We are IntechOpen, the world's leading publisher of Open Access books Built by scientists, for scientists

6,900
Open access books available

185,000
International authors and editors

200M
Downloads

Our authors are among the

154
Countries delivered to

TOP 1%
most cited scientists

12.2%
Contributors from top 500 universities



WEB OF SCIENCE™

Selection of our books indexed in the Book Citation Index in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?
Contact book.department@intechopen.com

Numbers displayed above are based on latest data collected.
For more information visit www.intechopen.com



Chapter 12

Hyperthermia: Cancer Treatment and Beyond

Ahmed Bettaieb, Paulina K. Wrzal and
Diana A. Averill-Bates

Additional information is available at the end of the chapter

http://dx.doi.org/10.5772/55795

1. Introduction

The three mainstays for cancer treatment include surgical removal of tumors, radiation therapy and chemotherapy, which have led to improved patient survival for certain types of cancer, but there is still much room for improvement. Cancer is one of the leading causes of death worldwide and accounted for 7.6 million deaths (13% of all deaths) in 2008 (World Health Organization, 2012). The 2012 Report to the Nation on the Status of Cancer indicated that there was a decrease in overall cancer mortality and incidence in the U.S.A. from 1999 to 2008, particularly for the four major cancer sites: lung, colorectum, breast and prostate [1]. However, there were increases in the incidence of other types of cancer, including those of the pancreas, kidney, thyroid and liver, as well as melanoma and adenocarcinoma of the esophagus, from 1999 to 2008.

Over the past decades, the struggle against cancer has led to the discovery of new strategies to fight this disease and to bring hope to patients. These new strategies include hyperthermia (also commonly known as thermal therapy or thermotherapy), biological therapies (e.g. immunotherapy), photodynamic therapy, laser treatment, gene therapy, and inhibitors of angiogenesis. Most of these strategies still need optimization, and in some cases (e.g. hyperthermia, photodynamic therapy), improved equipment is required. Moreover, a better understanding of the biological mechanisms involved in their anticancer action would certainly be beneficial. Hyperthermia is one of the few strategies to be adopted as a promising therapy among the alternative methods to treat cancer.

Hyperthermia is defined as moderate elevation in temperature. Hyperthermia can either have a pathological origin, resulting from the fever response of the organism to viral or bacterial infections, or may occur during exposure to high temperatures as during heat stroke. It is relatively recent as a clinical procedure, in which body tissues are exposed to elevated

INTECH
open science | open minds

© 2013 Bettaieb et al.; licensee InTech. This is an open access article distributed under the terms of the Creative Commons Attribution License (http://creativecommons.org/licenses/by/3.0), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

temperatures in the range of 39°C to 45°C. These high temperatures can damage and kill cancer cells with minimal injury to normal tissues [2]. During the last two decades, hyperthermia has been used as an efficient complement to standard cancer treatments such as radiation therapy and chemotherapy [2,3] (Figure 1). A further advantage is that hyperthermia can eliminate drug-resistant and radio-resistant tumour cells. Another form of hyperthermia involves very high temperatures (> 60°C), which can destroy or «cook» tumours by a technique known as thermal ablation (see review, [4]). The present review will address the therapeutic potential of moderate hyperthermia (39°C to 45°C).

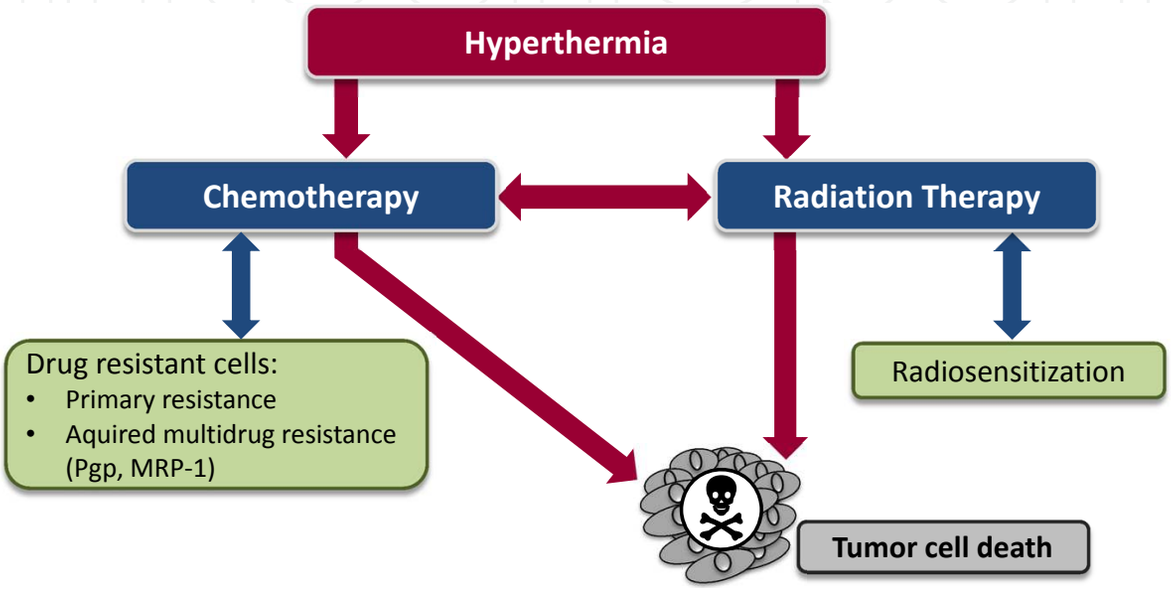


Figure 1. Hyperthermia complements standard cancer treatments such as chemotherapy and radiation therapy in destroying tumour cells.

2. Hyperthermia

2.1. Scientific history

The use of heat to treat disease, including cancer, is a concept that dates back to early Egyptian times, over 5000 years ago (see review, [5]). Indeed, the Egyptian medical papyrus recounts an attempt to treat breast cancer with a "heated stick" [6]. Likewise, many Greek doctors, among them Hippocrates, suggested cauterizing superficial tumours by using heated metal. Many ancient cultures, including the Roman, Chinese, Indian and Japanese cultures have used this concept for the treatment of a variety of diseases. During the late 1800s, there were numerous observations by astute clinicians of spontaneous remissions of cancer in patients suffering from a variety of infections [7]. Dr. William B. Coley found 47 case reports in which simultaneous infection seemed to have caused the remission of an incurable neoplastic malignancy (see review, [8]). In the late 1800s, he used “Coley’s Mixed Toxins” (bacterial pyrogenic toxins) as a deliberate fever-inducing treatment to control tumor growth [9]. Despite promising obser-

vations during several decades, these cancer treatments were difficult to administer in a controlled manner, and responses were unpredictable [10]. Using a different approach, Westermark reported the use of localized, non-fever producing heat treatments (42-44°C) by means of water-circulating cisterns that resulted in the long-term remission of inoperable cancer of the cervix [11]. As different techniques were developed, such as surgery, radiation therapy and chemotherapy, further development of hyperthermia for cancer treatment was put on the back burner. There was a resurgence of interest in the use of hyperthermia in cancer treatment based on scientific studies initiated in the 1960s and 1970s. A turning point was a study conducted in transplanted mouse tumors that illustrated novel biological phenomena: cytotoxicity of hyperthermia was dependent on time and temperature; increased sensitivity of large versus small tumors to hyperthermia (later attributed to vascular events); heat-induced thermotolerance of normal and tumor tissue; and hyperthermia-induced sensitization to radiation [12]. These promising observations led to quantitative experimental studies and a rapid increase in our understanding of the biological effects of hyperthermia. Furthermore, they frame the rationale for the clinical use of hyperthermia, and the development of more effective technologies for the precise application of heat to tumors and for the measurement of heat distribution in tumors by thermometry.

2.2. Cellular changes

Temperatures in the range of moderate hyperthermia can be non-lethal (39 to 42°C) or lethal (>42°C). Temperatures above 42°C were shown to kill cancer cells in a time- and temperature-dependent manner that was measured by the clonogenic cell survival assay [13]. However, despite numerous studies during at least three decades, which have improved our understanding of hyperthermia biology, the mechanisms involved in heat-induced cytotoxicity are still ill-defined [14]. Hyperthermia causes many changes in cells and leads to a loss of cellular homeostasis [15-17]. A key event appears to be protein denaturation and aggregation, which results in cell cycle arrest, inactivation of protein synthesis, and inhibition of DNA repair processes [18]. Other cellular effects of hyperthermia include: (1) the inhibition of DNA synthesis, transcription, RNA processing and translation; (2) increased degradation of aggregated/misfolded proteins through the proteasomal and lysosomal pathways; (3) disruption of the membrane cytoskeleton; (4) metabolic changes (e.g. uncoupling of oxidative phosphorylation) that lead to decreased levels of ATP; and (5) alterations in membrane permeability that cause increases in intracellular levels of Na^+, H^+ and Ca^{2+} (see reviews, [19,20].

Hyperthermia can cause changes in lipids but these appear to be reversible [21]. The viscosity of the plasma membrane decreases with increasing temperature [22], and this may be associated with altered transport functions of the membrane. Changes in membrane viscosity were linked to an elevation in the activity of the ATP-dependent sodium-potassium pump [22], which maintains Na^+ and K^+ levels across the plasma membrane against a concentration gradient. During hyperthermia, membrane permeability towards several compounds is altered, including polyamines, glucose, and anticancer drugs [23-25].

Despite the large number of documented cellular changes, the nature of the critical lesions that lead to cell death following heat treatment remains unknown. Proteins appear to be the first

target of hyperthermia in the clinically-relevant temperature range of 39 to 45°C (Figure 2). The alteration of cellular homeostasis after exposure to hyperthermia entails a certain number of post-translational modifications such as glycosylation, acylation, phosphorylation, farnelysation and ubiquitination [18,26]. Several studies reported that hyperthermia can cause DNA fragmentation and the formation of double strand breaks (DBSs) [27,28], which could arise from the inhibition of DNA repair mechanisms [21]. However, it appears that nuclear protein damage may be the key factor rather than direct DNA damage itself. Nuclear proteins, in particular, appear to be very sensitive to hyperthermia and undergo aggregation [21]. Nuclear protein aggregation has been linked to the inhibition of transcription and DNA replication.

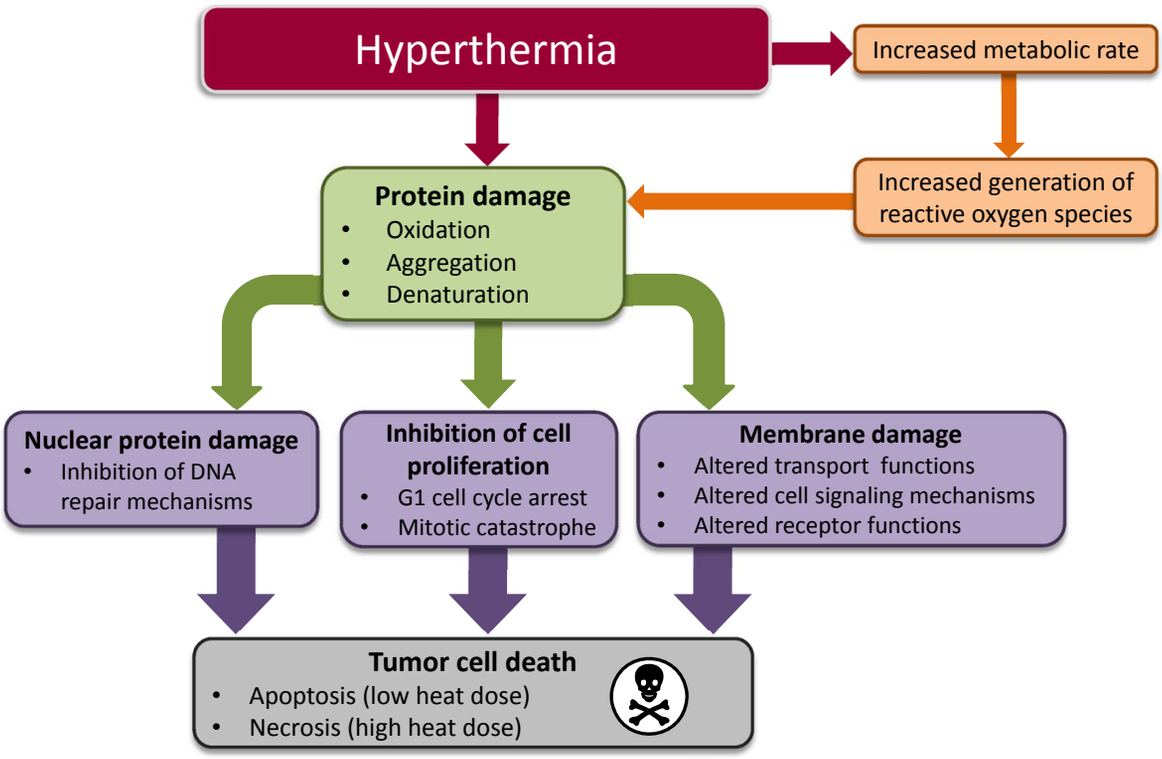


Figure 2. Hyperthermia-induced cellular changes that could lead to tumour cell death.

Elevated temperatures can increase the rates of biochemical reactions and this would increase cell metabolism, which should cause increased oxidative stress (Figure 2). Levels of reactive oxygen species (ROS) were shown to increase after exposure to both lethal (≥42°C) [29-31] and non-lethal (40°C) temperatures [32,33]. This would arise principally from the increased generation of ROS such as superoxide and hydrogen peroxide (H_2O_2), likely as a result of dysfunction of the mitochondrial respiratory chain. Other potential sources of increased ROS generation would be increased activity of superoxide-producing enzymes such as NADPH oxidase and xanthine oxidase at elevated temperatures. Hyperthermia could also increase the reactivity of these ROS; indeed, the cytotoxicity of hydrogen peroxide was increased at elevated temperatures (41 to 43°C) compared to the physiological temperature (37°C) [34]. Hyperthermia also inactivated cellular antioxidant defenses against H_2O_2 such as the pentose

phosphate pathway [35], which maintains the intracellular antioxidant glutathione in its reduced form, GSH [36]. An increase in the generation of ROS can cause oxidative damage to proteins, lipids and nucleic acids. A hyperthermia-induced decrease in tumor growth was accompanied by an increase in lipid peroxidation in rabbits [37]. Another consequence of increased ROS generation by hyperthermia is that molecules such as H_2O_2 can perturb mitochondrial membrane potential [38]. A temperature-induced increase in cell metabolism could also cause acidosis of the tumor tissue [39,40].

2.3. Cytotoxicity of hyperthermia

As a consequence of different cellular changes, hyperthermia causes mitotic catastrophe, permanent G1 arrest and a loss of clonogenic or reproductive cell capacity [21] (Figure 2). Cells can die by processes such as apoptosis and/or necrosis, which are dependent on the cell type as well as the temperature and duration of heat exposure [32,41]. Another consequence is that cells can become sensitized to other cytotoxic modalities such as radiation [16]. Hyperthermia was reported to cause centrosomal dysfunction and mitotic catastrophe [42], which have been implicated in thermal radio-sensitization [43]. Hyperthermia (42 to 44°C) has been reported to cause chromatin condensation and apoptotic DNA fragmentation (formation of DNA ladders) leading to apoptosis in many different cell types including HeLa cells [44], T lymphocytes [45,46], HL-60 leukemic cells [47], and mice embryonic fibroblasts [48]. In rats treated with whole body hyperthermia (41.5°C for 2 h), both the extent and kinetics of hyperthermia-induced apoptosis differed between two different tumor types (fibrosarcoma and colon carcinoma) [49]. Additionally, the same study revealed another important advantage; the induction of apoptosis was higher in tumor tissues in comparison to normal tissues. Most of the studies that have investigated the mechanisms of heat shock-induced cytotoxicity concluded that apoptosis is the main form of cell death and proposed the pro-apoptotic effects of hyperthermia as the potential desired outcome of hyperthermia in cancer therapy.

2.4. Hyperthermia and physiological changes

Several physiological factors including oxygenation, pH and blood flow were shown to play a role in the sensitivity of cells/tissues to moderate hyperthermia. The intrinsic sensitivity to heat varies significantly among different cell types. Several studies indicate that cancer cells are more susceptible to heat injury than normal cells [21,50]. This could be caused, at least in part, by the differential expression of heat shock proteins (Hsps) and other proteins involved in the cellular defense system against different stressors, including heat shock. However, there is no consistency in findings about heat sensitivity between tumor and normal cells [21]. The sensitivity of cells to heat also varies with phase of the cell cycle, where cells in S phase and mitosis were reported to be most sensitive [51].

Another reason for the use of hyperthermia in cancer treatment is the fact that tumor tissues are poorly vascularized in comparison to normal tissues. This may lead to a differential heating, with higher temperatures being achieved in tumors compared with normal tissue, where heat may be dissipated by circulating blood. Hyperthermia also appears to be complementary to other forms of treatment by being able to destroy tumor

cells that are relatively resistant to radiation therapy or chemotherapy. Tumor cells located in the hypoxic centers of tumors are relatively resistant to chemotherapy due to poor drug delivery. Several chemotherapeutic drugs also require oxygen to generate free radicals in order to cause tumor cytotoxicity. Further, most chemotherapeutic drugs are more effective against proliferating cells. However, hypoxia has been shown to cause decreased proliferation, which may partially explain the reason for resistance of tumor cells to chemotherapy [52-54]. Cells located in hypoxic areas of tumors are also resistant to radiation therapy.

Heating of human tumours is heterogeneous. Some areas of the tumour reach cytotoxic temperatures such as 43 to 45°C, whereas other areas only reach 39 to 42°C. It is more difficult to heat larger or deep-seated tumours to cytotoxic temperatures that are adequate to cause cell death or vascular damage.

Tumors are unable to adapt their blood circulation to the effects of high temperatures (≥42°C), which enables hyperthermia to cut off the supply of nutrients and oxygen, leading to lower interstitial pH and a collapse in tumor vasculature [55]. These conditions render cells more susceptible to heat treatment. Indeed, cells at lower (acidic) pH and decreased oxygen tension, as in the center of tumors, are more sensitive to heat treatment [56,57]. Cells in a nutrient-deprived environment are also more sensitive to elevated temperatures. This effect appears to correlate with changes in cellular ATP levels [58]. Cells that were deprived of glucose exhibited increased sensitivity to the cytotoxicity of hyperthermia [35]. This effect could be linked to a decrease in antioxidant defenses involving the glutathione redox cycle, since glucose metabolism, through the pentose phosphate pathway, is required for maintaining intracellular levels of GSH. On the other hand, heating at milder temperatures (e.g. 39° to 42°C) can increase tumor blood flow, which leads to improved tumor oxygenation [59,60]. This could render tumors more sensitive to radiation and certain anticancer drugs.

Hyperthermia (≥42°C) has been shown to cause vascular damage in rodent tumours, which leads to decreased oxygenation and necrosis [61]. Although, the vasculature of human tumours appears to be more resistant to hyperthermia than that of rodent tissues, hyperthermia has been shown to cause disturbances in the microcirculation of cancer tissue in human osteosarcoma [62].

Milder temperatures in the range of 40 to 41°C appear to be able to stimulate various elements of the immune system, thus increasing immune surveillance and protecting against tumor growth (see reviews, [63-65]). The exposure of immune effector cells (e.g. macrophages, T cells, and natural killer (NK) cells) to mild temperatures has been shown to: (1) enhance the migration of immune cells to target sites, which could allow better control of infection and tumor burden; (2) increase the expression of cell surface molecules (e.g. involved in antigen presentation); (3) increase the release of soluble factors involved in immune effector cell activity (e.g. pro-inflammatory cytokines including tumor necrosis factor-alpha (TNF-α), interleukin-1 (IL-1), IL-6, IL-10, and IL-12; (4) regulate immune cell proliferation; and (5) increase the cytotoxicity of immune cells against target (tumor) cells.

2.5. Thermotolerance: The other side of hyperthermia

The exposure of cells to lethal temperatures such as 43 to 45°C during short periods of time, ranging from 10 to 30 minutes, allows the development of tolerance towards subsequent exposure to multiple stresses; this phenomenon is termed "thermotolerance" [66,67]. Thermotolerance is an adaptive survival response induced by heat preconditioning whereby cells become resistant to a subsequent lethal insult such as that triggered by heat shock, reactive oxygen species (ROS), and environmental stressors including heavy metals [68,69]. If the level of stress is very low, cells attempt to survive by activating stress responses that protect essential biochemical processes such as DNA repair, protein folding, and the elimination of damaged proteins [70]. Once the stress stimulus is removed, cells can recover their normal cellular function. If the stress continues or is too severe, then the cell will likely die by apoptosis or necrosis.

The acquisition of thermotolerance is characterized by numerous biochemical and molecular changes. Thermotolerance is generally associated with the accumulation of Hsps [19,63,68,71-73]. Hsp expression is regulated by a stress-responsive transcription factor known as heat shock factor 1 (HSF-1), through its interaction with the heat shock element (HSE) [74]. In addition, changes in the expression of about 50 to 200 other genes, not traditionally considered Hsps, have been found during or after heat stress (see review, [20]). These include genes for transcription factors, protein degradation, DNA repair enzymes, metabolic enzymes, cell cycle arrest, transport and detoxification, and signal transduction. The reason for the induction of these other cell-protective pathways by heat shock is probably to protect nascent chain synthesis and folding, prevent protein misfolding and aggregation, and to promote recovery from stress-induced damage [75]. Proteomic analyses showed a change in the phosphorylation of 93 proteins between control RIF-1 and their thermotolerant derivatives, TR-RIF-1 cells [76]. These phosphorylated proteins are responsible for a range of cellular functions, which include chaperones, ion channels, signal transduction, transcription and translation, biosynthesis of amino acids, oxidoreduction, energy metabolism, and cell motility or structure.

The heat shock response is highly conserved in all organisms from yeast to humans, which suggests that it is important for survival in a stressful environment [74]. In addition to heat, the heat shock response can be induced by other insults such as oxidative stress, heavy metals, ethanol, toxins and bacterial infections.

The major classes of Hsps induced by the heat shock response are Hsp90, Hsp70, Hsp60, and Hsp27. Hsps appear to play an important role in thermotolerance. Many studies suggest a correlation between the accumulation of Hsp70 and the acquisition of a thermotolerant state in mammals, amphibians, insects [77-79] and fish [80]. Under conditions of stress, Hsp70s can prevent the formation of protein aggregates and assist the refolding of aggregated proteins into their native structures [19]. Other studies have shown that the state of thermotolerance correlated with an increase in the expression of Hsp110 [81]. Hsp110 is as effective as Hsp70 in preventing protein aggregation, and contributes, along with Hsp70 and Hsp40, to the refolding of denatured proteins. In addition to their protective role against a subsequent lethal

heat shock, Hsps are known to protect cells against other forms of stress, such as oxidative stress and radiation [82].

Hsps play an important and yet complex role in the regulation of apoptosis. The specific roles of different Hsps such as Hsp27, Hsp60, Hsp70 and Hsp90 in the regulation of the mitochondrial and death receptor pathways of apoptosis have been reviewed [82-85]. The induction of apoptosis through the Fas death receptor can be regulated by Hsp70 and Hsp27 [86,87]. Hsp27 and Hsp70 can regulate the death receptor pathway of apoptosis by preventing t-Bid translocation to mitochondria, which in turn inhibits cytochrome c release [88, 89]. Hsp90 was shown to be a negative regulator of caspase-2 activation [90]. Hsp27, Hsp70, and Hsp90 can attenuate apoptosis upstream of mitochondria [91], as well as interfering with apoptosome formation, post-mitochondrial events, and caspase activation [92]. Furthermore, Hsp70 and phosphorylated Hsp27 can protect cells against oxidative stress, a potent activator of apoptosis [93,94].

The development of thermotolerance by lethal hyperthermia has been the subject of intensive studies during the past three decades, whereas thermotolerance induced at mild, fever-range temperatures has received relatively little attention. Thermotolerance can be developed following exposure for shorter times (e.g. 30 min) to lethal temperatures (42 to 45°C) [68,71], or during continuous heating (e.g. 3 to 24 h) at non-lethal temperatures (39 to 41.5°C) [95,96]. The development of thermotolerance by exposure of cells to mild hyperthermia (40°C) for 3 to 24 h led to the accumulation of Hsps 27, 32, 60, 70, 90 and 110 [32,95]. This phenomenon is of notable importance for fundamental research given that it is a physiological fever-range temperature and suggests that thermotolerance could protect healthy tissue against stressors during clinical therapies. The treatment of BALB/c mice *in vivo* with fever-range whole body hyperthermia (39.5 to 40°C) for 6h led to increased expression of Hsp70 and Hsp110 in several mouse tissues [97].

Mild thermotolerance developed at 40°C created an apoptosis-resistant phenotype. The activation of the mitochondrial pathway of apoptosis by moderate hyperthermia (42 to 43°C) was attenuated in these thermotolerant cells [44]. Similarly, activation of the death receptor signaling pathway through the Fas receptor by lethal heat shock (42 to 43°C) was inhibited in thermotolerant cells [32]. Furthermore, thermotolerance developed at 40°C protected cells against the induction of apoptosis by oxidative stress (H_2O_2), mediated through the mitochondrial and death receptor pathways [33,38]. This apoptosis-resistant phenotype could be conferred by increased levels of both Hsps (Hsps 27, 32, 60, 70, 90, and 110 kDa) and antioxidants (catalase, manganese superoxide dismutase, glutathione) [32,33]. Mild thermotolerance also inhibited hyperthermia-induced ROS generation [32], and this could be explained by the ROS-inhibitory effect of Hsps such as Hsp27 and Hsp70 [93,94].

Hsps play overlapping roles in tumour development and growth by promoting cell proliferation and by inhibiting cell death pathways [98]. Hsp70 is a survival protein that is overexpressed in various malignant tumors and its expression correlates with increased cell proliferation, poor differentiation and poor therapeutic outcome in human breast cancer [99]. The increased expression of Hsp70 in tumors can prevent the activation of caspases and proteases, and thus abolish apoptotic cell death [98]. Moreover, the increased expression of Hsps appears to be involved in the acquisition of drug-resistant phenotypes. Several studies

have reported that Hsp27 may be involved in the development of resistance to chemotherapeutic agents such as doxorubicin and cisplatin [100-104].

2.6. Hyperthermia in cancer therapy

The biological rational for the use of hyperthermia in cancer treatment is very strong. Temperatures of 42.5°C and above are able to kill cancer cells. Findings from *in vitro* studies generally indicate that there is no intrinsic difference in heat sensitivity between normal and tumour cells [105]. However, a tumour selective effect of hyperthermia could occur at higher temperatures *in vivo*. In solid tumours, the vascular system is chaotic, which results in regions with hypoxia and low pH levels, compared to normal tissues. These conditions render cells more sensitive to the cytotoxic effects of hyperthermia. Therefore, hyperthermia can be beneficial by causing direct cytotoxicity to tumour cells, in addition to selective destruction of tumour cells in hypoxic and low pH environments within solid tumours. A further benefit is that mild hyperthermia can activate certain responses of the immune system, which could also provide protection against tumour growth [64,106]. In the clinic, hyperthermia has been shown to be most beneficial when used in combination with radiation therapy and/or chemotherapy.

2.6.1. Hyperthermia in combination with radiotherapy

One of the most promising aspects of hyperthermia in cancer treatment is the ability to eliminate radiation-resistant tumour cells [see review, 5]. Indeed, this renders hyperthermia as one of the most effective radiation sensitizers known. The basis for this effect is that hyperthermia has the ability to kill cells that are under conditions of hypoxia, low pH and that are in the S-phase of cell division, which are all conditions that render cells resistant to radiation. The mechanisms responsible for heat-induced radio-sensitization are not entirely understood, particularly for milder temperatures [21]. For temperatures of 43°C and above, nuclear protein damage is considered to be a critical event [107]. It was suggested that hyperthermia interferes with the repair of radiation-induced DNA damage. In support of this idea, hyperthermia increased the amount of radiation-induced chromosomal aberrations [13, 108]. It was suggested that heat-induced enhancement of chromosomal aberrations could arise from the inhibition of repair of radiation-induced DNA damage. Hyperthermia could exert its major effect on radio-sensitization by specifically inhibiting base excision repair of DNA damage [109,110].

2.6.2. Hyperthermia in combination with chemotherapy

The combined use of regional hyperthermia with systemic chemotherapy has considerable potential in cancer treatment mainly because localized heat delivery could enhance cytotoxic activity of anticancer drugs within a defined target region. This may lead to an improved therapeutic ratio by allowing targeting of chemotherapy, as can be achieved with radiation therapy. At present, targeted treatment with anticancer drugs can only be accomplished when they are administered either topically or intra-arterially. There is also evidence to suggest that the cytotoxic effects of hyperthermia and anticancer drugs may prove to be complementary. Tumour cells that are located in less well-vascularized regions of a tumour, such as the tumour

center, may be relatively resistant to systemic chemotherapy because they are exposed to lower concentrations of drug. The benefit of hyperthermia is that it kills cells most efficiently in the low pH and hypoxic environment of the tumour core. Furthermore, the temperature achieved in poorly vascularized regions of the tumour may be higher because of less efficient cooling by circulating blood. Another potential benefit is that regional hyperthermia at 40–43°C causes an increase in tumour blood supply [111]. Blood flow and vascular permeability, which are increased by hyperthermia, are critical factors for drug uptake [112].

Laboratory and *in vivo* studies have shown that the combined use of hyperthermia and chemotherapy leads to increased cytotoxic effects of several anticancer drugs such as cisplatin, anthracyclines, cyclophosphamide, ifosfamide, nitrosoureas, bleomycin, mitomycin, and nitrogen mustards such as melphalan [16,25,105,113-118]. Optimal heat enhancement of drug cytotoxicity generally occurs between 40.5°C and 43°C. For drugs such as cisplatin, alkylating agents, and nitrosoureas, interactions between heat and drug are more than additive (or synergistic), whereas in other cases, interactions are simply additive [119]. For bleomycin and Adriamycin, there is a threshold temperature of about 42.5°C to 43°C for enhancement of drug cytotoxicity. The antimetabolites (e.g. 5-fluorodeoxyuridin and methotrexate) and Vinca alkaloids or taxanes have independent interactions with hyperthermia. In general, the most effective heat-drug sequence is drug treatment immediately before heat delivery. The mechanisms of heat-induced enhancement of drug cytotoxicity are not well understood. Possible mechanisms include improved drug delivery to the tumour due to increased blood perfusion, increased intracellular uptake of drugs, and increased rates of reaction of drugs with cellular targets (e.g. increased drug alkylation, increased DNA damage).

2.6.2.1. Resistance to chemotherapeutic agents

One of the major limitations to the successful use of chemotherapy in cancer treatment is the development of resistance to multiple anticancer drugs. Cross-resistance occurs between different anticancer agents that have distinct structures and mechanisms of cytotoxicity. Multidrug resistance (MDR) is characterized by cross-resistance to four classes of commonly used anticancer drugs such as Vinca alkaloids, anthracyclines, taxanes, and epipodophyllotoxins. Classical MDR was discovered about 35 years ago and was initially related to the overexpression of the cellular 170-kDa protein P-glycoprotein (Pgp) [120], a member of the ATP-binding cassette (ABC) transporters. Pgp acts as an ATP-dependent transmembrane pump. Once anticancer drugs enter cells, they are immediately expulsed out of cells by Pgp. This results in decreased levels of drugs inside cells, rendering the drugs less effective against the tumour cells. In addition to Pgp, several other transporter proteins have been implicated in MDR in human cancer: multidrug resistance-associated protein 1 (MRP1), lung resistance protein (LRP) and breast cancer resistance protein (BCRP) [121]. MRP1 is a 190-kDa member of the ABC transporter family of proteins [122]. MRP1-mediated transport requires GSH, as well as ATP binding and hydrolysis. The overexpression of the protein MRP1 can cause cellular resistance to several anticancer drugs, including Adriamycin (doxorubicin), epipodophyllotoxins, and Vinca alkaloids such as vincristine, [123]. The substrate spectrum of MRP proteins also comprises amphiphilic anion conjugates of lipophilic compounds with glutathione (GSH),

glucuronate, or sulfate [124], as well as cysteinyl leukotriene (LTC4), prostaglandins, and the anticancer drug methotrexate [125].

Eventually, other distinct mechanisms were also implicated in the MDR phenotype [126]. These mechanisms engage other proteins involved in cellular defenses such as glutathione S-transferase (GST), an enzyme involved in the cellular detoxification of xenobiotics, which include certain anticancer drugs, toxins and environmental pollutants that undergo conjugation with the antioxidant GSH [127]. Other cellular defenses utilized by the MDR phenotype include metallothionein, thioredoxin, thymidylate synthase, dihydrofolate reductase, Hsps and topoisomerase II [126].

Clinical drug resistance appears to be a very complex and multifactorial problem [128] with multiple mechanisms involved. There is often overlapping substrate specificity between different drug transporters, and they are commonly co-expressed in many normal tissues and tumours. Overcoming MDR in cancer treatment presents a formidable challenge [129].

To date, three generations of inhibitors have been used to increase the efficacy of chemotherapy by inhibiting transporter-mediated drug efflux. However, the development of clinical inhibitors of ABC transporters as targets for clinical intervention in oncology has been difficult and new approaches are clearly needed. Clinical drug resistance is a major barrier which, if overcome, should lead to a significant improvement in patient survival.

2.6.2.2. Hyperthermia and reversal of resistance to chemotherapeutic agents

A beneficial effect of hyperthermia is its ability to reverse resistance to certain chemotherapeutic drugs [130]. Hyperthermia increased the cytotoxicity of anticancer drugs such as methotrexate [131], cisplatin [132], and mitomycin c [133] in cells exhibiting primary drug resistance. In addition, hyperthermia enhanced the cytotoxicity of melphalan in MDR Chinese hamster ovary CH^RC5 cells that overexpress Pgp [117]. CH^RC5 cells are resistant to anticancer drugs such as colchicine, Vinca alkaloids, Adriamycin, and melphalan [134].

Among the earlier strategies to overcome MDR, Pgp-modulating agents such as cyclosporin A and verapamil were developed. These chemosensitizers appear to act by decreasing Pgp-mediated efflux of anticancer drugs from cells, which allows increased accumulation of drugs to more cytotoxic levels inside cells. However, clinical studies showed that these chemosensitizers were effective only at toxic doses [128]. Therefore, chemosensitizers with improved MDR-reversing ability and lower toxicity need to be developed, as well as novel approaches. Hyperthermia (42 to 43°C) showed beneficial effects by reversing MDR involving Pgp when melphalan or Adriamycin was combined with Pgp modulators such as cyclosporin A [135,136]) or verapamil [137,138]. When combined with hyperthermia (43°C), the Pgp modulator PSC 833 reduced resistance to vinblastine in MDR K562 leukaemia cells and MESSA leiomyosarcoma cells [139]. Moreover, ultrasound-induced hyperthermia (USHT) increased Adriamycin cytotoxicity in the MDR human lung adenocarcinoma cell line MV522 [140]. The alkylating agent melphalan is mainly detoxified through conjugation with GSH, which can be catalyzed by GST [141]. In addition to overexpression of Pgp, CH^RC5 cells also overexpress the alpha and pi forms of GST, compared to the drug-sensitive AuxB1 cells [142]. Hyperthermia

was beneficial by enhancing melphalan cytotoxicity in MDR cells when GST was inhibited using ethacrynic acid [142].

2.6.2.3. Sensitivity of multidrug resistant cells to hyperthermia

Another important advantage for the clinical use of hyperthermia is that MDR cells overexpressing Pgp or MRP1 do not display cross-resistance to heat [25,143]. Indeed, these MDR cells exhibit equivalent sensitivity to the cytotoxic and apoptosis-inducing effects of hyperthermia (41-45°C) as their drug-sensitive counterparts. Moreover, drug-resistant sub-clones of human T-lineage acute lymphoblastic leukaemia (ALL) and acute myeloblastic leukaemia (AML) cells were as sensitive to hyperthermia as were the drug-sensitive sub-clones [144]. Results from these studies indicate that, in addition to enhancing drug cytotoxicity in resistant cells, hyperthermia alone can successfully eliminate MDR cells. Together, these findings clearly show that hyperthermia could be useful by destroying subpopulations of drug-resistant tumour cells, which have survived chemotherapy treatments, where the overexpression of Pgp and MRP1 is involved.

Apoptosis is considered to be a physiological mechanism for the elimination of damaged and abnormal cells, such as tumour cells. One of the hallmark characteristics of tumour cells is their ability to evade destruction by apoptosis [145]. The up-regulation of different anti-apoptotic proteins, to provide a survival advantage, has been a frequent explanation for the resistance of cancer cells to elimination by apoptosis [146]. The induction of death receptor and mitochondria-mediated signaling pathways of apoptosis by hyperthermia (41 to 43°C) in MDR $CH^{R}C5$ cells was compared to drug-sensitive CHO cells [147]. Differences were found between MDR and drug-sensitive cells in terms of induction of apoptosis by hyperthermia. For death receptor-mediated apoptosis, MDR cells contained higher levels of the anti-apoptosis protein c-FLIP and they had a lower level of activation of initiator caspase-8 and caspase-10 in response to hyperthermia. In the mitochondria-mediated pathway of heat-induced apoptosis, MDR cells showed higher mitochondrial levels of the pro-apoptosis proteins Bax and tBid, more pronounced mitochondrial membrane depolarization, and increased levels of the apoptosome protein Apaf-1 (apoptosis protease activating factor 1). The MDR cells appeared to show some resistance to death receptor-mediated apoptosis [147], in agreement with other studies in leukaemia cells [148, 149], but this resistance appeared to be compensated for by the pro-apoptosis changes in mitochondrial apoptosis. For the execution stage of apoptosis, the MDR and drug-sensitive cells showed similar levels of hyperthermia-induced caspase-3 activation, as well cleavage of caspase-3 substrates poly (ADP-ribose) polymerase (PARP) and inhibitor of caspase-activated DNase (ICAD) [147]. Similar levels of nuclear chromatin condensation were induced by hyperthermia, showing that overall, MDR cells are not resistant to hyperthermia-induced apoptosis compared to the drug-sensitive cells. In summary, MDR and drug-sensitive cells showed similar responses to heat in terms of clonogenic cell survival and apoptosis, which indicates that hyperthermia could be a promising strategy for eradicating MDR tumour cells in the cancer clinic.

2.7. Hyperthermia in the cancer clinic

2.7.1. Techniques to increase tumour temperatures

In the cancer clinic, hyperthermia is administered by exposing tumour tissues to conductive heat sources, or non-ionizing radiation (e.g. electromagnetic or ultrasonic fields). Hyperthermia can be applied by either invasive or noninvasive techniques, using externally applied power. To increase tumour temperatures, hyperthermia can be applied by several different techniques: local hyperthermia by external or internal energy sources, perfusion hyperthermia of organs, limbs, or body cavities, and whole body hyperthermia [150].

2.7.1.1. Local hyperthermia

Local hyperthermia entails elevating the temperature of superficial or deep-seated subcutaneous tumours while sparing the surrounding normal tissue, using external, intraluminal or interstitial heating modalities. The area can be heated externally with high-frequency waves (e.g. electromagnetic or ultrasound energy) aimed at the tumour from a device outside the body. To achieve internal heating, one of several types of sterile probes may be used, including thin heated wires, hollow tubes filled with warm water, implanted microwave antennae, radiofrequency electrodes and ultrasound. Local hyperthermia has allowed the use of hyperthermia in conjunction with other modalities of antineoplastic therapy. Local hyperthermia is more appropriate for the treatment of solid tumours, rather than blood diseases such as leukaemia. Despite advances in the technology of heating, the non-homogeneous character of the treatment region (i.e. tissue characteristics and blood flow) can often affect the uniformity of the heat dispersion in the treated area. This means that it can be difficult to obtain a uniform regional rise in the temperature that is reproducible [151-155]. Deep regional hyperthermia combined with chemotherapy, also known as hyperthermic intraperitoneal chemotherapy, is one of the promising methods for the treatment of prostate carcinoma [156,157] and bladder cancer [158].

2.7.1.2. Perfusion hyperthermia

This technique involves regional heating through the perfusion of a limb, organ (liver, pelvis, stomach), or body cavity using heated fluids [159-161]. In perfusion, the patient's blood can be removed, heated, and then pumped into the region that is to be heated internally. Perfusion hyperthermia can be applied with or without a cytotoxic drug. When applied to limbs without a cytotoxic agent, a temperature of about 43°C can be used for about two hours. Lower temperatures are used when perfusion is performed in combination with cytotoxic agents, to avoid drug toxicity.

2.7.1.3. Whole body hyperthermia

Externally-induced whole body hyperthermia can be used to treat metastatic cancers that have spread throughout the body. Whole body hyperthermia can be applied using different methods and involves heating the patient to a maximum temperature of 41.8 to 42°C. A newer

approach is to increase the temperature to about 40°C for a longer duration, and use a combination of mild hyperthermia with cytokines and/or cytotoxic drugs [118].

Many studies are focusing on improving the heating techniques. This is one of the main challenges that currently limit the clinical use of hyperthermia. Furthermore, improvements are required to heat effectively the deep-seated tumours that are localized in internal organs. The use of nanoparticles and the induction heating of magnetic materials that are implanted into tumors are among the new approaches that are currently being investigated for the improved application of hyperthermia.

2.7.2. Progress in the cancer clinic

In the cancer clinic, hyperthermia (40 to 44°C) is mainly used as an adjuvant to radiation and chemotherapy [2,5,16,150]. The major limitations of these conventional cancer treatments are lack of specificity and normal tissue toxicity. An important advantage of hyperthermia is that the cytotoxicity of radiotherapy and chemotherapy can be targeted to the tumour volume, thereby decreasing toxic side effects. The effectiveness of hyperthermia depends on the temperature rise and the duration of treatment at the elevated temperature. At least 19 randomized studies using a combination of hyperthermia with radiotherapy, chemotherapy or both, have shown significant improvement in clinical outcome in oncology patients, without a significant increase in side effects [150]. In all of these studies, the differences were very large. The combination of hyperthermia with radiation resulted in higher (complete) response rates, accompanied by improved local tumour control rates, better palliative effects, and/or better overall survival rates in many Phase II clinical trials [162-171]. These studies focused on many types of cancer including tumors of the head and neck, cervix, rectum, breast, brain, bladder, lung, esophagus, liver, appendix, prostate, peritoneal lining (mesothelioma), soft-tissue sarcoma and melanoma [2,3,105]. Based on results from a randomized study [171], radiation combined with hyperthermia was included in the 2007 Breast Cancer Guidelines for recurrent breast cancer and other localized cancer recurrences by the National Comprehensive Cancer Network (NCCN, U.S.A.).

Despite positive phase III trials, the clinical application of hyperthermia remains limited. This could be partly due to inadequate monitoring of tumour temperatures or thermal dose, during heat treatments. The temperature distribution throughout a tumour during clinical treatment is not homogeneous due to variable tissue properties and changes in blood flow [172] To ensure high quality of treatments, precise tumour temperature measurements and rigorous thermal dosimetric data are essential. Most hyperthermia centers obtain a sparse number of temperature measurements within intraluminal or interstitial catheters [173]. Thermal dose parameters are dependent on the number of measurement sites and on characteristics such as blood flow and tumor size [174]. It is eventually hoped that temperature measurements during hyperthermia treatment can be improved by measuring 3D thermal distribution in tumours by magnetic resonance imaging (MRI) techniques.

In general, hyperthermia treatments are well tolerated by patients. Hyperthermia can cause some toxicity, including skin burns, but this is usually of limited clinical relevance [166]. Normal tissue damage and toxicity do not generally occur during 1 hour of treatment with temperatures that are below 44°C [175]. Nervous and gastrointestinal tissues appear to be most sensitive.

3. Conclusion

Throughout the past two decades, hyperthermia has been used as a particularly efficient complement to standard cancer treatments such as radiation therapy and chemotherapy. Furthermore, considerable progress has been made in our understanding of the biology, physics and bioengineering involved in hyperthermia. Significant improvement in clinical outcome has been demonstrated for many different types of tumours, including head and neck, breast, brain, bladder, cervix, rectum, lung, esophagus, liver, prostate, melanoma and sarcoma [150]. In Europe, hyperthermia is a standard for the treatment of cervical cancer and some sarcomas. It is a successful alternative for the treatment of other types of cancer such as brain, bladder, rectal and esophageal cancer. Moreover, transurethral microwave thermotherapy (TUMT) has been found to be safe and effective as an alternative to surgery and drug treatment for chronic urogenital pathologies such as benign prostatic hyperplasia [176]. TUMT is a minimally invasive therapy that aims to maintain a good quality of life.

In spite of good clinical results, hyperthermia has received little attention [150]. Several problems associated with the acceptance of this promising treatment modality concern the limited availability of equipment for heating tumours, the lack of awareness concerning clinical results, and the lack of financial resources. Hyperthermia is currently under study in many clinical trials, particularly in Europe, Japan and the US, to improve and better understand this promising technique. Future areas of challenge and opportunity for hyperthermia include: improved understanding of thermal biology; improved technologies for delivery and monitoring of heat treatments in patients; successful high-quality clinical trials; and combination of hyperthermia with emerging cancer therapies [170].

Acknowledgements

Financial support is gratefully acknowledged from the Natural Sciences and Engineering Research Council of Canada (NSERC) (DAB).

Author details

Ahmed Bettaieb[1], Paulina K. Wrzal[2] and Diana A. Averill-Bates[3]

1 Department of Nutrition, University of California, Davis, California, USA

2 Department of Pharmacology and Therapeutics, McGill University, Montréal, Québec, Canada

3 Département des sciences biologiques, Université du Québec à Montréal, Succursale Centre-Ville, Montréal, Québec, Canada

References

[1] Eheman C. et al. Annual Report to the Nation on the status of cancer, 1975-2008, featuring cancers associated with excess weight and lack of sufficient physical activity. Cancer, 2012. 118(9): 2338-66.

[2] van der Zee J. Heating the patient: a promising approach? Ann Oncol, 2002. 13(8): p. 1173-84.

[3] Van der Zee J and MC Erasmus. Hyperthermia in addition to radiotherapy. Clin Oncol (R Coll Radiol), 2007. 19(3 Suppl): S18.

[4] Ahmed M and SN Goldberg. Basic science research in thermal ablation. Surg Oncol Clin N Am, 2011. 20(2): 237-58.

[5] Horsman MR and J Overgaard. Hyperthermia: a potent enhancer of radiotherapy. Clin Oncol (R Coll Radiol), 2007. 19(6): 418-26.

[6] Bryan CP. Ancient Egyptian medicine: the Papyrus Ebers Chicago: Ares Publishers. 1974.

[7] Storm FK. Background, principles, and practice. In: Storm, F.K. (ed.) Hyperthermia in cancer therapy. Boston: G.K. Hall; 1983. p. 1-8.

[8] Decker WK and A Safdar. Bioimmunoadjuvants for the treatment of neoplastic and infectious disease: Coley's legacy revisited. Cytokine Growth Factor Rev, 2009. 20(4): 271-81.

[9] Nauts HC, GA Fowler and FH Bogatko. A review of the influence of bacterial infection and of bacterial products (Coley's toxins) on malignant tumors in man; a critical analysis of 30 inoperable cases treated by Coley's mixed toxins, in which diagnosis was confirmed by microscopic examination selected for special study. Acta Med Scand Suppl, 1953. 276: 1-103.

[10] Stewart JR. Prospects for hyperthermia in cancer treatment. In: Urano M. and Douple E. (eds.) Hyperthermia and Oncology. Vol. 1. Utrecht: VSP; 1988. p. 1-12.

[11] Westermark F. Uber die behandlung des ulceration cervix-carcinoma mittels konstanter warme. Zentralbl Gynaekol 1998. 22: 1335.

[12] Crile G Jr. The effects of heat and radiation on cancers implanted on the feet of mice. Cancer Res, 1963. 23: 372-80.

[13] Dewey WC et al. Cellular responses to combinations of hyperthermia and radiation. Radiology, 1977. 123(2): 463-74.

[14] Milleron RS and SB Bratton. 'Heated' Debates in Apoptosis. Cell Mol Life Sci, 2007. 64(18): 2329-33.

[15] Lindquist S. The heat-shock response. Annu Rev Biochem, 1986. 55: 1151-91.

[16] Hildebrandt B et al. The cellular and molecular basis of hyperthermia. Crit Rev Oncol Hematol, 2002. 43(1): 33-56.

[17] Roti Roti JL. Cellular responses to hyperthermia (40-46 degrees C): cell killing and molecular events. Int J Hyperthermia, 2008. 24(1): 3-15.

[18] Lepock JR. How do cells respond to their thermal environment? Int J Hyperthermia, 2005. 21(8): 681-7.

[19] Richter K, M Haslbeck and J Buchner. The heat shock response: life on the verge of death. Mol Cell, 2010. 40(2): 253-66.

[20] Sonna LA et al. Invited review: Effects of heat and cold stress on mammalian gene expression. J Appl Physiol, 2002. 92(4): 1725-42.

[21] Sugahara T et al. Kadota Fund International Forum 2004. Application of thermal stress for the improvement of health, 15-18 June 2004, Awaji Yumebutai International Conference Center, Awaji Island, Hyogo, Japan. Final report. Int J Hyperthermia, 2008. 24(2): 123-40.

[22] Bates DA et al. Effects of thermal adaptation at 40 degrees C on membrane viscosity and the sodium-potassium pump in Chinese hamster ovary cells. Cancer Res, 1985. 45(10): 4895-9.

[23] Gerner EW et al. Factors regulating membrane permeability alter thermal resistance. Ann N Y Acad Sci, 1980. 335: 215-33.

[24] Lecavalier D and WJ Mackillop. The effect of hyperthermia on glucose transport in normal and thermal-tolerant Chinese hamster ovary cells. Cancer Lett, 1985. 29(2): 223-31.

[25] Bates DA and WJ Mackillop. Hyperthermia, adriamycin transport, and cytotoxicity in drug-sensitive and -resistant Chinese hamster ovary cells. Cancer Res, 1986. 46(11): 5477-81.

[26] Bensaude O et al. Heat-shock induced protein modifications and modulation of enzyme activities. EXS, 1996. 77: 199-219.

[27] Kuhl NM, J Kunz and L Rensing. Heat shock-induced arrests in different cell cycle phases of rat C6-glioma cells are attenuated in heat shock-primed thermotolerant cells. Cell Prolif, 2000. 33(3): 147-66.

[28] Lui JC and SK Kong. Heat shock protein 70 inhibits the nuclear import of apoptosis-inducing factor to avoid DNA fragmentation in TF-1 cells during erythropoiesis. FEBS Lett, 2007. 581(1): 109-17.

[29] Flanagan SW, PL Moseley and GR Buettner. Increased flux of free radicals in cells subjected to hyperthermia: detection by electron paramagnetic resonance spin trapping. FEBS Lett, 1998. 431(2): 285-6.

[30] Moriyama-Gonda N et al. Heat-induced cellular damage and tolerance in combination with adriamycin for the PC-3 prostate cancer cell line: relationships with cytotoxicity, reactive oxygen species and heat shock protein 70 expression. Eur Urol, 2000. 38(2): 235-40.

[31] Katschinski DM et al. Role of tumor necrosis factor alpha in hyperthermia-induced apoptosis of human leukemia cells. Cancer Res, 1999. 59(14): 3404-10.

[32] Bettaieb A and DA Averill-Bates. Thermotolerance induced at a fever temperature of 40 degrees C protects cells against hyperthermia-induced apoptosis mediated by death receptor signalling. Biochem Cell Biol, 2008. 86(6): 521-38.

[33] Pallepati P and DA Averill-Bates. Mild thermotolerance induced at 40 degrees C protects HeLa cells against activation of death receptor-mediated apoptosis by hydrogen peroxide. Free Radic Biol Med, 2011. 50(6): 667-79.

[34] Lord-Fontaine S and DA Averill. Enhancement of cytotoxicity of hydrogen peroxide by hyperthermia in chinese hamster ovary cells: role of antioxidant defenses. Arch Biochem Biophys, 1999. 363(2): 283-95.

[35] Lord-Fontaine S and DA Averill-Bates. Heat shock inactivates cellular antioxidant defenses against hydrogen peroxide: protection by glucose. Free Radic Biol Med, 2002. 32(8): 752-65.

[36] Przybytkowski E and DA Averill-Bates. Correlation between glutathione and stimulation of the pentose phosphate cycle in situ in Chinese hamster ovary cells exposed to hydrogen peroxide. Arch Biochem Biophys, 1996. 325(1): 91-8.

[37] Yoshikawa T et al. The role of active oxygen species and lipid peroxidation in the antitumor effect of hyperthermia. Cancer Res, 1993. 53(10 Suppl): 2326-9.

[38] Pallepati P and D Averill-Bates. Mild thermotolerance induced at 40 degrees C increases antioxidants and protects HeLa cells against mitochondrial apoptosis induced by hydrogen peroxide: Role of p53. Arch Biochem Biophys, 2010. 495(2): 97-111.

[39] Vujaskovic Z et al. Temperature-dependent changes in physiologic parameters of spontaneous canine soft tissue sarcomas after combined radiotherapy and hyperthermia treatment. Int J Radiat Oncol Biol Phys, 2000. 46(1): 179-85.

[40] Bicher HI. The physiological effects of hyperthermia. Radiology, 1980. 137(2): 511-3.

[41] Samali A et al. Thermotolerance and cell death are distinct cellular responses to stress: dependence on heat shock proteins. FEBS Lett, 1999. 461(3): 306-10.

[42] Nakahata K et al. Heat shock induces centrosomal dysfunction, and causes non-apoptotic mitotic catastrophe in human tumour cells. Int J Hyperthermia, 2002. 18(4): 332-43.

[43] Mackey MA and F Ianzini. Enhancement of radiation-induced mitotic catastrophe by moderate hyperthermia. Int J Radiat Biol, 2000. 76(2): 273-80.

[44] Bettaieb A and DA Averill-Bates. Thermotolerance induced at a mild temperature of 40 degrees C protects cells against heat shock-induced apoptosis. J Cell Physiol, 2005. 205(1): 47-57.

[45] Boreham DR et al. Heat-induced thermal tolerance and radiation resistance to apoptosis in human lymphocytes. Biochem Cell Biol, 1997. 75(4): 393-7.

[46] Mosser DD and LH Martin. Induced thermotolerance to apoptosis in a human T lymphocyte cell line. J Cell Physiol, 1992. 151(3): 561-70.

[47] Poe BS and KL O'Neill. Inhibition of protein synthesis sensitizes thermotolerant cells to heat shock induced apoptosis. Apoptosis, 1997. 2(6): 510-7.

[48] Buzzard KA et al. Heat shock protein 72 modulates pathways of stress-induced apoptosis. J Biol Chem, 1998. 273(27): 17147-53.

[49] Sakaguchi Y et al. Apoptosis in tumors and normal tissues induced by whole body hyperthermia in rats. Cancer Res, 1995. 55(22): 5459-64.

[50] Babbs CF and DP DeWitt. Physical principles of local heat therapy for cancer. Med Instrum, 1981. 15(6): 367-73.

[51] Yuguchi T et al. Combined use of hyperthermia and irradiation cause antiproliferative activity and cell death to human esophageal cell carcinoma cells-mainly cell cycle examination. Hum Cell, 2002. 15(1): p. 33-42.

[52] Fokas E, WG McKenna and RJ Muschel. The impact of tumor microenvironment on cancer treatment and its modulation by direct and indirect antivascular strategies. Cancer Metastasis Rev, 2012. 31(3-4):823-42.

[53] Cosse JP and C Michiels. Tumour hypoxia affects the responsiveness of cancer cells to chemotherapy and promotes cancer progression. Anticancer Agents Med Chem, 2008. 8(7): 790-7.

[54] Zhou J et al. Tumor hypoxia and cancer progression. Cancer Lett, 2006. 237: 10-21.

[55] Song CW. Physiological factors in hyperthermia. Natl Cancer Inst Monogr, 1982. 61: 169-76.

[56] Song SK et al. Increased intracellular Ca2+: a critical link in the pathophysiology of sepsis? Proc Natl Acad Sci U S A, 1993. 90(9): 3933-7.

[57] Wike-Hooley JL et al. Human tumour pH changes following hyperthermia and radiation therapy. Eur J Cancer Clin Oncol, 1984. 20(5): 619-23.

[58] Oleson JR et al. Biological and clinical aspects of hyperthermia in cancer therapy. Am J Clin Oncol, 1988. 11(3): 368-80.

[59] Song CW, H Park and RJ Griffin. Improvement of tumor oxygenation by mild hyperthermia. Radiat Res, 2001. 155(4): 515-28.

[60] Iwata K et al. Tumour pO2 can be increased markedly by mild hyperthermia. Br J Cancer Suppl, 1996. 27: S217-21.

[61] Song CW. Effect of local hyperthermia on blood flow and microenvironment: a review. Cancer Res, 1984. 44(10 Suppl): 4721s-4730s.

[62] Bogovic J et al. Posttreatment histology and microcirculation status of osteogenic sarcoma after a neoadjuvant chemo- and radiotherapy in combination with local electromagnetic hyperthermia. Onkologie, 2001. 24(1): 55-8.

[63] Calderwood SK, SS Mambula and PJ Gray Jr. Extracellular heat shock proteins in cell signaling and immunity. Ann N Y Acad Sci, 2007. 1113: 28-39.

[64] Peer AJ et al. Diverse immune mechanisms may contribute to the survival benefit seen in cancer patients receiving hyperthermia. Immunol Res, 2010. 46(1-3): 137-54.

[65] Wang XY et al. Current ideas about applications of heat shock proteins in vaccine design and immunotherapy. Int J Hyperthermia, 2005. 21(8): 717-22.

[66] Subjeck JR, JJ Sciandra and RJ Johnson, Heat shock proteins and thermotolerance; a comparison of induction kinetics. Br J Radiol, 1982. 55(656): 579-84.

[67] Samali A and TG Cotter. Heat shock proteins increase resistance to apoptosis. Exp Cell Res, 1996. 223(1): 163-70.

[68] Landry J et al. Synthesis and degradation of heat shock proteins during development and decay of thermotolerance. Cancer Res, 1982. 42(6): 2457-61.

[69] Martindale JL and NJ Holbrook. Cellular response to oxidative stress: signaling for suicide and survival. J Cell Physiol, 2002. 192(1): 1-15.

[70] Morimoto T et al. Hyperthermia enhances spectrin breakdown in transient focal cerebral ischemia. Brain Res, 1997. 746(1-2): 43-51.

[71] Landry J et al. Thermotolerance and heat shock proteins induced by hyperthermia in rat liver cells. Int J Radiat Oncol Biol Phys, 1982. 8(1): 59-62.

[72] Hayashi M et al. Reversal of P-glycoprotein associated multidrug resistance by new isoprenoid derivatives. Anticancer Drug Des, 2001. 16(4-5): 255-60.

[73] Kregel KC. Heat shock proteins: modifying factors in physiological stress responses and acquired thermotolerance. J Appl Physiol, 2002. 92(5): 2177-86.

[74] Akerfelt M, RI Morimoto and L Sistonen. Heat shock factors: integrators of cell stress, development and lifespan. Nat Rev Mol Cell Biol, 2010. 11(8): 545-55.

[75] Morimoto RI. The heat shock response: systems biology of proteotoxic stress in aging and disease. Cold Spring Harb Symp Quant Biol, 2011. 76: 91-9.

[76] Kim HJ, EJ Song and KJ Lee. Proteomic analysis of protein phosphorylations in heat shock response and thermotolerance. J Biol Chem, 2002. 277(26): 23193-207.

[77] Parsell DA and S Lindquist. The function of heat-shock proteins in stress tolerance: degradation and reactivation of damaged proteins. Annu Rev Genet, 1993. 27: 437-96.

[78] Favatier F et al. Variation in hsp gene expression and Hsp polymorphism: do they contribute to differential disease susceptibility and stress tolerance? Cell Stress Chaperones, 1997. 2(3): 141-55.

[79] Krebs RA and ME Feder. Deleterious consequences of Hsp70 overexpression in Drosophila melanogaster larvae. Cell Stress Chaperones, 1997. 2(1): 60-71.

[80] Podrabsky JE and GN Somero. An inducible 70 kDa-class heat shock protein is constitutively expressed during early development and diapause in the annual killifish Austrofundulus limnaeus. Cell Stress Chaperones, 2007. 12(3): 199-204.

[81] Easton DP, Y Kaneko and JR Subjeck. The hsp110 and Grp1 70 stress proteins: newly recognized relatives of the Hsp70s. Cell Stress Chaperones, 2000. 5(4): 276-90.

[82] Sreedhar AS and P Csermely. Heat shock proteins in the regulation of apoptosis: new strategies in tumor therapy: a comprehensive review. Pharmacol Ther, 2004. 101(3): 227-57.

[83] Beere HM. "The stress of dying": the role of heat shock proteins in the regulation of apoptosis. J Cell Sci, 2004. 117(Pt 13): 641-51.

[84] Beere HM and DR Green. Stress management - heat shock protein-70 and the regulation of apoptosis. Trends Cell Biol, 2001. 11(1): 6-10.

[85] Lanneau D et al. Heat shock proteins: essential proteins for apoptosis regulation. J Cell Mol Med, 2008. 12(3): 743-61.

[86] Schett G et al. Activation of Fas inhibits heat-induced activation of HSF1 and up-regulation of hsp70. Faseb J, 1999. 13(8): 833-42.

[87] Mehlen P, K Schulze-Osthoff and AP Arrigo. Small stress proteins as novel regulators of apoptosis. Heat shock protein 27 blocks Fas/APO-1- and staurosporine-induced cell death. J Biol Chem, 1996. 271(28): 16510-4.

[88] Paul C et al. Hsp27 as a negative regulator of cytochrome C release. Mol Cell Biol, 2002. 22(3): 816-34.

[89] Gabai VL et al. Hsp72 and stress kinase c-jun N-terminal kinase regulate the bid-dependent pathway in tumor necrosis factor-induced apoptosis. Mol Cell Biol, 2002. 22(10): 3415-24.

[90] Bouchier-Hayes L et al. Characterization of cytoplasmic caspase-2 activation by induced proximity. Mol Cell, 2009. 35(6): 830-40.

[91] Steel, R., et al., Hsp72 inhibits apoptosis upstream of the mitochondria and not through interactions with Apaf-1. J Biol Chem, 2004. 279(49): 51490-9.

[92] Beere, H.M., Stressed to death: regulation of apoptotic signaling pathways by the heat shock proteins. Sci STKE, 2001. 2001(93): re1.

[93] Musch MW et al. Induction of heat shock protein 70 protects intestinal epithelial IEC-18 cells from oxidant and thermal injury. Am J Physiol, 1996. 270(2 Pt 1): C429-36.

[94] Huot J et al. HSP27 phosphorylation-mediated resistance against actin fragmentation and cell death induced by oxidative stress. Cancer Res, 1996. 56(2): 273-9.

[95] Przybytkowski E et al. Thermal adaptation in CHO cells at 40 degrees C: the influence of growth conditions and the role of heat shock proteins. Radiat Res, 1986. 107(3): 317-31.

[96] Field SB and RL Anderson. Thermotolerance: a review of observations and possible mechanisms. Natl Cancer Inst Monogr, 1982. 61: 193-201.

[97] Ostberg JR, KC Kaplan and EA Repasky. Induction of stress proteins in a panel of mouse tissues by fever-range whole body hyperthermia. Int J Hyperthermia, 2002. 18(6): 552-62.

[98] Calderwood SK and DR Ciocca. Heat shock proteins: stress proteins with Janus-like properties in cancer. Int J Hyperthermia, 2008. 24(1): 31-9.

[99] Barnes JA et al. Expression of inducible Hsp70 enhances the proliferation of MCF-7 breast cancer cells and protects against the cytotoxic effects of hyperthermia. Cell Stress Chaperones, 2001. 6(4): 316-25.

[100] Huot J et al. Increased survival after treatments with anticancer agents of Chinese hamster cells expressing the human Mr 27,000 heat shock protein. Cancer Res, 1991. 51(19): 5245-52.

[101] Oesterreich S et al. Basal regulatory promoter elements of the hsp27 gene in human breast cancer cells. Biochem Biophys Res Commun, 1996. 222(1): 155-63.

[102] Oesterreich S et al. The small heat shock protein hsp27 is correlated with growth and drug resistance in human breast cancer cell lines. Cancer Res, 1993. 53(19): 4443-8.

[103] Garrido C. et al. HSP27 as a mediator of confluence-dependent resistance to cell death induced by anticancer drugs. Cancer Res, 1997. 57(13): 2661-7.

[104] Ciocca DR et al. Biological and clinical implications of heat shock protein 27,000 (Hsp27): a review. J Natl Cancer Inst, 1993. 85(19): 1558-70.

[105] Issels RD. Hyperthermia adds to chemotherapy. Eur J Cancer, 2008. 44(17): 2546-54.

[106] Skitzki JJ, EA Repasky and SS Evans. Hyperthermia as an immunotherapy strategy for cancer. Curr Opin Investig Drugs, 2009. 10(6): 550-8.

[107] Lepock JR. Role of nuclear protein denaturation and aggregation in thermal radiosensitization. Int J Hyperthermia, 2004. 20(2): 115-30.

[108] Dewey WC, SA Sapareto and DA Betten. Hyperthermic radiosensitization of synchronous Chinese hamster cells: relationship between lethality and chromosomal aberrations. Radiat Res, 1978. 76(1): 48-59.

[109] Kampinga HH and E Dikomey. Hyperthermic radiosensitization: mode of action and clinical relevance. Int J Radiat Biol, 2001. 77(4): 399-408.

[110] Kampinga HH et al. Resistance to heat radiosensitization and protein damage in thermotolerant and thermoresistant cells. Int J Radiat Biol, 1997. 71(3): 315-26.

[111] Song CW et al. Implications of increased tumor blood flow and oxygenation caused by mild temperature hyperthermia in tumor treatment. Int J Hyperthermia, 2005. 21(8): 761-7.

[112] Jain RK. Normalization of tumor vasculature: an emerging concept in antiangiogenic therapy. Science, 2005. 307(5706): 58-62.

[113] Hahn GM. Potential for therapy of drugs and hyperthermia. Cancer Res, 1979. 39(6 Pt 2): 2264-8.

[114] Marmor JB. Interactions of hyperthermia and chemotherapy in animals. Cancer Res, 1979. 39(6 Pt 2): 2269-76.

[115] Engelhardt R. Rationale for clinical application of hyperthermia and drugs. Strahlenther Onkol, 1987. 163(7): 428-9.

[116] Dahl O. Interaction of hyperthermia and chemotherapy. Recent Results Cancer Res, 1988. 107: 157-69.

[117] Bates DA and WJ Mackillop. The effect of hyperthermia on the uptake and cytotoxicity of melphalan in Chinese hamster ovary cells. Int J Radiat Oncol Biol Phys, 1989. 16(1): 187-91.

[118] Bull JM. An update on the anticancer effects of a combination of chemotherapy and hyperthermia. Cancer Res, 1984. 44(10 Suppl): 4853s-4856s.

[119] Kampinga HH. Cell biological effects of hyperthermia alone or combined with radiation or drugs: a short introduction to newcomers in the field. Int J Hyperthermia, 2006. 22(3): 191-6.

[120] Ling V. Multidrug resistance: molecular mechanisms and clinical relevance. Cancer Chemother Pharmacol, 1997. 40 Suppl: S3-8.

[121] Kuo MT. Redox regulation of multidrug resistance in cancer chemotherapy: molecular mechanisms and therapeutic opportunities. Antioxid Redox Signal, 2009. 11(1): 99-133.

[122] Cole SP et al. Overexpression of a transporter gene in a multidrug-resistant human lung cancer cell line. Science, 1992. 258(5088): 1650-4.

[123] Hipfner DR, R.G Deeley and SP Cole. Structural, mechanistic and clinical aspects of MRP1. Biochim Biophys Acta, 1999. 1461(2): 359-76.

[124] Jedlitschky G et al. Transport of glutathione, glucuronate, and sulfate conjugates by the MRP gene-encoded conjugate export pump. Cancer Res, 1996. 56(5): 988-94.

[125] Slot AJ, SV Molinski and SP Cole. Mammalian multidrug-resistance proteins (MRPs). Essays Biochem, 2011. 50(1): 179-207.

[126] Volm M. Multidrug resistance and its reversal. Anticancer Res, 1998. 18(4C): 2905-17.

[127] Mannervik B. The isoenzymes of glutathione transferase. Adv Enzymol Relat Areas Mol Biol, 1985. 57: 357-417.

[128] Baird RD and SB Kaye. Drug resistance reversal-are we getting closer? Eur J Cancer, 2003. 39(17): 2450-61.

[129] Tamaki A et al. The controversial role of ABC transporters in clinical oncology. Essays Biochem, 2011. 50(1): 209-32.

[130] Towle LR. Hyperthermia and drug resistance., In: Urano M. and Douple E. (eds.) Hyperthermia and Oncology, Vol. 4. Utrecht: VSP; 1994. p. 91-113.

[131] Herman TS et al. Reversal of resistance to methotrexate by hyperthermia in Chinese hamster ovary cells. Cancer Res, 1981. 41(10): 3840-3.

[132] Raaphorst GP, et al. A comparison of hyperthermia cisplatin sensitization in human ovarian carcinoma and glioma cell lines sensitive and resistant to cisplatin treatment. Cancer Chemother Pharmacol, 1996. 37(6): 574-80.

[133] Wallner KE, M Banda and GC Li. Hyperthermic enhancement of cell killing by mitomycin C in mitomycin C-resistant Chinese hamster ovary cells. Cancer Res, 1987. 47(5): 1308-12.

[134] Ling V and LH Thompson. Reduced permeability in CHO cells as a mechanism of resistance to colchicine. J Cell Physiol, 1974. 83(1): 103-16.

[135] Larrivee B and DA Averill. Melphalan resistance and photoaffinity labelling of P-glycoprotein in multidrug-resistant Chinese hamster ovary cells: reversal of resistance by cyclosporin A and hyperthermia. Biochem Pharmacol, 1999. 58(2): 291-302.

[136] Larrivee B and DA Averill. Modulation of adriamycin cytotoxicity and transport in drug-sensitive and multidrug-resistant Chinese hamster ovary cells by hyperthermia and cyclosporin A. Cancer Chemother Pharmacol, 2000. 45(3): 219-30.

[137] Averill DA and C Su. Sensitization to the cytotoxicity of adriamycin by verapamil and heat in multidrug-resistant Chinese hamster ovary cells. Radiat Res, 1999. 151(6): 694-702.

[138] Averill-Bates DA and B Courtemanche. The effect of hyperthermia and verapamil on melphalan cytotoxicity and transport in multidrug-resistant Chinese hamster ovary cells. Radiat Res, 1995. 143(1): 17-25.

[139] Dumontet C, F Bodin and Y Michal. Potential interactions between antitubulin agents and temperature: implications for modulation of multidrug resistance. Clin Cancer Res, 1998. 4(6): 1563-6.

[140] Liu Z, R Bendayan and XY Wu. Triton-X-100-modified polymer and microspheres for reversal of multidrug resistance. J Pharm Pharmacol, 2001. 53(6): 779-87.

[141] Awasthi S et al. Interactions of melphalan with glutathione and the role of glutathione S-transferase. Drug Metab Dispos, 1996. 24(3): 371-4.

[142] Turcotte S and DA Averill-Bates. Sensitization to the cytotoxicity of melphalan by ethacrynic acid and hyperthermia in drug-sensitive and multidrug-resistant Chinese hamster ovary cells. Radiat Res, 2001. 156(3): 272-82.

[143] Souslova T and DA Averill-Bates. Multidrug-resistant hela cells overexpressing MRP1 exhibit sensitivity to cell killing by hyperthermia: interactions with etoposide. Int J Radiat Oncol Biol Phys, 2004. 60(5): 1538-51.

[144] Uckun FM et al. Radiation and heat sensitivity of human T-lineage acute lymphoblastic leukemia (ALL) and acute myeloblastic leukemia (AML) clones displaying multiple drug resistance (MDR). Int J Radiat Oncol Biol Phys, 1992. 23(1): 115-25.

[145] Indran IR et al. Recent advances in apoptosis, mitochondria and drug resistance in cancer cells. Biochim Biophys Acta, 2011. 1807(6): 735-45.

[146] Reed JC. Dysregulation of apoptosis in cancer. J Clin Oncol, 1999. 17(9): 2941-53.

[147] Wrzal PK, A Bettaieb and DA Averill-Bates. Molecular mechanisms of apoptosis activation by heat shock in multidrug-resistant Chinese hamster cells. Radiat Res, 2008. 170(4): 498-511.

[148] Friesen C et al. Involvement of the CD95 (APO-1/FAS) receptor/ligand system in drug-induced apoptosis in leukemia cells. Nat Med, 1996. 2(5): 574-7.

[149] Ruefli AA et al. P-glycoprotein inhibits caspase-8 activation but not formation of the death inducing signal complex (disc) following Fas ligation. Cell Death Differ, 2002. 9(11): 1266-72.

[150] van der Zee J et al. The Kadota Fund International Forum 2004--clinical group consensus. Int J Hyperthermia, 2008. 24(2): 111-22.

[151] Fiorentini G and A Szasz. Hyperthermia today: electric energy, a new opportunity in cancer treatment. J Cancer Res Ther, 2006. 2(2): 41-6.

[152] Kamisawa T et al. [Thermo-chemo-radiotherapy for advanced biliary carcinoma]. Nippon Rinsho, 2006. 64 Suppl 1: 543-6.

[153] Eveno C et al. [Treatment of peritoneal carcinomatosis with surgery and hyperthermic peroperative intraperitoneal chemotherapy (HIPEC): new aspects and validated indications]. Bull Cancer, 2008. 95(1): 141-5.

[154] Kawai N et al. Effect of heat therapy using magnetic nanoparticles conjugated with cationic liposomes on prostate tumor in bone. Prostate, 2008. 68(7): 84-92.

[155] Kikumori T et al. Anti-cancer effect of hyperthermia on breast cancer by magnetite nanoparticle-loaded anti-HER2 immunoliposomes. Breast Cancer Res Treat, 2009. 113(3): 435-41.

[156] Kuylenstierna J and K Lantorp. [Dangerous drugs in health food store]. Lakartidningen, 1976. 73(33): 691.

[157] Tilly W et al. Regional hyperthermia in conjunction with definitive radiotherapy against recurrent or locally advanced prostate cancer T3 pN0 M0. Strahlenther Onkol, 2005. 181(1): 35-41.

[158] Rampersaud EN, Z Vujaskovic and BA Inman. Hyperthermia as a treatment for bladder cancer. Oncology (Williston Park), 2010. 24(12): 1149-55.

[159] Ginzburg GS, OV Galibin and AI Krasheniuk. [Polarographic study of the rate of dissociation of oxyhemoglobin]. Biofizika, 1972. 17(3): 446-52.

[160] Petrovich Z et al. Deep regional hyperthermia of the liver. A clinical study of 49 patients. Am J Clin Oncol, 1989. 12(5): 378-83.

[161] Petrovich Z et al. Regional hyperthermia of the liver. Strahlenther Onkol, 1989. 165(10): 721-3.

[162] Vernon CC et al. Radiotherapy with or without hyperthermia in the treatment of superficial localized breast cancer: results from five randomized controlled trials. International Collaborative Hyperthermia Group. Int J Radiat Oncol Biol Phys, 1996. 35(4): 731-44.

[163] Overgaard J et al. Randomised trial of hyperthermia as adjuvant to radiotherapy for recurrent or metastatic malignant melanoma. European Society for Hyperthermic Oncology. Lancet, 1995. 345(8949): 540-3.

[164] Valdagni R and M Amichetti. Report of long-term follow-up in a randomized trial comparing radiation therapy and radiation therapy plus hyperthermia to metastatic lymph nodes in stage IV head and neck patients. Int J Radiat Oncol Biol Phys, 1994. 28(1): 163-9.

[165] Datta NR et al. Head and neck cancers: results of thermoradiotherapy versus radiotherapy. Int J Hyperthermia, 1990. 6(3): 479-86.

[166] van der Zee J et al. Point-counterpoint: what is the optimal trial design to test hyperthermia for carcinoma of the cervix? Point: addition of hyperthermia or cisplatin to

radiotherapy for patients with cervical cancer; two promising combinations--no definite conclusions. Int J Hyperthermia, 2002. 18(1): 19-24.

[167] Sharma S et al. Side-effects of local hyperthermia: results of a prospectively randomized clinical study. Int J Hyperthermia, 1990. 6(2): 279-85.

[168] Harima Y et al. A randomized clinical trial of radiation therapy versus thermoradiotherapy in stage IIIB cervical carcinoma. Int J Hyperthermia, 2001. 17(2): 97-105.

[169] Sneed PK et al. Survival benefit of hyperthermia in a prospective randomized trial of brachytherapy boost +/- hyperthermia for glioblastoma multiforme. Int J Radiat Oncol Biol Phys, 1998. 40(2): 287-95.

[170] Hurwitz MD et al. Hyperthermia combined with radiation for the treatment of locally advanced prostate cancer: long-term results from Dana-Farber Cancer Institute study 94-153. Cancer, 2011. 117(3): 510-6.

[171] Jones EL et al. Randomized trial of hyperthermia and radiation for superficial tumors. J Clin Oncol, 2005. 23(13): 3079-85.

[172] Feldmann, H.J., et al., Changes in oxygenation patterns of locally advanced recurrent tumors under thermoradiotherapy. Adv Exp Med Biol, 1994. 345: 479-83.

[173] Paulides, M.M., et al., The clinical feasibility of deep hyperthermia treatment in the head and neck: new challenges for positioning and temperature measurement. Phys Med Biol, 2010. 55(9): 2465-80.

[174] de Bruijne, M., et al., Evaluation of CEM43 degrees CT90 thermal dose in superficial hyperthermia: a retrospective analysis. Strahlenther Onkol, 2010. 186(8): 436-43.

[175] Fajardo, J.E., et al., Chronic meningitis, polyarthritis, lymphadenitis, and pulmonary hemosiderosis. The Journal of pediatrics, 1982. 101(5): 738-40.

[176] Walmsley, K. and S.A. Kaplan, Transurethral microwave thermotherapy for benign prostate hyperplasia: separating truth from marketing hype. J Urol, 2004. 172(4 Pt 1): 1249-55.

IntechOpen

IntechOpen