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Heat-Shock Proteins Induced in Mammals by Whole Body Hyperthermia

Mitsuo KOSAKA[1], and Yutaka OKUMURA[2]

[1]*Department of Environmental Physiology, Institute of Tropical Medicine, Nagasaki University, 1-12-4 Sakamoto, Nagasaki 852, Japan.*
[2]*Department of Radiation Biophysics, Atomic Disease Institute, Nagasaki University, 1-12-4 Sakamoto, Nagasaki 852, Japan.*

INTRODUCTION

When living cells from bacteria to human tissue cells are exposed to heat shock, cells synthesize a set of several proteins called heat-shock proteins (1, 2, 19, 20). Heat-shock proteins are also induced in certain organs of intact mammals during elevation of body temperature (3-7, 10, 18). One major heat-shock protein is with the molecular weight of 70,000-74,000.

INDUCTION OF HEAT-SHOCK PROTEINS

When the body temperature of male Sprague-Dawley rats was brought to 42℃, four heat-shock proteins, with molecular weights of 70,000, 71,000, 85,000, and 100,000 (HSP 70, HSP 71, HSP 85, and HSP 100, respectively), were induced in various tissues of the rats (8). The HSP 70 was strongly induced by hyperthermia. Analysis of translation products of liver mRNAs from heat-shocked rats also showed increased synthesis of the four heat-shock proteins, indicating that these HSP-mRNAs were induced by hyperthhermia. Induction of the HSP-mRNAs was transient after hyperthermia. The induction of these heat-shock proteins was regulated at the transcriptional level. The amount of HSP 70-mRNA increased and was greatest 2.5 hr after hyperthermia; less was found at 6 hr. In contrast, HSP 71-, HSP 85-, and HSP 100-mRNA were most abundant in liver 6 hr after hyperthermia. All these HSP-mRNA were at the control level after 12 hr. The different inductions of mRNAs for the heat-shock proteins may reflect different regulation mechanisms for these proteins. The functions of heat-shock proteins are not understood. There is much evidence of a correlation between the production of heat-shock proteins and the development of thermoresistance of cells in a variety of cellular systems (14, 16). Treatment of rats at 41.8℃ for 1 hr increased the LD_{50} for rats given a second heat treatment 24 to 96 hr later (23). It is probable that heat-shock proteins may be involved in homeostatic control in response to heat shock against

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cells.

In order to know the effect of chronological ageing on the induction of heat-shock protein in human skin, HSP 72 was examined in organ-cultured samples of skin which were obtained from patients aged 17–86 years (17). Normal skin samples were obtained from skin surgery, and were cultured at 37℃ and treated hyperthermally at 45℃ for 1h. Detection of HSP 72 was carried out by indirect immunofluorescence method, using a monoclonal antibody specific for the HSP 72. After heat treatment at 45℃ for 1h, positive nuclear immunofluorescence was observed in all skin samples. When the skin explant was transferred from 45℃ to 37℃, nuclear staining was diminished in the epidermal cells at 1h after heat treatment. In 10 skin samples from the young group (aged less than 50 years), 6 showed positive nuclear immunofluorescence, and 4 showed weakly or faintly positive nuclear immunofluorescence at 1h after heat treatment. On the other hand in 11 skin samples from the old group (aged more than 50 years), 3 showed positive nuclear immunofluorescence, and 8 weakly or faintly positive nuclear immunofluorescence. The organ culture method is a simple system for examining the effects of heat at organ level which might occur in the body. This result indicates that there is an age-related dysfunction of the heat-shock response in normal human skin. There is the possibility of physiological implications that the skin samples from aged people are more sensitive to heat, and that the higher incidence of cell death by heat treatment may be related to the lower inducibility of the heat-shock protein in aged skin.

Heat-shock proteins are induced by various treatments of cells and organs of mammals other than heat; amino acid analogous, transition metals, oxidizing agents, poisons, anoxia, and tissue damage can all induce these proteins (1, 2, 20). The heat-shock response seems to be a way by which cells protect themselves against different stresses.

Ischemia of liver and cardiac stress of rats result in HSP 70–71 production in the liver and in the heat, respectively (4, 10). Administration of Na arsenite to rabbits induces HSP 74 in rabbit tissues (3). Thus, induced heat-shock proteins with the molecular weight of about 70,000 may have some function to protect cells against various stresses (1, 9, 15, 19. 20).

Temperature Regulation of Afghan Pikas, Weak Heat-Tolerant Rabbits

Afghan pikas (whistle rabbits, *Ochotona rufescens rufescens*) shows a character of weak heat-tolerance. The induction of heat-shock proteins in pikas was measured and compared with that in Wistar rats and albino rabbits in order to clarify the mechanism of its character of weak heat-tolerance (12, 13, 22). Heat-shock proteins were induced in liver, kidney, adrenal gland, spleen, brain and skeletal muscle of rabbits (HSP 68) and of rats (HSP 70) by heat-shock. However, induction of heat-shock proteins was not detected in any organs of pikas by heat-shock. These results suggest that the difficulty of induction of heat-shock proteins relates to the weak heat-tolerance in pikas. The lack of induction of heat-shock proteins might be a disadvantage to survive in hot environments.

HEAT-SHOCK PROTEINS IN TEMPERATURE REGULATION

Body temperature of tropical inhabitants is lower than that of temperate inhabitants. Extremely high fevers seldom exceed 41℃ or 42℃. Although the mechanism of these evidences is not yet clarified, we postulate a self-limiting mechanism, such as a negative feed back loop (11).

Mechanism of body temperature regulation in conditions of fever as well as of physiological hyperthermia was investigated. Proliferation of mouse microphage measured by the incorporation of methyl-^{3}H thymidine in cells was suppressed by a heat load of 39℃ for 2 hr. The activity of IL-1 secretion induced by LPS was also suppressed by the heat load. Suppression of proliferation and secretion of macrophage suggests that negative feedback loops exists to control production of heat. The induction of heat-shock proteins by the heat load in the macrophage was detected. The increase of HSP 70 induced in human monocytes was related to the inhibition of IL-1 secretion. It is assumed that IL-1 secretion affecting induction of fever is related to the induction of HSP 70.

In Fig. 1, our hypothesis of the explanation of interaction of heat-shock proteins and IL-1 is shown. LPS pyrogen cooperating with macrophage induces fever by IL-1 and PGE_2, and this fever reacts with macrophage resulting in suppression of proliferation. The suppression of proliferation inhibits IL-1 secretion which causes inhibition of heat production. IL-1 secretion is also suppressed by heat shock. On the other hand, fever induces heat-shock proteins by heat-shock transcription factor (HSTF). The biosyntheses of pre-IL-1 and release

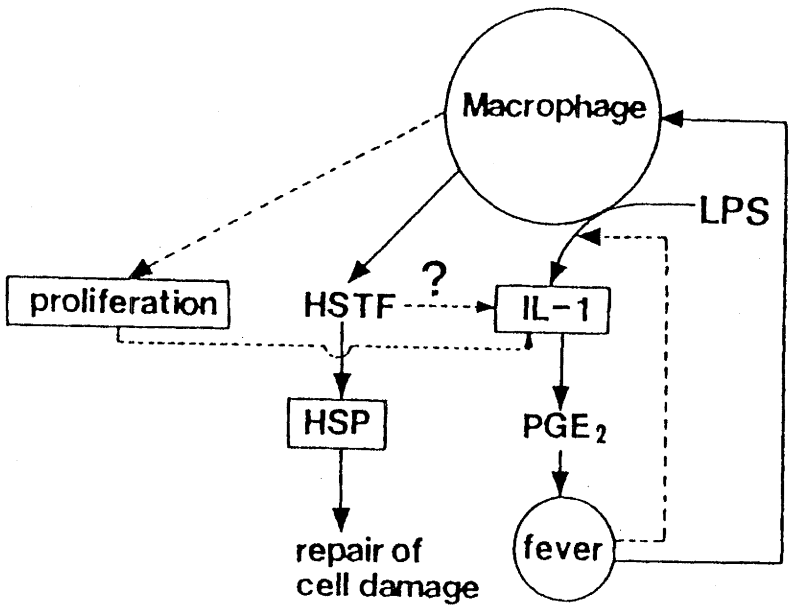

Fig. 1. A model explaining interaction of heat-shock protein and IL-1.

of mature IL-1 are inhibited by heat shock (21). During heat stress, the redistribution of HSTF to heat-shock element (HSE) is accompanied by binding to many additional chromosomal site and may act as a repressor of normal gene activity (24). It may be possible that HSTF binds to IL-1 gene and represses its gene expression. The negative feed back loops shown in Fig. 2 might be one possiblile model to explain a self-limiting mechanism of lower temperature of tropical inhabitants who were easily exposed to various infectious diseases in tropical environments.

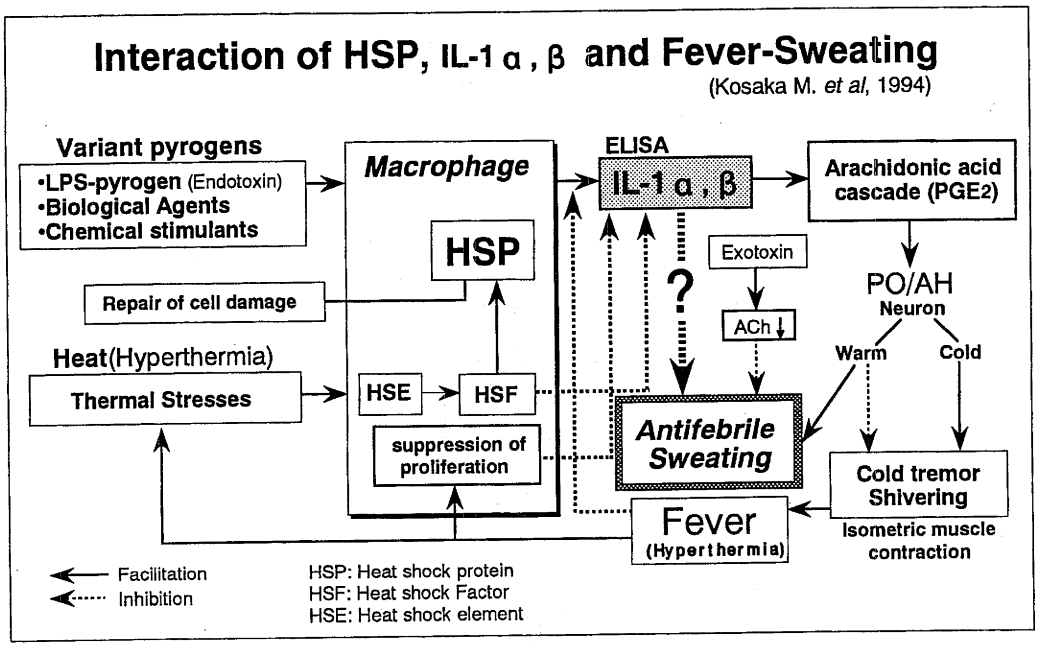

Fig. 2. A model explaining self-limiting mechanism of body temperature

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