The Effect of 5-Fluorouracil Chemotherapy on Energy Metabolism, Body Composition and Quality of Life in Patients with Colorectal Liver Metastases

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IV LIST OF ABBREVIATIONS

BIA	Bio-electrical impedance analysis
BMI	Body mass index
CEA	Carcinoembryonic Antigen
cm	centimetre
CRP	C-reactive protein
CT	Computed Tomography Scan
ECF	Extra-cellular fluid
EORTC	European Organisation for Research and Treatment of Cancer
FFM	Fat-free mass
FM	Fat Mass
5-FU	5-Fluorouracil
g	gram
kg	kilogram
KPS	Karnofsky Performance Status
l	litre
m	metre
mg	milligram
mm	millimetre
NS	not significant
r	rank correlation coefficient
TBW	Total body water
WHO	World Health Organisation

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VI. DECLARATION

I declare that the work presented in this thesis has been carried out solely by myself, except where indicated below.

Dr.D.C.McMillan and Sister. Elizabeth Kerr assisted in data collection for the study described in chapter 3.

VII. DEDICATION

To my mother, Maria,

for all her love, dedication and support.

VIII. SUMMARY

Chemotherapy for metastatic colorectal cancer has been used for palliation for many years. During this time 5-FU has remained the single most effective anti-neoplastic agent in the treatment of colorectal cancers. Whilst many studies have assessed the safety and efficacy of chemotherapeutic regimens using response rate, toxicity profiles and quality of life, few studies have evaluated the effect of chemotherapy on the metabolic response of the host in particular energy balance, body composition and quality of life.

The aim of this thesis was to examine the relationship between the systemic inflammatory response and body composition, dietary intake, resting energy expenditure, cytokine, hormone and protein metabolism in patients with colorectal liver metastases. In addition, the short and long term-effects of 5-FU based chemotherapy on the above measures of host metabolism, and quality of life in patients with colorectal liver metastases were assessed.

In chapter 3, the host metabolic response of 5-FU based chemotherapy was assessed in patients with colorectal liver metastases. In 25 patients, haematological and biochemical parameters were measured before and after 6 cycles of chemotherapy. In a subset of 11 patients, energy balance, body composition, acute phase protein response and quality of life were measured. The results of this study indicate that, in patients with colorectal liver metastases receiving 5-FU based chemotherapy, a number of changes occur in both haematological and biochemical parameters. These changes were consistent with those commonly associated with chemotherapy. However, it was of interest that positive acute phase proteins such as fibrinogen fell in concentration during the course of the study while other proteins such as globulins did not. These results are consistent with 5-FU based chemotherapy resulting in a reduction of the systemic inflammatory response.

Measured resting energy expenditure values were also noted to be higher than would be predicted whether expressed as kilocalories per day (median 113%) or per lean body mass derived from total body water (median 120%). There were no significant differences in energy intake or resting energy expenditure during the course of the study. Furthermore, in the present study where the majority of patients responded to treatment, there were no

alterations in any of the body composition parameters. These results are therefore consistent with previous work which concluded, in lung cancer patients, that there was evidence for tumour induced hypermetabolism independent of changes in gross body composition, although the absolute increase in hypermetabolism is small.

Global quality of life and Karnofsky performance status also did not change significantly during the course of the study. There was no change in physical function as measured by performance status and would suggest that the effect of 5-FU based chemotherapy, if any, on host energy metabolism is small. In summary, in patients with colorectal liver metastases, 5FU chemotherapy is 'fairly well' tolerated and is associated with a reduction in white blood cells, liver enzymes and acute phase proteins. In the sub-group there were no detectable effects on energy metabolism, body composition or quality of life.

Tumour markers such as CEA have been demonstrated to be effective in alerting clinicians of the possibility of disease progression in patients with colorectal cancer. Furthermore, the presence and the magnitude of a systemic inflammatory response has also been reported to be associated with disease progression in patients with colorectal cancer. However, overall, few 'host markers' have been assessed in the development of disease progression in patients with colorectal cancer.

Chapter four details a study of biochemical and haematological parameters in disease progression of patients with colorectal liver metastases receiving 5-FU based chemotherapy. Host factors that changed prior to CT defined progression were examined in 11 patients with colorectal liver metastases by comparing periods of 'responding disease' to 'progressive disease'. In addition, longitudinal assessment of patients prior to disease progression was assessed.

On comparison of parameters during a period of 'responding' disease and 'progressive' disease, an increase in white cell count, neutrophil count and C-reactive protein on disease progression was observed. With regard to longitudinal assessment, white cell count, C-

reactive protein and carcinoembryonic antigen were found to significantly change prior to CT-defined disease progression.

Therefore, the results of this study may suggest that 12-14 weeks prior to CT-defined disease progression there is tumour growth (as indicated by carcinoembryonic antigen levels) causing cell damage (as indicated by the transaminases) and necrosis, resulting in systemic inflammation, which in turn results in changes in liver protein production (C-reactive protein). There are increasing numbers of reports that suggest that this tumour-host inflammatory response is detrimental to the patient and may indeed promote disease progression in colorectal cancer. Since these events can be identified prior to CT-defined progression, markers of the acute-phase response such as white cell count and C-reactive protein, used in combination with carcinoembryonic antigen, may be useful in providing an 'early warning' of tumour progression in patients with colorectal liver metastases. Furthermore, it may be that the presence of an inflammatory response before starting chemotherapy is a poor prognostic indicator.

Chapter five describes a study that assessed the longitudinal relationship between leptin and energy metabolism, fat mass and appetite in patients with colorectal liver metastases undergoing 5-FU based chemotherapy. Sixteen weight stable patients with colorectal liver metastases undergoing chemotherapy were assessed fortnightly during 3 cycles of chemotherapy and compared to a group of nine normal subjects.

The results demonstrated a significant correlation between circulating leptin concentrations and percentage body fat. However, circulating concentrations of leptin were lower in the cancer group. In the cancer patients measured body fat was significantly greater than predicted body fat. Furthermore, circulating leptin concentrations were significantly correlated with measured but not predicted body fat mass. Changes in body fat mass of cancer patients over time were also correlated with leptin concentrations. These results therefore suggest that circulating leptin concentrations accurately reflect fat mass over time and that any effect of cancer on systemic concentrations is likely to be small.

The changes in circulating leptin concentrations were not associated with alterations in appetite, energy intake, resting energy expenditure or respiratory quotient. Furthermore, it was of interest to note that there was no association between the concentrations insulin/cortisol or their ratio and leptin concentrations, indicating that the control of energy balance in humans is more complex than previously thought.

In the present study, biceps and triceps skinfold thicknesses did not correlate with measured body fat mass. Given that bioelectrical impedance accurately measures fat mass in cancer patients the results would suggest that circulating leptin concentrations are a more robust indicator of fat mass than anthropometry in patients with cancer. Changes in circulating leptin concentrations reflected changes in body fat mass and suggest that leptin concentration may be a useful routine marker of nutritional status in cancer patients.

1.0 INTRODUCTION AND AIMS

1.1 Colorectal Cancer

1.1.1 General

In the United Kingdom, colorectal cancer is the third most common cancer after lung and breast cancer (The Cancer Research Campaign, 1999). There were over 32,000 new cases of colorectal cancer in 1995 and over 17,000 deaths attributed to the disease during 1997 (The Cancer Research Campaign, 1999).

More than 60% of patients who present with colorectal cancer are 60 years or older (The Cancer Research Campaign, 1999). There are marginally more cases of colon cancer in females than in males (male: female ratio 1.0:1.1), and more cases of rectal cancer in males than in females (1.0:0.7) (The Cancer Research Campaign, 1999).

Risk factors thought to be associated with colorectal cancer include a high consumption of red or processed meats, fat, alcohol and obesity. Possible preventative factors include a high fibre, fruit and vegetable intake, frequent exercise (The Cancer Research Campaign, 1999) and the administration of non-steroidal anti-inflammatory drugs (Smalley et al., 1999).

1.1.2 Pathogenesis of Colorectal Cancer

The majority of colorectal cancers, approximately 60%, are said to be 'sporadic (Ivanovich et al., 1999)'. Vogelstein and colleagues (1988) have described a multistep model for the genetic events in the progression of sporadic colorectal cancer. These events are commonly referred to as the adenoma-carcinoma sequence. They suggest that a cell must develop several defects through the activation of oncogenes and the inactivation of suppressor genes, in order to be fully malignant.

Normal Epithelium
↓ (chromosome 5 alteration)
Hyperproliferative Epithelium
↓
Adenoma Class I (<1.0 cm)
↓ (*ras* gene mutation)
Adenoma Class II (>1.0 cm)
↓ (chromosome 18 allele loss)#
Adenoma Class III (large adenoma with foci of carcinoma)
↓ (chromosome 18 allele loss)
Carcinoma
↓ (other chromosome loss)
Metastasis

Polyps that are more likely to undergo malignant transformation include those that are larger and have a greater degree of dysplasia (Kettlewell, 1994). Furthermore villous adenomas are more prone to malignancy than tubular adenomas (Kettlewell, 1994). Patients presenting with a solitary adenoma have a 1 in 20 risk of developing invasive cancer at 15 years. Those who present with multiple adenomas have 1 in 8 risk (McArdle, 1998).

Approximately 10% of the population are thought to have a genetic predisposition to colorectal cancer (McArdle, 1998). In 1% of these patients, hereditary factors are known to be the cause. Familial adenomatous polyposis coli (FAP) accounts for approximately 1% of all hereditary colorectal cancers and hereditary non-polyposis coli (HNPCC) accounts for 5-10% (Ivanovich et al., 1999).

1.1.3 Screening of Colorectal Cancer

At present there are two main methods of screening for colorectal cancer: faecal occult blood testing and flexible sigmoidoscopy.

Faecal occult blood testing is a relatively inexpensive method and appears to involve little patient discomfort, however compliance is variable and has been reported to be approximately 60% in some studies (Hardcastle et al., 1996). Studies have indicated that faecal occult blood testing leads to an increase in detection rates in patients with early stage tumours, Dukes A and B (Kewenter et al., 1991; Kronborg et al., 1996), which may in part decrease overall mortality from this disease (Mandel et al., 1993; Hardcastle et al., 1996; Kronborg et al., 1996).

Flexible sigmoidoscopy is also a quick and relatively simple method of screening. However, the disadvantage of this method is that some adenomas and early cancers cannot be observed due to limited anatomical access. Studies have indicated that in patients with colorectal cancer whose cancers can be visualised, mortality may be decreased (Neewcomb et al., 1992; Selby et al., 1992). Ransohoff and Lang (1993) advocate that sigmoidoscopy should be carried out in patients aged 50-75 years every 3-5 years.

At present there is a study being conducted by the Medical Research Council regarding the use of flexible sigmoidoscopy as a screening tool in the detection of colorectal cancer. The results are presently awaited.

1.1.4 Clinical Features of Colorectal Cancer

The majority of colorectal cancers (approximately 70%) arise in the sigmoid colon, rectosigmoid junction and rectum (Cancer Research Campaign, 1993). Right sided colorectal tumours often present with symptoms of iron deficiency anaemia, weight loss or a palpable right iliac fossa mass (Kettlewell, 1994). If obstruction of the bowel occurs, this is usually a relatively late presentation.

Left sided colonic tumours usually present with a change in bowel habit (alternating between constipation and diarrhoea), lower abdominal colic and distension (Kettlewell, 1994). Patients with rectal tumour usually present with bleeding per rectum, tenesmus or alterations in bowel habit (Kettlewell, 1994).

1.1.5 Surgery of Colorectal Cancer

The majority of patients with primary colorectal cancer will undergo surgical resection. According to the SIGN guidelines (1997), colorectal cancer should be based on the following 'oncological and anatomical principles':

- 'Colorectal cancer spreads in a predictable manner to the regional draining lymph nodes. Locoregional control is best achieved by surgery and is essential for satisfactory outcome.'
- 'Survival after 'curative resection' is related to the stage of the disease-depth of tumour penetration and/or the presence of lymph node metastases (biological markers of disseminated disease)'.
- 'The fixity of the primary tumour will determine resectability and the extent of spread and ultimate survival. Tethering or local infiltration is not necessarily a contradiction for radical surgery. Pre-operative radiotherapy should be considered for patients who are believed to be operable but have tethered rectal cancers. Fixed and irresectable tumours should be bypassed or dealt with by a proximal colostomy.
- 'In rectal cancers, stapling devices (which have facilitated low rectal anastomosis) and the modern assessment of distal colonic spread (> 1cm in 10% of patients; poorly differentiated poor prognosis tumours) have increased number of patients undergoing sphincter-saving surgery, particularly for tumours sited in the middle third of the rectum'.
- 'Mesorectal excision makes an important contribution to local control in patients undergoing resection of middle and lower third rectal tumours. There is evidence that the incidence of anastomotic leak is high after total mesorectal excision (TME); a temporary defunctioning stoma, therefore, should be considered'.
- 'In very low rectal cancers, colo-anal anastomosis is an option, but this will be influenced by body, build, size and local spread of tumour (circumferentially, distal resection margin)

and technical expertise. Randomised trials have not confirmed the value of specific techniques (e.g. 'no touch', 'high tie' or stapled versus handsewn techniques).

1.1.6 Pathology of Colorectal Cancer

A present the most widely used method of pathological staging in patients with colorectal cancer is Dukes' staging (Fielding, 1995). This was devised in 1932 to stage tumours of the rectum; however, it is now widely applied to tumours of the colon. This system is separated into 4 categories. Dukes A tumours are those that are limited to the bowel wall, Dukes B tumours are those tumours that have spread beyond the bowel wall. Tumours that have associated lymphatic spread are defined as being Dukes C tumours and those that have metastasised are defined as being Dukes D tumours (Williams et al., 1995).

Five-year survival rates of patients with Dukes' A, B and C tumours are approximately 90%, 70% and 30% respectively (Williams et al., 1995). However, Dukes staging only allows a 'crude' estimation of survival. In order to improve the prognostic value of Dukes staging, other significant pathological factors have been identified which are thought to affect outcome. These include the degree of differentiation, presence of venous invasion, character of invasive margin, peritumoural lymphocytic infiltration, number of nodes involved and the presence or absence of apical lymph node metastasis (McArdle, 1998). Although a TNM classification has been described, Dukes' classification or one of its modifications remains the most widely used method of pathological staging.

1.1.7 Outcome of Patients with Colorectal Cancer

The prognosis for patients with colorectal cancer is poor, approximately 30% of patients remaining alive after 5 years (Allum et al., 1994). This is thought to be partly attributable to the large number of patients (50%) who have locally advanced and/or metastatic disease at the time of initial presentation (McArdle et al., 1990; Allum et al., 1994; Hermanek et al., 1995). Furthermore, in patients who undergo apparently 'curative' resection, 50% die within 5 years. The poor prognosis of these patients may be due to the presence of 'occult' hepatic disease.

Finlay and McArdle (1986) observed 43 patients with colorectal cancer undergoing primary surgery. At this time, six patients were found to have overt hepatic metastases. Of the 37 remaining patients, 11 (29%) developed metastatic liver disease within 2 years. It was concluded that at the time of primary 'curative' surgery, as many as 29% of patients may have occult hepatic metastases.

1.1.8 Adjuvant Therapy for Colorectal Cancer

Due to the poor 5-year survival rates observed in patients with colorectal cancer, (despite undergoing 'curative' surgery), adjuvant treatments such as radiotherapy and chemotherapy have been introduced to treat 'occult' disease.

Adjuvant chemotherapy for colorectal cancer is widely used in patients with Dukes C colorectal cancers. It has recently been established that the use of the cytotoxic agent 5-fluorouracil in combination with the biomudulator folinic acid produces a small but significant improvement in survival in this patient group. Francini and colleagues (1994) reported a significant improvement in survival from six 5-day courses of 5-FU with high dose folinic acid. A meta-analysis (IMPACT STUDY, International Multicentre Pooled Analysis of Colorectal Cancer Trials Investigators, 1995) of three studies testing the same regime in Italy, France and Canada reported lower recurrence rates at three years and 5% better 3-year survival (83% vs. 78%). The US intergroup (1997) also reported lower rates of recurrence and a 4% improvement in survival at 3 years (75% vs. 71%). The study conducted by NSABP (Wolmark et al., 1993) of 1000 patients with colon cancer assessed the effectiveness of weekly 5-FU and folinic acid. Once again, lower recurrence rates and a moderate improvement in survival (84% vs. 77%) at three years was demonstrated.

The role of adjuvant chemotherapy in patients with Dukes B tumours is presently under debate. A meta-analysis of patients with Dukes B tumours in the IMPACT studies demonstrated a marginal improvement in survival of approximately 3% (International Multicentre Pooled Analysis of Colorectal Cancer Trials Investigators, 1995).

1.2 The Metastatic Process

A number of hypotheses have been made in order to explain the 'metastatic process'. It has been over 100 years since Paget (1889) asked the question 'what is it that determines what organ shall suffer in a case of disseminated cancer?' He observed 735 women dying of cancer of the breast and found that the site of metastatic disease was varied and could not be based on anatomical factors alone. He proposed that metastatic disease was not a random process, but that it was dependent on factors such as the environment that the individual organ offered i.e. 'the soil' and the attributes of the malignant cell i.e. 'the seed'. For metastasis to occur the 'seed' and the 'soil' had to be compatible.

Paget's theory has been reinforced with studies that have suggested that the metastatic process is not a random process, but one that is highly specialised. Fidler (1973) asked 'is the process of metastasis dependent on host or "soil" factors and/or is the process dependent on qualities unique to malignant tumour cells?'. Initially, a B16 mouse melanoma line was grown in tissue culture and then intravenously injected into syngeneic C57 BL/6 mice. Colonies of these cells that had proliferated in the lungs of the mice were then removed and the same procedure repeated to a total of 5 times. The results of the study demonstrated that the incidence of lung metastases significantly increased with each successive tumour line injected. It was concluded that as the syngeneic recipient of the tumour cells did not differ between successive cell lines, then the difference in the incidence metastases may be attributed to 'properties intrinsic to the various tumour cell lines' (Fidler, 1973).

Subsequently, Tarin and colleagues (1984) presented a study on a woman with malignant ascites from ovarian cancer that required peritoneo-venous shunting. They found that despite having a rapidly proliferating tumour that was implanting a large number of viable malignant cells into the blood stream, the lady did not proceed to develop any overt metastases elsewhere. They concluded that this study supported Pagets 'seed and soil' hypothesis as the cells that were malignant in the peritoneum did not appear to have the necessary properties to establish metastases in other organs (Tarin et al., 1984).

Weiss and colleagues (1983) assessed the metastatic behaviour of pulmonary metastases and compared them to their parent primary tumours after implanting them subcutaneously into mice. Tumours, which were used, included KHT osteosarcomas, B16 melanomas, Lewis lung and T241. The secondary KHT osteosarcomas and B16 melanoma cells gave rise to a greater number of metastases in comparison to the parent primary tumours. However, in contrast to these findings, the secondary Lewis lung and T241 tumours gave rise to fewer metastases in comparison to the primary parent cell lines. They concluded that the results of the study did not support the hypothesis that metastatic cancer cells originate from the 'progressive evolution of subpopulations of cancer cells with heritable metastatic phenotypes during tumour growth'.

From a study of 4728 autopsy reports on 47 sites of metastases, Viadana and colleagues (1978) proposed that metastases occur due to a cascade process. They proposed that the primary tumour does not act as the sole disseminating site for metastases to occur, but that key organs seed first and then cascade metastases elsewhere in the body. One of the criteria set for defining a key site was that it must be the first filter encountered by blood/lymph borne tumour cells. The frequency of metastases in specific organs was also considered in defining these sites. From the study it was proposed that solid tumours could be divided into 5 groups depending on the key sites of dissemination:

- Group 1:

The lungs are thought to be the key site for the dissemination of tumour cells. Metastases from this group of tumours appear to colonise the lung in the first instance and then disseminate to further sites. Tumours, which are included in this group, are head, neck, renal, endocrine and testicular cancers, cutaneous melanoma, and osteochondrosarcomas.

- Group 2

Tumours in this group include gastrointestinal tumours i.e. stomach, pancreas and colon, which have portal venous drainage. The key organ of dissemination in this case is the liver, which then seeds to the lungs and the entire body. Rectal adenocarcinoma appears to be an exception to this rule: tumours of the upper two thirds of the rectum appear to directly

metastasise to the liver and that those of the lower third, directly to the lungs. This is thought to occur due to differences in venous drainage between the two sites.

- Group 3:

These tumours are thought to disseminate to the lungs and the liver via independent routes. Seeding of these organs occurs independently via blood and lymphatic channels. Tumours included in this group are tumours of the upper third of the oesophagus, bladder, and female genital tract.

- Group 4:

Adenocarcinoma of the prostate is a tumour that can be classed as being in group 4 of the metastatic cascade. Metastases occur to the pelvis and vertebrae via the vertebral venous plexus. It is from this site that further metastases occur such as lung metastases.

- Group 5:

Includes breast cancer which is believed to have 3 sites of dissemination; lungs, liver and vertebrae. The lung are colonised via lymphatic channels, the liver by abdominal lymphatic vessels and the vertebrae via intercostal veins and vertebral venous plexus.

The metastatic cascade appears to explain the majority of metastasis as being due to anatomical factors. However, some tumours seem to be more particular in choosing which organs they metastasise to. Some tumours appear to bypass nearby organs and selectively colonise certain distal organs. In this case, the metastatic process may involve more specialised factors as those described in Paget's seed and soil theory.

1.3 Colorectal Liver Metastases

At the time of presentation it is thought that approximately 25% of all patients with colorectal cancer have metastatic disease (Finlay & McArdle, 1986).

Metastases to the liver are the commonest means of spread from colorectal cancer (Welch & Donaldson, 1979), followed consecutively by the lungs, adrenals, ovaries and bone (Sugarbaker, 1980). As many as 60% of patients with colorectal cancer will develop liver metastases (Welch & Donaldson, 1979), the liver being the sole site of recurrence in approximately 20% of patients (Weiss et al., 1986). Furthermore, hepatic metastases are found at autopsy in 80% of cases of disseminated colorectal cancer (Viadana et al., 1978).

The natural history of patients with colorectal liver metastases has been assessed by a number of investigators (Jaffe et al., 1968; Bengmark & Hafstrom, 1969; Oxley & Ellis, 1969; Wood et al., 1976; Bengtsson et al., 1981; Stangl et al., 1994)

STUDY	NO. OF PATIENTS	SURVIVAL
Jaffe et al (1968)	177	4.9 months*
Bengmark and Hafstrom (1969)	173	5.7 months**
Oxley and Ellis (1969)	112	<6 months
Wood et al (1976)	113	6.6 months**
Bengtsson et al (1981)	172	5.6 months**
Stangle et al (1994)	484	7.5 months*

*= median **=mean

From the table above, it is evident that prognosis for patients with untreated colorectal liver metastases is poor. However, it must be noted that these studies are limited and dated, as the majority of patients with this condition now receive some method of treatment, be it 'curative' or palliative.

It has been suggested that overall survival of patients with this condition is significantly affected by degree of hepatic replacement (Jaffe et al., 1968; Wood et al., 1976). Of particular

interest is the retrospective study conducted by Wood and colleagues (1976) at Glasgow Royal Infirmary. They assessed survival of patients with colorectal liver metastases in relation to the degree of hepatic replacement:

DISTRIBUTION OF LIVER METASTASES	NUMBER OF PATIENTS	MEAN SURVIVAL (MONTHS)	1-YEAR SURVIVAL (%)	3-YEAR SURVIVAL (%)
Widespread Bi-lobar Disease	87	3.1	6	
Widespread Unilobar Disease	11	10.6	27	10
'Solitary Metastases'	15	16.7	60	13

They concluded that survival of patients with colorectal liver metastases appears to be related to the extent of hepatic involvement of the tumour. These findings appear to be in agreement to a similar study conducted by Jaffe and co-workers (1968). They reported that the median survival times of patients with bi-lobar, uni-lobar and solitary colorectal liver metastases were 2.4 months, 3.1 months and 4.5 months respectively. Histological confirmation of colorectal liver metastases was only obtained in a few of the above-mentioned studies, and this may in part account for the differences in overall survival.

1.4 Treatment Options for Colorectal Liver Metastases

There are a number of treatment options that are available for patients with colorectal liver metastases. Treatments that are commonly offered to this patient group will be discussed:

1.4.1 Hepatic Resection

The past three decades have seen an increase in attempts to treat liver metastases by surgical resection. The post-operative mortality rate associated with this type of surgery is between approximately 2-5% (Wilson & Adson, 1976; Saenz et al., 1989; Scheele et al., 1991; Nordlinger et al., 1996). Reported 5-year survival rates have been encouraging (Wilson & Adson, 1976; Hughes et al., 1989; Scheele et al., 1991; Nordlinger et al., 1996; Fong et al., 1999):

STUDY	5-YEAR SURVIVAL RATES
Wilson and Adson (1976)	28%
Hughes et al (1989)	32%
Scheele et al (1991)	39%*
Nordlinger et al (1996)	28%
Fong et al (1999)	37%

* Actuarial 5-year survival

Nordlinger (1996) suggested that patients, who were older, with large metastases and undergoing hepatic resection in addition to resection of the primary tumour, were more at risk from post-operative death.

Many studies have attempted to identify patients who may have a 'less favourable' long-term prognosis after liver resection. Hughes and colleagues (1989) evaluated 800 patients who had undergone resection of colorectal liver metastases. Factors that were found to be detrimental to overall outcome were the presence of four or more metastatic lesions within the liver, a disease-free interval between the primary resection and liver resection of less than 1-year, size of pathological margin in liver resection, and the presence of associated lymphatic spread

from the primary tumour. Strong contra-indications to surgery were if radical removal of tumour was unable to be performed, and if there was tumour spread in the nearby hepatic and/or coeliac lymph nodes. Scheele and co-workers (1991) identified similar factors in their study of 266 patients receiving hepatic resection of colorectal liver metastases. The study by Nordinger and colleagues (1996) also identified carcinoembryonic antigen (CEA) levels of >30 ng/ml and age ≥ to 60 years as being 'less favourable' pre-operative indicators to hepatic resection. Fong (1999) also identified hepatic metastases >5cm in size and CEA levels > 200ng/ml as being poor prognostic indicators.

In conclusion, hepatic resection is the only potentially 'curative' treatment option in patients with colorectal liver metastases. Furthermore, this type of surgery is associated with post-operative mortality rates of less than 5% and 5-year survival rates of 25-40%.

However, as has been previously indicated only a minority of patients with this condition are suitable for such treatment, and as a result many patients receive palliative chemotherapy.

1.4.2 Chemotherapy

Chemotherapy for metastatic colorectal cancer has been used for palliation for many years. During this time one drug, 5-Fluorouracil (5-FU) has remained the single most effective antineoplastic agent in the treatment of colorectal adenocarcinomas (Schilsky, 1995).

1.4.2.1 5-Fluorouracil

Charles Heidelberger developed a group of anti-tumour agents known as the 5-Fluoropyrimidines (Heidelberger et al., 1957). The simplest derivative of this group is 5-FU. This is an analogue of the naturally occurring pyrimidine Uracil. The difference between Uracil and 5-FU is that in 5-FU, a fluorine atom is replaced at the carbon 5 position of the pyrimidine ring instead of hydrogen (Grem, 1996).

The main actions of 5-FU on the cell are:

- In vivo conversion of 5-FU to fluoro-2-deoxyuridine 5' phosphate (FdUMP) causing inhibition of the enzyme thymidylate synthetase and deoxyribonucleic acid (DNA) synthesis (Kufe and Major, 1981).

- Conversion of 5-FU to fluorouridine triphosphate (FUTP) which is incorporated into riboxynucleic acid (RNA) and therefore disrupts RNA function (Kufe and Major, 1981).

5-FU becomes cytotoxic only once it is metabolised in actively proliferating cells to form nucleotides (Diasio & Harris, 1989). It enters the cell via a carrier-mediated transport mechanism. It has a rapid volume of distribution and elimination. Approximately 80% of 5-FU is eliminated via catabolism and 20% via urinary excretion (Clingan, 1996). Pharmacokinetics studies of 5-FU have reported a primary half-life of approximately 5-20 minutes (MacMillan et al., 1978; Heggie et al., 1987). 5-fluorouracil is most cytotoxic during the S-phase of the cell cycle (Clingan, 1996).

1.4.2.2 Oral 5-Fluorouracil

5-FU may be administered orally. A randomised, double-blind study of one hundred patients with biopsy proven metastatic colorectal cancer compared oral administration of 5-FU to intravenous (IV) administration. Number of patients and sites of metastatic disease were as follows:

ROUTE OF ADMINISTRATION	ORAL 5-FU	IV 5-FU
Number of patients	47	53
Hepatic Metastases	23	22
Lung Metastases	12	17
Other Sites	12	14

Results of the study demonstrated an objective response rate of 26% in patients receiving IV administration in comparison to 19% of patients treated orally. Mean duration of response for patients receiving oral 5-FU was 11 weeks compared to 20 weeks for patients receiving intravenous 5-FU. Further analysis comparing the two modes of administration in patients with hepatic metastases only, displayed no statistical difference in response. However, mean duration of response was once again greater in patents receiving intravenous 5-FU (22 weeks compared to 10 weeks). Furthermore, it was observed that there was a wide variation in blood levels of 5-FU with oral administration. Due to this variability in absorption and significantly lower duration of response in patients receiving oral 5-FU, it was concluded that intravenous administration appeared to be a more effective mode of administration (Hahn et al., 1975). Due to these large variations in bio-availability (0-80%), 5-FU is rarely administered orally (Diasio & Harris, 1989).

1.4.2.3 Systemic 5-Fluorouracil

Single agent 5-FU may be administered by bolus injection. There have been many studies of bolus 5-FU in patients with colorectal liver metastases. Kirk Martin and colleagues (1990) assessed the effectiveness of bolus 5-FU in 33 patients with colorectal liver metastases. An overall response rate of approximately 20% was reported. Furthermore, duration of response within the liver was 6 months, time to tumour progression at all sites was 5 months and overall survival 10.5 months. A more recent study reported a response rate of 18% in 153 patients with advanced colorectal cancer receiving bolus 5-FU (Hansen et al., 1996). Median time to disease progression was approximately 5 months and overall median survival of 10 months.

Toxicities of bolus 5-FU tend to be dependent on dose. Dose-limiting toxicities include leucopoenia and oral mucositis. Other common toxicities include diarrhoea, phlebitis and mild conjunctivitis. The risk of toxic death from this regime is approximately 1% in patients with dihydropyrimidine dehydrogenase deficiency (Conroy, 2000). The effect of bolus 5-FU in patients with advanced cancer may, therefore be said to be poor, in relation to response rates and survival.

In an attempt to improve the therapeutic index of this drug, continuous intravenous infusion of this drug has been assessed (Lokich et al., 1989). Lokich and co-workers reported a response rate of 30% and a median survival of 10 months in 87 patients with advanced colorectal cancer receiving continuous infusion of single agent 5-FU (Lokich et al., 1989). Blijham and co-workers evaluated 110 patients with measurable advanced or metastatic colorectal cancer receiving 5-FU infusions. A response rate of 11% was reported and a median survival of approximately 9 months (Blijham et al., 1996). Hansen and colleagues (1996) reported a response rate of 28% in 159 patients with advanced colorectal cancer receiving infusional 5-FU. Median time to disease progression was 6 months with an overall median survival time of 13 months. The most common toxicity associated with continuous infusional 5-FU is the occurrence of plantar-palmar syndrome, where the feet and hands may become red, inflamed, tender and 'hacks' in the skin may form. Other toxicities such as mucositis, diarrhoea and neutropenia do occur, but to a lesser degree than in bolus 5-FU regimes (Conroy, 2000).

Single agent 5-FU appears to yield relatively low response rates with little impact on overall survival. Attempts have been made to increase the effectiveness of this drug by combining it with Folinic acid. Modulation of 5-FU with folinic acid is thought to be based on the premise that inhibition of thymidylate synthase may be increased in conditions where there is an excess of the folate co-substrate for thymidylate synthase, 5,10-methylene-tetrahydrofolate. The combination of thymidylate synthase, 5,10-methylene-tetrahydrofolate and an active metabolite of 5-FU (5-dUMP) appears to lead to the formation of a covalent ternary complex that has no enzyme activity and appears to gradually dissociate (Gebbia et al., 1993).

During the late 1980's several phase III trials were conducted to assess whether biomodulation of 5-FU with folinic acid was superior to single agent 5-FU (Petrelli et al., 1987; Erlichman et al., 1988; Nobile et al., 1988; Di Constanzo et al., 1989; Petrelli et al., 1989; Poon et al., 1989; Valone et al., 1989; Doroshaw et al., 1990; Labianca et al., 1991). Response rates varied from 15% to 48% for the combined regime and a significant increase in response rates compared to single agent 5-FU in all but two trials. However, with regard to overall survival only one study demonstrated any significant improvement (Erlichman et al., 1988).

In 1992, a meta-analysis of 9 clinical trials that compared single agent bolus 5-FU to 5-FU and folinic Acid was carried out by the Advanced Colorectal Cancer Meta-Analysis Project (1992). A total of 1318 patients were included in the study. Five hundred and seventy-eight patients received 5-FU and 803 patients 5-FU and folinic acid. Results of the meta-analysis reported a significantly higher response rate for 5-FU and folinic acid (23%) compared to 5-FU alone (11%). However, median survival between the 2 groups did not appear to significantly differ (11 months for 5-FU and 11.5 months for 5-FU and Folinic acid). Furthermore, when trials that had allowed cross-over of patients between the 2 arms of the study were excluded from the analysis, survival difference between the two groups remained insignificant.

Randomised clinical trials of folinic acid and 5-FU versus 5-FU alone

STUDY	N =	SCHEDULE	%COMPLETE OR PARTIAL RESPONSE	SURVIVAL
Nobile et al (1988)	82	5-FU 600mg/m^2 + FA 500mg/m^2 vs. 5-fu 600mg/m^2 weekly	16% vs. 5% (p=0.05)	No significant difference
Petrelli et al (1989)	318	5-FU 600mg/m^2+ FA 500 or 25 mg/m^2, 6 out of 8 weeks versus 5-FU 500mg/m^2 x 5 every 4 weeks	30% vs. 19% vs. 12% (p= < 0.01)	55 vs. 45 vs. 46 weeks (p=0.08)
Petrelli et al (1987)	41	5-FU 600mg/m^2 + FA 500mg/m^2 6 out of 8 weeks versus 5-FU 450mg/m^2 x 5	48% vs. 11% (p= 0.0009)	No significant difference
Valone et al (1989)	153	5-FU 400mg/m^2 x 5 + FA 200mg/m^2 x 5 every 4 weeks versus 5-FU 480mg/m^2 followed by 600mg/m^2/week	19% vs. 17% (p=0.4)	24 vs. 20 weeks (p=0.4)
Di Costanzo et al (1989)	181	5-FU 400mg/m^2 x 5 + FA 200mg/m^2 every 4 weeks versus 5FU 540mg/m^2 x 5	15% vs. 16%	25 vs. 21weeks
Labianca et al (1991)	182	5-FU 400mg/m^2 x 5 +FA 200mg/m^2 X 5 every 4 weeks versus 5-FU 400mg/m^2 x 5 every 4 weeks	21% vs. 10% (p = 0.046)	46 vs. 44 weeks
Poon et al (1989)	208	5-FU 370mg/m^2 x 5 + FA 20 or 200mg/m^2 x 5 vs. 5-FU 500mg/m^2 x 5	43% vs. 26% vs. 10% (p=0.001)	53 vs. 52 vs. 34 weeks (p=0.05)
Doroshaw et al (1990)	74	5-FU 370mg/m^2 x 5 + FA 500mg/m^2 x5 every 4 weeks versus 5-FU 370mg/m^2 x 5	44% vs. 13% (p=0.0019)	63 vs. 55 weeks (p=0.25)
Erlichman et al (1988)	124	5-FU 370mg/m^2 x5 + FA 200mg/m^2 x 5 versus 5-FU 370mg/m^2 x 5	33% vs. 7% (p < 0.0005)	54 vs. 41 weeks (p=0.05)

At present there remains controversy as to whether high dose or low dose folinic acid is required for optimal modulation of 5-FU. The meta-analysis (Advanced Colorectal Cancer Meta-Analysis Project, 1992) failed to identify optimal doses and schedules of folinic acid used in combination with 5-FU. At present, the most common regime used to treat advanced colorectal cancer in the USA is the 'Mayo' regime (FA 20mg/m^2/day IV + 5-FU 425mg/m^2/day with both drugs administered as an intravenous bolus for 5 consecutive days every 4-5 weeks).

In order to optimise the effect of 5-FU in combination with folinic acid, a different approach of administration has been assessed by De Gramont and colleagues (De Gramont et al., 1997). It was hypothesised that this regime would allow for the administration of higher doses of 5-FU and the prolongation of tumour cell exposure to the drug. This regime consists of high dose folinic acid (400mg/m^2) administered over two hours, followed by 5-FU intravenous bolus (400mg/m^2) and continuous 22-hour infusion of 5-FU for two consecutive days every two weeks. De Gramont reported a response rate of 33% when this regime was used in patients with advanced colorectal cancer, with a median progression-free survival of 28 weeks. It was concluded that bi-monthly high dose leucovorin, 5-FU bolus plus continuous infusion was effective in patients with advanced colorectal cancer. However, there did not appear to be any significant increase in overall survival with this regime.

In patients with advanced colorectal cancer who become resistant to 5-FU, some 'second line' chemotherapy drugs may be used. Those commonly used with be briefly discussed. Ralitrexed is a direct and specific inhibitor of thymidine synthase, the enzyme responsible for the synthesis of thymidylate, a nucleoside essential for DNA replication and repair. Since DNA, is the only cellular target for ralitrexed, it is thought that this may result in a reduced frequency of adverse effects. A study by Cunningham and colleagues (1996), demonstrated that ralitrexed was as effective as standard 5-FU and folinic acid regimes in terms of objective response rates and produced a median survival range of approximately 9.7 – 11.2 months, in comparison to 10.2 to 12.7 months with 5-FU and folinic acid regimes. Common toxicities with this regime include asthenia, diarrhoea, vomiting and transient increases in liver function tests. Research into the efficacy and safety of ralitrexed continues.

Irinotecan is commonly used in the second line treatment of metastatic colorectal cancer. It inhibits the enzyme topoisomerase 1, which is responsible for 'relaxing' coiled DNA prior to transcription or replication. This in turns, forms a stable complex with the DNA resulting in cell death during replication. Response rates of 12-24% have been reported in patients who have 5-FU resistant tumours (Conroy, 2000). Dose limiting toxicities include diarrhoea and neutropenia. Median duration of response is approximately 6 months (Ford & Cunningham, 1999; Conroy, 2000).

In summary, it would appear that the majority of patients still fail to benefit from systemic chemotherapy. Most cytotoxic drugs have a steep dose response curve. Pharmacokinetics suggest that if delivery of the drug to the tumour can be increased, then response rates may also be increased (Frei, III. & Canellos, 1980). Attempts to do this with systemic regimes have proven to be unsuccessful due to toxicity. In a bid to increase tumour exposure to cytotoxic drugs and decrease systemic toxicity some practitioners have turned to the use of regional administration of chemotherapy.

1.5 Regional Chemotherapy

Regional chemotherapy was first attempted by Clarkson and his colleagues in 1961 (Clarkson et al., 1962) and is based on the concept of targeted drug delivery. According to Widder and colleagues (1979) there are three levels of drug targeting:

- Selective delivery of the drug to the tumour bearing organ.
- Drug delivery direct to the tumour and not normal tissue within the target organ.
- The preferential uptake of cytotoxic drugs by the tumour cells within that organ.

Regional chemotherapy provides first level drug targeting. Hepatic metastases greater than 1cm in size are known to predominantly derive their blood supply from the hepatic artery (Ridge et al., 1987). Cytotoxic drugs that have been used for this type of regional administration into the liver include 5-fluoro-2'-deoxyuridine (FUDR) and 5-FU (Ensminger et al., 1978; Ensminger & Gyves, 1983). Approximately 94-99% of FUDR and 19-51% of 5-FU is thought to be extracted by the liver on first pass with hepatic arterial drug infusion (Ensminger et al. 1978). Studies conducted in the 1960's on the intra-arterial administration of FUDR or 5-FU, reported response rates of 50% or more (Clarkson et al., 1962) and prolonged survival in comparison to historical controls.

The first randomised controlled study comparing of intra-arterial chemotherapy (21 day infusion of 5-FU) to intravenous regimes (bolus 5-FU) was carried out by Grage and colleagues (1979). Sixty-one patients were entered into the study. There appeared to be no significant differences between the two regimes in terms of response rates (34% vs. 23%), duration of response, time to disease progression and overall survival (13.5 months vs. 15.4 months). Furthermore, there was a greater incidence of nausea and vomiting, diarrhoea, femoral-arterial thrombosis and bleeding and infection at the catheter site associated with intra-arterial infusion. Due to the toxicity observed, enthusiasm for intra-arterial chemotherapy of colorectal liver metastases at that time waned.

Due to the continuing poor response and survival rates with systemic chemotherapy, the development of an implantable refillable pump (Infusaid) revived interest in regional chemotherapy to the liver. FUDR was the drug of choice when using this pump for two reasons:

- The first pass extraction of FUDR is between 94-99% with minimal systemic toxicity, which appears to be suitable for patients with disease confined to the liver.
- The infusaid pump has a limited reservoir and a low infusion rate, therefore the cytotoxic drug has to be highly concentrated.

Early studies of regional administration with the infusaid pump reported a significant increase in tumour FUDR concentration, response rates and prolonged survival in comparison to historical controls. Balch and colleagues (1983) reported response rates in excess of 80% in patients receiving intra-arterial FUDR and prolongation of median survival in comparison to historical controls (26 months vs. 8 months). Niederhuber (1984) observed that of 50 patients receiving intra-arterial FUDR, 83% demonstrated a significant decrease in tumour size. However, associated with such response rates was significant local toxicity in the form of gastritis (17%-56), chemical hepatitis (32%-50%) and chemical cholecystitis (25%).

Results of the UK Pump study in 1994 (Allen-Mersh et al., 1994) reported that survival and quality of life was significantly better in patients receiving intra-arterial chemotherapy, compared with patients receiving symptom palliation only. Of the 59 patients receiving intra-arterial FUDR (0.2mg/kg/24 hours for 2 weeks with a 2 week respite) median survival was 13.5 months in comparison to 7.5 months for the 49 control patients. Furthermore, patients receiving regional chemotherapy had a superior quality of life.

In the 1980's, several randomised trials were conducted which compared intra-arterial administration of FUDR to either intravenous FUDR or 5-FU (Chang et al., 1987; Kemeny et al., 1987; Hohn et al., 1989; Kirk Martin et al., 1990; Rougier et al., 1992). In each study, there was a significantly higher response rate in the intra-arterial group.

STUDY	PATIENTS (IV)	PATIENTS (IA)	RESPONSE (IV)	RESPONSE (IA)
Chang et al (1987)	29	21	29%	62%
Kemeny et al (1987)	51	48	20%	50%
Hohn et al (1989)	65	50	10%	42%
Kirk-Martin et al (1990)	33	36	21%	48%
Rougier et al (1992)	41	81	9%	43%

However, the significant increase in response rates associated with intra-arterial administration of FUDR did not translate into a survival advantage.

In some studies, it was suggested that the insignificant differences in survival between intra-arterial and intravenous administration was due to factors such as small sample sizes or patients crossing from intravenous to intra-arterial arms of studies on development of disease progression. However, despite the absence of conclusive data on the survival advantage of intra-arterial chemotherapy, several thousands of infusaid pumps were inserted especially in the USA.

A recent meta-analysis (Meta-Analysis Group In Cancer, 1996) of 5 studies comparing intra-arterial chemotherapy to conventional systemic regimes confirmed that intra-arterial chemotherapy can achieve significantly higher response rates than systemic regimes (41% vs 14%) however, once again, the impact on survival remains unclear.

An important finding from these trials comparing conventional systemic regimes to intra-arterial FUDR administration was the differences in first sites of disease progression. Chang and colleagues (1987) observed that of the 13 patients who responded to intra-arterial treatment, 6 patients had the first site of relapse in extra-hepatic sites compared to 1 in 5 patients responding to intravenous therapy. Kemeny and co-workers (Kemeny et al., 1987) also noted that extra-hepatic disease progression developed in 56% of patients receiving intra-arterial therapy compared to 37% of patients receiving intravenous treatment. Disease

progression at extra-hepatic sites was reported by Rougier (1992) in 54% of patients receiving intra-arterial chemotherapy and 34% of patients receiving intravenous therapy.

Hence, it appears that with intra-arterial FUDR, the development of extra-hepatic disease progression appears to be more prevalent in comparison to systemic regimes. This may be due to the liver becoming 'saturated' with FUDR on intra-arterial administration due to its high first pass extraction rate (Ensminger et al. 1978), resulting in insufficient systemic levels of the drug. By concentrating on achieving high drug levels within the liver, 'occult' disseminated disease is ignored, which results in alteration of the natural disease process without any significant increase in survival.

Following results of studies using the regional administration of FUDR, further studies were directed at not only obtaining higher local tumour concentrations, but also adequate systemic levels of the drug. This was thought to be achieved by two methods:

- The combination of high dose regional and systemic chemotherapy
- Allowing high doses of regional chemotherapy to 'spill over' into the systemic circulation, at levels that are likely to affect 'occult' disseminated disease.

The first methods were explored by Safi and colleagues (1989). They compared 23 patients receiving regional FUDR alone to 21 patients receiving a combination of regional and systemic FUDR. The incidence of extra-hepatic recurrence rated decreased from 61% in patients receiving regional chemotherapy alone to 33% in patients receiving the combination of both regional and systemic regimes. However, there was no significant survival advantage observed between the two regimes.

The team at Glasgow Royal explored the latter approach. As 5-FU is known has a first pass extraction rate of 19-54% (Ensminger et al., 1978), they hypothesised that intra-arterial administration of this drug would not only produce high intra-hepatic levels but also 'adequate' systemic levels to treat any occult extra-hepatic disease. This would be due to

unmetabolised 5-FU 'spilling' over into the systemic circulation after first pass hepatic extraction.

Initially, Goldberg and co-workers (1990), carried out a pharmacokinetic study which demonstrated that 24 hour regional infusions of 5-FU conferred significant pharmacological advantage compared to intravenous or intra-arterial bolus infusions. A later study carried out Anderson and colleagues (1992), (based on previous reports of increased response rates with combination use of systemic 5-FU and folinic acid), compared pharmacokinetic profiles of intravenous and intra-arterial folinic acid. They reported a small but significant advantage with intra-arterial administration. Associated with this, however, was a high incidence of catheter thrombosis, which appeared to outweigh the pharmacokinetic advantages observed with intra-arterial folinic acid administration.

Anderson and colleagues (1992) subsequently conducted a phase I study of weekly 24-hour intra-arterial infusion of 5-FU (0.6g-2.0g/m^2/week) and systemic folinic acid (400mg/m^2). At the highest dose of 5FU, approximately 50% of patients developed WHO grade3/4 diarrhoea and for vomiting. It was concluded that the recommended dose for phase II studies should be 1.5gm/m^2 given intra-arterially over 22h.

The phase II study was conducted by Warren and colleagues (1994). Treatments were administered for 6 weeks, followed by a 2-week respite period. A total of 31 patients were entered into the study; 48% of patients responded to treatment with a further 39% of patients demonstrating stable disease. Median duration of treatment was 6 months and predicted median survival was 19 months. Compared to the toxicity observed with previous regional FUDR regimes, the toxicity associated with this regime was minimal.

Criticisms of previous studies of regional chemotherapy include the fact that different dosing schedules have been used. In order to achieve comparable intra-arterial and systemic regimes for further studies, a pharmacokinetic and phase I study was conducted by Kerr and co-workers (1995). The regime was based on the conventional systemic DeGramont regime in which folinic acid (400mg/m^2) is administered via intravenous infusion over 2 hours, followed

by a loading dose of 5-FU (400mg/m^2) over 15 minutes, followed by a 22 hour infusion of 5-FU (600mg/m^2). The same regime was repeated the next day. In the intra-arterial regimen the dose of infusional 5FU ranged from 0.8-1.84g/ms. Patients received intra-arterial chemotherapy over 48 hours for 2 days every fortnight. In this study significant toxicity occurred at 1.84g/m^2; hence for subsequent trials a dose of 1.6g/m^2 was used. Pharmacokinetic results of the study demonstrated that systemic levels of 5-FU were similar to those of the conventional De Gramont regime (1988).

A later study conducted by Howell and colleagues (1997) compared this 2-weekly regional 5-FU infusion with intravenous folinic in the treatment of patients with colorectal liver metastases. An overall response rate of 46% was observed in the 40 patients evaluated. Predicted median survival was 19 months. Sites of disease progression were the liver alone in 55% of patients, liver and lung in 16% and 29% at other sites. Once again, it was concluded that tumour response rates were comparable to those observed with intra-arterial FUDR. As extra-hepatic disease was the first site of disease progression in only 29% of patients, it was also suggested that therapeutic systemic levels were achieved in this study. At present the Medical Research Council is conducting a prospective randomised trial comparing regional to systemic chemotherapy, using the above regimen.

1.6 Evaluation of Response to Chemotherapy

From the previous literature (section 1.3), it is evident that a significant number of patients with colorectal cancer progress to advanced disease. In approximately 20% of patients, this will be in the form of liver metastases with no associated extra-hepatic spread (Weiss et al., 1986). A number of patients with this pattern of metastatic disease will receive palliative chemotherapy, either in the form of conventional systemic regimes or regionally. In order to assess the effectiveness of such regimes, response is commonly assessed by clinical examination, radiological scanning and the use of 'tumour markers'.

1.6.1 Clinical Evaluation

Evidence of increasing hepatomegaly, weight loss, fever and pain may indicate progressive metastatic disease in the liver. Other signs may include lower ankle oedema and the development of abdominal ascites. Some biochemical abnormalities that may be present include elevated alkaline phosphatase levels and abnormal liver function tests (Finlayson & Bouchier, 1995).

1.6.2 Computed Tomography Scanning

Few studies have been carried out which have evaluated the role of CT scanning in measurement of tumour response to chemotherapy. Conventionally response has been assessed using standard WHO criteria (Miller et al., 1981). More recently Dworkin and colleagues (1995) confirmed that tumour volume, rather than percentage hepatic replacement should be used to measure response to chemotherapy in patients with colorectal liver metastases.

1.6.3 Tumour Markers

The ability of CEA (a serum glycoprotein), CA-195 and CA-242 were assessed in patients with colorectal liver metastases receiving 5FU-based chemotherapy. The patients either

received 5-FU and folinic acid or 5-FU and interferon. When the tumour markers were compared to CT scan results, CEA appeared to correlate best with the findings (Ward et al., 1993).

The efficacy of intra-arterial (IA) chemotherapy was assessed by measuring CEA doubling time (CEA-DT) as a marker of response to treatment (Ichikawa et al. 1995). Ninety-two patients colorectal liver metastases were treated with hepatic arterial 5FU-based chemotherapy. All patients had a CEA level >10 ng/ml. When cumulative survival was assessed according to CEA-DT, patients with a CEA-DT <40ng /ml had a significant increase in survival if they were receiving injection rather than infusion chemotherapy (6 months median survival vs 2.4 months median survival time).

Acute-phase proteins have also been assessed in predicting response to chemotherapy. Simpson and colleagues (1995) carried out a study in 24 patients with colorectal cancer receiving recombinant interleukin-2, 5-FU and folinic acid. The acute-phase proteins included C-reactive protein, retinol binding protein, $\alpha 1$ anti-trypsin, transferrin and albumin. Eight percent of patients had a complete response and 21% of patients had a partial response. Of the patients who responded to treatment, all had a serum albumin level greater than 37g/l and a C-reactive protein value of less than or equal to 10 mg/l.

1.7 The Systemic Inflammatory Response and Cancer

1.7.1 Overview of the Acute-Phase Response

Cuthbertson first described the acute-phase response in the 1930's (Cuthbertson, 1979) when he carried out a study to investigate why fractures of the lower third of the tibia were slow to heal. It may be defined as being a 'concerted series of diverse systemic and local events that accompany inflammation resulting from tissue damage' (Banks et al., 1993). The response is termed as 'acute' as changes normally occur within hours or days of the initial insult. Characteristically, the acute-phase response is related to changes in the synthesis of certain hepatic proteins some of which are not generally produced in the healthy state (Dinarello & Wolff, 1995). These proteins include haptoglobin, specific protease inhibitors, complement components, ceruloplasmin, C-reactive protein and fibrinogen. These proteins produced by the liver have been termed as being acute-phase proteins.

Circulating concentrations of pre-albumin, albumin, retinol binding protein and transferrin, decrease with the acute-phase response and are sometimes termed as being 'negative acute-phase proteins' (Banks et al, 1993). Positive acute phase proteins are defined as being 'plasma proteins which increase in concentration by 25% or more in the first 7 days following tissue damage' (Thompson et al., 1992). The most important acute-phase proteins in the clinical setting appear to be fibrinogen and C-reactive protein (Dinarello & Wolff, 1995).

C-reactive protein is a non-glycosylated protein that consists of 5 identical 21k-Da non-covalently bound globular subunits (Banks et al., 1993). The exact role of C-reactive protein is unclear. It is thought to act as a inflammatory mediator in phosphorylcholine binding, complement activation and opsonization (Banks et al., 1993). It has also been suggested that it acts as a scavenger of DNA and is involved in the regulation of T- and B-cell interactions (Banks et al., 1993). Clinically, C-reactive protein appears to be the most useful acute-phase protein to assess the magnitude of the inflammatory response due to its specificity and ease of reproducibility in hospital laboratories (Thompson et al., 1992). Within 6-10 hours of the initial insult, C-reactive protein is known to rapidly increase and peak within 48 hours (Banks

et al., 1993). Due to its relatively short half-life, it is also known to quickly decrease as the magnitude of the inflammatory response decreases (Banks et al., 1993).

The synthesis of acute-phase proteins within the liver during inflammation is thought to be attributed to the actions of cytokines (Banks et al., 1993) that have been defined as being 'hormone like peptides that regulate numerous cellular responses' (Tramont & Hoover, 1995). The cytokines interleukin-1, interleukin-6 and tumour necrosis factor are termed as being 'pro-inflammatory cytokines' as they act as mediators during inflammation. Cells that synthesise these cytokines include phagocytes, leukocytes, T and B lymphocytes, mast cells, fibroblasts and endothelial cells (Grimble, 1996). There is also evidence that these cytokines initiate a number of metabolic effects that occur within the host during the acute-phase response (Tramont & Hoover, 1995).

One of the most obvious clinical effects of the acute-phase response is fever, which is thought to be caused by increased synthesis of prostaglandins near or in the hypothalamus (Tramont & Hoover, 1995). Prolonged weight loss may also result. Studies have reported an increase in resting energy expenditure in cancer patients with an inflammatory response (Falconer et al. 1994; Staal-van den Brekel et al. 1995). Anorexia has also been linked with the acute-phase response; however its exact mechanism is at present unknown. It may be induced by the action of cytokines (Grimble, 1996).

The role of leptin (a cytokine which has been reported to regulate appetite and body weight in mammals) in the inflammatory response has been recently been examined. Studies conducted in hamsters (Grunfield et al., 1996), mice (Sarraf et al., 1997) and in humans (Wallace et al., 2000) have reported an increase in circulating leptin concentrations as part the acute-phase response.

Increased gluconeogenesis and abnormal tolerance of glucose and lipid metabolism may be also be present during the acute-phase response (Dinarello & Wolff, 1995). Other metabolic effects of the acute-phase response include leucocytosis with an associated increase in the number of circulating immature neutrophils, thyroid dysfunction and

hypergammaglobulinaemia (Dinarello & Wolff, 1995). Changes in body composition such as muscle proteolysis are also known to occur (see section 1.8).

1.7.2 The Inflammatory Response and Cancer

It has been known for some time that a proportion of patients with cancer have evidence of an acute-phase response (Milano et al., 1978; Fearon et al., 1991; Falconer et al., 1994; McMillan et al., 1994; McMillan et al., 1995; Staal-van den Brekel et al., 1995; Scott et al., 1996). More recently, it has been hypothesised that the metabolic effects of the acute-phase response in patients with cancer may have an associated increase in resting energy expenditure which may promote weight loss in this patient group. In the context of patients with liver metastases, studies of host metabolism and the acute-phase response are limited.

Macfie and colleagues (1982) carried out a study to assess whether the presence of gastrointestinal malignancy had any effect on resting energy expenditure. Three groups of patients were included in the study; a control group, patients with local malignancy and patients with metastatic malignancy. Seventeen out of 19 patients with metastatic disease had liver metastases. All measurements were carried out pre-operatively. Resting energy expenditure was measured using indirect calorimetry and body cell mass by measurement of total body potassium.

The results of the study demonstrated that the mean total body potassium was significantly lower and the mean resting energy expenditure significantly higher in patients with disseminated disease in compared with controls. There was no statistical difference between patients with local disease and those with disseminated disease. From the study it was concluded that patients with malignancy, especially those with metastatic disease may have an increase in energy demand. However, it was also stated that this increase in energy demand is not large enough to be incorporated into planning nutritional support, but may over a period of time accumulate and become a significant factor in this patient group (Macfie et al., 1982).

Hansell and colleagues (1986a) carried out a study to investigate the relationship between resting energy expenditure and weight loss in patients with cancer. The study group consisted of 98 patients had malignant disease (of which 16 patients had liver metastases) and 38 patients had benign disease. Significant weight loss was defined as having lost more than 10% of pre-illness weight. Resting energy expenditure was measured using indirect calorimetry and lean body mass was measured by total body water measurements with tritiated saline. The results of the study demonstrated that there was a significant correlation between resting energy expenditure and body weight and lean body mass in all patient groups. No significant differences in resting energy expenditure in patients with and without liver metastases were observed (Hansell et al., 1986a).

Hansell and colleagues (1986b) conducted another study to investigate the effect that various tumour types had on resting energy expenditure. Patients with colorectal cancer, gastric cancer and non-small cell lung cancer were included in the study. A total of 20 patients had liver metastases from these groups. Resting energy expenditure was measured via indirect calorimetry and once again the results of the study demonstrated that there was no significant differences in resting energy expenditure in patients with liver metastases compared to those without (Hansell et al., 1986b).

Luketich and colleagues (1990) measured resting energy expenditure via indirect calorimetry both pre-operatively and post-operatively (5th post-operative day) in patients with colorectal, upper gastrointestinal, pancreaticobiliary, gynaecological, lung and 'miscellaneous' cancers. Of this group 11 patients had liver metastases. The aim of their study was to assess whether metabolic abnormalities in patients with cancer were associated directly with tumour metabolism, body cell mass or both. On analysis of the results each patients acted as his or her own control based on the Harris-Benedict resting energy expenditure equation. Fifteen percent of patients were defined as being hypometabolic, 51% normometabolic and 34% hypermetabolic. Patients with liver metastases were equally distributed throughout the three groups. The results demonstrated that patients who had a curative resection appeared to either remain normometabolic or become normometabolic after surgery. The converse of this occurred in patients who had a palliative resection as they either remained hypermetabolic or

their resting energy expenditure significantly increased. It was concluded that the tumour itself may be responsible for abnormalities in metabolism as the results of the study demonstrated that removal of the primary tumour appeared to bring about a normalisation of energy expenditure (Luketich et al., 1990).

Fredrix and colleagues (1991) measured resting energy expenditure in patients who had recently been diagnosed as having gastric or colorectal cancer. The control groups used in this study included 'healthy' subjects and patients with non-malignant disease of gastrointestinal origin. Resting energy expenditure was measured by indirect calorimetry. Fat free mass was calculated via bioelectrical impedance analysis. Nutritional status was assessed via serum albumin measurements, total-iron-binding capacity and percent ideal body weight. There was no significant difference in the proportion of patients who were hypermetabolic in each group and no significant differences in resting energy expenditure were found between patients with liver metastases and those without, which is in agreement with previous studies (Hansell et al. 1986a and b). The authors concluded that an increase in resting energy expenditure did not appear to be a major factor in the pathogenesis of cancer cachexia (Fredrix et al., 1991).

Weinmann and colleagues (1996) carried out a study to assess whether any peri-operative changes in metabolism and/or body composition were dependent on tumour stage. Thirty-two patients with colorectal cancer undergoing primary resection were included in the study; 14 of these patients had metastatic liver disease. Nutritional status, body composition (bioelectrical impedance analysis) and metabolic activity (indirect calorimetry) were measured 10-14 days prior to and after surgery. The results of the study demonstrated that there was no significant difference both pre-operatively and post-operatively in resting energy expenditure, oxidation of protein, body mass index and urinary excretion of 3-methylhistidine in patients with colorectal cancer irrespective of the presence of liver metastases. However, peri-operatively, patients with liver metastases did demonstrate a significant decrease in body cell mass resulting in a higher extracellular mass to body cell mass ratio compared to other groups. The authors of the study suggested that this partly may be due to the mobilisation, retention of protein and synthesis of protein by the tumour itself (Weinmann et al., 1996).

In summary, in cancer patients with a variety of tumours, the acute-phase response has been associated with a range of effects on the host metabolism. This response appears to be largely mediated by cytokines in particular interleukin-1, interleukin-6 and tumour necrosis factor. These metabolic parameters may contribute to an increase in resting energy expenditure and subsequent weight loss in this patient group. However, at present it is not clear whether the presence of liver metastases results in hypermetabolism. Macfie and colleagues demonstrated that patients with disseminated disease, of which 89% had liver metastases, had a significantly higher resting energy expenditure than control subjects. In contrast, several studies previously discussed in the text did not find any significant differences in energy expenditure in patients with liver metastases (Macfie et al., 1982; Hansell et al., 1986a; Hansell et al., 1986b; Luketich et al., 1990; Fredrix et al., 1991). None of the above studies incorporated measurements of acute-phase proteins such as C-reactive protein as markers of the inflammatory response.

Section 1.8 Body Composition In Patients with Advanced Cancer

In 1932, Warren carried out a study to determine the 'immediate causes of death in cancer'. From the 500 cases studied, he identified cachexia, 'progressive wasting and weakness, accompanied by increasing anaemia', as the commonest cause of death in this patient group. Furthermore, he reported a higher prevalence of cachexia in patients with colorectal, breast and stomach cancer.

In 1957, Craig and Waterhouse assessed changes in body composition in nine patients with advanced cancer. They measured total body water by dilution of deuterium oxide. From the study it was reported that patients with advanced cancer appeared to gain total body water (shared by both intra- and extra-cellular compartments) and lose body fat. The authors stated that patients suffering from insufficient calorific intake of non-malignant origin differed from cancer patients in that extracellular compartments of total body water appeared to increase.

In his review, Brennan (1977) questioned whether patients with advanced cancer responded to starvation in a different way from their non-malignant counterparts. He concluded that patients with cancer did appear to differ in that they were less able to conserve lean tissue mass. Furthermore, he stated that a decreased energy intake and inefficient utilisation of ingested foods appeared to further contribute to this state.

Warnold and colleagues (1978) measured body composition in twenty-nine patients with cancer and compared them to healthy controls (n=164). Body cell mass was measured by total body potassium measurements, total body water by isotope dilution techniques with tritiated water and body fat by a combination of total body water, body weight and body cell mass values. From the results of the study it appeared that body weight and body cell mass were significantly lower in patients with cancer compared with controls. Furthermore, body fat was found to be significantly lower in female patients with cancer.

Watson and Sammon (1980) carried out a study to investigate whether cachexia associated with malignant disease was different in any way from cachexia associated with benign

inflammatory disease. Ten patients with malignant disease and thirteen patients with benign inflammatory disease were included in the study. Body composition was measured using skinfold thickness measurements and radioisotope tracer methods. They reported that patients with cancer cachexia did appear to differ from patients with benign inflammatory disease, in that they had a higher percentage of lean body mass and had higher ratios of total body fat loss to lean body mass loss. They stated that this might imply that lean tissue is better preserved in malignant than in inflammatory state. However, it must be noted that measurements of the inflammatory state were not incorporated in the study.

A later study carried out by Cohn and colleagues (1981) assessed body composition in patients with haematological, lung, gastrointestinal and head and neck cancers and compared their findings to an age and sex matched control group. Assessment of body composition was made by measurements of total body nitrogen, potassium and water. Patients with haematological malignancies did not appear to significantly differ in weight, body fat, muscle mass and muscle protein content compared with controls. In patients with solid tumours, weight loss appeared to consist mostly of loss of muscle mass and body fat. Furthermore, in patients with severe wasting, muscle mass appeared to be predominantly lost and significant amounts of body fat retained.

Heymsfield and McManus (1985) assessed the 'tissue components of weight loss' in patients with cancer. They included three groups of patients in their study; healthy controls, patients with anorexia nervosa and patients with malignant metastatic disease. It appeared that weight loss observed in patients with cancer and anorexia nervosa could be attributed predominantly to loss of fat and skeletal muscle. However, patients with cancer appeared to lose less weight than their anorexia nervosa counterparts. This was thought to be due to factors such as the presence of ascites and tumour bulk. When visceral organ mass was assessed in patients with anorexia nervosa, the liver, heart, spleen and kidney volumes in addition to all other components of fat-free mass including muscle, appeared to decrease in size in accordance with body weight. Contrary to this, in patients with cancer, the spleen and liver tended to maintain size or enlarge and the heart and kidneys appeared to stay the same or decrease in size. Lean tissue mass appeared to rapidly decrease. This preservation of visceral mass in

patients with cancer, in comparison to patients with anorexia nervosa, was also thought to contribute to the observation that patients with cancer tended to lose less body weight. The authors concluded that triglycerides and skeletal muscle proteins appeared to be the primary energy sources involved in weight loss in patients with cancer (Heymsfield & McManus, 1985).

Macfie and Burkinshaw (1987) measured body composition in normal subjects, patients with benign gastrointestinal disease and patients with malignant gastrointestinal tumours (which included thirty-five patients with disseminated disease). Height, weight, body fat, total body nitrogen and total body potassium were measured. From the study it was reported that there was no significant difference in mean body fat or nonmuscle protein between the groups. There were also no significant differences noted in body composition between patients with benign disease and those with malignant disease. Furthermore, patients with metastatic disease did not significantly differ in body composition in comparison to patients with localised disease. It was concluded that the components of weight loss in patients with benign and metastatic disease were similar and that the findings of the study did not support the hypothesis that cancer patients adapted differently to starvation than patients with benign disease (Macfie & Burkinshaw, 1987).

Moley and colleagues (1987) measured body cell mass in three groups of patients; controls (n=233), patients with anorexia nervosa (n=18) and patients with untreated malignant disease (n=104). Body cell mass was measured by total body potassium. In comparison to control subjects, patients with cancer lost a significant amount of weight (12.7% for men and 13.9% for women). Furthermore, associated with this decrease in body weight there was a proportional decline in body cell mass. It was of interest to note that patients with metastatic disease tended to experience greater weight loss. However, body cell mass expressed both as an absolute value and as a percentage of body weight did not appear to differ in comparison to patients with localised disease. Of all the groups examined, patients with anorexia nervosa demonstrated the greatest weight loss. Unlike the untreated cancer patient, absolute body cell mass was significantly less than that of age/sex matched controls and when expressed as a percentage of body weight was 13.8% higher. When the patients with anorexia nervosa were

compared to those with malignant disease, body cell mass as a percentage of body weight was 30% higher. However, the groups were not age matched. From this study it was concluded that patients with anorexia nervosa appeared to demonstrate some sparing of body cell mass in response to starvation. However, the cancer patient appeared to have lost this lean tissue conserving mechanism and exhibited a significant decrease in both body cell mass and body fat during weight loss.

Preston and co-workers (1987) assessed body composition in six patients with non-small cell lung cancer. All patients had more than 25% weight loss from their pre-illness weight. Each patient was compared to 4 age, sex, and height matched control patients. Neutron activation analysis provided measurements of total body nitrogen, sodium, chlorine, calcium and phosphorus. Total body potassium was measured in a whole body counter.. They reported that patients with cancer had on average 82% less fat and 74% percent less muscle protein than controls. There was a disproportionate loss of muscle protein in comparison to fat. Furthermore, it was thought that this loss of muscle protein could be primarily attributed to skeletal muscle. They hypothesised that the protein stores from skeletal muscle were preferentially utilised for liver protein synthesis of acute-phase proteins.

The acute-phase response in patients with cancer has been discussed in the previous section (section 1.7.2). McMillan and co-workers (1994) carried out a study to assess whether the presence of an acute-phase response would affect body composition in this patient group. They also questioned whether the presence of an acute-phase response in certain patients would lead to the identification of patients who may have a 'metabolic' component to their weight loss. Thirty-one patients with gastrointestinal cancer and associated weight loss were included in the study. Body cell mass was estimated by total body potassium measurements. Albumin, total body water and twenty-four hour creatinine excretion were also measured. Patients with a C-reactive protein concentration of >5mg/l were defined as having an acute-phase response. The results of the study identified a significant reduction in total body potassium in patients with an inflammatory response when total body potassium was calculated as a percentage of predicted normal values (McMillan et al., 1994).

A later study carried out by McMillan and colleagues (1998) further investigated the relationship between the acute-phase response and body cell mass in patients with cancer. Longitudinal measurements of C-reactive protein, albumin and total body potassium were carried out over twelve weeks in seven patients with non-small cell lung cancer and eleven patients with gastrointestinal cancer. As in the previous study patients with a C-reactive protein concentration >5mg/l were defined as having an acute-phase response. They reported that there was a significant correlation between the change in total body potassium and C-reactive protein values over the 12-week period. In patients with a C-reactive protein concentration >30mg/l, there was a significant loss of total body potassium compared to patients without an acute-phase response. They concluded that the results demonstrated the significance of the relationship between the presence of an ongoing acute-phase response and the loss of body cell mass in patients with cancer (McMillan et al., 1998).

In conclusion, Warren in 1932 identified cachexia (weight loss) as the commonest cause of death in patients with cancer. The weight lost in cancer patients appears to comprise primarily of lean body mass and body fat (Craig and Waterhouse., 1957; Brennan, 1977; Watson and Sammon., 1980; Cohn et al., 1981; Moley et al., 1987; Preston et al., 1987). In patients with cancer and severe wasting, muscle mass appears to be predominantly lost and fat selectively retained (Cohn et al., 1981). Whether body composition in weight losing cancer patients is different from other disease states remains uncertain (Watson & Sammon, 1980; Macfie & Burkinshaw, 1987). The results from the two studies carried out by McMillan and colleagues (1994, 1998) suggest that there may be a relationship between the presence of an acute-phase response and body composition in patients with cancer.

In patients with liver metastases, studies on body composition are limited; few longitudinal studies appear to have been carried out in such patients. Furthermore, body composition studies do not appear to have been measured in patients receiving treatment regimes such as chemotherapy. Therefore, further work is required to be carried out in these areas to more fully understand the components of weight changes in patients with liver metastases and how this may be altered by chemotherapy.

1.8.1 Leptin and Cancer metabolism

The cloning of the *ob* gene and its encoded protein, leptin, from adipocytes has supported the hypothesis that appetite-regulating signals from adipocytes are integral components of the feedback loop between the periphery and the brain for energy homeostasis (Zhang et al, 1994; Flier, 1998). Leptin is an afferent signal from the periphery to the brain that regulates adipose tissue mass (Zhang et al, 1994). The concentration of leptin is positively correlated with body fat mass. Changes in plasma leptin concentrations in either direction, activate the efferent energy regulation pathways (Zhang et al, 1994). Leptin, via the central nervous system, reduces appetite and increases energy expenditure (Zhang et al, 1994). In the absence of leptin, such as in *ob/ob* mice, animals fail to limit their food intake and become obese.

In the presence of weight loss, the concentrations of insulin and leptin in the brain appear to decrease (Schartz & Seeley, 1997). This causes a cascade of hypothalamic responses that initiate changes in food intake, energy expenditure and peripheral metabolic metabolism, in order to promote an increase in food intake and storage of energy until energy deficit has been corrected. This occurs due to an increased hypothalamic production and release of neuropeptide Y, the production of which is normally suppressed by leptin after binding to a specific hypothalamic receptor (Inui, 1999). Furthermore, if a disease were to produce factors that induce or mimic the hypothalamic effect of excessive negative feedback signalling from leptin, the anticipated outcome would be prolonged anorexia and weight loss which is not accompanied by an adaptive response.

It has been suggested that proinflammatory cytokines are closely linked in the aetiology of cachexia (Licinio & Wong, 1997), with the administration of cytokines mimicking the metabolic changes of infection and cytokine-induced weight loss that are linked to anorexia. Cytokines such as IL-1 and TNF-α cause the production and release of leptin, despite the decrease in food intake that would normally suppress leptin expression (Sarraf et al, 1997). Therefore, it may be that an increase in leptin could contribute to anorexia, by preventing the normal compensatory mechanisms, in the face of decreased food intake. However, it must be

stated that these cytokines have also been reported to cause anorexia even the absence of leptin (Faggioni et al, 1997).

Studies investigating the role of leptin in cancer cachexia in humans remain limited. Recent studies assessing the effect of surgery in humans, have reported that leptin concentrations are increased in the acute inflammatory state and therefore these findings consistent with leptin having a role in anorexia of injury (Wallace et al, 2000; Stratton et al, 1997). Low or undetectable circulating leptin concentrations have been reported in patients with cancer-cachexia (Simons et al, 1997; Wallace et al, 1998). However, such levels of leptin did not appear to cause a decrease in energy expenditure or an increase in appetite. Therefore, it may be hypothesized that in cancer cachexia the leptin feedback mechanism may be dysfunctional, possibly at the hypothalamic level (Simons et al, 1997; Wallace et al, 1998). However, low circulating leptin concentrations did appear to correlate with the low percentage body fat of the patients. Moreover, the presence of a chronic inflammatory response (an increase in C-reactive protein) was not associated with increased leptin concentrations. Therefore it may be hypothesised, that leptin response differs between acute and chronic inflammatory states.

To date studies of leptin in cancer patients have been conducted in those with weight loss and therefore, the effects of the tumour and weight loss on host energy metabolism cannot be readily resolved. Moreover, no studies have assessed the longitudinal relationship between leptin and host energy metabolism in cancer patients.

1.9 Quality of Life in Patients with Colorectal Cancer

The World Health Organisation describes quality of life as being 'a state of complete physical, mental and social well-being and not merely the absence of disease' (World Health Organization Constitution, 1947). According to Bowling (1997) quality of life may be defined as being 'a grade of goodness'. It may also be described as being an expression of 'subjective well-being' (Cohen et al., 1996). Ringdal and Ringdal (1993) propose that quality of life may be described as 'reflecting the subjective feelings of well-being of the individual'

Quality of life was first measured in the 1940's by Karnofsky (Mor et al., 1984; Sullivan et al., 1995). It was originally designed to assess the level of patient activity and thus enable measurement of the patient's medical requirements. Studies have since demonstrated that the Karnofsky performance status is a valid global indicator of functional status in cancer patients (Yates et al., 1980; Mor et al., 1984).

Since then, many other quality of life tools have been devised for use in cancer patients. Maguire and Selby (1989) set up a working party to advise the Medical Research Council on the best available methods for assessment of quality of life in cancer patients. The instruments assessed were defined as being either a global measure of quality of life, an indicator of performance or scales that measured psychological dimensions only.

Global measures included Gough's visual analogue scale, Rosser and Kind's distress/disability matrix and Quality of life adjusted years. Performance indices included the Karnofsky performance status scale, the ECOG (Eastern Co-operative Oncology Group) scale, Katz activities of daily living and World Health Organisation scales. Scales, which were defined as measuring several dimensions, included the Iszak and Medalie Index, Priestman and Baum linear self-assessment system, Functional living index for cancer, Ontario Cancer Institute quality of life questionnaire, Padilla QL questionnaire, QL index, EORTC Quality of life Questionnaire and the Rotterdam Symptom Check list. Tools for assessment of psychological dimensions included Hospital Anxiety and Depression Scale and The General Health Questionnaire.

It was concluded that the Rotterdam Symptom Checklist appeared to be the best in assessing key dimensions of quality of life. Furthermore, the hospital anxiety and depression scale was thought to be of particular use in measuring levels of anxiety and depression in cancer patients. It was concluded that linear analogue scales may be of use in examining differences between treatment regimes (such as adverse effects or disease response). It was recognised that clinical interpretation of scales might prove to be difficult (Maguire & Selby, 1989). However, recently the EORTC (European Organisation for Research and Treatment of Cancer) QLQ-C30 has been shown to be both a reliable and valid tool for use in quality of life assessment in patients with various cancers (Aaronson et al., 1993; Ringdal & Ringdal, 1993).

In patients with colorectal cancer, studies of quality of life are limited (De Cosse & Cennerazzo, 1997). Sprangers and colleagues (1995) assessed quality of life in patients with colorectal cancer, in particular comparing stoma with non-stoma patients. They reviewed 17 studies that compared at least one of the following four aspects; physical, psychological, social and sexual functioning. Although there was conflicting evidence, some long-term effects of colorectal cancer surgery were identified. Both groups of patients appeared to be troubled with diarrhoea and at times, frequent and/or irregular bowel movements. Patients with a stoma appeared to have higher levels of psychological distress. Both groups had poorer social and sexual functioning; stoma patients appeared to fare worse than those without a stoma.

As has been previously described, metastases to the liver are the commonest means of spread from colorectal cancer (Welch & Donaldson, 1979), followed consecutively by the lungs, adrenals, ovaries and bone (Sugarbaker, 1980). As many as 60% of patients with colorectal cancer will develop liver metastases (Welch & Donaldson, 1979; Sugarbaker, 1980), with the liver being the sole site of recurrence in approximately 20% of patients (Weiss et al., 1986). As a result, many of these patients will receive chemotherapy as a form of treatment.

Traditionally biological markers of disease have been used as an end-point to evaluate the 'effectiveness' of treatment regimes in cancer clinical trials (Aaronson et al., 1993). As the majority of treatment regimes offered to patients with colorectal liver metastases are only

palliative, an increasing number of clinical trials conducted in this patients group are incorporating quality of life and performance status assessment (Allen-Mersh et al. 1994; Glimelius et al., 1994; Earlam et al., 1996).

For example, Allen-Mersh and colleagues (1994) randomised 100 patients with colorectal liver metastases to receive continuous hepatic-artery chemotherapy or symptom palliation only. Quality of life was assessed monthly using the Rotterdam Symptom Checklist and Hospital Anxiety and Depression Scale. In patients receiving symptom palliation only, 65% died within a year of diagnosis. Patients treated with chemotherapy survived a median 405 days compared with 226 days for patients receiving symptom palliation only. Quality of life was maintained in the treated group.

Sullivan and colleagues (1995) undertook a randomised double-blind trial comparing 5-fluorouracil and leucovorin to 5-fluorouracil plus placebo in 218 patients with metastatic colorectal cancer. The functional living index-cancer (a psychometrically validated questionnaire) and Karnofsky performance status was use to assess quality of life. This was completed at baseline and eight weekly thereafter. When comparing the two treatment regimes, no statistical differences were noted in tumour response rate, time to tumour response, duration of response, time to treatment failure and survival. The only difference in the drug toxicity profiles between the two groups was that grade 3-4 diarrhoea was more prevalent in patients receiving 5-fluorouracil and leucovorin. Both baseline functional living index-cancer and Karnofsky performance status appeared to predict survival. An increase in Karnofsky performance status scores was also associated with a response to treatment. However, in comparing the two treatment regimes, quality of life scores did not appear to be significantly different. They concluded that the functional living index-cancer did not appear to be of benefit in comparing drug efficacy between drug regimes.

Earlam and co-workers (1996) carried out a study assessing the relationship between tumour size, quality of life and survival in patients with colorectal liver metastases. They proposed that quality of life may be a 'preferable surrogate end-point' in assessing overall survival in chemotherapy clinical trials, as physical markers such as tumour response may not be as

relevant to the patient and may be open to error. Fifty patients were included in the study. Monthly quality of life questionnaires including the Sickness Impact Profile, Rotterdam Symptom Checklist and the Hospital Anxiety and Depression Scale were distributed until death.

A history of weight loss prior to entry into the study, baseline percentage hepatic replacement and alkaline phosphatase levels correlated with survival. There was also a significant correlation between physical, anxiety, depression and Karnofsky performance status scores and overall survival. Of interest was that physical symptom scores appeared to be able to predict survival better than tumour size measured by computed tomography scanning. Quality of life questions, which were observed to correlate best with survival, were ones on diarrhoea, eating, restlessness and the ability to work and sleep. It was suggested that tumour size may not be the only factor in influencing patients health and survival, but that 'tumour products' may have a significant role to play in this process. From the results of the study it was concluded that quality of life indicators may provide better estimation of survival in patients with colorectal liver metastases than tumour size (Earlam et al., 1996).

A subsequent analysis carried out by Earlam and colleagues (1997) assessed the effects of regional and systemic chemotherapy on quality of life in patients with colorectal liver metastases. It was postulated that quality of life may be useful in discriminating between treatments, which appear to be offering the same survival benefit. The study consisted of three study groups; 49 patients receiving symptom palliation only, 35 patients receiving systemic 5-fluorouracil and folinic acid and 51 patients receiving hepatic arterial floxuridine. Quality of life questionnaires used included the Rotterdam Symptom Checklist, the Sickness Impact Profile and the Hospital and Anxiety Depression Scale.

It was observed that patients receiving symptom palliation only had significantly higher hospital anxiety and depression scores compared with those patients receiving chemotherapy. Overall, quality of life and survival did not appear to significantly differ between the patients receiving regional and systemic chemotherapy. However, it was noted that quality of life scores did appear to differ between the two groups at specific time points. Patients receiving

systemic chemotherapy had significantly higher hospital anxiety and depression scores between months four and eight of treatment, and higher Rotterdam symptom checklist scores for sore mouths between months five to eight. It was concluded that due to side-effects, quality of life in patients receiving systemic chemotherapy appeared to be impaired. In contrast, hepatic arterial infusion was associated with similar survival benefit but quality of life appeared to be better maintained.

The above studies suggest that it would be of interest to examine both quality of life and markers of the acute-phase response in patients with colorectal liver metastases receiving 5-fluorouracil based chemotherapy.

1.10 Aims

To date, few studies have been carried out in patients with colorectal liver metastases to assess the relationship between the systemic inflammatory response and host metabolism. Furthermore, few studies have examined the effect of 5FU based chemotherapy on the systemic inflammatory response, quality of life in these patients and outcome.

The aims of this thesis are:

- To examine the relationship between the systemic inflammatory response and body composition, dietary intake, resting energy expenditure, cytokine, hormone and protein metabolism in patients with colorectal liver metastases.

- To examine the short and long term-effects of 5-FU based chemotherapy on the above measures of host metabolism, and quality of life in patients with colorectal liver metastases

2.0 METHODS

2.1 Assessment of Body Composition

2.1.1 Introduction

In order to examine the host response to chemotherapy in patients with colorectal liver metastases, it is important to ascertain the nature of tissue alterations occurring during this time (Ferrannini, 1988). However, it would appear that, as yet, no body compositional studies have been conducted in patients with colorectal liver metastases receiving 5-FU based chemotherapy.

2.1.2 Methods

Body composition may be described as consisting of 2 chemically distinct compartments, fat and fat-free mass (Durnin & Womersley, 1974):

- Fat is described as being anhydrous and potassium free. It is said to have a density of 0.900g/cc at 37 °C (Lukaski, 1987).
- Fat-free mass is reported to consist of 72-74% water, have potassium content of between 50-70mmols and the density at 37 °C 1.1g/cc (Lukaski, 1987).

Based on the premise that fat does not contain water, and that water contributes between 72-74% of total fat-free mass, total body water has been used as an index of metabolically active tissue (Lukaski, 1987). Methods of assessment of body composition which use this two compartment model of body composition include hydrostatic weighing (Durnin & Womersley, 1974; Deurenberg et al., 1994), isotope dilution (van Marken Lichtenbelt et al., 1994; McMurrey et al., 1997) and bio-electrical impedance analysis (Catalano et al., 1993; van Marken Lichtenbelt et al., 1994)

- Hydrostatic Weighing

Hydrostatic weighing is considered to be one of the most accurate methods of assessment of fat-free mass. It is based on the Archimedes principle that states that 'the volume of an object submerged in water equals the volume of water the object displaced' (Lukaski, 1987). With this method of assessment of body composition two basic assumptions are made:

- In the fat-free body, chemical composition is relatively static, and that the density of fat and fat-free mass are significantly different, in order to be able to distinguish between the two.
- The water content and ratio of bone mineral to muscle in the fat-free body is relatively constant.

In order to calculate percentage of body fat using underwater weighing, the equation proposed by Siri is commonly used (Fogelholm and van Marken Lichtenbelt, 1997):

$$\% \text{ BODY FAT} = \left[\frac{4.95 - 4.50}{\text{BODY DENSITY}} \right] \times 100$$

In this equation the density of fat is assumed to be 0.900g/cc and 1.100g/cc for fat-free mass (Lukaski, 1987). Hydrostatic weighing is reported to have a variability of approximately 1.2% (Forbes, 1994).

The advantages of using this technique are that the apparatus is relatively inexpensive. Fat-free mass and body fat can be measured at the same time, it can be readily repeated and has a low variability rate. A major disadvantage is that subject is required to be completely submerged in water. It is therefore considered to be unsuitable for use in children, the elderly and the infirm. Therefore, this method of body composition assessment was not regarded as being suitable for use in patients with advanced colorectal cancer due to the physical demands on the patient during the procedure.

- Isotope Dilution

The dilution of isotope labelled water is also used to determine Fat-free mass. Isotopes of hydrogen (in water) such as deuterium and tritium are among those commonly used. With this technique two assumptions are made (Lukaski, 1987):

- The isotope labelled water that is used, has the same body distribution volume as that of water.
- The isotope labelled water has similar exchange mechanisms to that of water.

Usually, the isotope is either administered orally or intravenously, after which there is an equilibrium period, which is then followed by a sampling period. During the sampling period, blood, urine or saliva is taken to measure the dilution of the labelled water and calculate total body water. Fat-free mass can then be calculated via the equation used by Pace and Rathburn (Fogelholm & van Marken Lichtenbelt, 1997):

$$\text{FAT FREE MASS (kg)} = \frac{\text{TOTAL BODY WATER (L)}}{0.732}$$

The fat mass may then be calculated by subtracting fat-free mass (kg) from body weight (kg). When using deuterium or tritium, this method of assessment of body composition may overestimate body water by 4-5%, due to the substances undergoing some exchange with non-aqueous hydrogen (Forbes, 1994).

The advantages of using this approach are that it is simple and inexpensive. The disadvantages are that the patients may be exposed to radiation and multiple blood samples are required. Moreover, due to the residual label, repeated measurements over a short period of time (i.e. within a week) may be less accurate. Therefore, this method of assessment was not used in the assessment of body composition in the proposed longitudinal studies.

- Bio-electrical Impedance Analysis

Bio-electrical impedance is based on the principle that 'the resistance (impedance) to the electrical current is proportional to the length of the conductor (which is lean tissue) and inversely proportional to its cross-sectional area' (Forbes, 1994). Bio-electrical impedance analysis may be carried out using single frequencies or multiple frequencies.

Commonly frequencies of either 50 kHz or 100 kHz are used for single frequency bio-electrical impedance analysis. This method allows estimation of total body water and furthermore estimation of fat-free mass using the equation derived by Pace and Rathburn (Chumlea & Guo, 1994). The measurement of bio-electrical impedance analysis at both low and high frequencies, allows total body water and intracellular and extra-cellular fluid volumes to be calculated. This is due to the observation that at high frequencies current is conducted by body tissues, whereas at low frequencies, it is predominantly conducted via extracellular fluids (Chumlea & Guo, 1994).

Oldham, evaluating 6 commercial bio-impedance analysers, estimated that in actual practice, precision of this method was approximately 5% (Oldham, 1996). Deurenberg and coworkers (1994) investigated whether there were any differences in results of various methods of body composition when the same subjects were assessed in 3 different laboratories. They reported no significant inter-laboratory differences in the relationship between total body impedance and fat-free mass measured by densitometry (Deurenberg et al., 1994). Catalano and colleagues (1993) found bio-electrical impedance analysis to be of use in the nutritional assessment of cancer patients.

The advantages of using bio-electrical impedance as an assessment of body composition are that it offers no hazard to the patient and allows repeated measurement of total body water and consequently fat free mass. Furthermore, it is portable, requires minimal patient co-operation and is a relatively reliable and accurate method of assessment of body composition. It was therefore chosen as a suitable tool for assessment of body composition in patients with colorectal liver metastases.

- Anthropometry

Anthropometry is an approach to the measurement of body composition, which was first devised in the late 19th century. As the major body store of fat is subcutaneous, various anthropometric measurements such as thickness of skinfolds and body circumferences have been used to compare individuals with standard reference populations (Durnin & Womersley, 1974; Hansell et al., 1986), for longitudinal follow-up (Harries et al., 1985) and to calculate the total body fat of an individual (Durnin & Womersley, 1974; Lean et al., 1996).

Anthropometry is founded on two basic assumptions (Forbes, 1994):

- The thickness of the subcutaneous fat mantle reflects total body fat.
- The site of measurement (used either individually or in combination) represents the average thickness of the whole mantle.

Triceps, biceps, subscapular and suprailiac skinfold thicknesses are frequently used (Durnin & Wormersley, 1974) . Body circumferences that are also regularly assessed include mid-arm circumference and waist circumference (Harries et al., 1985; Hansell et al., 1986; Lean et al., 1996).

Hansell and colleagues (1986) measured mid-arm muscle circumference and triceps skinfold thickness in 84 patients with various cancers, and compared them to 'expected normal values'. By doing this they reported that these patients appeared to have less muscle and fat than the standard population.

Harries and co-workers (1985) longitudinally measured mid-arm circumference, triceps, biceps, subscapular and suprailiac skinfold thickness in 28 malnourished patients with Crohns disease receiving controlled nutritional therapy. They reported that changes in weight correlated highly with mid-arm circumference measurements in this patient group. Furthermore, when they assessed intra and inter-observer variability in the assessment of skinfold thicknesses and mid-arm circumference, no significant bias was noted. Mid-arm

circumference measurements were reported to demonstrate the least variation and were found to be reproducible even in obese patients.

Body fat from anthropometric measurements is commonly calculated by the equations derived by Durnin and Womersly (1974). Lean and colleagues conducted a study to examine all simple anthropometric methods currently used to predict body fat. On estimation of body density (via underwater weighing), body fat could then be calculated using the Siris formula. Body density values used for comparison were measured by underwater weighing. The equations used were obtained from data from a healthy, adult, white population. Correlation coefficients observed between body fat and individual skinfold thicknesses and mid-arm circumference in males and females were:

MEASUREMENT	MEN- (R^2)	WOMEN- (R^2)
Biceps	0.730	0.695
Triceps	0.612	0.768
Mid Upper Arm Circumference	0.586	0.790

The best equation for prediction of body fat for men involved measurements of waist circumference, triceps skinfold thickness and age. For women, the most accurate method involved measurements of body mass index, triceps skinfold thicknesses and age. Other equations that used triceps skinfold values were also found to give 'good' predictions of body fat (>70.6% variance).

The disadvantages of using body anthropometry, as a method of assessment of body composition is that such estimates can vary between 6-24% (Forbes, 1994; Harries et al., 1985). Furthermore, it appears to have poor precision in obese subjects and those with firm subcutaneous tissue and that there may be regional variations in the subcutaneous fat layer. The advantages that it is cheap, safe, allows direct estimation of body fat and regional muscle. It is portable, may be repeated many times in a short time span, requires low levels of patient compliance and is of minimal discomfort to the patient. It was therefore deemed to be suitable

for use in the assessment of body composition in patients with colorectal liver metastases undergoing chemotherapy.

2.1.3 Summary

In conclusion, in order to examine the effects of chemotherapy in patients with colorectal liver metastases, it is important to assess the host response. One method of assessing this response is by longitudinal measurement of body composition. In the present studies, the two-compartment model of body composition was used (fat and fat-free mass). This was assessed using bio-electrical impedance analysis and skinfold anthropometry as previously described. In order to maximise the accuracy of these measurements, they were carried out in triplicate by the same person each time and the average values calculated. These methods of assessment were chosen because they are accurate, reliable, safe and involve little patient co-operation, and were suited to the sometimes frail and unwell population of patients studied.

2.1.4 Study Methods

- Bio-electrical Impedance Analysis

In the present studies multi-frequency bio-electrical impedance analysis was performed using a Xitron 4000B Complex Bio-impedance spectrum analyser (Xitron Technologies, Inc., San Diego, CA 92121, USA).

Prior to measurements being carried out, the patient was placed on a clinical couch. During the measurement the patient maintained a supine position with their arms relaxed at their side (ensuring at they were not in contact with the body) and feet slightly apart. At no point was the patient or any leads from the analyser in contact with any metal surfaces or other leads. The position of electrodes is very important when carrying out measurements using this analyser, as it will accurately measure parameters between the points of contact of the electrodes (Xitron type ISI4000). Therefore, physical landmarks were used to decrease the variability of measurements. Prior to the placement of electrodes, the skin was wiped with 70% isopropyl impregnated swabs and allowed to dry. Current source electrodes were placed on the right hand at the distal metacarpal joint and on the corresponding right foot at the distal metatarsal joint. Detection electrodes were placed between the right distal prominence of the radius and ulna and the between the right lateral and medial maleoli.

Prior to measurements carried out on each patient, the analyser was calibrated using a 422 ohm test resistor supplied by Xitron Technologies, Inc. Fifty frequencies between 5kHz and 1Mhz were used to measure resistance and reactance. The manufacturer supplied a modelling programme that carried out Cole-Cole analysis of the recorded spectrums. Quantitative statistics were used to assess the quality of the 'fit' of the data to this model. This type of analysis predicts the impedance at zero that has been reported to correspond with extracellular resistance and to infinity, which has been demonstrated to correspond with total body water volume values (Lukaski, 1987). The patients' height and weight were then combined with these values in order to derive intracellular, extracellular and total body water volumes.

- Anthropometry

Height was measured via a stadiometer (to the nearest centimetre) and weight was measured via beam-balance standing scales (model 424; Weylux; Cambridge, UK). Harpenden skinfold callipers (British Indicator Ltd., West Sussex, United Kingdom) were used in the present study. These callipers produce a constant spring pressure of $10g/mm^2$ throughout the skinfold range, which is the pressure advocated for this type of measurement to be carried out (Heymsfield et al. 1994). A stretch resistant tape was also used to accurately measure limb circumference and help in the location of sites. Triceps and biceps skinfold thicknesses were measured as well as mid-arm circumference. At this time the patient was standing upright, with their arms relaxed by their sides. The right arm of the body was used during each occasion. The triceps site was measured directly over the triceps muscle on the posterior midline of the upper arm, at the midpoint between the acromial process of the scapula and the olecranon of the ulna. The biceps site was located on the anterior surface of the biceps midway between the axiliary fold and the antecubital fossa. Midarm circumference was measured at the midpoint between the olecranon and the acromial process. At this time the tape was in a horizontal position, slightly touching the skin, but at no times compressing it. Each skinfold measurement was triplicated and the average score was then recorded to the nearest 0.5 millimetres.

2.2 Assessment of Energy Intake

2.2.1 Introduction

Energy intake may be defined as being 'the energy contained in the food consumed in the diet that is available for metabolism' (Seale & Rumpler, 1997). According to Beaton, 'there is not, and probably never will be, a method that can estimate dietary intake without error' (Beaton et al., 1997). Associated with certain subgroups of the population are specific errors inherent to them. For example, in the elderly, recall may be limited and any disabilities in sensory perception such as hearing, sight may complicate the assessment. In patients who are ill, dietary intake may vary considerably from day to day, often being dependent on the health of the individual at that time. The ability to remember and record which foods have been eaten may also vary, depending on how the individual feels that day. Furthermore, the patients 'net intake' may be affected by conditions such as vomiting and diarrhoea (Dwyer, 1994). Briefel and co-workers (1997) identified patients who were most likely to under-report dietary intake; they tended to be women, patients who were older, overweight and/or trying to lose weight.

Dietary intake may be measured retrospectively, prospectively or a combination of the two. Retrospective methods include 24-hour recall, food frequency questionnaire, semi-quantitative food frequency questionnaire and Burke-type dietary history (Dwyer, 1994).

2.2.2 Methods

- Prospective Methods

Prospective methods appear to be more suitable than retrospective recall for patients with advanced cancer. The main advantage of this type of dietary assessment is that it does not appeared to be highly dependent on the subjects' memory and recall ability, as food and drink intake is recorded at the time of consumption (Dwyer, 1994). Prospective methods of assessment of food intake include food diaries, weighed food records, telephone records,

photographic/video records, electronic records, duplicate portion analysis, intakes and outputs and direct observation by either trained observers or by video.

- Food Diaries

This type of dietary assessment involves the subject prospectively recording estimates of all food and drink consumed over the period of assessment. Commonly estimates are based on common 'crude' kitchen measurements such as cups, tablespoons etc. Food pictures/models in estimating portions may also be used. Advantages of using this technique include that food is recorded at the time of consumption and therefore does not rely as heavily on the subjects' memory. If the subject is adequately instructed, errors in estimating food intake may be reduced. Disadvantages are that the subject may find it obtrusive and estimations of intake may prove difficult. The individual must also be literate and physically able to write down records of intake. Due to these factors, subjects with high levels of compliance and motivation are required. They may also alter their intake on days of assessment and under reporting appears to be prevalent with this technique. Furthermore, sex differences in reporting exist, with women appearing to be competent than men (Dwyer, 1994). Lastly, the cost of coding and analysis can be expensive.

- Weighed Food Diary

All food and drinks consumed are weighed and recorded at the time of ingestion. The main advantage of this technique over estimations of portions is that overall accuracy of intake may be improved by up to 40% (Dwyer, 1994). Other advantages and disadvantages are similar to food diaries. However, one further disadvantage is that when subjects are eating/drinking away from home, they may not be able to provide weighed samples of food and drink intake e.g. restaurants/bars etc.

- Telephone Records

The subject reports food and drink intake as soon as it occurs. Advantages are that patient anonymity may be kept if required. Furthermore, less responsibility is put on the subject in comparison to written records, which in turn increases patient compliance. With this technique, patients who may have problems with transport (disability, distance) may be

assessed. Disadvantages are that it is assumed that portion sizes are actually eaten and validation studies on this technique are incomplete.

Other prospective methods of measuring dietary intake include photographic/ video tape recording, duplicate portion collection and analysis and food recording electronic devices.

2.2.3 Summary

Of the prospective methods discussed, food diaries appear to be best suited to patients with advanced colorectal cancer. Estimation of food portions with this method can be carried out using simple standard measures such as cups/ tablespoons etc and does not require food to be weighed prior to consumption. Also, the patient can carry the diary around with them and measure dietary intake when not at home e.g. in restaurants. Due to the portability and ease associated with of this method of assessment, it was considered to be suitable for use in patients with advanced cancer receiving palliative treatment.

2.2.4 Study Methods

Dietary intake was measured using a four-day food diary, which included two weekdays and two days at the weekend. They were asked to record entries immediately after they had been consumed and not at the end of the day. Portion sizes were estimated using simple standard kitchen measures such as cupfuls, tablespoonfuls and teaspoonfuls. Subjects were asked to describe food as being small, medium or large and thin, medium or thick. The method in which the foods were cooked, ingredients and brand names, if applicable, were also asked to be included. If tinned or packet foods were used then their weight were requested to be incorporated in the diary sheet.

2.3 Assessment of Energy Expenditure

2.3.1 Introduction

To sustain body physiological functions, energy is constantly expended by oxidative metabolism. Furthermore, ultimately all the energy produced by the body is lost as heat (Schutz & Jequier, 1994). At rest, the principle sites of energy (heat) production are the trunk, viscera and brain. During physical activity, the prime site of energy production is muscle (Kinney, 1988).

Energy expenditure can be expressed in either of two ways; total energy expenditure or resting energy expenditure. Total energy expenditure is the value obtained when the subject is undergoing 'normal' physical activity. Resting energy expenditure is carried out when the subject is at rest.

Methods of assessment of total energy expenditure include whole body direct calorimetry (Jequier, 1985; Jequier & Schutz, 1988; Murgatroyd et al., 1993), whole body indirect calorimetry (Murgatroyd et al., 1993; Jequier, 1985; Seale & Rumpler, 1997), the doubly labelled water method (Murgatroyd et al., 1993; Jequier & Schutz, 1988; Seale and Rumpler, 1997) and the bicarbonate method (Murgatroyd et al., 1993).

Measurement of total energy expenditure includes assessment both at rest and activity. However, with this approach, it may be difficult to separate the effects of chemotherapy on the host at rest, from the effects on activity. Therefore, measurement of resting energy expenditure was used as an appropriate end-point for the assessment of energy metabolism in the forthcoming studies.

2.3.2 Methods

- Whole Body Direct and Indirect Calorimetry

Direct calorimetry 'measures energy expenditure as the rate at which heat is lost from the body to the environment. This heat is transferred through non-evaporative heat loss

(radiation, convection, conduction) and through the evaporation of water' (Murgatroyd et al., 1993). As this method of assessment of energy expenditure is based on heat loss, and not influenced by metabolic processes involved in heat production, it may be considered to be one of the most accurate methods of assessment of energy expenditure. Furthermore, it is often used as a reference method for the assessment of energy expenditure.

Commonly, direct calorimetry is carried out as a whole body measurement with the patient at rest, in a sealed chamber. Non-evaporative heat loss can be measured by either of two ways; actively or passively. Active measurement involves measuring the rate at which heat is removed from the sealed, well-insulated chamber. This is commonly referred to as 'heat-sink' calorimetry. Passive measurement, more commonly known as 'gradient layer calorimetry', assesses heat loss via temperature gradients across a sealed, poorly insulated chamber. Evaporative components of heat loss can also be evaluated by measuring the latent heat of condensation (by condensing any water present in the sealed chamber) or the latent heat of vaporisation (by measuring the water content of air). With direct calorimetry, estimated oxygen and expired carbon dioxide levels are reported to be within 2% accuracy (Jequier, 1985; Jequier & Schutz, 1988). Advantages of using whole-body direct calorimetry in assessing energy expenditure include that it is accurate, allows direct measurement of energy expenditure and appears to be suitable for use in strict, controlled trials. Disadvantages include that it involves numerous small, detailed measurements (such as heat derived from food, excreta etc) in order to obtain accurate energy expenditure values. The equipment is expensive and requires skilled operators to use it. Furthermore, to combine invasive simultaneous measurements may prove to be difficult. The patient is also in an artificial, observed environment and this may cause behavioural changes, which may affect the overall results obtained. It may also prove to be extremely time consuming, which does not appear to be suitable for use in longitudinal studies in which repeated measurements at different time spans are required.

Indirect calorimetry 'measures energy expenditure as the rate at which heat is produced by the body' (Murgatroyd et al., 1993). Heat production is calculated from measurements of oxygen consumption and carbon dioxide production. Furthermore, indirect calorimetry also allows *in*

vivo fat and carbohydrate oxidation rates to be calculated (Murgatroyd et al., 1993). In an individual under basal conditions, direct and indirect calorimetry is reported to produce similar results (Jequier, 1985). However, events that increase heat production such as a meal/exercise do not directly cause an increase in heat loss. Due to this, direct calorimetry must be measured for at least 24-hours and this is considered to be a major disadvantage of this type of measurement in comparison to indirect calorimetry (Jequier, 1985). There are various methods of measurement of resting energy expenditure via indirect calorimetry.

One such method is whole body indirect calorimetry. This involves the patient being assessed in a sealed room at rest. During this time the room is supplied with a continuous, measured volume of fresh air. Samples of room air (including expired air) are then constantly analysed. The differences in oxygen and carbon dioxide concentrations in the inspired and expired air allow the calculation of energy expenditure. Furthermore, if required, protein oxidation rates (via estimations of urinary nitrogen) and alcohol oxidation rates (via breath samples) can be used in equations to calculate the contribution of different substrates to overall energy expenditure (Murgatroyd et al., 1993). Advantages of this technique include that it is accurate, provides information on substrate oxidation if required and appears to be of use in strict, controlled trials. Disadvantages are that it requires skilled operators with necessary computing and engineering skills. Furthermore, to carry out other measurements at the same time may prove to be problematic. The patient is in an artificial environment and this may lead to changes in behaviour during the period of assessment. It requires patients who are highly motivated and relatively fit. The equipment is expensive and 24-hour supervision is required. Lastly, this method of measurement can prove to be extremely time consuming. From the foregoing, it is clear that direct and indirect calorimetry via whole body chambers are not suitable methods of assessment of energy expenditure in patients with metastatic liver disease.

- Portable and Ambulatory Methods of Indirect Calorimetry

Indirect calorimetry may also be measured via portable/ ambulatory methods. These methods of assessment are thought to have an accuracy of between 2% and 4% (Murgatroyd et al., 1993). Such measurements are commonly carried out over relatively short periods

(approximately 30 minutes) and values extrapolated to give resting energy values per 24 hours.

- The Douglas Bag

The Douglas bag method allows measurement of energy expenditure to be conducted both at rest and during activity. The bag (which is gas impermeable) is carried on the subjects' back and expired air is transferred to the bag via the use of noseclips and mouthpieces. Advantages include that it involves the use of simple, robust, inexpensive equipment and provides accurate estimations of energy expenditure. Disadvantages are that leakage of carbon dioxide via diffusion from the bag is unavoidable, which affects the overall values obtained. Furthermore, the expired air has to be quickly analysed after collection. Due to the use of mouth pieces and nose-clips, measurements can not be carried out over long periods of time. Moreover, use of such equipment requires highly motivated individuals to participate in the assessments. Therefore, the use of a Douglas bag in patients with advanced cancer was not considered to be suitable to estimate energy expenditure.

- The Oxylog System

This system is commercially manufactured and is used to measure energy expenditure via indirect calorimetry. With this type of equipment, direct digital values of cumulative oxygen consumption are displayed every minute. Inspiratory volume is measured via a turbine flow meter, and polarographic cells measure partial pressure of inspired and expired oxygen. Advantages of using this system include that it is smaller and less cumbersome than the Douglas bag. It appears to be suitable for use at all levels of activity, including at rest. Instant gas analysis is provided with minute-by-minute results. However, it is only suitable for use over short periods of time due to the discomfort of the mouthpieces/ noseclips. Consequently, it was considered to be unsuitable for use in patients with advanced colorectal cancer receiving 5-FU based chemotherapy.

- Clinical/ Laboratory Indirect Calorimetry

Indirect calorimetry has also be carried out in clinical and/or laboratory situations. One method is via the ventilated hood system. This method allows air to be drawn over a subject's

head while they are at rest, and inspired oxygen and expired carbon dioxide values measured. The ventilated hood system commonly comprises of a pump, flow meter and a device to regulate air flow. Samples of air are then transferred to oxygen and carbon dioxide gas analysers. This approach was considered to be suitable for use in patients with colorectal liver metastases.

2.3.3 Summary

In conclusion, there are several methods for the assessment of resting energy expenditure. On evaluation of the previous literature, the ventilated hood system was considered to be the most suitable for use in patients with advanced colorectal cancer undergoing longitudinal assessment of resting energy expenditure.

2.3.4 Study Methods

The subjects were fasted overnight. Assessment of resting energy expenditure was carried out in the morning, after an overnight fast, using a ventilated hood system. The Deltatrac metabolic monitor (Datex Instrumentarium Corp., Helsinki Finland) was switched on for 30 minutes prior to calibration being carried out. It was then calibrated using a two-point calibration according to the Deltatrac Operators Manual.

Calibration was commenced by pressing the 'CAL' key, followed by the 'GAS' key, when the machine was in a 'measurement off' state. At this time, the Deltatrac automatically conducted a baseline check of the oxygen and carbon dioxide sensors. Furthermore, during this time, the carbon dioxide level of ambient air was measured and displayed. When 'GAS CALIBRATION' appeared on the screen, the calibration gas (Quick Cal gas-Datex Instrumentarium Corp., Helsinki Finland), was fed into the respiratory sample line until oxygen and carbon dioxide levels stabilised, and 'O_2 ACCEPTED' and 'CO_2 ACCEPTED' appeared on the screen. The readings were then adjusted, if required, to their nominal values (95% for O_2 and 5% for CO_2) and the Deltatrac returned to 'operator mode'. The equipment was serviced 6-8 monthly by qualified Deltatrac technicians.

The subject then lay on a bed for a period of 5-10 minutes prior to commencement of measurements. The canopy was carefully placed over the subjects' head and the plastic apron attached to the canopy was spread out loosely over the subject. The canopy was ventilated at a constant flow rate of approximately 40 litres/ minute. In the respirator, expired air was transferred into a 4 litre mixing chamber, and expired oxygen and carbon dioxide concentrations estimated.

The patient was instructed to lie on an examination couch quietly at rest in a relaxed, wakened state. Measurements were then carried out over a period of 25 minutes, with the first 5 minutes being discarded as the patient acclimatised to the hood. A printout of oxygen consumption, carbon dioxide production, respiratory quotient and 24 hour energy expenditure were then obtained each minute via a printout. Resting Energy Expenditure was calculated using the abbreviated Weir Equation (1949).

Resting Energy Expenditure (kcal/day) $= (3.9\ VO_2 + 1.1\ VCO_2)\ 1440$

VO2 = Oxygen Consumption

VC02 = Carbon Dioxide Production

2.4 Assessment of Tumour Response

2.4.1 Introduction

According to the World Health Organisation (Miller et al., 1981) it has become necessary to develop a 'common language to describe the results of cancer treatment and agree upon internationally acceptable general principles for reporting and assessing data'. On the reporting of response to treatments they advocate that generally four weeks should be the minimum duration of reported response. Furthermore, patients with measurable disease should be classified into the following criteria where possible (Miller et al., 1981):

- Complete Response: 'The disappearance of all known disease, determined by two observations not less than four weeks apart'.

- Partial Response: '50% or more decrease in total tumour load of the lesions that have been measured to determine the effect of therapy by two observations not less than four weeks apart. Bidimensional: single lesion, greater than or equal to 50% decrease in tumour area (mutilplication of longest diameter by the greatest perpendicular diameter); multiple lesions, a 50% decrease in the sum of the products of the perpendicular diameters of the multiple lesions. Unidimensional : greater than or equal to 50% decrease in linear tumour measurement. In addition there can be no appearance of new lesions or progression of any lesion'.

- No Change: 'A 50% decrease in tumour load cannot be established nor has a 25% increase in the size of one or more measurable lesions been demonstrated'.

- Progressive Disease: '25% or more increase in the size of one or more measurable lesions or the appearance of new lesions'.

2.4.2 Study Methods

World Health Criteria were used to define response in the following studies. A CT scan was carried out after each set of 6 cycles (approximately 3 monthly). If patients had a complete response, partial response or no change and acceptable toxicity profiles, then therapy was continued for a further 6 cycles. On disease progression patients received either best supportive care or second line chemotherapeutic drugs.

2.5 Assessment of Haematological and Biochemical Parameters

2.5.1 Haematological Parameters

Haemoglobin, red blood cell count, white blood cell count, differential and platelet count were measured using a S-Plus STKR flow cytometer (Coulter Corporation, Luton, United Kingdom). Fibrinogen was measured using a kinetic fibrinogen assay.

2.5.2 Biochemical Parameters

- Carcinoembryonic Antigen and Cortisol

These were both measured by immunoassay on the Bayer Immuno-1 analyser (Bayer PLC, Diagnostics Division, Newbury, Berkshire, United Kingdom).

- Albumin, C-reactive Protein and Alkaline Phosphatase

Albumin, C-reactive protein and alkaline phosphatase were measured on an Olympus AU5200 analyser (Olympus Diagnostic Systems. Eastleigh, Hampshire, United Kingdom). Albumin was measured by a dye-binding procedure. Intra and inter-assay CV'S were 1.2% and 3% respectively. C-reactive protein was measured by turbimetry after binding to a specific antibody. Intra and inter-assay were 5% and 7% respectively. Alkaline phosphatase method was based on the enzymatic conversion of p-nitrophenyl phosphate to p-nitrophenyl. Intra and inter-assay CV's were 3.6% and 5% respectively.

- Urea and Electrolytes, Liver Function Tests,

Urea and Electrolytes and Liver function tests were measured using a Bayer Adventia Analyser (Bayer PLC, Diagnostics Division, Newbury, Berkshire, United Kingdom).

- Insulin was measured using an ALPCO Insulin ELISA assay.

2.6 Assessment of Quality of Life

2.6.1 Introduction

Quality of life is a 'multidimensional construct that has become a prominent issue in modern cancer treatment' (Cella & Cherin, 1987). One reason for this is the development of more aggressive cancer treatments, which have questionable impact on cancer patients and their outcome. As a result, quality of life assessment has recently been included in a number of cancer clinical trials. With regard to patients with advanced colorectal cancer, quality of life assessment has been used to assess the efficacy of specific chemotherapy based regimes (Allen-Mersh et al., 1994) and to discriminate between various methods of administration of chemotherapy (Earlam et al., 1997). Furthermore, in a study conducted by Earlam and colleagues (1996), it was suggested that quality of life may be a better indicator of survival than physical markers such as tumour size.

Various quality of life tools that are commonly used in cancer clinical trials include the Karnofsky Performance Index, The Hospital Anxiety and Depression Scale, The Rotterdam Symptom Checklist, The General Health Questionnaire and the European Organisation for Research and Treatment for Cancer Modular Approach (Bowling, 1995).

Maguire and Selby (1989) suggested that quality of life tools used in patients with cancer should include assessment of adverse effects of treatment, physical functioning, social interaction, psychological functioning, sexual functioning and perceptions of body image. They also suggested that any measure should have the ability to assess one or more of these identified dimensions in a reliable and valid manner. Furthermore, they should be able to be completed by the patient within ten minutes, be easily administered and scored and be able to indicate any changes over time.

2.6.2 Methods

- The Karnofsky Performance Index

This index was initially developed in the 1940's as a measure of nursing workload (Bowling, 1995). Since then it has been widely used by clinicians as an indicator of functional status in patients with cancer. It is largely dependent on the physical dimensions of quality of life. The index consists of 11 categories that range from normal functioning (100) to dead (0). Each category combines information on the patients' capacity to function at work, at home, the severity of symptoms and personal/medical care requirements (Schaafsma & Osoba, 1994).

The scale largely measures the physical dimensions of quality of life. It is most commonly completed by a health professional, and not by the patient his/herself. Scores are summed to provide an overall score. However, it must be noted that the scoring procedure of the Karnofsky Performance Index has never formally been addressed (Bowling, 1995).

Mor and colleagues (1984) assessed the validity and reliability of the Karnofsky Performance Index as part of a national hospice study. It appeared to be significantly related to two other independent measures of functional ability. Furthermore, inter-rater reliability coefficients of >0.97 were reported. When the relationship between Karnofsky status and longevity was addressed in a population of terminally ill cancer patients, a correlation of 0.30 was reported which appeared to demonstrate its predictive validity.

Yates, Chalmer and McKegney (1980) also assessed the reliability and validity of the Karnofsky Performance Index. A Pearson correlation coefficient of 0.69 was reported between nurses and social workers scoring the same patient, which was considered to indicate a moderate degree of inter-rater reliability. Furthermore, average ratings for scores between the two sets of observers (nurses/social workers) were reported to be within 0.5 points of one another. A moderate degree of inter-rater reliability was demonstrated when a Pearson correlation coefficient of 0.66 was reported between home and clinic scores.

Advantages of the Karnofsky Performance Index include that it can be completed quickly and easily scored. Furthermore, studies have reported moderate levels of inter-rater reliability, validity and it has been suggested that it may have a predictive value in some groups of cancer patients.

However, it only measures the physical domains of quality of life, which limits its use as a global quality of life tool. It assumes that a patient with a low Karnofsky Index score, may have a poorer quality of life than patients with a higher degree of physical performance. The scoring system has never been formally assessed. Furthermore, interpretations of scales within the index are likely to vary, as each point appears to cover differing conceptual elements. Ultimately, the fundamental flaw of the Karnofsky Performance Index is that it involves patients being scored by another persons and not by themselves. Mercier and colleagues (1992) assessed the difference between Karnofsky index scores which were scored by the patient themselves (n=100) and by their clinician. Interclass correlation coefficients of 0.56 were reported, with perfect agreement in 25% of patient and physician pairs. When a *post hoc* analysis was carried out using a Pearson correlation coefficient, a higher correlation (r=0.633) was reported. It was concluded that there was a moderately low correlation between patient and physician scores. However, it was suggested that this was in part due to the patients' age and cancer site.

The Karnofsky Index was used in the forthcoming studies not as a quality of life tool but as a measure of functional status in patients with colorectal cancer receiving 5-FU based chemotherapy. It was scored by the same individual each time and used in the longitudinal follow-up of patients.

- The Hospital Anxiety and Depression Scale

This scale consists of 14 separate items that are set on two sub-scales; seven questions are based on anxiety and seven questions are based on depression. It can be self-administered or an interviewer can be used. Replies are based on feelings experienced over the past week and the subjects' rates their replies based on a four point scale. The hospital anxiety and

depression scale is scored by summing up the items on each of the two sub-scales; a score of ≥ 11 denotes significant morbidity.

The patients' scores do not appear to be affected by the presence of physical illness (Bowling, 1995). Moderate to high correlations (0.67-0.77) have been reported with other well-known anxiety and depression scales (Bowling, 1995). Furthermore, correlations of 0.41-0.76 for the anxiety scale and 0.30-0.60 for the depression scale have been reported on the assessment of the internal consistency of the scale (Bowling, 1995).

Maguire and Selby (1989) concluded that the hospital anxiety and depression scale 'seems a useful tool for measuring the psychological dimensions of quality of life in cancer patients'. They stated that the scale had the advantage that it was originally designed for use in patients with physical disease, and therefore, did not include items such as tiredness, which could be classified as being both due to mood disturbances and physical illness. Furthermore, factors such as the scale being self-administered, being quick and easy to apply and having the ability to assess change over time in patients with various cancers, makes it suitable for use in patients with cancer.

However, despite being a valid and reliable scale for the measurement of anxiety and depression, the scale was not thought to be suitable for use in patients undergoing chemotherapy. This was fundamentally due to the scale only measuring the psychological domains of quality of life, and not including factors such as recognised adverse effects of chemotherapeutic regimes, which have been reported as affecting overall quality of life in patients with cancer.

- The Rotterdam Symptom Checklist

The Rotterdam Symptom Checklist was originally devised by De Haes and colleagues (1990) as a tool to measure symptoms reported by patients with cancer participating in clinical research trials. Its content and structure were based on analyses of data from three previous studies that used varying symptom checklists (Bowling, 1995). Items were finally selected

based on factor analyses, the distribution of answers and judgements made by clinical oncologists.

The checklist was originally tested using eight items of daily living and 34 items based on symptoms. Scores were based on experiences over the past three days on a four point Likert-type rating scale (Bowling, 1995). The higher the score, the greater the severity of the symptoms. Validation originally took place in a Dutch study and since then it has been implemented in a number of British and Dutch Clinical trials.

Presently, the Rotterdam Symptom checklist consists of 30 symptom-based items, eight items based on the activities of daily living and one global quality of life item. It is usually self-administered and takes approximately five to ten minutes for completion. Overall scores can be summed up to provide two main sub-scales that measure the physical and psychological domains of quality of life (Bowling, 1995).

Maguire and Selby (1989) described the Rotterdam Symptom Checklist as being 'a good, clear and simple questionnaire which has been validated against independent interviews and found to have high sensitivity and specificity in measuring psychological dimensions'. On behalf of the Medical Research Council, they advocated the use of the Rotterdam Symptom Checklist, supplemented with other tools as required, such as the Hospital Anxiety and Depression Scale. Acceptable levels of internal reliability for physical distress and psychological components of the Rotterdam Symptom Checklist have been reported (Cronbach's alpha=0.71-0.88 and 0.88-0.94 respectively),(de Haes et al., 1990).

However, despite appearing to adequately assess the psychological and physical dimensions of quality of life, in patients undergoing cancer treatments, the checklist appears to fail to sufficiently address the social or sexual domains of quality of life. Therefore, it was not considered suitable for use in the present clinical studies.

- The European Organisation for Treatment of Cancer (EORTC) Modular Approach

During 1980, a quality of life study group was devised within the EORTC. Its overall objective was to develop a brief, standardised, quality of life measure which could be implemented in international clinical cancer trials.

Originally, the questionnaire consisted of 42 items, which was then reduced to 36 items. This has now been decreased to consist of only 30 items (EORTC QLQ-C30), which is the version currently promoted for use by the EORTC (Bowling, 1995). The EORTC QLQ-C30 is a multidimensional, cancer-specific, quality of life questionnaire. It is patient based, intended for self-administration and considered to be applicable across a heterogeneous group of cancer diagnoses (Ringdal & Ringdal, 1993).

It encompasses 5 functional scales which consists of physical, role, emotional, cognitive and social functioning. There are also three symptom scales (fatigue, nausea and vomiting, pain) and six single items (breathlessness, sleeping, appetite, constipation, diarrhoea and financial problems). Furthermore, there are two general quality of life items.

Items on physical functioning are answered 'yes' or 'no'. Questions regarding symptoms, anxiety, depression and limitations are answered using a choice of four responses:
1='not at all' 2='a little' 3='quite a bit' 4='very much'

The two questions based on overall physical condition during the past week and overall quality of life are scored using a seven point visual analogue scale, which ranges from 'very poor' to 'excellent'. The EORTC QLQ-C30 is regarded as a core questionnaire and may be supplemented by other modules such as lung, breast and head/neck cancers.

Niezgoda and Pater (1993) tested the EORTC QLQ-C30 on 96 mixed cancer chemotherapy patients in Canada. When compared to the Sickness Impact Profile, a moderate to strong correlation was reported with the following sub-scales of the questionnaire:

SUB-SCALE	R =
Physical	0.73
Emotional	0.48
Social	0.48
Cognitive	0.58
Fatigue	0.58
Role	0.55

When the pain scale was compared to the McGill pain scale, moderate correlations were reported:

PAIN SCALE	R =
Sensory/affective	0.57
Present pain intensity	0.53

Moderate to strong correlations between the sub-scales of the EORTC-QLQ C30 and the Cancer Rehabilitation Evaluation system were also reported:

SUB-SCALE	R =
Physical	0.71
Emotional	0.56
Pain	0.69
Finance	0.61
Social	0.46
Symptoms	0.55

Furthermore, a strong correlation (r=0.61) was reported between the psychological sub-scale and the General Health Questionnaire.

Practicality and reliability was assessed by Aaronson and colleagues (Aaronson et al., 1993) in 305 patients with resectable lung cancer from various countries. All sub-scales, except role

functioning met the minimal standards for reliability. Internal consistency coefficients (Cronbach's alpha) were within 'acceptable' limits (r=0.52-0.89). Furthermore, the questionnaire appeared to be responsive to changes over time.

2.6.3 Summary

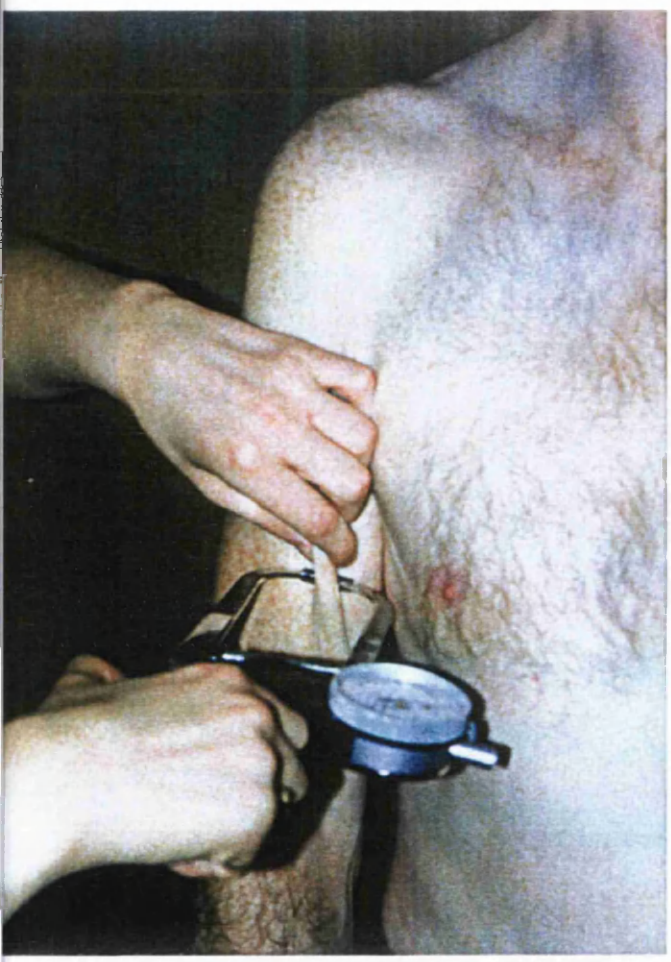
The EORTC-QLQ-C30 was considered to be the most suitable quality of life tools for use in patients with colorectal cancer receiving chemotherapy for the following reasons. The questionnaire was originally developed for use in patients with cancer. It measures commons symptoms which are associated with chemotherapy, which may affect the patients overall quality of life. It has also been reported to be reliable, valid and suitable for use in patients with various types of cancers, across a range of cultural backgrounds. The Karnofsky Performance Index was also used in the forthcoming studies, to complement measurement of the functional status.

2.6.4 Study Methods

The Karnofsky Performance Index was completed by the same oncology nurse prior to the administration of chemotherapy for a total of six cycles.

The EORTC QLQ-C30, (after instruction), was completed by patients immediately prior to their chemotherapy treatment. Guidelines written by the EORTC Study Group were used which detailed the scoring procedure required. Using this system, a score for each functional scale is obtained. The sum of the items incorporated in each separate category were then added, the total divided by the number of questions included in the category and a score obtained. A linear transformation was then conducted which converted this value to a percentage scale. The higher the value, the higher the level of functioning.

The EORTC QLQ-C30 was used under copyright restrictions and permission was requested and granted from the EORTC study group, EORTC Data Centre, Ave. E. Mounier 83, Bte 11, 1200 Brussels, Belgium.



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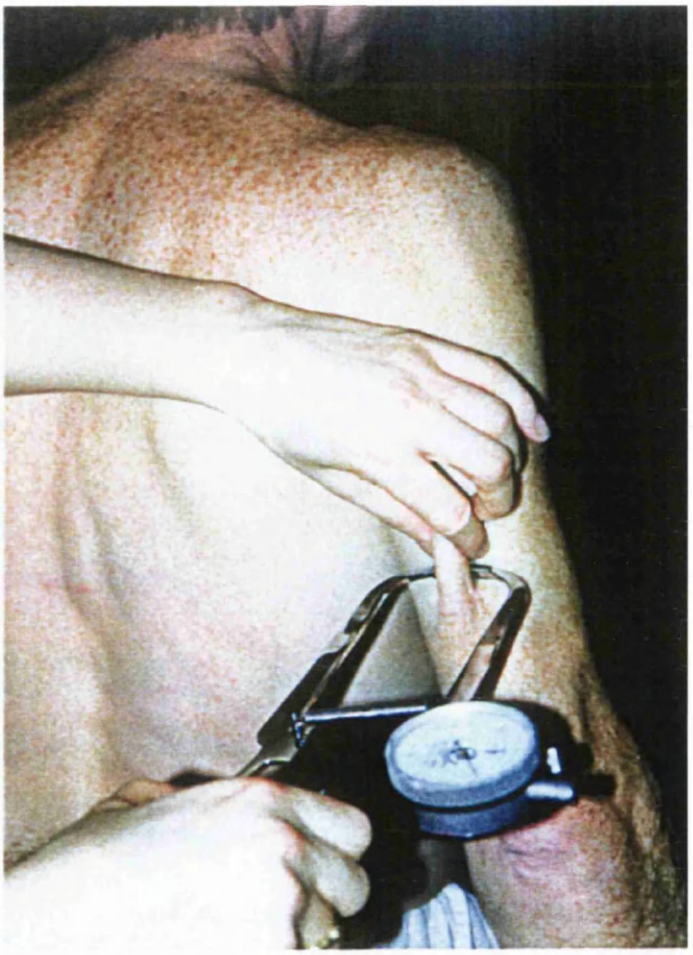

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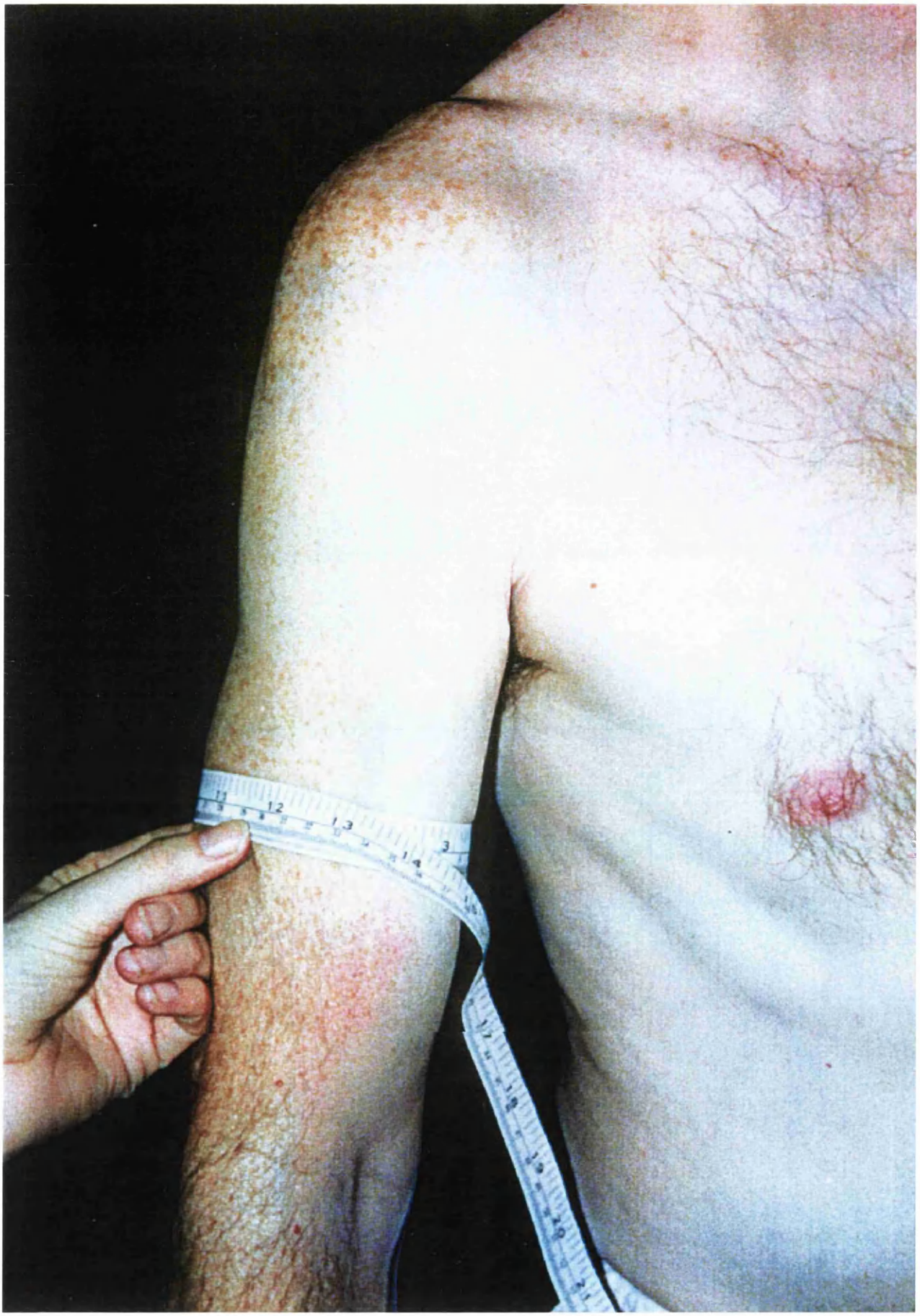

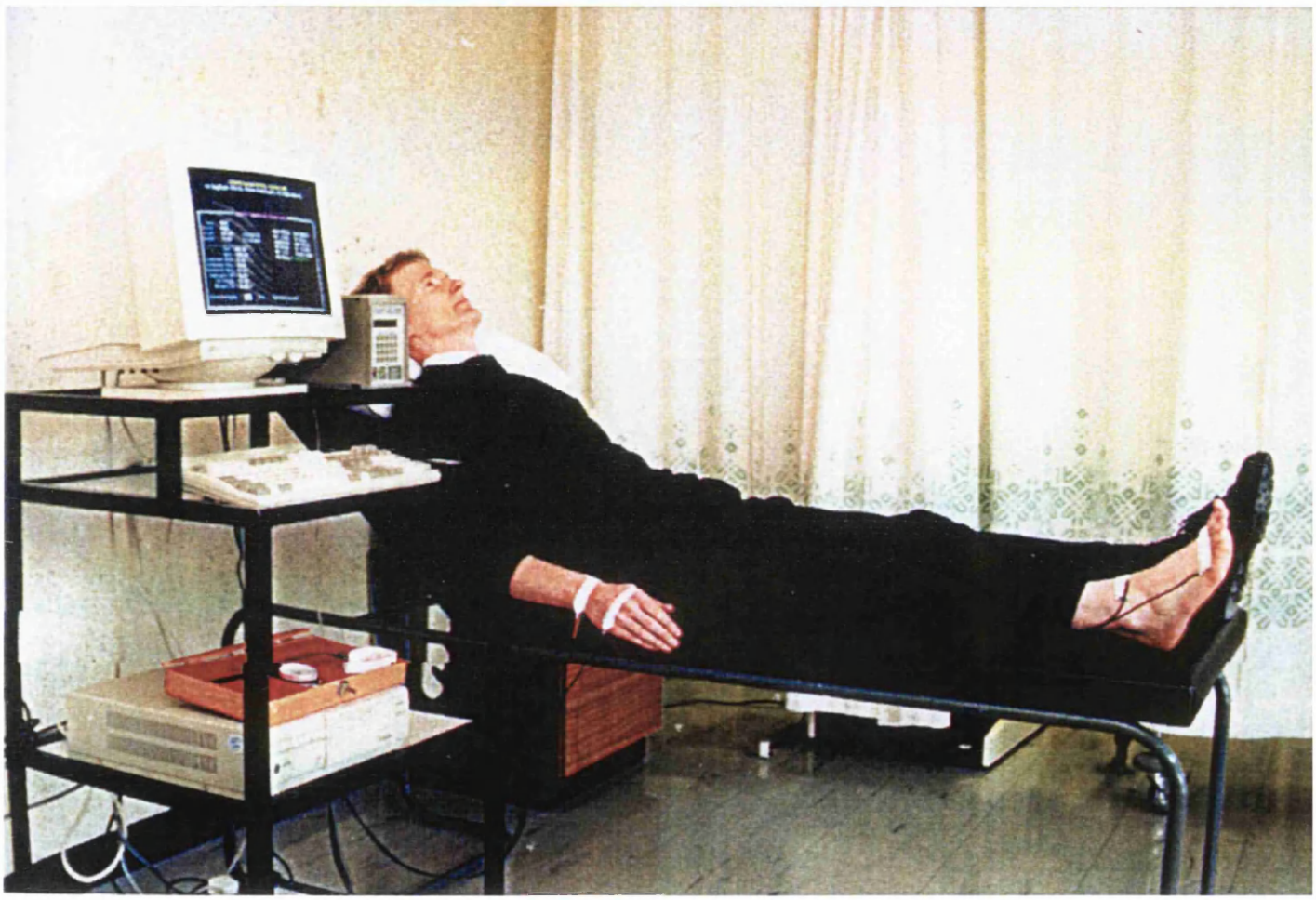

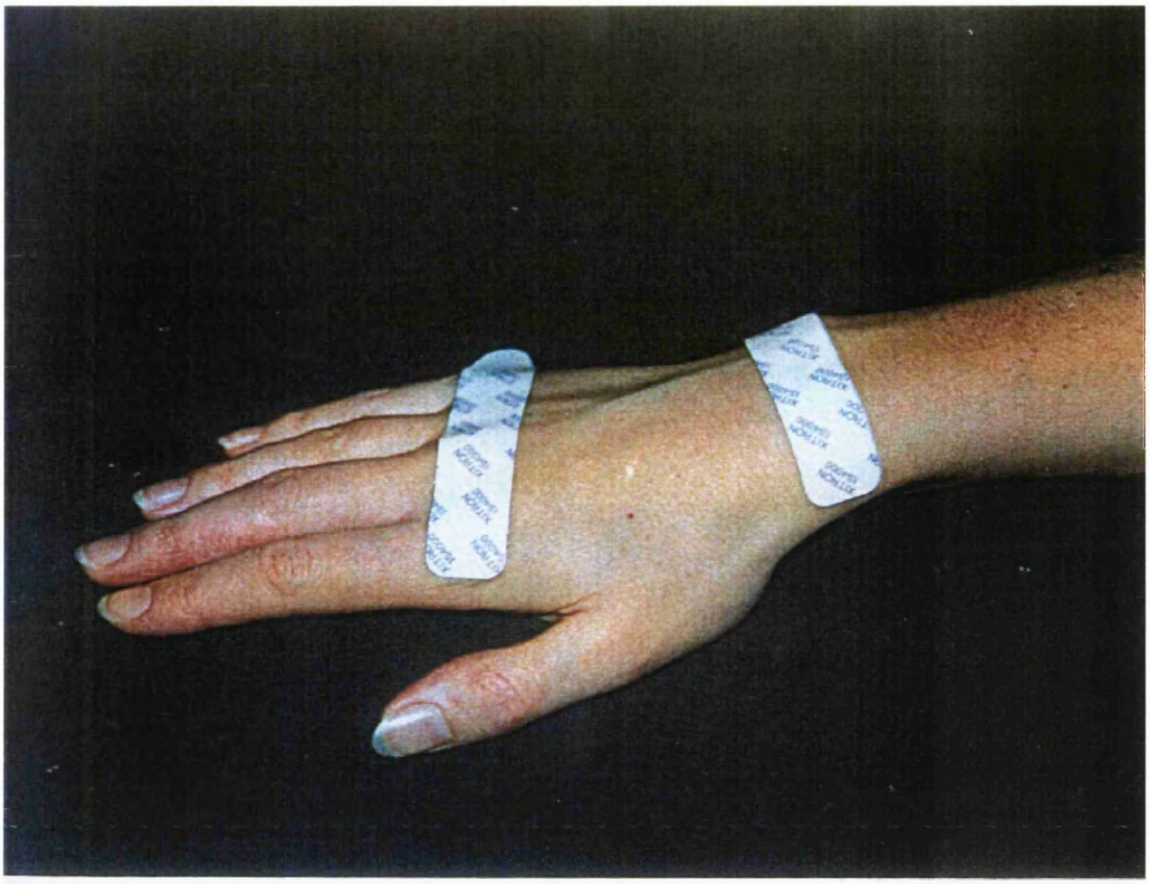

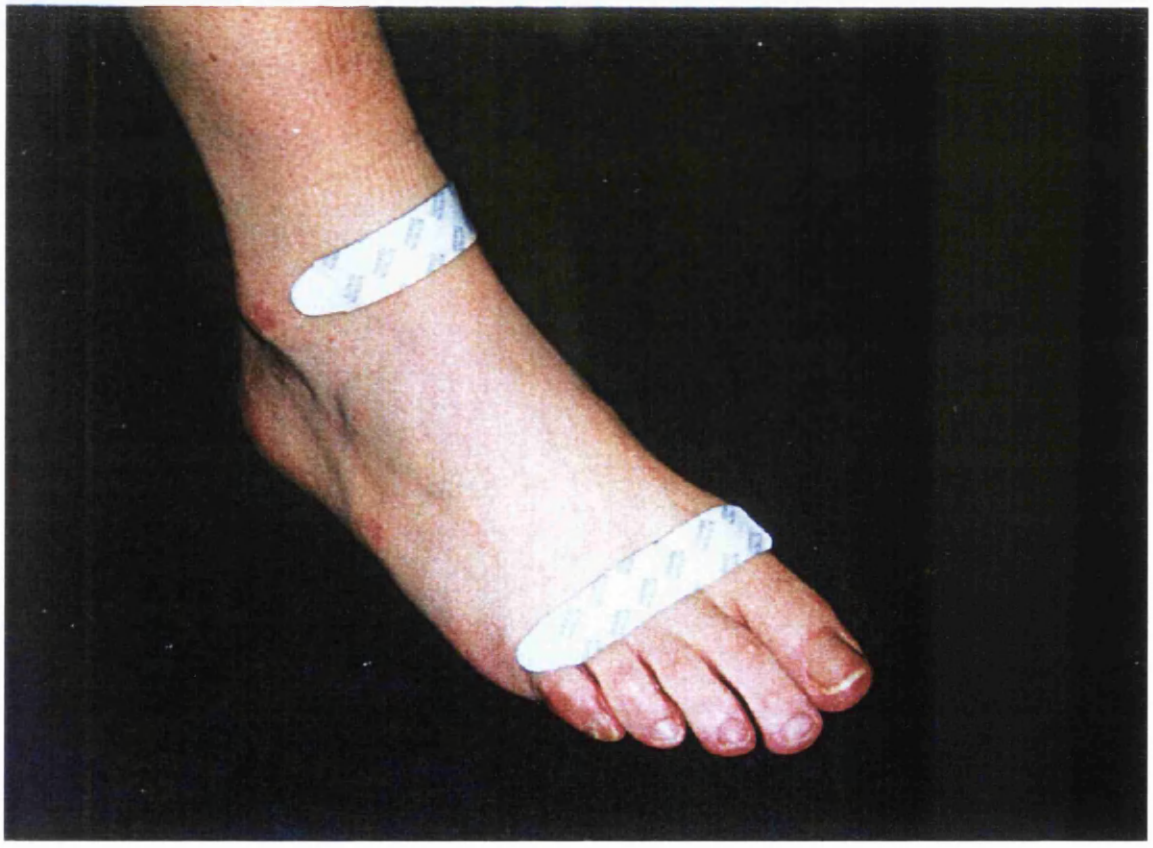

3.0

Longitudinal Study of the Host Metabolic Response to 5-FU Based Chemotherapy in Patients With Colorectal Liver Metastases

3.1 Introduction

Whilst many studies have assessed the safety and efficacy of chemotherapeutic regimens using response rate, toxicity profiles and quality of life (Advanced Colorectal Cancer Meta-Analysis Project, 1992; Dworkin et al., 1995; Meta-Analysis Group In Cancer, 1996; de Gramont et al., 1997; Earlam et al., 1997), few studies have evaluated the effect of chemotherapy on the metabolic response of the host in particular energy balance, body composition and quality of life.

Studies of the effect of chemotherapy on energy metabolism are limited (Jebb et al., 1994; Staal-van den Brekel et al., 1997; Demark-Wahnefried et al., 1997). With regard to the contribution of energy intake to energy balance, few formal assessments of dietary intake in patients receiving chemotherapy appear to have been conducted. Studies of resting energy expenditure in these patients are also limited. It has been proposed that patients with colorectal liver metastases are hypermetabolic. However, studies carried out in such patients have yielded conflicting results (Macfie et al., 1982; Hansell et al,. 1986a; Hansell et al., 1986b; Luketich et al., 1990; Fredrix et al., 1991; Weinmann et al., 1996).

The weight lost in cancer patients comprises of both lean body tissue and body fat (Craig & Waterhouse, 1957; Brennan, 1977; Watson & Sammon, 1980; Cohn et al., 1981; Moley et al., 1987; Preston et al., 1987). In patients with cancer and severe wasting, muscle mass appears to be disproportionately lost compared with that of fat (Cohn et al., 1981). Few studies have assessed the effect of chemotherapy on body composition (Jebb et al., 1994).

The relationship between the acute phase protein response and chemotherapy has been examined by a number of investigators in patients with various cancers (Milroy et al., 1989; Broom et al,. 1992; Simpson et al., 1995; Albuquerque et al., 1995). However, it remains

unclear whether the presence of a systemic inflammatory response in the host affects response to chemotherapy.

The aim of the present study was to examine the effects of 5-FU based chemotherapy on energy balance, body composition, acute phase protein response and quality of life in patients with colorectal liver metastases.

3.2. Materials and Methods

3.2.1 Subjects

Patients presenting with histologically proven colorectal liver metastases, objectively measurable disease, a life expectancy of more than 3 months, ECOG performance status <2 and adequate bone marrow liver, cardiac and renal function were studied. Patients were excluded from study if there was any clinical evidence of infection or the presence of any other cancer.

3.2.2 Experimental Design

Patients undergoing intravenous treatment received folinic acid 350 mg administered over a period of two hours, followed by 5-fluorouracil (400mg/m^2) over 15 minutes and 5-fluorouracil (600mg/ m^2) as a slow infusion over 22hr using an ambulatory pump. The same regime was repeated the following day.

The intra-arterial regime consisted of folinic acid 350 mg administered intravenously over a period of two hours, followed by 5-fluorouracil (400mg/m^2) intra-arterially over 15 minutes and 5-fluorouracil (1600mg/ m^2) as a slow intra-arterial infusion over 22hr using an ambulatory pump. The same regime was repeated the following day. The two regimens were therefore similar except for the difference in the concentration of the intra-arterial infusion.

Treatment was repeated fortnightly for 5 cycles, after which a CT scan was carried out to assess response to chemotherapy. The CT scans were carried out and reported by the same radiologist each time using the same instrumentation and technique. World Health Organisation criteria (Miller et al., 1981), as described in section 2.4, were used to grade the response to treatment as being complete, partial, static or progressive. All patients had 5 pulses of chemotherapy over no more than 10 weeks. Follow-up in this study was carried out until patients had completed their fifth cycle of either intravenous or intra-arterial 5-fluorouracil and folinic acid regimes.

Prior to each cycle of chemotherapy, venous blood samples were taken to measure haematological and biochemical parameters. In a subset of patients who agreed to take part, energy intake, energy expenditure, body composition and quality of life was also measured.

The ethics committee of Glasgow Royal Infirmary approved the study and all patients were informed of the purpose and procedure of the study.

3.2.3 Analytical Methods

Haematological Parameters: White cell count, neutrophil count, lymphocyte count, platelets, haemoglobin, red blood cell count, haematocrit, mean cell volume and mean cell haemoglobin were measured as described in section 2.5.1.

Biochemical Parameters: Circulating concentrations of sodium, potassium, chloride, urea, creatinine, adjusted calcium phosphate, bilirubin, alkaline phosphatase, aspartate transaminase, alanine transaminase, γ-glutamyl transferase, insulin, cortisol, total protein, albumin, globulin, fibrinogen, C-reactive protein and carcinoembryonic antigen (CEA) were measured as described in section 2.5.2.

Energy intake: This was assessed via a four day prospective food diary as described in section 2.2.4, prior to the commencement of each pulse of chemotherapy.

Energy Expenditure: Resting energy expenditure was measured using a ventilated hood system as described in section 2.3.4, prior to the commencement of each pulse of chemotherapy.

Body Composition: This was measured using bio-electrical impedance analysis and anthropometry as described in section 2.1.4.

Quality of Life: This was assessed using the EORTC QLQ-C30 questionnaire and the Karnofsky Performance Index as described in section 2.6.4.

3.2.4 Calculations

Predicted resting energy expenditure (in contrast to measured resting energy expenditure, see section 2.3.4) and predicted total body water (in contrast to measured total body water, see section 2.1.4) were determined using the following formulae and as previously described (Watson et al., 1998).

- Males

Predicted resting energy expenditure (kcal/day)= 66 + 13.8weight + 5.0height – 6.8age

Predicted total body water (l) = 2.447 – 0.09516 age + 0.1074 height + 0.3362 weight

- Females

Predicted resting energy expenditure (kcal/day)= 655 + 9.6weight + 1.8height – 4.7age

Predicted total body water (l) = -2.097 + 0.1069 height + 0.2466 weight

Lean body mass was determined using the following calculations (Cohn et al., 1981):

Lean body mass (kg) = Total body water (l)/ 0.73

3.2.5 Statistics

Data are presented as median and range. Where appropriate, differences over time were tested for statistical significance using the Wilcoxon signed rank test (Minitab Inc., PA, USA).

3.3 Results

Twenty-five patients were included in the study. Characteristics of patients are shown in Table 3.1. All patients were weight stable at the time of entry into the study. Twelve patients received intravenous 5-FU and folinic acid; 13 patients received intra-arterial 5FU and intravenous folinic acid. Two patients had a 20% reduction in dose in cycle 5, two patients had a 20% reduction in cycles 4 and 5 and one patient had a 20% reduction in dose in cycles 3,4 and 5.

After the 5 cycles of treatment 21 patients showed evidence of response or static disease and four had progressed.

Haematological Parameters (Table 3.2)

During the course of the study there was a reduction in white blood cell count ($p<0.01$) neutrophil count ($p<0.0001$) and platelets ($p<0.001$) and an increase in mean cell volume ($p<0.0001$) and mean cell haemoglobin ($p<0.0001$).

Biochemical Parameters (Table 3.3)

During the course of the study there was a reduction in circulating concentrations of potassium ($p<0.05$), the enzymes alkaline phosphatase ($p<0.01$), aspartate transaminase ($p<0.05$), alanine transaminase ($p<0.01$) and gamma GT ($p<0.01$), proteins fibrinogen ($p<0.05$) and C-reactive protein ($p<0.05$). There was an increase in circulating concentrations of urea ($p<0.05$), bilirubin ($p<0.05$) and insulin ($p<0.05$).

Of the 25 patients included in the study, there were 11 patients who agreed to undergo the measurement protocol having measurements of body composition, energy intake, resting energy expenditure and quality of life before and after chemotherapy. The characteristics of this group (Table 3.4) were not significantly different from the 25 patients initially entered into the study (Table 3.1).

During the course of the study there was no significant differences in energy balance (Table 3.5), body composition), (Table 3.6) or quality of life (Table 3.7).

3.4 Discussion

The results of this study indicate that, in patients with colorectal liver metastases receiving 5-FU based chemotherapy, a number of changes occur in both haematological and biochemical parameters.

Chemotherapeutic drugs are designed to reduce tumour growth either by cytotoxicity or cytostasis. However, as yet, the majority of these agents are not specific to malignant cells only, and as a result normal, rapidly dividing cells, such as bone marrow, are likely to be affected (Holmes, 1997; Pannall & Kotasek, 1997).

Mature blood cells, which are no longer undergoing cell division, are not affected by chemotherapy, it is the stem cells that are affected. This results in the inability of the bone marrow to adequately replace depleted blood cells. All types of blood cells are reported to have a predetermined life span (white blood cells-6 hours, platelets-8/10 days, erythrocytes-120days (Holmes, 1997). Due to this the effects of chemotherapy on bone marrow can predicted and the nadir (lowest point reached in the peripheral blood) estimated.

As white blood cells and platelets do not appear to be stored by the body in any significant amount, the effect of cytotoxic bone marrow suppression is normally first observed in the white cells and platelets. This is consistent with the results observed in the study, in which white blood cell count, neutrophil count and platelet count were all observed to decrease from baseline over the 5 cycles of chemotherapy.

Erythrocytes, as they have a life span of 120 days, are usually affected by treatment some weeks later (Holmes, 1997). Anaemia may occur as a result of chemotherapy, however, it is relatively uncommon for it to occur due to chemotherapy alone and the disease process itself often contributes to this state (Holmes, 1997). In the present study, anaemia was not observed

but there was an increase in mean cell volume and mean cell haemoglobin over the 5 cycles of treatment. This effect of 5-FU based chemotherapy does not appear to have been reported. Nevertheless, these changes may be precursors to clinical anaemia developing at a later stage.

The enzymes alkaline phosphatase, aspartate transaminase, alanine transaminase and γ-glutamyl transferase were observed to decrease during the course of chemotherapy. Alanine transaminase has been reported to be more 'liver specific; and the activity of alanine transaminase tends to persist longer than aspartate transaminase activity (Moss et al., 1987). γ-glutamyl transferase is thought to reflect cell turnover in the liver and kidneys (Marshall, 1995). The observed decrease in circulating aminotransferases and γ-glutamyl transferase activity from baseline values, on commencement of chemotherapy, may be due to disease response to the drug regime.

It was of interest that positive acute phase proteins such as fibrinogen fell in concentration during the course of the study while other proteins such as globulins did not. These results are consistent with previous results (Cwikiel et al., 1995) and suggest that 5-FU based chemotherapy results in a reduction in the systemic inflammatory response.

There are few clinical studies that have assessed host parameters such as energy balance before and during chemotherapy. Jebb and colleagues (1994) examined resting energy expenditure and body composition, before and after treatment in patients with small cell lung cancer. In patients who responded to treatment, there were significant reductions in the absolute and the percentage predicted energy expenditure in the absence of any change in body weight or fat-free mass. It was concluded that there was 'evidence for tumour induced hypermetabolism which is independent of changes in gross body composition, although the absolute increase is small' (Jebb et al., 1994). Moreover, as patients who responded to treatment displayed no significant changes in body weight, this implied that either total energy expenditure was decreased from factors such as physical activity or energy intake was increased. Both of these parameters were not formally assessed in this study.

Staal-van den Brekel and colleagues (1997) also assessed the effects of chemotherapy on energy metabolism in patients with small cell lung carcinoma. Furthermore, they also measured systemic levels of inflammatory mediators such as C-reactive protein, lipopolysaccharide-binding protein, soluble TNF receptor and soluble intracellular adhesion molecules. Twelve patients were entered into the study; resting energy expenditure was measured via indirect calorimetry and body composition with bio-electrical impedance analysis. Measurements were conducted prior to and one month after treatment. A significant decrease in energy expenditure expressed both as an absolute value and adjusted for fat-free mass was observed after treatment, irrespective of response to treatment. Energy intake was also measured in six of the twelve patients and this did not appear to significantly change. Body composition and weight also appeared to remain stable. C-reactive protein and lipopolysaccharide-binding protein were both observed to significantly decrease after chemotherapy. However, no correlation could be made between a decrease in energy expenditure and the acute-phase proteins measured. It was concluded that 'chemotherapeutic treatment attenuates the tumour-related metabolic derangements and acute-phase response' (Staal-van den Brekel et al., 1997).

Demark-Wahnefried and co-workers (1997) carried out a study in 18 breast cancer patients receiving adjuvant chemotherapy. Resting energy expenditure, diet-induced thermogenesis, energy intake, physical activity and body composition were measured prior to and during treatment. Resting energy expenditure was observed to decrease significantly mid-treatment and return to pre-treatment values after the completion of treatment. Energy intake and physical activity, were also observed to decrease during treatment. It was concluded that chemotherapy initiates a significant changes in body composition and energy metabolism and should be considered when breast cancer patients are given chemotherapy.

In the present study measured resting energy expenditure values were higher than would be predicted whether expressed as kilocalories per day (median 113%) or per lean body mass derived from total body water (median 120%). There were no significant differences in energy intake or resting energy expenditure during the course of the study.

The effect of chemotherapy on body composition was also examined by Jebb and colleagues (1994). They measured body composition using dual X-ray absorpitometry, prior to and approximately one month after treatment, in 28 newly diagnosed small cell lung cancer patients. Of the 28 patients, 18 responded to treatment. In patients who had responded to treatment, there was no significant change in body weight or gross body composition observed. In the present study where the majority of patients responded to treatment, there were no alterations in any of the body composition parameters. These results are therefore consistent with the observations of Jebb and co-workers (1994)

Quality of life studies have been conducted in patients with colorectal liver metastases receiving 5-FU based chemotherapy. Allen-Mersh and co-workers (1994) reported that hepatic arterial infusion of floxuridine was associated with maintenance of quality of life in patients with colorectal liver metastases.

In the present study global quality of life and karnofsky performance status did not change during the course of the study. These results are consistent with the above study where quality of life was maintained on chemotherapy. There was no change in physical function as measured by performance status and would suggest that the effect of 5-FU based chemotherapy, if any, on host energy metabolism is small.

In the present study we examined host metabolism over a period of 6 cycles of 5-FU chemotherapy (approximately 12 weeks). We chose this period of observation since we were able to ensure that, by the use of CT scans, the disease status of the patients had not changed. This allowed examination the effect of chemotherapy alone on host metabolism. Clearly, the present study can not address effects of chemotherapy beyond this time period and it may be that the relationship of host metabolism and chemotherapy could alter with time.

In summary, in patients with colorectal liver metastases, 5FU chemotherapy is associated with a reduction in white blood cells, liver enzymes and acute phase proteins. In the small sub-group there were no detectable effects on energy metabolism, body composition or quality of life.

Table 3.1

Baseline Characteristics of Patients with Colorectal Liver Metastases

	PATIENTS (N=25)
Sex (F/M)	8/17
Age (years)	59 (30-78)
Weight (kg)	70 (52-103.1)
BMI (kg/m^2)	24.2 (17.7-36.6)
IV/IA Chemotherapy	12/13
	Median (range)

Table 3.2

Haematological parameters in patients before and after 5 cycles of chemotherapy

	BEFORE CHEMOTHERAPY (N=25)	AFTER CHEMOTHERAPY (N=25)	P-VALUE
White cell count (10^9/l)	7.9 (3.9-17.2)	7.1 (3.4-12.1)	0.002
Neutrophil count (10^9/l)	5.2 (2.4-13.3)	4.1 (1.4-8.9)	<0.0001
Lymphocyte count (10^9/l)	1.6 (0.9-3.3)	1.6 (1.0-3.5)	0.492
Platelets (10^9/l)	277 (181-643)	201 (132-404)	<0.0001
Haemoglobulin (g/l)	12.8 (10.0-15.3)	13.0 (10.0-15.3)	0.692
Red blood cell count (10^{12}/l)	4.46 (3.75-5.51)	4.15 (1.75-5.02)	0.028
Haematocrit (l/l)	0.38 (0.30-0.44)	0.37 (0.30-0.42)	0.657
Mean cell volume (fl)	83.3 (74.3-91.4)	85.3 (79.9-97.8)	<0.0001
Mean cell haemoglobin (pg)	28.3 (24.4-32.7)	29.7 (26.4-30.4)	<0.0001
Median (range)			

Table 3.3

Biochemical parameters in patients before and after 5 cycles of chemotherapy

	BEFORE CHEMOTHERAPY (N=25)	AFTER CHEMOTHERAPY (N=25)	P-VALUE
Sodium (mmol/l)	138 (132-144)	138 (133-143)	0.896
Potassium (mmol/l)	4.3 (3.7-5.1)	4.2 (3.3-4.7)	0.011
Chloride (mmol/l)	103 (98-107)	105 (96-111)	0.065
Urea (mmol/l)	4.3 (1.8-6.8)	5.1 (2.1-8.2)	0.033
Creatinine (umol/l)	90 (63-112)	90 (55-126)	0.058
Adjusted Calcium (mmol/l)	2.45 (2.15-2.64)	2.40 (2.30-3.05)	0.648
Phosphate (mmol/l)	1.18 (0.65-1.40)	1.13 (0.75-1.35)	0.355
Bilirubin (umol/l)	12.0 (7.0-47.0)	14.5 (10.0-39.0)	0.040
Alkaline phosphatase (U/l)	715 (140-2490)	468 (165-1580)	0.001
Aspartate transaminase (U/l)	31 (19-118)	30 (15-76)	0.045
Alanine transaminase (U/l)	34 (10-171)	26 (9-104)	0.003
Gamma GT (U/l)	205 (20-750)	75 (19-440)	0.003
Insulin (mU/l)	6.70 (2.10-12.20)	14.1 (2.4-18.4)	0.018
Cortisol (ng/l)	460 (240-1040)	444 (30-850)	0.515
Total protein (g/l)	73 (58-81)	71 (61-82)	0.065
Albumin (g/l)	42 (32-49)	42 (29-49)	0.200
Globulin (g/l)	31 (23-41)	30 (12-53)	0.138
Fibrinogen (g/l)	4.0 (2.3-5.2)	2.4 (2.1-4.8)	0.015
C-reactive protein (mg/l)	22 (<5-163)	<5 (<5-183)	0.041
CEA (U/L)	1243 (21-22,620)	1028 (26-27,740)	0.480
Median (range)			

Table 3.4

Baseline characteristics of patients with measurements of body composition, energy metabolism and quality of Life

	PATIENTS (N=11)
Sex (F/M)	2/9
Age (years)	64 (48-76)
Weight (kg)	70.7 (60.8-103.0)
BMI (kg/m^2)	23.8 (19.0-36.5)
IV/IA Chemotherapy	5/6
	Median (range)

Table 3.5

Energy Expenditure before and after chemotherapy

	BEFORE CHEMOTHERAPY (N=11)	AFTER CHEMOTHERAPY (N=11)	P-VALUE
Energy intake (kcal/day)#	1584 (1269-1890)	1814 (1222-2503)	0.225
Resting energy expenditure (kcal/d)	1650 (1319-2627)	1691 (1353-2549)	0.248
Resting energy expenditure (kcal/kg/d)	22.8 (18.4-34.2)	22.3 (17.9-35.9)	0.139
Resting energy expenditure (kcal/kgLBM*/d)	34.6 (28.8-49.6)	32.3 (28.4-55.7)	0.374
Respiratory quotient	0.79 (0.72-0.92)	0.78 (0.72-0.92)	0.624

Median (range), # n=6, * derived from total body water estimate

Table 3.6

Body composition before and after chemotherapy

	BEFORE CHEMOTHERAPY (N==11)	AFTER CHEMOTHERAPY (N=11)	P-VALUE
Weight (kg)	70.7 (60.8-103.0)	74.4 (59.9-99.5)	0.534
BMI	23.8 (19.0-36.5)	24.3 (22.2-35.2)	0.534
Biceps skinfold thickness (mm)	5.3 (3.0-9.3)	5.5 (3.0-10.2)	0.929
Triceps skinfold thickness (mm)	11.0 (7.0-28.5)	11.6 (7.6-29.2)	0.169
Mid-upper arm circumference (cm)	26.5 (23.0-34.5)	27.3 (23.0-32.4)	0.919
Intracellular Water (l)	15.6 (11.8-21.6)	15.9 (12.5-19.5)	0.859
Total Body Water (l)	35.4 (26.6-48.0)	36.0 (27.3-45.6)	0.374
Lean body mass (kg)	48.3 (36.4-65.6)	49.2 (37.2-62.3)	0.374
Body Fat mass (kg)	24.3 (16.5-36.4)	22.7 (17.8-38.5)	0.515

Median (range)

Table 3.7

Performance status, appetite and quality of life before and after chemotherapy

	BEFORE CHEMOTHERAPY (N=11)	AFTER CHEMOTHERAPY (N=11)	P-VALUE
Karnofsky performance status	80 (60-100)	80 (80-100)	0.096
Appetite	7.0 (0.7-10.0)	8.2 (3.4-9.9)	0.533
EORTC QLQ C-30			
Physical functioning	60 (20-80)	60 (20-100)	0.221
Role functioning	83.3 (83.3-100)	83.3 (66.7-100)	1.000
Emotional functioning	75 (50-100)	75 (0-100)	0.337
Cognitive functioning	83.3 (66.7-100)	100 (33.3-100)	0.785
Social functioning	66.7 (16.7-100)	66.7 (16.7-100)	0.314
Quality of life	58.3 (33.3-66.7)	66.7 (16.7-75)	0.396
Fatigue	43.3 (22-66.7)	33.3 (11-100)	0.159
Nausea and vomiting	0 (0-33.3)	0 (0-83.3)	0.257
Pain	16.7 (0-33.3)	0 (0-33.3)	0.725
Dyspnoea	33.3 (0-33.3)	33.3 (0-33.3)	1.000
Sleep disturbance	33.3 (0-66.7)	33.3 (0-100)	0.660
Appetite loss	33.3 (0-66.7)	0 (0-100)	0.470
Constipation	0 (0-66.7)	0 (0-33.3)	0.257
Diarrhoea	0 (0-33.3)	0 (0-100)	0.059
Financial difficulty	0 (0-33.3)	0 (0-33.3)	0.317

4.0

Study of Biochemical and Haematological Parameters in Disease Progression of Patients with Colorectal Liver Metastases Receiving 5-FU Based Chemotherapy

4.1 Introduction

The use of 5-FU based chemotherapy for metastatic colorectal cancer is well established. This may be administered intravenously or intra-arterially (sections 1.4.2.3, 1.5).

Currently, response to such chemotherapeutic regimes is usually assessed using computed tomography at 3 monthly intervals. If a patient responds to treatment and has acceptable 'toxicity profiles', then the same type of treatment is usually continued. Alternatively, if a patient fails to respond to treatment they are commonly offered second line drugs or best supportive care.

However, it may be that there are changes in the tumour/ host response, prior to CT defined disease progression, which would alert the physician of disease progression. This would enable intervention at an earlier stage. Studies assessing the tumour/host response in patients with advanced colorectal cancer receiving chemotherapy are limited. For example, Grem and colleagues (1998), measured CEA (a tumour marker) concentration in 125 patients prior to and during 5FU based chemotherapy. They concluded that a consistent rise in CEA concentrations signals the need to reassess the patient's disease status.

Few host markers have been assessed in this context. It has been reported that the presence and magnitude of the systemic inflammatory response (as evidenced by circulating concentrations of C-reactive protein) is associated with the progression of colorectal cancer (McMillan et al., 1995).

The aims of the present study were to:

- Examine which host factors were altered on disease progression in patients with colorectal liver metastases.
- Examine if these identified factors altered prior to CT defined progression in patients with colorectal liver metastases.

4.2 Materials and Methods

4.2.1 Subjects

Patients presenting with histologically proven colorectal liver metastases, objectively measurable disease, a life expectancy of more than 3 months, ECOG performance status <2 and adequate bone marrow liver, cardiac and renal function were studied. Patients were excluded from study if there was any clinical evidence of infection or the presence of any other cancer.

Thirteen patients received intra-arterial 5-FU in combination with intravenous folinic acid and a further 12 patients received the same drugs intravenously.

4.2.2 Experimental Design

Patients undergoing intravenous treatment received folinic acid 350 mg administered over a period of two hours, followed by 5-fluorouracil (400mg/m^2) over 15 minutes and 5-fluorouracil (600mg/ m^2) as a slow infusion over 22hr using an ambulatory pump (Howell, JD et al, 1997). The same regimen was repeated the following day.

The intra-arterial regime consisted of folinic acid 350 mg administered intravenously over a period of two hours, followed by 5-fluorouracil (400mg/m^2) intra-arterially over 15 minutes and 5-fluorouracil (1600mg/ m^2) as a slow intra-arterial infusion over 22hr using an ambulatory pump. The same regimen was repeated the following day. The two regimen were therefore similar except for the difference in the concentration of the intra-arterial infusion.

Treatment was repeated two weekly for 5 cycles, after which a CT scan was carried out to assess response to chemotherapy. The CT scans were carried out and reported by the same radiologist each time using the same instrumentation and technique. World Health Organisation criteria (Miller et al. 1981), as described in section 2.4, were used to grade the response to treatment (complete, partial, static or progressive). Patients with static, partial or

complete responses were defined as having 'responding disease'. If this was associated with 'acceptable' toxicity profiles, treatment was continued for a further 6 cycles. Patients considered to have progressive disease either had best supportive care or were given second-line chemotherapy. Follow-up was continued until patients progressed either on intravenous or intra-arterial chemotherapy.

Prior to each cycle of chemotherapy, venous blood samples were taken to measure haematological and biochemical parameters. (as described in section 2.5).

In order to identify which host factors were altered on disease progression in patients with colorectal liver metastases, data from patients during a period of 'responding disease' (at 3 months) were compared to data from the same patients with a period of 'disease progression' (at least 6 months later). Disease status whether it was 'responding' or 'progressive' was defined using computed tomography scanning. All computed tomography scans were carried out by the same radiologist, using the same technique, same instrumentation and reported using World Health Organisation criteria.

The ethics committee of Glasgow Royal Infirmary approved the study and all patients were informed of the purpose and procedure of the study.

4.2.3 Statistics

Data are presented as median and range. Where appropriate, analysis between different disease states was performed using the Wilcoxon signed rank test, (Minitab Inc., PA, USA). Differences over time, where appropriate, were tested for statistical significance by analysis of variance using the Friedman rank test.

4.3 Results

Of the 25 patients assessed using the above criteria (Table 4.1), the data from 11 patients (Table 4.2) were used in the analysis. The remaining 14 patients were excluded due to the following factors. Three patients had a hepatic resection, three patients did not have disease progression, two patients died during periods of 'responding' disease and six patients did not have 'responding' disease.

The biochemical and haematological values of 11 patients were included in the analysis. Variables which changed significantly during progression compared with response included white cell count ($p<0.01$), neutrophil count ($p<0.01$), creatinine ($p<0.05$), alkaline phosphatase ($p<0.01$), alanine transaminase ($p<0.05$), aspartate transaminase ($P<0.01$), C-reactive protein ($p<0.05$) and carcinoembryonic antigen ($p=<0.01$) (Table 4.3).

These factors were then examined in order to determine whether such changes occurred prior to CT-defined disease progression. Of the 25 patients entered into the study, 14 patients were included in the analysis. Eleven patients were excluded as three patients had a hepatic resection, three patients did not have disease progression, two patient died during periods of 'responding disease', and three patients developed late progression and therefore were not included in the analysis. Data from four specific time points were used for this analysis; 4-6 weeks prior to disease progression, 8-10 weeks prior to disease progression and 12-14 weeks prior to disease progression.

Significant factors which predicted progression before CT were white cell count ($p \leq 0.05$), alkaline phosphatase ($p \leq 0.001$), C-reactive protein ($p \leq 0.05$), carcinoembryonic antigen ($p \leq 0.05$), and alanine transaminase ($p \leq 0.05$) (Table 4.4).

4.4 Discussion

Despite the small numbers of patients entered into the present studies, significant differences in a number of biochemical and haematological parameters were identified.

On comparison of these parameters during periods of 'responding' and 'progressive' disease, eight significant differences were identified. White cell count, neutrophil count, creatinine, alkaline phosphatase, alanine transaminase, aspartate transferase, C-reactive protein and carcinoembryonic antigen were all observed to be significantly different on disease progression. Furthermore, when these identified factors were examined longitudinally prior to CT-defined progression, white cell count, alkaline phosphatase, C-reactive protein, carcinoembryonic antigen, and alanine transaminase were all identified as being significantly different at various time points in comparison to the time of disease progression.

Some of these parameters have been previously reported to be altered in patients with malignancy, in particular, metastatic liver disease. Alkaline phosphatase is an enzyme which exists in all body tissues either at or in cell membranes (Moss et al., 1987). It is present in particularly high levels within the liver, epithelium of the intestine, bone, placenta and kidney tubules (Moss et al., 1987; Marshall, 1995). Serum alkaline phosphatase is thought to originate from the liver, biliary tract and skeleton (Moss et al., 1987). The actual function of this enzyme is as yet unknown, although it is thought to be involved in the calcification process in bone (Daly & Thom, 1988). Increased hepatic synthesis of alkaline phosphatase is known to occur during both intra and extrahepatic bleary obstruction. Parenchymal liver disease is also known to cause elevations in serum alkaline phosphatase (Moss et al., 1987). Causes of increased plasma alkaline phosphatase in malignant disease are commonly due to bony/hepatic primary or secondary tumours (Marshall, 1995). Tumours may also produce a form of alkaline phosphatase which are thought to be modified forms of nonplacental isoenzymes (Moss et al., 1987).

The transaminases are a group of enzymes that act as catalysts during the interconversions of amino acids and *a*-oxoacids via transfer of amino groups (Moss et al., 1987). Aspartate transaminase and alanine transaminase are both extensively distributed in tissue throughout

the body, and are present in equal amounts in the liver (Moss et al., 1987; Marshall, 1995). Levels of these enzymes are believed to reflect the amount of cellular damage (Marshall, 1995). In liver diseases with associated hepatic necrosis, both aspartate transaminase and alanine transaminase are commonly elevated, even before clinical signs and symptoms of the disease are evident (Moss et al., 1987). In patients with metastatic liver disease, five to ten-fold elevations in both enzymes may occur, with aspartate transaminase usually being present in higher levels (Moss et al., 1987; Marshall, 1995).

The presence of an acute-phase response, commonly identified with increased plasma levels of the acute-phase protein, C-reactive protein, has been reported in a number of patients with various malignancies. This response is believed to be mediated by the release of cytokines, which originate from host tissue and in some instances, by the tumour itself (section 1.7).

Carcinoembryonic antigen is normally present on the apical surface of the mucosal cell. Increases in concentration arise when it travels up the gland crypt of the cell and is lost in faeces. Only a small amount actually enters the blood supply. In patients with colorectal tumours, the changes in normal architecture of the cell diverts the secretion of carcinoembryonic antigen into faeces and redirects excretion into the blood stream and increases its usefulness as a tumour marker (Pannall & Kotasek, 1997). Carcinoembryonic antigen is also recognised as being elevated in 80-100% of patients with colorectal liver metastases (Marshall, 1995).

Overall, considering the results from the previous studies, the following observations were regarded as being of interest. On comparison of parameters during a period of 'responding' disease and 'progressive' disease, an increase in white cell count, neutrophil count and C-reactive protein on disease progression was observed. With regard to longitudinal assessment, white cell count, C-reactive protein and carcinoembryonic antigen were found to significantly change prior to CT-defined disease progression.

Therefore, the results of the present study suggest that 12-14 weeks prior to CT-defined disease progression there may be tumour growth (as indicated by Carcinoembryonic antigen

levels) which may cause cell damage (as indicated by the transaminases) and necrosis, resulting in systemic inflammation, which in turn results in changes in liver protein production (C-reactive protein). There are increasing numbers of reports that suggest that this tumour-host inflammatory response is detrimental to the patient and may indeed promote disease progression in colorectal cancer (McMillan et al., 1995; Nozoe et al., 1998; Nielsen et al., 2000). Since these events can be identified prior to CT-defined progression, markers of the acute-phase response such as white cell count and C-reactive protein, used in combination with carcinoembryonic antigen, may be useful in providing an 'early warning' of tumour progression in patients with colorectal liver metastases. However, such 'markers' of disease progression should only be considered in the absence of any signs or symptoms of sepsis, since this may cause a non-cancer rise in acute-phase proteins, and therefore affect the value of these inflammatory markers.

From the above it may be speculated that CEA in conjunction with markers of the acute-phase reponse, may improve the usefulness of CEA in predicting disease progression in this patient group. Indeed, there is some evidence that this approach is useful in primary colorectal cancer (Stamatiadis et al., 1992).

Table 4.1 **Characteristics of patients with colorectal liver metastases**

	Patients (n=25)
Sex (F/M)	8/17
Age (years)	59 (30-78)
Height (cm)	170 (158-187)
BMI (kg/m^2)	24.2 (17.7-36.6)
IV/IA	12/13
	Median (range)

Table 4.2 Characteristics of Patients with colorectal liver metastases and disease progression

	Patients (n=11)
Sex (F/M)	2/9
Age (years)	57 (30-71)
Height (cm)	173 (158-187)
BMI (kg/m^2)	23.6 (17.7—27.1)
IV/IA	4/7
	Median (range)

Table 4.3 Significant factors on comparison of 'Responsive' with 'Progressive' Disease.

Variable	Responsive Disease (n=11)	Progressive Disease (n=11)
White Cell Count (10^9/l)	5.5 (3.7-7.5)	7.2 (4.5-11.4)**
Neutrophil Count (10^9/l)	3.2 (1.8-4.9)	5.0 (2.5-8.8)**
Creatinine (umol/l)	88 (67-126)	84 (60-130)*
Alkaline Phosphatase (U/l)	580 (200-2260)	1020 (285-2260)**
Alanine Transaminase (U/l)	27 (18-110)	56 (26-137)***
Aspartate Transaminase (U/l)	34 (24-143)	84 (33-143)**
C-Reactive Protein (mg/l)	8 (0-126)	34 (0-175)*
Carcinoembryonic Antigen (U/l)	737 (19-49330)	4000 (26-72000)**
	Median (Range)	Median (Range)

*= $p \leq 0.05$, **= $p \leq 0.01$, *** = $p \leq 0.001$

Table 4.4 Haematological and Biochemical Parameters Prior to Disease Progression

Variable	Progression -12/14 weeks (n=14)	Progression -8/10 weeks (n=14)	Progression -4/6 weeks (n=14)	Progression (n=14)
White Cell Count (10^9/l)	7.4 (3.3-11.9)	5.7 (4.3-9.7) a*	7.6 (3-12.5)	8.2 (4.5-14.6)
Alkaline Phosphatase (U/l)	610 (245-1780)***	580 (250-1770)**	630 (250-2030)	1000 (305-2260)
C-reactive Protein (mg/l)	0 (0-78)*	8 (0-36)a*	24 (0-123)	24 (0-89)
CEA (U/l)	1027 (67-17098)**	1770 (76-21100)	1641 (97-58350)	2432 (137-72000)
Alanine transaminase (U/l)	22.5 (11-97)*	30.5 (13-180)	32.5 (11-176)	36 (18-137)

Median (range)

* = $p \leq 0.05$, ** = $p \leq 0.01$, *** = $p \leq 0.001$ compared with progression

Chapter 5

Longitudinal study of the relationship between leptin and energy metabolism, fat mass and appetite in patients with colorectal liver metastases undergoing 5-FU based chemotherapy.

5.1 Introduction

From the previous study chapters it is clear that over a course of 5-FU based chemotherapy (5 cycles) the effect on gross measures host energy metabolism is small. The effect of individual cycles of chemotherapy on the control mechanisms of host energy metabolism is less clear.

There has been extensive investigation into the protein metabolism of cancer patients undergoing 5-FU based chemotherapy (Schmoll et al., 1999). In contrast, there is less information available on fat metabolism in such patients.

In the past, adipose tissue in cancer patients has been considered difficult to estimate and relatively inactive and passive in the disease process. It is only comparatively recently that it has been demonstrated that adipose tissue participates actively in energy regulation, through a network of endocrine, paracrine and autocrine signals (Kemp et al., 1995).

With the recent discovery of leptin, an adipocyte secreted protein that regulates body weight in animals (Zhang et al., 1994; Pellymounter et al., 1994; Pellymounter et al., 1995), research into and our knowledge of appetite control mechanisms have increased greatly over the last few years. Recent evidence, in animals, suggests that circulating leptin concentrations accurately reflect adipose mass and regulate energy intake (primarily through appetite) and expenditure (primarily through substrate oxidation) through a control feedback loop involving neuropeptide Y (Stephens et al., 1995).

Information on the role of leptin in the energy balance of patients with cancer is limited. Recently, Simons and colleagues (1997) reported low or undetectable circulating leptin concentrations in patients with lung cancer and weight loss. These observations have been extended to weight losing gastrointestinal cancer patients, demonstrating that although circulating leptin concentrations are low, they are appropriate for the low percentage body fat

of these patients (Wallace et al., 1998). Indeed in both studies (Simons et al., 1997; Wallace et al., 1998) it appeared that cancer patients with detectable leptin tended to be less underweight and had a higher fat mass than those in whom leptin was undetectable. If the work in animals were to translate into the human situation this should result in improved appetite and decreased resting energy expenditure. However, it is well recognised that, in the main, appetite is decreased (O'Gorman et al., 1998) and resting energy expenditure is increased (Hansell et al., 1986a) in such weight-losing cancer patients.

To date studies in cancer patients have been conducted in those with weight loss. Therefore, the effects of the tumour and weight loss cannot be readily resolved. Moreover, no studies have assessed this relationship over time.

The aim of the present study was to examine the temporal relationship between leptin, appetite, energy metabolism and body fat mass in a group of advanced cancer patients without weight loss.

5.2 Materials and Methods

5.2.1 Subjects

Sixteen patients with histologically proven colorectal liver metastases were studied. Patients were required to have objectively measurable disease, adequate bone marrow function and a life expectancy of more than 3 months. Nine patients received intra-arterial 5-FU in combination with intravenous folinic acid and a further seven patients received the same drugs intravenously.

5.2.2 Experimental Design

Patients undergoing intravenous treatment received folinic acid 350 mg administered over a period of two hours, followed by 5-fluorouracil (400mg/m^2) over 15 minutes and 5-fluorouracil (600mg/m^2) as a slow infusion over 22hr using an ambulatory pump (Howell et al, 1997). The same regimen was repeated the following day.

The intra-arterial regime consisted of folinic acid 350 mg administered intravenously over a period of two hours, followed by 5-fluorouracil (400mg/m^2) intra-arterially over 15 minutes and 5-fluorouracil (1600mg/m^2) as a slow intra-arterial infusion over 22hr using an ambulatory pump. The same regimen was repeated the following day. The two regimens were therefore similar except for the difference in the concentration of the intra-arterial infusion.

The patients were studied two weekly for 3 cycles. Prior to each cycle of chemotherapy, venous blood samples were taken for the measurement of circulating concentrations of leptin, insulin, cortisol, C-reactive protein and albumin. Total body water was also assessed and the percentage body fat was estimated. Energy intake and resting energy expenditure were also measured. No dietary instructions were given and there were no food restrictions except that patients underwent an overnight fast prior to measurements.

Leptin and percentage body fat were also measured in age and sex matched normal healthy volunteers.

The study was approved by the Research Ethics Committee of Glasgow Royal Infirmary University NHS trust. All subjects were informed of the purpose and procedure of the study and gave written consent.

5.2.3 Methods

Appetite scores

The patients were asked at each assessment to record their appetite using a 10cm visual analogue scale, ranging from poor appetite to good appetite.

Insulin

This was estimated using an ALPCO Insulin ELISA Assay.

Cortisol

This was measured by immunoassay using a heterogeneous competitive magnetic separation assay (Bayer Immuno-1 Analyser, Bayer Diagnostics Division, Newbury, Berkshire, UK). The intra- and inter-assay coefficients of variation were 4% and 9%, respectively, over the sample concentration range.

Leptin

Serum leptin was measured by a radioimmunoassay using a sheep antibody raised against human recombinant leptin and an iodinated human leptin tracer as described in section 2.5.2.

Body Composition

Body fat was calculated using bioelectrical impedance analysis as described in section 2.1.4.

Energy Intake

This was estimated using a 4 day prospective food diary as described in section 2.2.4.

Resting Energy Expenditure

This was measured via indirect calorimetry via a ventilated hood system as described in section 2.3.4.

5.2.4 Statistics

Data are presented as median and range. Where appropriate, analysis between different disease states was performed using the Wilcoxon signed rank test, (Minitab Inc., PA, USA). Differences over time, where appropriate, were tested for statistical significance by analysis of variance using the Friedman rank test.

5.3 Results

The characteristics of the normal subjects and cancer patients are given in Table 5.1. The groups were similar in terms of age, sex, BMI, and percentage body fat mass. Overall, there was a significant correlation between circulating leptin concentrations and percentage body fat ($r= 0.709$, $p<0.0001$). However, circulating concentrations of leptin were lower in the cancer group ($p<0.05$)

In the cancer patients measured body fat was significantly greater than predicted body fat ($p<0.05$) which was calculated. Furthermore, circulating leptin concentrations were significantly correlated with measured ($r= 0.563$, $p<0.05$) but not predicted ($r= 0.446$, $p=0.083$) body fat mass.

The temporal relationship between the circulating factors, energy intake, energy expenditure and the measured fat mass in the cancer group is shown in Table 5.2. Each patient had between 3 and 5 observations. The cancer group were weight stable with no changes in body composition, appetite, energy intake, resting energy expenditure, respiratory quotient or the circulating concentrations of leptin, insulin, cortisol, C-reactive protein and albumin over the period of observation.

Within each patient, the last recorded measurements were compared with baseline. The median changes in the circulating concentrations of leptin were not associated with changes in appetite, energy intake, resting energy expenditure, respiratory quotient, insulin or cortisol. However, they were correlated with median changes in percentage body fat mass ($r=0.682$, $p=0.005$).

5.4 Discussion

In the present study the baseline leptin concentrations of normal subjects and cancer patients were significantly correlated with measured body fat mass. This is consistent with previous measurements of circulating leptin concentrations in weight-losing cancer patients which reported that leptin concentrations were appropriate for predicted body fat mass (Simons et al., 1997; Wallace et al., 1998). However, in the present study where controls were matched for weight and percentage body fat mass circulating leptin concentrations were lower in the cancer group. The basis of the difference between fat mass and the corresponding leptin concentration in normal subjects and cancer patients is not clear and merits further investigation.

Over the period of observation, patients were weight stable and there were no alterations in body fat mass and skinfold thickness. This enabled us to examine the temporal effect of the tumour on host leptin metabolism independent of weight loss. The changes in circulating leptin concentrations were not associated with alterations in appetite, energy intake, resting energy expenditure or respiratory quotient.

This would appear to contradict earlier work in normal subjects (Larsson et al., 1998). However, it is of interest that there is accumulating evidence in normal subjects that would suggest that circulating concentrations of leptin have less influence on appetite, energy intake, resting energy expenditure and fat oxidation than was previously supposed (Coleman & Herrmann, 1999). There is also a lack of association between leptin concentrations and appetite in other disease states such as anorexia nervosa (Stoving et al., 1999) and acquired immunodeficiency syndrome (Grunfeld et al., 1996).

It has been reported that the hormones insulin and cortisol have regulatory effects on circulating concentrations of leptin (Coleman & Herrmann, 1999). However, due to supraphysiological doses used in previous studies their relevance to the normal leptin metabolism is not clear. In the present study it was of interest that there was no association between the concentrations of these hormones or their ratio and leptin concentrations. Taken

together these results indicate that the control of energy balance in humans is more complex than previously thought.

It may be that appetite stimulation and the other central effects of leptin are blocked or overridden by other, as yet unknown, mechanisms resulting in unopposed weight loss. For example, Mantovani and co-workers (2000) reported an inverse correlation between circulating concentrations of leptin and the pro-inflammatory cytokine interleukin-6 suggesting that the host derived factors may over-ride the normal leptin feedback mechanism.

It is of interest that in the present study, changes in body fat mass of cancer patients over time were also correlated with leptin concentrations. These results therefore suggest that circulating leptin concentrations accurately reflect fat mass over time and that any effect of cancer on systemic concentrations is likely to be small..

In the present study, biceps and triceps skinfold thicknesses did not correlate with measured body fat mass. Given that bioelectrical impedance accurately measures fat mass in cancer patients (Simons et al., 1999), the results would suggest that circulating leptin concentrations are a more robust indicator of fat mass than anthropometry in patients with cancer and may have a role in assessing the nutritional status of patients with cancer.

In summary, the role of leptin in the regulation of energy balance in cancer patients is more complex than previously thought. It appears to be similar to that reported in normal subjects. This would indicate that the presence of cancer per se does not greatly alter circulating leptin concentrations. Changes in circulating leptin concentrations reflected changes in body fat mass and suggest that leptin concentration may be a useful routine marker of nutritional status in cancer patients.

Table 5.1

Baseline characteristics of normal subjects and patients with colorectal liver metastases.

	Normal subjects (n=9)	Cancer patients (n=16)	p-value
Sex (m/ f)	7/ 2	14/ 2	
Age (yrs)	65 (46-74)	61 (30-78)	0.677
BMI (kg/m^2)	25.2 (22.0-28.9)	24.7 (22.3-36.5)	0.760
Body fat mass (%)	33.0 (13.9-49.9)	33.4 (15.2-36.1)	0.760
Leptin (ng/l)	10.9 (0.6-31.2)	5.5 (3.3-13.4)	0.037
IV/IA Chemotherapy		7/ 9	

median (range)

Table5 2. Temporal measurements of leptin, appetite, energy metabolism and body fat mass.

	Baseline (n=16)	2 weeks (n=16)	4 weeks (n=16)	6 weeks (n=14)
Weight (kg)	75.4 (59.6-103)	75.8 (59.0-100.1)	75.8 (59.9-100.1)	73.9 (59.4-99.5)
BMI (kg/m^2)	24.7 (22.3-36.5)	25.1 (22.2-35.5)	25.1 (22.5-35.5))	25.4 (22.4-35.3)
Leptin (ng/l)	5.5 (3.3-13.4)	5.6 (2.5-12.9)	6.5 (2.7-15.4)	6.3 (2.5-15.2)
Insulin (mIU/l)	12.0 (2.1-41.0)	11.7 (1.3-27.7)	12.7 (4.4-48.9)	14.7 (3.1-36.6)
Cortisol (ng/ml)	462 (283-1040)	437 (231-730)	363 (289-648)	413 (226-680)
C-reactive protein (mg/l)	9 (<5-163)	12 (<5-113)	7(<5-67)	6 (<5-73)
Albumin (g/l)	43 (32-47)	43 (32-48)	42 (33-47)	42 (29-48)
Total body water (l)	38.3 (28.2-48.0)	36.8 (30.1-45.1)	38.3 (27.3-45.2)	38.2 (27.8-44.8)
Body fat mass (kg)	22.8 (9.3-37.2)	22.6 (15.3-38.5)	23.2 (13.5-39.1)	21.7 (13.2-38.3)
Biceps Skinfold Thickness (mm)	5.5 (3.3-9.3)	5.6 (3.4-9.0)	5.4 (3.4-9.2)	5.8 (3.0-10.2)
Triceps Skinfold Thickness (mm)	11.4 (6.1-28.5)	10.9 (6.4-28.0)	11.0 (7.6-28.4)	11.2 (7.5-29.0)
Mid Arm Circumference (cm)	28.4 (23.4-34.5)	28.2 (23.5-33.2)	27.5 (23.0-34.0)	28.3 (24.0-33.0)
Appetite	8.0 (0.7-10.0)	8.1 (1.0-10.0)	8.4 (3.2-10.0)	8.4 (3.4-10.0)
Energy intake (kcal/day)#	1596.5 (1269-1890)	1641 (914-2216)	1952 (1392-2435)	1814 (1222-2503)
Resting energy expenditure (kcal/ d)	1544 (1274-2060))	1593 (1226-1964)	1550 (1307-2126)	1566 (1219-1967)
Respiratory Quotient	0.82 (0.72-0.92)	0.81 (0.72-0.94)	0.81 (0.72-0.94)	0.83 (0.75-0.92)

Median (range) # n=6

6.0 Conclusion

Chemotherapy for metastatic colorectal cancer has been used for palliation for many years. During this time 5-FU has remained the single most effective anti-neoplastic agent in the treatment of colorectal cancers. Whilst many studies have assessed the safety and efficacy of chemotherapeutic regimens using response rate, toxicity profiles and quality of life, few studies have evaluated the effect of chemotherapy on the metabolic response of the host in particular energy balance, body composition and quality of life.

The studies presented in this thesis had a number of aims; firstly to examine the relationship between the systemic inflammatory response and the short and long term-effects of 5-FU based chemotherapy on biochemical and haematological parameters of the host response. Also, the effect of 5-FU based chemotherapy on body composition, dietary intake, resting energy expenditure and quality of life in patients with colorectal liver metastases was assessed.

Measured resting energy expenditure values were noted to be higher than would be predicted. However, there were no significant differences in energy intake or resting energy expenditure during the course of the study. In those patients who responded to treatment, there appeared to be no alterations in any of the body composition, quality of life and performance status parameters. It was of interest that positive acute phase proteins such as fibrinogen fell in concentration during the course of the study and was consistent with a reduction of the systemic inflammatory response.

The thesis studies also examined the relationship of the systemic inflammatory response and disease progression in patients with colorectal liver metastases receiving 5-FU chemotherapy. On comparison of parameters during a period of 'responding' disease and 'progressive' disease, an increase in white cell count, neutrophil count and C-reactive protein on disease progression was observed. This is consistent with tumour progression evoking a systemic inflammatory response. The basis of this response is not clear, however, it could be due to

either a non-specific response to necrosis produced by the growing tumour or to the malignant phenotype of the tumour. Nevertheless, these results are consistent with those found in other advanced tumours and would suggest that there may be a stereotypical host response to metastatic disease.

Finally, in this thesis, the longitudinal relationship between leptin and energy metabolism, fat mass and appetite in patients with colorectal liver metastases undergoing 5-FU chemotherapy was examined. The changes in circulating leptin concentrations were not associated with alterations in appetite, energy intake, resting energy expenditure or respiratory quotient. Changes in circulating leptin concentrations reflected changes in body fat mass and indicated that the relationship between circulating leptin concentrations and body fat mass was preserved in these patients. This would suggest that leptin concentrations may be a useful routine marker of nutritional status in cancer patients.

Therefore, from the results of the above studies, it would appear that in those patients who responded to 5-FU based chemotherapy the effects on host metabolism were small.

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