The tissue levels of paramagnetic ions are an important factor in the determination of $T_1$ values as observed by nuclear magnetic resonance (NMR) imaging. The increased levels of iron present in human disease states such as hemochromatosis lead to decreased $T_1$ values. The mean liver $T_1$ of three patients with iron storage disease was determined to be 130 msec, significantly different from the value of 154 msec, the mean for 14 normal controls. Whether NMR will be able to detect the increased copper levels in liver and brain in Wilson disease remains for further clinical trials to evaluate. NMR imaging, however, does serve as a noninvasive method for the diagnosis of states of iron overload and as a technique to follow progression of disease or response to medical therapy.

Iron and copper, when present in sufficient concentration in tissue, cause a decrease in the spin-lattice relaxation time ($T_1$) observed by nuclear magnetic resonance (NMR) imaging [1]. This enhancement of relaxation occurs because of the interaction between the magnetic moment of the paramagnetic ion and that of the hydrogen nucleus [2]. Because of this paramagnetic effect on $T_1$, NMR imaging may find use in the diagnosis of human disease associated with excessive iron or copper stores. Iron overload occurs in primary hemochromatosis, transfusion hemosiderosis, and alcoholic cirrhosis [3, 4]. Extremely high concentrations of copper are present in Wilson disease [5, 6] and various lymphomas [7], particularly Hodgkin disease, and in some cases of long-standing biliary obstruction and biliary cirrhosis [8]. In addition to its use in diagnosis, NMR imaging may prove to be valuable as a noninvasive means of following the progression of disease and its response to medical therapy.

To demonstrate the sensitivity of NMR in the detection of copper and iron, $T_1$ was determined for a standard set of solutions (in vitro) of various copper and iron ion concentrations. The feasibility of detection and quantitation of iron overload in vivo was then tested by observing the change in $T_1$ of mouse serum, homogenized kidney, and homogenized liver after the intravenous injection of iron dextran. Intramuscular injection of a therapeutic chelating agent was performed in an additional subset of experimental animals to test the feasibility of the use of NMR to follow response to therapy. After the practical use of NMR to detect elevated copper and iron levels had been demonstrated experimentally, a group of patients with iron overload were compared to a group of normal controls by NMR imaging of the abdomen. Correlation was made with CT scanning and an additional group of patients with early alcoholic cirrhosis examined by NMR.

Materials and Methods

NMR Spectroscopy

By quantitating the effect of $T_1$ of iron (+3) and copper (+2) ions in solution, the
concentrations of these ions that must be present in tissue to be detected by NMR can be determined. The $T_1$ values for a set of serial dilutions of ferric nitrate, iron dextran, and copper sulfate in saline were first determined experimentally. These in vitro $T_1$ measurements were made by the inversion-recovery technique using a JEOL FX-90Q NMR spectrometer operating at 90 MHz. $T_1$ values were calculated from the Fourier transform of the FID signal using the null method [9]. The precision of the null method in the determination of $T_1$ was checked by comparison with results obtained by semilogarithmic curve-fitting for a representative set of experimental data.

In the second phase of experimentation, the $T_1$ of murine serum, homogenized kidney, and homogenized liver was determined for animals before and after intravenous injection of iron dextran. Sterile water dilutions of 5:1 for serum, kidney, and liver were examined spectroscopically. Groups of three mice each were sacrificed and studied at time zero (control), 15 min, 75 min, and 24 hr after intravenous iron administration (0.5 mg/g). Two additional sets of animals received intramuscular injections of desferoxamine (an iron chelating agent, dosage 1.2 mg/g) 15 min after the iron injection and were sacrificed at 75 min and 24 hr. With this animal model, the feasibility of the use of NMR to study iron overload states and the response of such to chelate therapy could be investigated.

**NMR Imaging**

Three patients with iron storage disease (two with hemochromatosis and one with hemosiderosis) were examined by NMR imaging and compared to a control group of 14 normal volunteers. In each case a histologic diagnosis of elevated tissue iron levels was made by liver biopsy. Computed tomographic (CT) examinations of the abdomen were performed for comparison in two of the three patients. NMR imaging was performed with the Aberdeen 0.04 T magnet system described in detail elsewhere [10, 11]. Transverse 16-mm-wide slices were obtained at 2-cm intervals through the abdomen. Use of the spin-warp technique allowed a scan time of 4 min/slice with the capability of display of proton density or calculated $T_1$ NMR images [10]. Measurements of liver $T_1$ were obtained from appropriate regions of interest on the calculated $T_1$ images. CT scanning was performed with an Elscint Exel–905 system. A retrospective analysis of the NMR scans from patients in the clinical series at Aberdeen with alcoholic cirrhosis also was performed.

**Results**

**NMR Spectroscopy**

The effect of ferric ion in solution on $T_1$ is illustrated in figure 1A. Iron complexed by dextran was less effective in decreasing $T_1$ when compared with the free ion in solution. The lowest concentration tested, $5 \times 10^{-3}$ mg/ml of Fe$^{3+}$, continued to cause a statistically significant decrease in $T_1$. The effect on $T_1$ was concentration-dependent.

The effect of cupric ion in saline solution on $T_1$ also was concentration-dependent (fig. 1B). A two-fold reduction in $T_1$ was achieved with a $5 \times 10^{-2}$ mg/ml solution, the lowest concentration tested.
Table 1: Liver $T_1$ Values: Comparison of Patients with Iron Storage Disease with Normal Volunteers

<table>
<thead>
<tr>
<th>Subjects</th>
<th>No. Patients</th>
<th>$T_1$ (msec)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Iron storage disease . . .</td>
<td>3</td>
<td>130 (± 4)</td>
</tr>
<tr>
<td>Normal controls . . . .</td>
<td>14</td>
<td>154 (±11)</td>
</tr>
</tbody>
</table>

Note — $p < 0.01$ for $T_1$ between control group and group with iron storage disease.

Discussion

Tissue levels of paramagnetic ions serve as one basic factor in the determination of $T_1$ values as observed by NMR imaging. Copper and iron, both of which are paramagnetic when present in ionic form, occur in abnormally high levels...
in tissue in several disease states of man. NMR imaging, because of its physiologic basis, may provide a more accurate tool for diagnosis and surveillance of these disease states than those currently available in conventional radiology. In iron storage disease, mean levels of 1.8 mg/g (iron in liver tissue) are present when extensive hemosiderin deposits are visible on pathologic specimen [12]. Normal liver iron storage concentration is 0.1 mg/g. Similar changes also can be observed in the spleen in iron storage disease. From simple in vitro experimentation (fig. 1A), one can conclude that NMR is quite sensitive to the presence of iron in solution (specifically the ferric ion). The extension of this observation to clinical interpretation of NMR images will be discussed later.

Figure 1B reveals that NMR is also very sensitive to the presence of copper ion. Levels of greater than 250 mg/g of copper in dry liver by liver biopsy occur only in Wilson disease and long-standing biliary obstruction [6]. Levels as high as 700 mg/g of copper in liver have been reported in Wilson disease [8]. In contrast, the normal range is 15–55 mg/g. Extremely high concentrations of copper also have been reported in Hodgkin disease [7]. In addition, greatly elevated concentrations of copper are seen in the brain (particularly the thalamus and lenticular nuclei) in Wilson disease [5]. NMR imaging may be useful in the diagnosis of these diseases by the recognition of changes in T1 and their origin. The demonstration that elevated copper levels in these disease states cause a significant decrease in T1 on NMR imaging awaits further clinical trials.

In animal experimentation, the administration of pharmacologic doses of iron dextran to mice results in T1 changes that are detected readily by NMR (figs. 2–4). As previously noted, an increase in ferric ion concentration leads to a decrease in T1 (an enhancement of relaxation). NMR spectroscopy detected the initial high serum levels of iron (after intravenous injection) as a decrease in T1, with T1 recovering slightly toward normal as the iron was redistributed to other organ systems by 24 hr (fig. 2). The effect of intramuscular administration of a therapeutic chelating agent in lowering the final serum concentration of iron also was observed by NMR, as a more rapid increase in T1 toward normal.

Increased levels of iron in mouse kidney and liver after intravenous injection also were observed (by NMR) by the decrease in T1 of these tissues (figs. 3 and 4). Chelate therapy causes renal excretion of iron and secondary hepatobiliary excretion. Both of these effects were detected
NMR OF IRON AND COPPER DISEASE STATES

Fig. 8.—A, CT of man with hemochromatosis to illustrate increased attenuation of liver due to iron deposition. Apparently uninvolved (by CT) left lobe of liver was shown by NMR to be infiltrated by neoplastic disease. B, NMR (T₁ image) at comparable level. Involvement of left lobe of liver by hepatocellular carcinoma. Decreased T₁ of right lobe (dark gray) because of hemochromatosis.

by NMR as a further decrease in T₁. As followed by NMR, renal levels of iron began to return toward normal by 24 hr (T₁ increasing toward its original value). However, in liver, T₁ values remained low, indicating storage of iron in this organ with redistribution from other tissues.

Results from clinical imaging of patients by NMR indicate that patients with iron overload (hemochromatosis or hemosiderosis) can be distinguished clearly from the normal population by the decreased T₁ of liver tissue. As displayed on the gray scale for T₁ of the Aberdeen scanner, increased deposition of iron changes the appearance of liver from light (T₁ = 150 msec) to dark gray (T₁ = 130 msec). CT scanning of the same patient group also revealed the increased tissue levels of iron (the liver appears denser than normal). That hemochromatosis and other states of iron overload may be diagnosed by CT is well established [13, 14]. However, NMR is not subject to the bowel gas artifacts and problems inherent to standardization of Hounsfield units that CT manifests. Indeed NMR may be preferable to CT for examination of the liver because of the inherent improved soft-tissue discrimination [15].

Some cases of early alcoholic cirrhosis had T₁ values below normal for liver on NMR imaging. However, this was not a consistent finding. Either fatty infiltration or an increased hepatic level of iron may be responsible for this finding. Excessive alcoholic intake is a common cause of dietary iron overload that presents with hemochromatosis as a clinical syndrome [3].

The increased levels of iron present in the liver with hemochromatosis and hemosiderosis can be detected by NMR imaging. The pattern of additional organ involvement may make possible a more precise diagnosis of disease. This may obviate liver biopsy and provide a noninvasive method to observe the response of these diseases to medical therapy. However, the presence of fibrosis and cirrhosis in the liver results in an increase in T₁ [16]. This may alter the appearance on NMR imaging of diseases such as hemochromatosis and hemosiderosis in which one would expect T₁ to be decreased due to the presence of paramagnetic ions. In experimental work with mice, the tissue distribution of iron after intravenous injection and the subsequent excretion of iron after chelate therapy could be observed by NMR. Chronic aplastic anemia and thalassemia require frequent blood transfusions for survival of the patient. This therapy may lead to transfusion siderosis. However, NMR imaging could be used as a noninvasive tool for following the progressive accumulation of iron in the liver. Thus the need for chelate therapy in these patients could be defined more accurately. Myocardial deposition of iron, an important factor in morbidity and mortality, could also be followed.

In summary, NMR, like CT, can diagnose elevated tissue levels of iron. Yet NMR is noninvasive, without known harmful effects [17], and provides superior soft-tissue discrimination without air or bone artifacts. Thus NMR may find use in the diagnosis and investigation of iron storage diseases. Whether the increased tissue levels of copper in Wilson disease and Hodgkin lymphoma can be detected by NMR awaits further clinical trials.

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