Surgical Stress and the Heat Shock Response: Models of Stress Conditioning

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Overview

- Medical and Surgical Safety
  - Room for improvement
- Define Surgical Stress
- Introduce Stress Conditioning
- Hyperbaric Oxygen Therapy?

November 1999

INSTITUTE OF MEDICINE

Shaping the Future for Health

TO ERR IS HUMAN:
BUILDING A SAFER HEALTH SYSTEM

Health care in the United States is not as safe as it should be--and can be. At least 44,000 people, and perhaps as many as 98,000 people, die in hospitals each year as a result of medical errors that could have been prevented, according to estimates from two major studies. Even using

www.nap.edu/ openbook

2011- Goal of federal "Partnership for Patients" / HHS Secretary K. Sebelius

↓ errors by 40% = $10 Billion in Medicare savings

↓ deaths by 20,000/yr x 3 yrs

ELSEVIER
SAUNDERS

Neurol Clin 24 (2006) 795–806

NEUROLOGIC
CLINICS

Risk Estimates of Stroke After Coronary Artery Bypass Graft and Carotid Endarterectomy

Rima M. Dafer, MD, MPH

Department of Neurology, Stritch School of Medicine, Loyola University Chicago, 2160 South First Avenue, Maywood, IL 60153, USA

CVA post-CABG ~ 1.1 - 3.2% (up to 25%)
CVA post-CEA ~ 0.5 - 7%

Surgical Stress

Examples:

Ischemia-reperfusion

Inflammation

Hypoxia

Wounds

Radiation

Drug toxicity

Metabolic toxins

Hyper/hypothermia

Clinical Settings:

All surgery

Cardiovascular

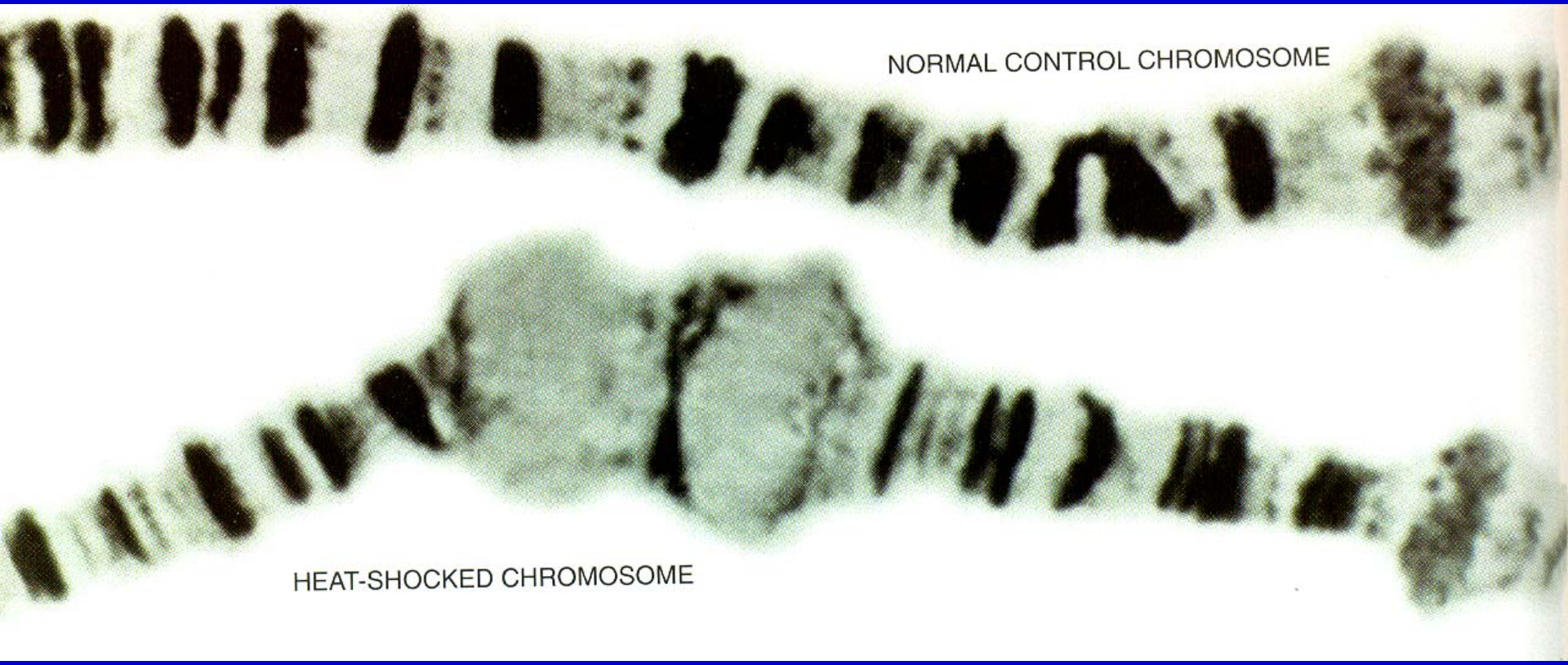
Trauma

Critical care

Transplant

Burns

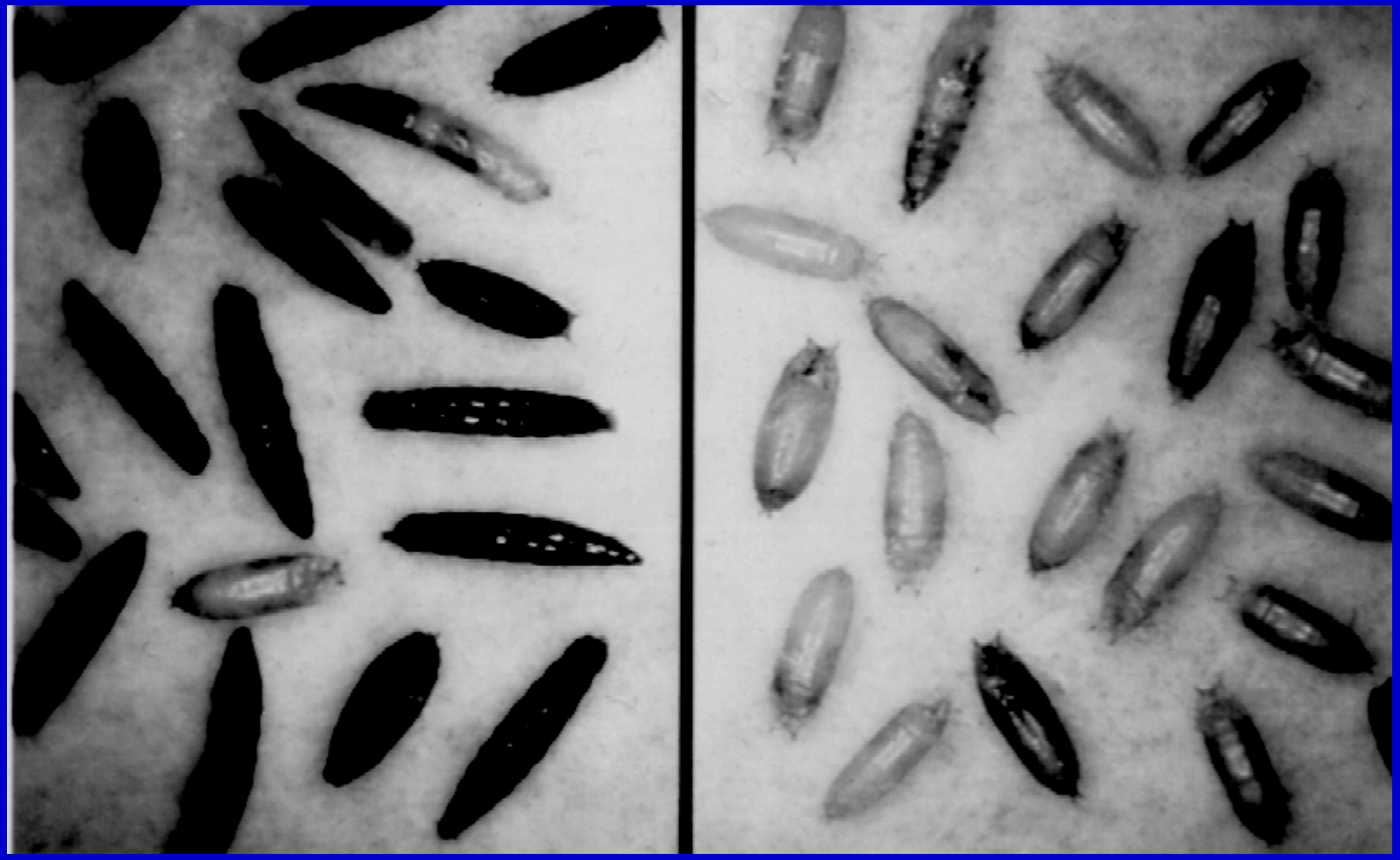
Heat Shock Response:
Birth of Molecular Biology



F. Ritossa, 1964 Italy
A. Tissieres, 1974 Switzerland
S. Lindquist, 1975 USA

Heat Shock Response-1960's

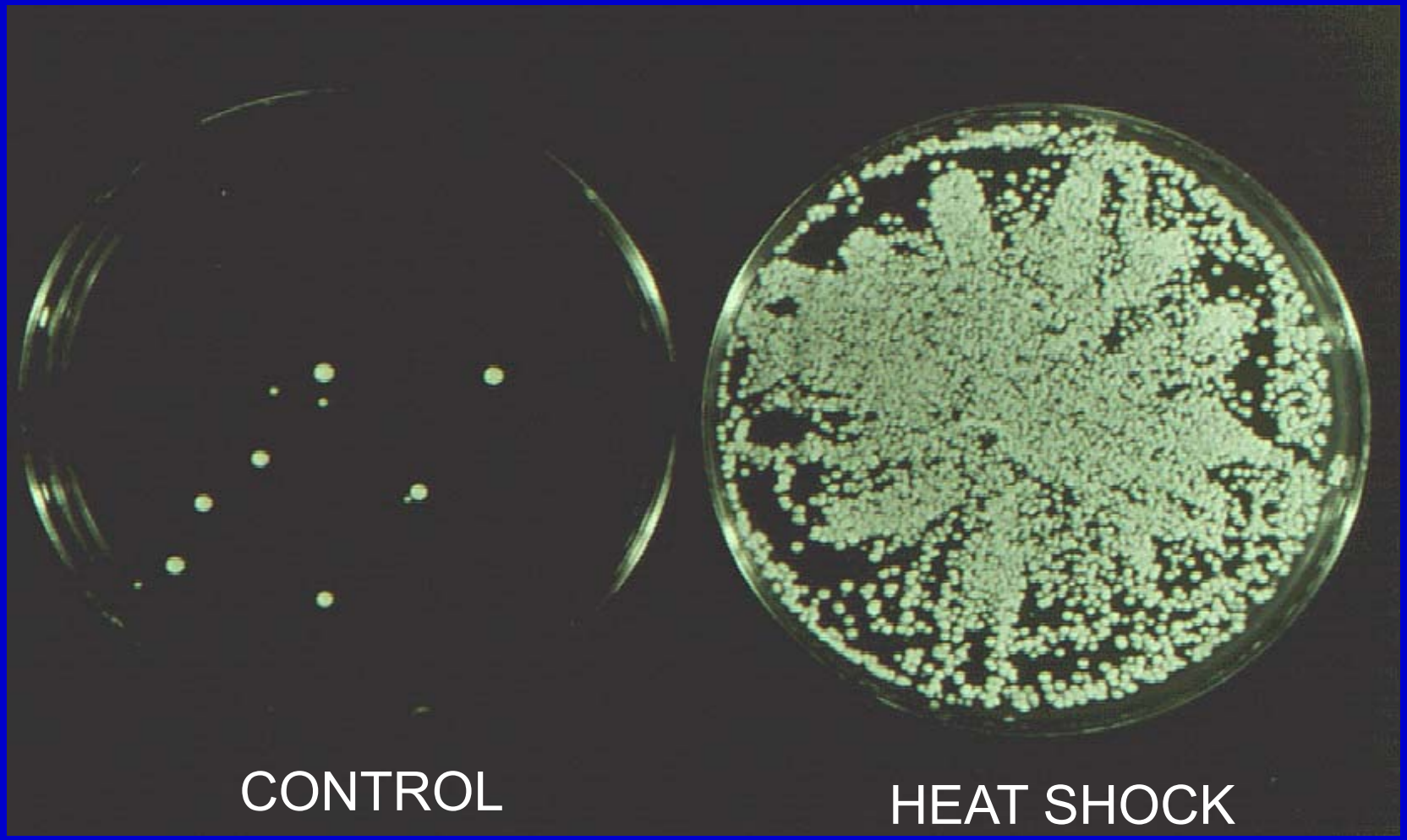
Prevents Thermal Death in *Drosophila*



CONTROL HEAT SHOCK

Thermotolerance

Yeast

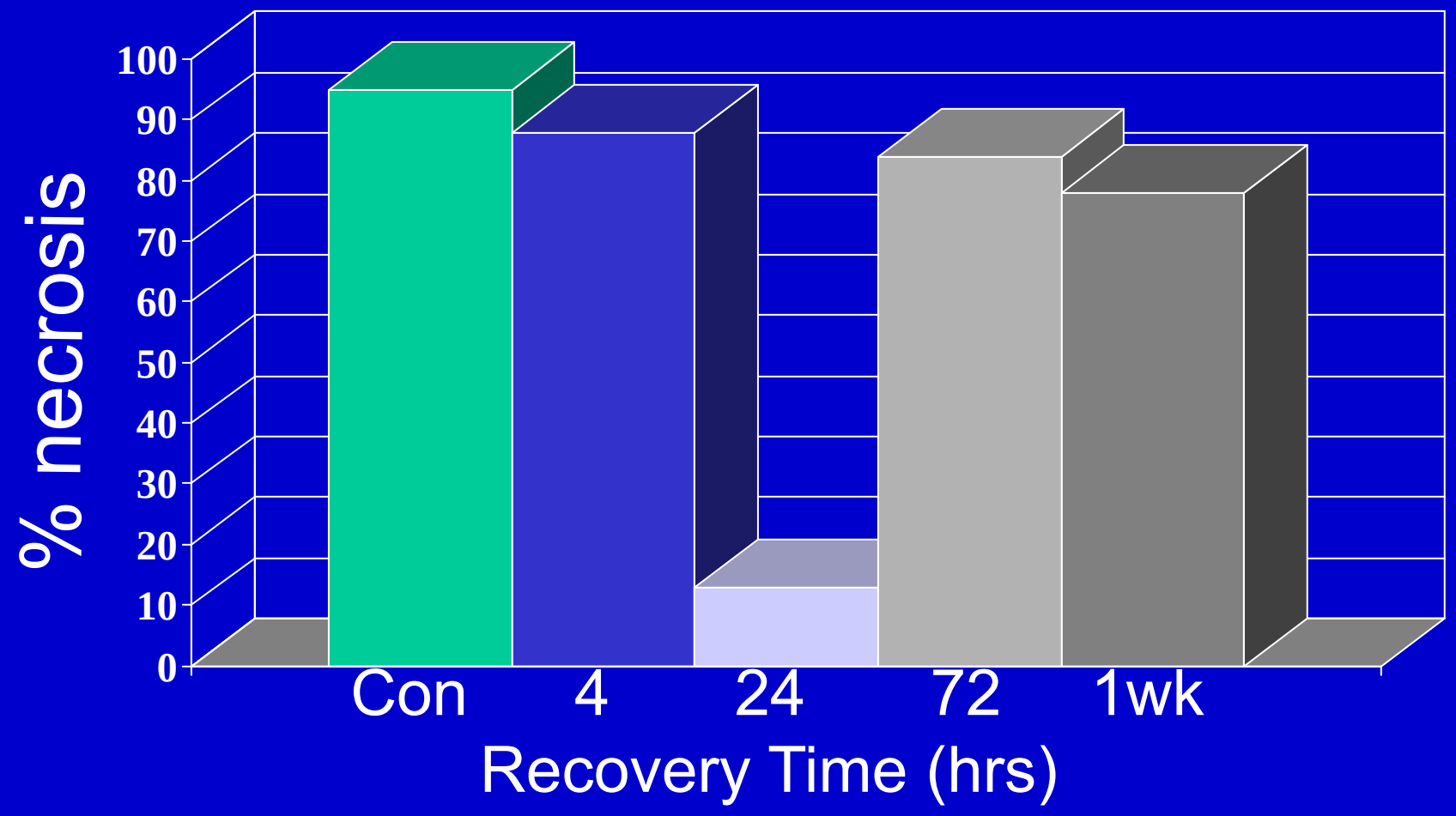


Lindquist, S., unpublished

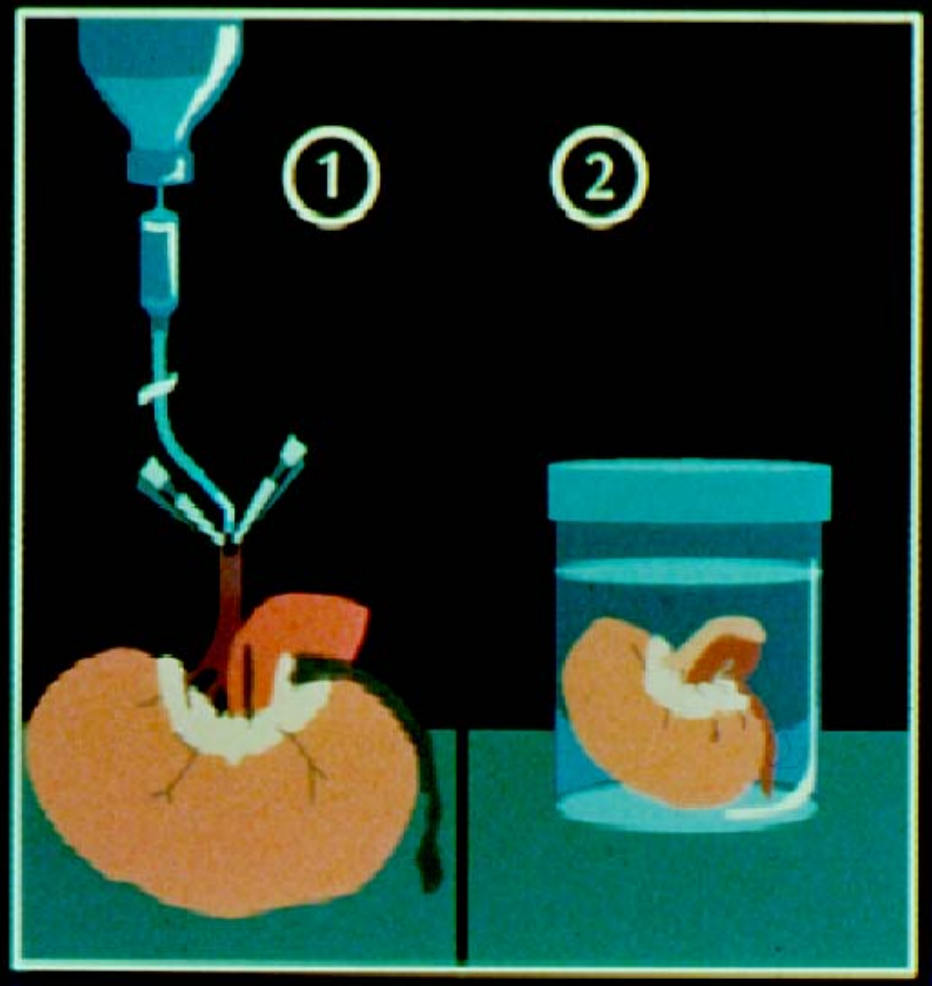
Thermotolerance:

Rodent in Vivo

Crile Jr., G. Cancer Res 1963;23:372.



Organ Preservation: A Model (1985)



1. Collins' solution
2. Simple cold storage

<u>TIME LIMITS (hrs):</u>

Heart- 5

Lung- 6-8

Liver- 24

Kidney- 48

Experimental Design:
Rodent Renal Isograft

SHAM

HEAT SHOCK (42.5 °C x 15 min)

RECOVERY (37 °C x hrs)

0 6-8 24

NEPHRECTOMY

PRESERVATION (4°C x 48hrs)

TRANSPLANTATION

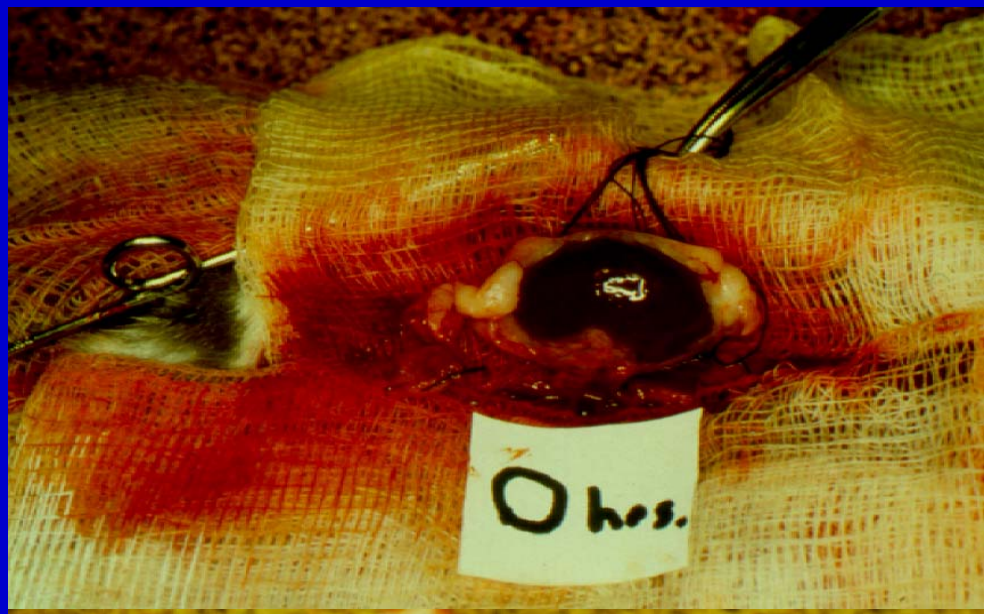
SURVIVAL 45 DAYS

Sham SC

0/7 functioned

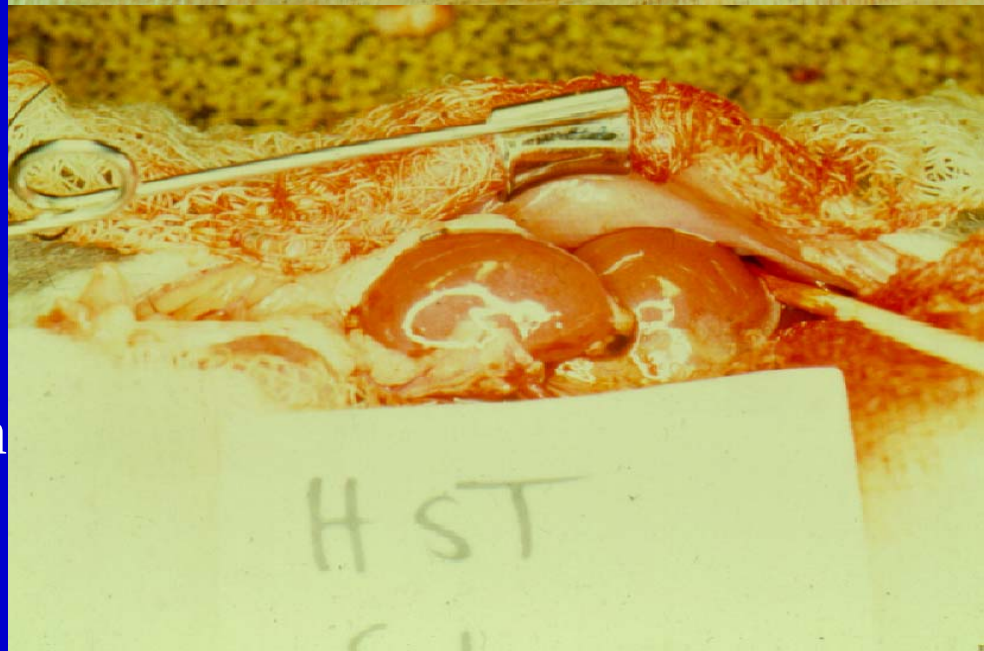
Stress/No recovery time

0/5 functioned



Stress Conditioned

4/5 normal function



Perdrizet, GA et al. Curr Surg 1989;46:23.

Heat Shock Protects Renal Isografts During Cold Storage

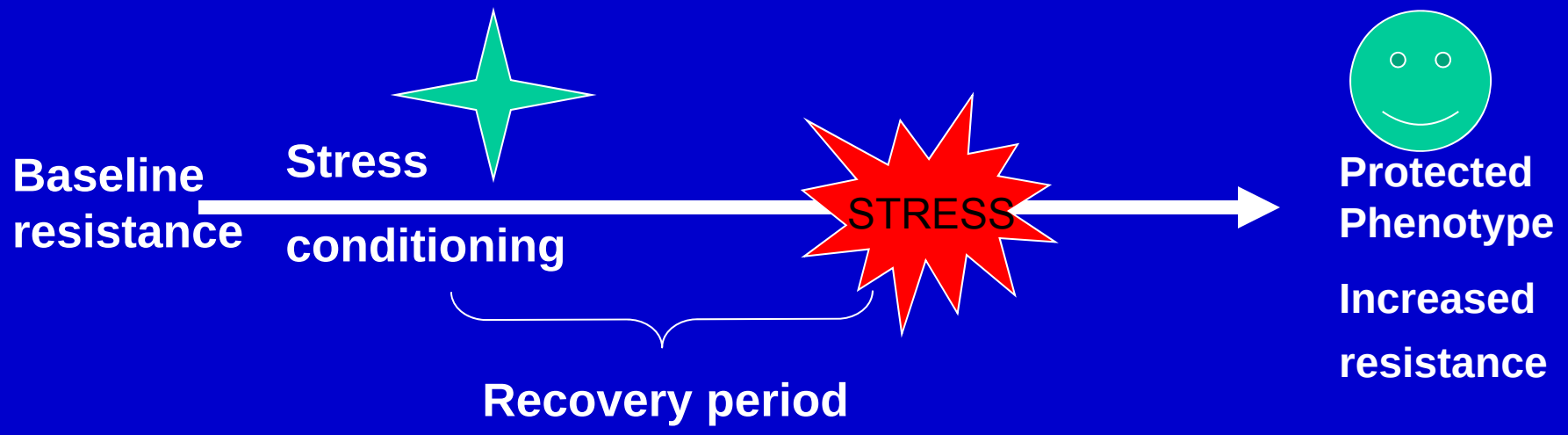
GROUP	KIDNEY GRAFTS (n)	EARLY FUNCTION (n)	SURVIVORS (n)
Sham	7	0	0
HS/0-4 hrs	5	2	0
HS/6-8hrs	5	5	4*
HS/24hrs	3	0	0

*$p < 0.002$ vs sham

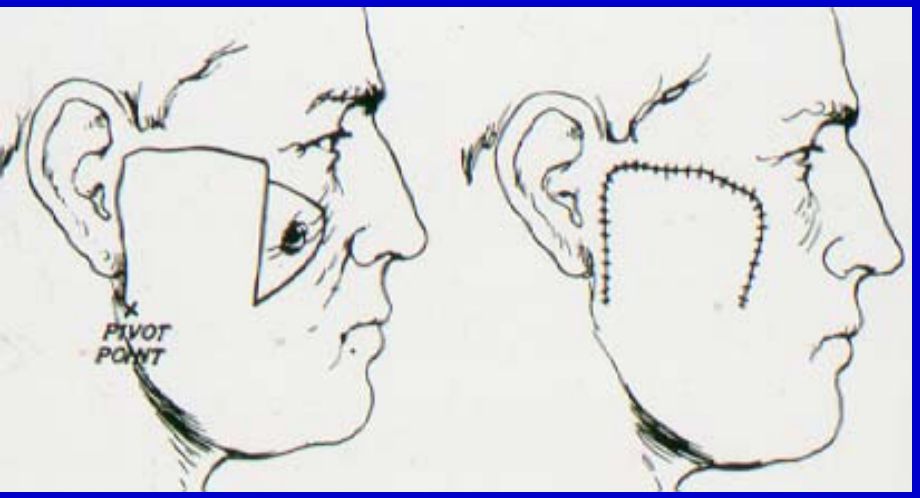
Perdrizet, GA et al. Current Surgery 1989;46:23.

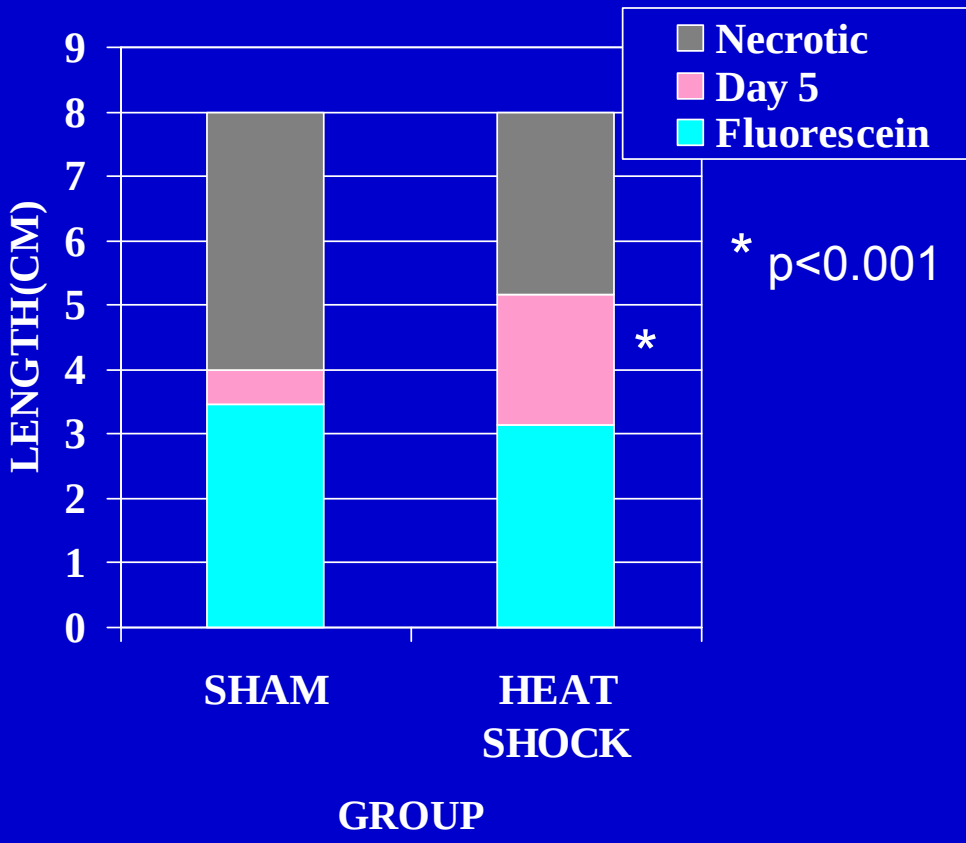
Stress Conditioning
Definition

Exposure of living cells or tissues to a <u>sublethal</u> stressor, followed by a recovery period prior to exposure to a <u>lethal</u> stressor.



Skin Flap Survival





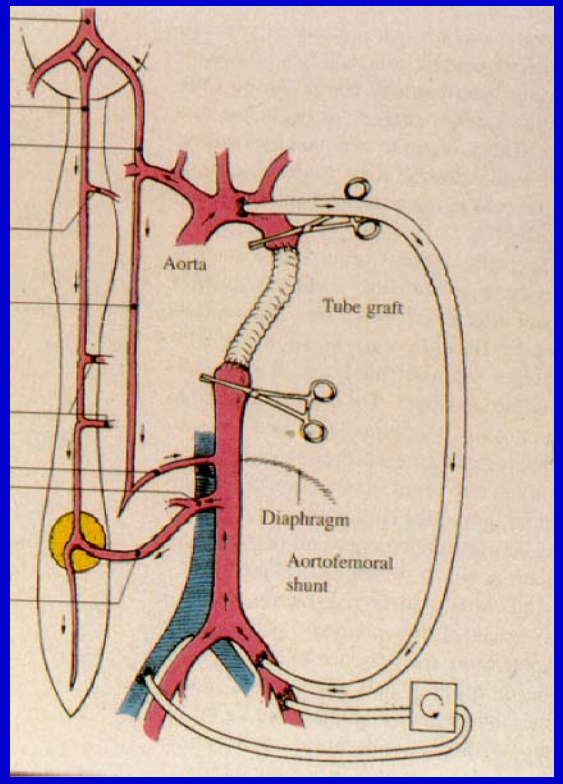
Groups: Sham vs Heat Shock

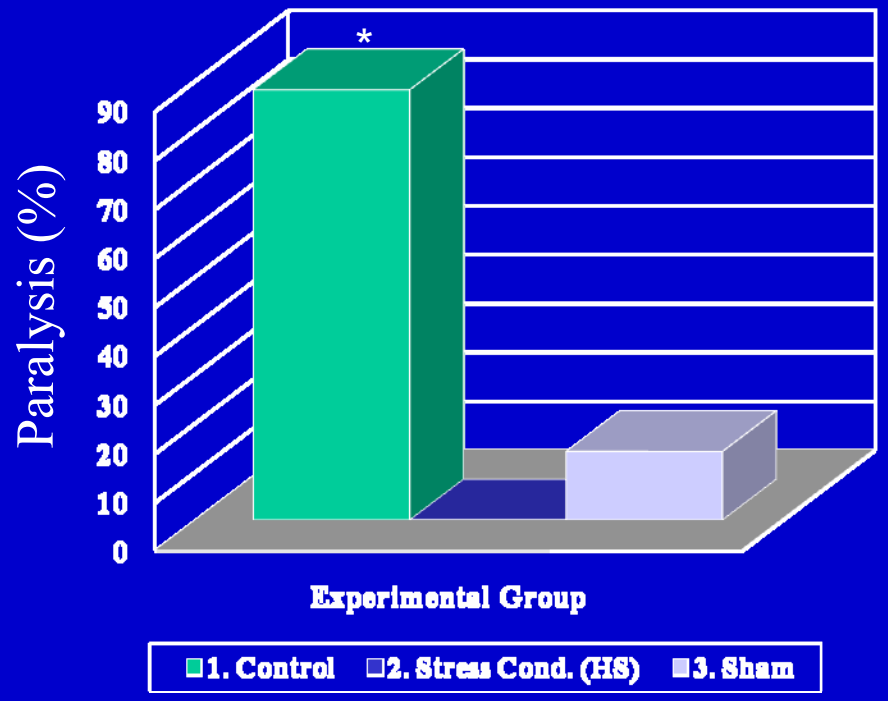
Rodent pedicle-skin flap

Day 5 viable length

Koenig, W., et al Plast Reconstr Surg 1992;90:659.

Acute Spinal Cord Ischemia: Incidence of Paralysis





*p < 0.05 1 vs 2 or 3

Perdrizet, G.A., et al. J. Thoracic & Cardiovas. Surg 2002; 124: 162-70.

Heat Shock Protein

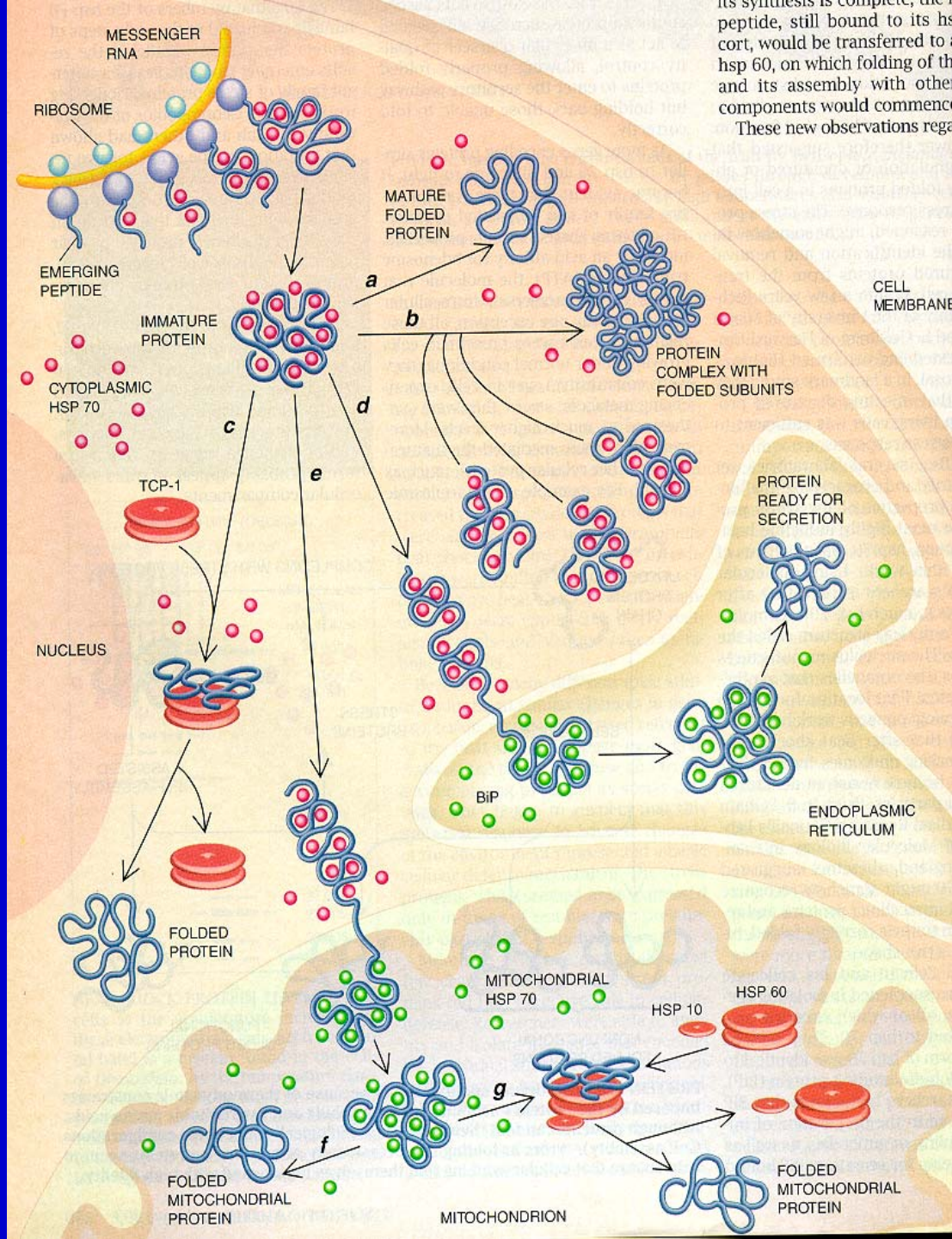
Cell Biology

<u>Basal cell functions:</u>

Protein synthesis

Protein transport

Mitochondrial function



Welch, W. Sci Amer 1993;268:56

Stress Conditioning
Historical Support

- Hans Selye - G.A.S.-1930
- Endotoxin tolerance-1940
- Thermotolerance-1970
- Heat-shock response-1980
- Ischemic preconditioning-1980
- Numerous-1990-present
    - Cytokines, oxidants, metals, fasting

The Physiology and Pathology of Exposure

to

STRESS

A treatise based on the concepts of the

GENERAL-ADAPTATION-SYNDROME

and the

DISEASES OF ADAPTATION

by

HANS SELYE

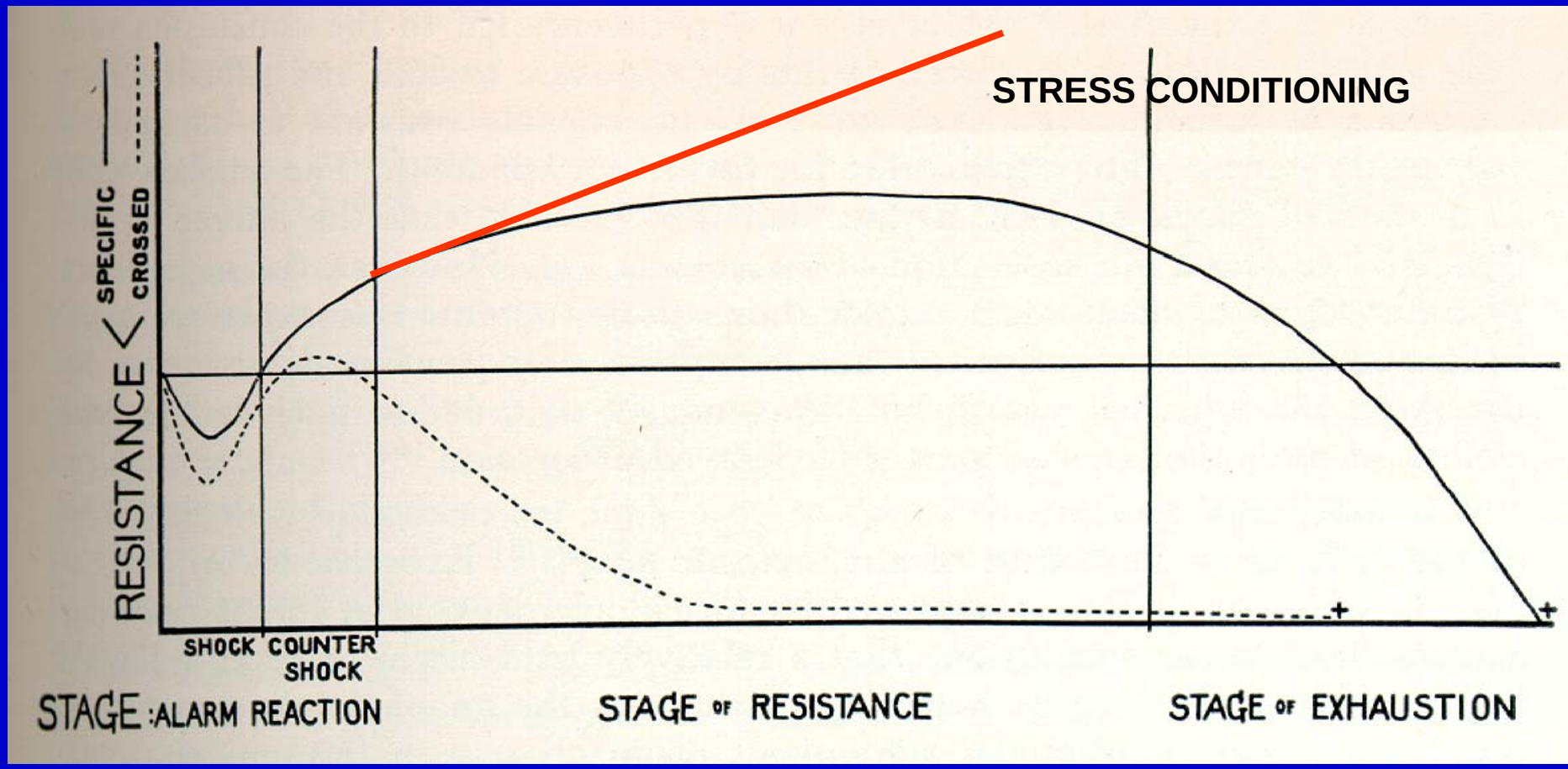
M.D., Ph.D. (Prague), D.Sc. (McGill), F.R.S. (Canada)

Professor and Director
of the
Institut de Médecine et de Chirurgie expérimentales
Université de Montréal

1950

Hans Selye, MD 1907-1982
Universite de Montreal

All Stress Responses Are Triphasic

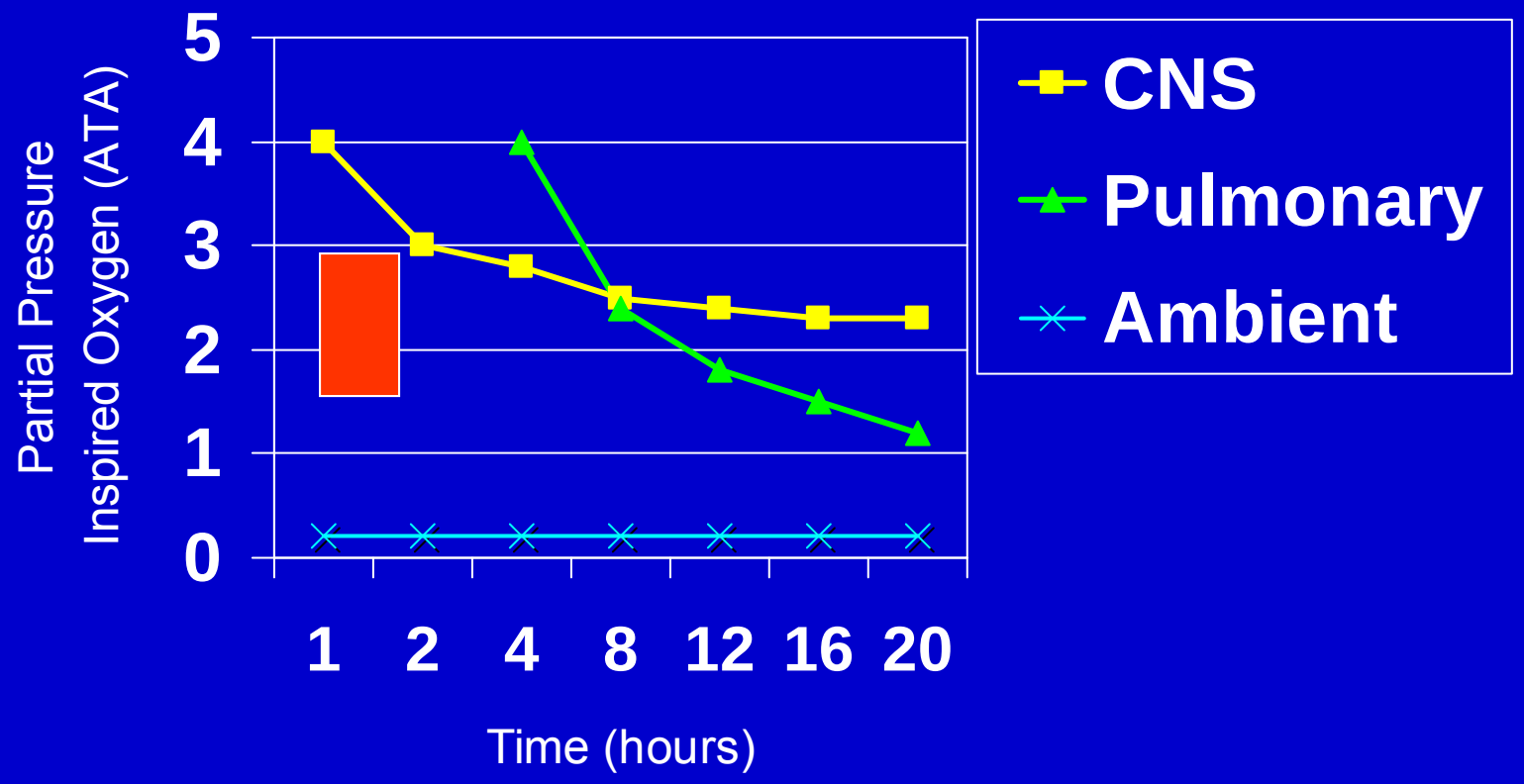


Selye, H. 1950, p55. The Physiology and Pathology of Exposure to Stress

Clinical Stress Conditioning Protocol?

- Is it possible to induce this response using a non-stressful agent?
  - Hyperthermia- 108ºF (42.5ºC)
  - Ischemia or hypoxia
  - Heavy metals
  - Ethanol
  - Oxygen?

Human Oxygen Toxicity Dose-Response Curves



- Dose range for clinical HBO_2 therapy

HBO_2 Pretreatment Increases Ischemic Tolerance: Animal Models

- Rodent brain
  - Chen, M.F., Liver 1992;18:110.
  - Wada, K., Neurosurg. 2001;49:160.
- Rodent heart
  - Kim, C.H., Pflugers Archiv Eur J Physiol 2001;442:519.
  - Cabigas , B.P., Cardiovas Res. 2006;72:143.
- Rabbit spinal cord
  - Dong, H., Anesth. 2002;96:907.
  - Nie, H., J Cereb Bld Flow & Metabolism 2006;26:666.

Hypothesis

- Exposure of human micro-vascular endothelial cell line will:
  - Confer protection against thermal and oxidant injury.
  - Increase expression of cytoprotective genes.

Methods

- HMEC-1 cell line, immortalized; CDC
- Hyperbaric oxygen
  - 2.4 ATA ≈ 1,800 mmHg O_2
  - 60 minute exposure time
- Cytoprotection Assays
  - Thermal or oxidant challenge
  - PI or MTT cell viability
- Gene expression-Microarray
  - Illumina Bead – WG-6 chip
  - Analysis- PBC and Ingenuity Pway
  - rt-PCR confirmation

OxyCure 3000

38,275 total genes on Microarray

Statistical Analysis

8,181 regulated genes

Godman CA, et al. Cell Stress & Chaperones 2010;15:431

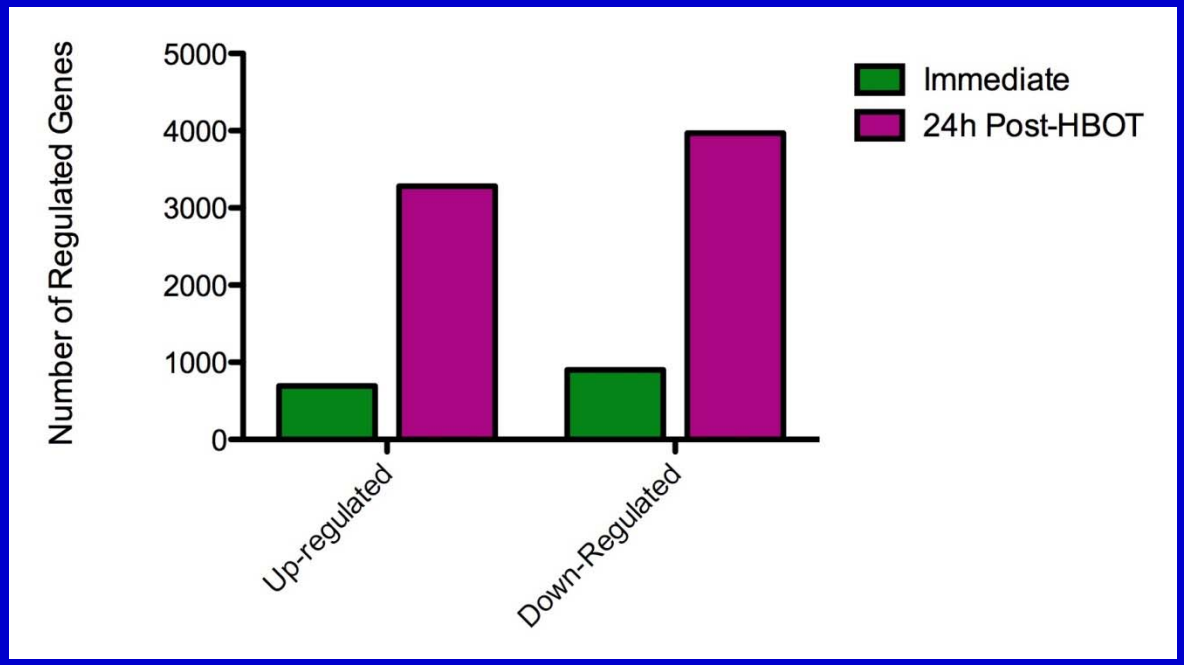
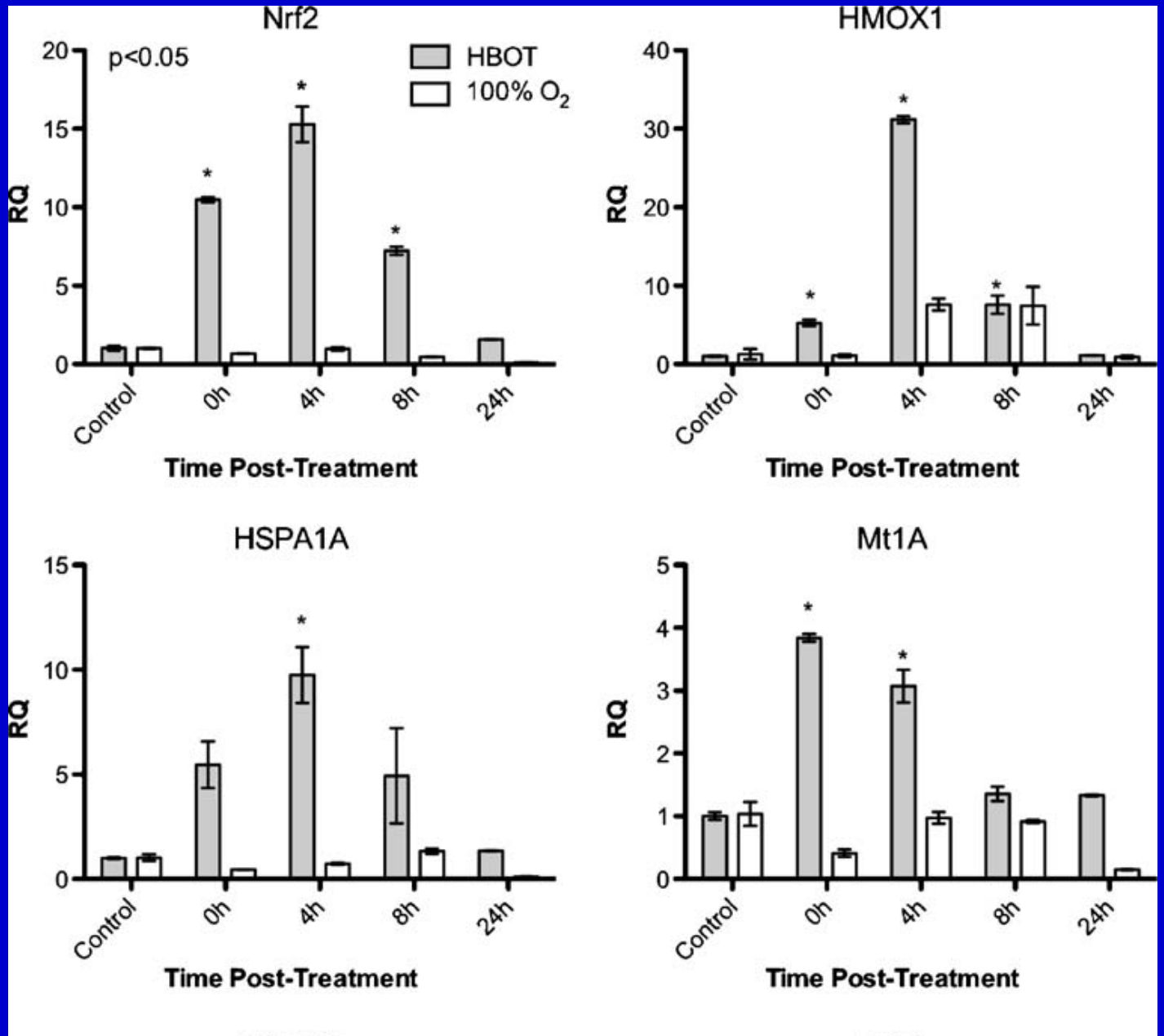


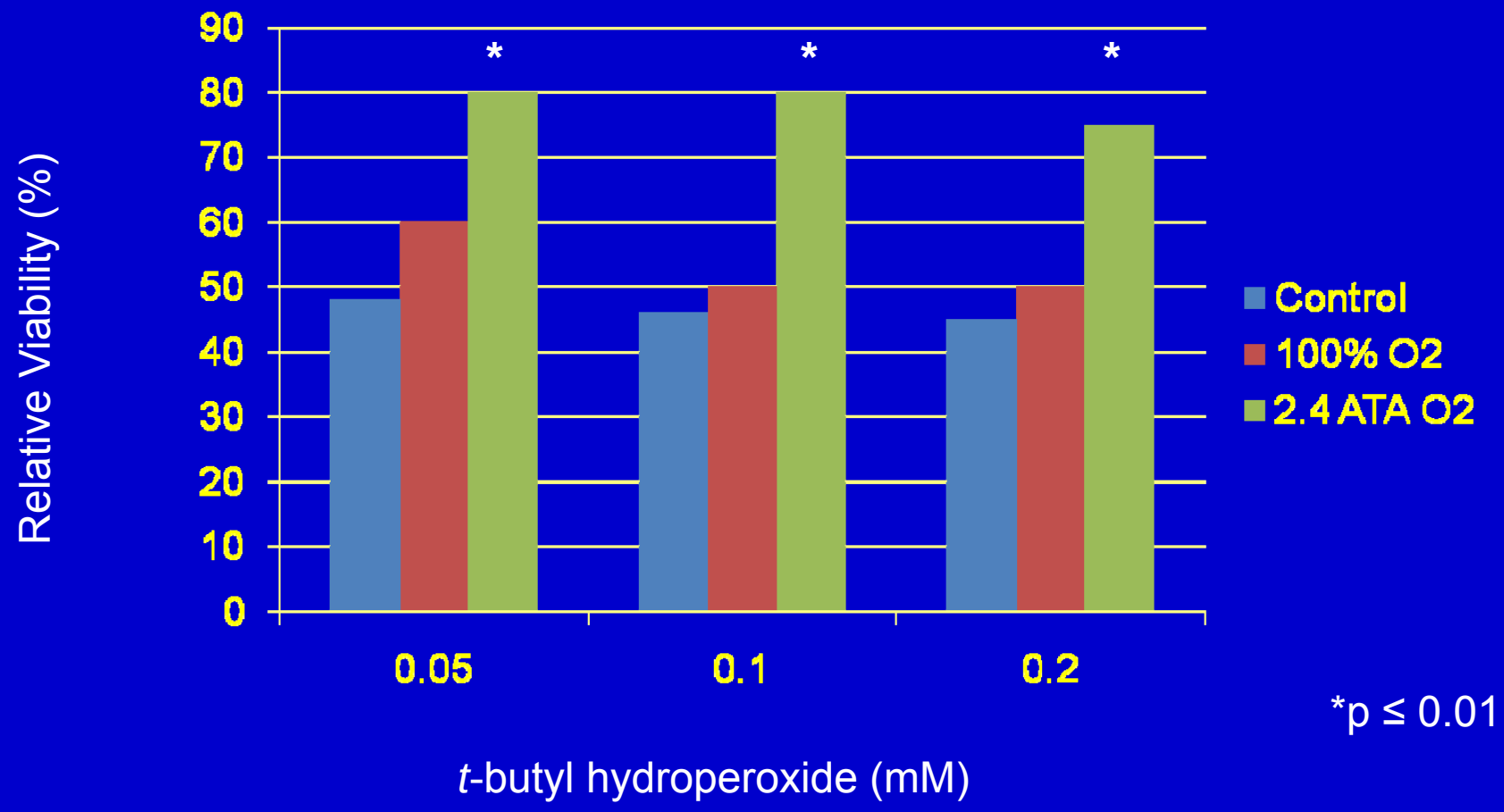
Table 3 Chaperone genes affected by HBOT

Official symbol	Official gene name	GeneID	HB0T-0h/ CNTL-0h	HBOT-24h CNTL-24h
DNAJB1	DnaJ (Hsp40) homolog, subfamily B, member 1	3337	1.83	
HSPA1B	Heat shock 70 kDa protein 1B	3304	1.81	
HSPA1A	Heat shock 70 kDa protein 1A	3303	1.73	
HSPH1	Heat shock 105 kDa/110 kDa protein 1	10808	1.49	
SERPINH1	Serpin peptidase inhibitor, clade H (heat shock protein 47), member 1, (collagen binding protein 1)	871	1.4	
HSPA2	Heat shock 70 kDa protein 2	3306	1.34	
DNAJ4	DnaJ (Hsp40) homolog, subfamily A, member 4	55466	1.32	
DNAJC11	DnaJ (Hsp40) homolog, subfamily C, member 11	55735		2.04
HSPA14	Heat shock 70 kDa protein 14	51182		1.96
HSPE1	Heat shock 10 kDa protein 1 (chaperonin 10)	3336		1.96
HSPH2	Heat shock 70 kDa protein 4	3308		1.92
HSPH1	Heat shock 105 kDa/110 kDa protein 1	10808		1.82
HSPC3	Heat shock protein 90 kDa alpha (cytosolic), class B member 1	3326		1.72
TCP1	t-complex 1	6950		1.72
DNAJC7	DnaJ (Hsp40) homolog, subfamily C, member 7	7266		1.59
DNAJC17	DnaJ (Hsp40) homolog, subfamily C, member 17	55192		1.54
DNAJC3	DnaJ (Hsp40) homolog, subfamily C, member 3	5611		1.54
DNAJC5	DnaJ (Hsp40) homolog, subfamily C, member 5	80331		1.47
DNAJC19	DnaJ (Hsp40) homolog, subfamily C, member 19	131118		1.45
HSPD1	Heat shock 60 kDa protein (chaperonin)	3329		1.45
SERPINH1	Serpin peptidase inhibitor, clade H (heat shock protein 47), member 1, (collagen binding protein 1)	871		1.44
DNAJA1	DnaJ (Hsp40) homolog, subfamily A, member 1	3301		1.39
HSPA8	Heat shock 70 kDa protein 8	3312		1.27
HSPA1A	Heat shock 70 kDa protein 1A	3303		−1.39

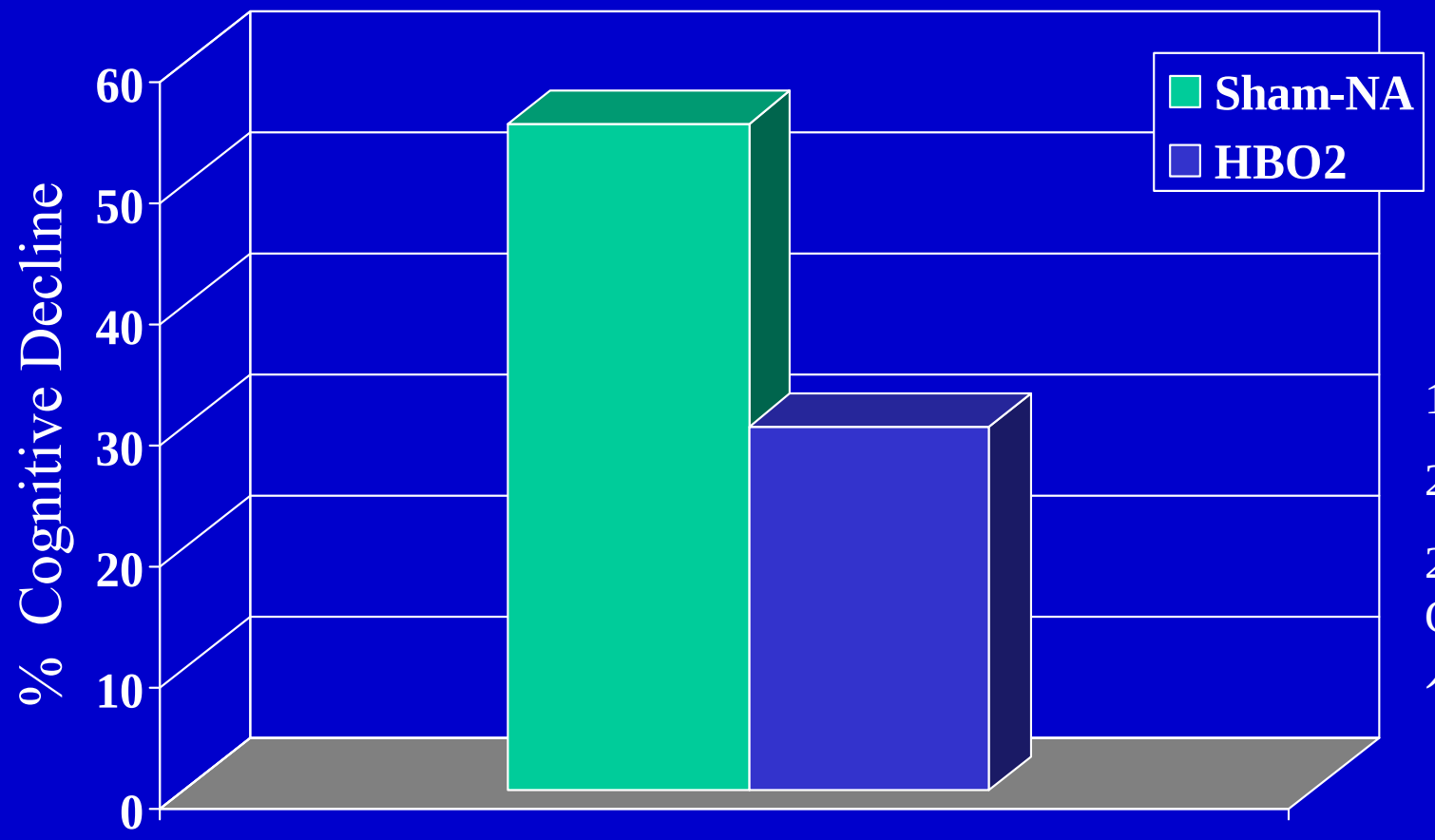
RT-qPCR- HBO_2 vs 100% O_2



HBO_2 Induces Oxidant Protection in HMEC-1 Cells



HBO_2 Pre-treatments Decrease Post-CABG Cognitive Decline



100% 0_2

2.4 ATA/60min.

24, 12, 4 hrs before CABG

↑ HSP70

Alex, J. et al J. Thoracic & Cardiovas Surgery 2005;130:1623.

Summary

- Surgery is stressful.
  - Prevent collateral tissue damage
- Stress Conditioning
  - Potent tissue protection
  - Preventative medicine
- HBO_2 as a safe pre-op stress conditioning agent?

Questions??

“The Light”

Fritz Eichenberg (1901-1990)

Univ. Rhode Island