake of nitrate, showed an inverse correlation with the risk of developing gastric cancer (24). These findings cast doubt on the importance of nitrates as a factor in the development of gastric cancer. Nevertheless, it should be emphasised that in the U.K. the largest proportion of dietary nitrate is derived from vegetables and that since these are abundant in vitamin C, they may prevent the formation of carcinogenic N-nitroso compounds by blocking the chemical reaction between nitrites and dietary amines in the stomach (25).

A high intake of salt may also be relevant to the development of gastric cancer. Salt has been shown to increase the uptake by the stomach of carcinogens in an animal model (26). Thus, in man, the salt in dried fish and other cured meats may be an important prerequisite to the carcinogenic effects of nitrites in the diet. The balance between dietary nitrite, ascorbic acid and nitrate is clearly of major importance and its complex interrelationship could account for the difficulties which still exist in this area of epidemiology.

However, there is other evidence favouring the Correa hypothesis. Conditions such as pernicious anaemia (27,28), hypogammaglobulinaemia (29) and gastric surgery for peptic ulcer (30,31) are associated with hypochlorhydria and predispose to gastric cancer. In a Scandinavian study, 10 (8%) of 123 patients with pernicious anaemia screened endoscopically were found to have gastric neoplasms. Five were carcinoid, 4 adenocarcinoma and 1 an adenomatous polyp (27). It has also been shown that the gastric juice of these patients contains bacteria capable of reducing nitrate to nitrite (32). Nevertheless, a recent study of the gastric aspirate from 3 groups of patients - 1 with pernicious anaemia, 1 following polygastrectomy and a control group, whilst confirming these findings, failed to show a direct correlation between an alkaline pH and an increase in the production of nitrosamines (16). Indeed the reverse applied - there was a definite trend in favour of increased nitrosamine production at an acid pH (16). These results suggest the formation of N-nitroso compounds in the stomach is more likely to occur at an acid pH which favours the chemical reaction between nitrites and amines (33), rather than at an alkaline pH which favours bacterial colonisation but not necessarily

nitrosation.

These results also imply that the Correa model for carcinogens in the stomach (20) is not totally accurate. Bacterial colonisation in an alkaline pH may not be an essential step. It could well be that the achlorhydria is a marker for abnormal gastric mucosa, such as the atrophic gastritis associated with aging or pernicious anaemia, and it is this alteration in the mucosal barrier rather than the bacterial colonisation which is important. Similarly, after gastric surgery patients develop dysplastic changes in the residual gastric mucosa as a result of bile reflux (34) which may predispose to gastric cancer

1.3 PATHOLOGY

Adenocarcinoma of the stomach accounts for 97% of gastric malignancies. There are several histological classifications of this tumour in current use. The classification devised by the World Health Organisation (WHO) recognises tubular, papillary, mucinous and signet ring forms of gastric cancer (35). It has not gained wide acceptance partly because many tumours have a mixture of more than 1 type, especially the tubular and papillary forms, but also because the 4 histological groups do not correlate well with known epidemiological data.

The most widely used classification was proposed by Lauren in 1965 (36). Lauren described 2 basic types of gastric cancer - intestinal and diffuse. The intestinal type is characterised by cells which grow in a tubular arrangement similar to small intestinal epithelium. They commonly have a striated border and are very cohesive, expanding into surrounding normal tissue in a well circumscribed fashion. There is little fibrous tissue reaction but often an inflammatory cell infiltrate. In the diffuse type the cells are usually single or arranged in indian file and infiltrate diffusely into surrounding tissue.

These cells may be devoid of cytoplasm or contain much cytoplasm leading to the so-called signet ring cell, where the nucleus is pushed to the side of the cell by the cytoplasm. There is usually a marked stromal reaction. However, in a considerable proportion of cases, approximately 15% (37,38), there is a mixture of intestinal and diffuse forms and in some reviews it has not been possible to classify a further 25% of cases (38).

It has been shown that these 2 histological types of gastric cancer, intestinal and diffuse, appear to have a distinctive aetiology (37). The diffuse type is taken to be the "endemic" form of gastric cancer. It affects both sexes equally and is associated with blood group A (37) and younger age groups (39,40). It appears to have a strong hereditary basis and has a similar incidence in high and low incidence areas (37,41). On the other hand the intestinal type is the "epidemic" form of gastric cancer (37,41). It is more common in males with a 2:1 male female ratio and is the most usual form of gastric cancer in the elderly (43). In countries with a high incidence of gastric cancer such as Japan, the increased number of cases are mainly of the intestinal type (37,41). These features point towards a strong environmental component in the development of intestinal type

of gastric cancer. However, it has not always been possible to demonstrate such a close correlation between the 2 different histological subtypes and these features. A recent report from the U.K. showed the reverse; there was a higher incidence of diffuse gastric cancer in the high risk areas for gastric cancer (38). One possible reason for this inconsistency is the heterogenous nature of the histology. In reality, the architecture of these tumours often have a mixture intestinal and diffuse forms and morphologically, commonly consist of both well and poorly differentiated cells, which must make histological assessment extremely difficult.

Kubo suggested that well differentiated tumours tended to be intestinal and poorly differentiated diffuse (43), and that these morphological characteristics are of more importance than the pattern of growth. Using this simple classification in Japan and New Zealand, he was unable to show any of the usual demographic and epidemiological correlations of the Lauren classification (44,45).

Ming has proposed a classification for gastric cancer with 2 histological categories; an expanding type and an infiltrating type (46). This classification, based on an analysis of 171

gastric carcinomas, has many similarities to that of Lauren, with the expanding type corresponding to intestinal type and infiltrating type corresponding to the diffuse type. Within the pattern of expanding or infiltrating types the cells can be well differentiated, poorly differentiated or undifferentiated. Ming also noted that age and sex distribution of expanding and infiltrating types were similar to the 2 histological subtypes of Lauren. Histologically the expanding type was commonly associated with intestinal dysplasia, which was rare with the infiltrating type.

The expanding type of gastric cancer constituted 67% of all cases. Of the expanding tumours 63% were fungating, 20% ulcerated, 10% polypoid, 4% superficial and 3% diffuse. The infiltrative carcinoma was diffuse in 68%, ulcerated in 27% and fungating in 5% of cases. Therefore, the histological type has a significant influence on the macroscopic appearance of the tumour. The infiltrative carcinoma with a diffuse macroscopic appearance gives rise to 'linitis plastica' or leather bottle stomach. The Ming classification does appear to have some merit although it offers relatively little advantage over the Lauren classification.

It is most important that a histological classification should be clinically and biologically relevant. Despite the obvious difficulties in classifying a proportion of cases, the Lauren classification is the closest to satisfying these criteria. Patients with the intestinal type of gastric cancer have a better prognosis than those with the diffuse type, independent of the type of surgical treatment (47). Also, with the the diffuse type microscopic infiltration occurs beyond the macroscopic boundary of the tumour therefore it is recommended that the surgical resection margins for diffuse tumours should be 5cm compared to the 2cm margin for intestinal tumours. Taking these factors and the epidemiological data into account I would suggest, where possible, the Lauren classification of gastric cancer should be used.

The most common sites for gastric cancer are the pylorus and pyloric antrum which account for 70% of cases. Carcinoma of the proximal third of the stomach and lower oesophagus now constitute up to 30% of cases compared to 15-20% 30 years ago (48). This change in the pattern of distribution may be related to the unique association between carcinoma of the cardia and lower oesophagus with Barrett's oesophagus (49). Thus the apparent

increase of carcinoma at these sites is due to a relative decline of carcinoma of the pylorus and pyloric antrum.

Barrett's oesophagus is characterised by a shifting of the squamo-columnar junction to at least 3cm above the lower most part of the oesophagus. Within the columnar epithelium there is usually gastric parietal and chief cells, junctional epithelium and epithelium which resembles intestinal mucosa with an abundance of mucous secreting glands (50,51). Epithelial dysplasia and carcinoma in situ are well described in Barrett's oesophagus (52,53) and result in it being a pre-malignant condition (52). Up to 86% of oesophageal adenocarcinoma are thought to arise in Barrett's oesophagus (52) and carcinoma is found in 7 - 46% of Barrett's oesophagus at diagnosis (53). The precise aetiology of Barrett's oesophagus is not established (53) although it is likely to be related to reflux of the gastric contents. Between 2 - 11% of patients with reflux oesophagitis develop Barrett's oesophagus (53) and a proportion of these will ultimately develop malignant change.

Efforts to ameliorate the risks of malignancy have produced the proponents of radical anti-reflux surgery because this is known to

reverse the epithelial change whereas medical treatment does not (55). However, since the risk of developing cancer in Barrett's oesophagus may be as low as 1 in 175 patient years (56), and the average age of patients with Barrett's oesophagus is 60 years, the decision regarding surgery is not clear-cut.

Predicting the patient most likely to develop malignant change within Barrett's oesophagus has been difficult. At the present time regular endoscopy for patients with severe dysplasia is appropriate. However, more sensitive markers are required. In the future, by examining endoscopic biopsies with oncogene probes such as c-Ha-ras p21, which is overexpressed in gastric carcinoma (57), it may be possible to detect those patients with Barrett's oesophagus at a high risk of developing malignancy.

1.4 MOLECULAR BIOLOGY

During the past 10 years there have been enormous advances in our understanding of the molecular biology of cancer. The learning curve seems to be exponential as we unravel the complex interrelationship between cellular oncogenes, viral oncogenes, polypeptide growth factors and cancer (58-61).

The proto-oncogenes are genes present in normal mammalian cells which are homologous with the viral genes responsible for the tumorigenic properties of RNA tumour viruses. They are also known as cellular oncogenes (59). In normal cells they are responsible for growth and maturation but when transferred to cell lines (transfection) they can promote the development of tumours. In addition, oncogenes are overexpressed in a variety of solid tumours and it has therefore been postulated that they are an integral part of the mechanism of carcinogenesis (58.59)

A number of oncogenes have now been identified in gastric cancer and gastric cancer cell lines. These include the genes c-Ha-ras, c-myc and c-erbB-2 (57,61-65). The c-Ha-ras p21 gene was found to have enhanced expression relative to normal tissues in a series of primary stomach

cancers and was also found in intermediate amounts in areas of dysplastic gastric mucosa (57). There was no correlation between tumour differentiation or depth of invasion and expression of c-Ha-ras although there was a tendency for more advanced tumours to express large amounts of c-Ha-ras (57). The presence of c-Ha-ras was demonstrated by in situ hybridisation using a monoclonal antibody to the oncogene on frozen and paraffin sections. Clearly this work needs further evaluation because it may have diagnostic and prognostic implications for the future. In addition the activation of the c-Ha-ras oncogene in a gastric cancer cell line was associated with a point mutation of the 12th codon (guanine to thymine) and this may be one of the ways in which the gene becomes oncogenic (62).

The c-erbB-2 oncogene is also amplified in gastric cancer. This oncogene codes for a protein which is similar to epidermal growth factor (EGF) receptor (65). The c-erbB-1 oncogene codes for EGF receptor and the possession of EGF receptor is known to be a poor prognostic factor in breast cancer (66). It is therefore of particular interest that EGF has also been found in primary gastric cancers and that the prognosis was worse for patients whose tumours elaborated EGF (67). The promoter regions for c-erbB-1 (68) and c-erbB-

2 (69) have recently been characterized. These areas regulate the production of receptors by the gene and therefore are targets for highly specific monoclonal antibody therapy. If the monoclonal antibody could be internalised within the nucleus without degradation it could effectively switch off receptor production and possibly control tumour growth.

The potential use of molecular biology in the diagnosis and treatment of cancer is enormous. However there is a caveat; oncogene over expression may only be an epiphenomenon - a marker of rapidly growing cells - and as such several steps removed from the fundamental initiation of carcinogenesis. Therefore it is wise to remain somewhat circumspect about their future role.

1.5 NATURAL HISTORY

In Europe and the USA gastric cancer presents late in its natural history. By the time of diagnosis the majority of patients have locally advanced disease or have evidence of metastases. Presenting symptoms related to gastric cancer are non-specific and include upper abdominal pain (78%), weight loss (67%), vomiting (48%), anorexia (45%) and weakness (42%) (70). These symptoms may predate the diagnosis by 12-24 months but patients usually present within 12 months of their onset. Once suspected the diagnosis can normally be confirmed using either barium studies or endoscopy (71,72).

Gastric cancer in general, is an intra-abdominal disease. The tumour typically spreads by local invasion of the stomach and surrounding structures. It metastasises to local and then regional lymph nodes and often spreads by the transcoelomic route to the peritoneum and omentum or occasionally to the ovaries giving rise to a Kruckenberg tumour. Haematogenous spread to the liver is via the portal vein. At laparotomy up to 70% of patients will have evidence of tumour in the regional lymph nodes, 25% will have peritoneal involvement and a similar proportion will have

liver metastases (70). There is direct infiltration of the pancreas in 23% and of the colon in 5%. The net effect of these findings is that relatively few patients have tumours which can be surgically cured despite the absence of metastases outwith the abdomen. Only 5-10% of patients will be alive 5 years after diagnosis and the majority of patients will die within the first 2 years (73-76).

The pattern of recurrence at post mortem reflects the original mode of dissemination of the tumour, except that liver metastases are more frequent (54%) and that lung metastases can be detected in almost 25% of cases (76). The pattern of failure after successful surgical removal of the primary tumour was studied by Gunderson and Sosia (77). In this series, 107 patients were subjected to periodic re-operation. Eighty-six developed recurrent disease and of these 87% had local-regional recurrence which was defined as tumour in the gastric bed, lymph node recurrence or a localised peritoneal recurrence. Distant metastases were present in 30% of patients but were the sole site of recurrence in only 5%. These findings suggest some potential for the improvement of the local control, and this will be amplified in Chapter Two.

While the rest of the World has made very little progress in the treatment of gastric cancer, the Japanese have engineered a remarkable triumph. This triumph is, at least in part, related to the point at which the diagnosis is made in the natural history. Intuitively the diagnosis must be made earlier than in Europe and the USA, which is reflected in the higher proportion of early gastric cancer diagnosed in Japan. This ultimately translates into survival advantages for the patient; the 5 year survival for patients with gastric cancer in Japan is now 50% (78). The earlier presentation in Japan can be explained by several factors. The Japanese screening programme undoubtedly leads to the detection of a significant proportion of early gastric cancer. However it may also serve to increase public awareness of gastric cancer and therefore encourage the Japanese individual to seek medical advice and investigation of dyspeptic symptoms earlier than his European counterpart.

However, there is some hope on the horizon for the UK. The study from Birmingham (79) and the report from Bristol (80) both suggest a higher proportion of early gastric cancer is now being diagnosed in this country, leading to a higher

resection rate and in the long term hopefully improved survival.

1.6 EARLY GASTRIC CANCER AND SCREENING FOR

GASTRIC CANCER

These 2 subjects are logically considered together because the main purpose of screening is to detect early gastric cancer, which can be readily cured by surgery.

Early gastric cancer is defined as tumour which has not penetrated the submucosa. Lesions may be protuberant, elevated, flat, depressed or excavated (81). Their average diameter is between 2-3 cm (42) but lesions as large as 12.5 cm may occur (82). This has led some to suggest because these tumours can be slow growing (83), almost regardless of when the diagnosis is established, their natural history is such that they remain confined to the submucosa and are therefore easily cured by surgery (84). However, this seems unlikely because the Japanese have increased the proportion of cases with early gastric cancer from 1% before 1951 (85) to over 40% in a population exposed to a screening programme with a concomitant improvement in overall survival (86).

In Japan over 4 million people are screened each year, with the detection of around 4000 new cases of gastric cancer (86). Indeed, it has become Japanese public health policy to make one

screening investigation available to all adults over the age of 40 years once every 3 years. The screening method involves the use of a double-contrast barium meal in conjunction with photo-fluorographic apparatus that takes 5-7 exposures per examination. The examination usually takes place in a mobile unit staffed by skilled technicians. Suspicious cases are referred for endoscopy. Although it seems likely that screening improves survival (86), in the absence of any randomised trial of screening versus no screening, it is not possible to unequivocally prove this assertion. At the present time case-control studies are used as an alternative to the randomised trial. In a recent case-control study from Nosetown in Osaka, the odds ratio of dying from gastric cancer in screened compared with unscreened individuals was 0.595 for males and 0.382 for females thus supporting the notion that screening improves mortality rates (86).

In the United Kingdom it is now accepted that early gastric cancer is identical to the Japanese variety (87). In the 1960s the detection rate for early gastric cancer was as low as 0.7% (88) although more recent data puts it at 10% (89). In Bristol the number of early gastric cancers doubled during the two ten year periods of

1965-75 and 1975-85. The survival rate of the patients with early gastric cancer was extremely good; 71% were alive at 5 years age (adjusted mortality 92%) (89).

However, although the screening programme has been successful in Japan the detection rate is low (1 in 1000 screened) and in the context of the lower rates of gastric cancer in the U.K. screening of all the population would be difficult to justify (90). Therefore an alternative has been to set up a clinic where only those patients with an exaggerated risk for the development of gastric cancer are endoscoped (79). In Birmingham all patients, from 6 general practices, who were over the age of 40 years and complained of dyspepsia were referred to a dyspepsia clinic for endoscopy. Over a 2 year period 15 gastric cancers were diagnosed and 2 (15%) had early gastric cancer (79). Sixty seven per cent of patients had a radical resection which is higher than the 18% reported previously from the same group (77). Nevertheless, it has been argued that even without a screening clinic the detection of early gastric cancer has increased (80) but our own data (Chapter Two) would not support this. The more frequent use of endoscopy in the latter part of the study was not associated with an improvement

in survival. This favours the notion that endoscopy outwith the context of a screening clinic may not be sufficiently sensitive. Therefore screening at risk groups such as those over 40 years of age presenting with a history of peptic ulcer surgery would seem appropriate. As with Barrett's oesophagus, oncogene probes (57) could be used to examine endoscopic biopsies, particularly those taken from patients with a previous history of peptic ulcer surgery or those in whom the initial endoscopic screen shows dysplastic changes.

1.7 SURGICAL TREATMENT AND STAGING

During the past 50 years there have been many reviews of the surgical treatment of gastric cancer which have produced remarkably similar findings. A summary of 8 studies (47,73-76,91-93) is shown in Table 1.3. Almost 80% of patients will have an operation. The remaining patients will be deemed unsuitable for surgery because of metastatic disease or because they are considered a poor operative risk. Approximately 60% of patients will have the tumour resected. In some of the reviews it is not stated what proportion of these resections are considered curative, but usually this applies to 50% of cases. In any event the assessment of curative resection is very subjective and may not accurately reflect the adequacy of the surgery. Laparotomy and biopsy or a palliative bypass procedure is performed in 20% of cases. The results for surgery have remained remarkably static during the 30 past years. The only real area of improvement has been the reduction in the operative mortality rates. These have declined in one group's experience from 22.5% in the period 1969-77 to 11.76% in the period 1978-82 (91). Gilbertson reviewed the surgical experience in gastric cancer of the

No. of patients	Operation rate %	Resection rate %	5 year survival %
7044	79	62	12

Table 1.3 Results of surgery taken from 8 studies
(1938-1982)

University of Minnesota between 1936-1963. This consisted of 1983 cases (75). He concluded that more extensive surgical excisions had not led to an improvement in survival. Indeed, he argued that patients fared worse under an aggressive surgical policy; the 5-year survival of patients having a curative resection between 1950-58 was 28% compared to a 17% survival for 1958-63 when more radical surgery was performed. This finding was explained by an increase in operative mortality for partial gastrectomy (25.6%) and total gastrectomy (33.3%) during the second period. However, with the recent improvements in postoperative care this argument may now be spurious.

The Japanese experience with gastric cancer is considerable and the results which they have obtained with surgery are much better than has been achieved in the USA or Europe. In a review of 4946 patients treated surgically in the National Cancer Centre, Tokyo between 1962-84 Maruyama (78) reported that in 4734 patients (95.4%) underwent tumour resection and that the 5 year survival for these patients was 51%. The postoperative mortality rate was only 1.6%. Admittedly over 50% of patients had tumours which had not penetrated the serosa but even so the results are impressive.

What can be learned from the Japanese experience? The emphasis must be on early diagnosis and low postoperative mortality. The Japanese perform, by UK standards, radical surgery (94) but their results cannot be taken to indicate that a radical approach is necessarily best because there are no randomised trials in the Japanese literature which have addressed this issue. Indeed such studies are sadly lacking in the world literature and hopefully the MRC Surgical Gastric Cancer Trial may help resolve this problem.

A detailed technical account of the of the surgical approaches to gastric cancer is beyond the scope of this thesis and the reader is referred to a recent review by Cuschieri (95). The 2 basic types of operation for gastric cancer are subtotal resection and total resection of the stomach. Subtotal resection can be curative for antral tumours but for proximal tumours total resection is likely to be necessary. Both may be combined with an R1 lymph node resection (left and right cardiac nodes, nodes along the greater and lesser curvature) or an R2/3 resection (R1 plus gastric artery nodes, coeliac artery nodes, common hepatic artery nodes, lymph nodes of the splenic hilum, supra and infra pyloric nodes, splenic artery nodes, lymph nodes in the hepato-duodenal

ligament, retro-pancreatic nodes, lymph nodes at the root of the mesentery, diaphragmatic and para-oesophageal nodes), splenectomy and for all except antral lesions the removal of the distal half of the pancreas. Clearly the operative mortality for the more extensive surgical procedures is higher and this must be carefully balanced against the improved outlook of survivors.

The staging of gastric cancer is extremely important because it allows valid comparisons of the results from different centres and different surgical approaches to this tumour. In 1984 a new staging system was devised to replace the 3 other systems in current use (UICC, The Japanese Research Society for Stomach Cancer (JRSSC) and The American Joint Committee on Cancer, for details see reference 95) and thus unify the description of gastric cancer in the USA, Europe and Japan. Essentially it is a TNM system which recognises 4 stages of gastric cancer (78). Its application depends upon the surgeon following the guidelines of the JRSSC (94) coupled with a careful histological analysis of the resected tumour and regional lymph nodes. It is summarised in Table 1.4 and the corresponding survival figures from Maruyama (78) show that the new staging system effectively separates survival by

Stage		% surviving at 5 years
I	Ia T1 N0	88
	Ib T1 N1	
	T2 N0	
II	T1 N2	65
	T2 N1	
	T3 N0	
III	IIIa T2 N2	38
	T3 N1	
	T4 N0	
IV	Any M1	5

T - Primary Tumour

T1:Tumour limited to the mucosa and submucosa

T2:Tumour involves the subserosa

T3:Tumour penetrates the serosa

N - Regional Lymph Nodes

N0:No metastases to regional lymph nodes

N1:Involvement of perigastric lymph nodes
within 3cm of the primary tumour

N2:Involvement of regional lymph nodes more
than 3cm from the primary tumour

M - Distant Metastases

M0:No evidence of distant metastases

M1:Evidence of distant metastases

Table 1.4 The new TNM Classification with
survival according to Maruyama (78)

stage. However the detailed nature of the lymph node sampling must inevitably mean that this staging system is unlikely to gain wide acceptance for routine use in the UK but will nevertheless be crucial to the conduct of future clinical trials.

1.8 CHEMOTHERAPY

During the past fifteen years there has been a sharp increase in the use of chemotherapy to treat disseminated gastric cancer. Initially the chemotherapy of gastric cancer consisted of single agent treatment, usually 5-FU. In the mid 1970s combination chemotherapy gained momentum as a sequel to the promising results obtained from the combination of 5-FU and the nitrosureas. There are now several different combinations of drugs in use which along with single agent data will be reviewed in this section.

1.8.1 Single Agent Chemotherapy

5-Fluorouracil (5-FU) has been the most widely used agent in gastric cancer and has an overall response rate of 22% with a response duration of between 4 to 5 months (96). The anti-tumour effect of 5-FU has not been improved by giving the drug using a relatively intensive schedule of 15 mg/kg/24hr x 5 i.v., then 7.5 mg/kg on alternate days to toxicity (97), nor by administering the drug as a continuous infusion (30 mg/kg/24hr) for 72 hours every 2 weeks (98). However, this latter method is associated with

virtually no systemic toxicity and may be worth exploring in combination with other agents. Otherwise, the most useful way of giving 5-FU is as an intravenous bolus at a dose of 750 mg/m² every week.

Mitomycin-C is an antibiotic alkylating agent which was developed in Japan. It has comparable activity to 5-FU, with a response rate of 24% (99) but is more toxic. In particular, its use is associated with cumulative myelosuppression which most commonly manifests as thrombocytopenia. Therefore the drug is now usually given as an intravenous bolus every 6 weeks. Another rather disconcerting finding is that mitomycin-C appears capable of inducing the haemolytic-uraemic syndrome. Fielding reported this condition in 8.5% of 251 patients receiving adjuvant mitomycin-C and 5-FU (100).

Adriamycin (doxorubicin) was investigated as a single agent for gastric cancer in the late seventies and was found to have modest activity, ranging from 15-24% (101,102). It has subsequently been incorporated into many combination regimens but is not without toxicity. Alopecia is virtually universal following its use although this may be prevented in some patients by the use of scalp cooling. Cumulative cardiotoxicity also occurs at

doses above $550\text{mg}/\text{m}^2$, but in general this is not a practical clinical problem in patients receiving what is ostensibly palliative treatment. Epirubicin a derivative of adriamycin which is less cardiotoxic has similar activity to the parent compound and in general offers little advantage (103). Mitozantrone a bis-substituted anthraquinone that binds to DNA, showed no activity in a group of 32 patients with advanced gastric cancer (104). This is disappointing because, in general, mitozantrone shares the same spectrum of activity against tumours as adriamycin with the advantage of not producing alopecia.

The nitrosoureas, carmustine (BCNU) and semustine (methyl-CCNU) have also been studied in gastric cancer. Methyl-CCNU has been used in combination with other cytotoxic drugs in a number of studies (102,105) and yet there is little evidence that it is an effective single agent. Moertel found only 8% of 37 patients responded (106). BCNU in limited testing had moderate activity (18%) in gastric cancer (107). The major drawback of the nitrosoureas is that they are emetogenic which presents a particular problem for patients with upper gastrointestinal malignancies. Other cytotoxic agents such as methotrexate have received remarkably little attention. A recent

report from the Gastrointestinal Tumour Study Group (GTSG) suggests little activity for methotrexate in patients who had been treated with previous chemotherapy but did show promising results for another folate antagonist triazinate (108).

Cisplatin has only recently been recognised to have important activity in gastric cancer (109-111), with response rates of 25% in studies which included patients previously treated with combination chemotherapy regimens. This is clearly encouraging and requires further investigation. Unfortunately, cisplatin is the most emetogenic of all the cytotoxic drugs in current use. Nevertheless, with modern antiemetics it should be possible to control the emesis and the most effective antiemetic for this purpose is high dose metoclopramide (see Chapter Three). Therefore the use of cisplatin should not be automatically excluded on the basis of vomiting.

Carboplatin, an analogue of cisplatin with less gastrointestinal and renal toxicity, has been tested in a small number of patients with gastric cancer but so far results are disappointing and suggest no significant activity (112). We have evaluated the cisplatin analogue TNO-6 (cis-1,1 di(aminomethyl)-cyclohexane sulfate

platinum II) in the treatment of gastric cancer (113) and the results will be presented in Chapter Three.

1.8.2 Combination Chemotherapy

Following the identification of a few moderately active agents against gastric cancer the search for more effective combination chemotherapy regimens began. In particular, it was hoped that using drugs in combination would improve not only response rates but also the duration of response which, for single agent chemotherapy, was usually only a few months (96). Moertel (106) reported a randomised study of 5-FU and methyl-CCNU versus methyl-CCNU alone in which response to the combination was 40%. Moreover, patients survived significantly longer following treatment with 5-FU and methyl-CCNU than with methyl-CCNU alone. However, subsequent experience with this combination failed to confirm these results and response rates were equivalent to single agent 5-FU (101,114). Adriamycin has been added to the combination of 5-FU and methyl-CCNU (102,105,115). When the results of these studies are combined the response rate is only 35% (14 out of 40 patients with measureable disease). Levi et

al (116) treated 35 evaluable patients with the combination of 5-FU adriamycin and BCNU (FAB) and found 18 (51%) of patients responded with a median duration of response of 10 months. However, a subsequent randomised study from this group showed no difference in the survival of patients treated with single agent adriamycin compared with FAB (117). Also Schnitzler et al (118) randomised 77 patients to either FAB or 5-FU and BCNU in combination (FB) and reported that only 24% of patients responded to FAB and 11% responded to FB with no difference in survival between the 2 groups.

Because cisplatin has shown such promising activity as a single agent (109-111) it has been incorporated into a number of combination chemotherapy regimens. Wagener et al (119) treated 20 patients with 5-FU, adriamycin and cisplatin. Nine (50%) of the 18 evaluable patients entered partial remission and 8 patients had stable disease. Similarly the combination of cisplatin, adriamycin and etoposide produced objective tumour regression in 10 (62.5%) of 16 patient (120). Unfortunately success with cisplatin containing regimens has not been universal. A study from the USA (121) which investigated 5-FU, adriamycin and cisplatin revealed that only 10 (29%) of 35 patients responded and a further study in which

cisplatin was combined with etoposide showed only 1 of 33 patients responded (122).

Triazinate the anti-folate has been combined with mitomycin-C to treat advanced gastric cancer. Using this combination O'Connell et al (123) at the Mayo Clinic have treated 33 patients, 29 of whom had failed previous chemotherapy. Nine (27%) of patients had a partial response. This is a particularly interesting study because of the high proportion of heavily pre-treated patients who went on to respond to second line therapy.

The most widely used regimen in gastric cancer is the combination of 5-FU, adriamycin and mitomycin-C (FAM) (124). This regimen was developed on the basis that it utilised 3 of the most active drugs against gastric cancer. Preliminary results suggested that over 50% of patients would respond to the combination but subsequent experience has shown the figure to be nearer 40%. A detailed review of this regimen will be undertaken in Chapter Three.

Also in Chapter Three, we will review the results of the most promising new regimen from the 1980s - 5-FU, high dose methotrexate and adriamycin (FAMTX) (125). This is an interesting combination which exploits the synergism of 5-FU

and methotrexate but unfortunately it appears to be particularly toxic.

1.8.3 Adjuvant Chemotherapy

Despite the relatively low activity of single and multiple chemotherapy regimens in advanced gastric cancer there have been a number of studies which have investigated the role of chemotherapy as an adjuvant to surgery.

As yet there is no convincing evidence that chemotherapy can improve the survival of patients after surgical resection of the primary tumour. In a large randomised series (616 patients), in the mid sixties, the Veterans Administration failed to show any benefit from intra-operative and adjuvant thiotepa - indeed the treatment group fared slightly worse (126,127). They also studied adjuvant floxuridine in 400 patients but again showed no difference in survival between control and treatment groups (128).

5-FU in combination with methyl-CCNU has been reported in 3 large series (129,130,131). The first of these from the GTSG showed significant benefit (40 deaths in 71 control patients; 29 deaths in 71 treated patients) at a median follow time of 48 months (129). However, in a similar

study where 200 patients were randomised to intravenous 5-FU and oral methyl-CCNU for a period of 2 years, or no treatment after surgical resection, there was no significant difference in disease-free survival or overall survival between the 2 groups (130). Moreover, 2 patients in the treatment group developed non-lymphocytic leukaemia probably as a result of exposure to methyl-CCNU (132). The third study of 5-FU and methyl-CCNU whilst showing an early survival advantage for the treatment group, showed no difference in survival between treatment and control groups at a median follow up of 42 months (131).

The British Stomach Group has conducted a randomised trial of 5-FU and mitomycin-C against no treatment after surgery. They were unable to show any overall survival benefit from chemotherapy in this large well designed study which accrued over 400 patients (133).

More recently, FAM has been used as an adjuvant to surgery in a multicentre study (134). Three hundred and thirty patients have been randomised to FAM or a non-treatment control arm. The interim report presented at ASCO in 1986 showed no significant difference in survival between the 2 groups and this trend continues (RC

Coombes Personal Communication)

Not surprisingly there have been a number of adjuvant studies from Japan. However, because adjuvant chemotherapy is now established as a standard treatment following surgery in many centres in Japan, the most recent studies do not possess non-treatment control arms (135,136). This is disappointing because many of the earlier studies in the Japanese literature, taken to indicate definitive evidence of the value of adjuvant chemotherapy show only marginal benefits. Imanga and Nakazota reported benefit from adjuvant mitomycin-C (0.08 mg/kg x 2 weekly for 5 weeks after surgery) in a randomised study of 520 patients with a 5 year survival of 68% in the treatment group compared to 54% in the control group (137). However these results could not be reproduced in a subsequent phase of the study where the same treatment formed one of 3 arms. The other 2 arms were mitomycin-C, 5-FU and cytosine arabinoside and a control arm. Survival was not significantly improved with either chemotherapy (137). In a recent study from Japan the survival of 2873 patients was analysed after random allocation to one of 3 treatments (136). Group A were given bolus mitomycin-C with no further therapy. Group B were given bolus mitomycin-C and

oral fluorouracil for 1 year and group C were given oral fluorouracil alone for 1 year. There was no difference in the 5 year survival of the 3 groups. Considering that a single dose of mitomycin-C is fairly close to a control arm these results suggest adjuvant treatment is not producing major improvement in the post operative management of gastric cancer.

At the present time there is no clear evidence that adjuvant chemotherapy is of value in gastric cancer. It is possible that only patients with a high risk of recurrent disease (such as those patients in whom the tumour has penetrated the serosa) should receive adjuvant chemotherapy in clinical trials, since this is the group most likely to benefit from this approach. Also, it is likely that more active cytotoxic drugs need to be identified in advanced disease before adjuvant treatment adopts an effective role.

1.9 RADIOTHERAPY

There is relatively little information on the efficacy of radiation therapy in the treatment of gastric cancer. Many reviews (138,139) cite the experience of Wieland and Hymmen who reported an 11% three year survival and a 7% five year survival in 82 patients treated with radiation (60Gy) alone (140). These results published in 1970 are taken to show that radiation therapy is effective in gastric cancer. They are further supported by the findings of Hoshi who gave radiation preoperatively and reported histological evidence of radiation damage to the resected tumour in 25-88% of cases depending on the dose of radiation administered (141). However, Falkson and Falkson found no responders in a group of patients with measurable lesions treated with radiation alone, compared to a 17% response rate for patients treated with 5-FU and a 55% response rate for patients treated with a combination of 5-FU and radiation (142).

The promising results from the combination of radiation and 5-FU were also shown by a study from the Mayo Clinic in which patients with unresectable gastric cancer were randomly allocated to radiation therapy (35-40 Gy) and

placebo or the same dose of radiation plus 5-FU given at a dose of 15 mg/kg daily over the 3 days prior to radiation (143). Forty eight patients entered the trial; the mean survival for the combined treatment arm was 13 months and 3 patients were alive at 5 years compared to a mean survival of 5.9 months for the radiation and placebo arm and all patients in this group were dead at 5 years (143).

More intensive chemotherapy regimens have subsequently been used in combination with radiation therapy. One study randomised 90 patients with locally unresectable gastric cancer to receive chemotherapy consisting of 5-FU and methyl-CCNU or radiation (50 Gy) given concurrently with 5-FU and then later the same combination of 5 FU and methyl-CCNU (144). Although, there was a median survival advantage for the chemotherapy alone group (70 weeks versus 36 weeks for the combined modality group) the analysis at 5 years favoured the combined modality treatment; 16% of the combined treatment arm were alive compared to 7% in the chemotherapy arm (144). The reason for this was the early, higher mortality related to the combined modality therapy. In particular during the first 5 months of this treatment there were more deaths due to

septicaemia in the combined modality arm.

A recent randomised study from the Eastern Cooperative Oncology Group (145) failed to confirm the Mayo Clinic data (143). Fifty seven patients with histologically confirmed, unresectable gastric cancer were allocated to 5-FU 600mg/m² once weekly or radiation therapy (40 Gy) combined with 5-FU 600mg/m² IV for the first 3 days of radiotherapy and then 5-FU 600mg/m² IV weekly. The survival of both treatment groups was almost identical; for 5-FU alone the median survival was 9.3 months, 5-FU plus radiation 8.2 months. Toxicity was more marked with combined modality treatment (145).

The toxicity related to combined modality therapy was also highlighted in a study from O'Connell et al (146). In this pilot study 18 patients with locally advanced gastric cancer were treated with 5-FU and adriamycin followed by concurrent radiation and 5-FU, and then maintenance therapy with 5-FU, adriamycin and methyl-CCNU. Patients experienced severe and prolonged nausea, weight loss and sepsis, and despite the achievement of local control of tumour all but 2 patients had progression of distant metastases.

Intra-operative radiation treatment has been

pioneered by Abe et al (147,148). The radiation is delivered as a single fraction (30-40 Gy) to the tumour bed and surrounding lymph glands. In a non-randomised trial (patients received intra-operative radiation therapy depending on the day of surgery) there were survival advantages for the group treated with intra-operative radiation therapy. This advantage was particularly marked in those patients where the tumour had penetrated the serosa and invaded surrounding structures such as pancreas. For this group the 5 year survival was 27% when treated with intra-operative radiation therapy compared to 0% when treated by surgery alone (147). These results have yet to be confirmed in a randomised trial.

There are few data on adjuvant radiation therapy in the treatment of gastric cancer. The British Stomach Group are currently evaluating this therapeutic approach and the results are awaited with interest (138).

Clearly, the precise role of radiation therapy in the management of gastric cancer has not yet been established. Combined modality treatment results in an escalation of toxicity with no proven survival advantage. Intra-operative radiation therapy is of interest but its efficacy remains to be confirmed in randomised trials.

CHAPTER TWO

PROGNOSTIC FACTORS IN
GASTRIC CANCER

2.1 INTRODUCTION

Although there have been a number of studies of the natural history and surgical treatment of gastric cancer from the U.S.A. and Europe, many use data derived from the 1930's and 1940's (73-76) or use data compiled from several centres (70). There are relatively few contemporary natural history studies (91) and we know of none from Scotland. Endoscopy became widely available in the 1970s and the effect that this technique may have had on the diagnosis and treatment of gastric cancer deserves investigation. This fact, coupled with the apparent gap in the literature of a natural history study from Scotland prompted this retrospective review. The present study describes the natural history of gastric cancer in a group of patients diagnosed consecutively in one centre in Glasgow (149).

2.2 PATIENTS AND METHODS

We reviewed the clinical records of all patients with histologically proven gastric adenocarcinoma diagnosed in Glasgow Royal Infirmary (G.R.I.) between January 1st 1974 and December 31st 1984. We included patients with tumour of the cardia which had spread to involve the oesophagus. All patients were registered with the West of Scotland Cancer Surveillance unit which records the date of diagnosis, date of death and the cause of death in all cancer patients from this region. The following data were abstracted: duration and type of symptoms, details of operative procedure, pathology of the resected specimen, and details of any chemotherapy. The sites of recurrent disease were recorded. Local relapse was defined as tumour recurrence identified endoscopically or by barium studies or tumour recurrence within the gastric bed (identified at re-operation, post mortem, or by abdominal ultrasound or C.T. scan). Distant relapse was defined as tumour recurrence at all other sites including distant peritoneal spread and liver metastases.

Patients with advanced gastric cancer were defined as those in whom it was not possible to

effect a surgical cure. Curative resections were defined as those in which there was no residual macroscopic disease after surgery.

Statistical Analysis

Data were analysed on the Glasgow University Computer (ICL 2976). Survival was analysed by the life table method. Using Cox's Regression Model (150), the effects on survival of the following independent variables were examined: duration of symptoms before diagnosis, type of surgery, lymph node involvement, serosal involvement, resection margin involvement, site of primary tumour, presence of liver metastases and tumour histology.

2.3 RESULTS

During the 11 year period 330 patients were diagnosed as having histologically confirmed gastric cancer in G.R.I.. Adequate clinical records and follow up were available for 328 patients and the remaining 2 patients have therefore been excluded from analysis. The diagnosis was rare under the age of 45 years and the male to female ratio was 1.45:1 (Table 2.1). At presentation (Table 2.2), 30% of patients had an abdominal mass and 6% were jaundiced.

Surgical Procedures

All except 51 patients (15.5%) had some form of surgical treatment. The types of operation and survival of patients for each subgroup are shown in Table 2.3 and Figure 2.1. Almost all of the long term survivors came from the curative resection group, a group which survived significantly longer ($p < 0.001$) than any other. Patients who had a palliative gastroenterostomy did not survive significantly longer than those having laparotomy alone, Celestin tube insertion or no surgery. The 11 years were arbitrarily divided into 2 periods. There was no change in the proportion of patients undergoing each surgical

Age (years)	Male (No.)	Female (No.)
<45	10	5
45-54	32	5
55-64	50	33
65-74	71	54
>75	31	37
Total	194	134

Table 2.1 Characteristics of 328 patients with gastric cancer

Presenting symptoms	Number of patients (%)
Anorexia and weight loss	272 (83)
Dysphagia and vomiting	210 (64)
Dyspepsia/abdominal pain	211 (64)
Unexplained anaemia	92 (28)
Emergency bleed/perforation	55 (13)

Table 2.2 Presenting symptoms in 328 patients with gastric cancer

Primary treatment	No. of patients (%)	Percentage surviving at years					Median survival (months)
		1	2	3	4	5	
Curative resection	128 (39.0)	63	47	36	30	24	21
Palliative resection	32 (9.8)	27	14	3	3	0	6
Gastroenterostomy	33 (10.1)	8	0	0	0	0	4
Celestin tube	26 (7.9)	8	4	4	4	0	2
Laparotomy alone	58 (17.7)	9	0	0	0	0	2
No surgery	51 (15.5)	12	6	3	3	3	1

Table 2.3 Survival of patients with gastric cancer according surgical to procedure

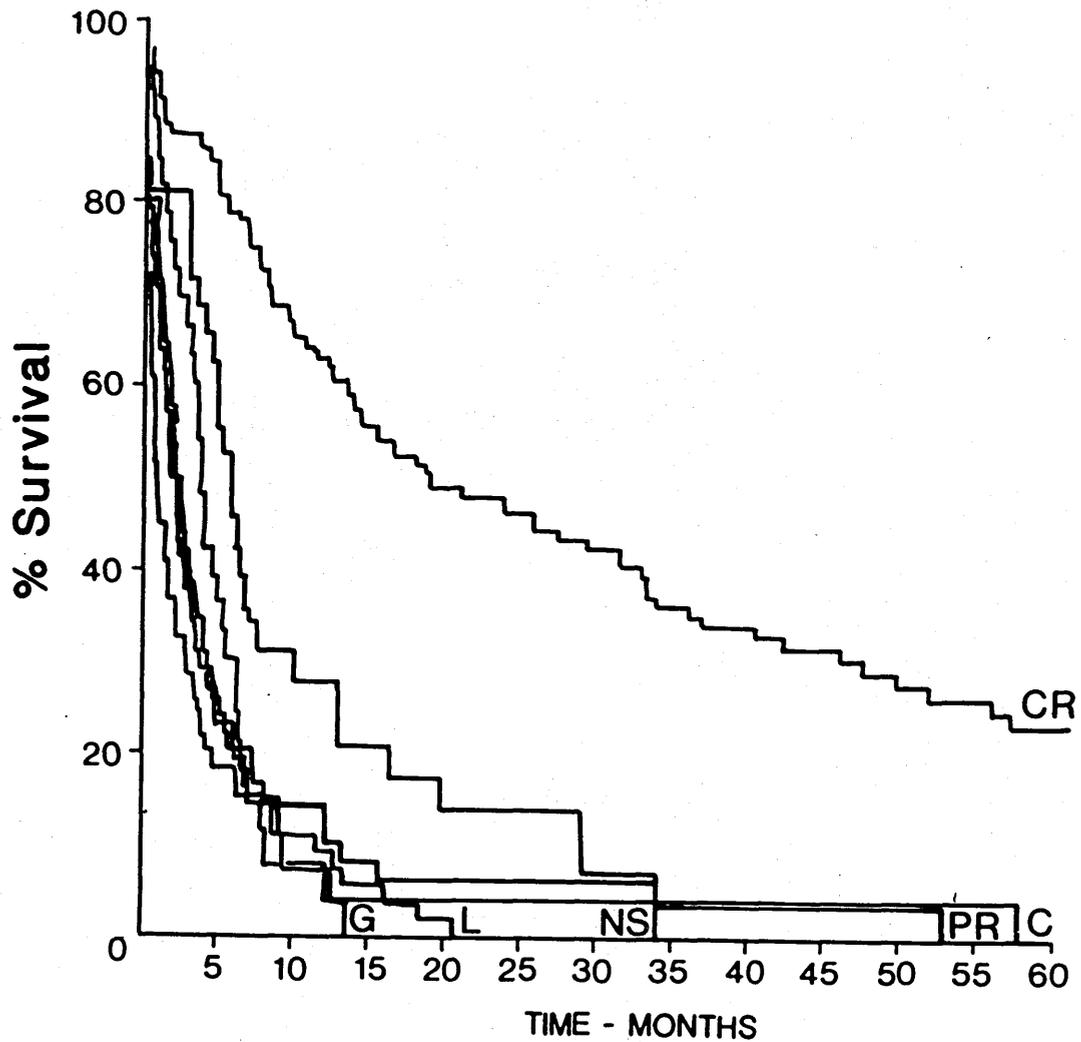


Figure 2.1 Survival by surgical procedure in 328 patients with gastric cancer. (CR, curative resection:n=128; PR, palliative resection:n=32; G, gastrectomy:n=33; C, celestin tube:n=26; L, laparotomy:n=58; NS, no surgery:n=51). Survival of CR versus PR: $p < 0.001$. Survival of PR versus G+L+NS: $p < 0.01$

procedure during the 2 periods. In particular, the number of patients having curative resection remained the same (Table 2.4). However, operative mortality (death within 30 days of surgery) for the curative resection group changed from 17.6% between 1974-79 to 8.4% between 1980-84.

We have not analysed operative mortality for the other groups because it was often extremely difficult to differentiate between deaths due to operation and those due to progression of disease.

Survival of Curative Resection Group

Despite curative resection, only 24% of these patients were alive 5 years after diagnosis (Table 2.3). To define the prognostic factors for this group, we examined the effects of site of primary tumour, serosal margin involvement, resection margin involvement and lymph node involvement (as determined by histological examination of the resected specimen) on survival. The results are shown in Figure 2.2 and in Table 2.5. All patients with resection margin involvement relapsed and died. Otherwise, serosal involvement was the best predictor of survival ($p=0.0004$). Indeed, all but 5 patients with resection margin involvement had serosal involvement. Lymph node involvement was only a significant factor when the

Operative procedure	Period	
	1974 - 1979	1980 - 1984
Curative resection	39.2	38.6
Palliative resection	10.8	9.0
Gastroenterostomy	8.5	11.6
Celestin tube	8.5	7.9
Laparotomy alone	22.3	14.8
No surgery	10.8	18.0

Table 2.4 Operative procedure by period

	No. of patients	Percentage surviving at years:					p value
		1	2	3	4	5	
Serosa							
Positive	90	54	35	26	18	13	0.0004
Negative	38	85	78	59	59	47	
Margins							
Positive	33	41	7	7	0	0	0.0005
Negative	95	69	58	43	36	28	
Nodes							
Positive	65	55	37	27	22	17	0.04
Negative	63	71	57	45	37	31	

Table 2.5 Survival by serosal involvement, resection margin involvement and lymph node involvement in 128 patients undergoing curative resection for gastric cancer

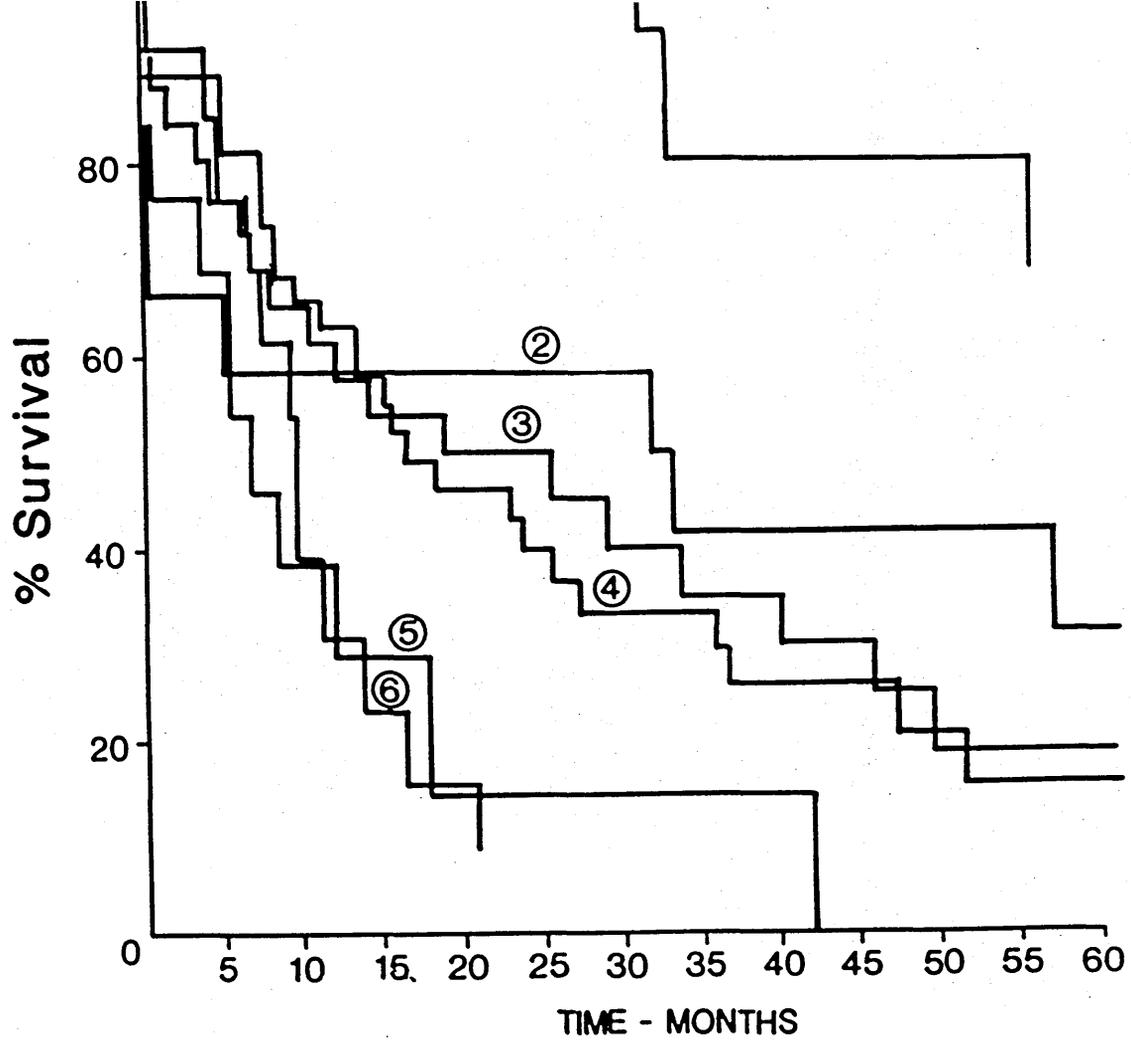


Figure 2.2 Survival by serosal involvement (S), resection margin involvement (M) and lymph node involvement (N) in 128 patients having a curative resection for gastric cancer

	S	M	N	Number
1	-	-	-	21
2	-	-	+	12
3	+	-	-	26
4	+	-	+	38
5	+	+	-	13
6	+	+	+	13

1+2 versus 3+4, $p=0.02$

3+4 versus 5+6, $p<0.01$

Lymph node	Resection		No.	Percentage survival	
	margin	Serosa		2 years	5 years
+ve	-ve	-ve	12	58	31
-ve	-ve	-ve	21	100	70
+ve	-ve	+ve	38	40	16
-ve	-ve	+ve	26	50	18
+ve	+ve	+ve	13	5	0
-ve	+ve	+ve	13	16	0
+ve	+ve	-ve	2	-	-
-ve	+ve	-ve	3	-	-

Table 2.6 The effect of lymph node involvement on survival in 128 patients undergoing curative resection for gastric cancer

serosa and margins were not involved (Table 2.6). Survival related to the site of primary tumour is shown in Table 2.7. Survival was poorer ($p < 0.05$) for patients with tumours of the cardia compared to the body or antrum. This was mainly because resection margin involvement was more frequent with tumours of the cardia (39.5%) than with tumours of the body (16.3%) or antrum (21.4%) ($p = 0.04$). The degree of differentiation of the cancer had no significant influence on survival (Table 2.8), although patients with well differentiated tumours had a better prognosis.

Endoscopy as a means of establishing the diagnosis had no effect on survival: the 102 patients undergoing endoscopy had a median survival of 663 days; the 26 patients who did not have endoscopy had a median survival of 570 days ($p = 0.22$). Analysis of the duration of presenting symptoms revealed that patients with a long history (>6 months) survived longer than patients with a short history (<6 months) (Figure 2.3). This finding persisted even when an adjustment was made, using Cox's Regression model, for the prognostic factors described above ($p < 0.001$).

Pattern of Recurrence

Seventy six percent of all patients having a

Site of primary tumour	No. of patients	Percentages surviving at years				
		1	2	3	4	5
Cardia	38	47	27	23	13	13
Body	43	68	54	45	35	28
Antrum	42	70	53	31	31	27

In 5 patients the site of primary tumour was not known

Table 2.7 Survival by the site of the primary tumour in 123 patients undergoing curative resection for gastric cancer

Differentiation	No of patients	Percentage surviving at years					Median survival (days)	p value
		1	2	3	4	5		
Well	18	71	53	53	46	39	>1000)	NS
Moderate	42	68	56	41	36	31	875)	
Poor	68	59	41	28	20	15	544)	

Table 2.8 Survival by histology in 128 patients undergoing curative resection for gastric cancer

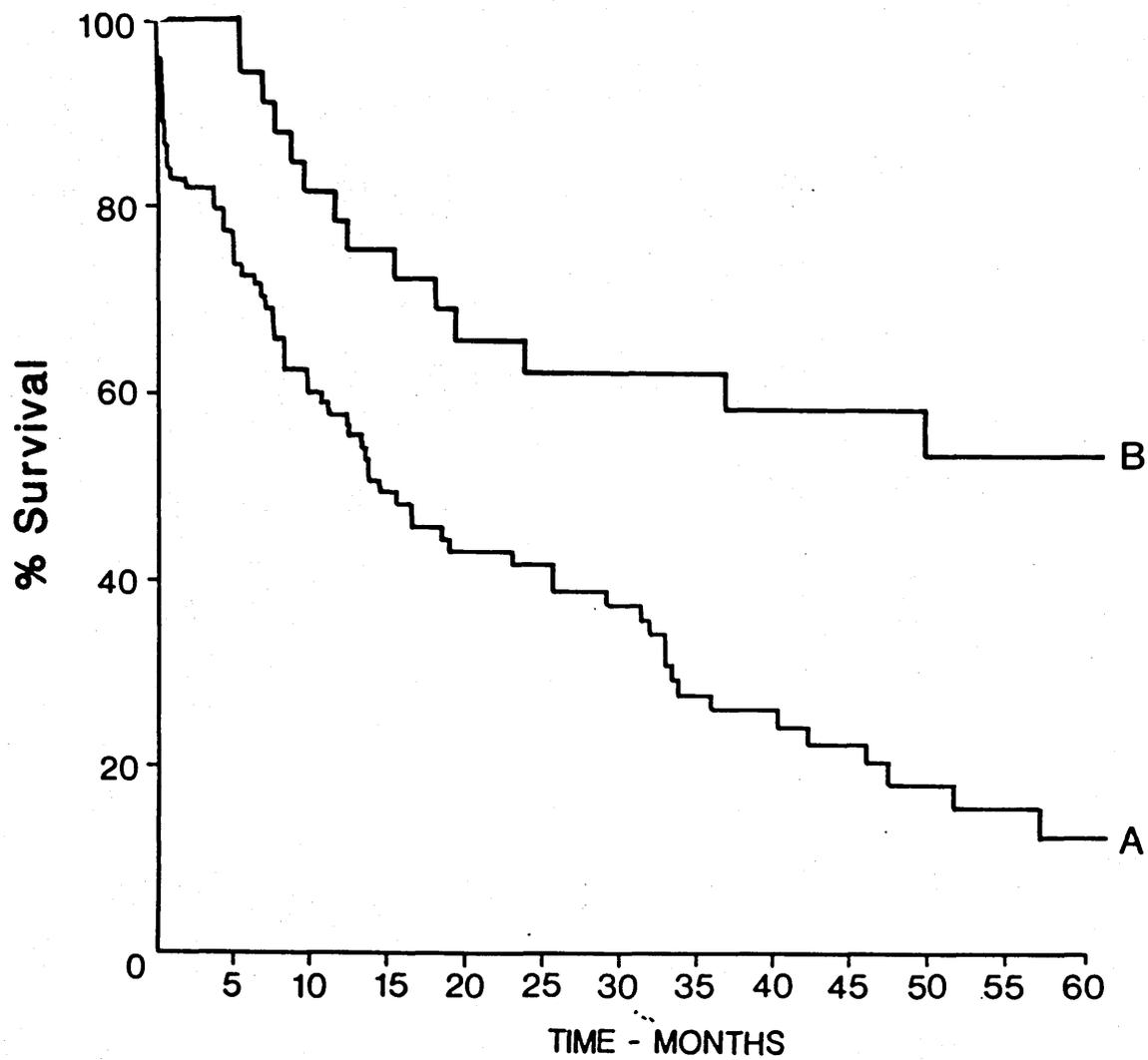


Figure 2.3 Survival by duration of symptoms in 121 patients having a curative resection for gastric cancer. A, <6 months (n=88); B, >6 months (n=33). p<0.001

curative resection are dead. The sites of recurrent disease are shown in Table 2.9. Of the patients with resection margin involvement, 53.2% developed local relapse alone or in combination with distant relapse.

Findings at resection		No of patients	Site of relapse (per cent)				
Serosa	Margins		None	Local	Distant	Both	Unknown
+ve	-ve	64	23.4	14.1	12.5	18.7	31.2
+ve	+ve	26	11.5	23.1	7.7	30.1	26.9

Patients with no serosal involvement have been excluded

Table 2.9 Pattern of recurrence after curative resection for gastric cancer

2.4 DISCUSSION

The survival of this unselected series of patients was related to the type of surgical procedure performed. All long term survivors were derived from the group of patients in whom there was complete clearance of macroscopic disease at the time of resection. However, although 39% of all patients had such a 'curative resection', only 11% achieved long term survival. Serosal involvement was the main determinant of survival. The group with serosal involvement had a 5 year survival rate of 13% compared to 47% for the group without. One explanation for the strong correlation between serosal involvement and survival, is that they have free cells in the peritoneal cavity, which is supported by the washings study (151). Indeed, I have also been able to demonstrate malignant cells in the peritoneum of a man with gastric cancer whose tumour had penetrated the serosa, but had not spread to lymph nodes (unpublished observations: St George's Hospital, London). Therefore it is possible that these patients have micrometastases at presentation.

Histological examination of the resected specimens showed that 24% of our curative

resection patients had tumour extending to the resection margin. The British Stomach Cancer Group reported a 22% incidence of resection margin involvement in their series of 390 patients (152), and Papachristou et al (153) found that following subtotal or total gastrectomy, 20% of their 350 patients had tumour involving the resection margin. This problem may be overcome if the resection margins are examined by frozen section at the time of operation (153). The prognosis for patients with resection margin involvement is known to be extremely poor (152), and none of our 31 patients in this category survived for more than 3 years. However, of our 31 patients, 84% also had serosal involvement which implies that their survival would have been limited, regardless of resection margin involvement. In Papachristou's study only 23% of the 73 patients with resection margin involvement developed anastomotic recurrence. The majority of patients died of metastatic disease before such recurrence became manifest (153). It is possible, therefore that resection margin involvement like serosal involvement, is a marker of aggressive metastatic stomach cancer. Lymph node involvement is also regarded as a major survival determinant in gastric cancer (154). Our experience suggests that

this is not the case and that spread of tumour to the lymph nodes only has prognostic significance in patients without serosal involvement.

In the curative resection group, patients with a long history of presenting symptoms had a better prognosis than those with a short history. This factor remained significant even when the data was subjected to multivariant analysis. This observation was first made in 1952 by Swynnerton and Truelove (73) and appears to apply to patients with early gastric cancer as well (88). It is possible that the subgroup of tumours associated with a long history behave less aggressively and are therefore more amenable to surgical intervention. A recent study from Japan (155) has shown that long term survivors after gastrectomy for gastric cancer have a high frequency of the HLA-DR4 antigen when compared to normal healthy controls and other gastric cancer patients. Thus it will be important to examine the HLA antigens of patients with a long duration of presenting symptoms.

Another factor described as having prognostic significance after surgery is the presence of human epidermal growth factor (EGF) in the resected tumour (68). Results from a group in Hiroshima suggest that patients with tumours

which possess EGF do considerably worse than those who do not. In this series of 158 patients with gastric cancer, EGF was detected using a combination of immunohistochemical and radio-immunoassay techniques in 40% of tumours. The survival of these patients was significantly shorter after surgery. Of interest, is that in 52 cases of early gastric cancer EGF could not be detected (68).

There is still debate about the best surgical approach to gastric cancer (156). In particular it remains controversial whether subtotal gastrectomy is as effective as total gastrectomy in affecting a cure (157,158). Pivotal to the argument is the consistently higher operative mortality of total gastrectomy (11.5-37.2%) compared to that of subtotal gastrectomy (3.2-23.7%) (157). Therefore although total gastrectomy should theoretically provide more longterm survivors, this advantage is offset by the greater number of early operative deaths, especially in older patients. Randomised studies are require to resolve this controversy but from our experience, length of history of presenting symptoms should be part of the stratification of such studies. It would also be advisable to incorporate an assessment of the cytology of the

peritoneal aspirates since this clearly has a major influence on survival (151).

The pattern of failure after resection for gastric cancer has been the subject of a number of reviews (77,158-160). In 2 post mortem studies loco-regional recurrence was found in 80% (158) and 96% (159) of patients following subtotal gastrectomy. However at post mortem almost 75-90% of patients also have distant metastases, these most commonly involving other abdominal organs (160). Our own data suggests that at least two thirds of patients have distant metastases at the time of relapse. Local recurrence alone was found in only 33% of patients but this is probably an overestimate, since it is likely that some of these patients have undetected metastatic disease.

As discussed in Chapter One, several classifications are used to describe the histology of gastric cancer (35,36,46). Most pathologists report at least the degree of differentiation of the tumour. Others divide gastric adenocarcinoma into the intestinal and diffuse types as described by Lauren (36), but as many as 30% of tumours may be unclassifiable or have a mixture of intestinal and diffuse forms (38). We were unable to undertake a retrospective review of the histology and classify it according to Lauren, therefore we

have reported histology simply in terms of the degree of differentiation. The degree of differentiation did not significantly affect the survival of patients having a curative resection, but there was a trend in favour of a better prognosis for patients with well differentiated tumours.

We have not described the incidence of early gastric cancer in our patients. By inference it is likely that the incidence was low because the overall prognosis of the surgically resected group was so poor. Houghton et al (80) found that early gastric cancer now comprises 12% of all gastric cancers and attributed this increased incidence to the more routine use of endoscopy. However, in our series, endoscopy as a means of diagnosis had no impact on survival. One possible explanation is that to obtain maximum benefit from endoscopy this procedure must be linked to a more comprehensive programme for early detection (79).

As a palliative procedure, resection of the gastric primary, even in the presence of solitary or multiple liver metastases, improves survival (161,162), and more importantly may overcome some of the distressing symptoms which can develop. In our experience, palliative resection was associated with a longer survival (6 months) when

compared to the survival of patients in the other advanced disease groups (1-4 months). However, patients who had a palliative bypass survived no longer than those who had a laparotomy alone or no surgical procedure. This of course does not necessarily mean that palliative bypass does not contribute to "quality of life" since measuring duration of survival ignores this factor, but it should mean that 'bypass' is not axiomatic for patients with irresectable tumour.

The prognosis for patients with gastric cancer remains very poor; the 5 year survival of the patients in this study was only 11%. Surgical intervention offers the only prospect of cure and is only likely to be successful in patients whose tumours have not penetrated the serosa. The best surgical approach is not established. Intra-abdominal micrometastatic disease is a major problem even in patients having an apparently 'curative' resection.

CHAPTER THREE

CHEMOTHERAPY IN GASTRIC

CANCER:

A CRITICAL APPRAISAL

3.1 TNO-6

3.1.1 Introduction

In pre-clinical studies TNO-6 was found to be less nephrotoxic in rodents than cisplatin but its activity against murine tumours was similar to that of cisplatin (163). A phase I study suggested that TNO-6 was less nephrotoxic in man and could be administered without hyperhydration (164). We thus initiated a study to evaluate the use of TNO-6 in patients with gastric adenocarcinoma. In addition because of the potential nephrotoxicity of this class of compounds, we decided to measure urinary N-acetyl- γ glucosaminidase (NAG). NAG is a lysosomal enzyme which is found in plasma and a number of tissues throughout the body, including the kidney. Elevation of urinary NAG has been shown to be a sensitive marker of cis-platin induced renal tubular injury (165). Therefore, estimation of urinary NAG was used in this study as a means of monitoring possible TNO-6 related renal tubular damage.

3.1.2 Patients and Methods

Patients

Sixteen patients (mean age 59 years, range 42-76) with histologically proven inoperable or recurrent gastric cancer entered this study. Thirteen were male and 3 female. Six had received previous chemotherapy (5-FU, adriamycin and mitomycin-C). Nine patients had liver metastases. Eligibility criteria were WBC $>4 \times 10^9/l$, platelet count $>100 \times 10^9/l$, normal serum creatinine, performance status Eastern Cooperative Oncology Group (E.C.O.G) grading < 3 and at least three weeks since completion of previous chemotherapy (6 weeks for mitomycin-C). Pretreatment evaluation included medical history, clinical examination, tumour measurement, serum urea and electrolytes, liver function tests, liver ultrasound, chest X-ray and full blood count.

Measurement of urinary NAG

Determination of NAG was based on the enzyme hydrolysis technique described by Lockwood and Bossmann (165). Using p-nitrophenyl-N-acetyl-B-D-glucosamide (Sigma Chemical Company Limited) as the substrate, NAG liberated p-nitrophenol which was measured spectrophotometrically. Samples of

urine collected 24 and 48 hours before and after TNO-6 were centrifuged, then dialysed to remove low molecular weight enzyme inhibitors (166) before enzyme estimation.

NAG estimation was performed on the final 6 patients entering the study. The urine collected for NAG measurement was also tested for protein, glucose and sediment.

Treatment protocol

TNO-6 $30\text{mg}/\text{m}^2$ was given by an intravenous infusion in 50ml of 5% dextrose every 4 weeks in the first 10 patients entering the study. The remainder received TNO-6 with hyperhydration consisting of 2 litres of dextrose given intravenously over the 2 hours before and after TNO-6 because the results of a phase I study which became available during the course of this investigation showed TNO-6 induced nephrotoxicity when given as a short intravenous infusion (167). If $\text{WBC} < 2.9 \times 10^9/\text{l}$ or $\text{platelets} < 75 \times 10^9/\text{l}$ treatment was delayed for 7 days or if $\text{WBC} < 1.0 \times 10^9/\text{l}$ or $\text{platelets} < 25 \times 10^9/\text{l}$ therapy was stopped. If creatinine clearance decreased by 20% from the pretreatment values, therapy was terminated. Patients were reviewed weekly for the first 4 weeks and every second week thereafter when

clinical examination was performed and urea and electrolytes, liver function tests, full blood count and platelets were estimated. All patients received a minimum of 2 cycles of therapy, unless disease progression was observed or disease toxicity occurred. Treatment was continued in responding patients. All patients received routine antiemetic therapy.

Assessment of response

The response to treatment was determined 4 weeks after chemotherapy. Death before this time was considered to reflect an inadequate trial of therapy. Duration of response and survival were taken from the initiation of chemotherapy. The definition of lesions used to assess response were as follows: measurable bidimensional - malignant disease measurable in 2 dimensions which included pulmonary, skin nodules and palpable primary tumours: measurable unidimensional - hepatomegaly. The liver was measured from below the costal margin in the midclavicular line and from the tip of the xiphoid process and the two values added together; evaluable, non-measurable - disease evident at endoscopy but not measurable.

Response criteria employed were those of the W.H.O. recommendations (168); complete response -

disappearance of all known disease for at least 4 weeks; partial response - greater than 50% decrease (for bidimensional lesions or those evaluable at endoscopy) or greater than 30% decrease (for unidimensional lesion) in total tumour size for at least 4 weeks without development of new lesions or progression of unmeasurable lesions; stable disease - less than 50% decrease or less than 25% increase in the size of measurable lesions or similar estimated changes in evaluable disease; progressive disease - 25% or more increase in the size of measurable lesions, estimated increase of 25% in evaluable disease or appearance of new lesions.

Assessment of toxicity

Toxicity of TNO-6 was assessed using W.H.O. recommendations (168).

3.1.3 Results

Response

The 16 patients with gastric adenocarcinoma were given 30 courses of chemotherapy. Five patients were not evaluable for response to TNO-6; 3 due to early death, 1 because radiotherapy was given to the gastric primary to control bleeding

and 1 patient in whom the only evidence of disease was an epigastric nodule which developed in the scar related to a previous gastric resection. The epigastric nodule was not biopsied and although it regressed on treatment with TNO-6 we do not think it should be regarded as an evaluable lesion since it may have been an inflammatory mass. Two of the early deaths occurred in patients who had received previous chemotherapy. Eleven patients were evaluated; 8 had progressive disease and 3 stable disease. The median survival for this group of 16 patients was 10.5 weeks with a range of 1 - 56 weeks.

Toxicity

Gastrointestinal toxicity data was available on all patients but information related to other toxicities was not available for the 3 patients who died early or the one patient given radiotherapy.

Gastrointestinal

Despite prophylactic antiemetic therapy, all but 3 patients experienced some degree of nausea and vomiting. This was considered severe (W.H.O. grade 3) in 11, moderate (W.H.O. grade 2) in 1 and mild (W.H.O. grade 1) in 1.

Renal

Renal function as determined by weekly serum creatinine and monthly creatinine clearance estimation, remained unchanged in 11 of 12 evaluable patients. In the remaining patient the creatinine clearance dropped from a pretreatment value of 113 ml/min to 38 ml/min 3 weeks after a single pulse of TNO-6. Proteinuria did not occur but during this time the patient developed obstructive jaundice due to tumour in the porta-hepatis and had an episode of septicaemia. He died from progressive gastric cancer 5 weeks after the course of TNO-6. Post mortem permission was refused.

Estimation of urinary NAG

The 6 patients studied for enzyme excretion received hyperhydration with TNO-6. All had elevated pretreatment urinary NAG activity (normal 0.43 ± 0.14 units/mmol creatinine) with jaundiced patients having the highest values. Twenty four hours after TNO-6 the NAG enzyme activity showed increases over pretreatment values of 1.6 to 7.4 fold (Figure 3.1). No significant changes with respect to serum creatinine and urine protein were observed in these patients following TNO-6.

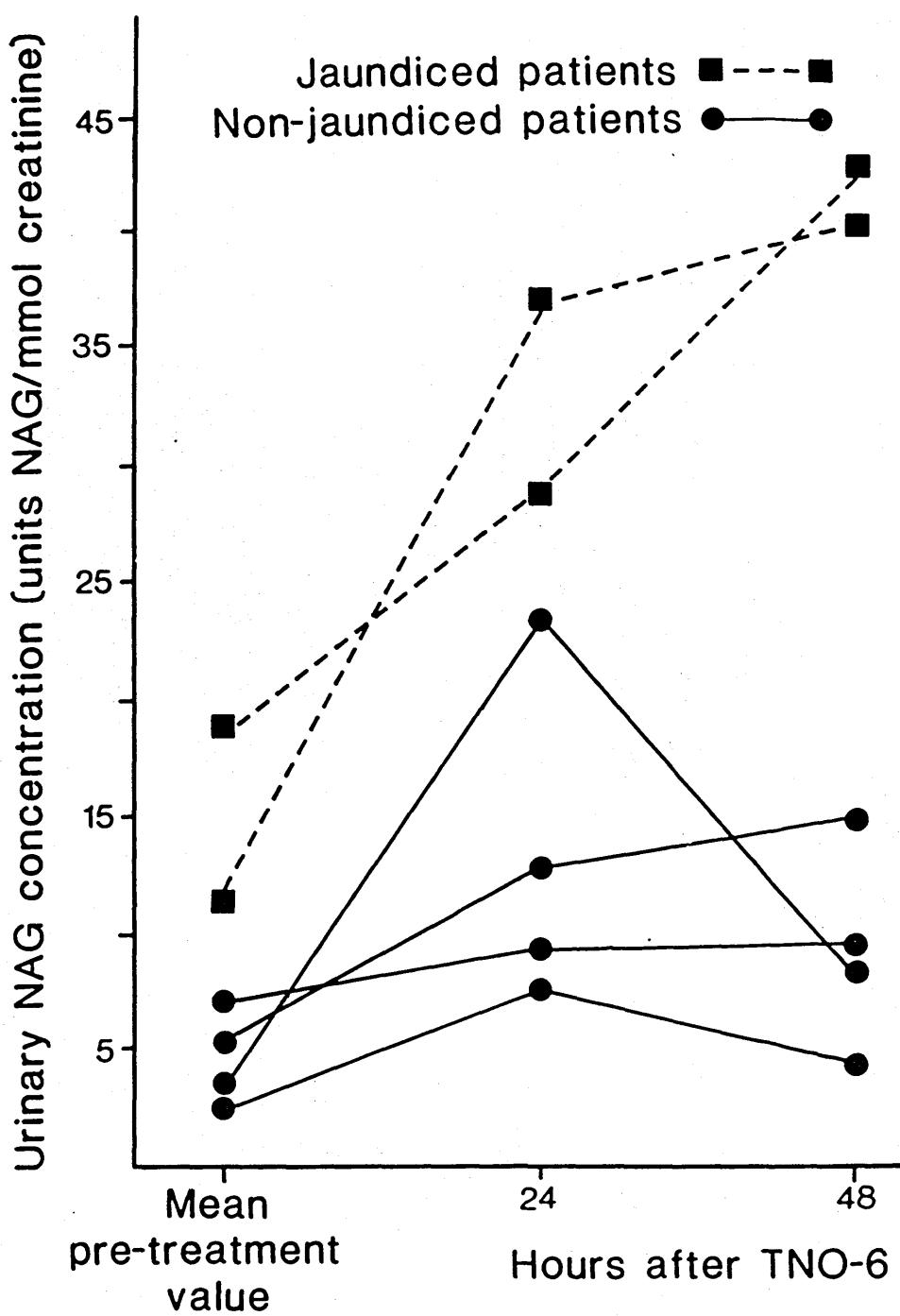


Figure 3.1 The effect of TNO-6 on urinary NAG in 6 patients

	White cell count	Platelet count
	$\times 10^9/l$	$\times 10^9/l$
Grade 0	2	8
Grade 1	5	1
Grade 2	3	0
Grade 3	2	2
Grade 4	0	1

Table 3.1 Haematological toxicity following TNO-6
as assessed by WHO grading (22).
(Nadir counts on 12 patients)

Other manifestations of toxicity

The majority of patients developed evidence of myelosuppression though it tended to be mild (Table 3.1). Only 1 patient, who had previous chemotherapy with F.A.M., required platelet transfusion. No cases of fatal infection were observed. No changes were observed in serial transaminases, bilirubin and alkaline phosphatase other than in those patients with hepatic metastases who developed progressive disease. Alopecia was not observed in any of our patients.

3.1.4 Discussion

This study was terminated prematurely because TNO-6 induced nephrotoxicity was reported from other centres conducting concurrent Phase II studies in patients with other neoplasms (128). However TNO-6 had no apparent activity in gastric adenocarcinoma, which was particularly disappointing, considering the activity of cis-platin in this tumour type. Four patients with gastric cancer were non evaluable, 3 due to early death, 2 of whom had prior chemotherapy. The higher incidence of early death in the pretreated group reflects the poor prognosis of these patients and highlights the difficulties of attempting to

identify new agents in gastric cancer, where currently most patients receive combination chemotherapy as part of their initial management.

Estimation of urinary N-acetyl-B-glucosaminidase is a sensitive method of detecting renal tubular damage (166). All patients, in whom NAG was measured, showed an increase following TNO-6, though none demonstrated concurrent alteration in serum creatinine or creatinine clearance, which reveals the insensitivity of these parameters in the early detection of the nephrotoxicity of cytotoxic agents. Although we found no definite TNO-6 induced alteration in renal function amongst our patients, other workers found significant elevation of serum creatinine in 13% of 61 patients receiving TNO-6 for a variety of neoplasms, even when hyperhydration was routinely employed (169). In addition, when TNO-6 is administered without hyperhydration it can produce renal failure and proteinuria (169). Consequently TNO-6 appears to offer no advantage over cisplatin in terms of nephrotoxicity and unlike cisplatin also produces glomerular damage. Emesis remained a difficult problem with TNO-6. It was similar to that encountered with cisplatin. Marrow suppression of a significant degree was recorded in 25% of patients, and in the absence of

nephrotoxicity was the dose limiting factor.

In conclusion, the cisplatin analogue TNO-6 produces nephrotoxicity that cannot be prevented by hyperhydration and has no significant activity in gastric cancer.

3.2 5-FU, ADRIAMYCIN AND MITOMYCIN-C (FAM)

3.2.1. Introduction

In 1979 MacDonald published the first report on the use of the combination, 5-FU, adriamycin and mitomycin (FAM) in advanced gastric cancer (124). Eighteen (50%) of 36 patients had a partial response to this chemotherapy, and the treatment was considered relatively non-toxic. The update on this report confirmed the activity of FAM in 42% of 62 patients (170). The duration of response was also encouraging (median 9 months) and responding patients survived longer. Therefore in June 1980 we initiated a phase II study of FAM in gastric cancer (171). The aim of the study was to assess the effectiveness of this regimen and to identify factors which might predict the response to therapy.

3.2.2 Patients and Methods

Eighty four patients with histologically proven advanced adenocarcinoma of stomach entered the study. None of the patients had received previous chemotherapy or radiotherapy. Three patients died from disease progression within 2

weeks of commencing treatment and are not considered further. The characteristics of the 81 evaluable patients are shown in Table 3.2 with performance status grading according to ECOG grading criteria. All patients in this study had disease which could be evaluated by one or more of the following means; clinical examination, radiology, hepatic ultrasonography, hepatic isotope scanning, abdominal computerised axial tomography or endoscopy. Formal evaluation of the response to therapy was made at the end of 2 complete cycles (16 weeks) though the patients were also continually assessed at regular outpatient visits. WHO criteria of objective response to therapy were used (168) as outlined in Section 3.1.2.

The treatment schedule which was identical to that used by MacDonald et al (170) consisted of 5-FU $600\text{mg}/\text{m}^2$ (days 1,8,29 and 36); adriamycin $30\text{mg}/\text{m}^2$ (days 1 and 29); mitomycin-C $10\text{mg}/\text{m}^2$ (day 1). The cycle was repeated every 8 weeks. All drugs were administered by bolus intravenous injection into a peripheral vein using standard precautionary techniques. Treatment was normally given on an outpatient basis if the patient was fully ambulant. Before each pulse of therapy the peripheral blood was monitored and if the white

Performance status	n
0	5
1	27
2	33
3	14
4	2

Histology

Poorly differentiated adenocarcinoma	54
Moderately differentiated adenocarcinoma	14
Well differentiated adenocarcinoma	10
Not available for review	3

Age: mean 54 years; range 33-77 years

Table 3.2 Characteristics of 81 evaluable patients
with advanced gastric cancer

blood count was less than $3.5 \times 10^9/l$ or platelet count less than $100 \times 10^9/l$ treatment was delayed for 7 days and thereafter modified as outlined in Table 3.3. Response to treatment was assessed after 2 cycles of chemotherapy. Patients demonstrating a complete response, partial response or stable disease received a further 2 cycles of chemotherapy. Treatment was then stopped but was re-instituted in those deemed to have a complete response if they subsequently relapsed. Patients considered to have progressive disease at the end of 2 cycles were not given further chemotherapy. In addition, the development of progressive disease in patients receiving their third or fourth cycle of chemotherapy led to cessation of chemotherapy. Duration of response and survival were measured from the start of treatment.

Statistical analysis

Differences in performance status and age between the groups showing complete response, partial response, stable disease and progressive disease were examined using the (non-parametric) Kruskal-Wallis analysis of variance. The significance of differences between tumour histology, primary tumour resection and presence

Drug dose	WBC x 10 ⁹ /l	Platelets x 10 ⁹ /l
100%	> 3.5	> 100
50%	2.5 - 3.5	75 - 100
0	< 2.5	< 75

Table 3.3 FAM treatment modification following marrow suppression in patients receiving chemotherapy for advanced gastric cancer

of hepatic metastases was calculated using Cox's empirical logistic transform (172) after correcting for multiple comparisons. Survival was analysed as outlined by Peto et al (173,174) employing the Logrank test to calculate the significance of differences between survival curves. The effects of independent variables such as response to chemotherapy, sex, age, performance status and presence of hepatic metastases on survival were examined using Cox's Regression Model (150)

3.2.3 Results

Response to treatment

(Table 3.4)

A complete response was obtained in 4 patients (5%), 1 of whom remains alive and disease free 5 years after commencing therapy (Patients' details see Table 3.5). In 24 patients (30%) a partial response was noted. Four patients with a partial response had successful resection of their primary tumour following chemotherapy. In 2 of these patients the tumour was originally deemed inoperable at laparotomy where a bypass procedure alone was performed. The other 2 patients did not have laparotomy prior to chemotherapy but were

	Complete response	Partial response	Stable disease	Progressive disease	p value
Patients n	4	24	8	45	
%	5	30	10	55	
Mean performance status	1.5	1.6	1.5	1.9	NS
Mean age	56	57	56	61	NS
Median duration of response (months)	10.5	-	-	-	
Median survival (months)	17.1		11	4.5	<0.001
Number with hepatic metastases	3		5	24	<0.001
Histology (3 patients not reviewed)					
Well differentiated	3		-	7	NS
Moderately differentiated	2		3	9	NS
Poorly differentiated	23		4	27	NS
Surgery					
None	8		2	14	NS
Laparotomy	6		2	13	NS
Laparotomy + bypass procedure	2		4	4	NS
Resection	12		-	14	NS

Table 3.4 Details of 81 patients with advanced gastric cancer treated with FAM

Site of disease in patients 1-4	Assessment of response	Duration of response (months)	survival (months)	Response to FAM on relapse
1 Anastamosis of Polya gastre- ctomy performed yrs earlier for gastric cancer	endoscopy CT scan of abdomen	58 and continuing	60	-
2 Resection margin after partial gastrectomy	endoscopy CT scan of abdomen	9	10 dead	none
3 L.supraclavicular adenopathy. R.pleural effusion after partial gast- rectomy for gastric cancer.No intra- abdominal recurrence	clinical chest x-ray	11	12 dead	none
4 Cervical/axillary lymphadenopathy with primary gastric cancer	clinical endoscopy barium meal	29	30	-

Table 3.5 Details of 4 patients showing complete response to FAM.

considered unsuitable for surgery as one had a massive primary tumour and the other had distant spread of disease in the form of bulky cervical lymphadenopathy. The overall duration of response was 10.5 months and the median survival of responders was 17.1 months. Response to chemotherapy usually occurred within the first cycle of therapy and was associated with stabilisation or improvement in performance status (PS) (mean PS before therapy 1.5; mean PS after therapy 1.3). Eight patients (10%) had stable disease with a median survival of 11 months. Forty-five patients (55%) had disease progression on therapy and all of these patients are now dead. The median survival for this group was 4.5 months. Mean age, performance status, tumour histology and incidence of primary tumour resection were similar in all groups. However, there was a highly significant difference ($p < 0.001$) between the presence of hepatic metastases in the non-responders and responders.

Survival of responders and non-responders are shown in the life table analysis graph (Figure 3.2). Response to chemotherapy was associated with a significant improvement in survival (Logrank test $\chi^2 = 35.8$ (1 d.f) $p < 0.001$). When the covariates sex, age, performance status and presence of

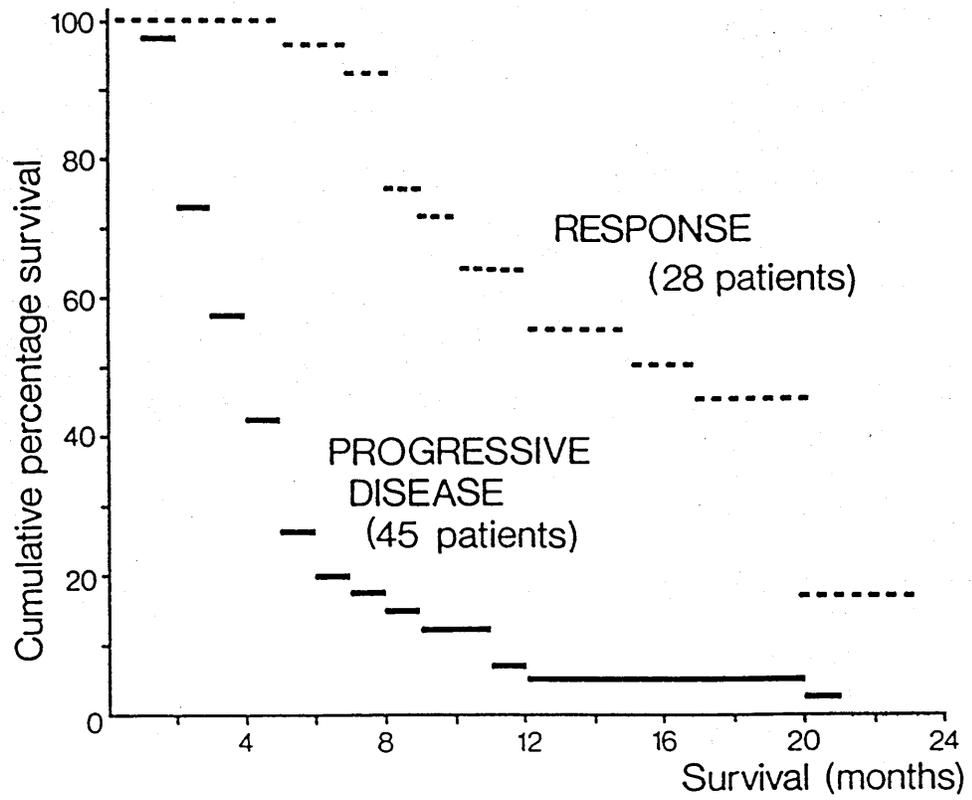


Figure 3.2 Survival of 81 patients with advanced gastric cancer treated with FAM

WBC nadir x 10 ⁹ /l			n	%
Grade*	0	4.0	25	42
Grade	1	3.0 - 3.4	14	23
Grade	2	2.0 - 2.9	10	17
Grade	3	1.0 - 1.9	8	13
Grade	4	1.0	3	5

Platelet nadir x 10⁹/l

Grade*	0	100	34	56
Grade	1	75 - 99	8	13
Grade	2	50 - 74	7	12
Grade	3	25 - 49	7	12
Grade	4	25	4	7

WHO grading (22)

Table 3.6 Myelosuppression in patients with advanced gastric cancer treated with FAM (60 patients with complete data available for analysis)

hepatic metastases were entered into Cox's Regression Model none reached the accepted level of statistical significance in explaining survival whereas response to therapy did ($p < 0.001$).

Toxicity

Full data on the degree of myelosuppression were available in 60 patients. Approximately 50% of those developed some degree of myelosuppression though this was only considered significant in 20% (WHO grade 3 and 4) (see Table 3.6). Nausea and vomiting was encountered in 60% of patients and 10% had prolonged gastro-intestinal side effects. One patient developed treatment related oral mucositis. Alopecia occurred in all patients completing two cycles of therapy. Scalp cooling was not used in any of these patients though they were all provided with a fashion wig, the colour and texture of which was matched to the patients own hair prior to commencing chemotherapy, in order to assure the best possible cosmetic result.

3.2.4 Discussion

This study is the largest single series of patients with advanced gastric cancer treated with FAM. The results confirm its activity in

adenocarcinoma of the stomach and relative lack of myelotoxicity, though gastrointestinal side effects were more frequently observed than in MacDonald's original (124) and subsequent reports (170). Alopecia developed in all patients completing 2 cycles of therapy. Scalp cooling which may prevent the development of alopecia associated with adriamycin containing regimens, was not routinely available for our patients at the time of this study.

Though the overall response rate in our study of 35% was slightly less than the 42 to 44 % reported by other workers, the duration of response and survival of the responding group were similar (124,175,176). With the difficulties of accurately assessing response rate in gastric cancer the survival figures are a more accurate reflection of benefit from therapy. This is highlighted by the single agent therapy data, where response rates of between 15 to 25% rarely produced a significant improvement in survival (96).

However, as less than 50% of patients will respond and all are subject to some drug induced toxicity, attempts have been made to improve the selection of patients most likely to benefit from treatment. Analysis by Frascini (176) and

MacDonald (170) of their 47 and 62 patients respectively, treated with FAM, failed to show any statistical correlation between performance status, tumour histology, site of metastases, or resectability of the primary tumour on response to treatment. In a recent review of 137 patients the Gastrointestinal Tumour Study Group found performance status ($p < 0.01$), presence of extra abdominal lymphadenopathy ($p < 0.05$) and pulmonary metastases ($p < 0.05$) to influence the response to treatment, though the fact that patients were treated with a variety of chemotherapeutic regimes makes overall interpretation more difficult (177). In our experience the presence of hepatic metastases was associated with a highly significant ($p < 0.001$) negative effect on response to treatment. Indeed, the response rate in patients without hepatic metastases was 51%.

We recognise that patients showing a partial or complete response to chemotherapy may represent a pre-selected group in that their incidence of hepatic metastases was low. It is arguable that they constitute a group with a relatively favourable natural history regardless of any effect of chemotherapy. On the other hand, further analysis of our patients with progressive disease reveals that median survival did not differ

significantly when patients without hepatic metastases (3 months) are compared to those with such metastases (5 months).

The application of this regime in clinical practice may seem limited in that more than half of the patients failed to show an objective response. This criticism should not detract from the frequent, and sometimes dramatic reduction in tumour mass that can accompany therapy with FAM, usually appearing during the first cycle of therapy and often associated with an improvement in performance status. Indeed, in one of our patients with tumour at the oesophagogastric junction, insertion of a Celestin tube was avoided following complete disappearance of dysphagia within 48 hours of administration of FAM.

Resection of gastric carcinoma following chemotherapy deserves comment. Although this applied to only 4 patients in the present study, they represent 25% of the group of 16 patients with unresected tumours who demonstrated a response to treatment. These 4 patients have survived 10, 20, 20 and 32 months respectively and 2 are still alive. Since patients undergoing primary resection have a longer survival than those having bypass procedures (161), secondary resection may also contribute to an improvement in

survival.

Of interest was a clinical curio which we have previously reported (178). One of the patients treated with FAM reported a dramatic improvement in his chronic obstructive airways disease following chemotherapy. This subjective improvement was confirmed by serial pulmonary function testing. It was mediated through an increase in FEV_1 and vital capacity indicating a reduction in airway obstruction. The mechanism of action of chemotherapy in this particular clinical situation is not clear. One possible explanation is that the chemotherapy sensitised the bronchi to the oral salbutamol which was continued throughout his chemotherapy. This has been proposed as one of the modes of action of steroids in asthma (179,180). Chronic obstructive airways disease need not be a contraindication to chemotherapy.

Chemotherapy with FAM offers, acceptable outpatient palliative therapy in 35% of all patients with advanced gastric cancer; the response rate was greater in those without hepatic metastases. In a small proportion of patients with inoperable tumours treatment with FAM rendered these tumours operable and these patients seemed to fare particularly well after chemotherapy. Nausea and vomiting remained a

significant problem and our efforts to deal with this are outlined in section 3.5.

3.2.5 My Involvement in the FAM Study

Although this data was generated from 4 centres in Scotland the majority of patients (n=53) were from Glasgow Royal Infirmary and I was personally involved in the management (including assessment of response and toxicity) of all of these patients.

3.3 5-FU, METHOTREXATE AND ADRIAMYCIN

3.3.1 Introduction

Bertino and his colleagues were the first to demonstrate a synergism between 5-FU and methotrexate (181). They showed enhanced cytotoxic activity in a variety of tumour models when methotrexate was given sequentially to 5-FU. Klein utilised this effect in the treatment of gastric cancer and reported a very high response (60%) in 30 patients treated with the combination of 5-FU, methotrexate and adriamycin (FAMTX) and emphasised the lack of toxicity (125). Other data from a small pilot study of adriamycin and methotrexate partly corroborated Klein's results by showing an objective response in 10 of 14 evaluable patients treated with this 2 drug combination (182).

Therefore we considered it appropriate to test FAMTX in our centre (183) given the apparent efficacy and low toxicity of this regimen.

3.3.2 Patients and Methods

Twelve patients comprising 7 males and 5 females with a mean age 61 years (range 41 - 78)

entered the study. One patient had received previous chemotherapy. All had measurable disease and only 1 patient had overt hepatic metastases. Performance status (ECOG grading) was I in 11 patients and II in 1. All patients had normal haematological indices prior to chemotherapy, a normal serum creatinine and a creatinine clearance >50 ml/min. Two patients had elevation of the serum bilirubin at 175 $\mu\text{mol/l}$ and 63 $\mu\text{mol/l}$ respectively, but the remaining patients had normal liver function. The regimen consisted of: Day 1 methotrexate 1.5 g/m^2 IV bolus followed 1 hour later by 5-FU 1.5 g/m^2 IV bolus and 24 hours later by folinic acid 50 mg/m^2 IV six hourly, combined with forced alkaline diuresis and continued until serum methotrexate levels were outwith the toxic range; Day 14 adriamycin 30 mg/m^2 IV, cycled every 28 days. Response to treatment and toxicity were assessed according to WHO criteria (168).

3.3.3 Results

The 12 patients received 35 courses of treatment. Following chemotherapy 2 patients had a partial response, 2 patients had stable disease, 7 patients had disease progression and 1 patient was not evaluable due to death from septicaemia. The

major toxicity was myelosuppression with a white cell count nadir $<1 \times 10^9/l$ in 4 patients and $1-2 \times 10^9/l$ in 4 patients. The 2 patients with abnormal liver function had nadir white cell counts of $>3 \times 10^9/l$. The median time of the white cell count nadir was day 13. Three patients developed life threatening septicaemia during the neutropenic phase and 1 further patient died of gram-negative septicaemia. Thrombocytopenia was observed in only 1 patient, with a nadir count of $54 \times 10^9/l$. Mucositis was recorded in 3 patients and an erythematous skin rash in 2. Although all patients were given routine antiemetic treatment, 4 still experienced nausea and vomiting. There was no nephrotoxicity.

3.3.4 Discussion

Clearly it was premature to report the activity of this regimen in gastric cancer since the confidence intervals on the response rate (16%) in such a small patient sample are wide (0 - 38%). However, the toxicity of the regimen deserves comment. In particular, we were alarmed by the high incidence of life threatening infection, occurring in the context of neutropenia, which had not been anticipated on the

basis of Klein's original report (125). The agents mainly responsible for the myelosuppression appear to have been methotrexate and 5-FU, which was reflected in the occurrence of the white cell count nadir on day 13 of the treatment cycle. In terms of myelosuppression adriamycin was less important, and it was of interest that the patients with abnormal liver function did not develop significant neutropenia. The reason for the more severe myelosuppression in our study when compared to Klein's experience (125) is not clear, but it certainly cannot be attributed to the use of inadequate 'folinic acid rescue'. Subsequent to our study, the EORTC also reported (184) that they were unable to reproduce the high response rate described by Klein. They treated 71 patients in a multicentre trial with only 22 (33%) responding patients and 4 toxic deaths (184). Therefore the toxicity and as yet unconfirmed efficacy indicates that this regimen should not be routinely used and that its inclusion in adjuvant protocols is potentially hazardous.

3.4 THE IMPACT OF CHEMOTHERAPY ON THE OVERALL SURVIVAL OF PATIENTS WITH ADVANCED DISEASE

3.4.1 Introduction

Although it is clear that patients with gastric cancer who respond to chemotherapy live longer (170,171), there is little evidence that chemotherapy improves overall survival rates. Indeed, in a recent randomised study from the North Central Cancer Treatment Group (185) in which 151 patients were allocated to 1 of 3 treatment regimens; single agent 5-FU, 5-FU and adriamycin, or FAM there was no survival advantage for any treatment arm. These findings are disconcerting and indicate quite clearly that there is a need for further randomised trials of combination chemotherapy versus no treatment in gastric cancer. At the present time there is only one small study in the literature which randomised 37 patients to either no treatment or a 5 drug combination of vincristine, methotrexate, cyclophosphamide, 5-FU and mitomycin-C. It showed almost identical median survival times for treated and control groups of 9.5 and 9 weeks respectively (186).

The retrospective review outlined in Chapter

Two allowed investigation of the contribution of chemotherapy to overall survival. During the last 5 years of the study a medical oncology unit was established in the centre and a larger proportion of patients with advanced disease received chemotherapy. This permitted comparison of the survival of patients with advanced disease during 2 periods, one when chemotherapy was rarely given and the other when chemotherapy was frequently given.

3.4.2 Patients and Methods

Two hundred and two patients in the retrospective review had advanced disease at presentation (this includes 2 patients who had curative resections subsequent to chemotherapy). Patients with advanced disease were defined as those in whom it was not possible to effect a surgical cure. Of these 202 patients, 50 were given chemotherapy. Forty five received FAM, 3 received TNO-6 and 2 received 5-FU alone. Using Cox's regression model (150) the effect of chemotherapy on survival was analysed taking into account the independent variables of type of surgery performed on each patient, tumour histology and presence of liver metastases.

	All patients		Excluding deaths within 14 days	
	median survival (days)	% surviving 1 year	median survival (days)	% surviving 1 year
No chemotherapy (n=152)	71	9	108	12
Chemotherapy (n=50)	160	24	155	23

All patients chemotherapy versus no chemotherapy $p < 0.001$)
 Exclude early deaths chemotherapy versus no chemotherapy $p = 0.02$

Table 3.7 The effect of chemotherapy on the survival of patients with advanced disease.

3.4.3 Results

The 50 patients who received chemotherapy survived significantly longer ($p < 0.001$) than those who did not (Table 3.7). To correct for patient selection for chemotherapy all deaths within 14 days of diagnosis were excluded. The significance value changed to $p = 0.02$ once these deaths had been excluded (Table 3.7). In a further effort to elucidate the effect of chemotherapy on survival we compared the survival of patients with advanced disease from the period 1974 - 1979 (when 8% of patients received chemotherapy) with the survival of patients from the period 1980 - 1984 (when 45% of patients received chemotherapy). We found no significant difference ($p = 0.4$) in survival rates, suggesting that chemotherapy did not influence prognosis.

3.4.4 Discussion

Treatment of gastric cancer with single agent chemotherapy results in tumour regression in approximately 10-20% of patients. This regression is normally short lived and of little biological significance to the patient (96). With the introduction of combination chemotherapy regimens

such as FAM, response rates in the region of 35-40% were reported (170,171) and it was observed that these responses could persist for longer (10 months) and that responding patients survived for a median of 17 months. Untreated, advanced gastric cancer has a very poor prognosis. A median survival of 4 months was reported in one North American series (187). Therefore it was considered possible that combination chemotherapy was prolonging overall survival. As a result, there have been few randomised trials comparing the survival of patients treated with combination chemotherapy, against a non-treatment control group.

In the absence of all but one controlled trial (186) this retrospective analysis provided valuable information on the effect of chemotherapy on overall survival. In the second period of the study, 45% of patients received chemotherapy compared to 8% of patients who received chemotherapy during the first period. In other respects the two patient populations were very similar. In particular, there was no major change in the type or proportions of surgical procedure performed, both groups were matched for the presence of liver metastases, the patients were drawn from a consecutive series, thus eliminating

the bias normally created by patient selection for a retrospective study, and information was available on virtually all patients for follow up. Using multivariate analysis, patients with advanced disease who received chemotherapy survived significantly longer than those who did not. However, when all deaths within 14 days of diagnosis were excluded from analysis, the difference was less marked, although still significant ($p=0.02$). The rationale for excluding early deaths was based on the assumption that patients selected for chemotherapy are usually more fit than those who are not, and that such patients are normally anticipated to live more than 14 days. The next step in the analysis was to compare the survival of patients with advanced disease during the 2 periods, one when chemotherapy was rarely given and the other when almost half of the patients were given chemotherapy. The survival of these 2 groups of patients was identical.

Why should a treatment which can produce tumour regression in 35-40% of patients fail to make an impact on overall survival? Chemotherapy is not devoid of side effects and may well contribute to an increased morbidity and mortality in patients who do not respond. Certainly this is

true of more toxic regimens (183,188), and this effect may outweigh the beneficial effects of chemotherapy in some patients. Thus, in order to optimise the use of chemotherapy, the administration of chemotherapy must be closely monitored so that toxicity is minimised. Side effects related to the gastrointestinal tract can be controlled by the appropriate antiemetic (section 3.5) and the timely use of antibiotics may prevent septicaemic episodes.

3.5 THE CONTROL OF NAUSEA AND VOMITING INDUCED BY CYTOTOXIC DRUGS

3.5.1 Introduction

Chemotherapy used in the treatment of gastric cancer is palliative. Therefore, optimisation of the use of chemotherapy depends on the selection of patients most likely to respond (such as those without liver metastases) and the minimisation of side effects. The most important side effects from the patients' point of view are nausea and vomiting, followed by alopecia (189). Alopecia may be prevented in some cases by the judicious use of scalp cooling (190). However, for patients with upper gastrointestinal malignancy nausea and vomiting is a particularly distressing symptom because it is often part of the symptom complex related to the tumour. Furthermore, these symptoms may exacerbate the anorexia commonly associated with gastric cancer and lead to more weight loss and general malaise. In view of this, as part of the strategy for the treatment of gastric cancer the development of effective antiemetic regimens was considered a priority. It was clear that if good control of emesis could be achieved, for any given patient, the balance would

shift in favour of active treatment.

There is a large number of antiemetics available but none is completely effective or devoid of side effects (191). We have therefore performed a number randomised, comparative trials in an effort to assemble the best antiemetic "package". Cisplatin is the most emetogenic of the cytotoxic drugs used to treat cancer and is by convention considered separate to other cytotoxic drugs in antiemetic trials. The nausea and vomiting associated with the administration of cisplatin has understandably deterred many clinicians from using this drug to treat gastric cancer. This therapeutic drawback is underlined by the data from Chapter Two which indicated that cisplatin is one of the most useful drugs in the treatment of gastric cancer.

The following data will relate to 4 randomised trials (2 of which included chemotherapy regimens containing cisplatin) and an open trial where the potential of a new antiemetic compound, a 5-hydroxytryptamine receptor (5-HT₃) antagonist, was tested (192-196).

3.5.2 Patients and Methods

Randomised trials

Patients with a variety of malignancies, receiving chemotherapy for the first time entered these trials.

Trial 1

Patients were randomly assigned by a double blind, double dummy crossover technique to one of the following antiemetics: metoclopramide given as a 50mg loading dose intravenously 30 minutes before chemotherapy, followed by a 12 hour intravenous infusion of 5mg/kg in 500ml physiological saline or chlorpromazine 25mg (1ml) given intramuscularly every 4 hours for 12 hours, beginning with the chemotherapy.

Trial 2

Patients were randomly assigned using a double blind crossover method to nabilone 2x1mg capsules combined with prochlorperazine (5mg) or nabilone 2x1mg and placebo. The first dose was given at 10pm the night before chemotherapy, and 3 further doses were given at intervals of 12 hours, although if vomiting did not occur the final dose was omitted.

Trial 3

This was a randomised open crossover trial where patients were assigned to nabilone and prochlorperazine or metoclopramide and dexamethasone and crossed on the second course. Nabilone was given as in trial 2. Metoclopramide was given as a 2mg/kg loading dose in 250ml of physiological saline administered as an IV infusion over 15 minutes before chemotherapy followed by an IV infusion of 3mg/kg in 500ml physiological saline over 8 hours. Just prior to chemotherapy dexamethasone 20mg made up to a 10ml volume with physiological saline was administered IV over 3 to 5 minutes. (More rapid injection can lead to an unpleasant paraesthesia).

Trial 4

Patients were randomly assigned using a single blind technique to one of 3 antiemetics. Domperidone 20mg tds or metoclopramide 20mg tds given orally on the day before chemotherapy and continued for 3 days with 20mg bd on day 4 and 20mg on day 5 after chemotherapy or dexamethasone given 4 mg tds on the day before chemotherapy and continued for 3 days, with 4mg bd on day 4 and 4mg od on day 5. On the day of chemotherapy the second dose of metoclopramide or dexamethasone was given

as an IV bolus (domperidone was not available for IV administration). If the first antiemetic regimen failed to control nausea and vomiting patients were randomly allocated to 1 of the remaining regimens for the next course of chemotherapy, and if that failed they were allocated to the third regimen. The chemotherapy was stratified into regimens of high (those containing intravenous doxorubicin, epirubicin or cyclophosphamide; there were 14 patients in each antiemetic group) and low emetogenic potential

Assessment of antiemetic efficacy and side effects

For trials 1-3 the number of episodes of emesis, duration of vomiting, severity and duration of nausea and any change in appetite were recorded for 24 hours following administration of chemotherapy, on an in-patient basis. Side effects of treatment including sedation, dizziness, dryness of mouth, dysphoria and extrapyramidal reactions were recorded, and graded none, mild, moderate or severe. All recordings were made by senior nursing staff or medical staff. After each antiemetic course the patient indicated the extent of emesis control by marking a cross on a 10cm linear analogue scale ranging from 'not at all sick' to 'very sick'. After the second antiemetic

course the patient completed a questionnaire to determine preference (if any) for either antiemetic regimen and outline reasons for this preference. For trial 4 nausea, vomiting and side effects were assessed on an in-patient basis for 24 hours after chemotherapy and the patients completed a daily diary of these symptoms for at least the following 5 days, or until the symptoms resolved.

Non-randomised trial

Patients with nausea and vomiting refractory to standard antiemetic therapy were selected for treatment with the 5-HT₃ antagonist GR38032F. Immediately before chemotherapy a dose of 4mg (base equivalent) of GR38032F (Glaxo group Research) was given by intravenous injection over 5 minutes and a further 4 mg orally was given at the same time. This oral dose was repeated 5 and 10 hours after chemotherapy. Nausea, vomiting and side effects were recorded on an in-patient basis for the 24 hours after chemotherapy.

3.5.3 Results

Randomised trials

Trial 1

Ninety five patients (52 women, 43 men; mean age 49) received chemotherapy regimens consisting of cisplatin or cisplatin analogues in 43 and regimens without cisplatin in 52. Ten patients given cisplatin and 11 of the others did not complete 2 courses because of death (10), violation of the protocol (7), stopping chemotherapy (2), refusal of intramuscular injections (1) and a dystonic reaction (1). For patients receiving cisplatin regimens there was complete control of nausea and vomiting in 13% when treated with chlorpromazine and 19% when treated with metoclopramide. For the regimens without cisplatin there was complete control of nausea and vomiting in 42% of patients treated with chlorpromazine and 46% with metoclopramide. Among patients who completed the crossover there were no significant differences in vomiting, nausea or appetite, except that among patients given regimens containing cisplatin the severity of nausea was significantly less in those who received high dose metoclopramide ($p < 0.01$) and a significant number of these patients preferred

high dose metoclopramide (22 v 5, $p < 0.01$); 6 had no preference. The assessment on the linear analogue scale in this chemotherapy group also favoured high dose metoclopramide ($p < 0.01$). No preference was shown in the group given regimens not containing cisplatin. In both chemotherapy groups there was no significant relation between the order of administration of active drug and drug preference (Fisher's exact test). Adverse effects except extrapyramidal reactions were similar for both antiemetics.

Trial 2

Thirty four patients (20 women, 14 men; mean age 55 (range 39-76)) entered the study. All were receiving chemotherapy regimens without cisplatin. Four patients did not complete 2 courses: 3 died and chemotherapy was stopped in 1. There was complete control of nausea and vomiting in 85% of patients given nabilone and prochlorperazine and 74% of patients given nabilone. Among the patients who completed the crossover there were no significant differences in nausea, vomiting, appetite, assessment on the linear analogue scale or adverse effects other than those on the central nervous system (euphoria, dysphoria or disorientation) which were significantly less

common with the combination of nabilone and prochlorperazine ($p < 0.01$). Other side effects such as sedation (80%), dizziness (60%) and dry mouth (80%) occurred with similar frequency in both groups. A significant number of patients preferred the combination (15 v 1, $p < 0.001$); 14 had no preference) mainly because it was associated with less CNS side effects. There was no significant relation between the order of administration of the drug and drug preference (Fisher's exact test).

Trial 3

Eighty patients (47 women, 33 men; mean age 42 (range 18-68)) entered the study. All patients were receiving cisplatin in a dose range of 20-50mg/m² or the cisplatin analogues carboplatin or JM9. Ten patients failed to crossover because of change of chemotherapy in 5, protocol violation in 4 and extrapyramidal reaction in 1. There was complete control of nausea and vomiting in 24 patients (32%) given metoclopramide and dexamethasone and 14 patients (19%) of patients given nabilone and prochlorperazine. This difference was not significant but the scores for emesis on the linear analogue scale were significantly better for metoclopramide and dexamethasone (2.2+/-0.32) compared to nabilone and

prochlorperazine (3.5+/-0.37), p=0.018. With regard to side effects the following were significantly more frequent with nabilone and prochlorperazine; dizziness (p<0.001), sedation (p<0.01), dryness of the mouth (p<0.001) and dysphoria (p<0.05). Extrapyramidal reactions were only seen with the metoclopramide and dexamethasone combination.

There was no significant patient preference for either antiemetic (31 v 26; 13 no preference). However for patients given carboplatin, a significant proportion preferred nabilone and prochlorperazine (p=0.013). This was mainly because patients preferred the antiemetic given by the oral route. There was no significant relation between the order of administration of antiemetic and antiemetic preference.

Trial 4

Twelve of 20 (60%) of patients randomised to receive dexamethasone had no nausea or vomiting. This was significantly better than metoclopramide where control of nausea and vomiting was seen in only 5 of 20 (25%) patients (p=0.005). With domperidone, 8 of 20 (40%) patients experienced no nausea and vomiting, which was not significantly better than metoclopramide or significantly worse than dexamethasone.

Of the 35 patients who failed first line therapy, 12 received a second option. Of these, 6 received dexamethasone and only 1 experienced nausea and vomiting. The remaining 6 either received domperidone (n=3), or metoclopramide (n=3) but all experienced nausea and vomiting. No patient was given metoclopramide or domperidone as a third line antiemetic. Dyspepsia was similar with all 3 antiemetics (39-52%) and was in general extremely mild. The median duration (range) in days of the symptoms were as follows: nausea 3.5 (1-14); vomiting 2 (1-13); retching 2 (1-14); time for appetite to become normal and general activity to return to normal 3 (2-10) and 3.5 (2-14) respectively.

Non-randomised trial

Thirteen women and 2 men (mean age 57) took part in this open study. In 13 patients the chemotherapy regimen was unchanged from that which had caused nausea and vomiting previously and in 2 patients there was a minor modification of chemotherapy. The details of previous antiemetic therapy and chemotherapy regimens are shown in Table 3.8. No patient was given cisplatin. The 15 patients received 31 courses of GR38032F. Only 1 patient experienced nausea and vomiting. In a

Antiemetic (and dose x 4 daily)	No of patients
Domperidone 20mg+dexamethasone 4mg	6
Domperidone 20mg only	5
Dexamethasone 4mg only	2
Metoclopramide 20mg+dexamethasone 4mg	1
Metoclopramide 10mg+dexamethasone 4mg	1

Regimen (and dose in mg/m²)

Anthracycline 40+cyclophosphamide 500+ vincristine	6
Epirubicin 90-120	3
Mitozantrone 8+methotrexate 30	2
Doxorubicin 50+cyclophosphamide 450	2
Doxorubicin 40+etoposide 100	1
5-FU 750+epirubicin 40+cyclophosphamide 750	1

Table 3.8 Details of previous chemotherapy and previous antiemetic therapy in patients given GR38032F

subsequent course of treatment this same patient experienced transient mild nausea but no vomiting. None of the other patients had any nausea, retching or vomiting. Side effects reported were dryness of mouth in 1 patient, mild diarrhoea in 1 patient (who had a history of chemotherapy-associated diarrhoea) and mild sedation in 1 patient.

3.5.3. Discussion

The 4 randomised trials accrued 269 patients and provided important information on the efficacy of the various antiemetics tested (for summary see Table 3.9). Although the majority of these patients had tumours other than gastric cancer, the findings are relevant to the management of this tumour because there is no evidence that patients with gastric cancer react in a unique way to chemotherapy. For patients receiving cisplatin, trials 1 and 3 established the superiority of high dose metoclopramide over other antiemetics. In trial 3 metoclopramide was combined with intravenous dexamethasone because this combination has been shown to be better than metoclopramide alone (197). Also, in trial 3 metoclopramide was given as a loading dose (2mg/kg) IV followed by a

Best antiemetic

Trial 1

Cisplatin regimens high dose metoclopramide

Non-cisplatin regimens chlorpromazine

Trial 2

Non-cisplatin regimens nabilone and
prochlorperazine

Trial 3

Cisplatin regimens high dose metoclopramide
and dexamethasone

Trial 4

Non-cisplatin regimens dexamethasone

Table 3.9 Summary of the results of the 4
randomised antiemetic trials

continuous IV infusion (3mg/kg) because we have shown that this is the best way to achieve steady state plasma levels of metoclopramide in the region of 850-1000mg/ml (198) - the level at which metoclopramide functions optimally as an antiemetic (199). Therefore, at the present time for patients receiving cisplatin alone or in combination with other cytotoxic drugs we would recommend the use of high dose metoclopramide and dexamethasone.

However, these findings did not apply to the cisplatin analogue carboplatin. The group of patients who received carboplatin had an overall preference for nabilone and prochlorperazine. This preference was not based upon superior antiemetic efficacy. It was due to the oral route being the preferred method of receiving antiemetic therapy by these patients. This presumably reflects the fact that carboplatin is given as a short IV infusion and cisplatin is given as a prolonged IV infusion with IV fluids. Thus, the route of administration of antiemetic therapy is irrelevant to the patient on cisplatin but is extremely important to the patient on carboplatin.

With regard to the patients who were given chemotherapy regimens which did not include cisplatin the best antiemetics appeared to be

dexamethasone and the combination of nabilone and prochlorperazine. The mechanism by which dexamethasone is antiemetic is unknown but our experience would suggest it is very effective with few associated side effects. Nabilone is a semi-synthetic cannabinoid and when combined with prochlorperazine it is an extremely useful antiemetic treatment. Sedation and dry mouth are the most frequent side effects and these may be reduced in frequency by using a slightly lower dose of nabilone (1mg bd). Prochlorperazine was combined with nabilone to reduce the incidence of adverse effects on the central nervous system related to nabilone. It has been shown that combining a phenothiazine with the naturally occurring cannabinoid delta-9-tetrahydrocannabinol (THC), reduces the incidence of CNS side effects related to THC (200). Our double blind trial indicates that this effect occurs when a phenothiazine is combined with nabilone.

The dopamine antagonist, metoclopramide, when given in low dose (20mg tds) was not effective in the treatment of emesis caused by cytotoxic drugs, and yet in high dose it was. At these higher dosage levels metoclopramide antagonises 5HT at the 5-HT₃ receptor and it has been suggested that this might be one of the ways

in which metoclopramide is antiemetic against cytotoxic drugs (201). Using an animal model of cytotoxic drug induced vomiting the 5-HT₃ receptor antagonist, GR38032F, was shown to be a potent antiemetic (202). Our clinical trial confirms these results. Moreover, GR38032F was effective in patients who had failed conventional antiemetic treatment and its use was associated with very few side effects. These findings suggests that this compound and other 5-HT₃ receptor antagonists will have an important role in the future management of the nausea and vomiting induced by cytotoxic drugs.

With the advent of better antiemetic treatments, effective palliation with chemotherapy of the common solid tumours, including gastric cancer, should become more realistic. Chemotherapy which produces objective tumour regression in only 50% of cases will be of more clinical value because it can be administered with the minimum of side effects.

CHAPTER FOUR

THE
SUBRENAL CAPSULE ASSAY
AS A MEANS OF PREDICTING
RESPONSE TO CHEMOTHERAPY

4.1 INTRODUCTION

The six day subrenal capsule (SRC) assay was developed by Bogden et al (203) as an in vivo xenograft system that could be used to investigate the effects of cytotoxic agents on a variety of human tumours from xenografts established in nude mice, and on primary surgical explants of human tumours (204). The technique involves the establishment of a first generation xenograft by means of transplantation of fresh human tumour (or tumour from a xenograft) under the renal capsule of normal immunocompetent mice. Initially, nude mice were used as the recipients of the tumour but later it was shown that it was possible to use normal immunocompetent mice (205). It was also shown that the growth rate of the xenografts could be inhibited by the administration of cytotoxic drugs to the mice, and that the degree of growth inhibition provided a precise in vivo method of measuring the sensitivity of the transplanted tumour to the administered cytotoxic drug (205). Also, because the technique involved the use of immunocompetent mice, rather than immunodeficient athymic nude mice, transplantation was possible without the creation of a highly sterile environment. These factors indicated that the SRC

assay should be very cost effective and the short time frame of the assay was seen as a major advantage when compared to the time taken to grow xenografts subcutaneously in nude mice. In addition, the use of intact solid tumour fragments was considered to have greater biological relevance than other systems such as the in vitro human cloning assay of Hamburger and Salmon (206)

As outlined in Chapter Three, chemotherapy regimens will usually produce objective tumour regression in only 30-45% of patients. Nevertheless, those patients who respond to chemotherapy benefit in terms of survival, but the price is the unnecessary toxicity of those who do not respond. We therefore felt that this was a suitable tumour in which to study a predictive chemotherapy system such as the SRC assay. Preliminary information from Bogden's group suggested that it should be feasible to grow primary surgical explants from gastric cancer (207), and therefore we initiated a study to investigate the possible role of the SRC assay as a chemopredictive test in gastric cancer.

4.2 MATERIAL AND METHODS

Surgical Explants

Resected specimens were placed immediately into tissue culture medium No 199 (TC 199) and taken to the pathology department where a sample approximately 1 cm^3 was removed from the primary tumour. The tumour was dissected into $4 \times 0.5 \text{ cm}^3$ pieces and returned to the tissue culture medium for transfer to the laboratory. All patients received their operation in the centre where the laboratory was located, therefore transportation time for specimens was minimised. In the laboratory the tumour was trimmed of extraneous connective tissue and necrotic areas. It was prepared for transplantation by dissection into pieces approximately 1 mm^3 using a template placed under a glass petri dish. After dissection, the pieces of tissue were put in a separate petri dish containing TC 199 and placed on ice. The sizes of the cut pieces of tumour were checked using a Zeiss stereoscopic microscope with a micrometer scale calibrated so that 1 micrometer unit (MU) was equivalent to 0.1 mm. Average diameters ranged from 10-15 MU. Pieces smaller or larger than this were discarded. A specimen was retained for histological analysis. The mice (C57BL/6 x DBA/2

F1, hybrid) age 14-20 weeks were ear marked, weighed, and anaesthetised with diethyl ether. The left side of each animal was shaved and wiped with a 70% alcohol solution, then an incision was made laterally in the region of the left kidney, which was subsequently exteriorised (Figure 4.1). An incision was made in the renal capsule (Ziegler iridectomy knife with a 4 mm blade) and the tumour introduced using a trochar (supplied by MacCarthy's Surgical Ltd, Dagenham, UK) (Figures 4.2 and 4.3). The inside of the trochar was made wet by placing it in TC 199 before insertion of the tissue. Once under the renal capsule (Figures 4.4 and 4.5) 2 perpendicular diameters of tumour were measured using the stereoscopic microscope. After measurement the kidney was carefully resited in the abdomen, and the incision was closed using a stapler gun (Figure 4.6). Each control group had 5-8 mice and each treatment group had 4-6 mice. All surgery was carried out by one operator (David Cunningham).

Six days later the mice were sacrificed by cervical dislocation, re-weighed and the tumour re-measured in situ. An evaluable assay was one in which the mean increase in the sum of the 2 perpendicular diameters was 0.5 MU (which is the same criterion used by Bogden et al (203)).

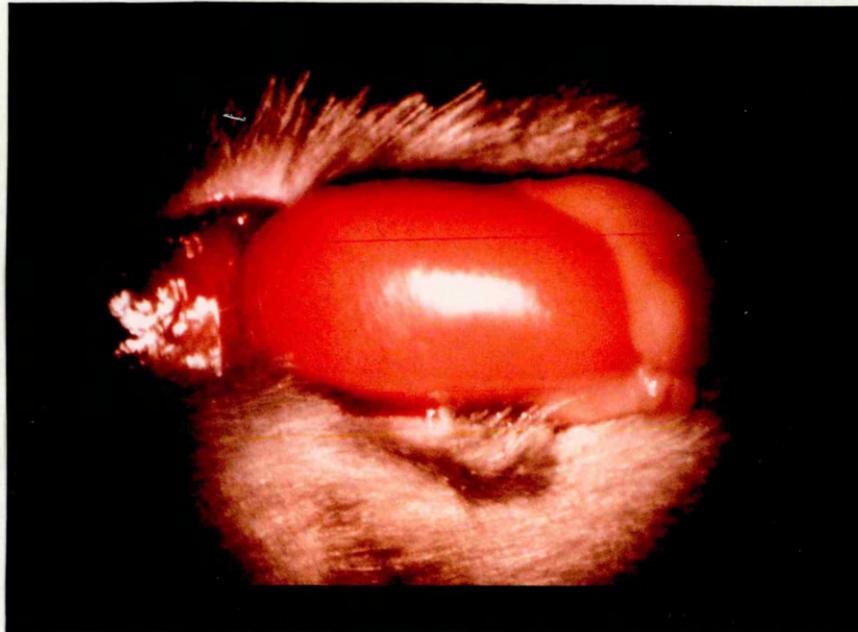


Figure 4.1 After an incision in the left flank the left kidney was exteriorised.

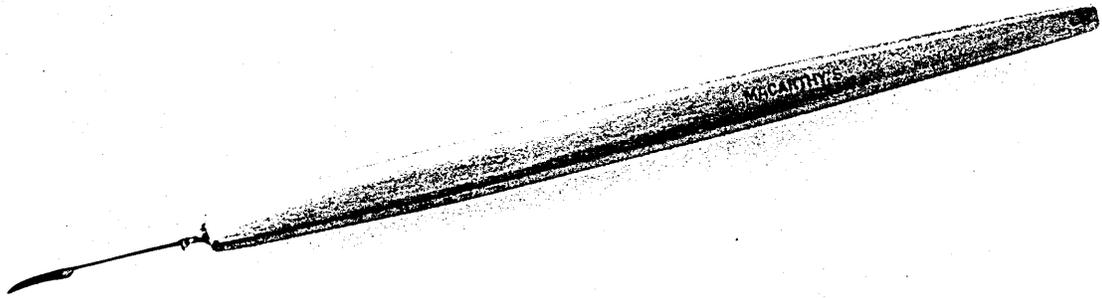


Figure 4.2 Ziegler knife

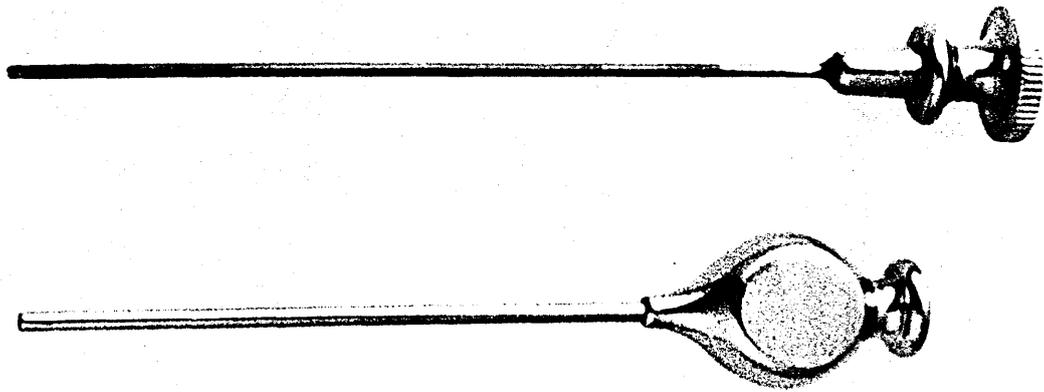


Figure 4.3 The trochar used to introduce tumour under the renal capsule

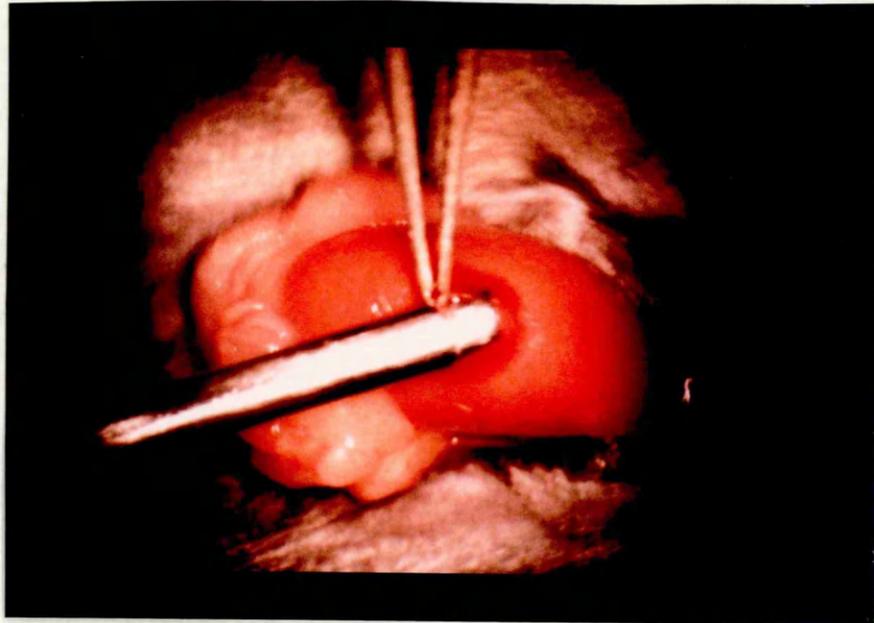


Figure 4.4 The trochar is under the renal capsule loaded with tumour

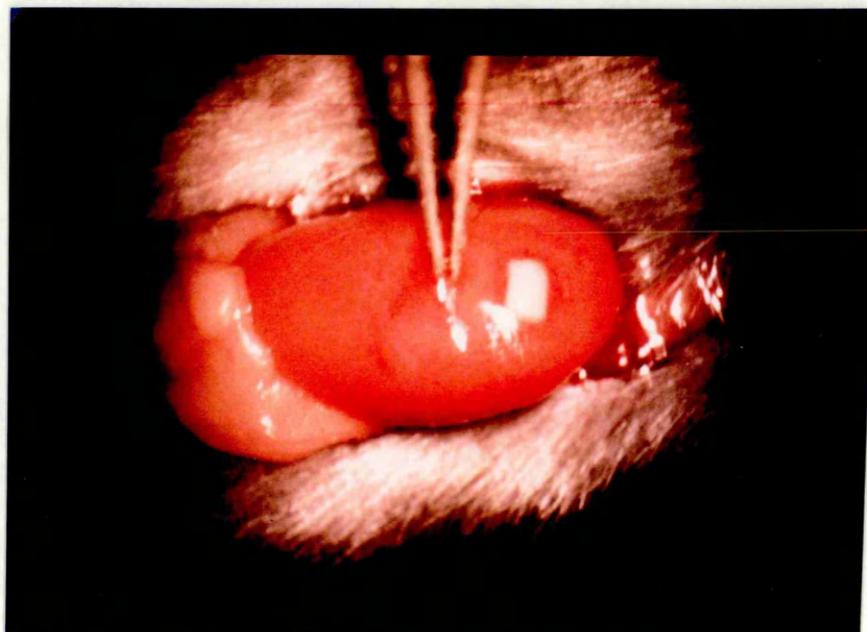


Figure 4.5 The appearance after the tumour has been placed in position



Figure 4.6 The incision after closure with staples

Thereafter, the tumour bearing kidney was removed and put in formal saline.

Chemotherapy

Chemotherapy was given on Days 1,3 and 5 as outlined in Table 4.1. The drugs tested were those of a 4 drug combination chemotherapy regimen which we were using to treat advanced gastric cancer. It was proposed that this regimen would be used to treat on relapse, patients from whom we had obtained tumour tissue for this study at the time of initial surgical resection.

Pathology

Sections, prepared from the mouse kidney and implanted tumour, were stained with haematoxylin and eosin. They were examined by one pathologist without the knowledge of the group of origin of the specimen (i.e. control or treatment). An Inflammation Score was devised in which the degree of inflammatory infiltrate was graded as follows: none=0, mild=1, moderate=2 and marked=3.

Tumour from an animal allograft and a human xenograft

In addition to the tissue from surgical implants, tumour was also obtained from a human

Drug	Dose mg/kg/injection	Method Administration
Cisplatin	3.3	Subcutaneous
Epirubicin	6.6	Intravenous
Methotrexate	6.6	Subcutaneous
5-Fluorouracil	80.0	Subcutaneous

Table 4.1 Details of chemotherapy administered to the mice

xenograft and an animal allograft for transplantation. The xenograft was Wils tumour, which is a human lung adenocarcinoma established in nude mice and the animal allograft was the Walker 256 tumour which is a rodent sarcoma grown in Wistar rats.

4.3 RESULTS

Surgical Explants

Specimens of gastric adenocarcinoma were obtained from 17 patients. Sixteen were from the primary tumour and 1 was from a lymph node metastasis. The specimens retained for histological analysis all showed tumour. From the 17 gastric tumours a total of 418 xenografts were transplanted and the mean time for transplantation of one xenograft was 4.8 minutes. the mean time for tumour resection to completion of transplantation was 3hrs 45minutes (range 2hrs 10mins - 4hrs 40mins). Fourteen of the 17 tumours gave evaluable assays in the control groups with a mean increase in the sum of the tumour diameters of 4.1 M.U. (range 1.6 - 10.5).

However, when the xenografts were examined histologically tumour cells were present in only 26 (6%) and the remaining xenografts consisted of fibrous tissue and a lymphocytic infiltrate. It should be emphasised that although a small number of xenografts contained tumour, this consisted of a few acini or single tumour cells as shown in Figure 4.7. The extent to which the xenografts were infiltrated by lymphocytes was significantly influenced by the administration of cytotoxic

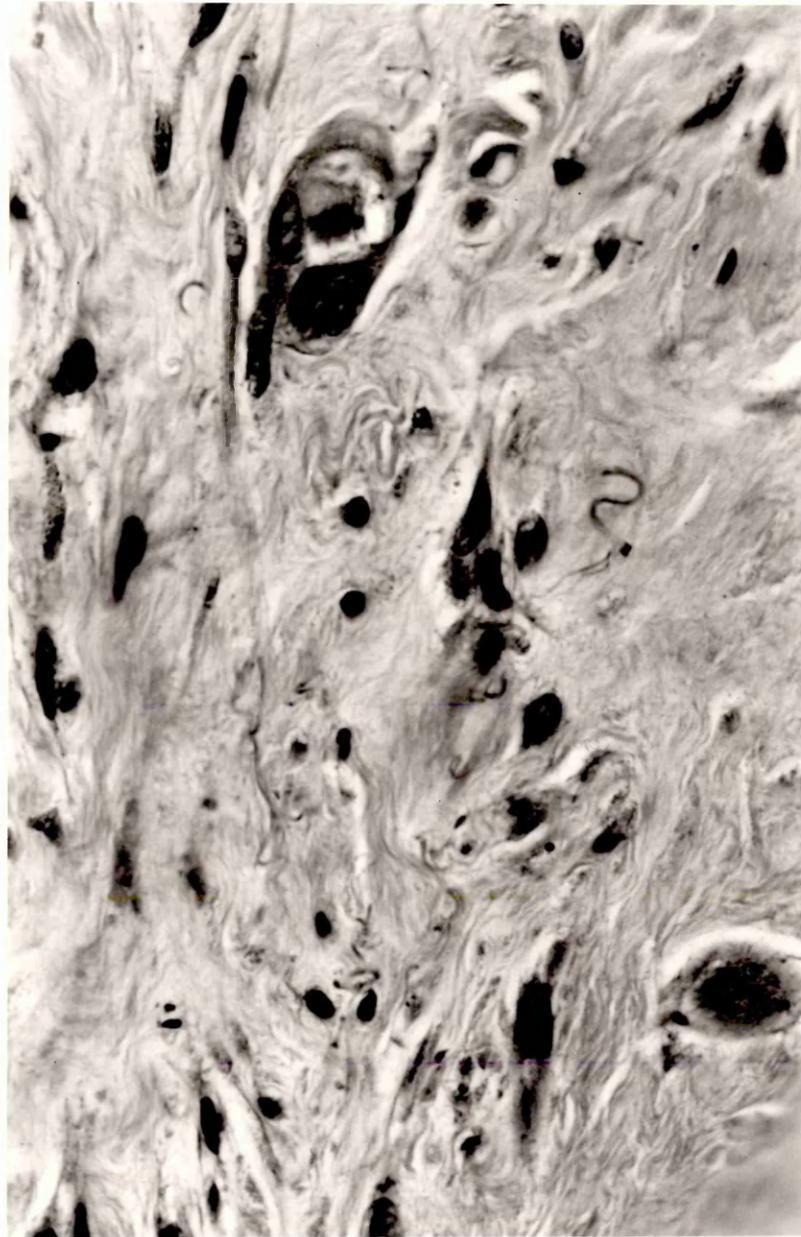
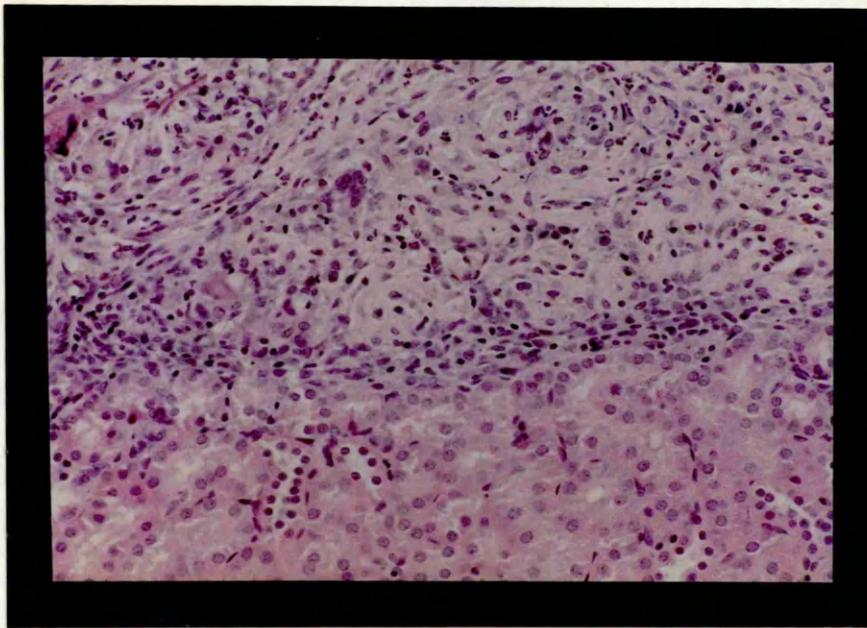


Figure 4.7 Section of the subrenal capsular xenograft six days after implantation. A single residual acinus of gastric carcinoma is present. This finding was exceptional, most animals showing no evidence of tumour. (The figure appears refractile because of the presence of collagen.) (H and E x 753)

drugs. Xenografts from control groups (Figure 4.8) invariably had a pronounced lymphocytic infiltrate compared to treated groups (Figure 4.9). These findings were reflected in the Inflammation Score which is represented in figure 4.10. There was a significantly positive correlation ($p < 0.001$) between the Inflammation Score and the increase in size of xenografts (Spearman's Rank Correlation). In the case of one tumour, rather than giving the treatment groups chemotherapy, the animals were sacrificed on days 1, 3 and 5 and the tumours were examined histologically. This revealed, as might be expected, a progressive increase in the amount of lymphocytic infiltrate over the 5 day period. Also, there was no tumour seen in the transplanted tissue from day 1, despite the presence of viable tumour in the specimen which was examined histologically just before transplantation.

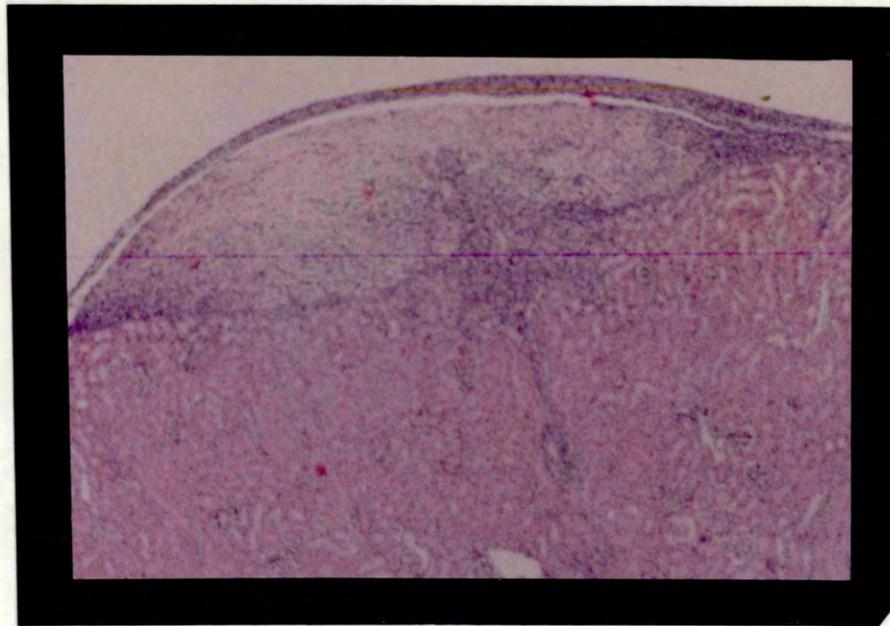
Tumour from the human xenograft and the rodent allograft

In both the xenografts there was considerable growth over the 6 day period. The mean increases in tumour diameters for the Wils tumour and the Walker 256 tumour were 11 MU and 65 MU respectively. Histology of the Wils tumour taken from a control and a treatment group is



A

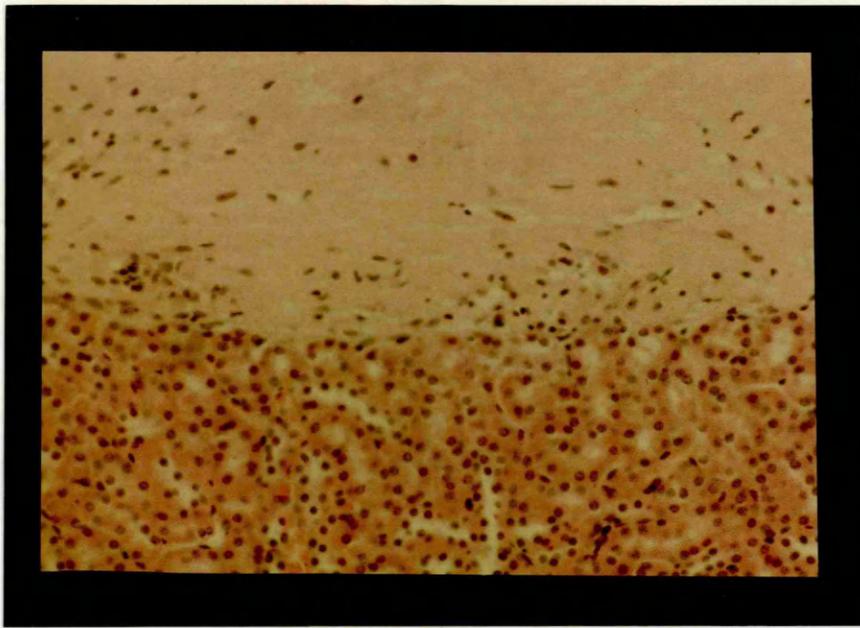
H and E x 250



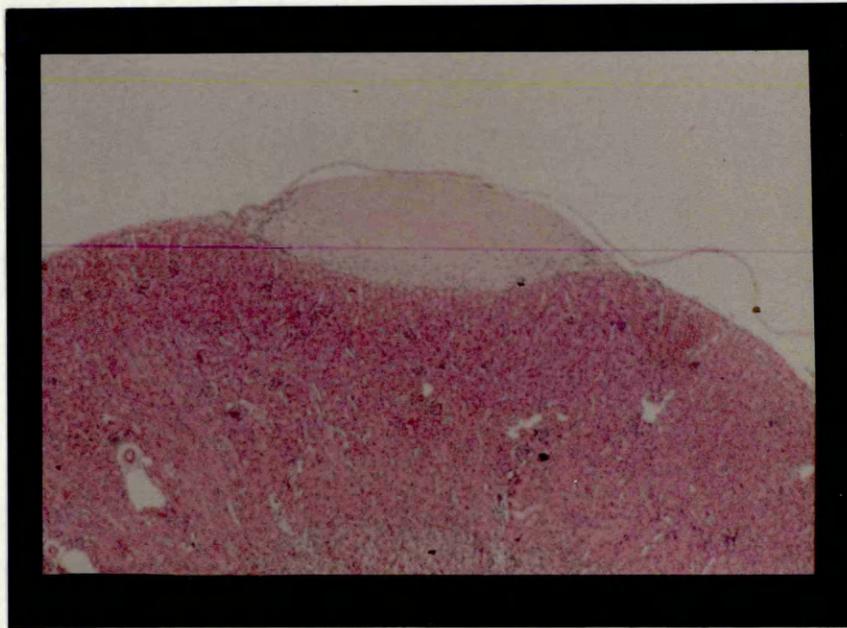
B

H and E x 40

Figure 4.8 Sections of the xenograft from control animals. An intense lymphocytic infiltrate is present which extends into the renal parenchyma.

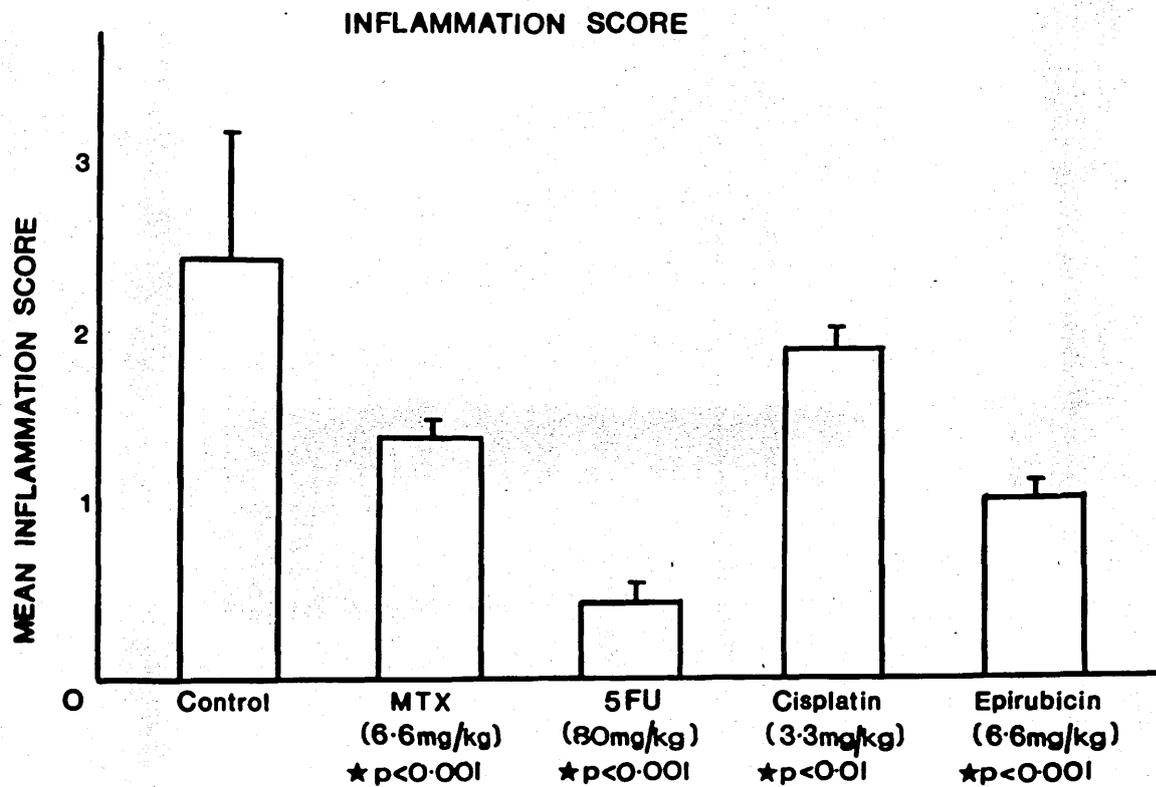


A H and E x 250



B H and E x 48

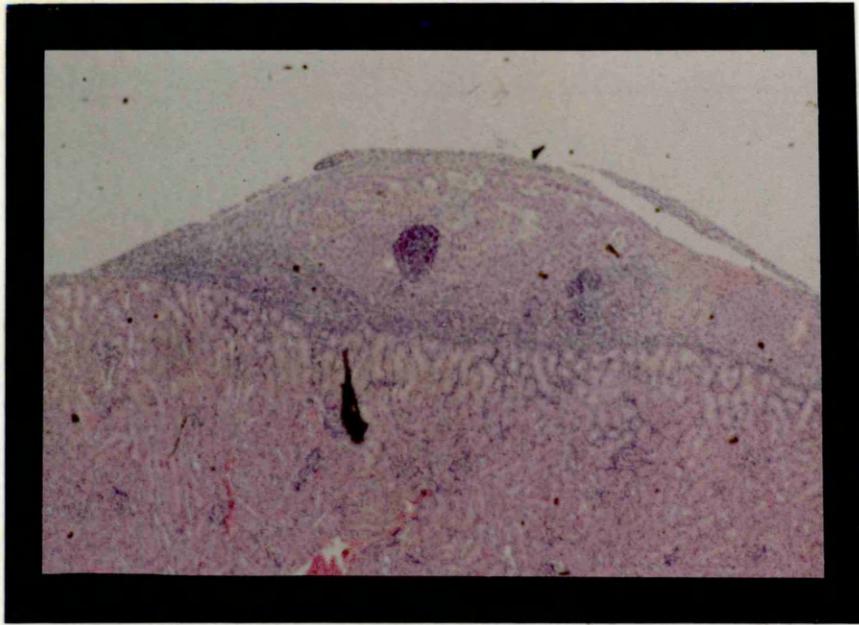
Figure 4.9 Sections of xenograft from a 5-FU treated animal. In contrast to Figure 4.8 there is no significant inflammatory cell infiltrate.



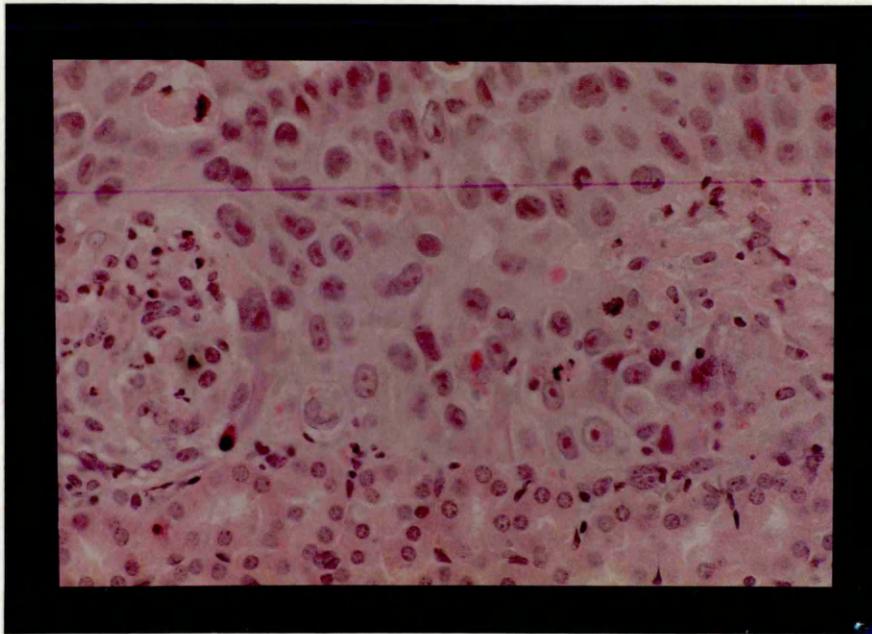
★WILCOXON'S SIGNED RANK TEST

Figure 4.10 Inflammation Score for 418 xenografts derived from primary surgical explants.

shown in Figures 4.11. A typical xenograft from the Walker tumour, on day 6, is shown in Figure 4.12. and the histology in Figure 4.13. Histology of both xenografts confirmed the increase in size was due to tumour. There was persistence of the lymphocytic infiltrate but in view of the marked tumour growth, this made a negligible contribution to the increase in xenograft size.



A H and E x 40



B H and E x 300

Figure 4.11 Sections of the Wils tumour taken from a control (A) and treatment group (B).

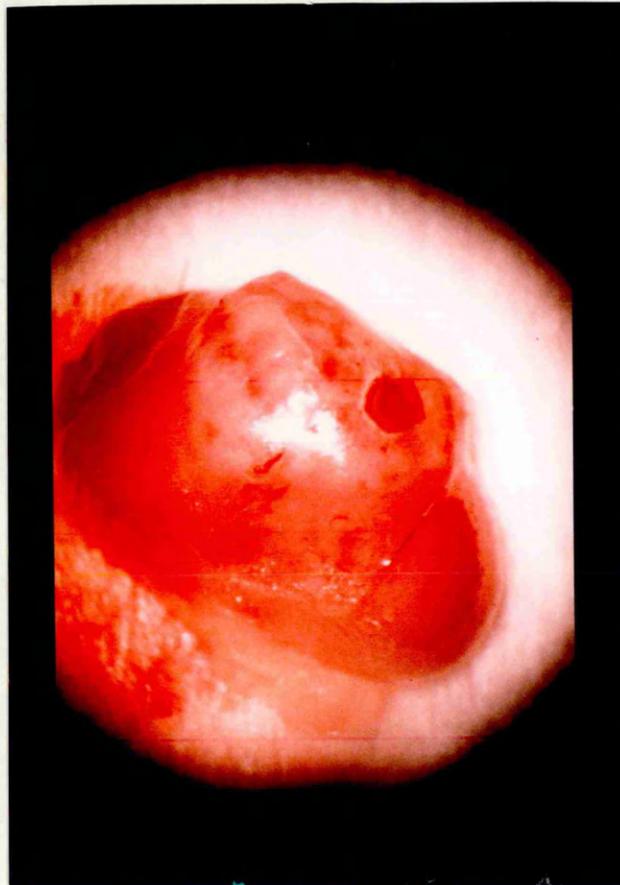


Figure 4.12 Macroscopic appearance of the Walker 256 sarcoma six days after transplantation under the renal capsule.

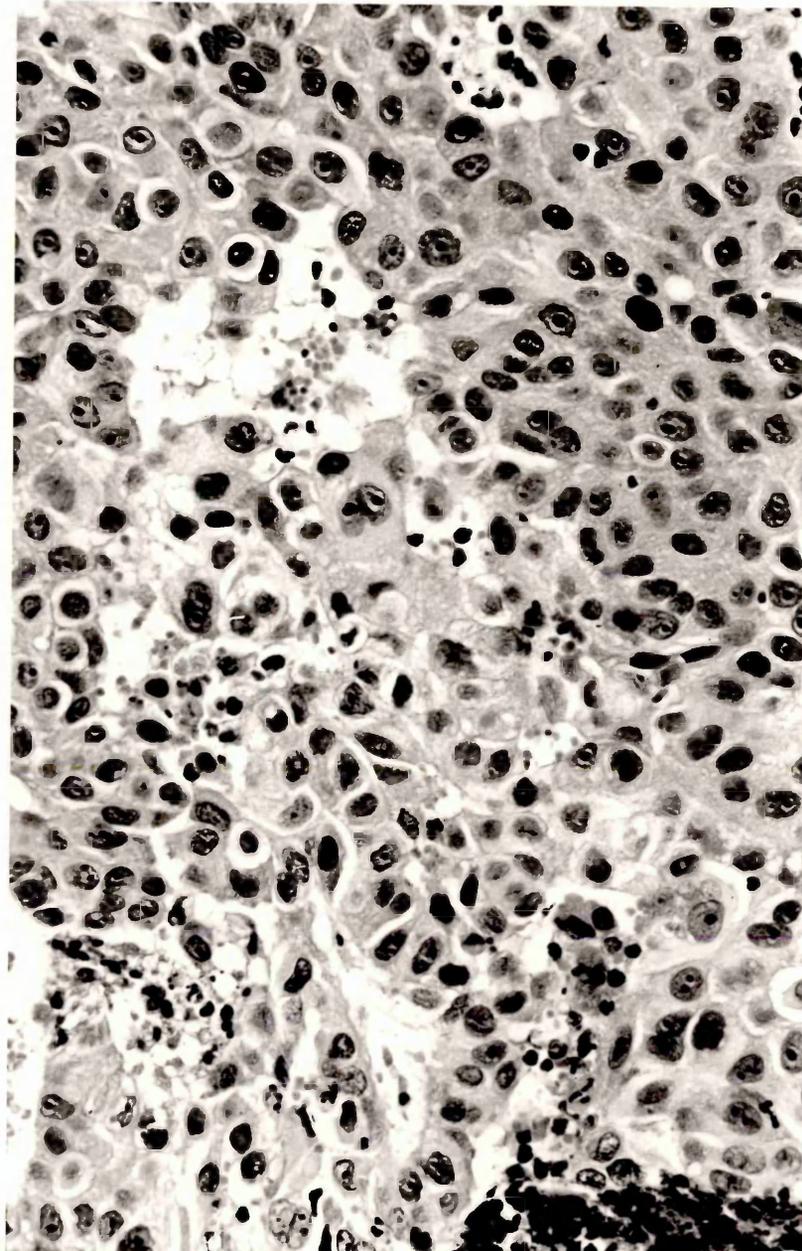


Figure 4.13 Section of Walker 256 tumour six days after subrenal capsular transplantation. Areas of necrosis are present, but most of the tumour is viable. (H and E x 395)

4.4 DISCUSSION

Although the original aim of this study was to investigate the usefulness of the SRC assay as a chemopredictive test for gastric cancer, it became clear from the histological analysis of the transplanted xenografts that this would not be feasible. However, we have demonstrated a very important size artefact related to the infiltration of the xenograft by lymphocytes. This is disappointing because the SRC assay theoretically offers several advantages over other chemopredictive systems such as the clonogenic assay (206). The SRC assay maintains cell to cell contact and does not depend upon the highly selective cell population of the clonogenic assay in which tumour colonies grown on agar are derived from a very small proportion of the original tumour (206).

When Bogden et al (205) first introduced the SRC assay, it was suggested that the short time frame required for tumour growth would render any immunological response to the implanted tumour irrelevant. Subsequent experience was to the contrary; Edelstein et al (208) have shown that 6 days after transplatation, the tumour is usually infiltrated by mouse "response cells".

Nevertheless, there was still debate about the significance of this infiltrate, especially in terms of its interference with assessment of the assay. Using flow cytometry, Aamdal et al (209) demonstrated that for human tumours established in nude mice and subsequently transplanted under the SRC, that the contribution of the lymphocytic infiltrate to the overall volume of the xenograft was 15-25%. On the basis of these results they concluded that the inflammatory infiltrate was of little consequence to the measurement of xenograft growth.

For primary surgical explants from gastric cancer we have shown conclusively that there is an inherent defect within the SRC assay. Apparent growth, as a result of lymphocytic infiltration occurred in control xenografts which did not contain viable tumour cells. The amount of lymphocytic infiltration was reduced by administration of cytotoxic drugs, so that xenografts exposed to cytotoxic drugs were smaller than the controls. Using the parameters of tumour measurement in this situation is inadvisable, and might lead to the erroneous assessment of a cytotoxic agent's activity against a given tumour. Thus, for tumour derived from primary surgical explants, histological validation of the assay is

essential. Levi et al (210) modified the SRC assay to include an histological scoring system which encompassed the extent of the tumour necrosis and the amount of lymphocytic infiltration. This is more reliable than tumour measurement alone, but has the disadvantage of making the SRC assay more complex, and time consuming to perform.

Despite these findings, Bogden continues to report "a high rate of evaluable specimens" without reference to histological validation (211) and in a recent review article concluded "host cellular reactions are not artefactual to a tumour size parameter for evaluating tumour response" (212). Similarly a recent study from France emphasised the value of the SRC assay as a chemopredictive test in cervical cancer but made no mention of the histology of the xenograft (213).

It may be argued that the "take rate" of the assay could be improved by only using tissue confirmed by frozen section to contain viable tumour cells. However, growth of tumour using the SRC technique was still unsuccessful in 4 of our 17 tumours where the presence of viable tumour cells was confirmed histologically by frozen section prior to transplantation.

Our experience with the transplantation of

human and animal tumour xenografts under the SRC has been limited to 2 tumours, both of which grew consistently well within the SRC assay. The growth was on such a large scale that routine histological validation was not essential and provided no obvious advantage over macroscopic tumour measurement. Moreover, in this context the "size artefact" related to lymphocytic infiltration was less relevant. Indeed, we have successfully used the SRC assay to investigate circumvention of drug resistance with verapamil in Wils tumour (214) and to examine low density lipoprotein as a cytotoxic drug targeting vehicle in the walker 256 tumour (215).

The use of the (SRC) assay as a chemopredictive test in gastric cancer did not prove feasible. Important defects within the SRC assay were identified. In particular, a lymphocytic infiltration of the dead, transplanted tumour tissue can produce apparent tumour growth. Thus, stricter criteria of tumour growth including histological confirmation of viable tumour in the transplant are essential to prevent misleading results from the SRC assay.

CONCLUSIONS

In this thesis we have defined the group of patients with gastric cancer who do well after surgical resection. Patients in whom the tumour has penetrated the serosa do badly, even if they have an apparently curative resection. For this group of patients, surgery alone is insufficient treatment and some form of adjuvant therapy is clearly required. The best candidate at the moment is chemotherapy but so far the results of adjuvant chemotherapy have been disappointing. Another possible approach involves the use of intraperitoneal chemotherapy given in the adjuvant setting because as outlined in Chapter Two, tumours which have penetrated the serosa are often associated with free cancer cells in the peritoneum. Cisplatin, one of the most active drugs in gastric cancer, has been given by the intraperitoneal route to treat ovarian cancer but not gastric cancer. This route of administration effectively delivers 15 times the equivalent intravenous dose to the peritoneum and because it is absorbed via the peritoneum it also has a systemic effect (216). This systemic effect would be necessary to eradicate micrometastatic disease outwith the peritoneum.

The Japanese experience has shown that it is possible to improve the prognosis for patients

with gastric cancer by making an early diagnosis. In the U.K. preliminary results from a dyspepsia clinic, in which patient groups with an accentuated risk of developing gastric cancer are screened have been encouraging. If these results are confirmed it would be worthwhile establishing similar clinics throughout the country.

With regard to advanced disease, further trials of chemotherapy are required to determine new, active cytotoxic drugs and the best combination of cytotoxic drugs for clinical use. Promising additions to the list of active single agents include cisplatin and triazinate but the bench mark remains 5-FU. Outwith clinical trials, the patients most likely to benefit from chemotherapy are those without liver metastases. For some of these patients successful treatment may result in sufficient tumour regression that will allow the resection of a primary tumour initially deemed to be inoperable. Therefore, the potential benefits from chemotherapy of this subgroup of patients are considerable, particularly if side effects of treatment such as nausea and vomiting can be abolished, and with the new 5-HT₃ antagonists this may become possible. For the patients with liver metastases, regional chemotherapy such as 5-FU given via the

hepatic artery deserves investigation and could be combined with systemic treatment. Other possible therapeutic avenues include intra-hepatic microspheres (217) which can be used as a vehicle for targeting cytotoxic drugs against the tumour. There will also be patients for whom immediate cytotoxic drug therapy may be inappropriate; for example it would be difficult to justify the use of chemotherapy in an elderly patient with asymptomatic liver metastases. Such a patient should be carefully reviewed and only offered chemotherapy if the tumour gives rise to symptoms.

By focusing on these areas eventually it may be possible to improve the outlook for our patients with gastric cancer. Certainly, there is no place for therapeutic nihilism or the remarks of Mayo Robson made in 1900 will remain equally relevant at the end of this century.

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