Tumor biology and physiology

Michael R. Horsman
Professor of Experimental Radiotherapy

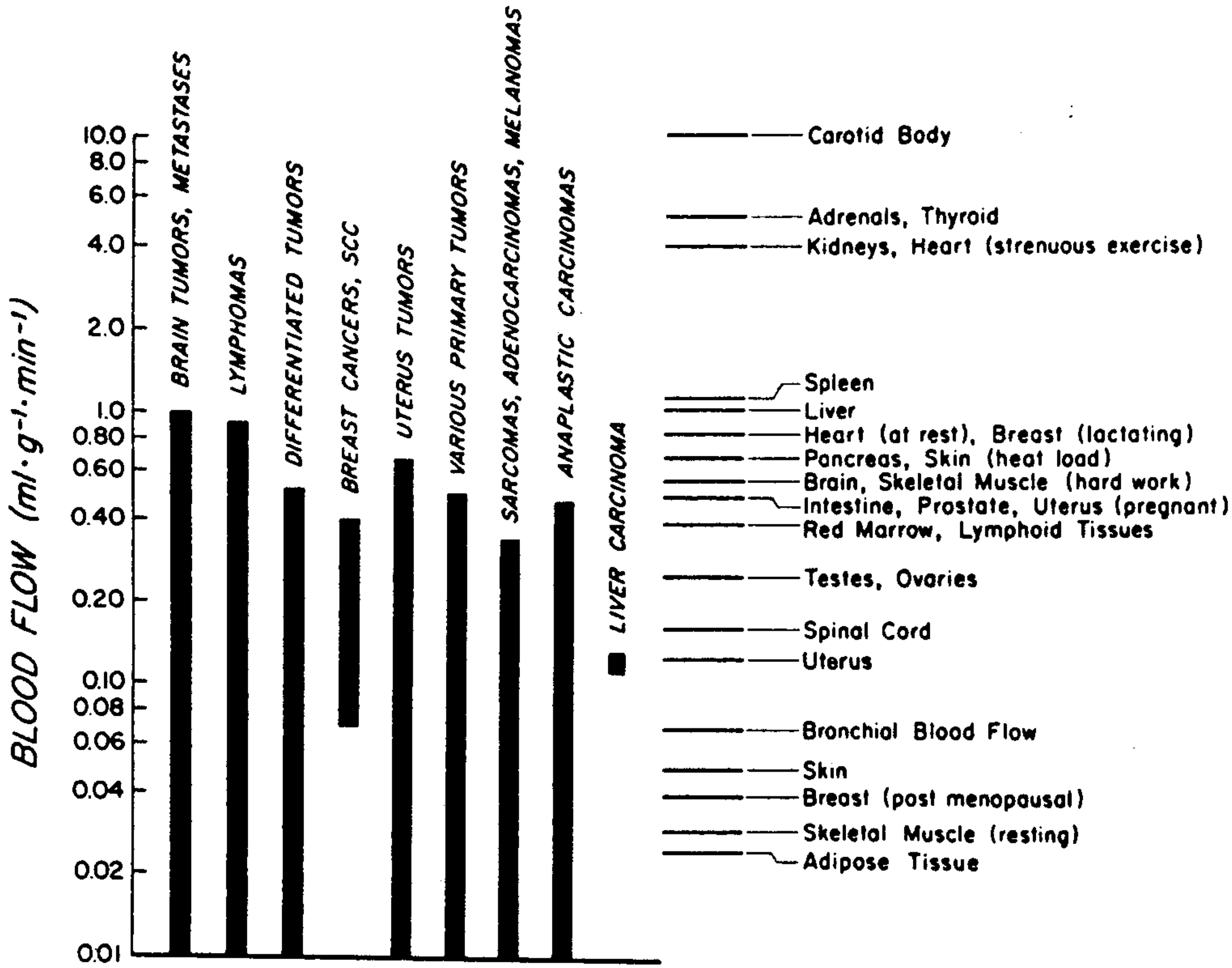
Experimental Clinical Oncology-Dept. Oncology
Aarhus University Hospital
Aarhus, Denmark.

Relationship between the vascular supply of tissues and heat

- Blood flow influences the tissue response to heat
  - ability to heat the tissue
  - determines the microenvironment

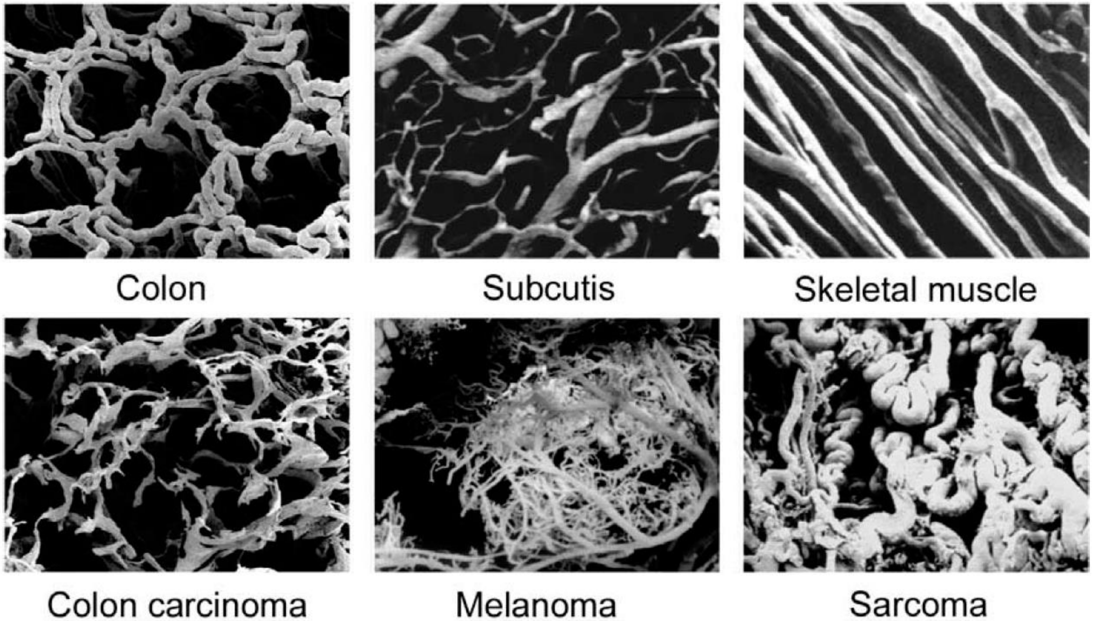
- Hyperthermia changes blood flow in tissues and this will affect tissue physiology

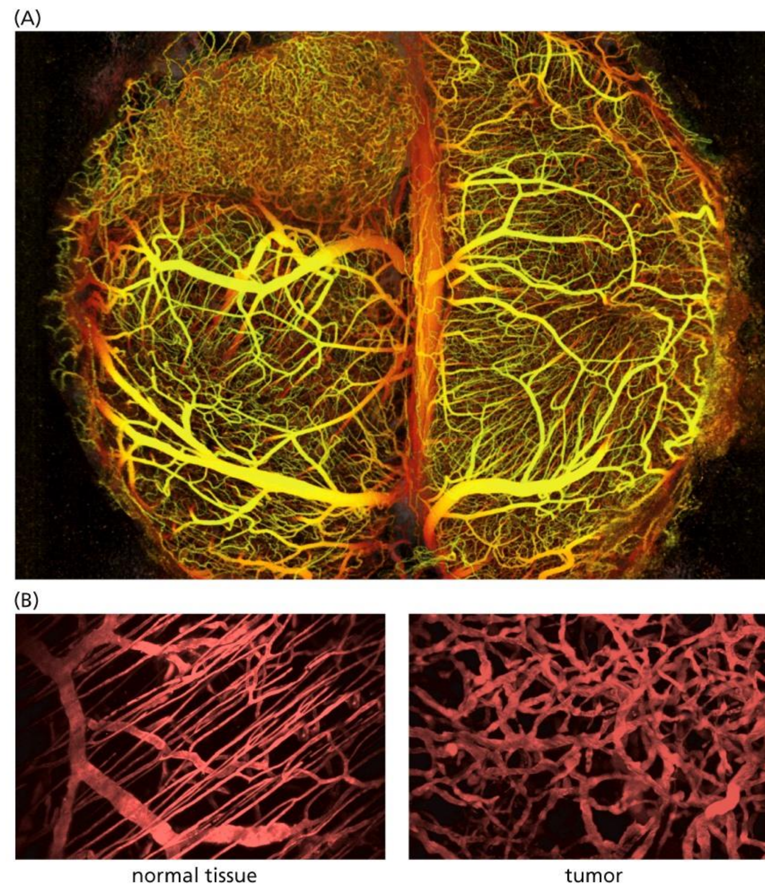
Blood flow in tissues



Vaupel et al. (1989) Cancer Res. 49:6449-65

Tumour .v. Normal tissue vessels



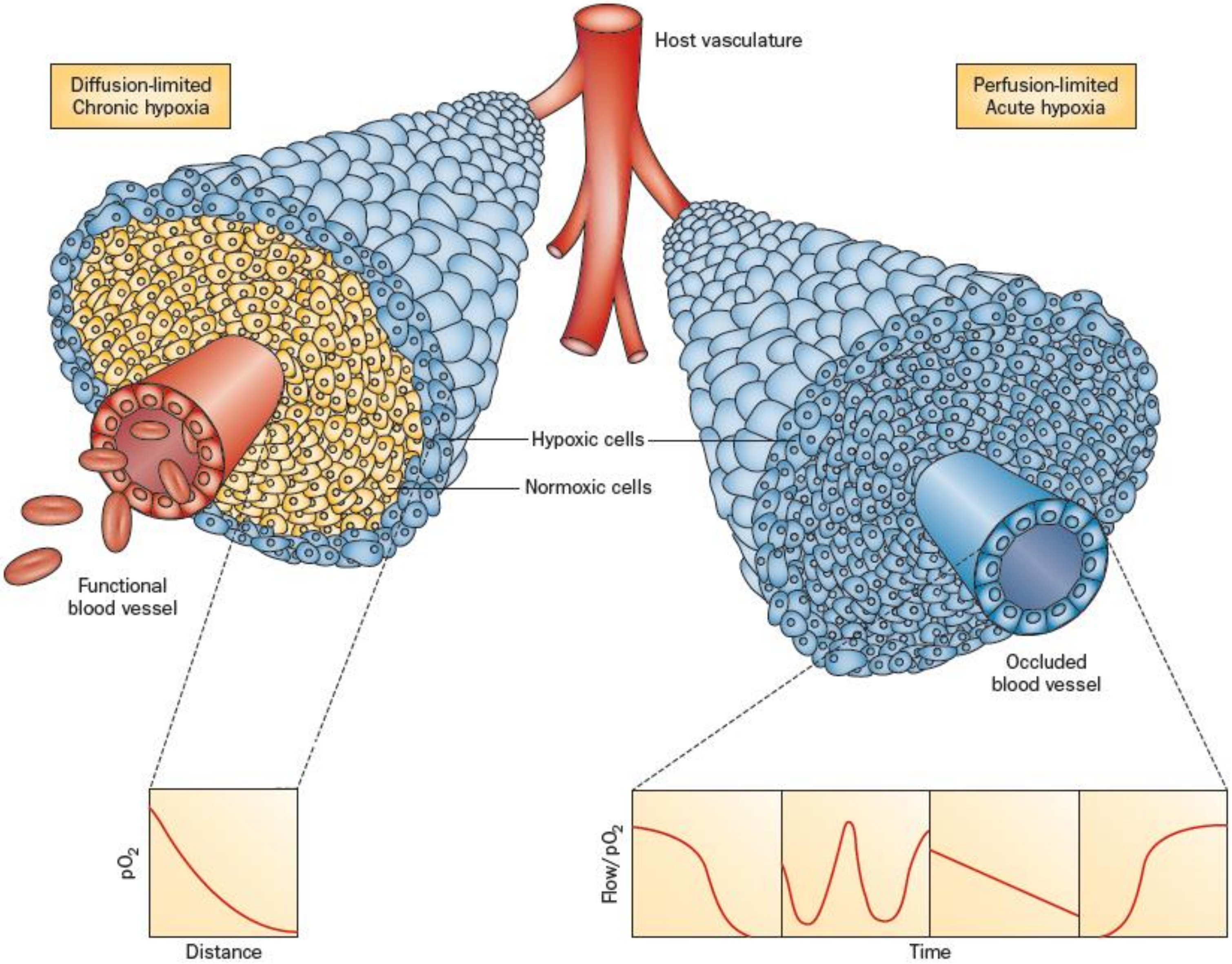


Major Structural and Functional Abnormalities :

- ***Abnormal vascular density***
- ***Contour irregularities***
- ***Loss of hierarchy***
- ***Lack of regulatory control mechanisms***
- ***Structural defects in vessel walls***
- ***Increased vascular permeability***
- ***Flow irregularities***
- ***Cellular aggregations/blockage***
- ***Increased haematocrit***

Weinberg (2014) The Biology of Cancer (2nd Ed.)

Horsman & Vaupel (2016) Frontiers in Oncology Research Topics



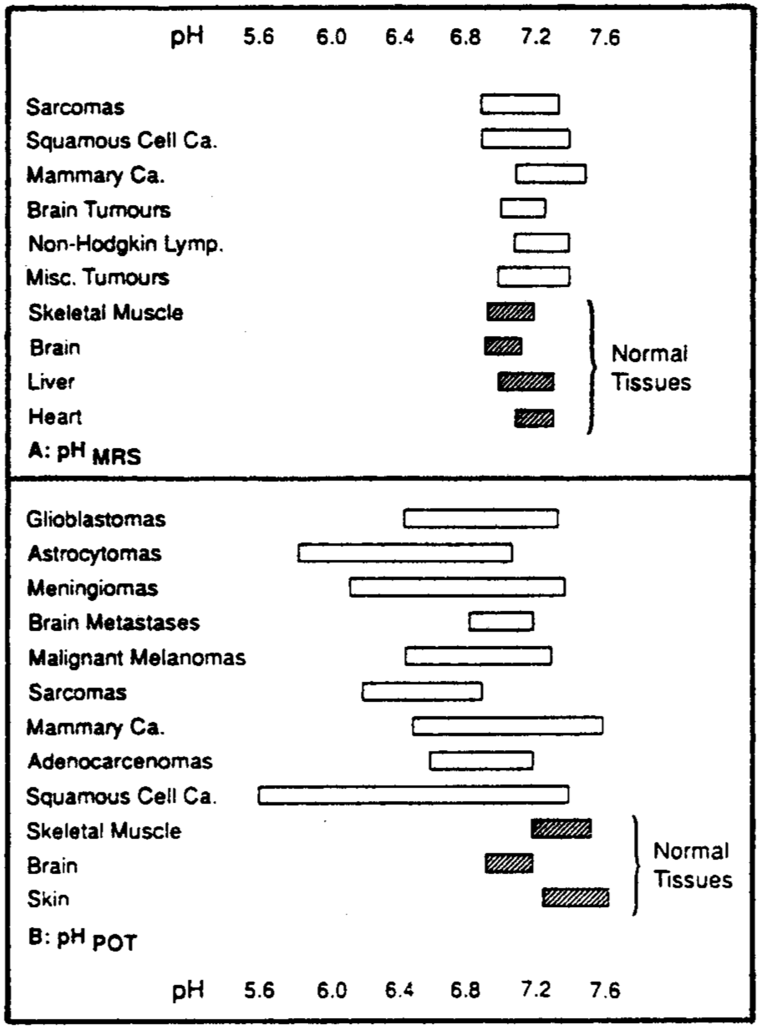
Horsman et al. (2012) Nat. Rev. Clin. Oncol. 9:674-687

Oxygenation and pH

Table 5 *Comparison between the mean pO_2 values in normal tissues and in human malignancies*

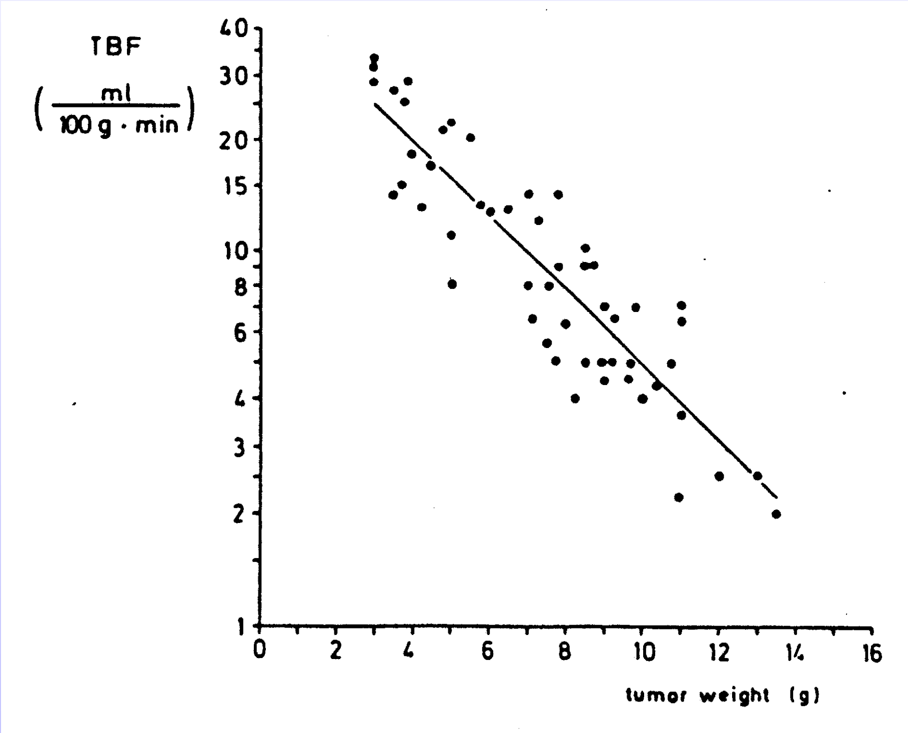
Tumor type	pO_2 (normal tissue)/ pO_2 (tumor)[a]	Refs.
Cervix cancer		
Stage 0	1.6	91
Stage 1	2.4	91
Stage 2	3.2	91
Stage 2	1.4–1.8	101
Squamous cell cancers	1.7	105
	2.4	106
	2.5	84
	4.4	86
	6.3	107
Breast cancer	1.4	88
	2.0	105
	2.4	84
	4.4	107
Melanomas	6.3–6.7	86, 106, 107
Soft tissue sarcomas	2.8	107
	6.3	86
Malignant lymphomas	1.5	105
	2.2	106
Adenocarcinomas	7.1	86
Basal cell epitheliomas	5.6	107

[a] Ratio of mean O_2 tension in normal tissue to mean O_2 tension in tumors.

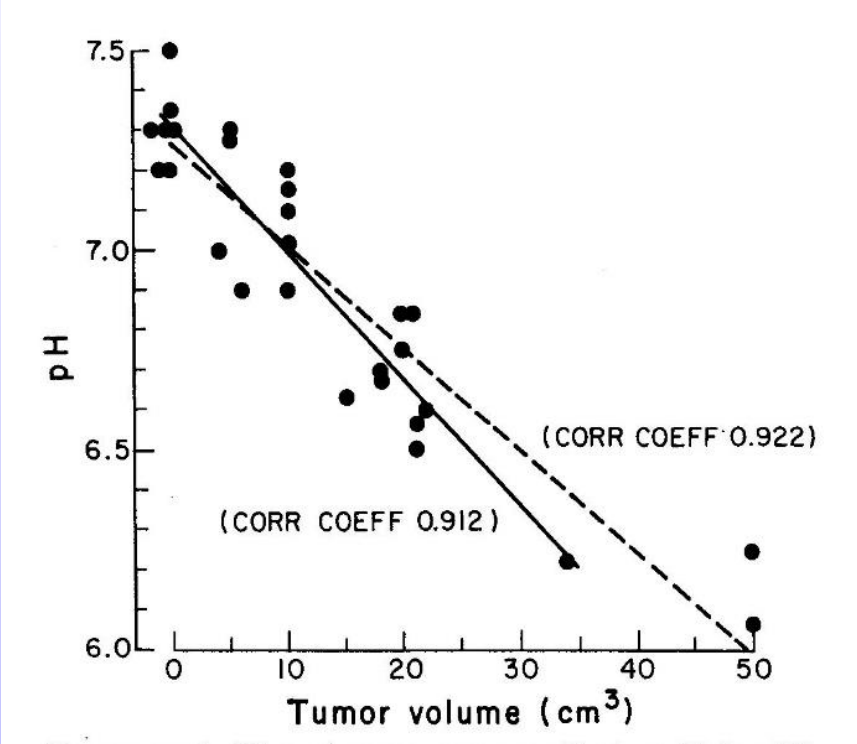


Vaupel et al. (1989) Cancer Res. 49:6449-6465

Effect of tumor size



Vaupel (1979)

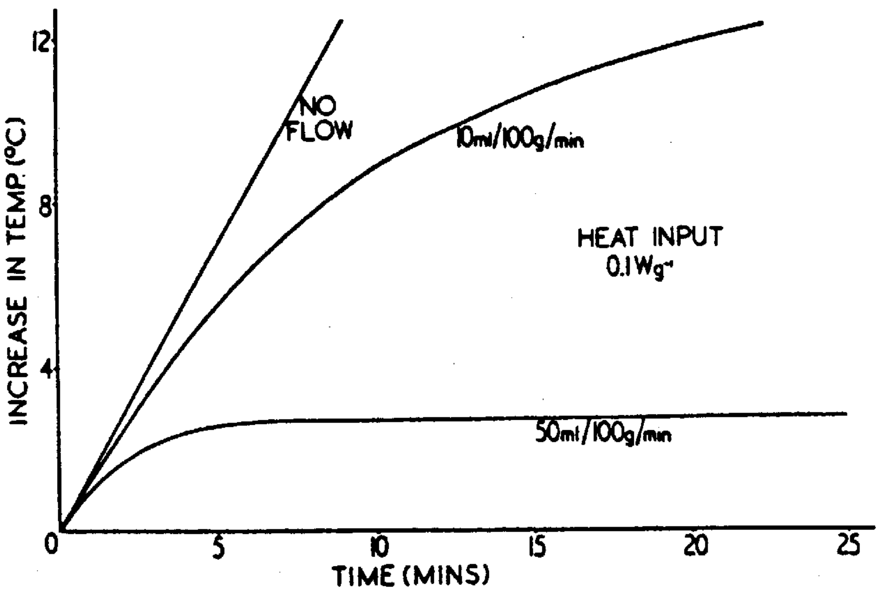


Jain et al. (1984) JNCI 73:429-436

Relationship between blood flow and temperature

$$\Delta T = \frac{I}{kS_t}(1 - e^{-kt}) \quad \text{and} \quad k = \frac{Fp}{100\lambda}$$

Where, ΔT = increase in temperature of a tissue
I = heat imput rate per g of tissue
S_t = specific heat of the tissue (J/g/°C)
F = tissue blood flow (ml/min/100g)
p = density of the tissue (g/ml)
λ = describes the ratio of solubility of heat in tissue and blood

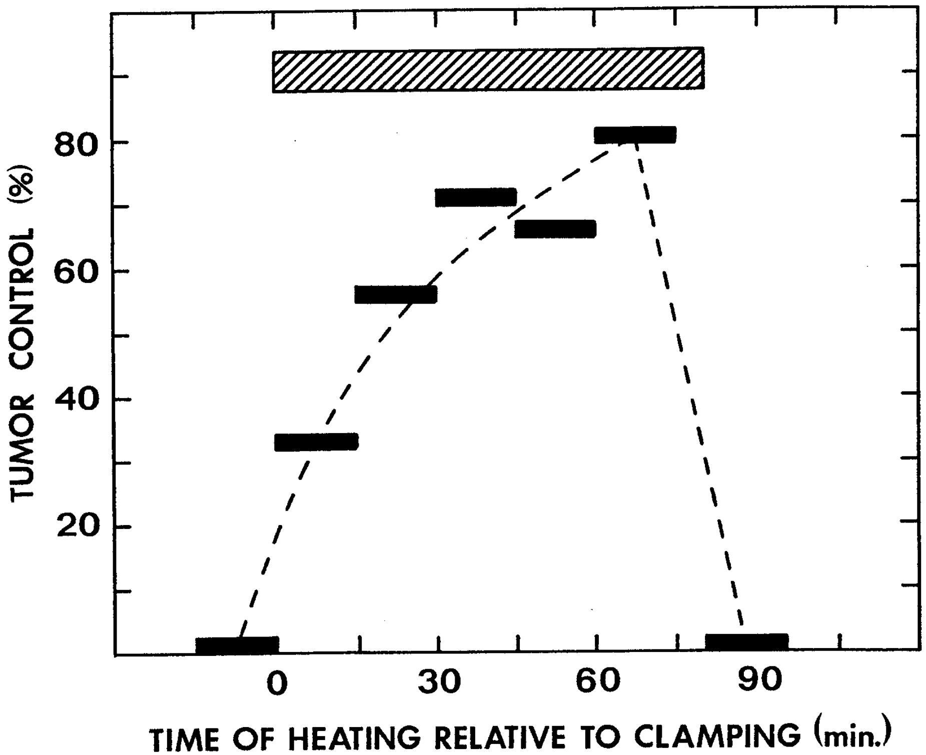


The increase in tissue temperature is inversely proportional to the rate of blood flow, assuming:

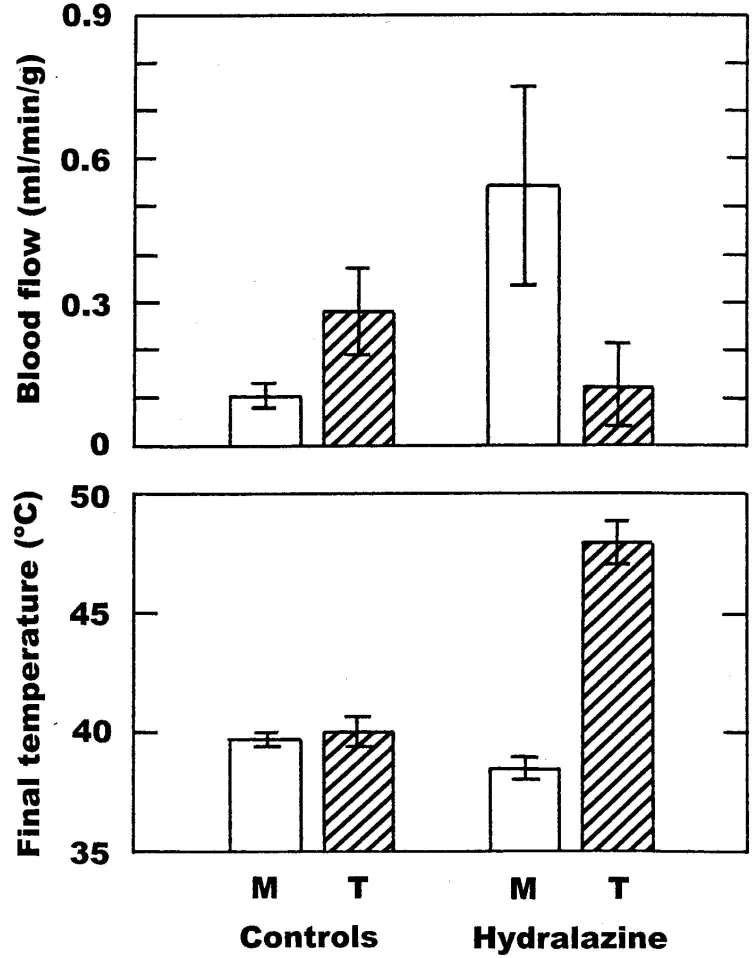
(1) the equilibrium distribution of heat between blood and tissue is immediately obtained
(2) heat is removed from the tissue only by the blood flow
(3) the blood flow is constant
(4) there is no re-circulation of heat
(5) the tissue is homogeneous

Patterson & Strang (1979)
IJROBP 5:235-41

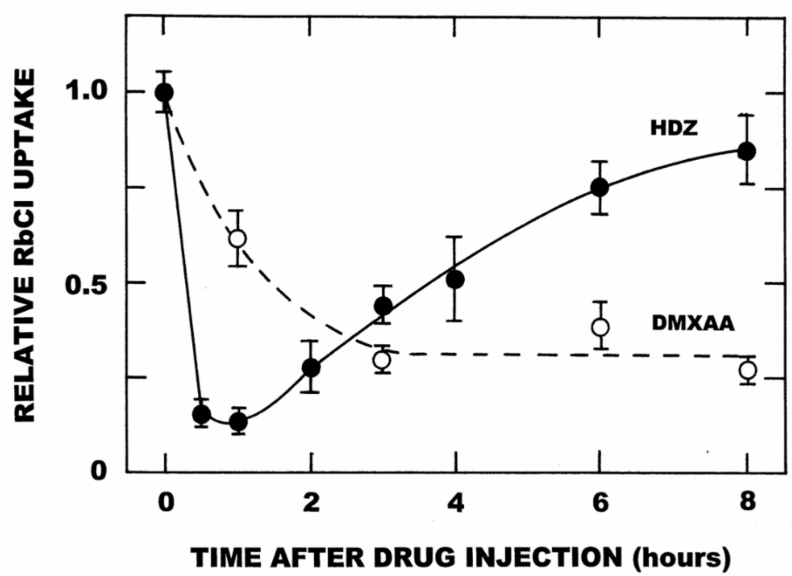
Effect of modifying tissue blood flow

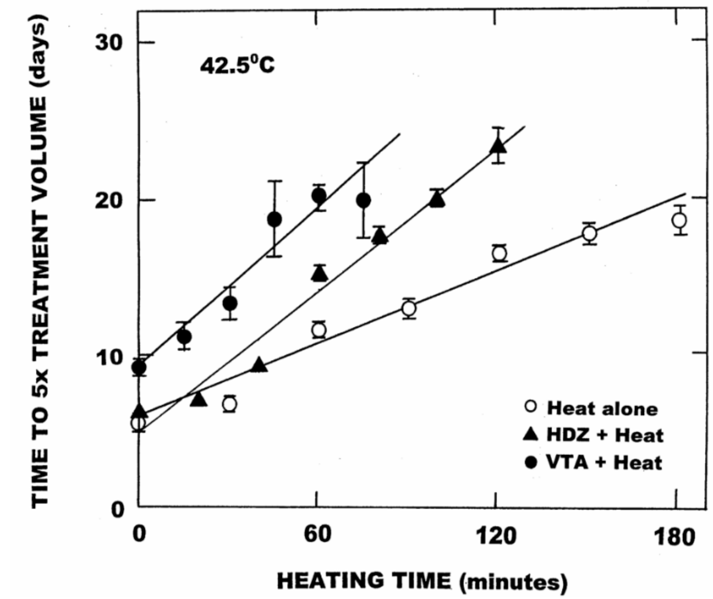


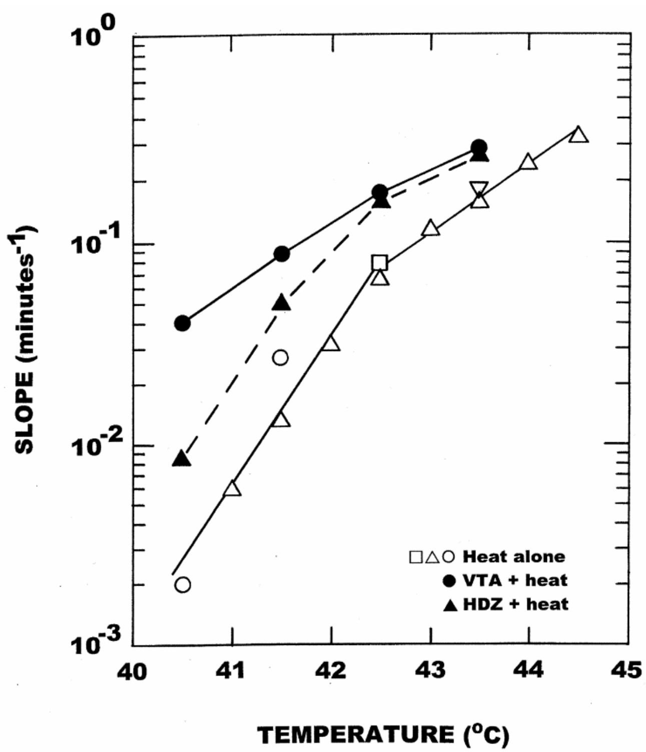
Hill & Denekamp (1978)
Br. J. Radiol. 51:997-1002



Voorhees & Babbs (1982)
EJCCO 19:1027-33







	HDZ	DMXAA
Temperature required	41.5ºC	41.5ºC
Effective temperature	42.2ºC	42.5ºC
Actual temperature	41.7ºC	41.7ºC
Decrease in pO_2[1]	41%	42%
Decrease in pH[2]	0.40 units	0.21 units

[1]Based on % pO_2 values ≤ 5mmHg
[2]Obtained from NMR studies

Exploiting tumour pathophysiology to improve the therapeutic potential of hyperthermia

- Modifiers of the microenvironment
  - pH modification
  - Transient modifiers of blood flow

- Vascular targeting agents:
  - Angiogenesis inhibitors (AIs)
  - Vascular disrupting agents (VDAs)

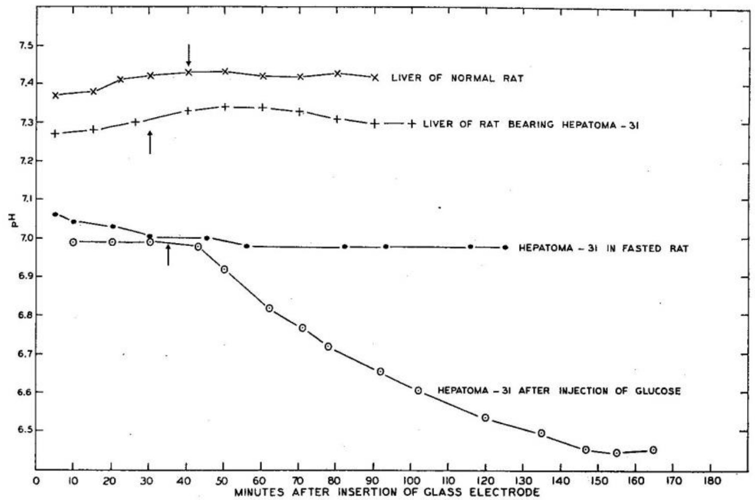
Injecting glucose

Table 2.
The effect of hyperglycemia on the rodent tumor pH.
Glucose was given intraperitoneally.

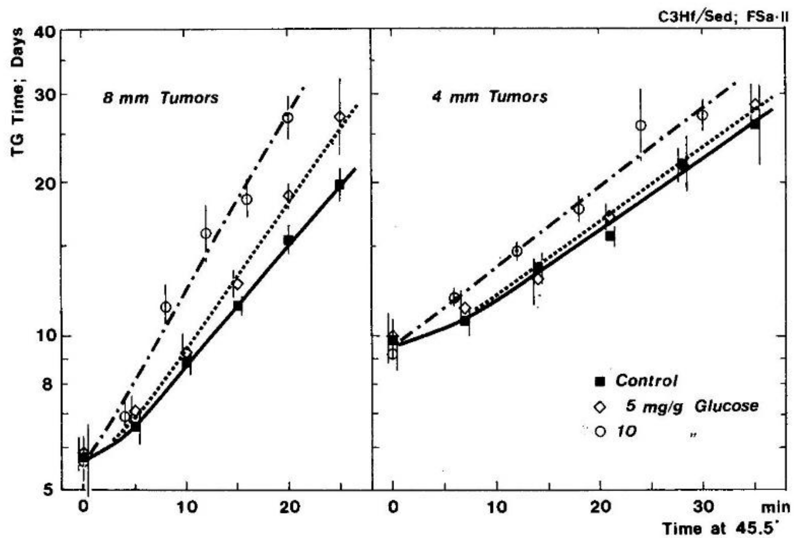
Tumor	Before	Glucose (mg/g)	After	Investigators	
Rat hepatoma	6.99 (6.81–7.10)	6.0	6.42 (6.40–6.45)	Kahler and Robertson	(1943)
Rat hepatoma	7.02 (6.72–7.22)	6.0	6.73 (6.35–6.99)	Kahler and Robertson	(1943)
Mouse Sarcoma	7.0	5.0	6.6	Naeslund and Swenson	(1953)
Rat hepatoma	6.96 ± 0.17	6.0	6.46 ± 0.22	Eden *et al.*	(1955)
Rat sarcoma	6.95 ± 0.25	6.0	6.55 ± 0.27	Eden *et al.*	(1955)
Rat lymphosarcoma	7.00 ± 0.20	6.0	6.50 ± 0.30	Eden *et al.*	(1955)
Rat sarcoma	7.04 ± 0.11	6.0	6.67 ± 0.14	Eden *et al.*	(1955)
Rat Harderian-gl.ca	7.00 ± 0.11	6.0	6.54 ± 0.23	Eden *et al.*	(1955)
Rat sarcoma	7.01 ± 0.16	6.0	6.63 ± 0.22	Eden *et al.*	(1955)
Rat fibrosarcoma	6.83 ± 0.24	6.0	6.48 ± 0.28	Eden *et al.*	(1955)
Rat hepatoma	7.06 ± 0.22	6.0	6.62 ± 0.27	Eden *et al.*	(1955)
Rat TVIA 1.0–2.5g	7.0 (6.8–7.1)*	Con.Inf.	6.5 (6.0–7.0)	Jahde and Rajewsky	(1982)
Rat TVIA 4.0–6.0g	6.9 (6.7–7.1)*	Con.Inf.	6.1 (5.5–6.7)	Jahde and Rajewsky	(1982)
Rat Walker 256 ca	6.98 ± 0.13	6.0	6.0	Jain *et al.*	(1984)

* Continuous infusion

Urano (1988) Hyperthermia & Oncology 1:161-200

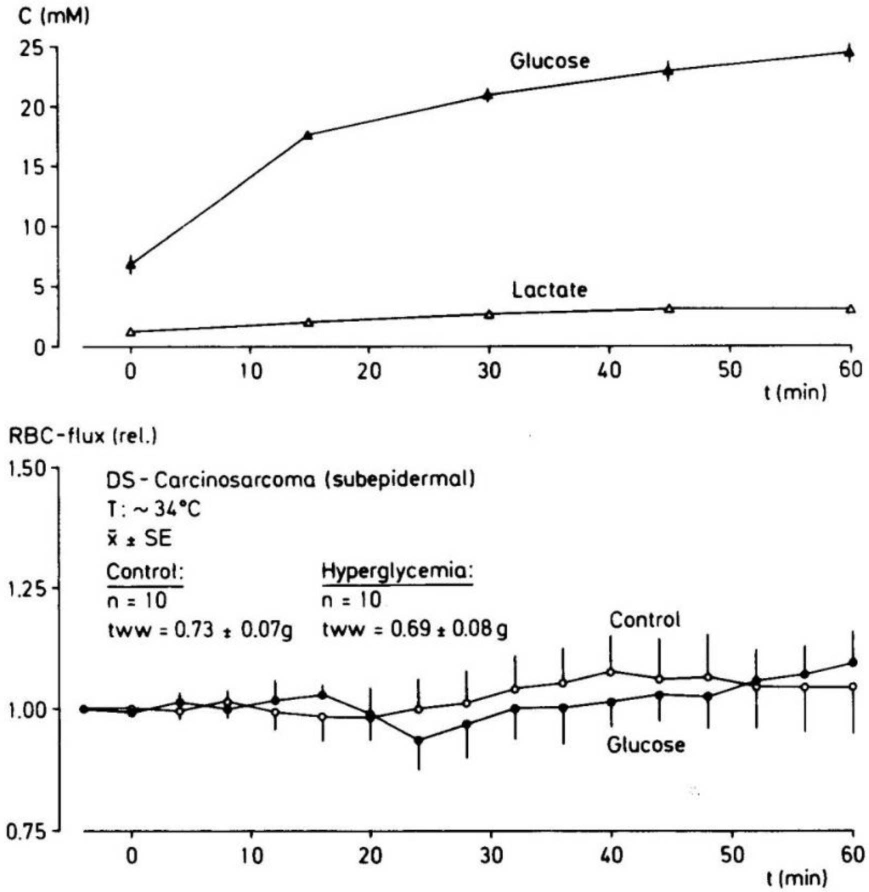


Kahler & Robertson (1943) JNCI 3:495-501



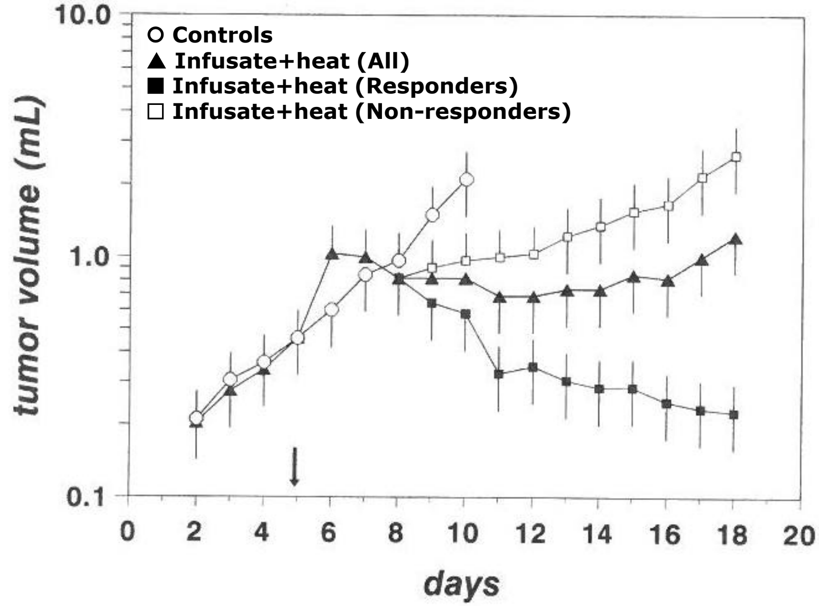
Urano (1988) Hyperthermia & Oncology 1:161-200

Infusing glucose



Vaupel et al. (1989) Int. J. Hyperthermia 5:199-210

Infusate (glucose/lactate/buffer; 60 min) + Heat (43^{O}C; 30 min)



Mueller-Klieser et al. (1996) Int. J. Hyperthermia 12:501-511

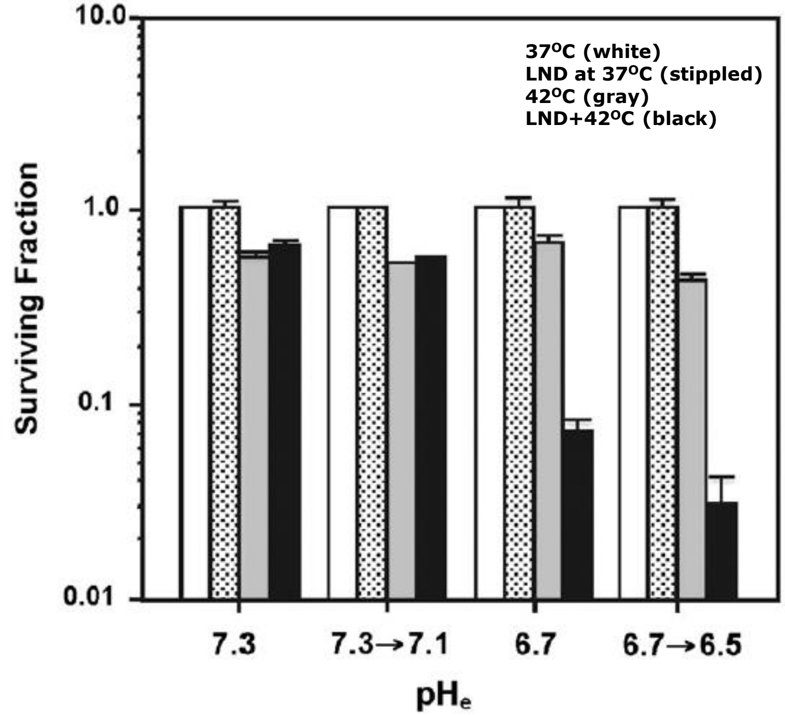
Lonidamine

Table I. Summary of the effects of 150 μM LND on pH_i in DB-1 cells.

Treatment pH_e	Control	150 μM lonidamine
7.3	7.20 ± 0.07 (6)	7.15 ± 0.16 (3)
7.3–7.1	7.05 ± 0.04 (3)	6.93 ± 0.15 (4)
6.7	6.76 ± 0.04 (5)	6.30 ± 0.21 (9)
6.7–6.5	6.52 ± 0.15 (7)	6.09 ± 0.26 (6)

The pH_i values (mean ± SD) are after 60 min at 37 °C. Extracellular pH (pH_e) was measured immediately following the 60 min of pH_i monitoring. The number of separate experiments is in parentheses.

Lonidamine for 60 min prior to and during heating (42^{O}C, 2h)

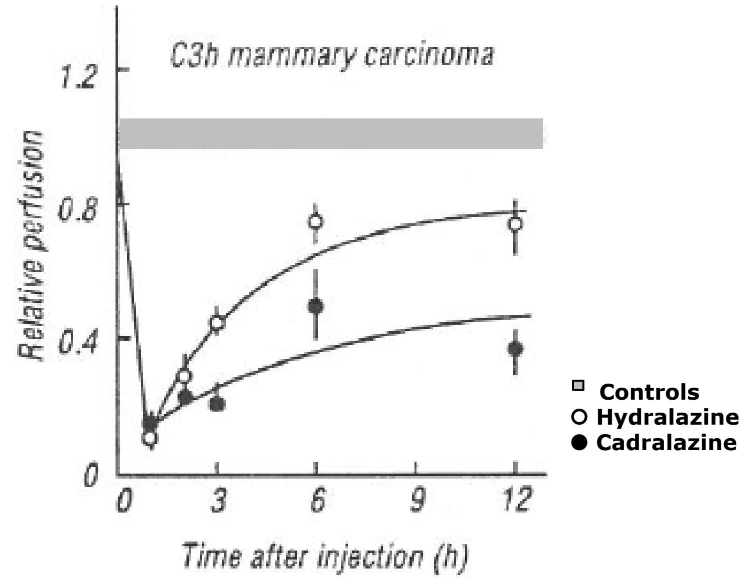


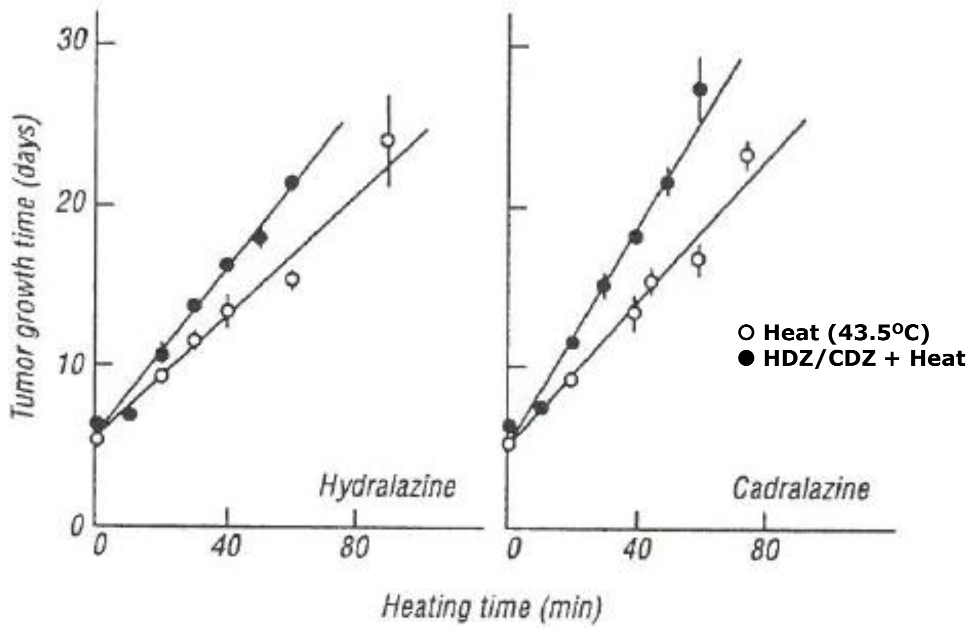
Coss et al. (2014) Int. J. Hyperthermia 30:75-78

Transient modifiers of tumour blood flow

- Hydralazine/Cadralazine
- Nitroprusside
- 5-hydroxytryptamine
- Noradrenaline
- Angiotensin II
- Anaesthetics
- Glucose
- Hb-O_2 affinity modifiers
- Embolizing agents
- Certain drugs
- NO inhibitors
- Bioreductive drugs
- Chemical radiosensitizers

Hydralazine and Cadralazine (5 mg/kg; i.v.)





Horsman et al. (2000)
Expt. Oncol. 22:32-37

Vascular targeting agents

AIs

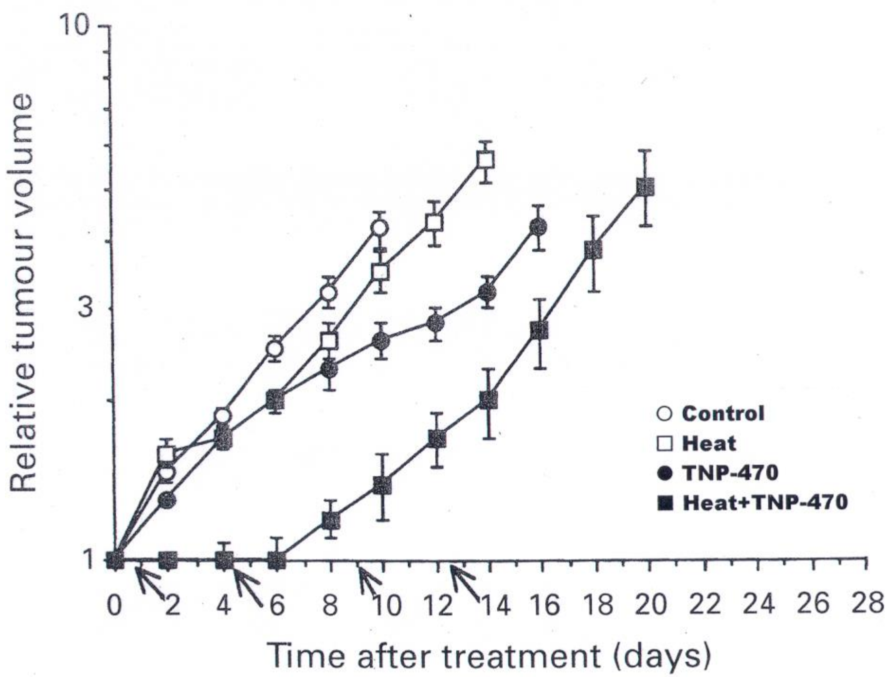
- TIMP
- Thalidomide
- Suramin and analogues
- Fumagillin and TNP470
- Cytokines
- CAI
- Endostatin
- Angiostatin
- Thrombospondin
- Arginine Deiminase
- Anginex
- Anti-VEGF(R) Ab
- Bay 43-9006 (Sorafenib)
- SU5416
- SU6668
- SU11248 (Sunitinib)
- PTK787/ZX 222584
- ZD6474
- EGFR inhibitors
- COX-2 inhibitors
- Chemotherapy

VDAs

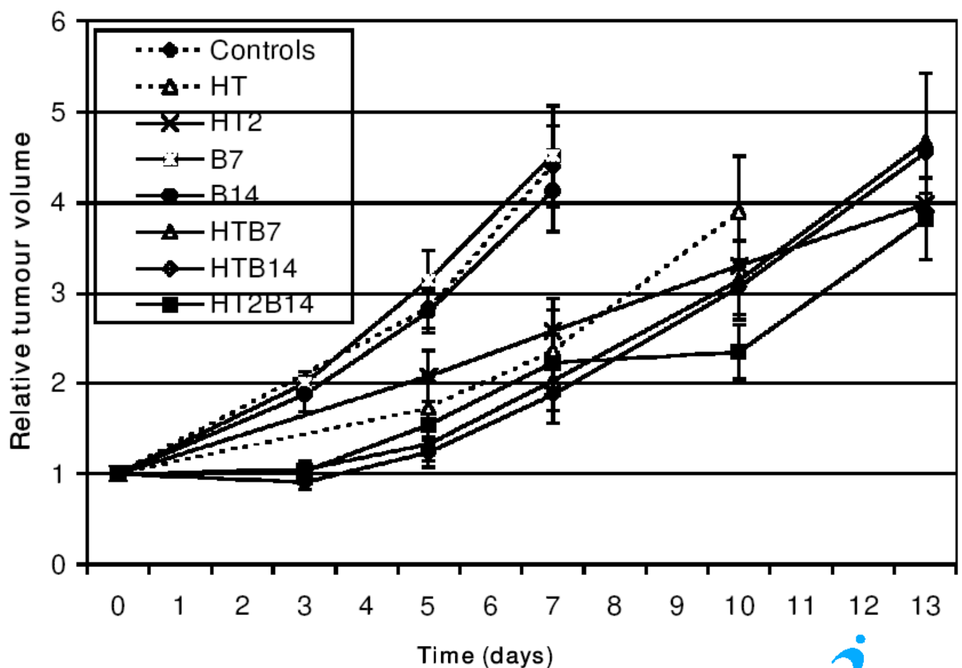
- Hyperthermia
- Photodynamic therapy
- LAK cell therapy
- Tumour necrosis factor
- Interleukins
- Interferon-gamma
- Vinka alkaloids
- Colchicine
- Arsenic trioxides
- Dolastatins
- FAA
- DMXAA
- CA4DP
- AVE8062
- ZD6126
- OXi-4503
- MN-029
- NPI-2358
- Ligand-based approaches
- (Ab, peptides, growth factors)
- Radiation

Combination studies with AIs and hyperthermia

AIA	Tumour	Heating	Reference
TNP-470	ESO-2 human oesophageal	43^{O}C	Yano et al (1995)
	NSC-8 human gastric cancer	43^{O}C	Yano et al (1995)
	SCCVII mouse carcinoma	42 – 44^{O}C	Nishimura et al (1996)
Batimastat	BT_4An rat glioma	44^{O}C	Eikesdal et al (2002)



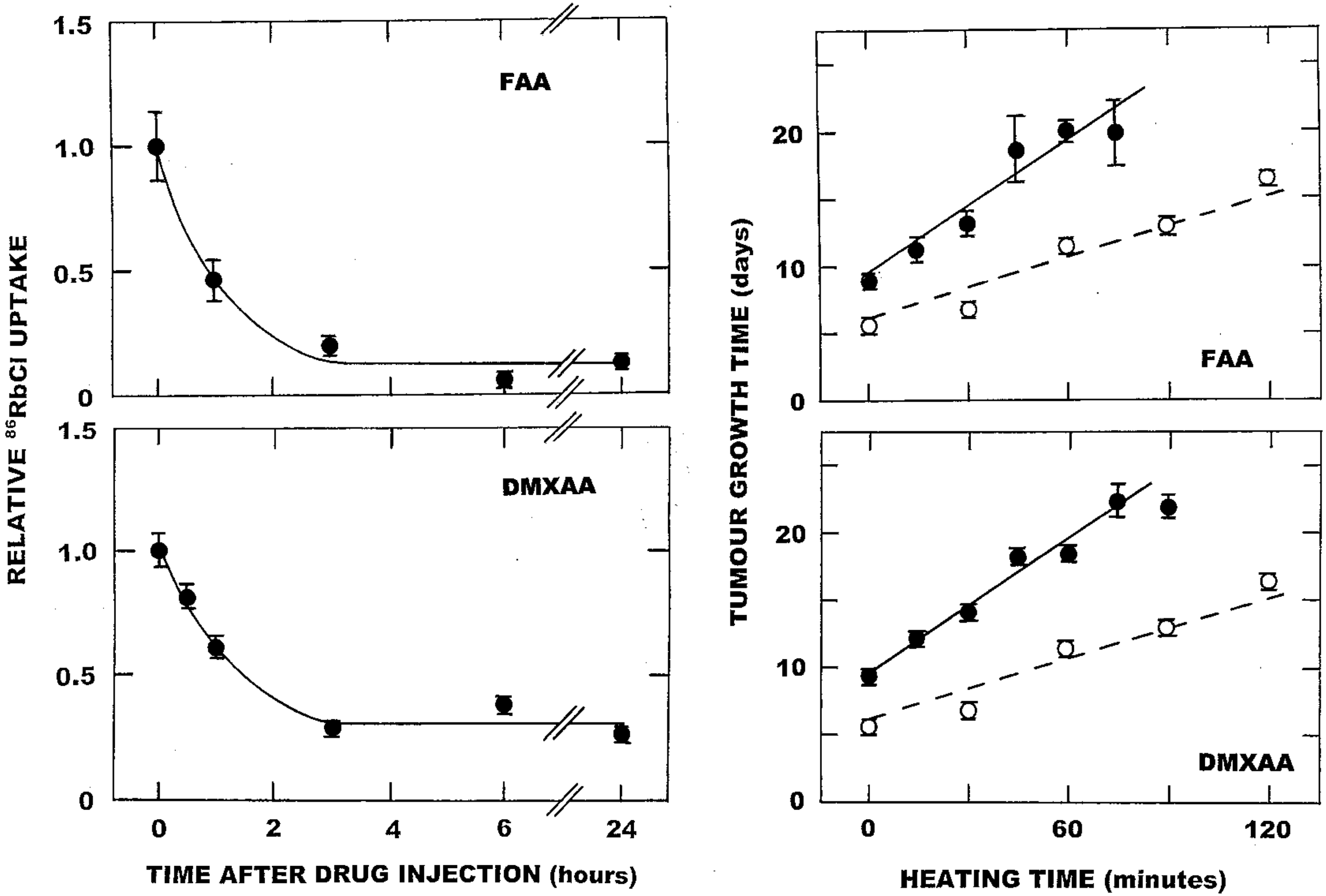
Nishimura et al. (1996) BJC 73:270-274



Eikesdal et al. (2002) IJH 18:141-152

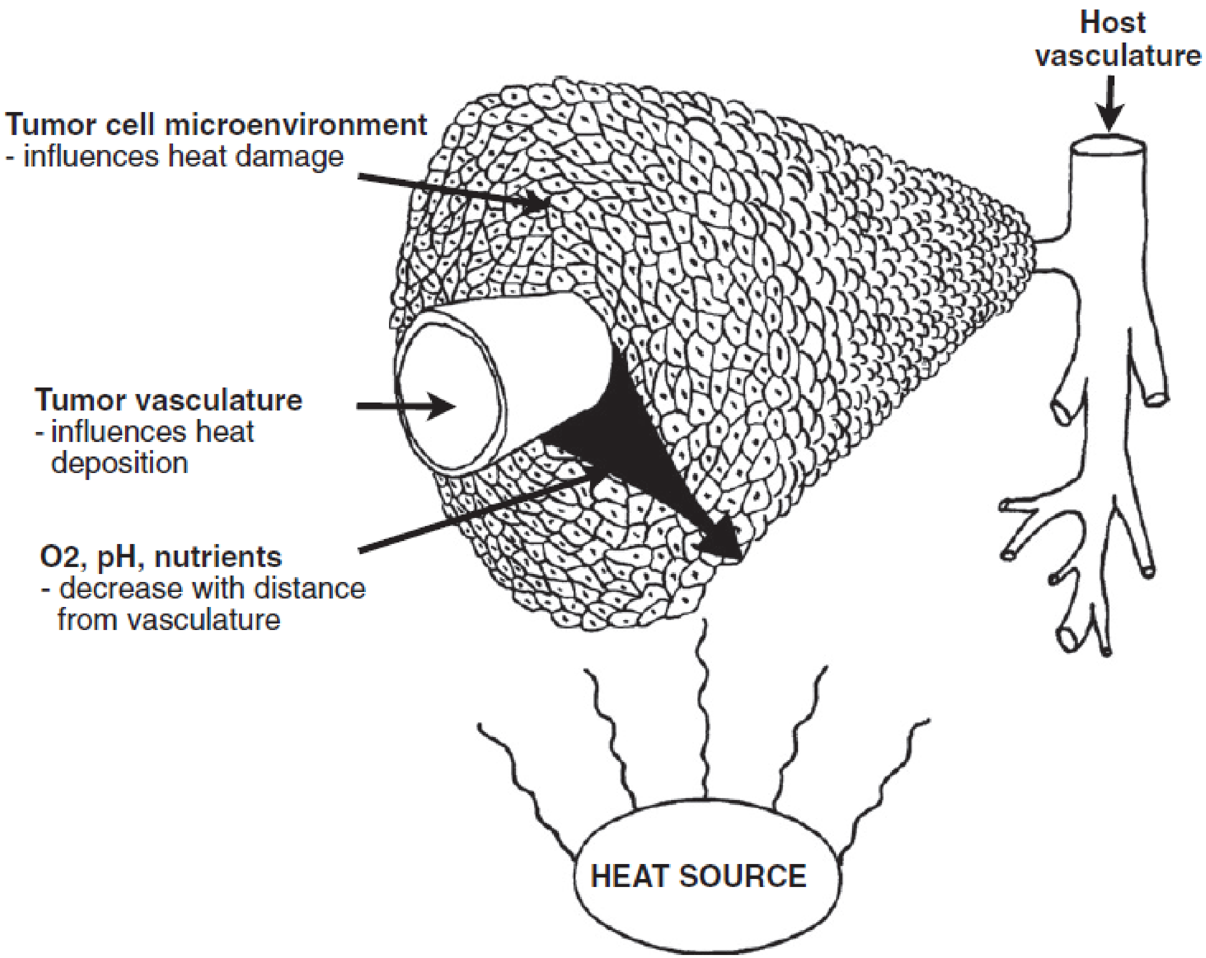
Combination studies with VDAs and hyperthermia

VDA	Tumour	Heating	Reference
TNF	DS-carcinosarcoma	43 + 44^{O}C	Kallinowski (1989)
	SCK mammary carcinoma	42.5^{O}C	Lin (1996)
ATO	SCK mammary carcinoma	41.5 – 42.5^{O}C	Griffin (2000, 2003)
	FSaII fibrosarcoma	42.5^{O}C	Griffin (2000, 2003)
VBL	BT_4An glioma	44^{O}C	Eikesdal (2001)
FAA	C3H mammary carcinoma	40.5 – 42.5^{O}C	Horsman (1991, 1996, 2001)
	B16 melanoma	43^{O}C	Sakaguchi (1992)
DMXAA	C3H mammary carcinoma	39.5 – 42.5^{O}C	Murata (2001, 2004)
CA4P	BT_4An glioma	44^{O}C	Eikesdal (2000, 2001)
	C3H mammary carcinoma	40.5 – 42.5^{O}C	Murata (2001)
OXi4503	C3H mammary carcinoma	39.5 – 42.5^{O}C	Hokland (2007)



1.5
1.0
0.5
0
FAA
DMXAA
RELATIVE 86RbCl UPTAKE
0
2
4
6
24
TIME AFTER DRUG INJECTION (hours)
20
10
0
TUMOUR GROWTH TIME (days)
60
120
HEATING TIME (minutes)

Effect of Heat



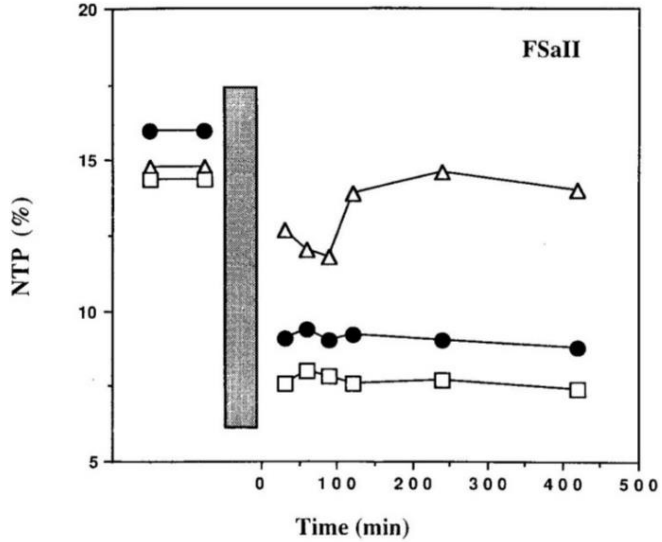
Response to Hyperthermia

Factor	Temperature	
	low	high
Tumour blood flow	↑	↓
Haemoglobin - oxygen saturation	↑	↓
Oxygen consumption	↑	↓
Energy status	↑	↓
Lactate	–	↑
pH	–	↓

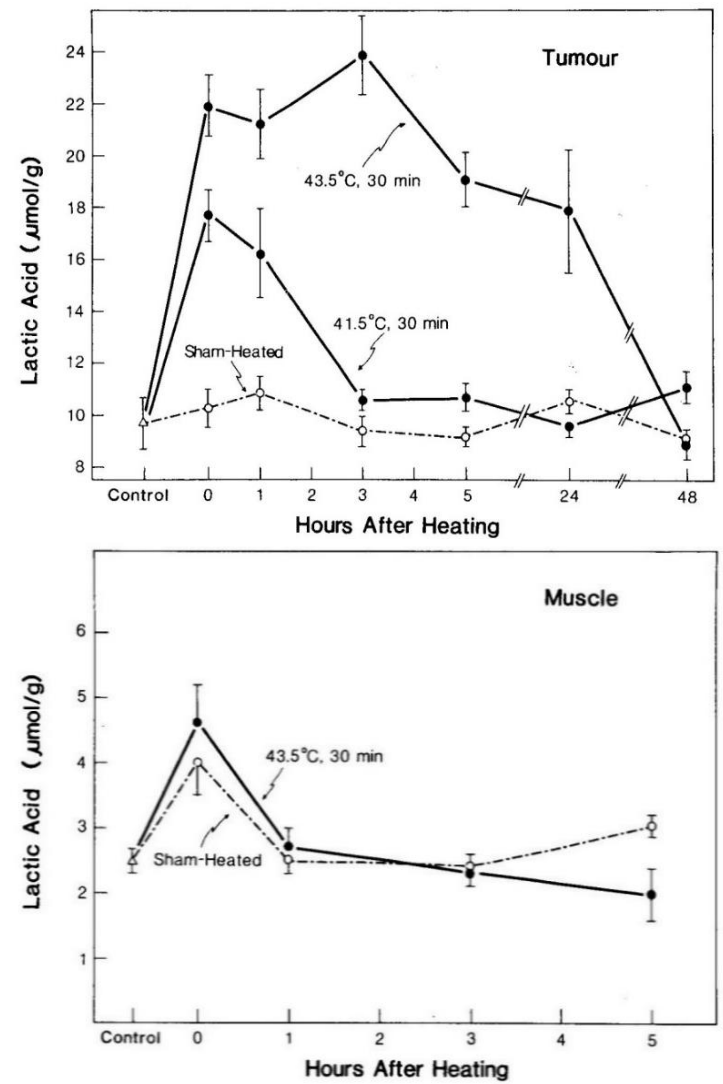
↑ increase; ↓ decrease; – unchanged

Hokland et al. (2010) Int. J. Hyperthermia 26:264-272

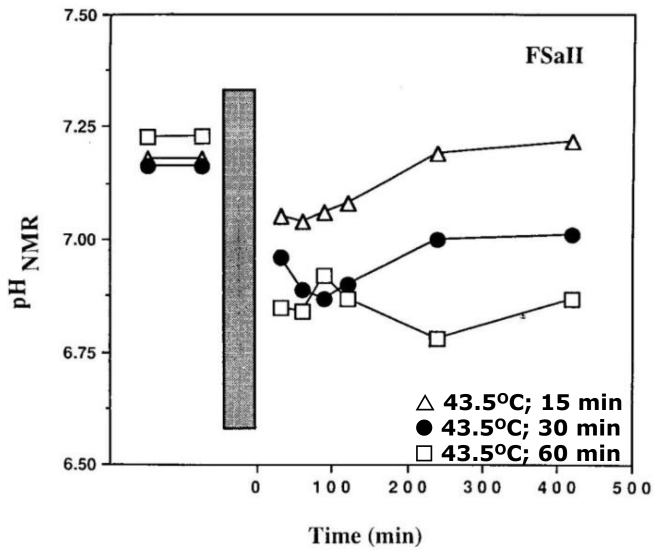
Energy status



Lactate

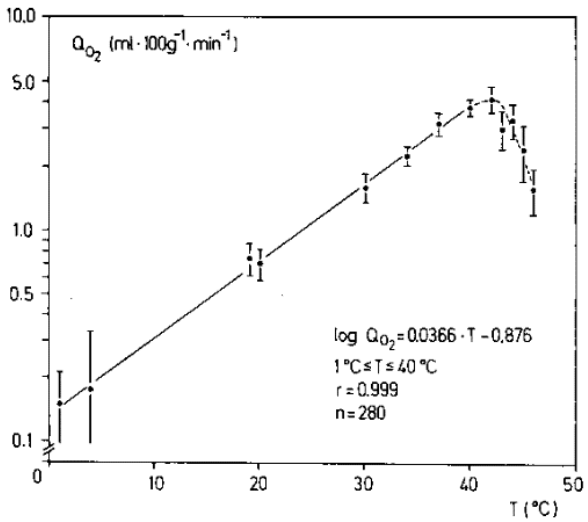


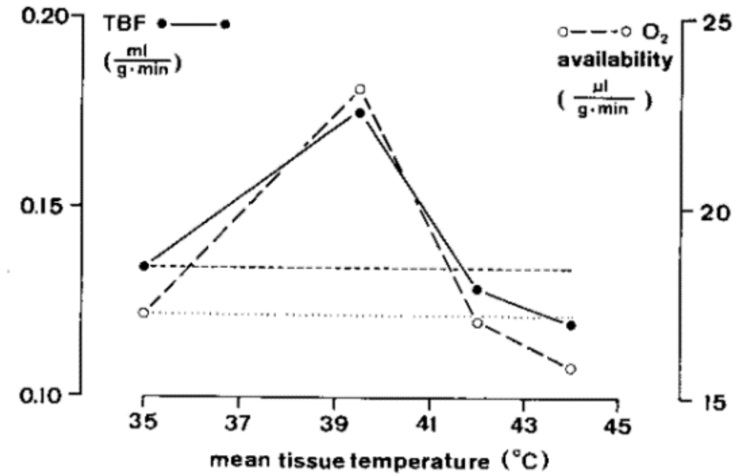
pH

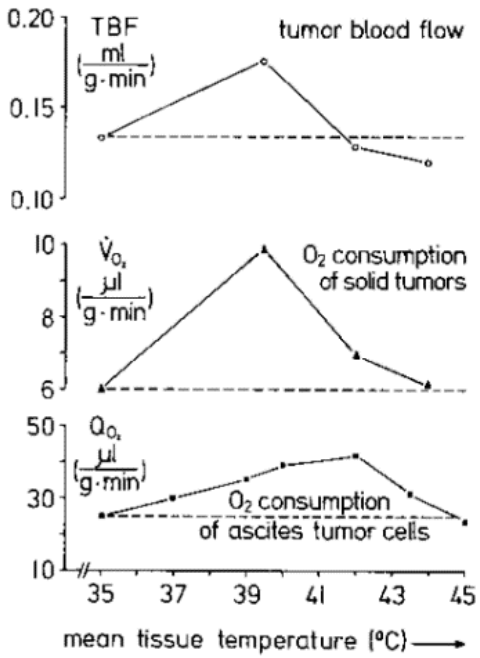


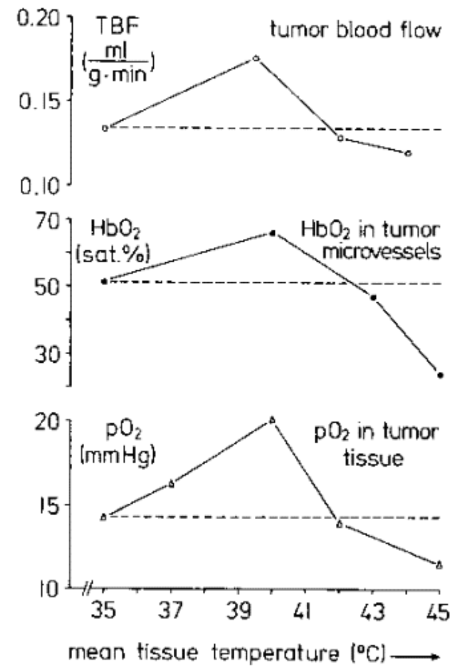
Vaupel et al. (1990) Int. J. Hyperthermia 6:15-31

Lee et al. (1986) Int. J. Hyperthermia 2:213-222



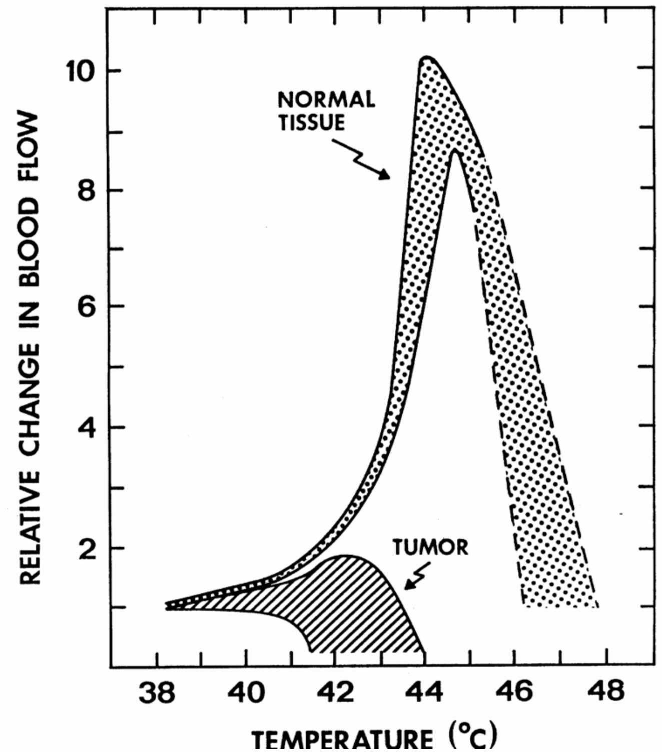




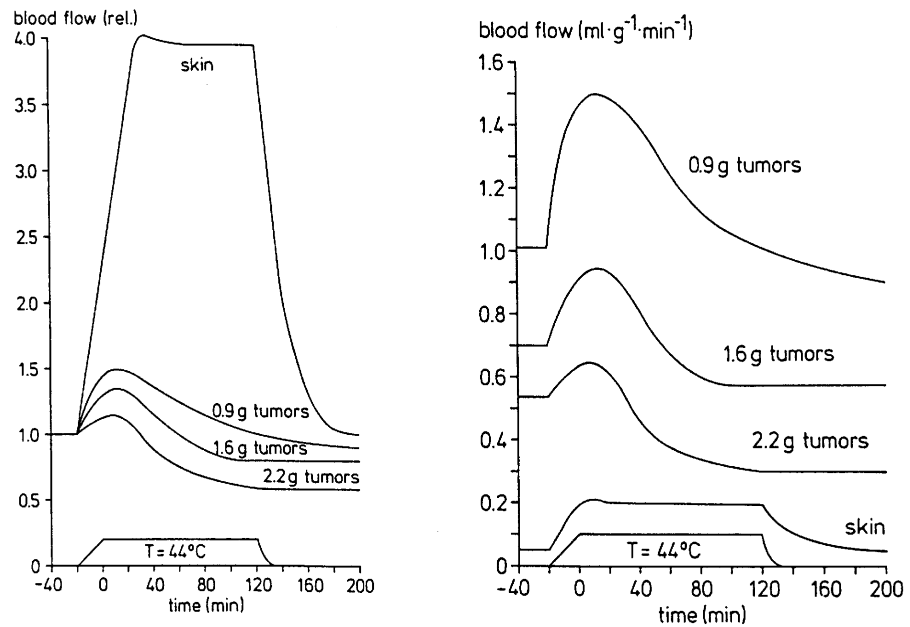


Vaupel & Kallinowski (1987) Recent Results in Cancer Res 4:71-109

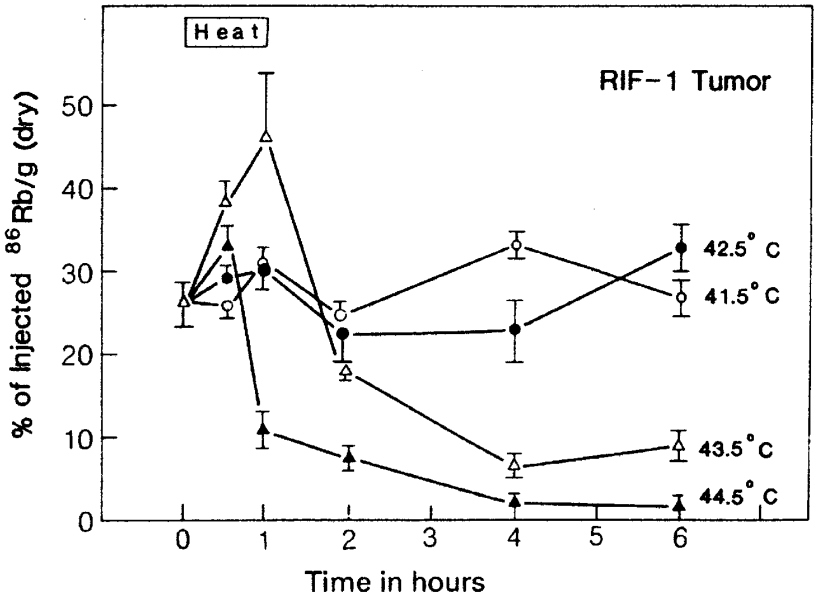
Effect of heat on tissue blood flow

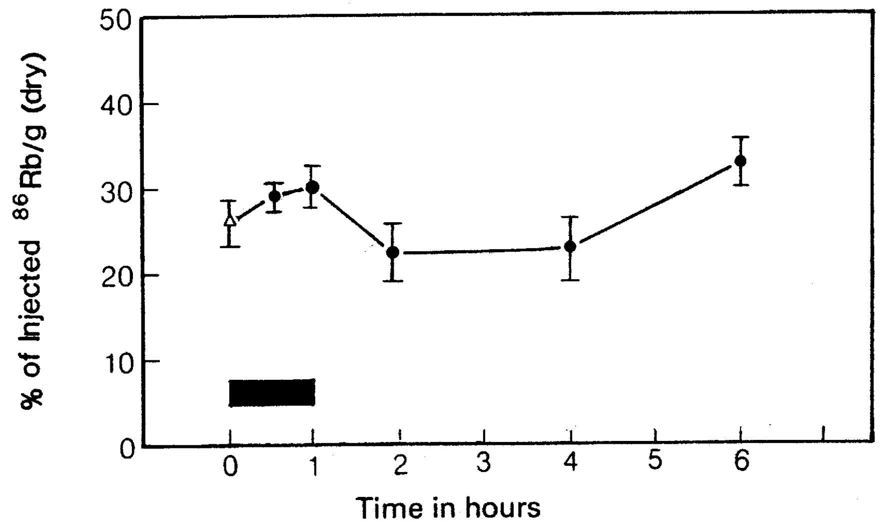


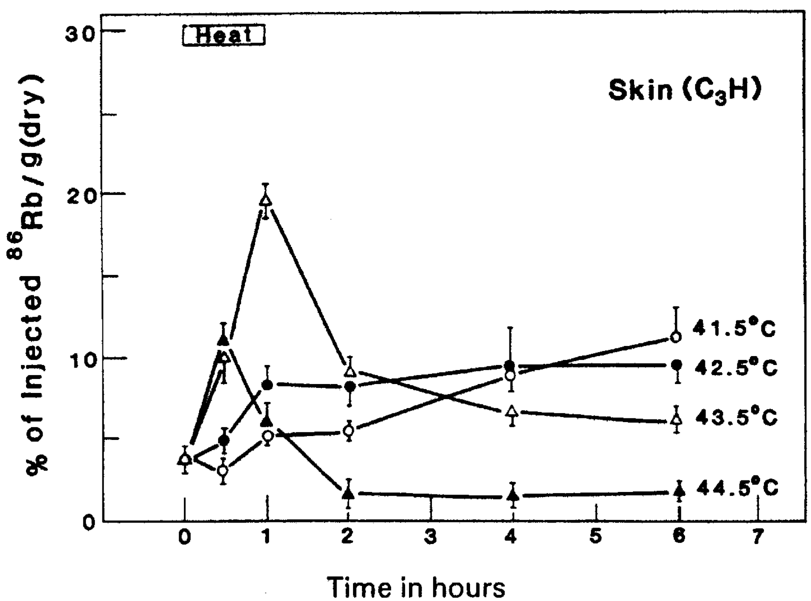
Song (1990) Cancer Res. 44:4721s-4730s



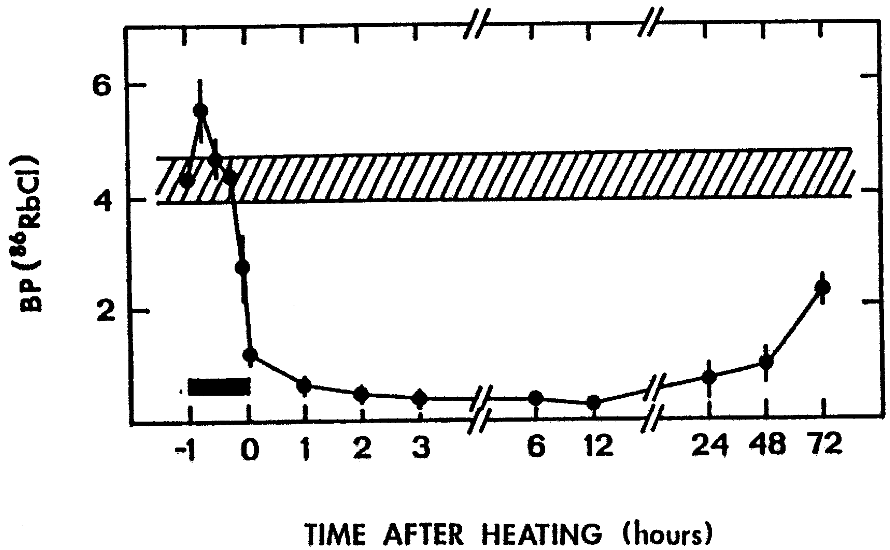
Vaupel (1993)







Song (1991)



Gyldenhof et al. (1996)
Hyperthermic Oncology 780-782

Hyperthermia and tumor reoxygenation

INT. J. HYPERTHERMIA, 1995, VOL. 11, NO. 3, 315–322

Review
Eugene Robertson Special Lecture
Hyperthermia from the clinic to the laboratory: a hypothesis

J. R. OLESON†

Department of Radiation Oncology, University of Utah Health Sciences Center, 50 N Medical Drive, Salt Lake City, UT 84132, USA

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Recently reported thermal isoeffective dose–response relationships in human tumours confirm the existence of an effect of hyperthermia in combination with radiotherapy. The prognostically important thermal doses are based upon the lowest temperatures achieved within tumours, and these thermal doses are well below those used in most laboratory studies that have provided the rationale for hyperthermia treatment. Direct thermal cytotoxicity and thermal radiosensitization are insignificant at these low thermal doses. Other explanations for the mechanism of hyperthermia effect appear warranted. We hypothesize that hyperthermia at low thermal doses causes reoxygenation and hence direct radiosensitization *in vivo*.

Key words: Hyperthermia, thermal dose, hypoxia, reoxygenation

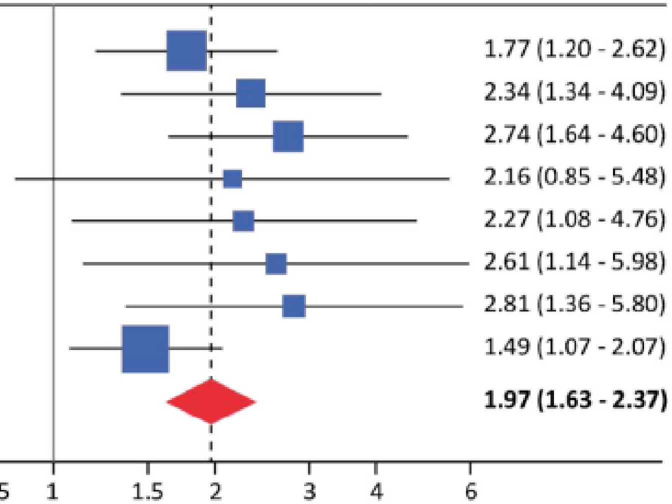
1. Introduction

Cytotoxic and radiosensitizing effects of hyperthermia (HT) reported *in vitro* and in laboratory animal tumours for fractionated HT (43–44°C for 30–60 min) and fractionated radiotherapy (RT) do not explain the recently reported thermal dose–effect relationships in human tumours (Oleson *et al*. 1993). We hypothesize that the most significant effect of HT as it has been used in human tumours is in producing indirect radiosensitization through tumour reoxygenation. We exclude from consideration here the consideration of continuous HT (<41°C) with low dose-rate brachytherapy for which combination there is a well-defined biological effect apart from the oxygenation status of cells (Armour *et al*.1991, Spiro *et al*. 1991). In this paper we review evidence leading to the above hypothesis.

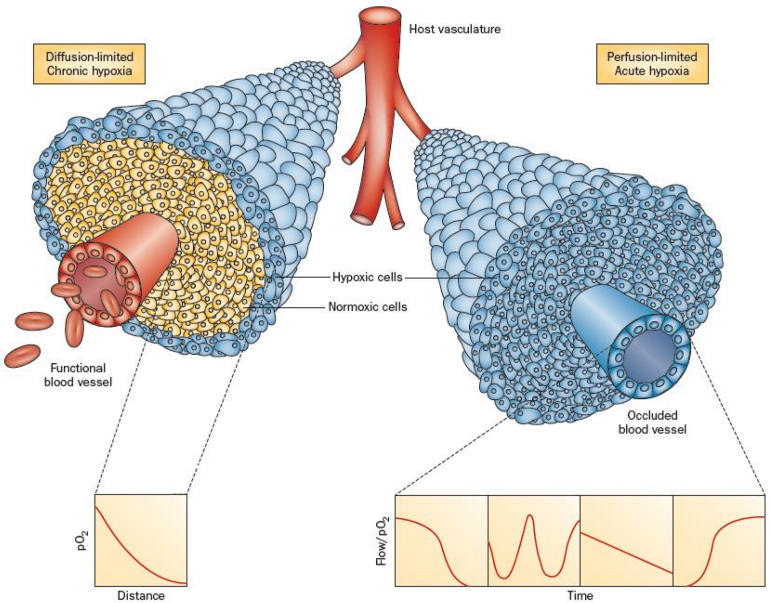
Meta-analysis of randomised clinical trials of radiation (RAD) + hyperthermia (HEAT)

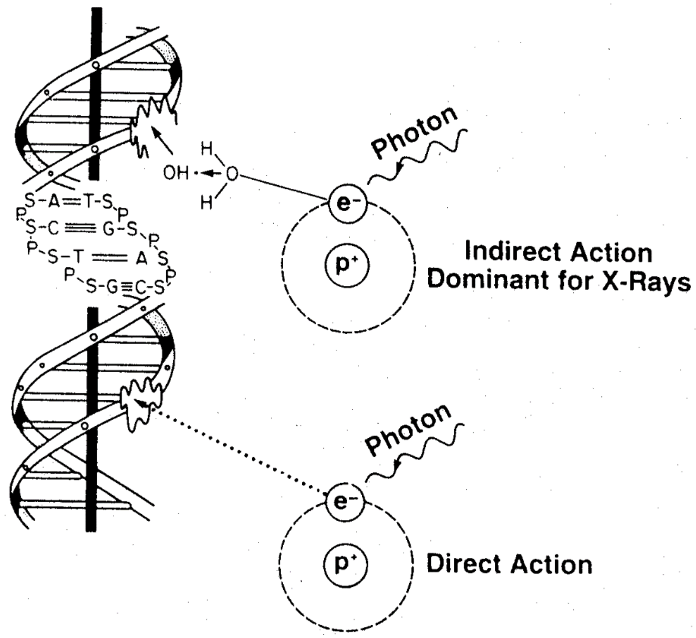
Tumor site	Trials	All pt.	RAD+HEAT Failure	RAD+HEAT All	RAD+HEAT Without failure	RAD ALONE Failure	RAD ALONE All	RAD ALONE Without failure	Endpoint: CR/loco-regional control Odds ratio (95% CI)
BREAST	6	419	86	229	62 %	98	190	48 %	1.77 (1.20 - 2.62)
HEAD & NECK	4	207	45	104	57 %	66	103	36 %	2.34 (1.34 - 4.09)
UTERINE CERVIX	4	249	41	125	67 %	71	124	43 %	2.74 (1.64 - 4.60)
LUNG	1	80	11	40	73 %	18	40	55 %	2.16 (0.85 - 5.48)
RECTUM	2	258	104	128	19 %	118	130	9 %	2.27 (1.08 - 4.76)
BLADDER	1	101	14	52	73 %	24	49	51 %	2.61 (1.14 - 5.98)
MELANOMA	1	128	28	63	56 %	45	65	31 %	2.81 (1.36 - 5.80)
MIXED	4	610	177	306	42 %	204	304	33 %	1.49 (1.07 - 2.07)
OVERALL	**23**	**2052**	**506**	**1047**	**52 %**	**644**	**1005**	**36 %**	**1.97 (1.63 - 2.37)**

I-squared = 0.0%, p = 0.48



Elming et al. (2019) Cancers





Direct effect

$RH \rightarrow RH^+ + e^-$

$RH^+ \rightarrow R\cdot + H^+$

Indirect effect

$H_2O \rightarrow H_2O^+ + e^-$

$H_2O^+ \rightarrow H^+ + OH\cdot$

$RH + OH\cdot \rightarrow R\cdot + H_2O$

In absence of oxygen or in presence of –SH

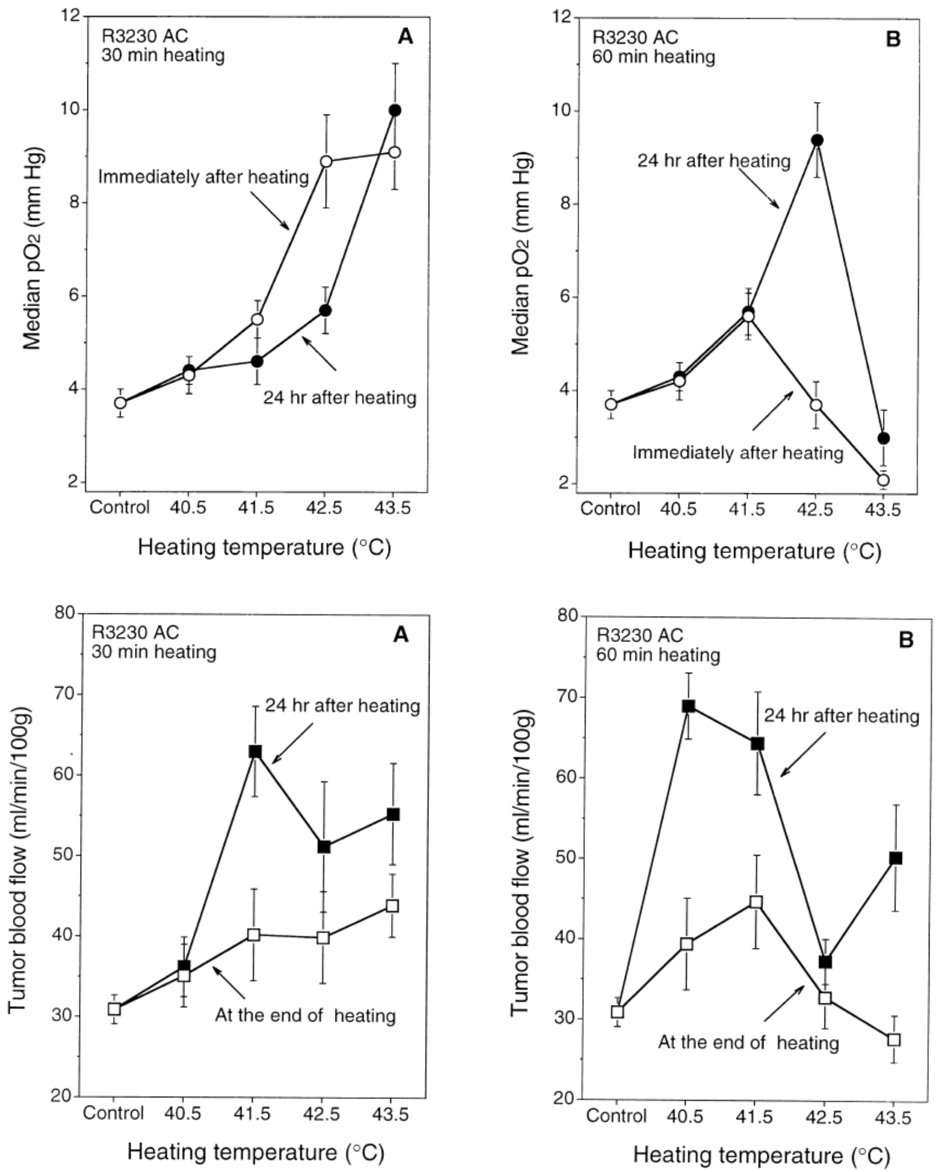
$R\cdot + SH \rightarrow RH + S\cdot$

(Target restitution)

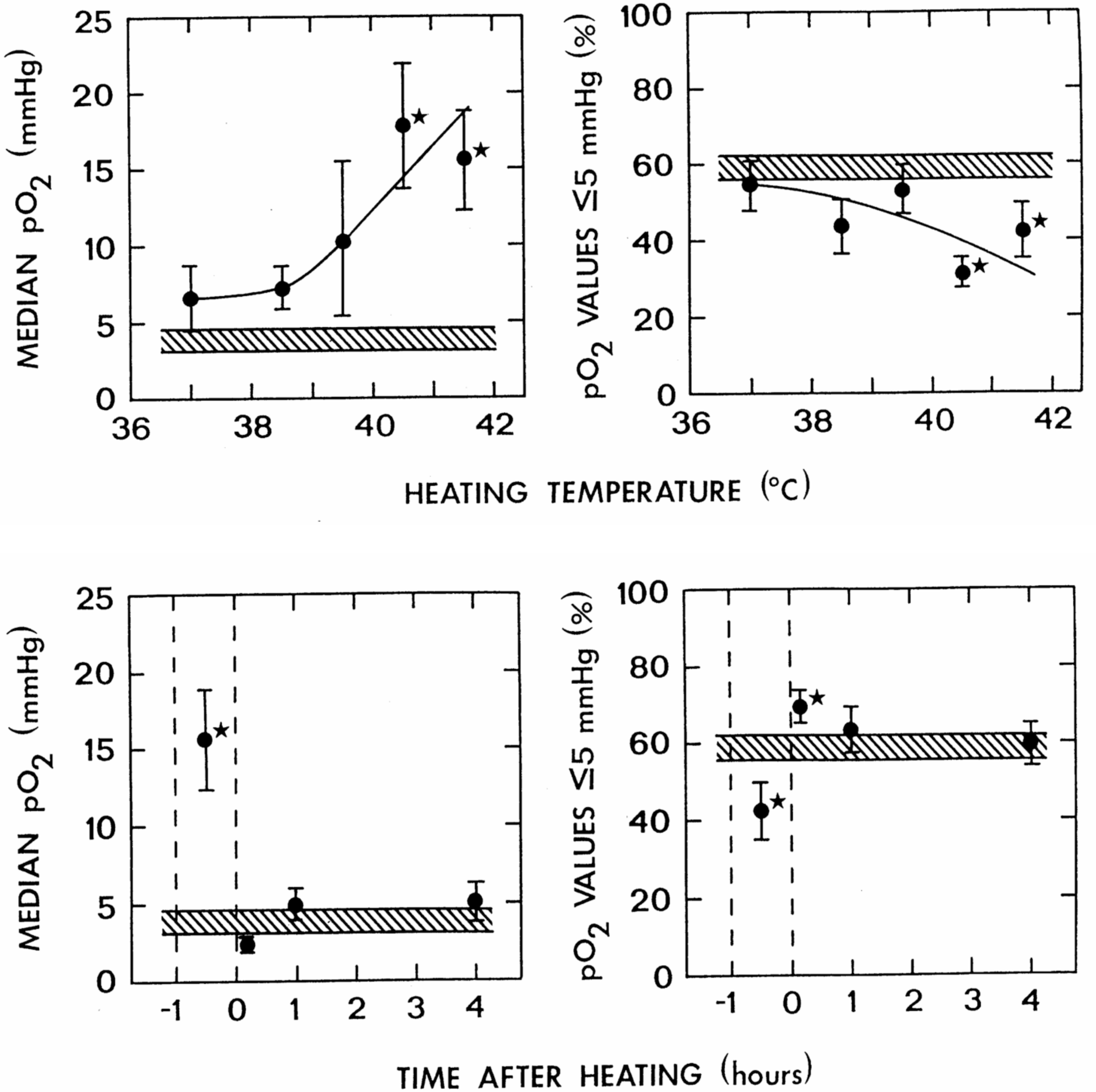
In presence of oxygen

$R\cdot + O_2 \rightarrow RO_2\cdot \rightarrow ROOH$

(Oxygen fixation hypothesis)



Shakil et al. (1999) Int. J. Radiol. Oncol. Biol. Phys. 43:859-865



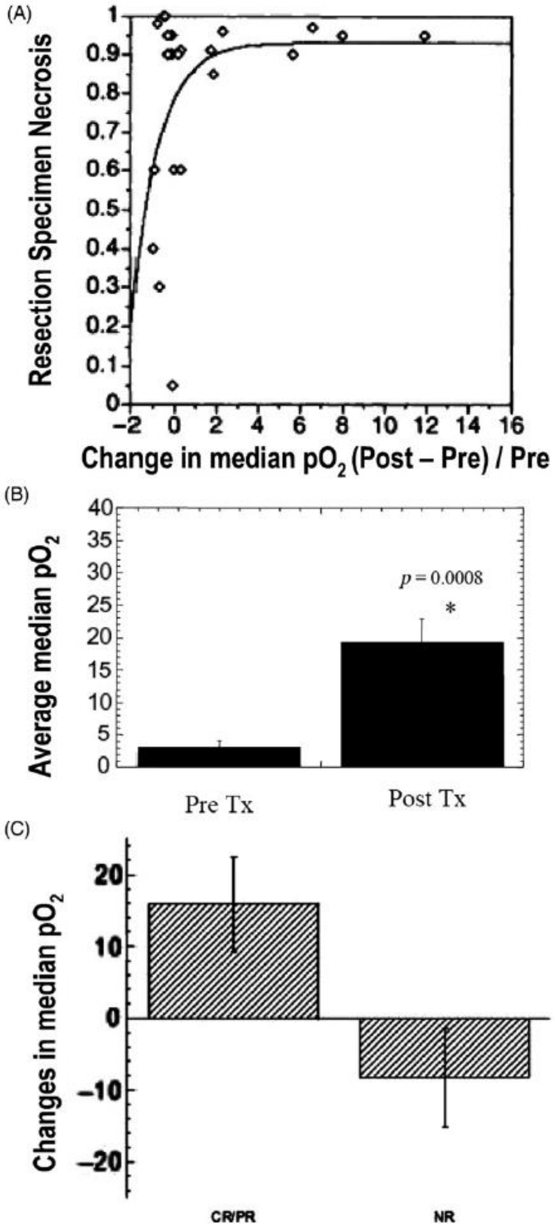
Horsman & Overgaard (1997) Int. J. Hyperthermia 13:141-147

Hyperthermia and oxygenation

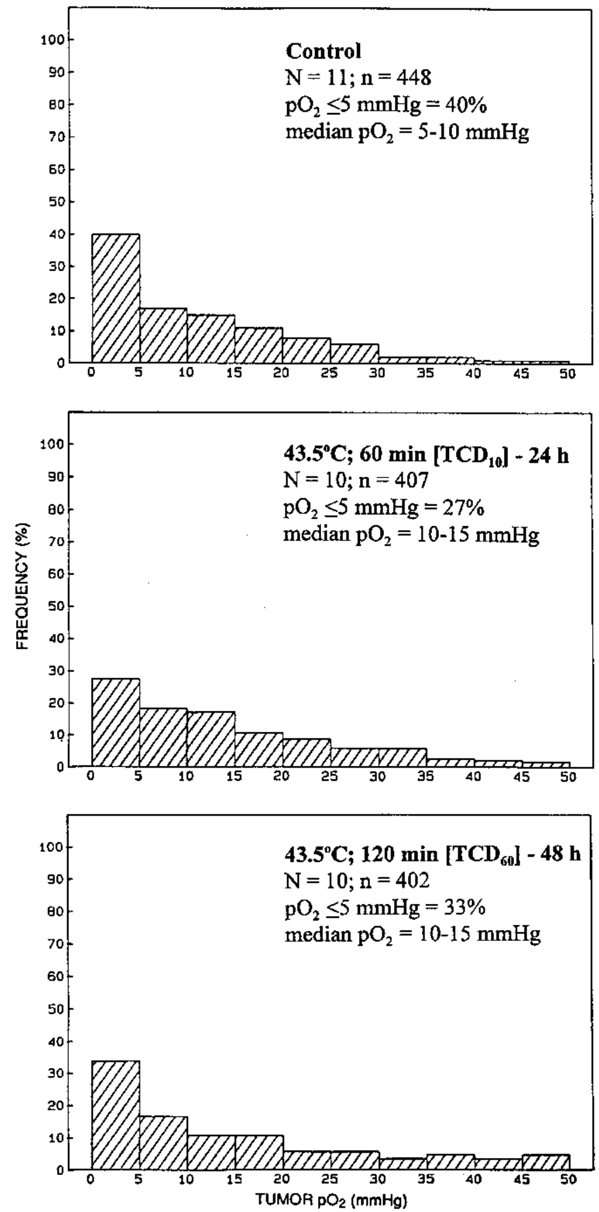
- Brizel et al. (1996) Cancer Res 56:5347-50
  - human soft tissue sarcomas
  - pO_2 increased after heating and this increase correlated with necrosis.

- Vujaskovic et al. (2003) IJH 19:498-506
  - breast cancer
  - after heating pO_2 increased in tumours that were originally hypoxic.

- Jones et al. (2004) Clin Cancer Res 10:4287-93
  - locally advanced breast cancer
  - pO_2 after hyperthermia increased in responders (CR + PR) and decreased in non-responders.

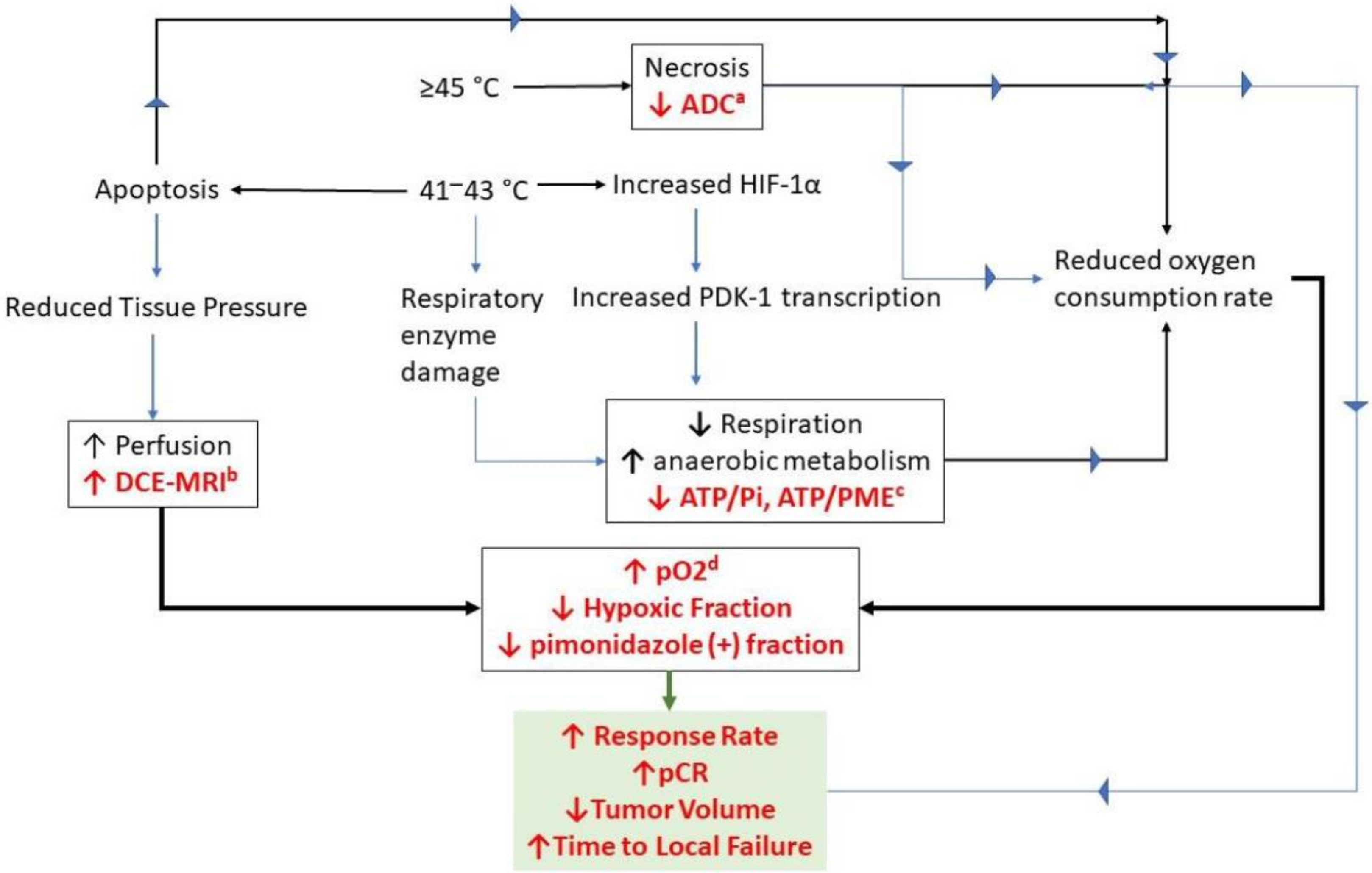


Duke University studies

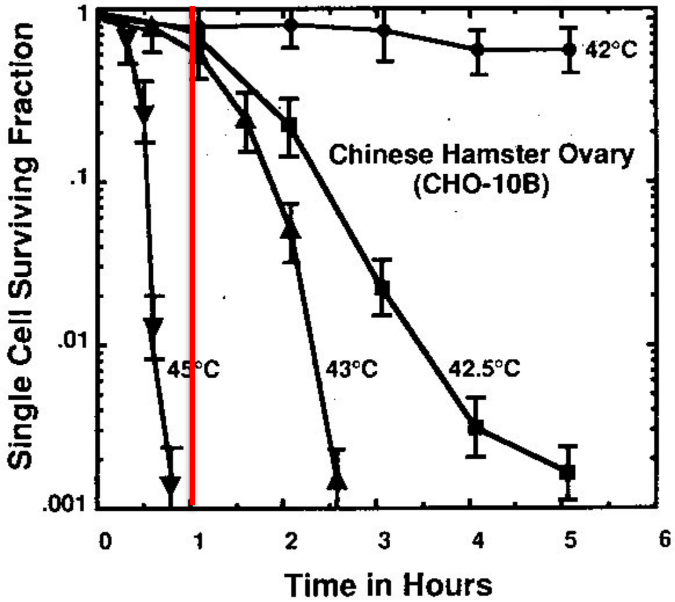


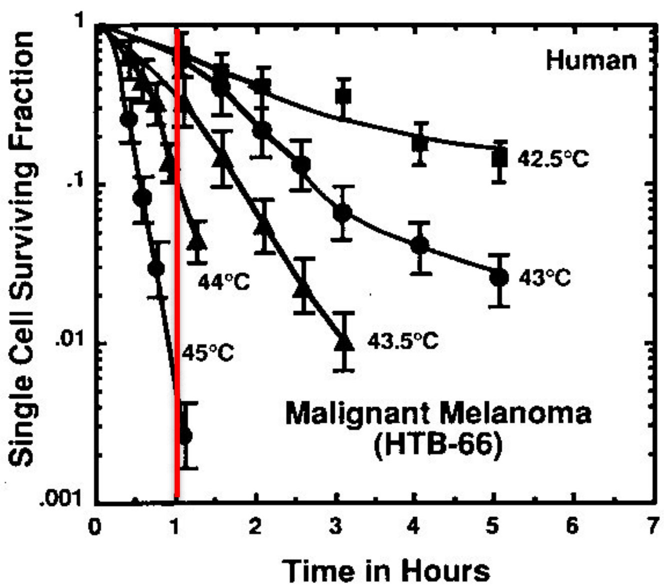
Hetzel et al (1992) Rad Res 131:152-6

Possible mechanisms for reoxygenation following heat

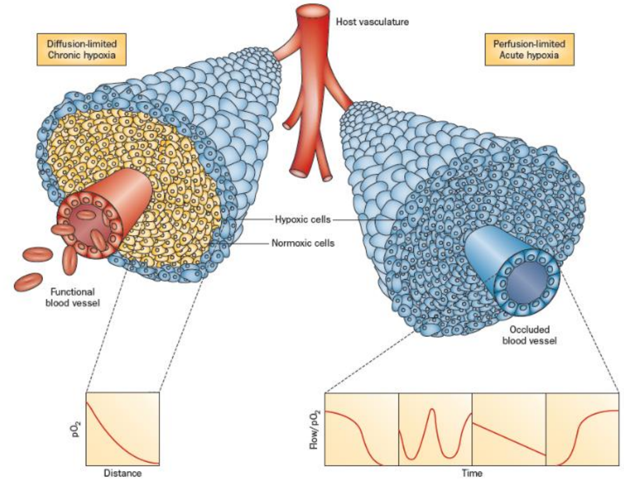


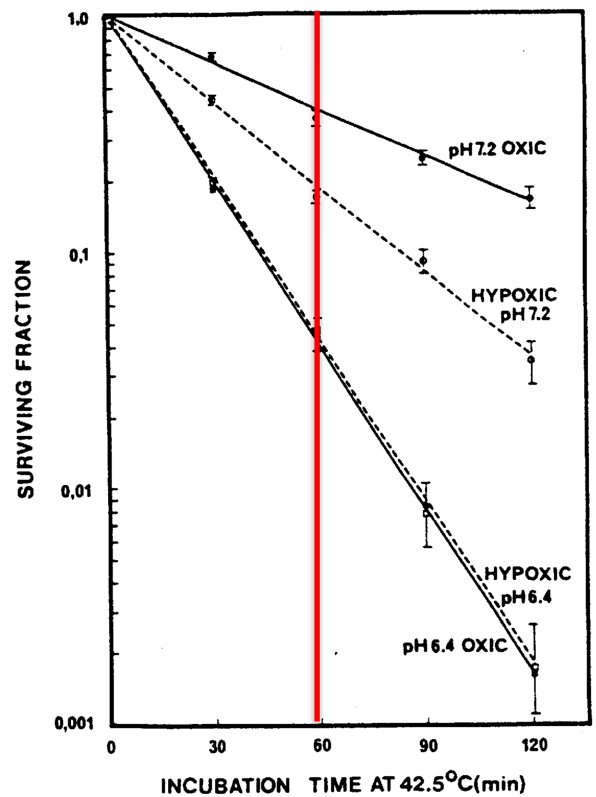
Dewhirst et al (2022) Cancers 14:1701





Roizin-Towle & Pirro (1991)
IJROBP 20:751-756





Overgaard & Bichel (1977)
Radiol. 123:511-514