HYPERTHERMIA

PHYSIOLOGICAL CHANGES ASSOCIATED WITH WHOLE BODY HYPERTHERMIA FOR CANCER TREATMENT

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Preface

Over the last ten years hyperthermia has been introduced as an experimental therapy that is available to the clinician in the treatment of malignant disease. Intensive experimental and clinical research is continuing into various aspects of treatment.

Since the importance of enviromental factors in hyperthermic cellular destruction was realised, a large number of investigators have undertaken studies aimed at ellucidating the role of hyperthermia on cells under the influence of such factors as nutrition, oxygenation and hydrogen ion concentration. Investigations are continuing into temperature/time relationships for killing of both normal and malignant tissues, while other studies are focusing attention on heat dose fractionation and thermal induced thermal resistance.

Efforts have been made to achieve greater understanding of the structure and functioional limitations of tumor microcirculation, of disturbances brought about in it during hyperthermia, and the resultant changes occurring in oxygenation and acid base balance at the local level.

Additional studies are in progress with the aim of investigating the role of hyperthermia in potentiating damage to normal and malignant tissues brought about by radiotherapy and the sequential effects of the two modalities. The effects of combined chemotherapy and thermotherapy are also under intensive investigation.

In a clinical setting hyperthermic treatment may be given in two ways - locally or systemically. In the case of local hyperthermia attempts are made to maximise the temperature rise in the malignant tissues while minimising the effects on normal tissues - in some cases by employing active cooling. During whole body hyperthermia treatment (WBHT) the entire patient is heated and a body temperature of 41.8°-42°C is reached. It is usually necessary to carry out

this treatment either under general anaesthesia or under a generalised sedation technique.

Since 1978 hyperthermia treatment, both local and WBHT has been undertaken in the Rotterdam Radiotherapeutic Institute. This work has been carried out by the department of Experimental Radiotherapy and the department of Anaesthesia of the Erasmus University, Rotterdam.

This thesis will first discuss some relevant aspects of the theoretical and experimental background to the treatment. Then certain aspects of WBHT will be discussed, including heating techniques and anaesthesia for this type of therapy. Finally the results will be presented of some experiments that were carried out to determine the causes of some post treatment complications.

It should be noted that this thesis restricts itself to early pathophysiological changes induced by hyperthermia treatment and no attempt will be made to describe or interpret the effects of hyperthermia on experimental tumors or clinical malignancy. The clinical trials of Whole Body Hyperthermia inevitably involved a large group of specialists from a large range of scientific disciplines. It is intended that the results of Whole Body Hyperthermia Treatment should be dealt with in separate publications that are in preparation by colleagues in the field. It is hoped that the thesis presented here will provide helpful guidelines for any anaesthetist presented with the task of anaesthetising patients for Whole Body Hyperthermia Treatment. It is also probable that general conclusions from these studies will be applicable to future work in deep local body heating. With these aims in mind only the most important aspects of thermobiology have been presented and the reader should not expect to find a full review of all the work that has been performed on this subject.

CHAPTER I

THERMAL SENSITIVITY OF NORMAL TISSUES AND THE INTACT ANIMAL

Bligh (1979), has stated that constituent metabolizing cells of virtually all animals and plants are destroyed by temperatures outside the approximate range of 0°C to 45°C.

It would seem, however, that the intact animal is less able to withstand high temperatures than its constituent tissues. Reed et al. (1964) working in dogs have shown that it is possible to heat the perfused liver, via portal vein infusion, to a temperature of 46°C for short periods of time, provided that hepatic hypoxia is avoided. Raised enzyme levels indicating possible liver damage gradually reduced to normal within 21 days. Lung tissue of the dog has been shown to tolerate temperatures up to 43°C for periods as long as 60 minutes without significant permanent disturbance of structure or function - Reed (1965).

In the intact animal, Frankel et al. (1963) have presented evidence of developing generalised tissue hypoxia in dogs at body temperatures between 42° and 43°C, and Magazanik et al. (1980) have confirmed the occurrance of increasingly severe metabolic acidosis in dogs at rectal temperatures above 42°C. It should not, however, be forgotten that temperatures of different tissues may vary widely in the same animal when that animal is subjected to hyperthermia; Dickson et al. (1979), working with adult pigs have demonstrated that, at a rectal temperature of 42°C, the temperatures of the internal organs may vary considerably. The temperature measured in the liver, for instance, is constantly 0.1-0.4°C higher than that in the rectum. Fletcher et al (1980), working with anaesthetised rats, have reported that liver temperatures may be

as much as 2°C higher than rectal temperatures.

Shibolet et al. (1976), have stated that levels of temperature incompatible with life appear to begin, in man, around 42°C, though this cannot be taken as an absolute limit, in view of the report of Ferris et al. (1938) of recovery of two out of four heat stroke patients admitted to hospital with rectal temperatures in excess of 44°C.

The concept of critical thermal maximum (CTM) was introduced by Cowles and Bogart in 1944 and Hutchinson (1961) has defined it as the minimum raised body temperature that is lethal for the animal in question. Wright (1976), working with mice has established a CTM of approximately 44°C. This was the temperature at which heat induced convulsions and loss of the righting reflex was seen and usually occurred after about 90 minutes in a 40,8°C, 50 percent relative humidity, environment. They noted that the CTM was higher in animals preacclimatized at 30°C than those acclimatized at 15°C. They noted no change in CTM caused by prior hyperthermic exposure to either a core temperature of 42°C or after survival from CTM exposure. Hubbard et al. (1977), in a study using rats, related mortality to the product of the rise in °C above 40.8°C and the minutes of exposure and found a positive correlation between the two. They also made the observation that long term survivors (in the non-exercising and heated group of animals) had a faster cooling rate than those animals that died following the experiment. Bynum et al (1977), by raising the body temperature by 1.5°C per hour followed by passive cooling, established a CTM of 43.4°C for anaesthetized dogs. Above this threshold all dogs died. This confirmed the work of Shapiro et al. (1973) who found a value between 43°C and 44°C in awake non-exercising dogs.

Bynum et al. (1978), commenting on the reports of Pettigrew et al. (1974a) on the maintenance of patients under general anaesthesia at rectal temperatures of 42°C for up to eight hours without persistent side effects, considered there to be a need for raising the CTM of humans to above 42°C or that the whole

concept of CTM should be modified. They went on to suggest that "the concept of CTM be expanded to include a combination of exposure time and body temperature which results in both CTMs and CTMc injuries". CTMs they defined as the thermal maximum necessary to achieve specific subclinical biothermal injuries and CTMc as that necessary to achieve the clinical syndrome of heat stroke and shock. To aid in the application of these concepts they used the term Tequivalent 42 (Teq 42). This is the thermal dose administered expressed as if it were in minutes at a temperature of 42°C. They further demonstrated correlation both between Teq 42 and mortality in heat stressed dogs and between Teq 42 and mortality in mammalian cell cultures.

Temperature Time Relationships for tissue damage

One of the first studies of the relationship between the temperature and time of maintainance of that temperature and the succeeding tissue damage was performed by Moritz and Henriques (1947) who studied the thermal reaction of the skin of pigs - this varied from erythema to transepidermal necrosis. They established, that between 44°C and 51°C, for every one degree rise of temperature the tissue required only half the exposure time to sustain the same degree of damage. They also established that there was a critical temperature for damage above which only a very small rise of temperature was necessary for a rapid increase in the rate of damage. Tests were also carried out on human volunteers and it was found that there was little or no difference in thermal sensitivity or reaction as between porcine or human skin.

Similar temperature/time relationships were obtained by Crile (1963) and Suit (1977) using mice feet. The latter found a two thirds reduction in heating time required to produce the same effect following clamping of the arterial supply at 43°C. This would indicate the importance of an intact circulation, and hence the ability to transport heat away from overheated areas, in the

maintainance of thermal homeostasis.

Morris et al (1977) used tails of baby rats as an assay system and found similar results in that doubling the heating time or a rise in temperature of 1°C had a similar effect on necrosis or stunting of tail growth between 41°C and 46°C. Clamping of the tails, and hence occlusion of the blood supply, gave a reduction in heating time by a factor of 3 - equivalent to a temperature rise of 1.5°C. They also established that at a critical point an increase of only 20% in heating time or less than 1/2°C was all that was required to increase necrosis from 0% to 100% - indicating the presence of a very steep dose response relationship at around the critical temperature.

Similar temperature/time relationships have been demonstrated by Okumura and Reinhold (1978), who employed an assay method using thermal damage to the skin of rats. They confirmed that when the skin temperature was lowered by 1°C twice the heating duration was necessary to produce the same degree of thermal damage.

The above results would indicate the vital necessity of uniform warming and very accurate temperature monitoring during clinical hyperthermia (Field and Bleehan 1979).

CHAPTER II

CELLULAR THERMOBIOLOGY

ARE MALIGNANT CELLS MORE SENSITIVE TO HEAT THAN NORMAL CELLS?

As pointed out by Field and Bleehan (1979) in a recent review, the most important question to be answered is whether hyperthermia is more damaging to tumors than to normal tissue. A great deal of work has been done in both tissue cultures and in intact animal experiments.

In 1941 Vollmar attempted to quantitate survival of malignant cells following hyperthermia (Jensen sarcoma and Ehrlich ascites carcinoma from rodents) in comparison with normal splenic fibroblasts; she based her measurements on subjective assessment of growth rate of tumors, following exposure to various temperatures for 3 hours. It was concluded that whereas normal cells were resistant to thermotherapy at temperatures up to 43°C, malignant cells were killed at between 40°C and 42°C. Auersperg (1966), on the other hand, in a study based on the use of vital dye exclusion as an end point for cell survival, concluded that malignant epithelial cells derived from Carcinoma line C4 and C27 were less sensitive to heat than normal human fibroblasts obtained by skin biopsy.

Bender and Schramm (1966) using microscopic evidence of increase of cell numbers after incubation at 37°C as the criterium of cell survival after treatment, studied a number of cultures of human and animal tissues; organ and embryonic cultures were used as controls. They demonstrated that malignant cells were 1-2°C more thermosensitive than normal cells.

In a much quoted study,Chen and Heidelberger (1969), using plating efficiency as an indicatior of survival, determined that C3H mouse ventral

postate cells, that had been malignantly transformed by methylcholanthrene were more themosensitive than normal controls. Pretreatment at 42.5°C for four hours did not effect the rate of malignant transformation, which, the authors stated, demonstrated true acquisition of thermal sensitivity and not that only the most heat sensitive cells had been transformed.

It may be that the method of malignant transformation has an influence on the acquisition of thermal sensitivity, as Ossovski and Sachs (1967) found that cells transformed by a strain of polyoma virus were more stable to heat than untransformed cells. Similarly thermosensitivity may vary between different cell lines.

Kim et al. (1974) found less colony formation ability, and thus greater thermal sensitivity in malignant HeLa cells than in normal mouse fibroblasts following hyperthermia at 42°C.

Harisiadis et al. (1975) used colony counting following post-treatment incubation at 37°C as an index of survival for short exposure (less than 30 minutes) to temperatures of 45°C. They were unable to demonstrate increased thermo-sensitivity of malignant cell lines (V79 and CHO derived from the chinese hamster and a cloned hepatoma cell line in the Morris 5123 tumor), when compared with normal cells derived from rat liver.

Giovanella et al. (1976) state that is necessary to define malignancy and to decide what constitutes a neoplastic cell when considering the question of thermo-sensitivity. They have defined human cells as being neoplastic only when the cells have been cultured from human neoplasms, are aneuploid and capable of producing malignant tumors when injected into thymus deficient nude mice. On this basis, working entirely with material of human origin, they demonstrated higher thermo-sensitivity in four malignant cell lines when comparing them with normal, presumably euploid, cell lines derived from human foetal material. In these experiments the number of surviving cells following treatment were counted and expressed as a percentage of control (untreated)

numbers. Statistical significance was apparant after two hours at 43°C in the case of colon carcinoma cells (compared with foetal intestinal cells), and after 4 hours in the case of melanoma cells under the same conditions (comparing with foetal melanocytes). It also required 4 hours treatment - this time at 42.5°C - to differentiate between survival of terato-carcinoma cells in comparision with foetal neuroepithelial cells. In the case of adult fibrosacoma cells, which were compared with adult fibroblasts, an exposure time of eight hours at 43°C resulted in significant differences. It should be noted that the percentages of normal cells destroyed by the treatment were not negligible; for instance, after eight hours of treatment, though about 95 per cent of melanoma cells had been destroyed, at the same time about 70 per cent of normal melanocytes had been destroyed. Nonetheless, this study does suggest that, using the abovementioned definitions of neoplastic cells, normal cells might be more thermo-resistant than their non-malignant counterparts.

Dewey et al. (1977), in a short review of cellular processes under hyperthermia, considered that, in general, there are no consistant differences between normal cells and malignant tumor cells in their thermosensitivity and their reactions to hyperthermic temperatures. They admit however, that studies of morphological changes and inhibition of metabolic reactions such as respiration and macromolecular synthesis do tend to indicate that normal cells are more resistant to thermal stress than undifferentiated malignant cells. Cavaliere et al (1967) for instance, have shown that Novikoff hepatoma and Ehrlich Ascites carcinoma cells had a lower oxygen consumption at 42°C than at 38°C; normal and regenerating liver cells showed similar consumptions at the two temperatures. Similar results were obtained by Muckle and Dickson (1971) who observed reduction in oxygen uptake by rabbit VX2 carcinoma cells at 42°C in comparison to 37.5°C, while anerobic glycolysis was not increased in either malignant or normal cells at hyperthermic temperatures.

Some recent work by Symonds (1982) deserves mention. When comparing heat

sensitivity of normal mouse haemopoietic stem cells from bone marrow (CFU-S cells) and L1210 clonogenic mouse leukemia cells taken from bone marrow, it was shown that there was a markedly greater heat sensitivity in the leukemic cells. However, when L1210 cells were grown as an ascites tumor they were not significantly more sensitive to hyperthermia than CFU-S cells. The effect of the presence of normal healthy marrow cells on the mortality of the malignant cells, was investigated by heating a mixture of L1210 asities cells with healthy marrow cells. Thermo-senstivity of the L1210 cells was increased by this mixing and it was postulated that this was caused by release of lysosomal enzymes from heat damaged haemopoietic stem cells capable of penetrating the membranes of adjacent malignant cells, rendered unusually permeable by heat.

As Field and Bleehen (1979) have pointed out, there are as yet no clear reasons why tumor cells would be more heat sensitive per se than other cells. Probably most authorities would agree that there is negligable difference between normal and neoplastic cells in this respect.

FACTORS EFFECTING SENSITIVITY OF CELLS TO HYPERTHERMIA

Cell Cycle

Cells synthesizing DNA are relatively resistant to x-rays and hence their sensitivity to radiation depends on the stage in the cell cycle. This has been demonstrated by a number of workers including Sinclair and Morton (1965) who used synchronized V79 chinese hamster cells and demonstrated most x-ray resistance in the later part of S-phase with equal sensitivity before (G1) and after (G2) this period. Dewey et al.(1970) have shown that G1 cells are two or three times as sensitive to radiation as S-phase cells.

When considering heat damage the reverse of the above is observed - i.e.

cells are more sensitive in S-phase than in G1 phase. This has been demonstrated by Westra and Dewey (1971) and Dewey et al.(1971). The latter authors considered the possibility that heat inactivation of mammalian cells resulted from denaturation of proteins.

Palzer and Heidelberger (1973),working with HeLa cells, found late S and early G2 phases to be the most thermosensitive parts of the cycle. Cells in other phases, following similar heat dosages, exhibited survival that was seven times that of those in the late S and early G2 phases.

Effect of Hypoxia and pH

As will be discussed in a later part of this thesis, cells of malignant tumors are frequently in a hypoxic milieu and at a low pH. It is therefore of considerable importance to know if these conditions will decrease or increase the thermal sensitivity of these cells.

The fact that hypoxia and low pH are interrelated to a certain extent makes consideration of the separate contribution of the two variables rather difficult - especially in intact tissues. Separation is only feasable using cell cultures.

Gerweck et al. (1974), showed that hypoxic asynchronous CHO cells were at least as sensitive as their oxygenated controls to hyperthermia at 45.5°C whereas the hypoxic cells were more resistant to radiation. Hahn (1974) also demonstrated that hypoxia was of little importance when considering the effects of hyperthermia on HA1 cells at 43°C. Tnese results were confirmed for chinese hamster V79 cells and mouse EMT6 cells by Power and Harris (1977).

In contrast, Harisiadis et al. (1975) found that hypoxic V79 Hamster cells were 'dramatically more susceptible to killing by heat than areated cells'. They also found, as others have done, that the reverse was true in the case of radiation. This work might be criticised on the grounds that the investigators

made no attempt to standardise the pH of the different cell groups. Recent work by Leith et al. (1982), working with human colon cells in media buffered to a constant pH of 7.4, have demonstrated no difference between oxic and hypoxic cell suspensions in terms of hyperthermic inactivation.

It is thus probable that the process of cell killing by hyperthermia is, in contrast to that of radiation, largely independant of the degree of cell oxygenation.

Effect of pH

Gerweck and Rottinger (1976) demonstrated a ten-fold decrease in survival of CHO cells at pH 7.0 at 42°C compared to pH 7.4. At pH 6.7 decrease of survival was 500-fold. Overgaard and Bichel (1977) maintained both oxic and hypoxic cells (JB-1E tumor cells) at 42.5°C at a pH of 6.4 or 7.2. At pH 7.2 the decreased survival, assesed using clonogenic assay methods, was slightly enhanced by hypoxia. This increased mortality they attributed to a accumulation of lactic acid in the hypoxic cells caused by anerobic glycolysis, which is apparently unimpaired under hyperthermia - as earlier shown by Muckle and Dickson (1971), Cavaliere et al. (1967), Mondovi et al. (1969), and Dickson and Shah (1972). Overgaard and Bichel (1977) went on to demonstrate a very much greater depression of survival after treatment at 42.5°C in cells maintained at a pH of 6.4 and under these conditions there were no differences between oxygenated and hypoxic cells. The same authors, (Bichel and Overgaard, 1977) demonstrated that at pH 7.2 plateau cells were more sensitive to heat than exponentially growing cells. They were even more sensitive at pH 6.4, but the relative sensitivity of the exponentially growing cells had increased. This observation of the greater thermal sensitivity of plateau phase cells, as confirmed by Hahn (1974), is of importance due to the fact that many cells in solid tumors in animals are nonpoliferating and hence are relatively radioresistant. A similar situation may well exist in human tumors and hence these

observations have important therapeutic consequences.

Freeman et al. (1980), working with CHO cells, confirmed earlier work on the increased sensitivity of cells at low pH and investigated the effect of pre- or post-hyperthermic incubation at various pH values. The most important of their findings with regard to the present discussion was the finding that post-treatment incubation in an acid medium caused augmentation of the thermal damage. This effect was not however seen by Gerweck (1977), who also used CHO cells.

In a short review of the role of tissue environmental factors, Overgaard and Nielsen (1980) have postulated that the enhanced response to hyperthermia under acidic conditions may be effected in three different ways. There may be a direct increase in sensitivity to heat as shown by a decreased Do (heating times to reduce survival to 1/e in the exponential part of the survival curve) as shown in the study of Bichel and Overgaard (1977). In the second case there may be a reduced ability to repair sublethal damage - this is demonstrated by a reduced shoulder on the survival curve and can be seen in the study by Nielsen and Overgaard (1979). Lastly, they suggest that there may be a mechanism whereby reduced survival is due to lowering of the level at which thermal resistance occurs during prolonged heating.

The mechanisms whereby increased thermal sensitivity is bought about by exposure to low pH conditions are uncertain. Overgaard (1977) initially suggested that lysosomal destruction of malignant cells was being accelerated at a low pH and Overgaard and Poulsen (1977) have demonstrated increased proteolytic activity following hyperthermic treatment under acidic conditions when PNJ ascites tumor cells were incubated with cytochrome C. This was interpreted as meaning that lysosomal activity may account for the greater thermal sensitivity of malignant cells in vivo than similar cells treated in vitro (Overgaard 1977).

It is not clear if extracellular pH per se is the most important factor or

if it is just causing a modification of intracellular pH. Haveman (1979) has claimed that the change of intracellular pH is of importance in heat sensitivity, whereas Dickson and Oswald (1976) stated that there was no relationship between intracellular pH and hyperthermic cell destruction.

Nutritional state of the cells

A number of studies have been carried out on the nutritional state of cells in vitro in which the heat sensitivity has been assessed. Conflicting results have been obtained. Kase and Hahn (1975) demonstrated increased hyperthermic survival of unfed plateau phase cells compared with exponentially growing cells; but Hahn (1974) has shown that unfed cells become more sensitive to heat. The study by Bichel and Overgaard (1977), already alluded to, would indicate that pH is the dominant factor determining heat sensitivity and Overgaard and Nielson (1980) considered that increased heat sensitivity of unfed plateau phase cells, (Hahn 1974) might be due to increased environmental acidity.

Thermal Resistance

Gerweck (1977) has demonstrated that the shape of cell survival curves in response to temperature may be biphasic at marginally lethal temperatures. CHO cells at pH 7,4 were used and they demonstrated that at 41°C the initial decrease of survival leveled off after 2,5 hours and that further treatment at 41.5°C caused no further cell killing. A similar levelling occurred after three hours at 42°C, whereas at 43°C cell killing proceeded exponentially after 2 hours of heating. Relative survival of cells heated for 4 hours at 42°C was decreased by low pH and it was demonstrated that at 42°C the resistant tail of the survival curve developed after 3.5 hours at a pH of 7.0 (instead of the 3

hrs at pH 7,4). At pH 6.7 the shoulder of sub-lethal heat damage that was seen in the previous curves, practically disappeared and the survival decreased exponentially. Similar results have been obtained by Bauer and Henle (1979) who went on to demonstrate that the temperature compatible with biphasic survival curves could be increased by acute heat conditioning for ten minutes at 45°C; this causes acute thermotolerance. This phenomenom will be discussed below.

Thermotolerance

Thermotolerance is a term given to a certain degree of thermal resistance that is acquired as a result of a previous (sub-lethal) hyperthermic dose.

Palzer and Heidelberger (1973) working with HeLa cells demonstrated increased survival following thermotolerance development during factionated dosage of 42°C hyperthermia. This thermotolerance developed after 2 treatments of one hour each separated by an interval of 3 hours and declined by the time the separation interval had reached 8-10 hours. They concluded that their cells were recovering from sub-lethal hyperthermic damage.

Gerner et al (1976) also working with HeLa cells demonstrated that the degree of thermotolerance acquired following a first thermal dose depended on the magnitude (temperature/time product) of that dose. They demonstrated that cell metabolism was necessary by showing that tolerance did not develop when the cells were incubated at 0°C in the period between the two hyperthermic exposures. This has been confirmed by Stickney et al (1980), who have presented results that suggest that manipulation of polyamine biosynthesis can be effective in regulating the development of thermotolerance and have shown that if polyamine decarboxylation is inhibited thermotolerance does not develop.

Nielson and Overgaard (1979) investigated the effect of extracellular pH on

thermotolerance and recovery of hyperthermic damage in vitro. The L1A2 cells that they studied developed maximum tolerance 10 hours after the first thermal dose. The degree of tolerance in cells grown in a pH adjusted modified Krebs-Ringer phosphate buffer was lower in conditions of increased acidity. The pattern of development with time, however, did not change. They also demonstrated that a low pH in the interval between hyperthermia treatments was particularly responsible for the observed inhibition of thermotolerance development. Nielson (1981) studied unfed L1A2 cells in plateau phase and found that, though the overall pattern of thermotolerance development was similar, the degree of tolerance was less than that found in exponentially growing cells. It was also noted that this was not due to the increased environmental acidity in the plateau phase cells.

Gerweck and Bascomb (1982) investigated the effect of hypoxia on the development of thermotolerance. They concluded that, using CHO cells, 'reduction in oxygen concentration during and between heat treatments effected neither the magnitude nor the kinetics of thermo tolerance development'. Therefore it may be concluded that hypoxia alone is not an important factor in causing change in thermotolerance but that pH is the most important environmental factor determining this phenomenon.

Li et al (1982) in experiments using plateau phase chinese hamster HA1 cells came to the conclusion that the duration of the first (sensitising) dose had very little to do with the rate of development or the decay of thermotolerance. They also found that a low sensitising temperature 41°C or 42°C caused maximum thermotolerance to develop earlier than when a high sensitising temperature (43°C) was used. They hypothesized that thermotolerance is an non-specific defence mechanism of cells and that sublethal damage caused by the first thermal dose triggers a mechanism that can only act successfully when the cells are maintained at a temperature below 43°C - hence the apparent inconsistency of their findings.

Step Down Heating

This is a term used to describe an interesting phenomemon in temperature time heating relationships. A marginally lethal or non-lethal temperature may become toxic when preceeded by acute heat treatment of higher temperatures such as 45°C. Henle and Leeper (1976), while studying the combined modalities of hyperthermia and radiation on CHO cells, demonstrated reduced survival following incubation at 40°C given after a pretreatment at 45°C.

Henle et al. (1978) presented similar results, again using CHO cells. They showed that increased cell killing was dependent on the temperature of the post treatment incubation. They also produced evidence that preincubation at a low hyperthermia temperature of 40°C reduced cell killing at 45°C. This they attributed to enhanced cellular capacity to accumulate sub-lethal damage, though it might also be considered as being due to induced thermotolerance.

Modifying Treatments

Search is continuing for chemical substances that will modify the effects of hyperthermia. Not only will these modifications indicate possible causes of hyperthermic damage but may point the way to more effective clinical treatment.

Henle and Leeper (1982) have recently reported studies of hyperthermia on CHO cells in which inhibitors of macromolecular synthesis were used. Exposure of the cells to cyclohexamide for 2 hours before a 2 hour treatment at 45°C increased survival by a factor 1,8. When the drug was administered during a 7 hour 37°C incubation following a 10 minute 45°C heat treatment a 50% decrease in subsequent thermotolerance development resulted. Lucanthine, a reversible inhibitor of RNA synthesis, and hydroxyurea, an inhibitor of DNA synthesis had no effect on the development of thermotolerance. This would indicate that thermotolerance may require synthesis of new proteins.

Yatvin (1977) has studied membrane effects and cell killing by hyperthermia. Using a B coli mutant, K1060, in which the unsaturated fatty acid (UFA) content of the cell membranes could be altered, he concluded that the higher the UFA content, and hence the lower the viscosity, the lower was the hyperthermic killing rate. By increasing membrane fluidity with Procaine he was able to increase cell killing. Yau (1979) has confirmed this increase in hyperthermic mortality caused by Procaine and has found it to be dose dependent. These findings suggest, at least in part, that thermal killing may be attributed to changes in cellular membranes caused by hyperthermic exposure.

Azzam et al (1982) incubated chinese hamster V79 cells at various hyperthermic temperatures in 87% Deuterated Water (D2O) (which has been shown to cause stabilization of macromolecular structures). Below 43°C the thermoresistant tail of the survival curves occurring in untreated controls was eliminated in the cells incubated in D2O, but above 43°C thermoprotection was provided. This may indicate stabilisation of either cellular membranes or DNA and enzyme structures in the cell.

Glycerol has been shown to significantly increase the temperature at which 50% protein denaturation occurs (Back et al. 1979) and Henle and Waters (1982) have recently demonstrated that it can protect CHO and HeLa cells against thermal killing. This protection was concentration dependent. They also demonstrated that glycerol could protect cells at low extracellular pH and could increase thermal resistance of cells undergoing step down heating. An interesting observation was that, during long term nutrient deprivation (8.5 hours), glycerol increased cell killing at 41.5°C. Therefore protection by glycerol must be dependent on the presence of, or a metabolic by-product of, some unknown deficient nutrient.

Discussion of the interactions of hyperthermia and chemotherapeutic agents will be discussed a later section.

COMBINED CELLULAR EFFECTS OF HYPERTHERMIA AND RADIATION

There is a large amount of work that indicates that hyperthermia potentiates radiation damage in cells (Ben-Hur et al. 1974; Kim et al. 1974; Power and Harris 1977; Ross-Riveros and Leith 1979) .

Hyperthermia has been shown to cause a steeper slope of the survival curve following radiation and, in most cases, causes a reduction in the 'shoulder' of the curve. As already mentioned hyperthermia sensitivity and radiobiological sensitivity occur at different stages in the cell cycle. Kim et al. (1976) using synchronized cultures of HeLa cells has shown that combined treatment of hyperthermia and radiation resulted in various intensities of enhanced cell killing throughout the cell cycle. The greatest synergism occured in cells in the late S phase.

Ben-Hur et al. (1974) used split dose experiments with CHO cells to demonstrate that hyperthermia was inhibiting repair of sub-lethal damage. This view of the action of the combined modalities was supported by Dewey et al. (1977), and Suit and Gerwick (1979).

Suit and Gerwick (1979) have summarised experimental evidence and have concluded that there is no evidence that sensitisation to radiation by hyperthermia is greater in malignant than in cells of normal tissue origin.

Effects of Hypoxia

Hypoxic cells are less sensitive to radiation than are well oxygenated cells and the oxygen enhancement ratio (OER) for radiation is in the order of 2.5 to 3 (Suit and Gerwick 1979) - in other words cell killing is increased when the cells are oxygenated. In the case of hyperthermia the OER is approximately unity i.e. there is in general no greater thermosensitivity in oxygenated as opposed to hypoxic cells.

A number of studies have been carried out into the influence of hyperthermia on radiation OER in the mammalian cells. Robinson et al. (1974) and Kim et al. (1975) used mouse bone marrow and HeLa cells respectively. They demonstrated reduced OER's at hyperthermic temperatures. Power and Harris (1977), however, in studies with EMT6 mouse tumor cells and V79 chinese hamster cells showed no change in OER at 43°C.

These studies would indicate that differences exist between different types of malignant cells in their sensitivity to combined heat and radiation and would point to the necessity, in some cases, of sequential application of the two modalities.

Effect of pH

The effect of low pH on the cell killing of the combined modalities of radiation and hyperthermia is of no small importance. This is especially so when it is realised that both normal and malignant cells are probably equally sensitive to the combined treatments (Suit and Gerwick 1979) and that there are areas of low pH in tumors (Bicher et al. 1980).

Rottinger and Mendonca (1982) have demonstrated that both human glial cells and chinese hamster ovary cells showed improved post radiation survival if maintained at a low pH following treatment.

Lunec and Parker (1980) working with HeLa-S3 cells established that these were more sensitive to hyperthermia at pH 6.7 than at 7.4. They went on to demonstrate a slight increased hyperthermic potentiation of radiation damage at the lower pH value.

Freeman et al. (1981a) demonstrated slight radioprotection of CHO cells when radiation was carried out at an acid pH - thus confirming the work of Haveman (1980) who obtained similar results working with murine carcinoma cells. Freeman et al. (1981a) went on to show that both heat treatment alone

and combined heat and radiation in acid medium produced more cell killing than treatments in alkaline medium. Subsequent recovery from heat damage was less in an acid pH. Freeman et al. (1981b) have produced similar results working with Madcap-37 cells and in addition, showed that an acid environment prolonged the duration of hyperthermic radiosensitisation when heat preceeded radiation.

Timing of Heat and Radiation

Sapareto et al. (1978), in an extensive investigation into the combined effects of X-irradiation and hyperthermia on CHO cells, studied not only the timing of radiation in relation to heat, but also investigated the effects of the two modalities on cells in different stages of the cell cycle. They concluded that when relatively sublethal heat doses were given, (as would be the case in whole body hyperthermia treatment), maximum decrease in survival of asynchronous cells occurred when heat was given during, or following radiation. They noted the fact that a five minute difference in timing could well make a 6 fold difference in the resulting cell survival. They also found that the regime of heat application immediately before, or immediately after, sensitized S phase cells to radiation. These cells are normally radioresistant but, under these conditions, there were no differences in survival between S phase and G1 phase cells.

Dewey and Freeman (1980) have postulated that, though the maximum effect on cells occurs following simultaneous administration of hyperthermia and radiation, it may be possible to obtain a therapeutic gain by administering heat after radiation damage in tumours is repaired. This would be due to the enhanced thermal killing observed at low pH occurring in malignant tissues, the characteristics of which will be discussed in another chapter.

COMBINED CELLULAR EFFECTS OF HYPERTHERMIA AND CYTOTOXIC AGENTS

Several anticancer agents have been shown to have increased activity at hyperthermic temperatures and to acquire increased cytotoxicity at tempertures that are obtainable during WBH or local hyperthermia treatment. Indeed some substances, not usually considered as cytotoxic at normal body temperatures, may acquire such activity under hyperthermic conditions. Field and Bleehen (1979) have reproduced a classification of types of drugs and their actions at elevated temperatures. As they have pointed out, this is a somewhat arbitory classification, but it may be a useful one and is used below.

Drugs Which Show no Threshold Effect

These drugs are cytotoxic at normal temperatures and are more so at increased temperatures. Johnson and Pavelec (1973), while studying the increased cytotoxicity of thio-TEPA under hyperthermia on chinese hamster fibroblasts, demonstrated a doubling of cell killing at 40°C in comparison to 37°C which they considered to be a purely chemical effect consistent with that predicted by reaction kinetics i.e. the cellular inactivation rate was linear with the rise in temperature.

Other agents falling into this group are the nitroso-ureas and Cis-platinum; Hahn (1979) using chinese hamster HA1 cells has demonstrated the increased efficiency of cell killing produced by these drugs at increased temperatures.

Drugs exhibiting a threshold temperature effect

a) cytotoxic drugs

These drugs, though they may be cytotoxic at 37°C, appear to have a change

in action at a critical temperature of 43°C. Above this level their cell killing effect is considerably increased.

Braun and Hahn (1975) have demonstrated this effect with bleomycin and attributed it to the fact that the 43°C hyperthermia was preventing recovery of potential lethal damage caused by the chemotherapeutic agent. Hahn and Strande (1976), working with chinese hamster cells, demonstrated this effect using adriamycin. This lethal effect was short lived but prolonged heating (30 minutes or more) caused a decrease in the cell killing. This effect was also seen if heating was applied before the administration of the drug. These effects they attributed to a biphasic permeability of the cell membranes to adriamycin, which was first increased by heat and later decreased.

Donaldson et al. (1978) have also demonstrated this type of hyperthermic protection against cytotoxicity occurring in the case of actinomycin D. They showed in addition that heat, administered both before and after the drug may cause decreased cell killing and were able to demonstrate that this was not caused by altered membrane permeability. They have rightly pointed out, as have many others, the need for careful timing of administration of chemotherapeutic agents when given as part of a hyperthermic regime.

b) non-cytotoxic agents

A further sub-group may be described as containing drugs not normally having cytotoxic properties at 37°C but which induce increase cell kill at 43°C or above. The polyeme antibiotic amphoteracin B is not cytotoxic at 37°C or 41°C, as shown by Hahn et al. (1977), but these investigators demonstrated that it produces a high degree of cell killing at 43°C.

Li et al. (1977) have demonstrated that some organic solvents, used to dissolve water insoluble chemotherapeutic agents, may themselves become highly cytotoxic at raised temperatures.

Effects of Hypoxia

The effect of hypoxia on chemotherapeutic agents may well be important as pointed out by Field and Bleehen (1979).

Roizin-Towle and Hall (1978) have demonstrated that hypoxic V79 chinese hamster cells are more sensitive to bleomycin under both hypothermia (17.5°C) and hyperthermia (42.5° C) than are oxygenated cells. At 37.5°C, on the other hand, oxygenated cells were more sensitive to the drug than hypoxic ones.

A certain amount of work has been done on combination of electronaffinic nitroimidazole radiosensitizers and hyperthermia. Stratford and Adams (1977) have demonstrated that though Misonidazole showed no toxicity (at the tested concentrations) against aerated V79 cells at 37°C and 41°C, an effect was seen at 37°C against hypoxic cells and this was very much increased by exposure to 41°C.

It would appear that cell killing potentiation by Misonidazole is not limited to hypoxic cells. Roizin-Towle and Hall (1978) have demonstrated that aerated V79 cells exhibit decreased survival following combined treatment with Bleomycin and Misonidazole as compared with Bleomycin alone. The cytotoxic effect of heat in combination with misonidazole has been confirmed by Stone (1978) using oxygenated cells from mouse 3HC mammary tumors.

CHAPTER III

THERMOBIOLOGY OF TUMORS

Tumors and normal tissues are not only composed of cellular material characteristic of that tissue but also contain blood vessels. The blood contained in these blood vessels assist in the transport of oxygen and other necessary organic and inorganic substances to the tissue, and removal of carbon dioxide and other products of cellular metabolism.

Tumors often contain areas of inadequate circulation and hence part of the tumor may be hypoxic. Due to the inability of the circulation to remove acid metabolic products of the hypoxic metabolism, the tumor may also be at a lower pH than the surrounding normal tissues.

Heat is also transported about the body by the circulation. For instance, cool blood flowing through a heated tissue tends to cool it down and if the circulation to one part of the tissue is less than to another part, the first will be cooled less than the second. Hence, during whole body hyperthermia differential heating and cooling of tumors and their overlying normal tissue may occur. Though this explanation is the one usually offered to explain this phenomenon (LeVeen et al. 1976), Gullino et al. (1978), using 'tissue isolated' tumors, were unable to produce a change in the difference between the temperature of the tumor and the surrounding tissue brought about by changes that they induced in tumor blood flow.

Another aspect of hyperthermic therapy that must be considered is the effect of hyperthermia on the circulation to the tumor and the resultant changes brought about in the internal milieu of that tumor. The following section will attempt to deal with the characteristerics of tumor circulation

circulation and consequent tumor environmental effects that are the result.

TUMOR CIRCULATION

Tumors may be regarded as having a macrocirculation and a microcirculation. The macrocirculation is provided by blood vessels derived from neighbouring structures and adjacent tissues. The amount of this circulation will vary, depending on the total blood flow to the area of the body in which the tumor is situated. It will also vary according to the amount of blood that is admitted to the microcirculation of the tumor. At certain levels of hyperthermia the microcirculation in tumors may be increased.

Eddy (1980) working with a squamous cell carcinoma in a hamster cheek pouch preparation has demonstrated vasodilatation in tumor vasculature at 43°C. Song et al. (1980) using the Walker 256 carcinoma implanted subcutaneously in the flank of rats has shown increases in blood flow in small tumors (0.3-0.7 gms) at 43°C whereas larger tumors (2-5 gms) showed a decrease in blood flow at the same temperature. These changes will alter the amount of blood admitted to the microcirculation.

Whole body hyperthermia may cause different changes in blood flow in different parts of the body, and it has been shown that splancnic blood flow may be significantly decreased under these conditions (Faithfull 1982). Though total flow through the microcirculation of a locally heated tumor may be increased if situated in, for instance, the skin, the same tumor in, for example, the liver may show dramatic decreases in its microcirculation at the same temperature if the organism is subjected to whole body hyperthermia.

Measurement of Tumor Blood Flow

Measurements of total blood flow to tumors are reported by a number of

authors using a variety of techniques. These techniques can be divided into two distinct categories (Applegren 1978) - volume flow measurements and the use of radioactive tracers.

a) Volume flow measurements

Volume flow measurements consist of direct or indirect measurement of flow into or out of the tumor. If the circulation of a tumor can be isolated to a certain extent within the body, total venous outflow can be measured. Pioneering work in this respect was performed by Gullino and Grantham (1961). Similar methods have been used by Grantham et al. (1973) and Vaupel (1975). The arterial inflow into a tumor can be measured indirectly by venous occlusion plehtysmography provided the tumor is in a suitable position - for example subcutaneously implanted in the foot or tail of small animals (Kjartansson et al. 1976).

b) Use of Radioactive Tracers

The second method of measuring blood flow consists in the use of radioactive tracers whose concentration in the tissues are measured and give a reflection of the blood flowing through that tissue. Radioactive microspheres have been used by Blachard et al. (1965) and Rankin and Phernetton (1976). Takacs et al. (1975) have measured total tumor flow using a radioactive isotope of rubidium, Rb86. The method of measurement was devised by Sapirstein (1958) and can be used with I131 antipyrine and C14 antipyrine uptake (Allen et al. 1975).

Measurement of microcirculatory flow

The microcirculation or local tumor blood flow may be estimated by a variety of clearance techniques and Xe131 has been extensively used for this

purpose. Authors using this method include O'Brien and Veall (1974) and Robert et al. (1967). Na24 clearance has also been used to measure flow in malignant tissues by Peterson et al. (1969). Additional methods of measuring local tumor blood flow included distribution of dyes such as lissamine green as used by Owen (1960) and the use of Evans blue by von Ardenne and Reitnauer (1980b). Reinhold (1971) has used serial microangiograms. Heat clearance techniques have been used by Müller-Klieser et al. (1982).

Peterson (1979) has summarised the results of various studies in tumor blood flow and concludes that blood flow per unit of tissue volume decreases as tumor increases in size and this is reflected in the incidence of central tumor necrosis. Blood flow through tumor tissues is, in general, lower than the normal tissue of origin and is essentially inhomogeneous.

Direct observations of BA1112 sarcomas, implanted in rats, have been made by Endrich et al. (1979) using modified Algire chamber techniques. They were able to confirm both temporal and functional blood flow inhomogeniety in growing tumors. They demonstrated greater mean blood flows in peripheral actively growing areas of the tumor (in comparison with the surrounding normal tissues), but they measured decreasing flows towards the central (necrotic) areas of the preparations.

EFFECTS OF HYPERTHERMIA ON TUMOR CIRCULATION

Direct microscopic observation of the microcirculation under conditions of hyperthermia is difficult. It has, however, been accomplished.

Reinhold (1971) using a modification of the Algire Chamber System (Algire 1943) has developed a method of growing 'sandwich tumors' - sheets of a tumor of approximately 50 microns in thickness complete with their microvasculatures.

These sheets are grown in situ on the host animal and the preparation is sufficiently translucent to allow light microscopy and study of growth and reaction of the microcirculation under various conditions. Reinhold and van den Berg-Blok (1981) using a similar 'sandwich' preparation studied the effect of 42°C and 42.5°C hyperthermia on a Rhabdomyosarcoma BA1112 in rats. They demonstrated a latent period of about one hour at 42.5°C before vascular changes occurred. These were first manifested as a slowing of the microcirculation and after three hours microcirculatory damage was evident. On the following day the tumors demonstrated extensive central necrosis.

At 42°C essentially the same damage occurred but to a much lesser extent. Additional treatments with misonidazdole, glucose or 5-thio-D- glucose at 42°C caused similar degrees of damage to those obtained at 42.5°C thus indicating a degree of 'thermosensitization' produced by these substances.

In a further paper (Reinhold and van den Berg-Blok in press), the same authors studied 'step-down' heating. They pointed out that using the '$t_{\frac{1}{2}}$ per °C' rule (whereby a rise of treatment temperature of 1°C requires half the time exposure to produce the same damage) the 50% circulation stoppage time was essentially the same for similar thermal doses, but that with step down heating the microcirculation in the surrounding tumor bed was more impaired than was to be expected from similar temperature/time related treatments at lower temperatures. This would indicate a possible increased tumor destruction following step down heating.

Eddy (1980) has used a transparent cheek pouch chamber in the Syrian hamster. Temperature of the in vivo preparation was controlled by allowing water to flow through the compartmentalised chamber. Following an initial decrease of vascular diameter at all temperatures, at 43° and 45° vasodilatation occurred, followed at 43° by occurence of petechiae and signs of endothelial degeneration in the presence of persistent hyperaemia. At 45° haemorrage and thrombosis was accompanied by a complete shutdown of circulation

and was followed by coagulation necrosis.

Indirect studies of tumor vasculature have been carried out by Song (1978) who studied subcutaneously implanted Walker 256 carcinoma preparations in rats, locally heated in a water bath for 60 minutes at a temperature of 43°C. Vascular volume studies using Cr51 labelled red cells revealed significant increases in skin and muscle blood flow but not in the tumors. Similar changes were found in vascular permeability studies (using I125 labelled albumin) - again no changes in the tumor tissue. This lack of vascular changes in tumors under hyperthermia would account for the differential heating of tumors and normal tissues that has been claimed during hyperthermia by LeVeen et al. (1976) and Kim et al. (1977).

Song et al. (1980) went on to correlate the effects of hyperthermia on vascular function, pH, and cell survival. They demonstrated no change in blood flow in Walker 256 carcinomas subcutaneously implanted in rats and heated for one hour at 43°C, while at the same time skin and muscle blood flow increased to 3 or 4 times the value at normothermia. Only in smaller tumors did the blood flow increase during one hour heating at 45°C. Blood flow in the larger tumors was significantly decreased at three hours after a heat treatment of one hour at 45°C.

In the case of SCK tumors (i.e. a mammary adenocarcinoma) which were implanted subcutaneously in mice, they demonstrated marked slowing of blood flow at temperatures as low as 41°C ceasing completely at 45°C. Hence, it is reasonable to postulate that different tumors (or different species) may be more or less sensitive to hyperthermia depending on the vulnerability of their vascular system or of the malignant cells in the tumor system.

The pH changes demonstrated by Song et al. (1980) correlated fairly well with the vascular sensitivity at various temperatures. At effective temperatures a fall in pH was observed. This would show partial return to prehyperthermia values during a recovery period. Further falls were seen when

heating was reapplied.

MILIEU INTERNE OF TUMORS

In view of the importance of the acid/base and oxygenation state of cells exposed to hyperthermia, a number of studies have been carried out into the pH and pO2 status of tumors both under normothermic and hyperthermic conditions. Bicher et al. (1980), working microelectrode systems with subcutaneously implanted C3H mouse tumors, confirmed that, in general, tumor pO2 is very inhomogenous, being extremely low in some areas that are at the same time, at a very low pH. They showed that though there was an initial rise of pO2 with temperature in tumor, muscle and brain tissue, the 'break point' (i.e. the temperature above which the pO2 fell precipitously) was much lower in tumor than in the other two tissues. They also demonstrated, using hydrogen clearance methods, a fall in blood flow occurring at or near the breakpoint. The same authors demonstrated that the average interstitial pH of tumors was 6.8 and this value was decreased to a mean value of 6.2 following one hour of 43°C hyperthermia.

Vaupel (1979) reporting results of other investigations (Vaupel et al. 1974; Vaupel 1976; and Vaupel 1977), has demonstrated that oxygen consumption, per gram of tumor, decreases when a tumor becomes larger. This is accompanied by an exponential decrease in total blood flow per unit weight (Vaupel 1974). Under conditions of slightly increased temperature, Vaupel et al. (1977) demonstrated that total tumor blood flow and oxygen consumption (in DS carcinomas implanted in rat kidneys) rose significantly when the temperature was raised from 37°C to 39.5°C. When the temperature was further raised to 42°C, the values of both these parameters fell to below those occurring at

37°C.

Vaupel (1979) using polarographic methods and oxygen microelectrodes of 2-5 microns tip diameter has demonstrated that (in all but most superficial areas of 0-180 microns depth) marked differences of pO2 exist only in the region of blood vessels. For the most part a monotonous pattern of very low pO2 predominates. Respiratory hyperoxia, as expected, had little effect on the oxygen tensions in tumor tissue, and haemoglobin oxygen saturations were only slightly increased from the very low values obtained at normoxia (Vaupel et al. 1978; Vaupel et al . 1980).

Vaupel (1980) during a study using one micron tip diameter pH microelectrodes has essentially confirmed the work of Bicher et al. (1980) by demonstrating that exposure to 43°C for one hour could reduce the pH value in experimental tumors by a mean of 0.54 units as compared with preheating values. He also noted that the pH of large tumors (mean 7.21) was distinctly higher than small tumors (mean of 6.75). Von Ardenne and Reitnauer (1980) have published similar evidence for a decrease in pH following hyperthermia.

Reports on pH in human tumors are scarce. Naeslund and Swenson (1953) reported 5 pH measurements in gynaecological tumors; the mean value was 6.94. Pampus (1963) reported a mean value of 6.84 in miscellaneous brain tumors, whereas Meyer et al. (1948) working on specimens after surgical removal obtained very low values of 5.54 - 6.75. A recent study by van den Berg et al. (1982) have reported mean pH values of 7.29 in mammary tumors in humans. Although these values were higher than those reported in other publications they were, never-the-less significantly less than subcutaneous values (mean value of 7.63). The same authors using the same electrode systems reported a mean pH of 7.15 in 24 measurements of rat Rhabdomyosarcomas and a mean of 7,59 in rat muscle.

Faithfull et al. (1980) have reported the results of pH monitoring under Whole Body Hyperthermia at 41.8°C. In the few cases that they reported they

observed no significant overall change in the pH as a result of hyperthermia though temporary changes could sometimes be induced by variations in arterial blood pressure, cardiac output and pCO2. Bicher and Mitagavaria (1981) have reported pH and pO2 changes in malignant tissues of mice under local hyperthermia. They found a decrease of pH of 0.5 - 1 unit following one hour treatment at 43°C. PO2 decreased to very low levels at 46°C. A sharp decrease in blood flow occurred at around the 'breakpoint' for oxygen (43°C). This would indicate that circulatory failure might well be the cause of the hypoxia that they observed.

Glucose uptake in malignant tumors has been measured by a number of authors, but always in 'tissue isolated' tumors. Vaupel et al. (1980) using DS carcinoma in rats have found that glucose uptake is significantly increased at 39°C and returns to the initial normothermia level after further heating and temperature stabilization at 42°C. Arterio-venous differences and glucose extraction did not change under constant hyperthermia for 30 minutes. The same is true for lactate release. The changes were similar in both large and small tumors. However, Vaupel et al. (1976) have shown that the uptake of glucose per gram of tissue is less in large tumors than in small ones. On the other hand, Gullino et al. (1978) have failed to demonstrate any consistent change in glucose uptake under hyperthermia of 42°C for one hour. These authors used Walker 256 and MTW9A tumors in rats. This would once more indicate differences in hyperthermic metabolism in different tumor systems.

The phenomenon that decrease of interstitial tumor pH occurs following massive intraperitoneal administration of glucose has been known for many years (Eden et al. 1955). This is attributed to build-up in the tumor of lactic acid produced by anoerobic glycolisis due to an inadequate circulation to the malignant tissues. This phenomenon has recently been demonstrated in Rotterdam (van den Berg, personal communication) and the drop in pH following the administration of glucose was as much as 0.6 pH units. This phenomenon is not observed

with other sugars such as glactose or fructose (Eden et al. 1955, Voeghtlin et al. 1935).

This hyperacidification of tumors occurring in the presence of increased anaerobic glycolysis is part of the central concept of the 'cancer multi- step therapy' (van Ardenne and Reitnauer 1980a; von Ardenne and Krüger 1980). The build-up of lactic acidosis is said to occur predominately at the venous end of the capillaries and, due to the stiffening of erythrocytes under the influence of low pH (Smid-Schönbein et al. 1973), vascular occlusion occurs. Vaupel et al. (1980) have confirmed that this mechanism can be operative in areas of low pH (6.4) under conditions of 42°C hyperthermia. Sluggish flow in the capillary beds would then tend to increase relative viscosity of the blood and under conditions of low sheer (low blood velocity) stasis would occur. This would lead to further acidification under the influence of anerobic glycolysis and so retrograde stiffening of the erythrocytes will occur. This is said to produce virtually complete destructon of the tumor circulation (von Ardenne and Reitnauer 1980).

CHAPTER IV

HEATING TECHNIQUES

Basically, heating techniques for whole body hyperthermia (WBHT) in use over the last few years divide into two types. In the first technique the skin is used as the major heat exchange organ. Blood is warmed by flowing through the heated and vasodilated skin and the heat is thus distributed to the rest of the body. The lungs may also be used as a secondary heat exchanger. A second method is warming by use of an extracorporeal circulation circuit and heat acquisition by passage through an extracorporeal heat exchanger. Different heating techniques will be briefly described while the technique using the Siemens cabin in use in Rotterdam will be described in more detail.

Having considered the section on thermobiology in this thesis, it may reasonably be asked why a treatment temperature of 41.8°C is used for Whole Body Hyperthermia Treatment when considerable evidence has been presented that maximum benefit in terms of cellular destruction takes place at considerably higher temperatures. The reason for choosing this temperature is partially explained in Chapter I where it was pointed out that the intact animal is more sensitive to high temperatures than its constituent tissues. It was pointed out by Shibolet et al. (1976) that 42°C should be taken as the point above which survival in man may be jeopardized. In dogs this level is probably between 43°C and 44°C (Bynum et al. 1977; Shapiro 1973). Evidence will be presented in a later chapter indicating that 41.8° - 42°C is the point above which in man unacceptable organ toxcity - in particular liver hepatic toxity, begins to appear.

The question arises as to if in fact 41.8°C is a temperature at which

therapeutic gain can be expected. As pointed out in the preface, this thesis is restricted to a discussion of the short term effects of WBHT but, suffice it to say that numerous studies of WBHT revealed that clinical benefit may be gained from treatment in the 41.8° - 42°C range of treatment temperatures (Pettigrew et al. 1974b; Larkin et al. 1977; Moricca et al. 1979; Bull et al. 1979; Parks et al. 1979; Herman et al. 1982, and many others).

Hot Wax

This technique was devised by Henderson and Pettigrew (1971) in Edinburgh, who introduced the revival of interest in WBHT in recent years. A mixture of paraffin wax B.P. (melting point 43°C) is heated to a temperature of 45°C. The anaesthetised patient is placed in a bath-like container and the molten wax is poured over him. The wax, as it comes into contact with the cooler skin surface, begins to solidify and, in so doing, the latent heat of fusion is released, producing yet more heat to warm the blood flowing through the skin without increasing the temperature.

The patient may produce sweat under the influence of the rising body temperature, but this sweat is prevented from evaporating by the wax which forms a seal against the surface of the body. A further advantage of wax is that the body of the patient tends to float in it and hence pressure points are reduced to a minimum. Pressure necrosis and burns in certain points, for instance the wrists, elbows, heels etc may easily occur following Whole Body Hyperthermia treatment and are caused by occlusion or reduction of blood supply due to pressure on skin that, due to its raised temperature, probably has a greater oxygen consumption than normal and hence a greater susceptability to damage.

Using this method it is possible to raise the temperature to 42°C in one hour (Pettigrew and Ludgate 1977). The plateau temperature is maintained by

allowing the body to come into thermal equilibrium by removing or peeling back portions of the solidified wax and allowing a greater or lesser degree of evaporation of sweat from the skin surfaces so exposed. The hot wax method has been applied by MacKenzie et al. (1975); Pettigrew et al. (1974); and Pettigrew and Ludgate, (1977).

A slightly less messy modification of the original technique consists of first placing the patient in a large plastic bag - a not inconsiderable undertaking when the patient is anaesthetised! This modification has been practised by Blair and Levin, (1977) and Greenlaw et al. (1980). A disadvantage of the modification is that access to the patient is somewhat more limited than in the original wax-only technique.

Hot Water

a) Immersion

A logical progression in the total emersion theme is to use water in place of the wax in the 'Pettigrew technique' (as this has become known). Water emersion has been used by Versteegh et al. (1980) and Losheck et al. (1981). The latter placed the patient in a plastic bag to prevent maceration of the skin following prolonged contact with water.

Water has a number of advantages over wax. It is cheap, and freely available and its temperature can be rapidly changed by use of a thermostatic mixer tap. It is essential to ensure good mixing of the water circulating through the bath to avoid 'hot spots'. A further advantage of water, shared with the Pettigrew technique, is the buoyancy of the patient in water and prevention of possible pressure points. A disadvantage, or rather a slight complexity of the water technique (and presumablly also the wax technique) is that pressure transducers used in vascular pressure monitoring must be caliberated with reference to the depth of water or wax above the patient

(Versteegh et al. 1981). When draining the water at the end of treatment care should be taken to ensure that the decrease of external pressure from the surrounding water does not result in the production of a situation in which the veins of the patient dilate and 'hypovolemic shock' is produced (Pomp, personal communication). Using this technique the body temperature can be raised to 41.8°C in approximately 50 minutes depending on the build (and hence surface/volume ratio) of the patient (Losheck et al. 1981).

b) Heating blankets and heating suits

The use of heated water circulating blankets to produce WBHT has been employed by Larkin et al. (1977), Barlogie et al. (1979), Moricca et al. (1979), Larkin (1982), and Herman et al. (1982). The techniques employed are all roughly the same in that the anathetised patient is wrapped in water circulating blankets, the temperature of which is adjusted to a temperature warm enough to ensure rise in core temperature but cool enough to prevent the occurrence of skin burns. Using this technique heating times vary from 1½-2 hours (Larkin et al. 1977), to a maximum of 2.6 hours (Barlogie et al. 1979). Water temperatures vary from 42°C (Barlogie et al. 1979) to 50°C (Morrica et al. 1979).

Heated water circulating suits, in which the patient is entirely enclosed in the suit apart from the head, hands and feet, have been employed by Bull et al. (1979); Ostow et al. (1982); and Lees et al. (1980). The latter authors also used an insulating blanket over the patient during treatment and achieved a warming rate of 3°C per hour, which is about the same as reported by other groups of investigators.

Hot Air

a) Using the lungs as a heat exchanger

In an early report, Henderson and Pettigrew (1971) reported a technique of warming the respiratory gases by utilization of large amounts of heat produced by the absorption of carbon dioxide by 'soda lime' (calcium hydroxide and sodium hydroxide) in a closed anaesthetic circuit. The same soda lime is used for absorption of the carbon dioxide expired by the patient as for the carbon dioxide that is added from outside the circuit.

Using this technique, in combination with the wax method, the authors were able to raise the body temperature by 5-6°C in 20 to 40 minutes.

b) Using the skin as a heat exchanger

Transfer of heat to the surrounding air is one of the principles of the Pomp-Siemens Hyperthermia Cabin which will be described in greater detail in a later section. Basically, this is a semi-sealed chamber in which the patient lies exposed to hot air produced by two 1.2 Kw heaters. The air temperature is controlled by a thermostat and is recirculated by axial ventilation in the cabin.

This method, with or without the additional use of a 400 watt, 27 MHZ generater, has been used by a number of investigators including Pomp (1975); Neumann et al. (1974); Wüst et al. (1975), Reinhold et al. (1982); van der Zee et al. (1979) and Faithfull et al. (1982).

Heating times vary depending on the air temperature of the cabin (this is usually kept at 50-60°C), the temperature to be reached and the use of the RF generator. Neuman et al. (1979) using RF heating reached 40.5°C in about 10 minutes. Others using the standard heating coils report that a mean time of about 165 mins is required to reach 41.8°C (van Rhoon, personal communication).

Radiation Heating

Heating using infra red radiation has been used by Heckel (1975). The apparatus employed was able to raise the body temperature to only 40°C in about 2½ hours. This method is generally believed to be unsuitable.

Pyrogen Therapy

One of the pioneers of cancer therapy by hyperthermia was W.B. Coley who introduced a method of producing Hyperthermia by injection of a pyrogenic vaccine containing a mixture of bacterial products (Coley 1893). The results were very unpredictable, though some good results were obtained in securing remissions of the disease process.

Fever therapy has been intermittently applied since then in the treatment of malignancy. Warren (1935) used a combination of pyrogenic fever and external heating (produced by radiation heat from carbon filament lamps) to achieve a treatment temperature of 41.5°C. Shoulders and Turner (1942) combined fever therapy and deep x-ray therapy. In some clinics this method of production of hyperthermia was recently still in use (Nauts 1975).

Extracorporeal Circulation

Initial reports of the use of extracorporeal circulation through a heat exchanger to induce hyperthermia appeared in 1979 (Parks et al. 1979). Other investigators subsequently using the method include; Sutherland (personal communication), Hermann et al. (1982), Bull et al. (1982), Eisler et al. (1982). The technique is usually accomplished by establishment of a surgically prepared arteriovenous shunt in the ingunal region. Blood is then allowed to flow through the heat exchanger. Oesophageal temperature can be raised to 41.5°

in as little as 22 minutes (Parks et al. 1979) though Herman et al. (1982) report a mean warming time to 42°C of 1.3 hours.

THE ROTTERDAM TECHNIQUE

This technique was developed over the course of 45 treatments under Whole Body Hyperthermia at 41.8°C and is based on the use of the Pomp-Siemens Cabin - a diagram of which is presented in Figure 1.

Description of the Pomp-Siemens Hyperthermia Cabin

The patient lies on a mattress filled with granules (1-2 mm in diameter) of expanded polystyrene which is situated in the centre of the cabin (Pomp 1965). The granules ensure reduction of pressure points. On each side and at the foot end is a space about 10 cm wide extending down and below the level of the patient. This is closed by a perforated plywood board which covers the space in which the 1,2 KW heaters are located. These heaters are elongated and are situated one on each side of the patient. At the foot end near the top of the cabin (the 'roof' and sides are made of thick perspex), is a temperature sensor for the thermostat. Air is circulated through grills in the plywood cover at the foot end by an axial ventilator. In this way hot air, which rises through the perforated board at the sides of the cabin, is circulated slowly around the cabin (velocity less than 0.5 m/sec).

The patient's heads extends outside the cabin and the semicircular opening in front of the neck of the patient is closed with towels - a semicircular clamp is present for this purpose. Full access to the head of the patient is thus ensured. Further access to the patient is provided through circular

openings about 15 cm in diameter that are provided along the sides of the cabin - these are closed by small sliding doors when not in use.

The Siemens cabin, as delivered, also contains a radiofrequency system of heating. A coil about 40 by 50 cm is incorporated into a small mattress which is positioned under the trunk of the patient. This 27 MHZ coil can be connected to a 400 watt radiofrequency generater and can thus provide additional heating. Also supplied is a 433 MHZ microwave applicator which can be positioned above the top of the cabin (the top of which consists in part of a sort of roller blind which can be opened). After connection to a 250 watt generator additional local heating can be administered. The above mentioned heating systems were not employed due to the unacceptable degree of interference caused to all monitoring equipment.

Patient Treatment Regimes

In the first treatments, the only form of heating which was used was supplied by the two 1.2 Kw heating elements already described. The air temperature was maintained at 50-60°C. Using this form of heating alone patients were heated to 41.8°C in a mean time of 165 minutes or at a rate of 1.75°C per hour.

In an attempt to reduce cooling of the patients by evaporation of the large quantities of sweat produced during the procedure, the patients' exposed skin was covered in later treatments with sheets of thin plastic film. A regime of heating and humidifying of the respiratory gases to 42-43°C and warming of all infused fluids to 42°C was also instituted in an attempt to reduce warming time as it may well be advisable, from the point of view of tumor thermotolerance development, to warm the patient as quickly as possible. Using hot air in combination with plastic film and heating respiratory gases and infusion fluids, the mean speed of warming was increased to 1.9°C per hour. A small decrease in warming time resulted (mean of 165 minutes to mean of 150 minutes).

A further and more marked improvement of warming speed was produced by the

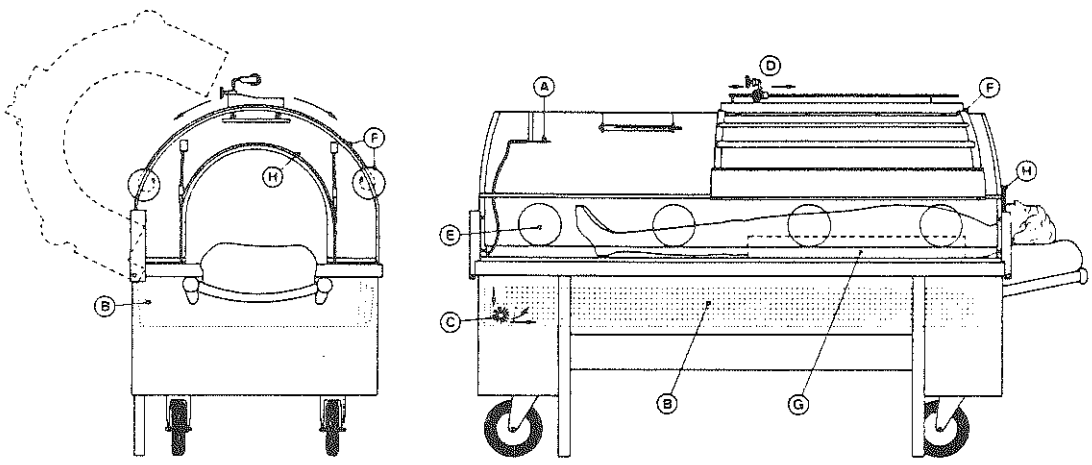

Fig. 1. The Pomp Siemens Hyperthemia Cabin. Size of the main body (without head holder) 187 cm in length and 92 cm in width.

A = Sensor for thermostat.
B = Air heating elements.
C = Axial ventilator (not shown in front-view diagram).
D = Adjustable holder for microwave applicators (applicators not shown).
E = Ports for handling, infusions, etc. (sliding doors not shown).
F = Adjustable plastic roller top.
G = Position for radiofrequency coil under patient (not shown).
H = Holder for sealing cloth.

use of a water circulating mattress placed under the patient. Warm water at a maximum temperature of 46°C was circulated through the mattress using a Churchill Thermocirculator LTSM thermostatically controlled heater circulator. Using the addition of this heating method the mean heating rate was increased to 2.6°C per hour and the mean warming time was reduced to 108 minutes. A further advantage of the addition of plastic film, heating of inspiratory gases, heating of infusion fluids and the incorporation of the warming blanket is evident, as van Rhoon and van der Zee (1981) have demonstrated that, using this regime the uniformity of temperature distribution is much better during the plateau of 41.8°C.

During the plateau phase of treatment the temperature of the patient is easily regulated by lowering the temperature of both the water in the circulating mattress and the hot air inside the cabin. Excessive rise in temperature can be easily adjusted by opening of the access windows or the roller blind at the top of the cabin. In the case of falling temperature the temperature of the circulating water can be quickly adjusted.

Cooling of the patient at the end of plateau phase occurs rapidly once the cabin is opened and the plastic film is removed. If necessary, further cooling by suitably placed fans can be undertaken. Excessive cooling of the skin, for instance by swabbing with alcohol solutions, should be avoided as this may actually slow cooling of the body due to vasoconstriction of the skin blood vessels resulting in decreased heat dissipation.

It is, as mentioned above, probably desirable to heat the patient under WBHT as fast as possible and it may be that future WBH treatments should be carried out using one of the faster methods of heating such as water immersion or extracorporeal circulation. To what extent the physiological changes that occur as a result of heating are dependent on the heating method employed remains to be elucidated.

CHAPTER V

ANAESTHESIA

It is very difficult to describe in any detail a 'typical' anaesthetic administered to a group of patients. Every administration of an anaesthetic is inevitably tailored to the requirements and reactions of the individual patient in question. An anesthetist is constantly trying, by pharmacological means, to steer the physiological state of the patient in the direction in which it should be progressing - or at least in the direction in which the anesthetist in question considers that it should be progressing.

A final physiological measurement that may be produced in, for instance, analysis of the cardiovascular state of the patient at one particular point in the hyperthermia treatment, may very well represent the outcome of opposing forces. The thermophysiological reflexes of the patient may be forcing some parameters in a direction completely opposite to that in which the anaesthetist considers to be desirable.

For instance, consider the mean systemic arterial pressure during the cooling phase of the treatment. As will be seen later, there are very significant falls in pressure at a point 15 minutes after the initiation of cooling. This is a physiological reaction of the patient. The anaesthetist, who is constantly monitoring these changes, may consider that the pressure has fallen (or is about to fall) to unacceptably low levels and may attempt to counteract this fall by rapid intravenous infusion of crystalloid or colloid solutions.

The end result that occurs in the data of 'the' patient (actually the mean etc of all patients) is thus a momentary glimpse, at one spot in time, during the continuously changing physiological state of the patient, under the very

unphysiological conditions of anaesthesia and hyperthermia treatment.

Preoperative Preparation

The patients were all seen by the anaesthetist on the evening before treatment. A final assessment of their general fitness, and in particular their fitness to undergo the proposed treatment, was carried out. Over the previous weeks all patients had undergone a vigorous 'workup' with particular regard to their cardiorespiratory systems.

Where possible, all patients had undergone extensive pulmonary function tests and, with few exceptions, all had very reasonable lung function as assessed by pulmonary mechanics and blood gas data. It is of importance at this point to note that, in general, the worse the pulmonary function the greater the likelihood that the patient would require post-treatment pulmonary support and artificial ventilation. It should be stressed that it is absolutely imperative that a well equipped intensive care unit be available, staffed by personnel capable of, and regularly employed in, the treatment of patients with cardiorespiratory disorders.

The cardiovascular system was assessed by the usual means and most patients underwent bicycle ergonometry. They were required to perform at 120 watts. This high requirement, (or aimed for performance - in the later stages of the trial patients were not required to perform as well), was initially set because of the high degree of stress that the cardiovascular system was expected to undergo. This stress has been commented on in the literature (Euler-Rolle et al. 1977; Bull, 1982) mostly however, without production of figures for cardiac work etc. As will be seen later, our patients did not appear to be greatly stressed from a cardiovascular standpoint. However, in view of the expected stress, no patients were accepted with a history of myocardial infarction or angina pectoris. Patients with hypertension were not initially accepted,

though mild controlled hypertensive patients were admitted to the trial at the end of the series.

All patients underwent isotopic cerebral scanning to exclude secondary malignant deposits in the brain. This requirement was to be dramatically vindicated in one patient whose pretreatment workup was performed in another clinic. The physician in charge, though requested to carry out this investigation, did not consider it necessary and, moreover, neglected to inform the hyperthermia group that it had not been carried out. The day after treatment, the patient developed hemiplegia and a subsequent isotopic brain scan revealed an intracerebral secondary deposit. The hemiplegia subsided after a few days and was hence probably due to odema and not to hemorrhage into the tumor as first feared.

Anaesthesia and Monitoring

The patients received premedication consisting of papaveretum 0.3-0.4 mg/kg body weight and scopolomine 0.004-006 mg/kg body weight. This was administered intramuscularly one hour before the patient was bought to the hyperthermia room.

In cases where a catheter was passed into one of the hepatic veins, the patients were first brought to the x-ray department, where a 7 French 80cm long Cordis femoro-renal A2 catheter was advanced through an 8 French Cordis 501-608 catheter introducer system inserted, under local infiltration anaesthesia, into the left femoral vein in the groin. It was advanced under fluoroscopy until its tip lay in one of the hepatic veins. Through this catheter samples were taken, during the hyperthermic treatment, of hepatic venous blood and central venous pressure was measured with a Hewlett Packard 1280C pressure transducer and displayed on a Hewlett Packard compact monitor 78341A. Many patients were given intravenous diazepam (0,1 to 0,2 mg per kg) during insertion of this, and

other intravascular monitoring lines, which were, when possible, inserted before induction of anaesthesia.

An intravenous infusion was set up, usually in a vein in the dorsum of the left hand, and an intra-arterial catheter was inserted into the radial artery in the left wrist and advanced into the brachial artery. Arterial pressure was measured with a Gould Statham P231D pressure transducer connected to a Siemens E2150 pressure module, displayed on a digital display module E2160 mounted on a Sirecust 323 unit.

A 7 french gauge KMA thermodilution Swan-Ganz catheter was inserted through another 8 french Cordis 501-608 catheter introducer system inserted into the left subclavian vein and the catheter was advanced under pressure monitoring into the pulmonary artery. The pulmonary arterial pressure was measured with a Hewlett Packard 1280C pressure transducer and displayed on the Hewlett Packard compact monitor. This gave digital readout of pulmonary artery pressures and, after inflating the occlusion balloon, the pulmonary capillary wedge pressures.

Cardiac output was measured by thermodilution using a KMA thermodilution cardiac output computer, model 3500. The injectate, which was injected randomly in the respiratory cycle, consisted of 10 ml of 5 percent dextrose solution at room temperature.

Anaesthesia was induced with an intravenous injection of a 1 percent solution of methohexitone (1 mg per kg bodyweight). After muscular relaxation had been achieved using a nondepolarising relaxant, (in most cases d-tubocurarine was employed using an intravenous dosage of 0,5 mg per kg) the patient was intubated with a cuffed endotracheal tube. Intermittent positive pressure ventilation was carried out using a Siemens Elema servo- ventilator 900A with a mixture of 33% oxygen and 66% nitrous oxide. The frequency of ventilation was kept at ten inspirations per minute and the tidal volume was adjusted to obtain an initial end tidal expired carbon dioxide concentration of between 3 and 4 percent.

Every 10-15 minutes the following measurements were taken:- heart rate, systolic, diastolic and mean arterial pressures, cardiac output, systolic, diastolic and mean pulmonary artery pressures, pulmonary capillary wedge pressures and central venous pressure. Measurements were also taken of expired carbon dioxide percentage, and carbon dioxide minute production using a Siemens Elema carbon dioxide analyser 930. The electrocardiogram was continuously monitored.

At the end of the hyperthermia treatment, the non-depolarising muscle relaxants, which were administered when necessary throughout the treatment in small intravenous increments of 0,1 to 0,2 mg per kg bodyweight, were reversed using neostigmine 0,3 mg per kg preceeded by atropine 0,16 mg per kg. The patients were then removed from the cabin and transferred to the intensive care unit.

Modification of Standard Anaesthetic Technique

As mentioned above, most patients were maintained under nitrous oxide and oxygen with administration of intermittant doses of the muscle relaxant d-tubocurarine as required. In some patients pancuronium bromide was used as the relaxant. This made very little difference to the pattern of changes in the physiological state of the patient induced by the hyperthermia treatment. This is what one would expect when it is realised the enormously greater degree of vasodilatation of the cardiovascular system bought about by the hyperthermia in comparison to that bought about by d-tubocurarine (in comparison to pancuronium).

Some patients received low concentrations of halothane (0.25-1%) or enflurane (0.5-1.5%) during treatment. Again these concentrations had little effect on the overall cardiovascular status of the patients. It was only when the patients were maintained solely on nitrous oxide-oxygen and halogenated

volatile anaesthetic agents using no nondepolarising muscle relaxants that the effects of these agents were noticable. In these cases - one patient had enflurane and one had halothane - we observed lower than normal mean values for arterial pressure and cardiac output during treatment.

One patient received large quantities of fentanyl in an attempt by an anaesthetist, other than the author, to control tachycardia. This resulted in hypotension requiring treatment with dopamine infusion. It could, of course, be argued that this was coincidental and that the patient had an inherant tendency to become hypotensive. However, the same patient received hyperthemic treatment one week later under a 'standard' (non fentanyl containing) anaesthetic and had no hypotensive problems.

It is the conclusion of the author that patients react best when a minimal number of medicaments are administered.

No cases of awareness under anaesthesia occurred in the present series. Wilson and Turner (1969) have advocated the use of opiate premedication in the prevention of awareness, though it has been pointed out that this is not always effective (Faithfull 1969). Most patients had amnesia for at least the first 4 or 5 hours of the post-treatment period, and sometimes for 12 hours or more. The relatively heavy premedication was given in order to provide: a) sedation, b) analgesia during the introduction of monitoring lines, and c) amnesia for the rather strange and dramatic experience of being placed in, (what must seem to most patients), an apparatus resembling a glass coffin.

It is interesting to note that the requirements for relaxant drugs appear to be very much increased during the hyperthermic period and the early cooling phase of treatment. The same remark applies to the volatile agents and the two patients, already alluded to the above, required progressively higher concentrations of fluothane and enflurane to allow them to tolerate intermittent positive pressure ventilation.

Increased requirement for anaesthetic agents has been commented on by

Pettigrew and Ludgate (1977). At first sight it would appear reasonable to suggest that there might be increased excretion or breakdown of drugs under hyperthermic conditions. However, urinary excretion was certainly not increased in these patients as the majority were anuric at this point in treatment. It is doubtful if hepatic destruction of drugs is increased in view of the probable decrease in hepatic metabolism occurring under hyperthermia and the fact that hepatic blood flow and oxygen consumption are significantly reduced under these conditions (Faithfull 1982). The hepatic effects of hyperthermia are considered later.

Fluid balance

Fluid balance under anaesthesia for hyperthermic treatment has so far not been discussed. The maximum sweating rate in man is very high and may amount to a litre or more per hour (Scott 1966) and it would seem reasonable to suggest that patients undergoing treatment would lose vast quantities of fluid. It was, however, very rapidly recognised that if an attempt was made to replace fluid with crystalloid solutions (on the basis of calculation of losses) the patients became oedematous. This overload with water was commented on by Mackenzie et al. (1975) who tried (unsuccessfully) to use the Central Venous Pressure as a guide to fluid replacement.

Oedema occurring under hyperthermia is usually first noticable in the conjunctiva. The fingers may become swollen (a further good reason for insistence on removal of rings etc) and in extreme cases the face becomes very swollen and oedematous. The first case anaesthetised by the present author developed massive pharyngeal oedema. This was due to attempts to maintain systemic arterial pressure and replace all calculated losses with crystalloid solutions. Pulmonary oedema was never observed in our patients. Herman et al. (1982), also commented on the 'capillary leak' syndrome and found increases in alveolar-arterial oxygen differences at the end of treatment which they

attributed to pulmonary oedema. Our alveolar arterial pO2 gradients showed no changes as can be inferred from Table III, chapter VII. Arm and leg oedema has been commented on by Ostrow et al. (1981) as occurring in all patients and resolving within 48 hours of the therapy.

It is of course possible to attempt to counteract the development of oedema by increasing urine output by the use of diuretics such as frusemide. This however, only brings on the necessity of administering yet more fluid, if the dogma of fluid replacement is followed.

Urine output usually falls rapidly as plateau temperatures are reached and at the end of the hyperthermic phase of treatment at 41.8°C (the 'plateau temperature'), most patients were anuric if no attempts were made to stimulate urine production. There is always a diuresis, beginning at the end of the cooling phase and continuing into the post treatment period. The reason for this may be that receptors in the great veins are stimulated in response to the rising central venous pressure that is seen during cooling. This is discussed later.

Logawnay-Malik et al. (1979) have studied the effects of hyperthermia on renal function in dogs and have shown progressive decreases in renal plasma flow and glomerular filtration rates with progressive hyperthermia (maximum rectal temperature of 42°C) and reversal of these changes as the animals were cooled. These changes they could not wholly explain by the systemic hypotension produced by the hyperthermia and they attributed them partly to antidiuretic hormone release (Hellman and Weiner 1953).

Early in the trial it was noticed that some patients developed burning and blistering on their toes. It was argued that this might be caused by inadequate cooling of the tissues as a result of sluggish circulation in the capillary beds in the extreme periphery of an extremely vasodilated and possibly relatively underfilled circulation. As a result it was decided to administer 500 ml of a Dextran solution (molecular weight 40,000) as warming was commenced

with the aim of: a) 'desludging' the periphery and b) causing an 'osmotic filling' of the circulation. This treatment combined with insulation of the feet under a single layer of flannel decreased the incidence of blistering.

At the beginning of the trial it was noticed that many patients, though apparently awake after the reversal of nondepolarising relaxants were in a very confused state during which no real contact was possible with the patient. They were frequently very restless and in some cases intravenous monitoring lines were accidently pulled out during this phase. Administration of tranquillizers such as diazapam did little to ease the situation. Disorientation following hyperthermia has been reported by Smith et al. (1980) and would appear to be quite widespread (Birch 1980; Sutherland, 1980; personal communications).

An interesting phenomenon was often seen at this stage. The patients would lie in bed quite quietly and would not react until stimulated. They would then appear to have a form of Morrow 'startle' reflex. This may be self-perpetuating in the form of a short jactitating reaction of a almost epileptiform character.

We considered that these behavioural changes might be caused by small degrees of cerebral oedema though no obvious oedema was ever present on inspection of the optic fundi. Cerebral oedema was also not seen in a series of dogs treated at 41,8°C (van Rhoon, personal communication). However, cerebral oedema and congestion is generally present in heat stroke (Shibolet et al. 1976) and hence the policy was adopted of administering 200 ml of 20% manitol solution at the start of cooling. At this stage 500 cc of plasma was infused in an attempt to counteract any excessive falls in systemic arterial pressure that were frequently seen at this stage of treatment. Since this regime was introduced, the instance of restlessness has been much reduced.

The fluid administration that is now given during the 5 to 6 hours of treatment is approximately: 500 ml Dextran, 500 ml plasma, 200 ml 20% manitol and 3-4 litres of Ringers lactate solution.

CHAPTER VI

CARDIOVASCULAR SYSTEM

FIXED POINT MARKER ANALYSIS DURING TREATMENT

In view of the unavoidable differences in rates of warming in different patients it was not feasable to analyse the data on a time base and therefore a fixed point marker analysis system was used. For each patient fixed points in the treatment were marked and measurements were thus analysed for each patient at the same stage of treatment. During cooling, which was usually very rapid, analysis was at fixed time intervals. The markers were chosen as follows: Marker 1 - immediately before induction of anaesthesia; Marker 2 - at the commencement of warming following insertion of thermocouples and covering with plastic sheeting; Marker 3 - on reaching plateau temperatures of 41.8'C; Marker 4 - after one hour at plateau; Marker 5 - at the end of plateau; Marker 6 - 15 minutes after the start of cooling; Marker 7 - 30 minutes after the start of cooling; Marker 8 - 45 minutes after the start of cooling.

Means $\pm$ 1 SEM were calculated for every single marker point. Statistical significance of difference between values at two marker points were tested using Student's t-test (two-sided). To exclude patient variance a paired t-test was used. A '2p' value of less than 0.05 was chosen as the lowest level of significance.

TABLE I Cardiovascular parameters during Whole Body Hyperthermia

	Marker 1	Marker 2	Marker 3	Marker 4	Marker 5	Marker 6	Marker 7	Marker 8
	Before Induction of Anaesthesia	Beginning of Warming	On Reaching Plateau of 41.8'C	After one hr at 41.8'C	At End of Plateau	After 15 mins of cooling	After 30 mins of cooling	After 45 mins of cooling
Body Temperature (°C)	37,10 + 0,12 (20)	37,22 + 0,10 (27)	41,74*** + 0,03 (30)	41,82*** + 0,03 (30)	41,82 + 0,03 (30)	40,52*** + 0.12 (29)	39,45*** + 0,12 (25)	38,89*** + 0,13 (17)
Heart Rate (beats min-1)	92,6 + 4,2 (21)	84,2 + 3,0 (29)	125,6*** + 3,9 (30)	143,3*** + 4,3 (30)	145,6 + 4,1 (30)	140,6* + 4,1 (27)	136,4*** + 3,5 (25)	134,0** + 4,3 (15)
Systolic Systemic Arterial Pressure (mm Hg)	117,8 + 4,7 (23)	106,4** + 4,3 (30)	109,3 + 4,6 (30)	95,4*** + 3,9 (29)	95,7 + 3,6 (30)	90,7* + 3,9 (29)	94,6 + 3,3 (28)	99,9 + 4,6 (17)
Diastolic Systemic Arterial Pressure	69,6 + 2,4 (23)	65,3 + 2,6 (30)	56,5** + 2,3 (30)	50,0*** + 2,1 (29)	50,0 + 2,1 (30)	46,3* + 2,2 (29)	48,8 + 1,7 (28)	51,8 + 2,7 (17)
Mean Systemic Arterial Pressure (mm Hg)	87,8 + 3,7 (23)	81,5 + 3,4 (30)	75,4 + 3,1 (30)	66,1*** + 2,5 (28)	65,5 + 2,4 (30)	61,4** + 2,6 (29)	65,6* + 2,0 (28)	68,2 + 2.9 (17)
Cardiac Index (L min-1 m-2)	4,19 + 0,37 (20)	3,33** + 0,33 (22)	7,50*** + 0,41 (22)	7,02 + 0,36 (22)	7,34 + 0,32 (21)	6,82 + 0,45 (21)	6,62 + 0,30 (22)	6,32* + 0,41 (13)
Stroke volume Index (ml)	45,3 + 3,2 (19)	36,5** + 3,3 (21)	59,2*** + 3,0 (22)	49,4*** + 3,0 (22)	49,8 + 2,6 (21)	47,9 + 2,7 (19)	48,4 + 2,3 (19)	47,7 + 2,1 (11)
Pulmonary Capillary Wedge Pressure (mm Hg)	5,9 + 0,9 (20)	8,1** + 0,7 (26)	8,7 + 0,6 (26)	8,5 + 0,7 (24)	8,5 + 0,5 (22)	9,7** + 0,6 (26)	11,0** + 0,7 (22)	10,0 + 0,7 (13)

Left Ventricular Work Index (Kgm m min-1)	4,8 ± 0,5 (20)	3,4** ± 0,3 (22)	7,4*** ± 0,6 (22)	5,7*** ± 0,4 (21)	5,9 ± 0,4 (20)	5,3 ± 0,6 (20)	4,9 ± 0,3 (20)	5,3 ± 0,4 (11)
Central Venous Pressure (mm Hg)	3,0 ± 1.09 (8)	5,7** ± 1,2 (11)	6,8 ± 1,3 (11)	7,5 ± 1,5 (11)	7,7 ± 1,4 (10)	8,4* ± 1,5 (10)	11,9* ± 1,7 (8)	11,8 ± 1,4 (4)
Systemic Vascular Resistance Index (dynes sec cm-5)	2126 ± 276 (8)	2639 ± 215 (11)	880*** ± 55 (10)	791* ± 63 (10)	783 ± 91 (10)	831 ± 91 (9)	772 ± 78 (8)	783 ± 141 (4)
Systemic Pulmonary Arterial Pressure (mm Hg)	23,1 ± 2,0 (18)	20,2** ± 1,4 (23)	24,7*** ± 1,7 (23)	24,4 ± 1,5 (21)	22,6 ± 1,6 (17)	24,6 ± 4,2 (15)	26,4 ± 1,2 (13)	24,9 ± 2,7 (7)
Diastolic Pulmonary Arterial Pressure	8,6 ± 1,0 (18)	9,8 ± 0,5 (23)	10,5 ± 1,0 (23)	10,6 ± 1,2 (21)	10,4 ± 0,9 (17)	11,9 ± 1,3 (15)	12,5** ± 0,8 (13)	13,1 ± 1,1 (7)
Mean Pulmonary Artery Pressure (mm Hg)	14,1 ± 1,1 (20)	13,7 ± 0,8 (25)	16,4* ± 0,9 (25)	15,8 ± 1,2 (23)	15,8 ± 1,0 (19)	17,3 ± 1,3 (19)	18,3 ± 1,3 (16)	17.4 ± 1,5 (9)
Right Ventricular Work Index (Kgm m min-1)	0,56 ± 0,10 (8)	0,31** ± 0,06 (11)	1,11*** ± 0.15 (10)	0,88 ± 0,12 (11)	0,84 ± 0,08 (10)	0,83 ± 0,18 (8)	0,79 ± 0,12 (8)	0,77 ± 0,12 (4)
Pulmonary Vascular Resistance Index (Dynes sec cm-5)	178 ± 25 (20)	151* ± 19 (22)	90*** ± 9 (22)	89 ± 14 (21)	91 ± 12 (17)	93 ± 16 (17)	92 ± 16 (15)	107 ± 24 (7)

Means ± 1 SEM. Number of patients between brackets. Significance on paired t-test in comparison with previous marker point.

* = $p < .05$ ** = $p < .01$ *** = $p < .001$

Results

A number of the measured cardiovascular parameters are presented in Table I. Graphical presentation of the changes in the systemic circulation is shown in Fig. 2 and those of the pulmonary circulation in Fig. 3.

It should be noted that the expression 'cardiac index' (cardiac output per m2 of body surface area) has been used throughout this thesis. This was used in an attempt to make comparison possible between individual patients whose surface area lay between 1,52 and 2,12 square metres (Dubois and Dubois 1916). The expression of cardiac output as the cardiac index is quite common practice though this has been criticised by Burch and Giles (1971) who have suggested that it might be more accurate to express cardiac output in terms of a more readily measurable variable such as body weight.

The beginning of the warming period (marker 2) is usually between 45 and 60 minutes after the induction of anaesthesia. When comparing cardiovascular values at this time, with those occurring just before induction of anaesthesia (marker 1), it can be seen that a number of changes have taken place. There has been a reduction in the cardiac index, and, because there has been no significant fall in heart rate, a fall in stroke index must have taken place. The fall in mean arterial pressure is negligable though the systolic pressure has fallen significantly. There has been a fall in left ventricular work index. Though there have been no marked changes in mean pulmonary artery pressure, the pulmonary vascular resistance has fallen and right ventricular work index is also very significantly decreased. Both pulmonary capillary wedge pressure and central venous pressure have increased in comparison to the preinduction period.

By the time that plateau temperature of 41.8°C has been reached (marker 3), a number of very pronounced changes have occurred. There has been a large and highly significant increase in both cardiac index and heart rate. Because the

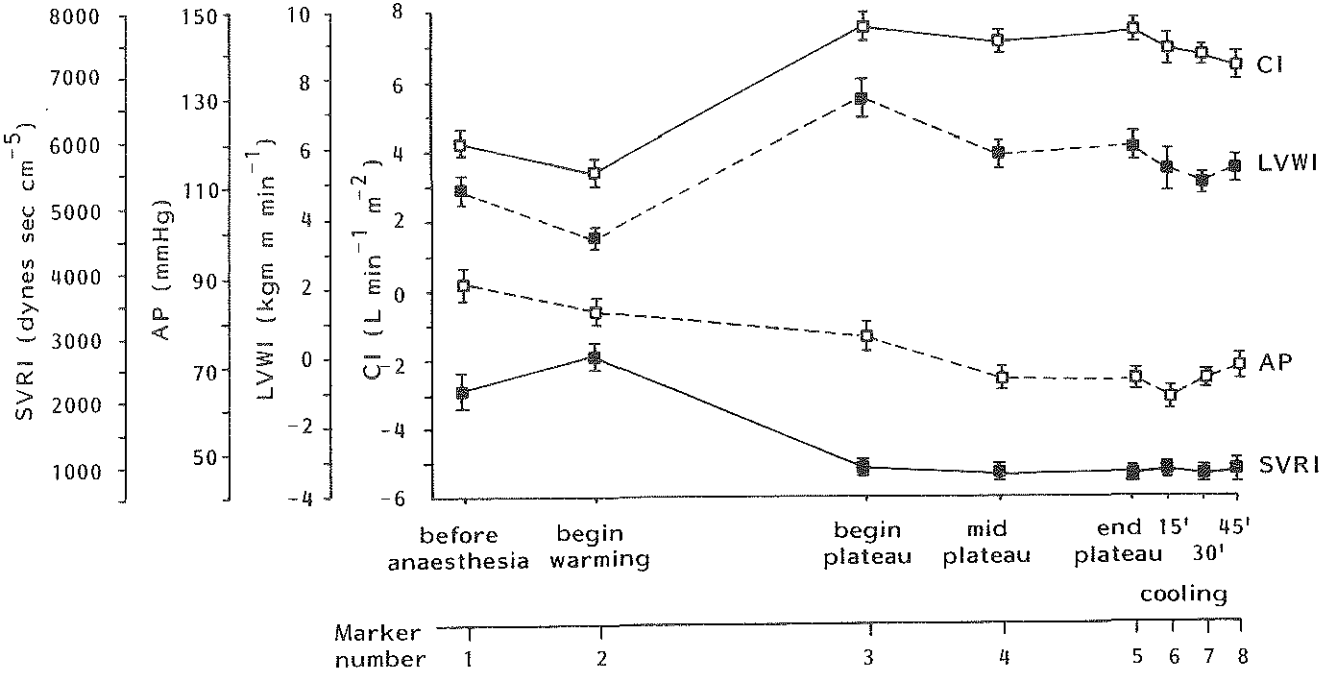

Fig. 2. The effects of Whole Body Hyperthermia (2 hrs at 41.8°C) on a number of variables in the systemic circulation. From top to bottom cardiac index (CI), Left ventricular work index (LVWI). Mean systemic arterial pressure (AP), and systemic vascular resistance index (SVRI). Means ± SEM.

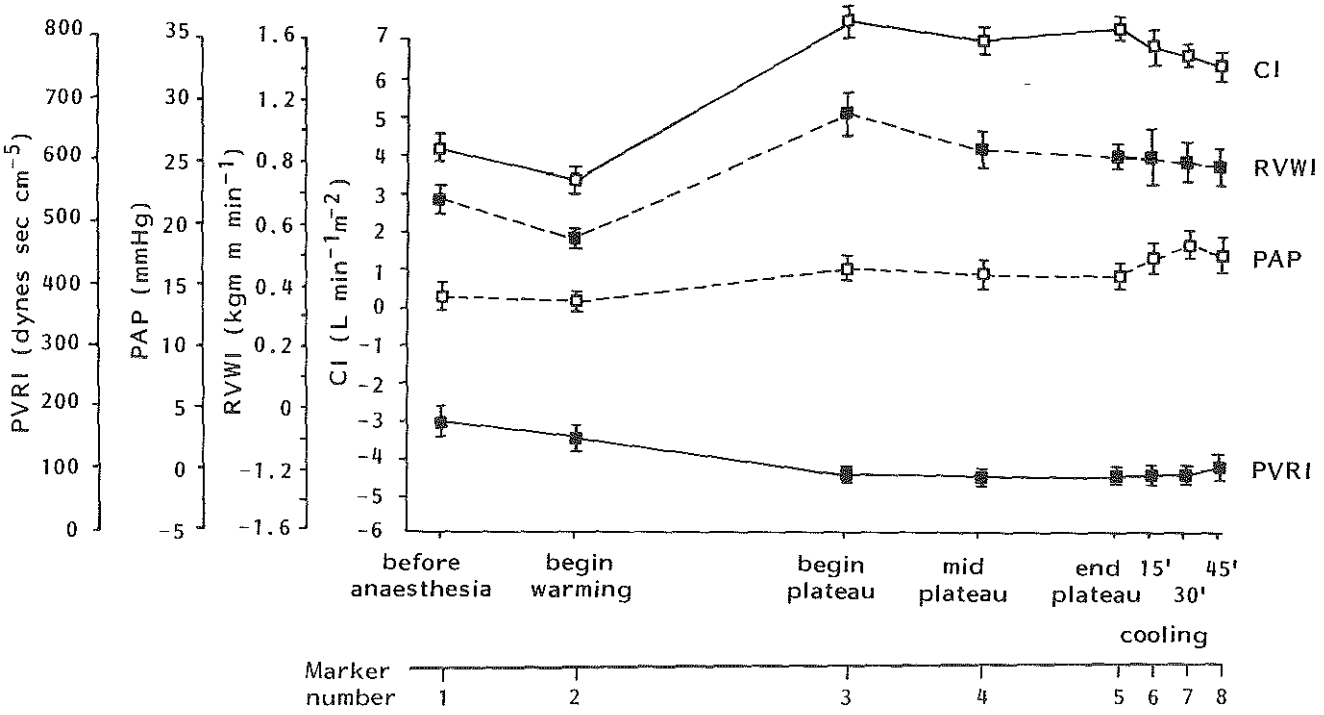

Fig. 3. The effects of Whole Body Hyperthermia (2 hrs at 41.8°C) on a number of variables in the pulmonary circulation. From top to bottom cardiac index (CI), right ventricular work index (RVWI), mean pulmonary artery pressure (PAP), and pulmonary vascular work index (PVRI). Means ± SEM.

cardiac index has increased proportionally more than heart rate, a large increase in stroke index has taken place. There have been no significant changes in either the pulmonary capillary wedge pressure or the central venous pressure and, as would be expected, there were marked decreases in both the systemic and pulmonary vascular resistances. Whereas the systemic resistance has decreased to 34% of its value at marker 2 the pulmonary resistance has decreased to only 54%. Hence, though the mean systemic pressures (especially the diastolic pressures) have decreased, there has been a significant rise in mean pulmonary artery pressures. Both left and right ventricular work indices have increased enormously but whereas the left ventricle has increased its work by 118% the right ventricular work has increased by 275%.

During the first half of the plateau period (between marker 3 and marker 4) many of the cardiovascular changes occurring during warming continue. There is a further fall in the systemic vascular resistance causing very marked concomitant falls in the systolic, diastolic and mean arterial pressures. No changes occur in the pulmonary vascular resistance and there are no marked decreases in pulmonary artery pressures. The cardiac index does not decrease and thus, whereas the left ventricular work decreases to a very significant extent, there is no decrease in right ventricular work. The heart rate continues to rise and this results in a fall in stroke index.

It should be noted that at this moment (marker 4), the left ventricular work is not significantly more than it was before the induction of anaesthesia. The right ventricular work, on the other hand, is very significantly raised above the preinduction values and remains so throughout the plateau phase.

During the second half of plateau (marker 4 to marker 5) there are no significant changes in cardiovascular haemodynamics, but once the cooling period commences, the most obvious and constant change seen is a very significant fall of mean arterial pressure at 15 minutes of cooling (marker 6), followed by a significant rise at 30 minutes (marker 7). Over the same period

the mean pulmonary artery pressure is steadily rising, as are the pulmonary capillary wedge pressures and central venous pressures. The cardiac index does not decrease to any marked extent. There are no changes in either the pulmonary or systemic vascular resistances and whereas the left ventricular work significantly decreases, the right ventricular work remains constant.

During the next 15 minutes of cooling, (marker 7 to marker 8), there has been a fall in cardiac index, but there are no relevant changes in systemic and pulmonary vascular resistances or left and right ventricular work. Every 15 minutes of cooling, a progressive fall in heart rate takes place which, after 45 minutes (marker 8), is not significantly faster than at the beginning of plateau. No changes in the stroke index were observed during cooling.

Changes in systemic and pulmonary pulse pressures are also presented in Table I. In the case of the systemic system a fall in pulse pressure after induction is followed by a marked rise as plateau is reached. Similar changes occur in the pulmonary circulation but, whereas by the time midplateau is reached (marker 4), there has been a very significant decrease in the systemic arterial pulse pressure, the pulmonary pulse pressure does not change. Systemic arterial pulse pressures are not then significantly different from those occurring before treatment (marker 1).

Discussion

When the conscious individual is subjected to thermal stress sufficient to raise the body temperature, considerable anxiety may be caused. Though Rowell et al. (1969) have trained experienced subjects to relax and not to hyperventilate under these conditions this is not usually feasible under whole body hyperthermia treatment. Barlogie et al. (1979), Blair & Levin (1977), Larkin et al. (1977), Parks et al. (1979), Pettigrew & Ludgate (1977) have employed general anaesthesia and Bull et al. (1979), Bynum et al. (1979) and Ostrow et al. (1981) have used a generalised sedation technique. The different techniques

employed may affect the cardiovascular changes taking place, but MacKenzie et al. (1975) have commented that they obtained similar changes in cardiovascular parameters when using different anaesthetic tecnhiques. During one technique, the patients were artifically ventilated while in the other, spontaneous respiration was permitted.

Most authors, commenting on cardiovascular effects of whole body hyperthermia, report changes taking place in heart rate. Though absolute figures given for heart rates found at plateau temperatures vary, fairly good correlation between different reports is obtained when the changes are expressed as change in the number of beats per minute per 1°C rise of body temperature (beats min-1 °C-1). It is questionable if this is justifiable as there may not be a linear change in pulse rate betwen 37° and 42°C. The heart rates reported by various authors have been calculated on this basis and are presented in Table II (assuming basal temperature at the beginning of warming to be 37°C if this is not stated). As pointed out by Pettigrew et al. (1974b), the heart rate continues to rise after reaching plateau temperature and, where possible, both maximum heart rates and those obtaining at the beginning of plateau are presented. Both Pettigrew et al. (1974) and MacKenzie et al. (1975) have pointed out that stable narcosis is necessary for a constant heart rate.

Falls in systemic arterial pressure during whole body hyperthermia treatment have been observed by Dubois et al. (1980), Larkin et al. (1977), Barlogie et al. (1979), Lees et al. (1980) Kim et al. (1979) and Hermann et al. (1982). Rises have been seen by Pettigrew et al. (1974b) and Euler-Rolle et al. (1977). Moricca et al. (1979), Ostrow et al. (1981) and Bynum et al. (1978), on the other hand, found little change in the mean arterial pressures.

Many authors have described a rise in pulse pressure during warming - presumably this is caused by the gross vasodilation that is occurring. The resulting arterial pressures are then due to the overall balance between the

TABLE II Reported values of heart rate changes during Whole Body Hyperthermia Treatment.

Authors	Mean heart rate at start of warming	Plateau temperature	Mean heart rate change at beginning of plateau in beats min-1 °C-1	Mean of the maximum heart rate change during plateau in beats min-1 °C-1
Barlogie et al (1979)	91	42°C		8,1
Moricca et al (1977)	not stated	41.8°C		10,0
Ostrow et al (1981)	105	41.8°C		11,5
Kim et al (1979)	87,6	41.5°C		11,9
Larkin et al (1977)	90	42°C		14,4
Pettigrew et al (1974 b)	not stated	42°C	8,5	11,0
Bynum et al (1976)	96	41.8°C	12,0	14,0
Present study	92.6	41.8°C	9.2	12.8

The changes are expressed in change of number of beats per minute per degree centigrade rise in body temperature (beats min-1 °C-1).

increased cadiac output and the decreased peripheral resistance that occurs.

Surprisingly, the literature on whole body hyperthermia treatment contains very little comment on changes in cardiac output. Herman et al. (1982) reported that, in 4 patients at plateau temperature, cardiac index values were apparently twice base-line. Parks et al. (1979) only mention that cardiac output was always increased averaging 10.8 litres per minute in the 14 determinations made. In patients 'having marginal cardiac function' they measured pulmonary capillary wedge pressures and comment that they were 'little affected'. As no details of systemic pressures were given it is impossible to calculate the systemic resistance or left ventricular work from these figures. Kim et al. (1979) give figures indicating cardiac indices increasing by an average of 72% during warming to 41,5°C; our own figures indicate a 140% rise up to 41.8°C, remaining the same during the plateau temperature. The lower figures from Kims' group may well be attributable to the depressant effect of the continuous infusion of thiopentone and fentanyl that they used. In another publication from the same group of investigators (Bull et al. 1979), in which the patients received either a 'low dose' infusion of ketamine or a similar thiopentone/fentanyl mixture, the cardiac index rose by 118%.

It should be noted at this point that when percentage changes are referred to in results from the present study, these indicate mean paired percentages. Hence the values given may vary slightly from those that the reader may calculate using the figures given in table I.

From the figures of Lees et al. (1980) it may be calculated that, in their patients, left ventricular work increased and systemic vascular resistance decreased by 25% and 51% respectively; their report also showed that a small dose (1.25 mg) of Dehydrobenzperidol could, at 41.5°C, cause an average decrease of 19% in cardiac work and a 16% decrease in systemic resistance. Unfortunately there is no mention of the phase of plateau during which this effect was seen. They noted a return of mean arterial pressure (which had decreased 50%) to

pre-droperidol levels within 5 minutes.

There appear to be very few available publications on the haemodynamic changes taking place in the pulmonary circulation under whole body hyperthermia treatment. The filling pressure of the right ventricle (the central venous pressure) has been fairly widely reported but published results are very variable. Pettigrew et al. (1974b) comment that a rise of 5-10 mm Hg in central venous pressure occurs on warming - values then return to prewarming levels once hyperthermia is established. Lees et al. (1980) noted an average decrease of central venous pressure of 2.6 mm Hg, whereas Moricca et al. (1979) and Herman et al. (1982) noticed no significant changes. In our own patients the only significant changes that took place in the central venous pressure values occurred during cooling. Both Pettigrew & Ludgate (1977) and Mackenzie et al. (1977) comment that central venous pressure is not a good index of the need for fluid replacement. This accords with our own experiences that administration of colloidal solutions in an attempt to maintain central venous pressure at a certain level only leads to interstitial oedema without significant alteration of cardiovascular parameters.

There are few available publications in which pulmonary artery pressure changes were mentioned. Parks et al. (1979) noted that the pressures were little affected, but Lees et al. (1979) reported an average rise in mean pulmonary arterial pressure of 1.3% - It is assumed that this was not statistically significant. The latter publication is the only one available from which calculations of right ventricular work and pulmonary vascular resistance can be made. From the available figures it may be concluded that their patients experienced an average rise in right ventricular work of 110% and a fall in pulmonary vascular resistance of 7%. These results may be compared with the results of calculations using figures obtained in the present investigation (table I), in which the right ventricular work rose by 215% and the pulmonary vascular resistance fell by 50%.

Very few authors have commented on changes occuring during the cooling period following whole body hyperthermia. Bynum et al. (1978) show figures demonstrating no changes in mean arterial pressure half an hour after cooling had started. The cooling rate of their subjects was 2.7°C (rectal) over the first half hour, which corresponds quite well with our rate of 2.4°C (pulmonary artery) over the same period. At this moment the mean arterial pressures of our patients were also not significantly changed though, as we have mentioned above, there was a significant fall after 15 minutes of cooling. Herman et al. (1982) noted precipitous decreases in blood pressure during cooling.

The above mentioned 'dip' effect was also noticed by Rowell et al. (1969) who cooled conscious volunteers (average blood temperature 38.84°C) by rapidly lowering the skin temperature. They also noted, as was seen in the patients in the present study, increases in the central venous pressure associated with, in their subjects, and increase in central blood volume. The increase in stroke volume that they observed was not seen in the present study. In their subjects the experiments were terminated when 'the subjective sensations of thermal stress became intolerable'. It is thus possible that the pulse rates of their subjects were, at that stage, 'artifcially' elevated by psychological stress (the average rate was 149 beats/min) and that they fell rapidly with the psychological relief of cooling - hence the rising stroke volume even in the presence of a falling cardiac output. Our patients were unconscious and were, it was to be expected, free from anxiety - their heart rates fell only 6% (from means of 145.6 to 136.4 beats per minute) in the first half hour of cooling.

In conclusion, attention should be drawn once again to the different relative amounts or work performed by the left and right ventricles during hyperthermia and to reiterate that, though the left ventricular work was not significantly raised above pretreatment levels during the midplateau phase the right ventricular work was very significantly raised at this stage. These important results, which are graphically illustrated in Fig. 4, may have

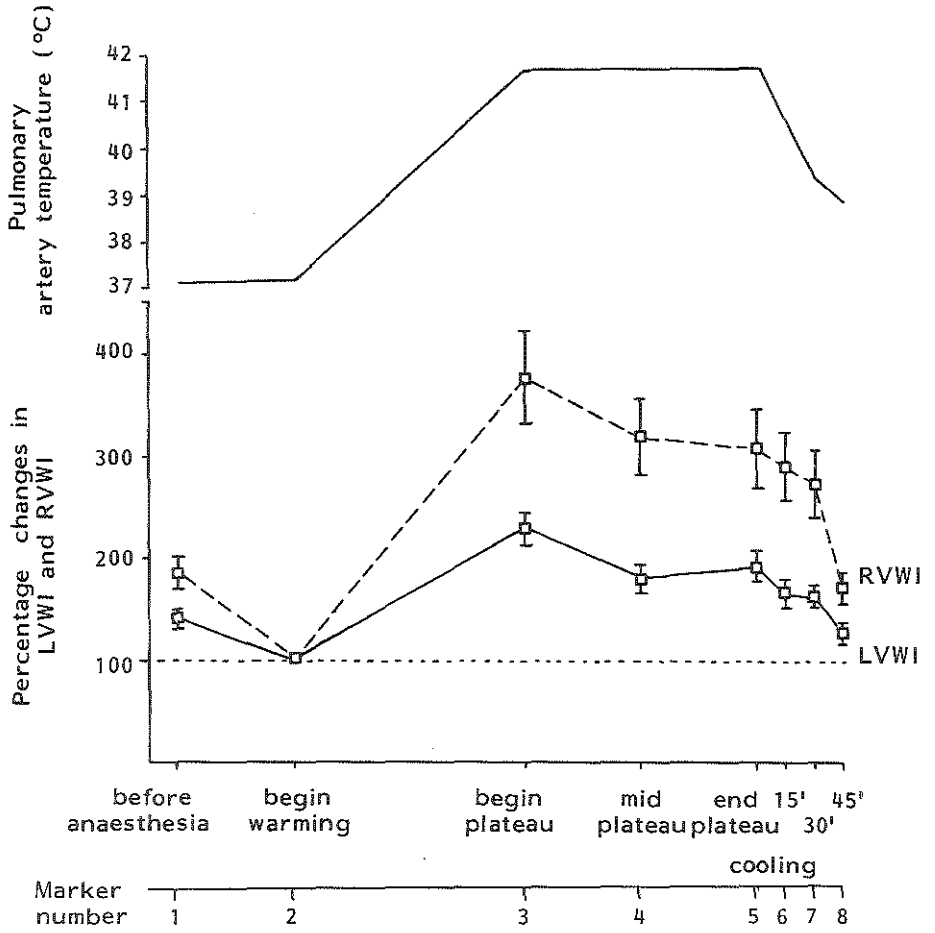

Fig. 4. The effects of Whole Body Hyperthermia (2 hrs at 41.8°C) on percentage changes in left ventricular work index (LVWI), and right ventricular work index (RVWI). Means ± SEM. The 100 percent value is taken as being the beginning of warming. The top graph shows the mean body temperature changes occurring.

considerable clinical relevance when assessing the fitness of patients to undergo whole body hyperthermia treatment.

The question naturally arises as to why the decrease in systemic vascular resistance should have been so much more than that of the pulmonary vascular resistance. The former decreased by mid plateau to 31.6% of its value at the beginning of warming whereas the latter decreased to 49.8% of its pre-warming value - These differences can account for the rise in mean pulmonary artery pressure and the proportionately greater increase of right ventricular work.

Resistance to flow through a vascular bed depends on two factors. Firstly, there is the resistance to steady flow, and secondly, the resistance to pulsatile flow (impedence). Pulmonary vascular resistance defined (as in the calculated results earlier presented) as the ratio of the average pressure decrease across the pulmonary vascular bed to the average flow is very limited in that it fails to take into account the pulsatile nature of the flow (Foëx 1980).

The concept of vascular impedance was introduced by Randal and Stacy (1956) and was applied by Caro and McDonald (1981) to the pulmonary circulation. Milnor et al. (1966) in a study in dogs have demonstrated the effect of heart rate and pulmonary blood flow on the oscillatory component of hydraulic input power. At a fixed pulse rate oscillatory power varies with the square of the flow. They demonstrated, however, that flow could be increased with less imput power increase if the pulse rate increased while the stroke volume remained constant. Hence, high pulse rates may prevent excessive rises of right ventricular work in the patients undergoing WBHT and one should be wary of trying to decrease heart rate by means of beta blockers (Euler-Rolle et al. 1977; Moricca et al. 1979). From the above brief discussion it is clear that the pulsatile nature of pulmonary vascular flow will tend to cause the increase in right ventricular work to be proportionately more than is the case in the systemic circulation. In the latter the pulsatile component of work is much less and hence steady work output predominates.

FIXED POINT MARKER ANALYSIS DURING RECOVERY FROM HYPERTHERMIA

The last marker in the fixed point analysis of the cardiovascular variables during treatment was marker 8. This was 45 minutes after the beginning of active cooling. The majority of measured variables were still at a level signifiantly different from those obtained before the induction of anaesthesia.

A further fixed point marker analysis was performed beginning in the Intensive Care Unit after the patient had been removed from the hyperthermia cabin and the non-depolarising muscle relaxants had been reversed by neostigmine. The first point for this new analysis was 2 hours after the beginning of cooling and at this time the following parameters were significantly higher than before induction of anaesthesia: body temperature, heart rate, cardiac index, mean pulmonary artery pressure and right ventricular work index. The variables that were still significantly reduced were: mean systemic arterial pressure, systemic vascular resistance index and pulmonary vascular resistance index. Pulmonary capillary wedge pressure and central venous pressure had raised values at the end of treatment (marker 8), but were not significantly different from the beginning of treatment at this first marker point in the Intensive Care Unit. Stroke index had increased from the end of treatment and was significantly increased once more, though the mean value was not as high as the maximum value obtained on attainment of the plateau temperature. Subsequently all the variables mentioned above were analysed every hour and compared with marker 1 (at the beginning of the treatment).

Results

Figure 5 shows the time at which the above mentioned parameters became not significantly different from the beginning of treatment. It should be noted

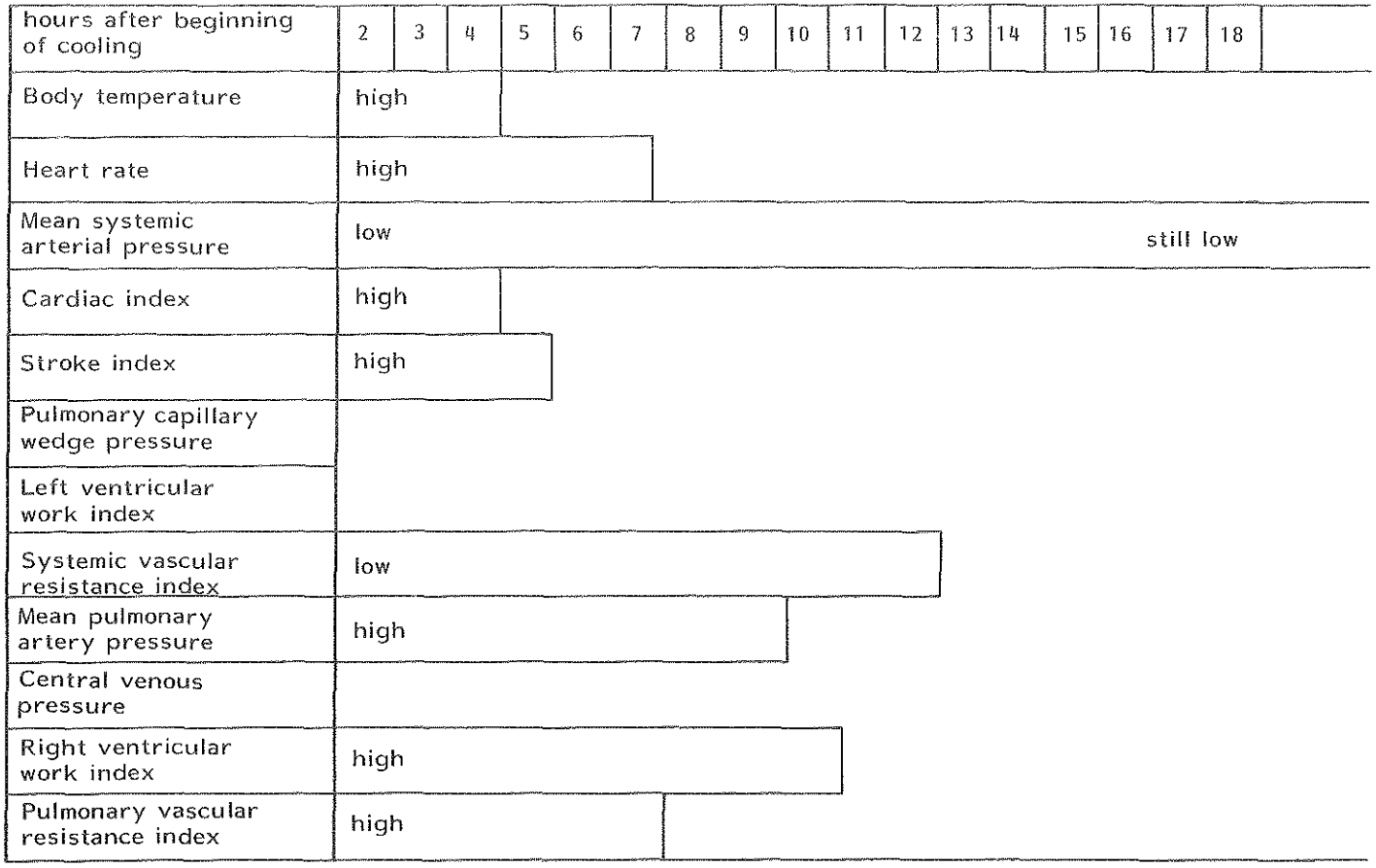

Fig. 5. Time course for the return of various cardiovascular parameters to normal following WBHT. Outside the histogram bars the values are no longer statistically different (on a paired Student's T-test) from the pre-treatment values. Indication is given at the beginning of the chart of the direction of abnormality.

that body temperature assessed as pulmonary artery temperature, measured by the thermistor tip of the Swan Ganz catheter, returned to normal 5 hours after opening of the hyperthermia cabin and hence 5 hours from the beginning of cooling. This point coincides with normalisation of the cardiac index and is followed by stroke index at 6 hours and pulmonary vascular resistance index at 7 hours.

Heart rate remained significantly elevated for 3 hours after body temperature was normal. Pulmonary artery pressure does not return to normal values until 10 hours after the beginning of cooling and though left ventricular work was not significantly raised at any stage during the patients stay in the intensive care unit, the right ventricular work did not return to normal until 11 hours of cooling had elapsed. Thirteen hours after the cabin was opened the systemic vascular resistance returned to normal values. The only remaining 'abnormal' parameter was then the mean systemic arterial pressure. This remained significantly lower than it was before treatment was commenced and even 18 hours after the end of plateau (the end of this analysis) it was still significantly reduced - that is not to say that the pressure at this stage was at a level that caused any anxiety. At 18 hours of cooling the mean pressure was 71.6 mm Hg as opposed to 87.7 mm Hg at the beginning of treatment.

It should be noted that the numbers of patients making up the statistical group were in general smaller the longer the observation time continued. This was due to a number of factors. As was mentioned in the section on anaesthesia a number of patients at the beginning of the study were in a very confused state and, in some cases, intravascular monitoring lines were pulled out or became otherwise inoperational. Without a fully patent and suitably functioning Swan Ganz catheter it is, of course, impossible to measure pulmonary artery pressure, pulmonary capillary wedge pressure and cardiac output. As these parameters are all required for several calculations, it was hence impossible

to calculate left and right ventricular work, and pulmonary and systemic peripheral resistance indices for all patients over a prolonged period of time.

It was, of course, quite possible to record those parameters that could be easily measured by non-invasive means such as heart rate and systemic blood pressure. Temperature was measured rectally if the Swan Ganz themistor was not available.

Discussion

Few authors have commented in much detail on the cardiovascular state of patients following WBHT. Hermann et al. (1982) reported 30 WBHT treatments - 16 using warming blankets and 14 using extracorporeal circulation. In 7 instances they noted mean blood pressure decreases to 60-70 mm Hg for up to 2.5 hours after treatment. They also noted that decreases of pressure occuring at elevated temperatures, were accentuated at the end of therapy when patients were actively cooled.

Pettigrew et al. (1974b) observed that a persistent tachycardia with low blood pressure could develop in patients with 'sensitive' tumors. Barlogie et al. (1979) commenting on the diastolic pressure (which had decreased during treatment), noted a rapid return to normal during the ensuing 12 hours, while Bull et al. (1979) noted that 5 out of 14 patients had systolic blood pressures between 70 and 90 mm Hg for 30 minutes to 3 hours post treatment. This 'postoperative hypotension' was also noted by Smith et al. (1980).

It would seem, from the above, that hypotension is a fairly frequently occurring phenomenon in the early recovery phase following WBHT. Apart from the above mentioned comment by Pettigrew et al. (1974b) there have been no comments on pulse rate changes nor have there been reports of changes we noted in other cardiovascualar parameters. The most likely explanation is that these determinations were never performed.

CHAPTER VII

OXYGEN TRANSPORT AND OXYGEN CONSUMPTION

The cardiovascular and respiratory systems together provide the function of transporting oxygen from the lungs to the cell. This may be regarded as their most important function (Nunn 1977b).

The amount of oxygen transferred from the lung to the cells of the body per minute has been termed the oxygen 'flux' by Nunn and Freeman (1964). This is determined by the cardiac output per minute and the oxygen content of the arterial blood. The latter is in its turn dependent on the haemoglobin content of the blood and the arterial oxygen saturation of the blood.

In theory, one gram of haemoglobin can combine with 1.39 ml of oxygen. Previously the figure was taken as 1.34 ml, but the former figure was adopted following the determination of the molecular weight of haemoglobin (Braunitzer 1963). Some studies (Prys-Roberts et al. 1971) have indicated that the factor 1.34 may give good results at normal acid base values and when calculating arterial/mixed venous oxygen content differences over a wide range of differences. On the other hand, Gregory (1974) studied oxygen capacities of foetal and adult blood by determination of their oxygen and carbon monoxide combining power. In the adult he came to a mean value for oxygen combining power of 1.306 ($\pm$ 0.006 S.E.M.) ml of oxygen combining with 1 gram of haemoglobin. This mean value was used in all calculations used in this thesis. It is perhaps important to realise that it is largely unimportant, in the present study, which factor is used. Content differences are being estimated and hence any inaccuracy in the absolute values obtained will automatically cancel each other out.

In addition to the oxygen that is carried, reversably bound to haemoglobin

a very small amount of gas is carried in physical solution in the blood. At normal arterial pO2 of the blood approximately 0.25 ml of oxygen is carried in this way in 100 ml of blood. This solubility is dependent on the oxygen partial pressure (PaO2) of the arterial blood and when breathing 100% oxygen the level of dissolved oxygen rises to 1,5 ml per 100 ml of blood. The amount of dissolved oxygen also rises with decreasing temperatures (Nunn 1977c) and conversely increasing the temperature will decrease the amount of dissolved oxygen. As mentioned in the section on anaesthesia, the patients were maintained during treatment on 33% oxygen. The mean PaO2 at raised body temperatures at markers 3 (beginning of plateau), 4 (mid plateau) and 7 (after 30 minutes of cooling) were 117.8($\pm$ 5.8 S.E.M.), 110.7 ($\pm$ 3.05 S.E.M.), and 110.0 ($\pm$ 5.44 S.E.M.)respectively.

In view of the small amounts of oxygen carried in solution at these partial pressures at the raised body temperatures obtaining, these have been ignored in the calculations presented below. The oxygen saturation ofthe blood was corrected for pH, temperature, and pCO2 using the computer routine described by Kelman (1966) and this corrected value was used in all ensuing calculations.

Oxygen flux was calculated as:

$$\frac{CI \times SaO2 \times Hb \times 1.306}{100}$$

where CI = Cardiac Index in ml per minute

SaO2 = Percentage arterial oxygen saturation

Hb = Haemoglobin concentration in grams per 100 ml blood

This calculation results in the oxygen flux in ml per minute per m2 of body surface area.

Oxygen consumption was calculated as:

$$\frac{CI \times (CaO2 - CvO2)}{100}$$

where CI = Cardiac Index in ml per minute

CaO2 = Arterial Oxygen Content per 100 ml of blood

CvO2 = Mixed venous oxygen content per 100 ml of blood

CaO2 is derived from the equation:

$$CaO_2 = Hb \times SaO_2 \times 1.306$$

and CvO2 is derived from the equation

$$CvO_2 = Hb \times SvO_2 \times 1.306.$$

when SVO2 = Percentage mixed venous oxygen saturation

Results

Figure 6 is a graph of the calculated oxygen consumptions of patients during whole body hyperthermia treatment at the points at which it was assessed. These points were: before induction of anaesthesia (marker 1), after induction of anaesthesia and at the beginning of warming (marker 2), at attainment of plateau temperature of 41.8°C (marker 3), after one hour at plateau (marker 4), and half an hour after commencement of cooling (marker 7).

Figure 7 is a graph of the calculated oxygen flux during this treatment period. As stated in the figure legends, significance indicated is in comparison with the previous data point. There were no significant differences in oxygen consumption between marker 1 and marker 7. In other words, after 30 minutes of cooling there was no longer any significant increase in oxygen consumption compared with the consumption when the patients were awake (and normothermic). At 30 minutes of cooling the mean pulmonary artery temperature was 39.4°C ($\pm$ 0.31 S.E.M.). The oxygen flux, on the other hand, was still significantly raised at this point above the awake values ($p < .01$).

It is of great importance to know, not so much the absolute value for oxygen flux or oxygen consumption, but the ratio between the two. Figure 8 is a graphical representation of this ratio obtained by dividing the oxygen flux by the oxygen consumption. It should be noted that, during warming, there is a highly significant rise in this value and that, after 30 minutes

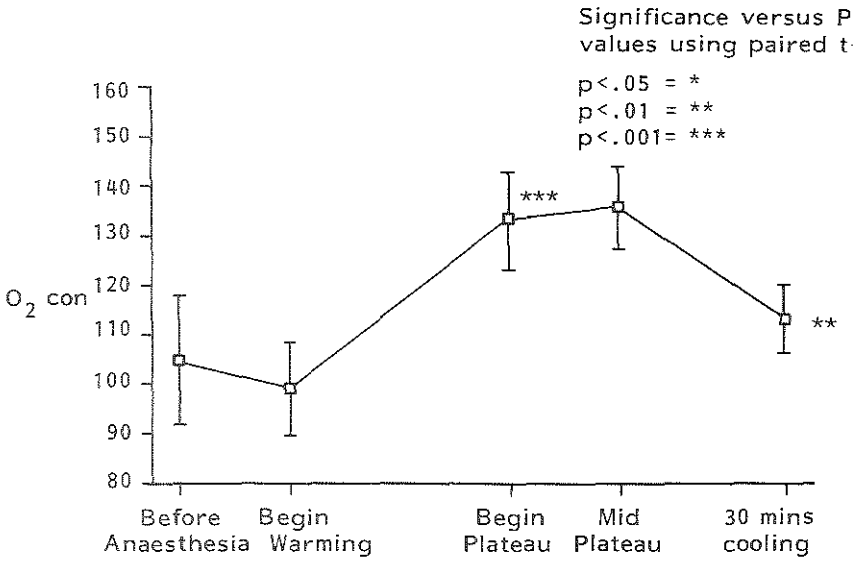

Fig. 6. The effect of whole body hyperthermia (2 hrs at 41.8°C) on oxygen consumption per m^2 body surface area (O_2 con). Means ± SEM.

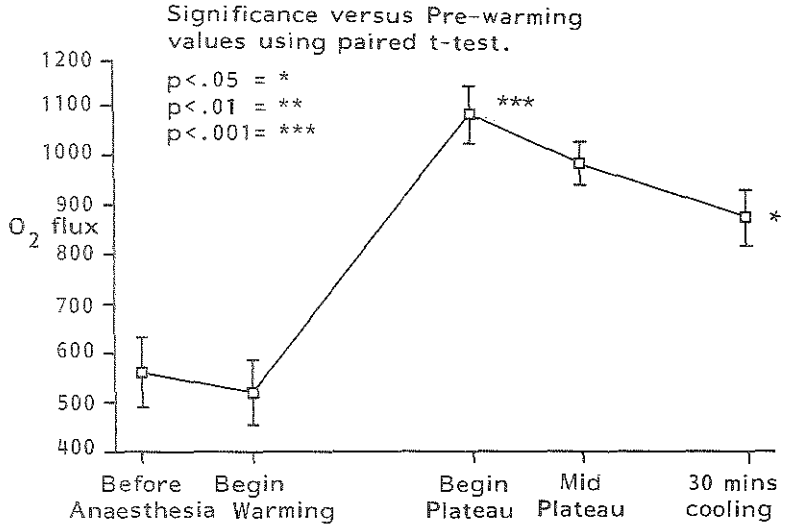

Fig. 7. The effect of whole body hyperthermia (2 hrs at 41.8°C) on oxygen flux in ml per m^2 body surface area (O_2 flux) Means ± SEM.

Significance versus Pre-warming values using paired t-test.

$p<.05$ = *
$p<.01$ = **
$p<.001$ = ***

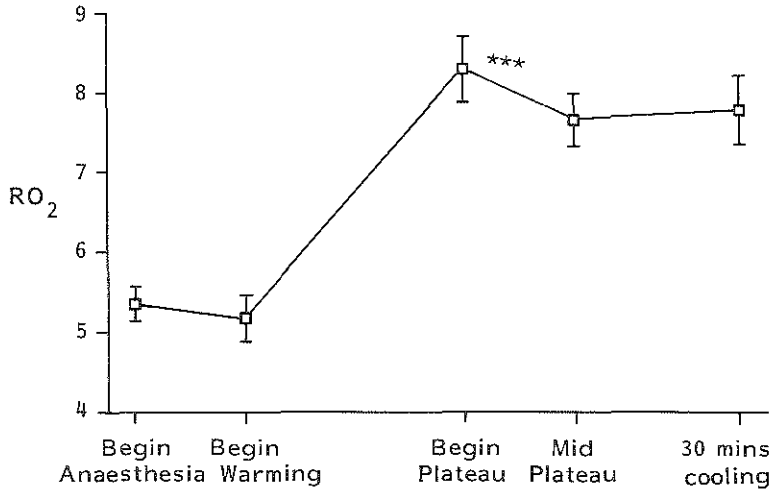

Fig.8. The effect of whole body hyperthermia (2 hrs at 41.8°C) on $\frac{\text{Oxygen Flux}}{\text{Oxygen Consumption}}$ (RO_2) Means ± SEM.

of cooling, it is still significantly higher than when the patients were awake ($p < 0.05$). The figures from which the calculations for the above graphs were obtained together with figures used in the graphs are presented in Table III.

Discussion

A number of studies of basal oxygen consumption have attempted to link the figures obtained with various physical characteristics of the subjects such as age, sex, height, weight and body surface area. Most studies find a relationship between the surface area and the oxygen consumption. Altman and Dittmer (1971) quote figures from Baldwin (1948) for oxygen consumption. These mean figures are: 131 x surface area for males between 35 and 49, 132 x surface area for males between 50 and 69, and 129 x surface area for females between 50 and 79. The above figures have been quoted as all the patients, in whom oxygen consumption was estimated before induction of anaesthesia, fall into these groups.

Using the above figures and surface area values for our patients, we arrive at a mean figure for oxygen consumption of 130.8 ml min-1 m-2. The figures presented in this study give a mean of 104.8 ml min-1 m-2, which is a reduction of 19.9% on the estimated figures. Nunn (1977a) has produced figures for predicted values for oxygen consumption during uncomplicated anaesthesia based of figures of basal data from Aub and Dubois (1917) and Boothby and Sandiford (1924). Using these figures it was calculated that the patients, in whom oxygen consumption was measured under anaesthesia and before warming, should have had a mean oxygen consumption of 105.1 ml min-1 m-2 instead of the calculated mean of 99.1 ml min-1 m-2 - a decrease of 5.7% on the calculated figures. The fact that the percentage mean decreases at the two marker points vary, may be accounted for by the fact that the mean body temperature of the patients had risen slightly by the time marker two had been reached - in some

Table III: Oxygen Consumption and Flux under Hyperthermia

	Marker 1 Before induction of anaesthesia	Marker 2 Beginning of Warming	Marker 3 on reaching Plateau of 41.8°C	Marker 4 After 1 hour at 41.8°C	Marker 7 After 30 mins of cooling (Tp° = 39.45°C ± 0,12)
Cardiac Index (L min-1 m-2)	4,19 ± 0.37 (20)	3,33 ± 0,33** (22)	7,50 ± 0,41*** (22)	7,02 ± 0,36 (22)	6,62 ± 0,30 (22)
Arterial Oxygen Saturation (%)	95,43 ± 0,65 (8)	98,29 ± 0,36*** (15)	94,10 ± 1,58* (16)	93,03 ± 1,02 (25)	94,50 ± 0,89 (18)
Haemoglobin Concentration (gm/100 ml)	9,17 ± 0,73 (9)	9,83 ± 0,57 (14)	9,44 ± 0,52*** (16)	9,70 ± 0,36 (25)	9,22 ± 0,33** (29)
(CaO2-CvO2) ml per 100 ml blood	2,77 ± 0,15 (8)	3,04 ± 0,21 (12)	1,86 ± 0,09*** (14)	2.01 ± 0,12 (21)	1,81 ± 0,08 (20)
Oxygen Flux (ml min-1 m-2)	561,1 ± 72,0 (8)	520,8 ± 65,5 (12)	1078,9 ± 58,8*** (13)	980,7 ± 44,7 (21)	870,17 ± 56,7* (18)
Oxygen Consumption (ml min-1 m-2)	104,8 ± 13,2 (8)	99,1 ± 9,5 (12)	133,4 ± 10,0*** (13)	136,0 ± 8,3 (19)	113,2 ± 7,1** (17)
Oxygen Flux / Oxygen Consumption	5,37 ± 0,22 (8)	5,18 ± 0,30 (12)	8,32 ± 0,42*** (13)	7,67 ± 0,34 (19)	7,81 ± 0,44 (16)

The effects of Whole Body Hyperthermia (41.8°C for 2 hours) on a number of oxygen flux and consumption parameters. Means ± 1 S.E.M. The sample markers are given in brackets. Significance on paired t-test in comparison with previous marker point. * = $p < .05$, ** = $p < 0.1$, *** = $p < .001$.

cases warm water was being circulated through the heating blanket and the cabin heaters were already switched on. It would therefore appear from the above discussion that, either the patients had lower than normal oxygen consumption values or that there was a slight systematic error in the method of measurement.

Prys-Roberts (1974) has produced, based on various data (Brandfonbrener et al. 1955; Schröder et al. 1966; Frohlich et al. 1969; Cournand et al. 1944; Stead et al. 1944; and Emirgil et al. 1967) figures to show that there is a good correlation between cardiac index and age and has presented the formula: Cardiac Index = 4.16 - 0.02 (age in years). Using this formula we arrive at a theoretical mean figure of 3.04 litres per minute in the patients in whom oxygen consumption was measured before anaesthesia was induced. This compares with the actual measured mean value of 3.85 litres per minute. This means that the patients, as a group, were somewhat hyperdynamic.

As a group in order to achieve an oxygen consumption of 130.8 ml min-1 m-2 (which was calculated above to be their oxygen consumption according to the figures of Baldwin (1948)) the patients should have had a mean arterial-mixed venous oxygen content difference of:

$$\frac{130.8 \times 100}{3.85} = 3.40 \text{ ml per 100 ml of blood.}$$

The mean measured difference was 2.77 ml per 100 ml of blood.

It is unlikely that constant errors of measurement would occur in either cardiac index measurement or arterial and mixed venous oxygen saturation estimations. It must therefore be concluded that the error was in sampling of the mixed venous blood from the Swan Ganz catheter, with consequent withdrawal of a mixture of pulmonary artery blood and blood that was being withdrawn from pulmonary capillaries and veins, and was hence partially or wholly reoxygenated. This would result in decreased arterial mixed venous oxygen content difference and hence lower than mean oxygen consumption. As the position of the Swan Ganz catheter was not moved after placement, it is likely that similar

errors of a similar magnitude would have occurred each time a sample was taken and hence the resultant values obtained would be low by the same proportion. It may thus be concluded that though the oxygen consumption figures that are presented in this thesis are probably too low, it may still be possible to draw valid conclusions from them.

The oxygen flux, as can be seen by reference to Table III and figure 7, is greatly increased under conditions of hyperthermia. The oxygen carried by the blood, (as previously stated, the minimal amounts of oxygen carried in solution have been disregarded), increased from a mean value of 561.1 ml min-1 m-2 in the sedated awake state to a mean value of 1078.9 ml min-1 m-2 on reaching plateau. This was due to the large increase of cardiac index under conditions of hyperthermia. The flux decreased slowly but was still 980.7 ml min-1 m-2 at mid plateau. These rises in oxygen flux of 92.1% and 74.6% respectively, are very much greater than the corresponding mean increases of oxygen consumption, which amount to 27.3% and 29.8% respectively.

As Nunn and Freeman (1964) have pointed out, the oxygen flux is usually considerably in excess of oxygen consumption and the normal ratio is about 4. They have suggested that a flux of 400 ml min-1 would be the lowest tolerable for a short period under anaesthesia. This would correspond to an oxygen ratio of about 1.6. It can thus be seen that the oxygen ratio can be regarded as a sort of safety factor - the higher the ratio the larger the reserve of oxygen that can be used at any one moment, the lower the percentage extraction of oxygen from the blood, and hence the lower the arterial/mixed venous oxygen content difference. This we saw in the patients that we studied, who had reduced arterial mixed venous oxygen content differences at hyperthermic temperatures.

The increases in oxygen consumption during whole body hyperthermia treatment, as presented above, were relatively modest. Lees et al (1979) have presented figures from patients undergoing WBHT at 41.8°C under

thiopentone/fentanyl anaesthesia. Their oxygen consumption figures showed a mean rise of 58.5% over normorthermic control values. Our own figures indicate an increase of 34.6% over the normothermic anaesthetised controls. Our patients were paralysed and ventilated while those of Lees et al. (1979) were breathing spontaneously - indeed they were often grossly hyperventilating with average increases of nearly 200% in minute volume (Lees et al. 1980). The muscular work of breathing, and hence the oxygen consumption of the respiratory muscles, was thus increased.

Myocardial Oxygen Consumption

Blain et al. (1956) give a mean figure of oxygen consumption of the myocardium of 9.1 ml of oxygen consumed per 100 grams of myocardium as the normal consumption in resting subjects. The normal weight of the heart of man varies with body length and sex varying from 219 (± 30 SD) grams for females of 135 cm of length to 378 (± 40 SD) grams for males of 200 cm length (Ziek, 1942).

Using the normogram for heart size presented by Bove and Scott (1971), the expected cardiac oxygen consumption for our group of patients was calculated to be 27.4 ml per minute or 16.2 ml per minute per m2 of body surface area. The mean value for those patients in whom oxygen consumption was calculated at marker 1 (before induction of anaesthesia) was 18.0 ml per minute per m2 of body surface area.

Theye (1972) in a study in dogs under halothane anaesthesia has presented the formula:

$$vO2 = work \times 2.1 + 1.4$$

where:

vO2 = left ventricular oxygen consumption in ml per minute per 100 gm of heart tissue

and work = left ventricular work in kilogram metres per minute.

TABLE IV Myocardial oxygen consumption under Whole Body Hyperthermia

Time	Mean calculated Right Ventricular oxygen consumption	Mean calculated Left Ventricular oxygen consumption	Mean total cardiac oxygen consumption	Mean whole body oxygen consumption	% change in total body oxygen consumption from Marker 1 accounted for by myocardial oxygen consumption
Marker 1	3,4	20,7	24,1	104,8	
Marker 2	3,1	16,9	20,0	99,1	71,9%
Marker 3	4,3	28,5	32,8	133,4	37,3%
Marker 4	4,0	24,0	28,0	136,0	21,7%
Marker 7	3,9	20,9	24,8	113,2	34,0%

Calculated mean right and left ventricular oxygen consumption in ml per m2 body surface area in comparison with total body oxygen consumption.

Applying this formula to the patients in this study, a mean oxygen consumption of the left ventricle at marker 1 of 20.4 ml per minute per m2 of body surface area was obtained.

Vinten-Johansen et al. (1982) studied left ventricular oxygen requirements in dogs under pressure loading or volume loading conditions. They determined that volume loading conditions had greater oxygen requirements than was previously appreciated; being consistently greater than that obtained for pressure loading conditions. There was, however, no statistical significance between the two regression lines. Using the results presented in this study, (assuming equal distribution of volume and pressure loadings), left ventricular minute oxygen consumption at marker 1 was calculated to amount to 21.0 ml per m2 of body surface area. As can be seen the values for left ventricular work calculated by the two different methods correspond quite well with each other. The first calculation gave an oxygen consumption figure of 20.4 ml per m2 body surface area and the second a value of 21.0 ml per m2 body surface area - the mean value was 20.7 ml per m2 of body surface area.

Using the data presented in the two publications (Theye, 1972 and Vinten-Johansen et al. 1982) of right ventricular work, one can calculate mean oxygen consumptions for both right and left ventricles for each marker point at which total body oxygen consumption under WBHT was estimated. These are presented in the accompanying table IV. It can be seen that the estimated total myocardium oxygen consumption increased by about 14% for every 1°C rise of body temperature. The rest of the whole body increased its oxygen consumption by about 6% per 1°C rise in temperature. As can be seen approximately 37% of the increased oxygen consumption occurring at marker 3 can be accounted for by increase in myocardial oxygen demand.

In the case of the cardiovascular system we have a very good indicator of organ oxygen consumption which has been shown to have good correlation with oxygen uptake - this is the work that is performed by the heart. It is there

fore reasonable extrapolate cardiac work in terms of oxygen consumption under hyperthermic conditions. Without direct measurements of hyperthermic oxygen consumption of other organs it may be unwise to further discuss percentage contributions of other organ systems to whole body oxygen consumption. As Hales et al. (1979) have pointed out there exist basic differences in the circulatory responses to different species to heat stress would it would therefore be unwise to draw too many conclusions about oxygen consumption in man under hyperthermia based on extrapolation from animal experimentation.

Factors affecting oxygen transport to the tissues

The oxygen saturation of the haemoglobin in the blood is affected principally by the partial pressure of oxygen. A graph of the relationship between the two parameters forms the foundation of the S-shaped oxyhaemoglobin dissociation curve.

Various factors will cause a shift of the curve to the right or the left, thus raising or lowering the p50 value (the value at which the haemoglobin is 50% saturated with oxygen). A shift to the right implies a decreased oxygen saturation for the same partial pressure. Hence arterial desaturation may occur; but at the same time oxygen will be released from haemoglobin more easily at the pO2 levels existing in the tissues. The curve is shifted to the right and the p50 is raised by increasing hydrogen ion concentration (lowered pH), pCO2, temperature, ionic strength or haemoglobin concentration. The effect of a rise of temperature is to be seen from the figures in table III for arterial oxygen saturation under WBHT, where it can be seen that the maximal decreases of SaO2 were observed at midplateau. At this point there had been a mean statistically significant decrease in SaO2 of 5.6% from the post anaesthetic induction value. We must of course assume that, at the same time, that no changes had taken place in intrapulmonary shunt values. This assumption is

a reasonable one to make in view of the fact that the alvelor arterial pO2 differences did not increase, as mentioned above.

In view of the easier release of oxygen to the tissues afforded by the shift to the right of the dissociation curve it is unlikely that tissue hypoxia would be occurring, especially in view of the increased oxygen flux that was present. This view is supported by the fact that no significant blood arterial or mixed venous pH changes occurred during treatment.

A further factor causing changes in the position of the dissociation curve is the level of 2,3-diphosphoglycerate (2,3-DPG) in the erythrocytes. A decease of 2,3-DPG will shift the curve to the left and this may hinder tissue oxygenation. Kim (1979) has measured 2,3-DPG during hyperthermia and has found no changes.

The final pathway of oxygen into the cell is by diffusion. Diffusion coefficients for oxygen have been measured under normothermic conditions (Erdmann and Krell 1976), but work in this field still needs to be done in the case of hyperthermia.

In conclusion, it may be stated that no excessive rises in oxygen consumption occurred in our patients and that the rise in oxygen flux that took place was more than adequate to supply the needs of the body. There was no reason to suspect the occurrence of generalised tissue hypoxia.

CHAPTER VIII

SIDE EFFECTS AND COMPLICATIONS FOLLOWING WHOLE BODY HYPERTHERMIA

A number of complications and side effects of whole body hyperthermia have been reported in the literature. Both major and minor complications will be briefly discussed. Minor complications include the very commonly occurring generalised lethargy, which may last for several days after treatment. Minor burns at pressure points have been described by Pettigrew et al. 1974; Mackenzie et al. (1975); Barlogie et al. (1979); Lees et al. (1980); Hermann et al (1982) and many others.

Gastrointestinal disturbances have been seen by many authors including Larkin et al. (1977); Moricca et al. (1979); Smith et al. (1980) and Ostrow et al. (1981). These are presumably due to fluid accumulation as a result of the capillary leak syndrome described by Herman et al. (1982) and previously mentioned in this thesis. Our patients often suffered from the resulting diahroea and vomiting.

Herpes simplex may occur following treatment and has been seen by Pettigrew et al. (1974a); Mackenzie et al. (1975); Barlogie et al. (1979) and Smith et al. (1980). In the present study this complication was also seen and is probably due to activation of the latent herpes virus. Post hyperthermia fevers have been commented on by a number of authors including Bull et al. (1979) and Barlogie et al. (1979). These did not form a feature of the present series of patients, but further details of temperature patterns in the recovery phase will be given and discussed later.

Haematological Changes

A number of authors have commented on haematological changes following WBHT. Pettigrew et al. (1974b) have noted haemoglogbin decreases averaging 0.8 gms per 100 ml within 30 hours of each treatment. As can be seen from table III, Chapter VII, there were highly significant decreases in haemoglobin during warming in our patients (between markers 2 and 3) and also at the onset of cooling (between markers 4 and 7). These decreases were almost certainly due to haemodilution consequent upon vasodilatation and net movement of fluid into the vascular space aided, during warming, by the osmotic effect of the administration of Dextran 40 and, during cooling, by the manitol that was given. This view is also held by Larkin et al. (1977), who commented that the only significant changes in haemoglobin and haematocrit were explainable by volume shifts. The haemoglobin values in our patients remained significantly lowered, (in comparison with those obtaining at the beginning of treatment), until 8 hours after the commencement of cooling. At 24 hours after cooling there were no longer any significant differences to be seen.

Many authors report a leucocytosis following WBHT. Pettigrew et al. (1974b) reported a mean rise of 67% in white cell count to a mean of 13,700 per mm3 after 4 hours treatment, returning to normal within 24 hours. Both Larkin et al. (1977) and Bull et al. (1979) comment that the leucocytosis was caused by a rise in the polymorphonuclear cells accompanied by absolute mean falls of lymphocyte counts. This has also been our own experience. Barlogie et al. (1979) reported marked neutrophilia in some patients but with no overall significant trends in the group as a whole. Herman et al. (1982) found neutrophilic leucocytosis of more than 12,000 per mm3 in 7 of 16 treatments using blanket technique but saw neutropenia (less than 1,000 per mm3) in 3 out of 14 treatments in which extracorporeal warming techniques were employed. This might indicate differences caused by heating technique per se and Parks et al.

(1979), who also used extracorporeal heating, noted, in contrast to most authors, that the leucocyte counts were little affected.

In the patients reported upon in this thesis the total white cell count was significantly raised (in comparison with the day before treatment) for up to 4 hours after cooling commenced but this significance had disappeared by the following morning (± 18 hours after opening of the hyperthemia cabin). It is probable that the total body white cell count was still significantly raised at 6 hours of cooling, but the probable haemodilution that was present prevented the leucocyte concentration from being significantly raised. The lymphocyte count remained significantly depressed for 3 days following WBHT. This may possibly be caused by destruction in the reticulo-endothelial system as seen by Fletcher et al. (1980) following whole body hyperthermia in rats.

Leucocytosis induced by WBHT may have considerable significance as Grogan et al. (1980) have demonstrated that polymorphonuclear cells, from patients that have undergone WBHT, showed an increase in bacteriocidal activity.

Coagulation Changes

Reduction in the number of circulating platetlets would appear to be a frequent occurance during and following hyperthermia, though some authors (Bull et al. 1977), have not reported this effect. Pettigrew et al. (1974a) reported four deaths associated with evidence of disseminated intravascular coagulation in a series of 51 patients. In three cases recent tumor necrosis was evident.

Barlogie et al. (1979) noted a decrease in the mean platelet count from 243,000 to 147,000 per mm3, 24 hours after treatment. The count returned to normal after about one week. They also observed prolongation of the prothrombin and partial thromboplastin times by an average and 4 and 6 seconds at 24 hours post treatment. These were accompanied by mild to moderate decrease of fibrinogen levels (minimum 100 milligrams per 100 ml during treatment) and increases

in fibrin split products in 4 out of 13 patients. These changes they attributed to a low grade disseminated intravascular coagulation syndrome.

Larkin et al. (1977) noted no appreciable changes in prothrombin or partial thromboplastin values but commented on the fact that platelet counts were occasionally noted to drop dramatically. This they attributed to prior chemotherapy. Parks et al. (1979) noted that platelet counts, which reduced from mean 235,000 to 160,000 per mm3, might remain reduced for several months (even in the presence of normal megakaryocyte activity). Fibrin split products remained within normal limits and they noted that there was a correlation between the magnitude of the patient's tumor mass and the level of thrombocytopenia that was observed.

Herman et al. (1982) regularly saw platelet counts post treatment depressed below the 70,000 per mm3 level with decreases in fibrinogen accompanied by prolongation of prothrombin and partial thromboplastin times. The platelet reductions were more common after heating using extracorporeal circulation than after using water circulating blankets. One patient out of 11 died from disseminated intravascular coagulation.

In the present series of patients there were no significant changes in thrombocytes to be observed at mid plateau (after one hour at the treatment temperature of 41.8°C), but 1½ hours later, and ½ an hour after cooling had commenced, there had been a highly significant fall ($p<.001$). The mean value before treatment of 262,000 per mm3 had fallen to a mean value of 127,000 at this point. The mean value at mid plateau was 222,000. The thrombocyte count remained low and was still highly significantly lowered (mean of 88,000 per mm3) three days post treatment. Fibrinogen levels showed highly significant falls at marker 7 (½ hour after commencement of cooling) but had recovered by 2 days post treatment.

Electrolyte Changes

Many authors have reported changes in blood electrolyte concentrations during and following WBHT. The three electrolytes most commonly changed are magnesium, phosphate and calcium. These all tend to fall during treatment as reported by Larkin et al. (1977), Barlogie et al. (1979) and Herman et al. (1982), though the falls in calcium concentration seen by the latter authors were not remarkable. Phosphate levels tended to return to normal levels at 24 hours and magnesium at 48 hours. Bull et al. (1979) observed similar patterns. Ostrow et al. (1981) reported lowered calcium levels for more than 24 hours.

Potassium levels were seen by Pettigrew et al. (1974b) to rise during warming and treatment and to fall in the post-treatment phase. Low levels of potassium have been seen following treatment by Larkin et al. (1977), Barlogie et al. (1979), Ostrow et al. (1981) and Versteegh et al. (1981).

In the present series of patients similar results to the above were seen, and highly significant decreases in both phosphate and magnesium were observed, but whereas phosphate levels had returned to normal at 48 hours the magnesium levels had not returned to normal until 72 hours after treatment. Calcium concentrations remained low after treatment and there were still highly significant decreases at 72 hours after the beginning of WBHT. Potassium levels were also still significantly lowered for up to 72 hours.

In general no great problems have been reported in maintaining sodium and chloride levels during and following treatment. This accords with the present findings that though there were significant falls in these electrolytes, the changes were of minimal clinical significance.

Miscellaneous Changes in Blood Chemistry

Some authors have commented on changes in glucose levels taking place as a

result of WBHT and both Larkin et al. (1977) and Ostrow et al. (1981) have shown that there may be considerable rises during treatment. Barlogie et al. (1979) have shown that mean increases may still be considerable (50% rise) 24 hours after treatment.

In the present series mean rises of 100% in glucose levels were seen during the cooling phase of treatment and in the intensive care unit. These returned to normal somewhere between 6 and 18 hours after cooling was commenced. These rises in glucose levels are almost certainly sympathetically mediated and Kim et al. (1979) demonstrated significant rises in both adrenaline and noradrenaline levels in the plasma of patients undergoing WBHT.

In the present series of patients urea and creatinine levels in the blood were monitored - once during cooling (marker 7), and then at 24, 48, 72 hours post-treatment. Highly significant rises were seen in both parameters (in comparison with the pretreatment values) at marker 7, and whereas the creatinine levels had returned to normal by 24 hours the increased urea levels did not return to normal for 72 hours. Increases in blood urea and creatinine levels have also been observed by Versteegh et al. (1981) returning to normal within 5 days. Other investigators (Bull et al. 1979), on the other hand, have found no changes in creatinine or creatinine clearance levels. Impairment of renal fuction under hyperthermia is probably quite common and Logawney-Malik et al. (1979), in a study in dogs, have observed reversable decreases of renal plasma flow and glomerular filtration rates under 42°C hyperthermia.

From the above discussion it may be concluded that, in the present series, none of the complications or side effects mentioned caused an insuperable problem in the safe clinical treatment of patients under WBHT. The sometimes severe hepatic problems that were encountered will now be discussed in the next chapter.

CHAPTER IX

THE LIVER AND HYPERTHERMIA

Reports of raised serum enzyme levels suggesting the presence of impairment of hepatic function are very widespread in literature concerning whole body hyperthermia treatment.

It would appear that 41.8°C should, at the moment, be taken as the upper safe limit for whole body hyperthermia treatment. Many authors have commented on the correlation between the temperature at which the patients were treated and the incidence and severity of raised serum enzyme levels. Pettigrew et al. (1974b) reported that there were no significant changes in SGOT, SGPT and LDH, following 47 treatments at or below 41.8°C whereas after 17 treatments at temperatures between 41.8° and 42°C, even though the temperatures were only held for between 10-40 minutes, dramatic rises in SGOT (25 fold), and SGPT (8 fold) occurred. The bilirubin levels increased following the higher treatment temperatures to levels of 1.56 mg per 100 ml of blood and were almost 3 times as high as those following treatment at 41.8°C or below. Similar findings have been reported by Blair and Levin (1977) and by Pettigrew and Ludgate (1977). Morrica et al. (1979) commented that signficant increases in SGOT, SGPT, LDH and gamma-GT only occurred when the treatment temperature was held between 42° and 42.3°C - though again the high temperture was only held for short periods (15-20 minutes).

Larkin et al. (1977) held patients' temperatures at 42°C for 2 hours and observed an eight-fold increase in mean SGOT levels at 24 hours after treatment together with twelve-fold increases in CPK and almost three-fold increases in LDH. These values had returned to normal 96 hours post-treatment. The alkaline phosphatase values were not affected.

Barolgie et al. (1979), treated patients at 42°C and noted that all patients had some elevation of CPK levels peaking at 24 hours after thermotherapy. They showed figures to indicate mean eight-fold increases at 24 hours. They made the observation that greatest rises were seen following the first treatment and patients did not necessarily have increased CPK levels following subsequent treatments, even if the first treatment had resulted in substantial rises of the enzyme. One patient developed myoglobinuria. Myocardial CPK fraction was normal. An LDH rise, not associated with haemolysis, was attibuted to change in liver function, though changes in SGOT and alkaline phosphatase were not consistently noted in the patients with marked LDH elevations.

Lees et al. (1980), while agreeing that the level of the treatment temperature is important, have suggested that mechanical ventilation of the lungs might also play a role in the occurrence of hepatic damage by reducing hepatic blood flow. However, Libonati et al. (1973) have demonstrated that there were no changes in total splancnic blood flow or oxygen consumption as between patients spontaneously breathing or those whose ventilation was controlled. To the best of our knowledge no comparative studies have been performed under hyperthermic conditions.

Rises in the levels of serum enzymes follwing WBHT are not confined to those patients in whom the temperature was raised above 41.8°C, though the severity of the changes may be very much reduced by not exceeding this level. Blair and Levin (1977) found a mean ten-fold increase in SGOT following 10 WBH treatments where the temperature was held above 41.8°C but also a 2.6-fold increase in the 25 treatments in which the temperature was maintained at 41.8°C or below. The corresponding mean values in bilirubin were seven-fold and 1.5-fold respectively. One of the patients from the first group, who was on phenobarbitone treatment for epilepsy, died on the fourth postoperative treatment day in liver failure. A relationship was found between the incidence of liver failure and the ingestion of alcohol or enzyme inducing drugs.

TABLE V Serum Enzyme changes following Hyperthermia

	1 Day before Treatment	During Cooling (Marker 7)	1 day after Treatment	2 days after Treatment	3 days after Treatment
SGOT u/l	12,9 ± 2,4	15,1 ± 3,8	48,4 ± 7,8***	210,1 ± 68,4**	235,7 ± 125,7
(up to 19)	(28)	(24)	(27)	(30)	(28)
SGPT u/l	13,6 ± 2,7	9,8 ± 2,1	27,9 ± 4,6*	141,1 ± 39,5**	322,3 ± 172,5
(up to 10)	(28)	(24)	(27)	(28)	(15)
CPK u/l	21,7 ± 3,3	20,0 ± 6,2	156,1 ± 41,1**	99,1 ± 20,8***	53,5 ± 10,5**
	(25)	(27)	(23)	(22)	(12)
LDH (total) u/l	157,2 ± 11,8	151,3 ± 14,8	242,9 ± 22,9**	437,1 ± 166,8	632,6 ± 277,2
(up to 240)	(27)	(27)	(25)	(21)	(7)
LDH1 Percent	24,3 ± 1,4	25,3 ± 1,4	25,6 ± 1,6	24,2 ± 1,8	15,5 ± 5,7
(18-38)	(26)	(26)	(24)	(18)	(4)
LDH2 Percent	43,4 ± 1,4	42,7 ± 1,5	40,8 ± 1,4	44,1 ± 2,9	36,0 ± 8,6
(28-48)	(26)	(26)	(24)	(18)	(4)

LDH3 Percent (12-32)	20,8 ± 1,2 (26)	19,8 ± 1,1 (26)	18,5 ± 1,0 (24)	14,2 ± 1,3** (18)	14,8 ± 4,7 (4)
LDH4 Percent (2-10)	7,6 ± 0,5 (26)	7,2 ± 0,6 (26)	5,8 ± 0,8 (24)	6,5 ± 1,0 (18)	11,5 ± 4,9 (4)
LDH5 Percent (0-6)	3,9 ± 0,4 (26)	5,0 ± 0,8* (26)	9,0 ± 1,6** (24)	11,6 ± 4,4 (18)	22,3 ± 11,8 (4)
Alkaline Phosphatase u/l	29,7 ± 2,3 (26)	22,6 ± 2,7*** (19)	31,6 ± 4,3 (16)	34,3 ± 3,4 (18)	38,0 ± 6,5 (4)
gamma-GT u/l	25,6 ± 3,2 (26)	22,4 ± ± 4,0 (16)	25,7 ± 4,4 (13)	28,7 ± 4,6 (14)	34,3 ± 7,7 (4)

Serum enzymes before, during, and following Whole Body Hyperthermia Treatment (2 hours at 41.8°C). Means ± 1 SEM. Significance in comparison with 1 day before treatment using paired t-test. x = $p < .05$, xx = $p < 0.1$, xxx = $p < .001$. Normal values of the estimating laboratory are given in brackets under the name of the enzyme in question.

Bull et al. (1979) noted in five of fourteen patients treated for the first time for one hour at 41.8°C 'transient elevations occurring at 24 hours' in SGOT (median 2.4-fold increase) and SGPT (median 3.7-fold). Six days later the values were the normal range. CPK values were elevated but the isoenzymes did not shift from the fraction associated with skeletal muscle. Ostrow et al. (1981), noted significant elevations of LDH ($p < .05$) and SGOT ($p < .001$) 24 hours following four hours treatment at 41.8°C. Herman et al. (1982) noted that 'SGOT, CPK and LDH generally increased during the first 48 hours following therapy and then decreased to normal in the next two days'. Of their 30 treatments, 28 were at 42°C or above for periods of 2.5 to 3 hours.

The serum enzyme results that were obtained in the present series of treatments carried out at 41.8°C show similar trends to those mentioned above. Results for SGOT, SGPT, CPK, LDH and isoenzymes, alkaline phosphatase and gamma-GT are shown in the accompanying Table V.

The only changes taking place in the patients as a group at marker 7 (half an hour into the cooling period), in comparison to the day before treatment, occurred in the LDH5 isoenzyme fraction - this was then significantly increased - and in the serum alkaline phosphatase value, which had decreased to a highly significant extent.

One day after treatment significant rises in the following enzyme levels were seen: SGOT (mean 4-fold increase), SGPT (mean 2-fold increase), CPK (mean 7-fold increase), LDH (mean 1.5 fold increase), and LDH5 fraction (mean 2-fold increase).

At two days after treatment further mean rises have taken place in SGOT (now a 16-fold increase), SGPT (now a 10-fold increase), LDH (now a mean 2.7 -fold increase - this was no longer statistically significant) and LDH5 fraction (now a mean 3-fold increase - again no longer significant).

One should be wary of drawing too many conclusions from the figures presented for values obtained three days following treatment. By this time

most patients with an uneventful recovery following the hyperthermia had been discharged from hospital. If not, they were considered sufficiently 'normal' not to warrant performance of further investigations. The figures presented in the table thus consist of results from patients already having abnormalities in their serum enzyme levels. The results presented are thus biased towards abnormal levels.

Description of the enzyme changes occurring following WBHT is easy - interpreting them is considerably more difficult. From the results presented and after reviewing the work of others in this field it is tempting to suggest that a mixed syndrome might exist.

The first part of the syndrome may be composed of non-specific effects of heat on the body with particular reference to the skeletal muscular system. This tends to be reflected in changes in CPK, levels of which have been observed to rise by many authors including ourselves. From results of Bull et al. (1979) and Barlogie et al. (1979) it may be concluded that there is little CPK leakage from damaged myocardium and that the bulk of the enzyme originates in the skeletal musculature. CPK isoenzyme levels were not estimated in the present series of patients. However, the LDH isoenzyme fraction originating from cardiac muscle (LDH4) showed no rise. This would indirectly support the previous suggestions made concerning the origin of the high levels of CPK that were observed.

We cannot confirm the observations of Barlogie et al. (1979) that CPK levels were greatest following a patient's first hyperthermia treatment. There was, however, a tendency for the first treatment to produce higher levels of SGOT, SGPT, and LDH. The LDH5 fraction (which originates from the liver) also showed this trend. It should be stressed that the above remarks are only 'impressions' and, due to the low numbers of patients involved, cannot be supported by analysis of statistical significance.

In order to separate somewhat the supposed liver damage, (as reflected by

SGOT and SGPT levels), from hyperthermic muscular damage reflected by CPK changes, the results were divided into two groups. The 'normal liver' group consisted of those cases in which the maximum SGOT levels never exceeded a value of twice the laboratory normal values (up to 19 units per litre). Twice normal values were chosen as opposed to normal values due to the small numbers of patients that showed no rise in SGOT - in only five cases did the SGOT never rise above 'normal values'. The 'normal liver' group was made up of patients from ten treatments. The rest made up the 'abnormal liver' group. The CPK values following treatment in the two groups were analysed using a Students t-test. At no time were statistically significant differences seen between the groups. This would indicate that the CPK changes were largely independent of SGOT changes.

The pattern of changes observed in CPK, SGOT, and SGPT following static hyperthermia is probably species dependent in view of the findings of Hubbard et al. (1979). They observed that CPK activity in rats following hyperthermia was a less sensitive index of thermal stress than transaminases. They suggested that the release patterns of the three enzymes could be useful in differential diagnosis between heat and work induced damage. This differential pattern does not appear to exist in man.

A number of studies into hepatic integrity under hyperthermic conditions have been carried out in perfused liver models in rats. Barrows et al. (1978) observed significant decreases in bile secretion after 30 minutes at 43°C and dramatic increases in SGOT and SGPT leakage from the isolated liver after 45 minutes at the same temperature. They demonstrated relatively linear increases from 60 to 90 minutes at temperatures ranging from 39 to 42°C.

Collins et al. (1980) in an isolated perfused rat liver model using the ratio of 3 hydroxybutyrate to acetoacetate concentration in the blood draining from the liver as an indication of hepatic function (Krebs 1968) were able to demonstrate progressive decreases in function between 37° and 43°C.

Administration of palmitate significantly increased the ratio at all temperatures indicating a protective effect of fatty acids in the thermally stressed perfused liver. Collins and Skibba (1980) demonstrated that hyperthermia (43°C) largely inhibited macromolecular synthesis in the perfused rat liver by suppressing precursor incorporation into hepatic DNA, RNA and protein. Decreased gluconeogenesis, decreased urea synthesis and a decrease in lactate consumption have been demonstrated in similar models by Skibba and Collins (1978).

In summary it may be concluded that CPK changes following WBH are a reflection of thermal damage to skeletal musculature and that, in our experience, rises in SGOT, followed at a later stage by SGPT, are a reflection of hepatic damage which tends to be most severe following a patient's first WBH treatment.

CASE REPORT OF A CASE OF FATAL HEPATIC NECROSIS

One patient in the present series died of hepatic necrosis and a case report will be presented.

The patient, Mrs D, was a 51 year old housewife, who was admitted to the hyperthermia unit for combined radiotherapy and whole body hyperthermia treatment. Four years previously she had attended the outpatients clinic complaining of pain in the left upper jaw, the severity of which had increased over the last year. Examination revealed a swelling in the alveolar process of the maxilla, extending into the soft palate. Biopsy revealed a cystic adenocarcinoma and a resection of the left upper jaw was carried out under general anaesthesia. No halothane had been administrered on this, or any other occasion. One month before the operation she had undergone a course of local

radiotherapy of 20 x 200 Rads and 3 months after the maxillary resection she had also received a course of radiotherapy - this time she received 20 x 150 Rads.

When presenting for hyperthermia the patient was symptom free, but on a routine follow-up chest x-ray scatterd metastatic deposits had been observed. Old tuberculous scarring was also visible - the patient had undergone drainage of a tuberculous empyema forty four years previously. Before the hyperthermia treatment, she received a prophylactic course of isoniazid. Further medical history was uneventful.

The patient presented with a palato-maxillary prosthesis in the left upper jaw. Obvious trismus was present and the mouth could not be further opened than about 2 centimeters (as measured between the upper and lower incisor teeth). Blood pressure was 140/90 and auscultation of heart and lungs revealed no obvious abnormality. She was well nourished and she weighed 69 kg.

No contraindictions to whole body hyperthermia treatment were noted on preliminary examination. Heart function, as assessed by bicycle ergometry, was good, and the patient performed well at 120 watts. This resulted in a maximum pulse rate of 150 beats per minute and resulted in no changes in heart rhythm on the electrocardiogram. No repolarisation disturbances were seen. Pulmonary function testing revealed that the patient had a vital capacity of 3,5 liters with a 1 second forced expiratory volume of 72 percent of the vital capacity. The maximum breathing capacity was 150 liters per minute. All three values may be considered as normal. The arterial blood gas and acid/base values were normal.

Isotrophic cerebral scanning revealed no abnormality, but on liver scanning a zone of hyperactivity was seen at the hylum, indicating the presence of a space occupying lesion. Further investigation with cholescintography revealed good hepatocyte function, but there was somewhat delayed excretion into an enlarged gall bladder.

Routine laboratory values revealed no gross abnormality : Alkaline phosphatase was 28 U/l; gamma-GT 11 U/l; SGOT 9 U/l; SGPT 5 U/l; LDH 172 U/l and Bilirubin <10 micromol/l. LDH fractions were: LDH1 - 15%; LDH2 - 56%; LDH3 - 15%; LDH4 - 10%; LDH5 - 4%. The haemoglobin concentration was 7,8 mmol/l.

Hyperthermia Treatment

On the day of treatment the patient was premedicated in the usual way with 15 mg of papavaratum and 0.25 mg of hyocine given intramuscularly one hour before she was brought to the hyperthermia treatment room. She was placed in the hyperthermia cabin - the perspex cover was first removed - and an infusion of Ringer lacatate solution was started after insertion of a cannula into a vein on the dorsum of the left hand.

An intraarterial catheter was inserted percutaneously into the radial artery of the left wrist, after first establishing that good collateral circulation was present from the ulnar artery. This catheter was advanced into the brachial artery and connected in the usual way to the pressure transducer. A Swan Ganz catheter was inserted into the left subclavian vein and advanced, under protection of a intravenous bolus of 100 mg of lignocaine, into the pulmonary artery.

Anaesthesia was induced with 100 mg of methohexitone intravenously, preceeded by 5 mg of d-tubocurarine to prevent fasiculations following the suxamethonium, 100 mg of which was administered after the patient was asleep.

In view of the trismus that was present, the patient was intubated using the retrograde intubation technique (Faithfull, 1982). An epidural catheter was passed through a Tuoey needle, inserted through the cricothyroid membrane. After passage through the larynyx, this catheter was retrieved from the nasopharyx and attached to a suction catheter, which was then drawn down through the vocal chords. An 8.5 mm diameter armoured latex cuffed endotracheal tube

was passed over the suction catheter and into the larynyx. The epidural catheter and suction catheter were then removed through the endotracheal tube. This intubation was accomplished rapidly and without difficulty.

The patient was further relaxed with 25 mg of d-tubocurarine after the action of the suxamethonium was seen to be over. A further 10 mg was administered 10 minutes later when it became clear that paralysis was not complete. The patient was ventilated with a Siemens Elema Servo ventilator 900A with a mixture of 33% oxygen and 66% nitrous oxide. Tidal volume was 900 ml and the frequency was 10 respirations per minute.

Warming was commenced after covering the patient with plastic foil and this was accomplished using hot air in the cabin and circulation of warm water through the warming mattress on which the patient lay. The body temperature was raised from 37.8°C to 41.8°C in 90 minutes.

During the initial warming period the left ventricular work index rose sharply from 4 kg m min-1 at 37.8°C to 7.7 kg m min-1 at 39.5°C. This was more than usual for the group of patients as a whole. Small (5 mg) incremental doses of dehydrobenzperidol were administered and this resulted in decreases in systemic vascular resistance and left ventricular work.

At 41.6°C body temperature, the left ventricular work again showed a tendency to rise and 0.5% halothane was added to the inspired mixture. Ten minutes later this was increased to 1% and the patient was maintained on this concentration until an hour and a quarter of plateau had elapsed. Mrs D. began to 'fight' the ventilator at this stage and 1.5% halothane was given for a further 15 minutes by which time the mean systemic blood pressure had slowly decreased to 50 mm Hg. This was considered to be rather low and the vaporiser was turned off. The systemic pressure rapidly recovered to a mean value of 64 mm Hg.

It should be noted that the cardiac index was considered to be quite adequate at all times and never fell below 5 l min-1 m-2. It should further be

noted that mean arterial pressures between 50 and 60 mm Hg were not at all uncommon in the present series of treatments. These levels were seen at one of the fixed marker points (3, 4 or 5) in 15 of the 30 treatments.

After switching off the halothane 10 mg of d-tubocurarine was given. Further movement during the last minutes of the plateau phase was controlled with 1% halothane. This was switched off as the mean systemic pressure again started to fall - this time the lowest level reached was 55 mm Hg.

Cooling was uneventful - a dip in mean arterial pressure from 60 to 57 mm Hg was noted. At this stage the arterial line became blocked but further measurements revealed systolic blood pressures (measured with a von Recklinghausen oscillotonometer) remained above 90 mm Hg for the rest of the treatment.

Arterial oxygen saturation at plateau was 94.9%. This was well within one standard deviation for the group, as was the oxygen consumption, oxygen flux and the ratio of the two. It might be suggested that changes in liver function might result, in part from hypotensive episodes occurring during the plateau phase of treatment. However, statistical analysis (Student t-test) of SGOT values 48 hours following treatment revealed no differences between the 9 treatments in which mean arterial values were between 55 and 50 mm Hg and the 7 in which pressures were between 60 and 55 mm Hg. There were also no differences between this group and the 14 treatments where the pressures were always above 60 mm Hg. In short there was no indication that the changes in cardio-respiratory parameters was abnormal from the group of patients as a whole.

Post-treatment Period

Immediately after treatment slight electrolyte disturbances were found such as are found after every treatment. Levels of calcium, potassium and phosphate were low but caused no anxiety.

Five hours after opening the cabin the potassium level in the blood had

fallen to 3.1 mmol/l - this was corrected to 4.6 mmol/l by 9 hours of cooling by administration of 2 gm of potassium chloride in the intravenous infusion. By the following morning the calcium level had fallen to 1.76 mmol/l and 1 gm of calcium chloride was given intravenously. The phosphate levels had fallen from 1.14 mmol/l pretreatment to 0.66 mmol/l at this time (this is just within the 'normal' level for the estimating laboratory).

Haematological investigations the following morning gave indications of a mild intravascular coagulation syndrome. This normally recovers by 2 days post treatment without further therapy, but, in this case, on the second post treatment day, abnormalities were still present. Fibrinogen levels had decreased to 0.4 gm/l (pretreatment level 2.6 gm/l) and thrombocytes were at a level of 29,000 per mm3 (pretreatment level was 230,000 per mm3). Fibrin breakdown products were detected in the plasma at levels between 40 and 80 mg/l. Treatment was instituted with low dosage of heparin and fresh plasma. Following this regime the clotting parameters gradually improved. At this time (2 days) prothrombin time was 3.9 x normal, thrombin time was twice normal and parital thromboplastin time was 1.3 x normal.

It should be noted that, in the recovery period, the cooling rate of the patient in the intensive care unit was very much slower than in the group as a whole. Between two to five hours after cooling had been commenced, all temperature measurements revealed values that lay more than one standard deviation higher than the mean of the group. In the series of patients here described, it was noted that slower cooling tended to be associated with evidence of liver function changes following treatment.

In order to further investigate this phenomenon, two groups of patients were retrospectively studied. One group consisted of four patients in whom the highest levels of SGOT were obtained two days after treatment. The other group was composed of patients in whom the lowest levels of the enzyme were found. The difference between the two groups with respect to the SGOT levels was very

significant ($p<.01$). The body temperature differences between these two groups of patients was analysed using a Student's t-test. It was found that the body temperature of the high SGOT group was always significantly higher than in the low SGOT group between 12 and 18 hours after cooling had started. The differences at earlier stages in the cooling period were also largely significant. At all times between two hours of cooling and eleven hours of cooling significant differences in body temperature was found between the two groups ($p<.05$) with the exception of two hours of cooling ($p<.2$), seven hours of cooling ($p<.1$), and ten hours of cooling ($p<.1$).

It is interesting, at this point, to remember the work of Hubbard et al. (1977), which has already been referred to in chapter I, in which they were able to find a relationship between cooling speed and survival in hyperthermically stressed rats. It may be that the severity of liver damage in patients could be reduced by more attention to rapid cooling.

It is, of course, allmost impossible to determine if the slower cooling was as a result of the production of pyrogenic substances by an already compromised liver or if the damage was caused by prolongation of the 'hyperthermia' - though this was very low grade in the post treatment phase. In view of the fact that Pettigrew and Ludgate (1977) held patients' temperatures at, or just below, 41.8°C for 'about 4 hours' without observing significant toxicity would point to the former possibillity as being more probable.

At two hours of cooling (on entry into the intensive care unit) Mrs D's pulse rate was more than one standard deviation lower than the mean of the group as a whole, and her mean systolic pulse pressure was more than two standard deviations lower than the mean of all patients. Cardiac index was, however, normal and, on clinical grounds, there was no reason to suspect that her oxygen flux was diminished. As the arterial line had unfortunately become irretrievably blocked at the end of treatment, no blood gas data are available for the intensive care recovery phase of this patient. The heart rate remained

low, (more than one SD), for a further hour and the mean arterial pressure was low, (more than one SD), for a further three hours. Otherwise there were no marked differences, from the group of patients as a whole, in the measured or calculated cardiovascular parameters during the recovery phase of this patient.

Measurements of the serum enzyme levels the day following treatment were as follows: SGOT 47, SGPT 16, CPK 191, and LDH 266 U/l. LDH isoenzymes were as follows: LDH1 22%, LDH2 48%, LDH3 13%, LDH4 7%, and LDH5 10%. All these values were within one SD of the mean of all treatments with the exception of LDH2 which was more than one SD raised, and LDH3 more than one SD lower. The total bilirubin level was 20.7 micromol/l (13,2 micromol/l was in the conjugated form).

Two days after treatment the following values were seen: SGOT 1540, SGPT 805, CPK 149, and LDH 3586 U/l. LDH1 was 3%, LDH2 7%, LDH3 2%, LDH4 12%, and LDH5 had risen to the huge value of 76%. All these values were outside two standard deviations of the mean of the group and it was obvious the patient was severely ill. The bilirubin level had risen to a total of 62.4 micromol/l (46.5 micromol/l was in the congugated form).

The patient's condition continued to deteriorate and at three days post treatment the SGOT had risen further to 1738 U/l and the SGPT to 2002 U/l - the LDH was 2185 U/l. The total bilirubin had risen to 118 micromoles per litre. The patient died on the eighth post treatment day in hepatic coma. Renal function had remained good throughout the course of the hepatic problems.

Autopsy revealed massive hepatic necrosis and a few small metatstatic seconday deposits were seen at the hylum of the liver. No indications of thrombotic processes or bleeding were observed.

Discussion

The cause of the fatal hepatic necrosis in this patient was not

immediately obvious, but as soon as it was realized that the patient had received halothane during treatment, suggestions were made that this was the cause of the disaster. A number of very highly placed, and otherwise very knowledgable physicians are under the impression that halothane is a highly 'hepatotoxic' agent.

Early findings by Little et al. (1958) indicated that hepatic functions were not more affected following anaesthesia by halothane than following anaesthesia by cyclopropane and ether. Bunker and Blumemfeld (1963) reported two cases of fatal hepatic necrosis following halothane but they concluded that it was not possible to implicate the anaesthetic in either case. They pointed out that in the U.S.A. 15-20 patients per year, might, on epidemiological grounds, be expected to contract infectious hepatitis within 5-10 days of surgery. Mushin et al. (1964) after a retrospective study of 15,774 halothane, and 6,123 non-halothane anaesthetics concluded that there was no evidence to show whether halothane was producing a specific toxic or allergic affect. They were also unable to detect any important differences with respect to the liver pathology between the halothane and the non-halothane group. They also concluded that there was 'unlikely to be a graduated response on the liver related to dose'.

In 1966 the subcommittee of the national halothane study presented its findings in the U.S.A. They performed a comparative study of halothane and other anaesthestics as to the incidence of fatal massive hepatic necrosis within six weeks of anaesthesia. They examined 82 cases of necrosis. Of the 9 'unexplained' cases, 7 had received halothane for the final operation, and 4 had received 2 halothane anaesthetics within 6 weeks of the final operation. They studied more than one million anaesthetics and the committee commented that 'the study failed to establish a causal relationship between halothane and hepatic necrosis'. Klion et al. (1969) concluded that there was no evidence that preexisting liver disease increased the risk of adverse hepatic reactions

to halothane.

Mushin et al. (1971) examined the pattern of incidence of previous anaesthetics and found that there was a significant association between post-anaesthetic jaundice and a previous administration of halothane when the interval between the two halothane administrations was 4 weeks or less. Simpson et al. (1971) however, considered that it was still not possible to conclusively rule out viral infection as a cause of liver damage in an individual case. Later Inman and Mushin (1978) further reviewed jaundice following multiple exposures to halothane and found that 75% of patients reported as 'posthalothane jaundice' had received a previous halothane anaesthetic within 4 weeks.

In a recent review Cousins (1980) has discussed possible precipitating factors in the production of 'halothane hepatitis' and pointed out that under conditions of decreased hepatic oxygen delivery, binding of reactive halothane intermediates (which may be capable of direct hepatic damage) might be increased.

From the evidence available it was not considered that Mrs D. had suffered from hepatic hypoxia during WBHT. At plateau temperature she had a high cardiac index (6.2 l min^{-1} m^{-2}) and normal mixed venous oxygen saturation (72.1%) in the presence of an adequate haemoglobin level (10.0 gm per 100 ml of blood). She had not received halothane within 4 weeks of WBHT - indeed she had never received the drug. It was therefore not considered that the use of halothane had contributed to her tragic demise.

A further factor that might possibly have had an influence on the hepatic function of this patient was the pretreatment by isoniazid. Hepatoxcity following administration of this drug has been reported and these effects are often more severe if isoniazid administration is repeated after a period of abstinence (Repertorium Verpakte Geneessmidelen, 1976). The incidence is low (0.1% - Elis, 1980) and tends to occur during the early months of therapy. The

prognosis is almost always good and, if treatment is interrupted, elevated chemical values return to normal (Meyer and Hoigné 1980).

The question arose as to the likehood of isoniazid producing enzyme induction. This is especially important in view of the reports of Cousins et al. (1979) of hepatoxcity produced in rats by hypoxic administration of halothane in the presence of phenobarbitone. Blair and Levin (1977) reported 3 cases of hepatic failure following WBHT. Two patients had been regular consumers of alcohol and the third had received longterm treatment of epilepsy with phenobarbitone. The authors suggested that alcohol and enzyme inducing drugs could increase the incidence of liver damage above 42°C. No details of anaesthetic techniques or cardiorespiratory parameters were given for these patients.

It is not clear whether isoniazide can, in fact, produce enzyme induction and it is not included in the list of more than 200 enzyme inducing drugs given by Conney (1967).

It was concluded after lengthy discussion that neither halothane nor isoniazid had a significant influence on the course of the hepatic necrosis in this case. It was clear that further work was necessary to elucidate the cause of hepatic necrosis after WBHT.

HEPATIC BLOOD FLOW ESTIMATIONS IN PATIENTS UNDER HYPERTHERMIA

As a result of the above conclusions, it was decided to estimate the total splanchnic blood flow during subsequent WBH treatments. Monitoring of sodium, potassium, lactate, ammonium, SGOT and SGPT levels in hepatic venous blood was carried out after sampling through a catheter introduced via the femoral vein into one of the hepatic veins.

The continuous infusion technique for estimation of hepatic blood flow was introduced by Bradley et al. (1945). This technique is based on the Fick principle and was introduced using bromsulphthalein (BSP). The use of this substance has largely been superceded by the use on indocyanine green (ICG) which has been shown to have no extrahepatic extraction routes (Caesar et al. 1961). Estimated hepatic blood flow (EHBF) is calculated from the formula:

$$EHBF = \frac{\text{ICG infusion rate (mg mins-1)}}{\text{ICGa (mg ml-1)} - \text{ICGhv (mg ml-1)}} \times \frac{1}{I - Ht}$$

where:

ICGa = Systemic arterial concentration of ICG

ICGhv = hepatic venous concentration of ICG

and Ht = the percentage haematocrit of the patients blood.

Dye determinations were performed photometrically using a Carl Ziess PMQ11 spectrophotometer. Determinations of ICG contents were performed in undiluted plasma. As the blank densities of the plasma appear to change during treatment, a blank correction was used as applied by Winkler and Tygstrup (1960). Dye concentration was calculated as:

$$\frac{D805 - f \times D600}{E805 - f \times E600}$$

where:

D = optical density at 805 or 600 namometres

E = extraction coefficient of ICG in plasma at the same wavelengths,

and f = plasma blank factor ie $\frac{\text{plasma blank at 805}}{\text{plasma blank at 600}}$

A 15 mg loading dose of ICG was given intravenously before anaesthesia was induced and continuous infusion was carried using a Hospal K10 infusion pump at a dose of 0.333 or 0.167 mg per minute. At least 20 minutes were allowed for equilibriation before estimations were made. Samples of arterial and hepatic venous blood were measured for estimation of total liver blood flow at the following times: before induction of anaesthesia (marker 1), at the start of warming (marker 2), beginning of plateau (marker 3), mid-plateau (marker 4),

TABLE VI Hepatic blood flow studies during hyperthermia

Treatment number and maximum SGOT post treatment	Marker 1 Before induction of anesthesia		Marker 2 At beginning of warming		Marker 3 on reaching 41.8°C		Marker 4 after one hour at 41.8°C		Marker 7 After half an hour of cooling	
	EHBF	Extraction of ICG	EHBF	Extraction of ICG	EHBF	Extraction of ICG	EHBF	Extraction of ICG	EHBF	Extraction of ICG
Treatment 20 Maximum SGOT - 18	2300 (39,6%)	24,4%	1350 (39,5%)	32.2%	1960 (20,2%)	20,3%	2580 (24,7%)	20,5%	2470 (29,8%)	16,46%
Treatment 21 Maximum SGOT - 336			2250 (35.9%)	44.7%			7640 (50.9%)	13.4%		
Treatment 22 Maximum SGOT - 72	2180 (35.3%)	38,4%	1380 (33,0%)	47,1%	4020 (30,2%)	19,0%	6890 (51,7%)	10.7%	5220 (36,4%)	13,1%
Treatment 25 Maximum SGOT - 29			1880 (49.8%)	64.8%	3410 (34.4%)	57,6%	5900 (52.9%)	45,7%	3050 (29.7%)	42,7%

Estimated hepatic blood flow (EHBF) using indocyanine green (ICG) clearance techniques. EHBF is expressed as ml per minute. The figures in brackets respresent EHBF as a percentage of the cardiac output. Hepatic ICG extraction is expressed as percentage.

and half an hour after opening the hyperthermia cabin (marker 7). In addition to total splanchnic blood flow the hepatic extraction ratios for ICG were calculated from the following formula:

$$\text{Extraction} = \frac{ICGa - ICGhv}{ICGa}$$

Results

Results of ICG estimations of total splanchnic blood flow from 4 WBH treatments are shown in Table VI. The flows are also expressed as a percentage of the cardiac output. Details are also given of the calculated extraction of ICG, and of the maximum SGOT levels measured following treatment. As can be seen, estimated flows increased dramatically under conditions of hyperthermia, while at the same time ICG extraction decreased. The greatest changes occurred in patients having the greatest rises in SGOT following treatment.

Discussion

Values for liver blood flow in the awake state have been estimated by Cooperman et al. (1968) using ICG clearance methods. They measured the flow in six subjects and they range between 1.38 and 2.93 litres per minute, with a mean of 2.06 liters per minute. The two measurements obtained in the this study in the awake patients fall into that range. In four patients Cooperman et al. (1968) measured cardiac output. The splanchnic blood flows ranged from 27.9 to 50% of the cardiac output - again our figures fall into that range, though figures of 25% of cardiac output are often quoted (Strumin 1980). Cooperman et al. (1968) studied the effect of anaesthesia with nitrous oxide/oxygen and hyperventilation under d-tubocarine. This is the same sequence as was used in the patients reported here. Splanchnic blood flow in their subjects fell to a mean of 1.4 l/min (range 0.73 to o.41) and this represented a mean of 25% of cardiac output (range 12.4 to 29.5%). Our estimated flows did not fall to this extent. Mean flows of the patients in this series were 1.7 litres per minute and this represents a mean of 35.5% of the cardiac output.

When looking at the estimated flows presented in Table VI the most obvious change to be seen, under hyperthermia, is a very large increase in splanchnic blood flows - in three cases to 50% or more of the cardiac output. It is most unlikely that splanchnic blood flow would rise to these levels under conditions of hyperthemia in view of the large increase in skin blood flow that is known to occur (Wyss et al. 1974). This increase in skin blood flow, which is partially mediated by a decrease in venomotor reflex tone, (Zitnik et al. 1971), may reach large proportions and the flow may be such that there is virtually no difference in blood gas concentrations between blood in the radial artery and that in the forearm vein once body temperature has reached 38°C (Lees et al. 1980).

Further discussion of splanchnic blood flow will be given in a later section, but it should be noted that in all cases ICG extraction had decreased by the time midplateau (marker 4) had been reached, indicating decreased ability of the liver to excrete the drug.

There were no significant differences in the blood concentrations of sodium, potassium, SGOT, SGPT, ammonium or lactate between blood drawn from the general systemic circulation and hepatic venous blood in the nine patients in whom the values were estimated.

Oxygen saturation of hepatic venous blood was followed during WBHT in seven treatments. There was a significant ($P<.05$) fall at marker 3 (beginning of plateau). This fall became nonsignificant when the one patient in the group who had a marked rise of SGOT at 48 hours, was excluded. He had a 48 hours SGOT value of 336 units as against the other six with a mean value of 35.8 units (SEM $\pm$ 3.2). His values for both SGOT at 48 hours and oxygen saturation of the hepatic blood at markers 3 and 4 (beginning and mid plateau) were all more than 2 standard deviations outside the mean of the group.

In view of the unreliability of the estimated total splanchnic blood flow in these patients, it is almost impossible to draw any firm conclusions from

the arterial and hepatic venous saturation values. It can only be stated that in the one patient referred to hepatic venous oxygen content was very low and amounted to 5.6 ml/100 ml of blood.

It became clear that, in man, estimation of total hepatic blood flow under hyperthermia using ICG clearance methods was unreliable and it was decided that animal experiments should be performed.

EXPERIMENTAL STUDIES OF HEPATIC BLOOD FLOW

Dye Clearance Methods

Immature female Yorkshire pigs weighing between 24 and 26 kg were used in the study. Anaesthesia was induced with an intraperitoneal injection of sodium thiopentone (30 mg per kg body weight). After intubation with a cuffed endotracheal tube the animals were ventilated using a Bennett anaesthesia ventilator with a gas mixture of 33% oxygen and 67% nitrous oxide. The expired carbon dioxide was measured with a Goddard type 146 capnograph and the ventilation was adjusted to maintain an expired concentration of carbon dioxide between 4 and 5%. Pancuronium bromide was used to maintain muscular relaxation and this was administered intravenously using a Braun Melsungen Perfuser adjusted to administer the drug at a rate of approximately 0.5 mg per kg per hour.

A 5 mg loading dose of indocyanine green was injected intravenously and it was then administered using a continuous infusion with another Braun Melsungen Perfusor appartus at a rate of 4.75 mg/hr. Through a skin incision in the groin a 7 French 80 cm cordis femoro- renal A2 catheter was introduced into the femoral vein and advanced under x-ray control till its tip lay in one of the

main hepatic veins - this position was checked by injection of radio-opaque dye (Urografin-Schering AG). This catheter was used to sample hepatic venous blood. Arterial blood was sampled through a 1.85 mm bore catheter introduced into the femoral artery through the same incision.

After a minimum of 30 minutes of indocyanine green infusion, heparinized hepatic venous and arterial blood samples were withdrawan and centrifuged. The concentration of indocyanine green in the supernatant plasma was estimated by absorption spectometry at 796 nanometers wavelength using a Vitatron UC 200S photomometer and comparing the extinction so obtained with a calibration curve previously constructed using freshly prepared indocyanine green solution.

The estimated hepatic blood flow (EHBF) was calculated from the formula:

$$\text{EHBF (ml min-1)} = \frac{\text{ICG infusion rate (mg min-1)}}{\text{ICGa (mg ml-1)} - \text{ICGhv (mg ml-1)}}$$

Where:

ICGa = Arterial concentration of ICG

and

ICGhv = Hepatic venous concentration of ICG

Hepatic extraction percentage (E) was calculated using the formula:

$$E = \frac{ICGa - ICGhv}{ICGa}$$

In two of the six pigs a further estimation was performed after at least one hour's pause to check the reproducability of the measuring methods.

Using a water circulating blanket and a Churchill thermo-circulator LTCM thermostatically controlled heater circulator four of the pigs were heated to the temperature range of 39° to 40°C and the EHBF estimations repeated. The blood temperature in the pulmonary artery was taken as core temperature and this was measured using the thermistor tip of a 7 French gauge KMA thermo-dilution Swan Ganz catheter. Two of the four pigs were further heated till their temperatures were above 40°C and again EHBF estimations were repeated.

TABLE VII Indocyanine green clearance studies in pigs

Pig number	Before Warming		39° - 40°C		More than 40°C	
	EHBF	ICG Extraction	EHBF	ICG Extraction	EHBF	ICG Extraction
1	48,1	16,6				
2	61,1 55,1	25,57 26,3				
3	52,4	11,3	137,5	3,5		
4	57,9	16,1	550,0	2,1		
5	40,7 40,7	18,0 22,4	112,0	2,6	infinite	zero
6	47,0	19,9	39,2	22,7	infinite	zero

Estimated hepatic blood flow (EHBF) in six yorkshire pigs (24-26 kg body weight) at various temperatures measured by indocyanine green (ICG) clearance techniques. Blood flow is expressed in ml per kg body weight per minute and ICG extraction as percent.

Results

The results of the hepatic blood flow estimations and ICG extraction rates are presented in Table VII. As can be seen, at normothermia, there is little difference between the various animals in terms of estimated flow, but in the range of 39° to 40° an enormous range of calculated flows were obtained. Some of the flows are clearly incorrect. In both animals above 40°C body temperature there was no measureable difference between ICG concentrations in the hepatic venous blood and the arterial blood. Calculations thus yielded 'infinite' blood flow and zero extraction.

Discussion

EHBF in pigs has been measured by various authors using ICG clearance techniques. Lucke and Hall (1978) using Pietrain pigs with a mean body weight of 67,3 kg obtained average flows of 16.3 ml kg-1 min-1 - whereas Imamura and Clowes (1975) employed pigs with a mean body weight of 25,8 kg and estimated mean flows of 44,2 ml kg-1 min-1. The weights of their pigs compare well with those used in the present study (mean body weight 25,2 kg), as do flows obtained. The pigs in this study had mean flows of 50,7 ml kg-1 min-1.

Imamura and Clowes (1975) obtained good correlation (correlation coefficient of 0,8) between their flows measured by indocyanine green clearance and direct measurements obtained by a method employing two Swan Ganz thermodilution catheters positioned in the inferior vena cava above and below the openings of the hepatic veins. Mean flows of 43,6 ml kg-1 min-1 were obtained using this method.

In view of the clearly incorrect estimations made by ICG clearance methods under hyperthermic conditions it was decided to further pursue the study using

other methods.

Thermodilution Techniques

It was decided to employ the dual Swan Ganz catheter method of measuring total hepatic blood flow as used by Imamura and Clowes(1975). Pigs of similar body weight to those used in the ICG clearance studies were anaesthetized as described above.

Via an incision in the upper right side of the throat of the animal, a seven french gauge swan ganz thermodilution catheter was inserted as high as possible into the right internal jugular vein. This was advanced under pressure monitoring through the right atrium and right ventricle until its tip lay in the pulmonary artery. It was connected to a KMA thermodilution cardiac output computer model 3500. Cardiac output could then be measured by injection of 5% dextrose solution through the proximal line of the Swan Ganz catheter.

A seven french femororenal catheter was inserted into the left femoral vein in the groin and advanced under fluoroscopy until its tip lay in one of the hepatic veins as described earlier. Into the other femoral vein two further seven french Swan Ganz thermodilution catheters were inserted. One was advanced until its thermister tip lay in the thoracic part of the inferior vena cava half way between the openings of hepatic veins and the exitus of the vena cava into the right atrium. The other Swan Ganz was positioned with its tip just distal to the exit of the hepatic veins into the inferior vena cava. The position of these catheters was checked under fluoroscopy by injection of a radiopaque dye (Urografin-Schering AG).

Using this experimental arrangement in conjunction with the KMA thermodilution cardiac output computer, it was possible to measure the flow at the tips of both catheters. Hence, by subtraction, total blood flow from the

hepatic veins, and hence total splanchnic blood flow could be estimated.

It was found that this technique was only applicable to pigs of 24 kg or more bodyweight. Otherwise it was not possible to insert sufficient length of the lower catheter to enable measurements to be made. This was due to the 30 cm distance between the proximal injection opening of the Swan Ganz catheter and the thermistor measuring tip. A standard catheter was used to ensure a good mixing of the injectate.

Two flow measurements were taken at two phases of the respiratory cycle fifty per cent apart and the Bennett ventilator was set to deliver a 1:1 inspiratory/expiratory ratio. Recent work by Janssen et al. (1981) would indicate that the mean of the two results so obtained should have a good correlation with the mean flow during the respiratory cycle. In total, for each measurement 3 paired measurements were made and the results averaged.

In order to determine the reproducibility of the method, paired estimates of liver blood flow (separated by 15-45 minutes) were performed on six pigs.

The pigs were then heated as described in the previous section on dye dilutional methods. Measurements were performed at the following points: normothermia (between 37° and 38°C); between 39° and 40°C body temperature; between 40° and 41°C; and above 41°C. Body temperature was again taken as the temperature measured by the thermistor tip of the Swan Ganz catheter positioned in the pulmonary artery.

Arterial and mixed venous blood samples were taken at the time of estimations of liver blood flows, and blood gas and acid/base values were estimated using a Radiometer ABL1 acid base laboratory. The oxygen saturations of the blood and haemoglobin concentrations were measured using a Radiometer OSM2 hemoximeter.

From the above measurements the total splanchnic oxygen consumptions were calculated using the formula:

TABLE VIII Splancnic blood flow in pigs

Paired thermodilution estimations of total splancnic blood flow	Indocyanine green clearance estimations of total splancnic blood flow
64.2	41.8
63.8	61.1
52.5	55.1
44.6	52.4
50.4	57.9
48.8	40.7
52.4	47.0
52.4	40.7
60.7	
53.5	
46.2	
48.5	
	significance using Student's T test
Mean 53.2	50.4
SE ±6.5	NS ±7.6
n (6)	(8)

Paired thermodilution estimations of total splancnic blood flow and estimations of total splancnic blood flow using ICG clearance techniques. Pigs at normal body temperatures.

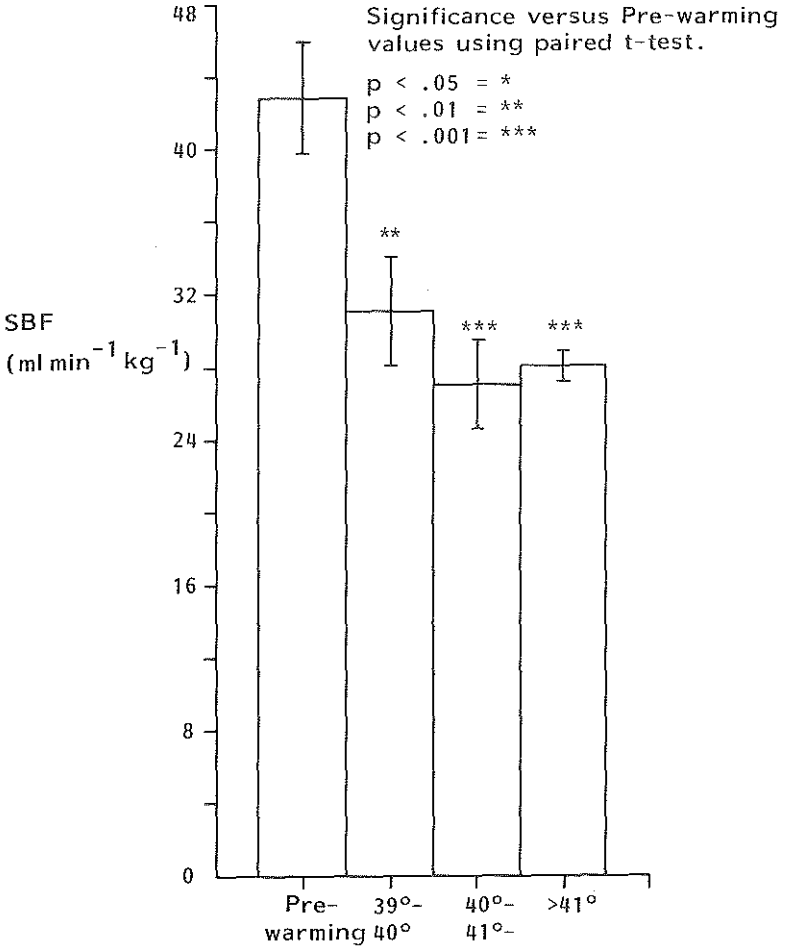

Fig. 9. The effect of Hyperthermia on splancnic blood flow (SBF) in pigs. Means ± SEM.

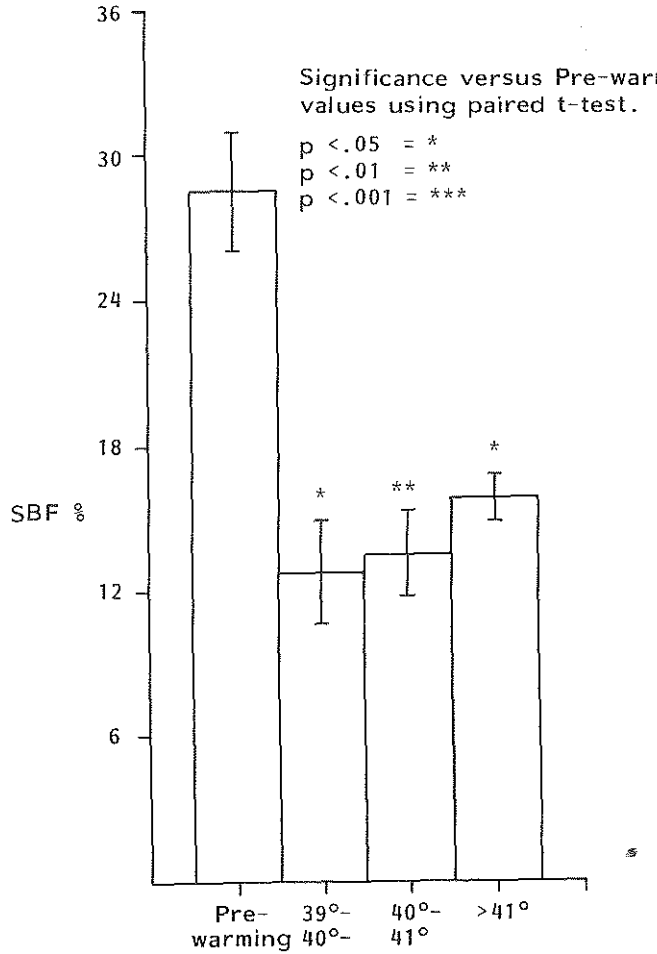

Fig. 10. The effect of Hyperthermia on splancnic blood flow as percent of cardiac output (SBF%) in the pig. Means ± SEM.

$$\text{Splanchnic oxygen consumption} = \frac{(CaO2 - ChvO2) \times CO}{10}$$

where:

CaO2 = oxygen content of systemic arterial blood in ml/100 ml blood

ChvO2 = oxygen content of hepatic venous blood in ml/100 ml blood

and CO = cardiac output in litres per minute

CaO2 was derived from the formula:

$$CaO2 = Hb \times SaO2 \times 1.306,$$

and ChvO2 was determined from the equation:

$$ChvO2 = Hb \times ShvO2 \times 1,306$$

where:

SaO2 = percentage oxygen saturation of systemic arterial blood

and ShvO2 = percentage oxygen saturation of hepatic venous blood.

Results

In Table VIII results are presented of the paired thermodilution estimations of liver blood flow. Also presented from comparison are the (largely unpaired) estimations of liver flow using the ICG clearance techniques. It should be noted that these were performed in a different group of animals, and hence no comparison can be made between the individual figures in the two columns.

There was no statistically significant difference (using the Student's t-test) between the results obtained by the two methods. The correlation coefficient between the two measurements making up the pairs of the thermodilution measurements was 0.81 ($P < .05$).

Figure 9 is a histogram representation of the changes of total splancnic blood flow against temperature. As can be seen, statistically significant reductions in flow occurred at all temperatures above 39°C.

In figure 10 these changes are plotted as a proportion of the cardiac output - again, all falls are significant.

In figure 11 splancnic oxygen consumption per kilogram bodyweight is plotted

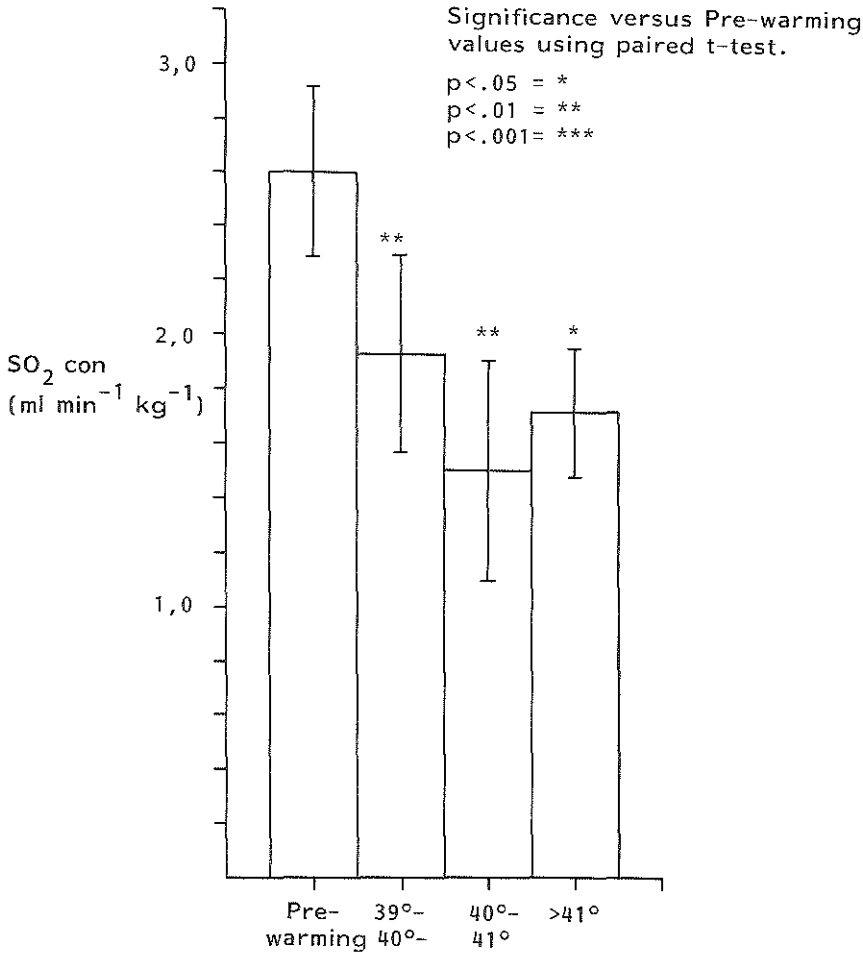

Fig. 11. The effect of Hyperthermia on splancnic oxygen consumption (SO_2 con) in the pig. Means ± SEM.

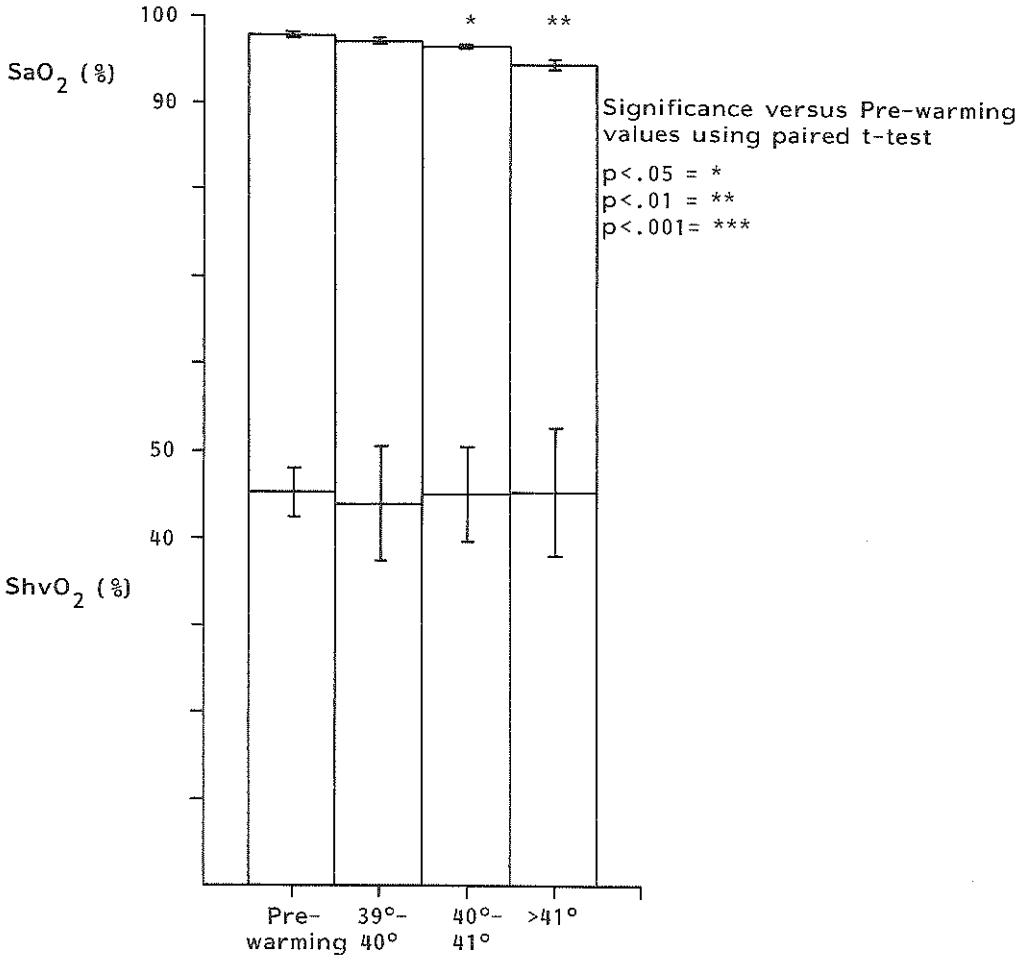

Fig. 12. The effect of Hyperthermia on arterial oxygen saturation (SaO_2) and hepatic venous oxygen saturation ($ShvO_2$) in the pig. Means ± SEM.

against temperature. Significant decreases occurred when the animals' body temperatures were raised above 39°C.

In figure 12 changes with temperature of both systemic arterial oxygen saturation and hepatic venous oxygen saturation are presented. Whereas the arterial saturation fell significantly on raising body temperature, there were no significant changes in the hepatic venous oxygen saturations.

Discussion

Investigations of hepatic blood flow under hyperthermic conditions have been carried out in a number of animal models and also in man. In the latter case the rise in body temperature was not nearly as much as is induced during WBHT.

Traks and Sancetta (1959) have studied the effects of raised body temperature in normal human subjects using bromsulphalein (BSP) clearance and found no change in estimated hepatic blood flow. It should be noted that the average body temperature was only raised by 0.8°C. Using the same techniques under conditions of induced pyrexia, Bradley and Conan (1947) found that EHBF increased significantly with concomitant reduction in BSP extraction.

Using ICG clearance methods, Rowell et al. (1970) have recorded decreases in EHBF in spontaneously ventilating human volunteers. The blood temperature in the right atrium was between 39.05'C and 39.44'C and the mean decreases in flow amounted to 34%. This compares quite well with the percentage fall that was observed in the pig (29,2%) between normothermia and 39°- 40°C. Rowell et al. (1970) noted that the splancnic blood flow began to decrease near the time that the blood temperature began to rise. They stated that the hepatic extraction remained constant at 85% and they postulated that splancnic vascular resistance might have been increased via central baroreceptor reflexes in the presence of a decreasing mean arterial pressure. However, in a further study,

Rowell et al. (1971) demonstrated that falling mean arterial pressure or aortic pulse pressure were not major causes of the splancnic vasoconstriction in response to heating in the human subject.

Using radioactive microspheres to study the splancnic blood flow in unanaesthetised baboons Hales et al. (1978) have demonstrated a 35% reduction in flow when the body temperature was raised by a mean of 1.7°C. The study further revealed that there is no rise in cardiac output under these conditions. This is in contrast to man, in whom very large increases in cardiac output are found (Rowell et al. 1970). The baboon would appear to differ from man in a number of its thermoregulatory mechanisms as a lack of human like active skin vasodilation under conditions of hyperthermia was demonstrated in heat stressed baboons by Wyss and Rowell (1976).

In a study using radioactive microspheres, Hales (1973) has studied blood flow changes in the splancnic area of hyperthermic sheep. Hyperthermia at mean temperatures of 40.2°, 40.7° and 42.3°C resulted in mean decreases in splancnic flow of 20%, 42% and 53% respectively. In this study there were no marked changes in cardiac output.

In experiments in anaesthetised cats and rabbits, Walther et al. (1970) have demonstrated that local heating of the spinal cord causes decrease in activity in cutaneous sympathetic effects with corresponding increase in viseral sympathetic outflow. Similar effects were later demonstrated in decereberated rabbits by Iriki and Kozawa (1976a). These results would indicate that thermoregulatory responses of the sympathetic nervous system are capable of functioning after loss of hypothalamic integration. In addition it has been shown that vagal activity is decreased during spinal chord warming (Iriki and Kozawa 1976b).

Local heating of the spinal cord has also been shown to decrease blood flow the in superior mesenteric and splenic arteries of anaesthetised dogs, with concomitant increase in flow in the dorsalis pedis artery (Hales and

Dampney 1975).

From the above discussion, it may be concluded that the thermoregulation responses of the splancnic circulation in response to hyperthermia is basically similar in all mammalian species. Cardio-cutaneous responses to heating may vary but it would appear that splancnic blood flow is almost always reduced under these conditiones. It is tempting to suggest that similar reductions in total splancnic hepatic flow and oxygen consumption to those found in pigs in the present study, may be occuring in man undergoing WBHT and that these physiological responses might be,in part, responsible for the hepatic toxicity that was observed following treatment.

It would appear that estimated hepatic blood flows obtained under hyperthermic conditions employing dye clearance methods are unreliable and there is a need for the development of alternative clinical methods directed towards this end.

Future Possibilities

If the assumption is accepted that changes in liver function following WBHT are, at least partially, caused by decreases in total splanchnic blood flow during treatment, it would seem logical to attempt to block the increase of sympathetic discharge that is taking place. This has been demonstrated in experimental models (as mentioned above) by Walther et al. (1970) and Iriki and Kasawa (1976a).

If spinal epidural blockade with local anaesthetics is performed sympathetic discharge from the thoraco-lumbar outflow will be decreased. Epidural anaesthesia has been employed by both Pettigrew and Ludgate (1977), and Blair and Levin (1977) in order to increase cutaneous vasodilation and hence by, increasing heat flux from the skin surface, to speed the warming of

the patient. The latter have demonstrated a mean rise of body temperature during the first hour of heating of 1.7°C (in three treatments) without epidural blockade, as opposed to a rise of 3.1°C per hour (in 32 treatments) after blockade. Unfortunately the temperatures at which the patients were held at plateau and also the duration of treatments vary between the two groups and no conclusions can be drawn as to the prevention of rises of serum enzymes or bilirubin levels following treatment.

Alpha-adrenergic blockade of the sympathetic nervous system might also be expected to decrease sympathetic tone in the splanchnic vascular bed. Hence less decrease of blood flow to the liver should occur during WBHT. This treatment might, indeed, have other advantages than the increase of splanchnic blood flow.

It has been shown that platelets can take up circulating catecholamines from plasma and that this effect is both time and temperature dependent (Born and Smith 1970) and it has recently been confirmed by Zweifler and Julius (1982) that patients with pheochromocytoma have greatly increased platelet levels of catecholamines. Zwiefler and Romero (1975) have shown that platelets from patients with pheochromoacytoma exhibit greater sensitivity to adenosine diphosphate (ADP) aggregation than do those of normal controls. They have further demonstrated that this effect can be prevented by the alpha-adrenergic blocking agent phentolamine.

As already mentioned, Kim et al. (1979) have confirmed the presence of increased concentrations of circulating catecholamines in patients undergoing WBHT. In some cases the concentrations, that they measured, were comparable with those found in patients suffering from phaeochromocytoma. It might therefore be postulated that alpha-adrenergic blockade during WBHT might prevent catecholamine release. Uptake of these substances by the platelets, which may take place at an faster than normal rate at raised temperatures, would then not occur. In this way the occurance of disseminated intravascular coagulation

following WBHT might be diminished or prevented.

SUMMARY OF THE EFFECTS OF HYPERTHERMIA ON THE LIVER

Rises in serum enzymes, indicating both muscle damage and hepatic impairment have been seen following whole body hyperthermia treatment. The most useful enzymes to follow as indicators of liver damage would appear to be SGOT and SGPT, which tend to reach their highest levels at 48 hours folowing treatment.

Evidence from published work is available to indicate that a body temperature of 41.8°C should, at the moment, be considered an upper safe limit of treatment temperature. However one patient in the present series died of hepatic necrosis, a case report of which is reported in this thesis. It would appear that total splancnic blood flow is significantly reduced under hyperthermic conditions and that indocyanine green clearance techniques are unsuitable for estimating this reduction. Diminution of flow is almost certainly mainly mediated through sympathetic vasoconstriction of the splancnic bed. It would seem logical to try to block this effect by alpha blockade of the autonomic nervous system.

Total splancnic oxygen consumption is reduced at hyperthermia. This is not as a result of decreased blood flow, with consequent desaturation of hepatic venous blood , but as a result of altered hepatic cellular metabolism at high temperatures.

SUMMARY AND CONCLUSIONS

In Chapter I, the thermal sensitivity of normal tissues in the intact animal were discussed and it was pointed out the intact animal was more sensitive to temperature rise than some of its individual organ systems. The concept of critical thermal maximum was introduced and a discussion was presented on the relationship between hyperthermia and the duration of maintainence of the raised temperatures.

In Chapter II a short review of cellular thermobiology was presented. It was concluded that malignant cells are probably no more sensitive than normal cells from the same tissue of origin. Thermosensitivity of cells at various stages of the cell cycle under different nutritional conditions was discussed. It was concluded that though hypoxia per se had little effect on thermosensitivity, the pH of the incubation medium was of great importance. Low pH conditions, both during and following hyperthermia, can greatly increase cellular thermosensitivity and can markedly increase cell mortality. A short discussion of thermal resistance and thermotolerance was presented together with evidence that thermotolerance development is an active cellular 'defence' mechanism. Mention was made of the effects of 'step-down heating' and of studies of substances that may protect or sensitise cells to hyperthermia.

The discussion of cellular thermobiology was further expanded in Chapter II to include combined cellular effects of hyperthermia and radiation. Again the effect of low pH is of importance in cell mortality. A short discussion of the timing of hyperthermia and radiation was presented and the chapter was concluded with a few remarks on combined treatment with hyperthermia and cytotoxic agents.

In chapter III the thermobiology of tumors was considered. Studies have

revealed that microcirculatory blood flow of malignant tumors is essentially inhomogenious. Evidence is presented that flow can be greatly decreased or even completely obstructed under conditions of hyperthermia. Tumors have been found to be generally hypoxic and at a lower pH than surrounding normal tissues. Under conditions of hyperthermia, the pH is further reduced and oxygenation is impaired to a greater extent than under normothermia. Glucose uptake and metabolism was discussed, as were the possible mechanisms whereby tumor circulation is compromised under hyperthermia.

In Chapter IV a discussion of the various heating techniques in use for Whole Body Hyperthermia treatment of patients was presented. These encompass skin heating with hot wax, water and hot air. The use of extracorporeal heat exchangers was mentioned. A detailed description of the apparatus in use in Rotterdam for WBHT was presented together with an outline of the development and modification of the heating methods employed.

A detailed description of anaesthetic techniques in use in the present study was presented in Chapter V. Also included was a description of the pretreatment fitness criteria for inclusion of patients in the study, and a description of monitoring procedures during treatment. Fluid balance under WBHT was considered. The most important conclusions were that a very light general anaesthetic should be administered and that care should be taken not to attempt to alter haemodynamic parameters by administering large quantities of colloidal solutions as this may lead to oedema formation. It was further noted that urine output may be greatly diminished under WBHT.

In Chapter VI, results were presented of studies of the reaction of the cardiovascular system to WBH. This treatment is associated with large decreases in both systemic and pulmonary vascular resistance. Whereas the systemic pressure is reduced at the plateau temperature of 41.8°C, pressure in the pulmonary artery is increased. A considerable tachycardia occured, but did not cause undue problems. Though cardiac output was greatly

increased, left ventricular work was not raised to levels markedly more than those obtaining in the awake sedated patients. On the other hand, right ventricular work was greatly increased. The mechanisms responsible for this paradox were discussed. The pattern of 'normalization' of cardiovascular variables in the post treatment period was also presented and discussed.

In Chapter VII details were presented of whole body oxygen consumption and of the oxygen flux to the tissues under conditions of hyperthermia. Factors affecting the oxygen supply were discussed and it was concluded that there was no evidence of the occurrence of tissue hypoxia. Increases of oxygen consumption were modest, (34.6% increase over normothermic anaesthetized controls) and it was shown that 37% of these increases could be accounted for by increases in cardiac work.

In Chapter VIII, a number of side effects and complications that occurred following WBHT were described and discussed. These included haemotological and electrolyte changes. The occurrence of a mild intravascular coagulation syndrome in a number of patients was discussed. The complications occurring did not cause undue anxiety.

In Chapter IX the, sometimes severe, changes in serum enzyme levels were discussed and it was postulated that these were caused by a mixed post hyperthermic syndrome. Skeletal muscular damage was reflected by raised CPK levels, whereas rises of SGOT and SGPT levels (which tend to be most severe following a patient's first hypethermic exposure) were indications of liver damage. A case report was presented of a patient who died in hepatic failure following hyperthermia treatment. The etiology of this tragedy and the possible hepatoxicity of the halothane anaesthesia used in this case were extensively discussed. No firm conclusions could be drawn as to the cause of this fatality.

Results of estimations of total splanchnic blood flow under hyperthermia in patients and in experimental animals (pigs) were presented. It was

concluded that the technique of indocyanine green clearance gave unreliable results under these conditions. Further studies in pigs of splanchnic blood flow and oxygen consumption were presented and it was seen that both values were significantly decreased above a body temperature of 39°C. The cause and possible prevention of these changes was discussed.

On the basis of the data obtained from patients and experimental animals it was concluded that the liver was the limiting organ with respect to the safety of Whole Body Hyperthermia Treatment. Further methods to closely monitor liver function during any hyperthermia treatment involving either WBHT or large volume local heating must be refined.

As a final conclusion, it may be stated that this thesis has demonstrated that Whole Body Hyperthermia Treatment under general anaesthesia is entirely feasible. Provided that certain precautions and adequate monitoring regimes are observed, morbidity can be kept to an acceptable minimun. Once the problems of liver toxicity can be overcome, it may be possible to raise treatment temperatures and provide vastly improved clinical results from this form of treatment.

GLOSSARY OF ABBREVIATIONS

AP	Mean Systemic Arterial Pressure
C14	A radioisotope of Carbon
ChvO2	Content of oxygen in hepatic venous blood
CI	Cardiac Index
CO	Cardiac Output
CPK	Creatine Phosphokinase
Cr51	A radioisotope of Chronium
CTM	Critical Thermal Maximum
Cvo2	Content of oxygen in the mixed venous blood
D	Optical Density
D2O	Deuterated water
DNA	Desoxyribo Nucleic Acid
2,3,DPG	2,3, Diphosphoglycerate
E	Optical Extinction
EHBF	Estimated Hepatic Blood Flow
Hb	Haemoglobin concentration in the blood
Ht	Haematocrit
I131	A radioisotope of Iodine
ICG	Indocyanine Green
ICGa	Arterial Concentration of Indocyanine Green
ICGhv	Hepatic Venous Concentration of Indocyanine Green
Kw	Kilowatt
LDH	Lactic Dehydrogenase
LVWI	Left Ventricular Work Index
MHz	Megahertz
Na24	A radioisotope of Sodium
O2con	Whole body oxygen consumption per m2 of body surface area
OER	Oxygen Enhancement Ratio
p	Statistical probability of chance occurence
p50	Partial pressure of oxygen at which haemoglobin is 50% saturated
paO2	Partial pressure of oxygen in the arterial blood
PAP	Mean Pulmonary Artery Pressure
pCO2	Partial pressure of carbon dioxide
pH	Concentration of hydrogen ions in a solution
pO2	Partial pressure of oxygen

PVRI	Pulmonary Vascular Resistance Index
Rb86	A radioisotope of Rubidium
RO2	Ratio of whole body oxygen consumption and oxygen flux
RVWI	Right Ventricular Work Index
SaO2	Saturation of oxygen in the arterial blood
SBF	Splanchnic Blood Flow
SBF%	SBF expressed as a percentage of the cardiac output
SD	Standard Deviation
SEM	Standard Error of the Mean
SGOT	Serum Glutamate Oxoloacetate Transaminase
SGPT	Serum Glutamate Pyruvate Transaminase
ShvO2	Saturation of oxygen in hepatic venous blood
SO2con	Splanchnic Oxygen Consumption
SvO2	Saturation of oxygen in mixed venous blood
SVRI	Systemic Vascular Resistance Index
VO2	Oxygen consumption
WBHT	Whole Body Hyperthermia Treatment
Xe131	A radioisotope of Xenon

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