Metallothionein gene expression in brown adipose tissue

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Reduced environmental temperature triggers a range of behavioural and physiological responses to maintain body temperature. Brown adipose tissue (BAT) is a major site of thermoregulatory heat production [1], and during cold exposure thermogenesis in the tissue is rapidly stimulated, through the release of noradrenaline by the sympathetic nervous system. There is a substantially increased requirement for oxygen by BAT during thermogenesis and this may be accompanied by an increase in free radical generation.

Metallothionein (MT) is a low molecular weight metal-binding protein which is induced directly by metals such as Zn, Cd and Cu, and by a wide range of stress factors which elevate for example, steroids, cytokines and catecholamines [2]. MT protein levels in the liver and kidney have also been shown to increase when rats are exposed to 4°C for 24 h [3, 4]. Since hepatic MT levels increase on cold exposure and the protein has a proposed role as an antioxidant, we have investigated MT gene expression in BAT.

Groups of 6 male Hooded Lister rats (~260 g body weight) were maintained at 25°C (control), or exposed to 6°C for either 6 or 24 h. A further group at 25°C were injected s.c. with ZnCl₂ at a dose of 10 mg Zn.Kg⁻¹ body weight, and killed 6 h after the injection. Liver, kidney, interscapular BAT and epididymal white adipose tissue were removed and snap-frozen in liquid nitrogen. Total RNA was extracted, separated by agarose gel electrophoresis and transferred to a nylon membrane by capillary blotting, as described elsewhere [5]. Detection of MT-1 mRNA was by chemiluminescence using a specific 28-mer antisense oligonucleotide probe [6] labelled with digoxigenin. 18s rRNA was also measured by the same technique using a 35-mer antisense oligonucleotide with a consensus sequence for a wide variety of species [5]. MT-1 protein in soluble tissue extracts was measured by radiolimunnoassay [7].

Cold exposure caused a time-related increase in hepatic MT-1 gene expression; Zn injection, however, had a much more profound effect on liver MT-1 mRNA levels (Fig. 1A). In contrast, Zn had no effect on MT-1 gene expression in BAT, whereas cold exposure clearly induced transcription of this gene, the effect being greater at 24 h (Fig. 1B). Analysis of MT-1 protein was consistent with these results, and the Zn levels in BAT increased gradually over a number of days as compared to the rapid response of MT over the initial 24 h in the cold. Further studies are in progress to investigate both the mechanism of induction and the function of MT in BAT.

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