Short Communication

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The Effect of Temperature Acclimation on the Spin-lattice Relaxation Time of Brown Adipose Tissue

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Summary Cold acclimation induced an increase of spin-lattice relaxation time ($T_1$) of proton nuclear magnetic resonance in rat brown adipose tissue with concomitant increase of water content, while heat acclimation did not effect $T_1$ of this tissue, although its water content was significantly decreased. Temperature acclimation did not affect either $T_1$ or water content of neck muscle.

The notion that various physiological states of tissues might be characterized by water molecules existing in different physical states has been developed recently through the observations of changes in water proton signals obtained by the use of nuclear magnetic resonance (NMR) spectroscopy (Beall et al., 1976). Although the NMR spin-lattice relaxation time ($T_1$) is well correlated with tissue hydration, it is also dependent upon the motional freedom of the water molecules.

Small mammals such as rats acclimate to cold by dramatic transition of heat production from shivering to more efficient non-shivering thermogenesis especially in conjunction with an enhanced thermogenesis and marked hyperplasia of brown adipose tissue (Smith and Horwitz, 1969). Moreover, the responses of this tissue to humoral regulators such as noradrenaline and glucagon are considerably modified by temperature acclimation (Kuroshima and Yahata, 1979). However, it is not yet known precisely which cellular mechanisms regulate a high exothermic process of this tissue (Nicholls, 1977).

The present study concerned with the changes in NMR $T_1$ of brown adipose tissue and skeletal muscle due to temperature acclimation in order to know whether physical state of water in these tissues is involved in the acclimated changes in relation to non-shivering thermogenesis.

Adult male rats of Wistar strain were used as experimental animals. They were acclimated for 4 to 5 weeks to 25°C at 50% relative humidity (warm-acclimated controls), 5°C (cold-acclimated rats) and 33°C at 30–45% relative humidity (heat-acclimated rats). These animals were killed by cervical blood letting. The

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interscapular brown fat and neck muscle were quickly removed and packed into glass tubes with 10 mm diameter. $T_1$ was measured by a 90°-90° pulse sequence method as the time for $(M_0 - M_{s(t)})/M_0$ (magnetization ratio) to decay $1/e$ of its value at $t=0$ through the data points plotted on a semilog scale by the use of 60 MHz pulsed NMR apparatus (JNM-FSE-60C, JEOL Ltd., Tokyo, Japan). $M_{s(t)}$ is the recovery of magnetization as a function of the time $t$ between two pulses and $M_0$ is the thermally equilibrated magnetization (Tanaka et al., 1978). The measurements were performed immediately after excision of the tissues at room temperature of 25°C. Relation between magnetization ratio and the time $t$ was linear in both neck muscle and brown fat. Water content was determined by drying the sample in an oven at 100°C until a constant weight was achieved, usually for 24 hr. The statistical significance of the results was tested by the Student $t$-test.

As shown in Table 1, $T_1$ was greater in the neck muscle with higher water content than that in the brown adipose tissue with lower percentage of water.

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<tr>
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<th>Neck muscle</th>
<th>Brown adipose tissue</th>
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<tbody>
<tr>
<td></td>
<td>$T_1$</td>
<td>Water</td>
</tr>
<tr>
<td>Warm-acclimated controls (7)</td>
<td>0.73±0.022 75.5±0.21</td>
<td>0.30±0.017 30.7±1.63</td>
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<tr>
<td>Cold-acclimated rats (8)</td>
<td>0.72±0.045 75.4±0.08</td>
<td>0.39±0.007 44.6±0.64</td>
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<tr>
<td>NS</td>
<td>NS</td>
<td>$&lt;0.001$</td>
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<tr>
<td>Heat-acclimated rats (8)</td>
<td>0.78±0.015 76.3±0.22</td>
<td>0.28±0.009 25.0±0.73</td>
</tr>
<tr>
<td>NS</td>
<td>NS</td>
<td>$&lt;0.01$</td>
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Mean±standard error. Number in the parenthesis indicates the number of animals. NS: not significant.

This result is consistent with the previous findings (Inch et al., 1974; Saryan et al., 1974). Temperature acclimation did not affect either $T_1$ or water content of the neck muscle. On the other hand, cold acclimation induced significant increase in $T_1$ of brown adipose tissue with concomitant increase of water content. Heat acclimation did not effect $T_1$ of this tissue, although its water content was significantly decreased. Figure 1 shows the relationship of $T_1$ to the percent water content in the brown adipose tissue from all experimental groups. As water content increased, the $T_1$ rose. It should be noticed that the protons in the lipid molecules would significantly affect $T_1$ of lipid-rich tissues. $T_1$ of protons in the white adipose tissue exhibits the shortest value among the tissues due to low water content and protons of lipid molecules (Inch et al., 1974). Cold acclimation caused a fall (Moriya and Itoh, 1969), while heat acclimation an increase (Yahata and Kuroshima, unpublished observation) in lipid content of the brown adipose tissue. Therefore, the decreased content of fat might be responsible for the
Fig. 1. Proton spin-lattice relaxation time ($T_1$) vs. percent water content plotted for brown adipose tissue from temperature acclimated rats. ◯, warm-acclimated controls; △, heat-acclimated rats; ×, cold-acclimated rats.

Elevated $T_1$ in the cold-acclimated brown adipose tissue. However, it is also conceivable that factor(s) other than water and fat contents influences $T_1$ of water protons in the brown adipose tissue, inasmuch as $T_1$ of heat-acclimated brown adipose tissue was not changed despite of decreased water and increased fat contents.

It has been argued that water molecules interact with cellular macromolecules to such an extent that the physical state of cellular water differs from that of ordinary water. This view seems to be substantiated by the following findings that longer $T_1$ values in cancerous tissues are not strictly correlated with a non-specific increase in the percentage of tissue water (HAZLEWOOD et al., 1974), and significant changes in $T_1$ are observed by manipulation of chromosome condensation cycles independent of changes in water content (BEALL et al., 1976). Although poorly understood, some possible physical mechanisms underlying the elevated relaxation rate of water protons have been proposed; those include a lower degree of intra-cellular water order, quantitatively fewer macromolecule-bound paramagnetic ions or free radicals, and a larger fraction of relatively free intracellular water in rapid exchange with a fraction having a short relaxation time (SARYAN et al., 1974). Therefore, an elucidation of extent to which the modified $T_1$ in the temperature-acclimated brown adipose tissue is associated with its thermogenic capacity awaits further study.

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REFERENCES


HAZLEWOOD, C. F., CLEVELAND, G., and MEDINA, D. (1974) Relationship between hydration and proton nuclear magnetic resonance relaxation times in tissues of tumor-bearing and


