Sequential MR Studies of Intracerebral Hematomas in Monkeys

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It is well recognized that the MR appearance of intracranial bleeding changes with the age of lesion. It is also well known that hemoglobin in stagnating blood undergoes oxidation to methemoglobin, a substance that lowers the relaxation times of surrounding water protons. To study these phenomena in a controlled way, about 3 ml of blood was injected into the right frontal lobe of two rhesus monkeys, and they were scanned sequentially for up to 2 months in a Picker NMR scanner (B_o = 0.25–0.5 T). The image intensity of the blood changed during the first week, consistent with the lowering of T1 and T2. On the inversion-recovery scans the initial appearance of the blood was less bright than was the contralateral white matter, reversing after 3–5 days. The opposite was true on spin-echo images. T1 and T2 values were calculated for all images. In parallel experiments, several milliliters of freshly drawn blood was placed in test tubes and relaxation times were measured in a bench-top analyzer at 0.25 T over a period of 10 days. The relaxation times dropped markedly, at a rate that depended on sterility, temperature, etc., closely approaching the expected result for complete conversion of hemoglobin to methemoglobin. Ten blood samples with different methemoglobin concentrations were prepared by adding varying doses of sodium nitrite. The change in 1/T1 was found to be roughly proportional to the methemoglobin concentration for values up to 40%, and the initial slope was consistent with published data.

Since the original observation of Sipponen et al. [1], there has been considerable interest in the appearance of extravasated intracranial blood on magnetic resonance (MR) scans [2–4]. It has been noted that the signal intensity, and hence the visual appearance, of the blood is not constant, but changes with the passage of time. In fact, there are times when the blood appears very similar to normal brain tissue, thereby creating an obvious identification problem for the diagnostician. It has been suggested [5] that this change in MR signal is largely due to the chemical change of hemoglobin to methemoglobin, a substance with a well known relaxing effect on nuclear spins [6, 7].

To study this phenomenon in a controlled way, we injected blood into the brain of rhesus monkeys and scanned them sequentially. We found that the pattern of the resulting MR changes in the hematoma paralleled the reported observations in human subjects. We also followed similar MR changes in blood samples in vitro and correlated these changes with the amount of methemoglobin formation. The emphasis of this experiment was on the early stage (days 0–6) after the hemorrhagic event, that is, in the period of major diagnostic uncertainty.

Materials and Methods

Two rhesus monkeys were used as subjects. About 3 ml of freshly drawn venous blood was injected through a surgical burr hole into the depth of the right frontal lobe. Both MR and CT scans were obtained daily at first, with the frequency then decreasing, for a total scanning period of up to 2 months. The MR scans were obtained on a Picker instrument with a field strength of 0.25 T for the first monkey and of 0.5 T for the second monkey. Each set

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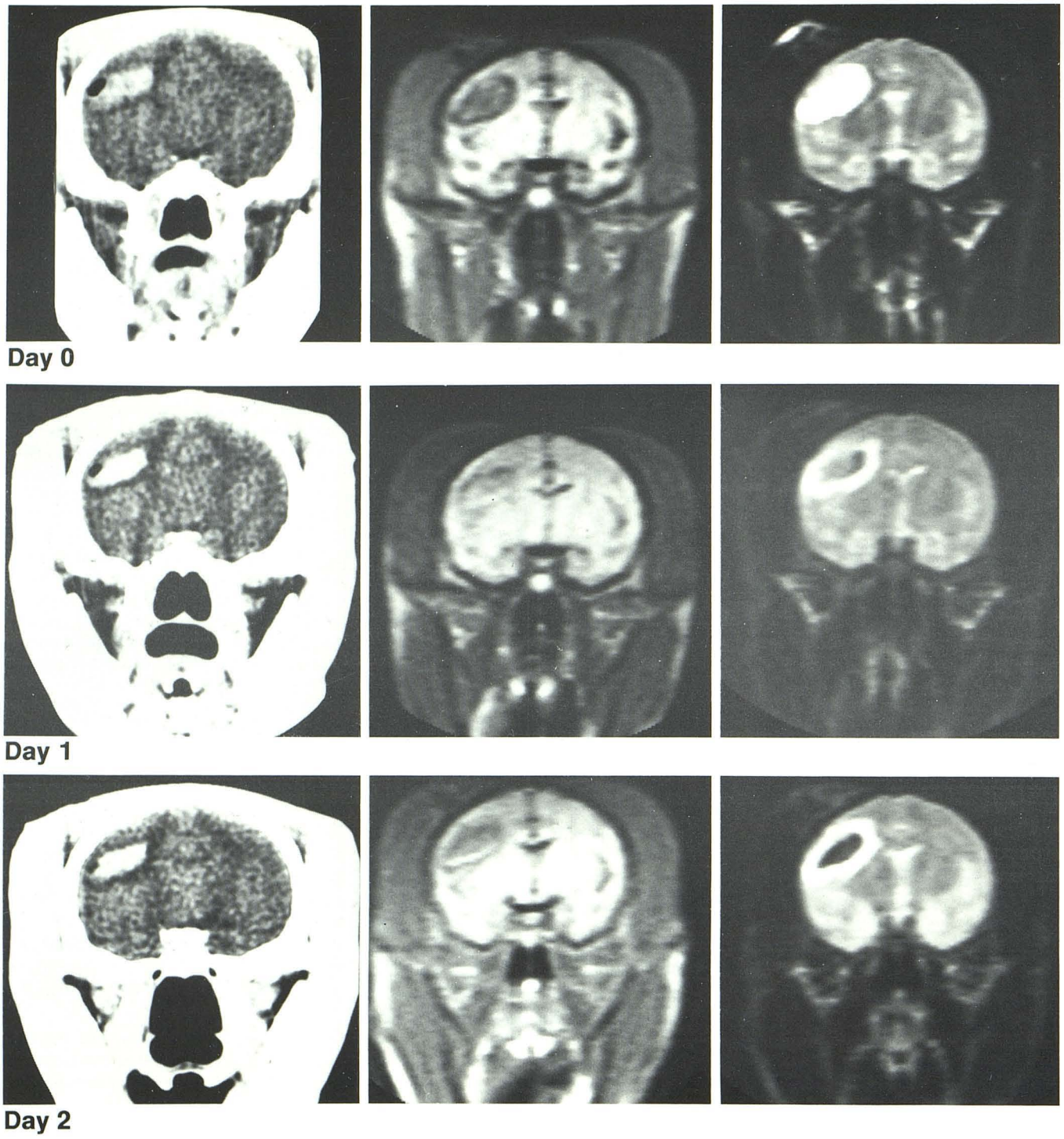
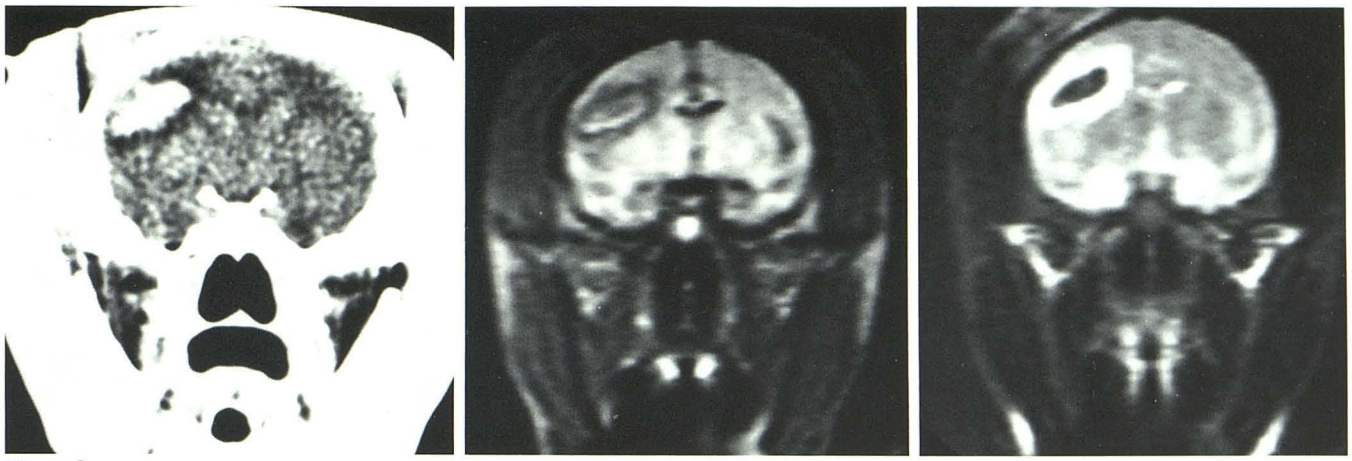


Fig. 1.—Monkey 2 at intervals of 2 hr (day 0) to 2 months after surgery. CT scan (left), IR (T1-weighted) image (middle), and SE (T2-weighted) image (right). Hematoma in right frontal lobe is surrounded by halo of edema. Edema consistently appears dark on CT scans, dark on T1-weighted scans, and bright on T2-weighted scans, although separation of edema from blood on T2-weighted images is not visible until day 1.



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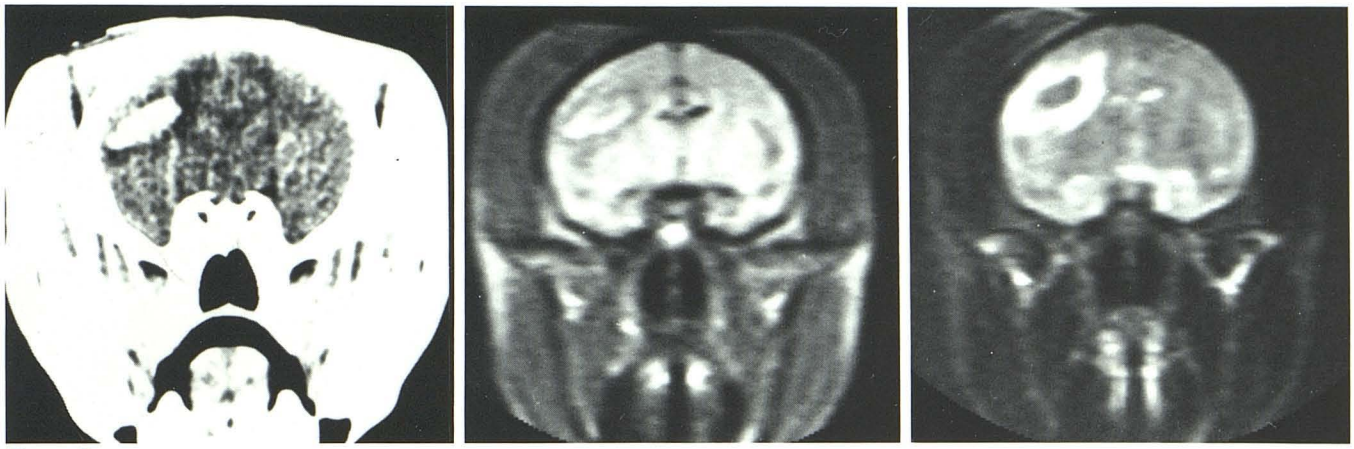

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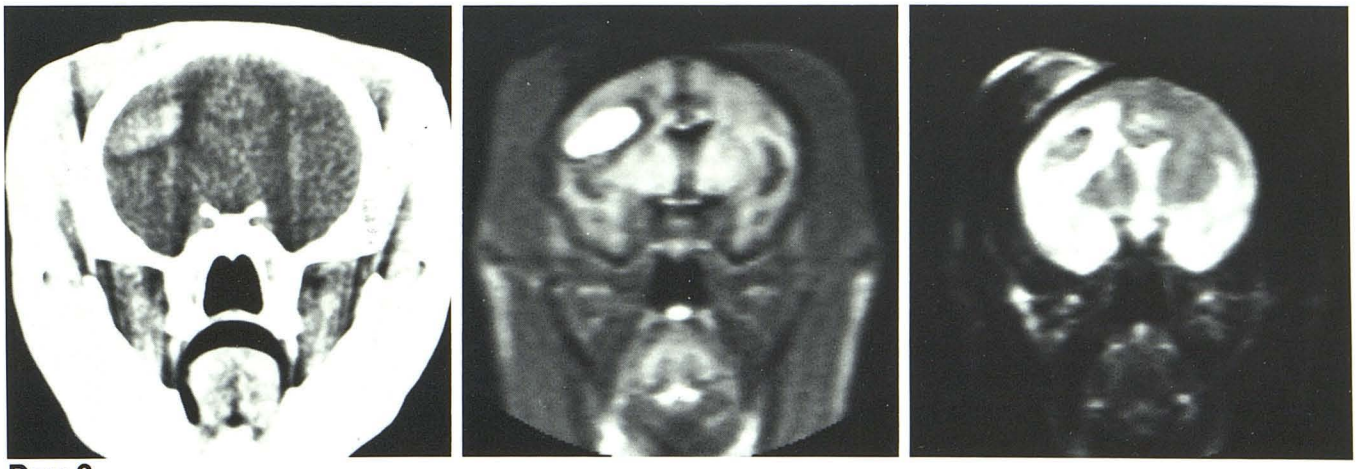

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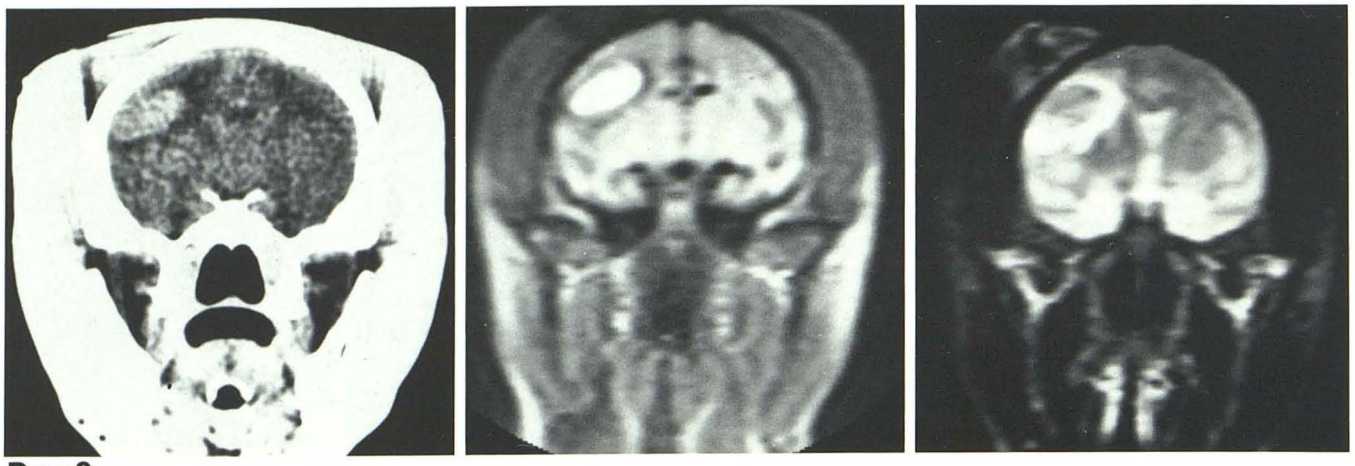

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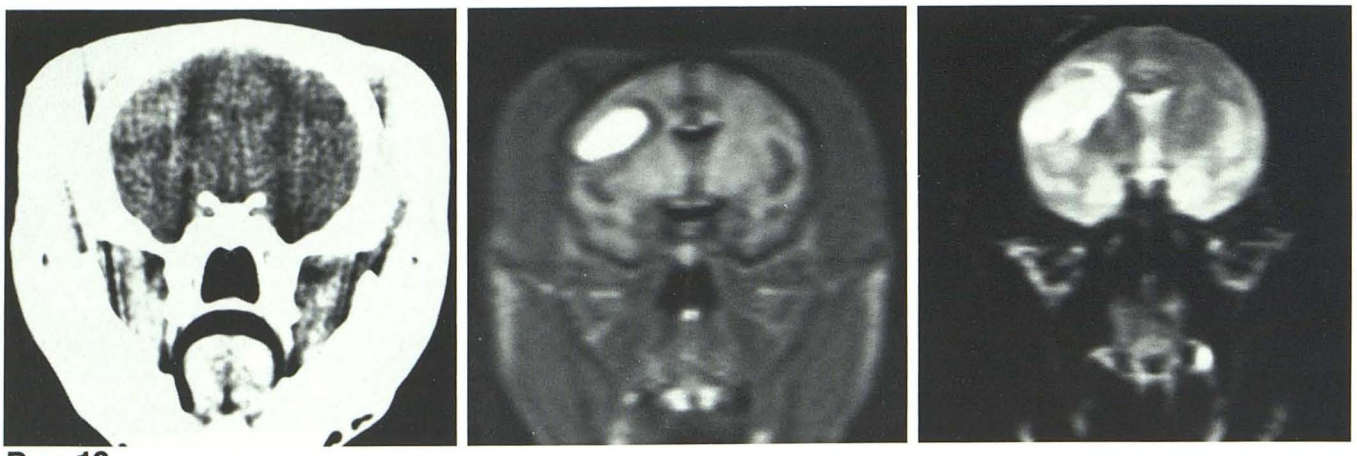

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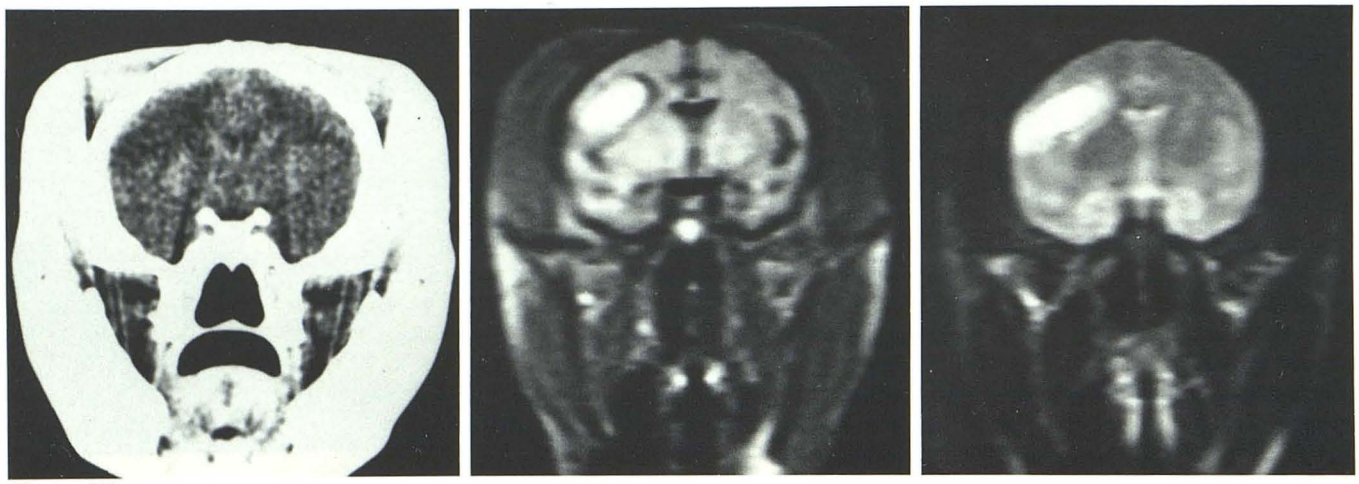
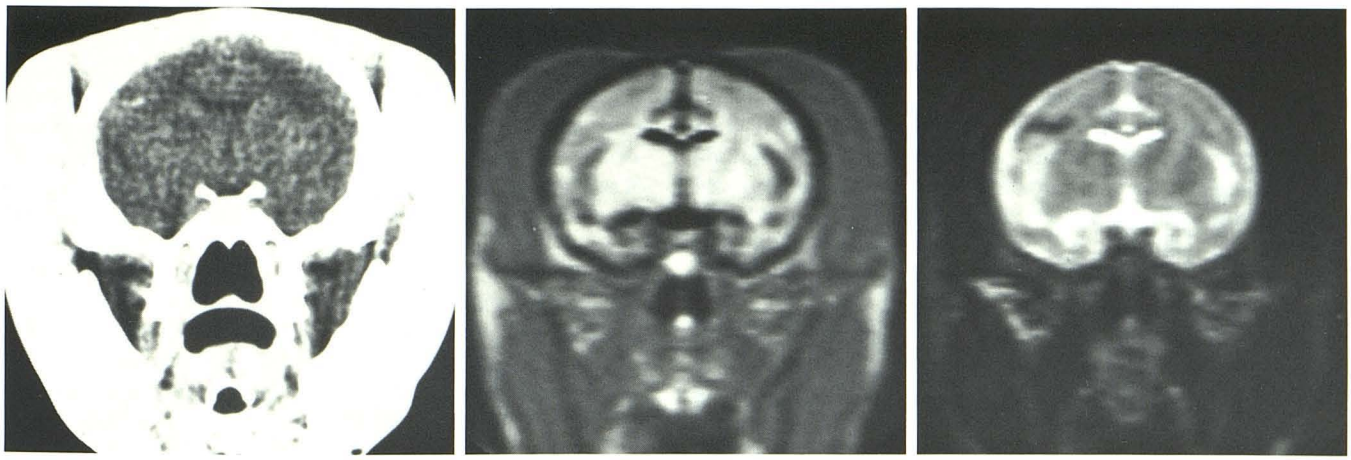


Day 15



2 Months

TABLE 1: **MR Pulse Sequences Used in This Study**

Sequence	Times in msec for Monkey 1 (Monkey 2)		
	Repetition Time	Echo Time	Inversion Time
Density-weighted	1700 (2500)	40	...
T1-weighted	1700 (2500)	40	400 (600)
T2-weighted	1700 (2500)	80 (120)	...

of scans included a T1-weighted pulse sequence, a T2-weighted sequence, and a density-weighted sequence (table 1). CT scans were obtained on a General Electric 9800 scanner.

Relaxation times were calculated by two methods: the Picker program for generating T1 and T2 images and a hand calculator programmed to solve the signal equations for T1 and T2. Regions of interest were selected at the Picker console for the hematoma, the surrounding edema, and contralateral white matter. The CT scans were used as a guide to ensure that the regions were properly chosen.

Bench studies were also performed on four samples of fresh blood placed in a test tube and incubated at 37°C for up to 10 days. Both heparin and EDTA were used as anticoagulants. MR relaxation times were measured in a Praxis II analyzer with a field strength of 0.25 T. To correlate relaxation times directly with methemoglobin concentration, another series of 10 samples was prepared with varying concentrations of sodium nitrite, a substance known to facilitate the conversion of hemoglobin to methemoglobin. The methemoglobin concentrations, blood gases, and T1–T2 values were then measured. As a control, sodium nitrite was also added to a water sample.

Results

In both monkeys, the hematoma remained visible for at least a month, surrounded by a halo of edema that helped to delineate the blood. Figure 1 shows the sequential CT and MR images of the second monkey. On the CT images the blood initially appeared as a hyperdense lesion that gradually returned to normal brain density. The surrounding halo was seen as a hypodense ring. On the inversion-recovery (IR), or T1-weighted, images, the blood initially appeared dark, but soon became bright relative to the contralateral brain tissue, finally returning to normal brain intensity. The surrounding edema was always dark. On spin-echo (SE), or T2-weighted, images, the situation was reversed. The blood initially appeared bright, becoming dark after several days, while the halo of edema always appeared bright. Later (10–14 days) another reversal took place in the T2-weighted images, so that the lesion center appeared hyperintense on both MR images. Finally, at 2 months, a transverse band of low signal intensity on both IR and SE images was noted as the only residue of the hemorrhagic lesion. Noteworthy is the appearance on the corresponding CT image of a hyperdense area quite similar in appearance and location to the hypointense MR area.

The opposite behavior of the two types of MR images in the early stage is consistent with the shortening of relaxation times T1 and T2 from the high values characteristic of normal blood. The calculated values of T1 and T2 are shown in figure 2. The values obtained from the Picker program agreed with the hand calculator values. Note that the relaxation times of the hematoma initially were longer than those of normal brain tissue, but showed a marked drop during the first 3–5 days. The T1 values remained somewhat low relative to contralateral tissue, while the T2 value reversed, becoming very large on day 15, consistent with the visual appearance previously noted. Not included in the graphs are the 2-month data, at which time the blood was completely resorbed and the relaxation rates were indistinguishable from those of normal brain tissue.

The results for monkey 1 (not shown) were similar in the general trends, although the lowering of T1 and T2 was not so marked.

The in vitro blood samples also showed a lowering of T1 and T2 with time. Figure 3 shows T1 values plotted versus time for a typical blood sample, compared with the in vivo results from one of the monkeys. The rate of change varied somewhat. Also shown in figure 3 is the expected relaxation time T1 that would be produced if all the hemoglobin were converted to the methemoglobin form. This point is calculated from published results of Koenig et al. [6], adjusting the data

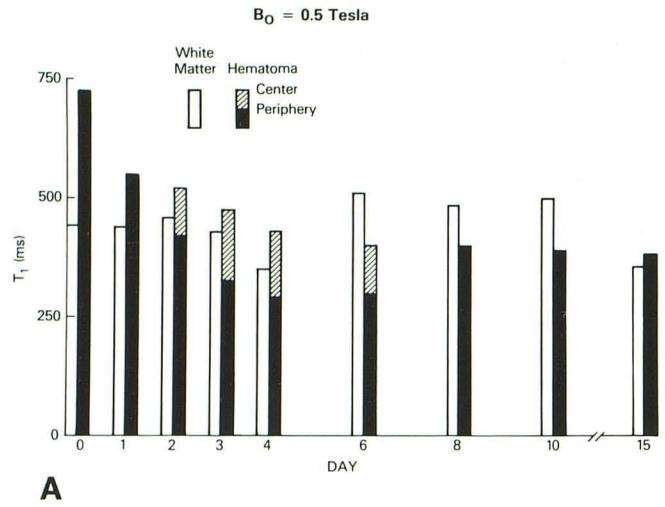


Fig. 2.—T1 (**A**) and T2 (**B**) values of hematoma and contralateral white matter for monkey 2 vs. number of days after injection of blood.

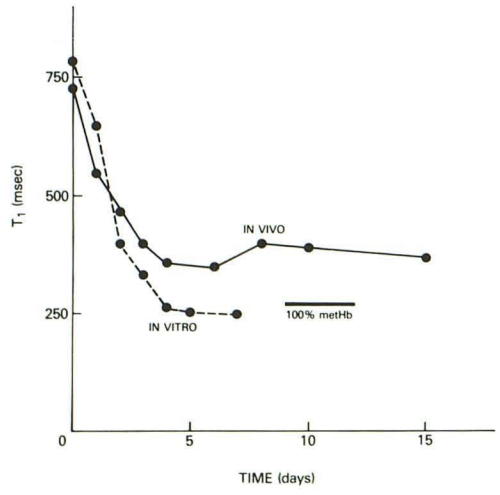


Fig. 3.—Relaxation time T1 vs. number of days after preparation for one of the in vitro samples (*broken line*) and for monkey 2 (*solid line*). *Thick horizontal line* represents expected end point assuming 100% conversion to methemoglobin [6].

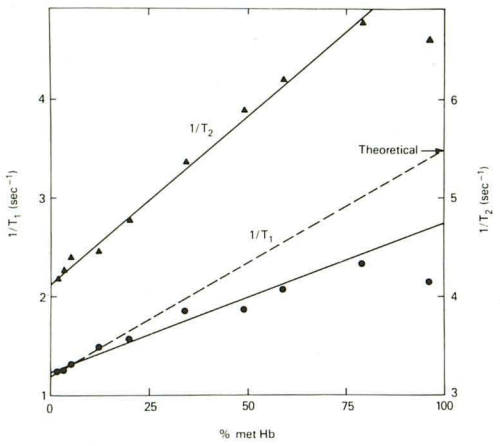


Fig. 4.—Relaxation rates 1/T1 and 1/T2 vs. methemoglobin concentrations (*solid lines*) for blood samples prepared with differing amount of sodium nitrite. Theoretical 1/T1 end point (*arrow*) is based on published data for methemoglobin [6]. *Broken line* fits low-dose points with theoretical end point.

for the estimated effect of differences in pH and temperature. We note that in vitro changes occur more quickly and approach more closely the expected end result than do in vivo changes.

The results for the samples doped with sodium nitrite are shown in figure 4. In this case we plotted the reciprocal of T1, called the relaxation rate, instead of T1 itself, because it is the rate 1/T1 that theoretically should vary linearly with the amount of methemoglobin formed. Note that the rates are approximately proportional to the methemoglobin concentration, except for the sample with the largest dose of sodium nitrite. Also shown in this figure is the estimated 1/T1 if all the hemoglobin were converted to methemoglobin, as above. Note that the first five or six data points extrapolate fairly well to this value, suggesting that the higher doses of sodium nitrite may have an additional effect on proton relaxation, apart from the conversion to methemoglobin. The sodium nitrite added to water had no effect on relaxation times, as expected.

Discussion

In all studies, both in vivo and in vitro, relaxation times of blood were initially longer than those of normal brain tissue, but became shorter within several days. This results in a period of "indistinguishability" during which the MR appearance of the blood is similar to that of normal brain tissue. In our studies, this was generally at about 24–48 hr. However, the factors affecting MR relaxation may well vary among actual clinical lesions, and it may be difficult to predict the actual window of indistinguishability.

The mechanism causing the early lowering of T1 and T2 is, by necessity, related to physiochemical changes in the extravasated blood. The conversion of hemoglobin to the methemoglobin form as a first step in denaturation, as well as the effect of methemoglobin on MR relaxation of surrounding

water protons, are both well known phenomena [6–8]. Indeed, our in vitro relaxation times correlated well with the measured or estimated methemoglobin concentrations. However, the in vivo relaxation times failed to reach the final value that would be expected if all of the hemoglobin were converted to the methemoglobin form. This may be partly due to other phenomena, such as resorption of hemoglobin and cell lysis.

The shorter relaxation times caused by denaturation of hemoglobin to methemoglobin occur because the structure of methemoglobin allows a closer approach of free water to the paramagnetic iron atom than does normal hemoglobin. However, the situation is complex, with "inner sphere" and "outer sphere" effects both contributing. Recent measurements on the field dependence of T1 have shed new light on the subject [6]. It is interesting to note that in our experiment the changes do not occur uniformly throughout the hematoma, but appear earlier in the peripheral part. This is particularly noticeable in some images (e.g., fig. 1, day 3). The reason for this effect is not clear. We can only speculate that the inner core of the hematoma is somehow protected from the chemical changes affecting the hemoglobin.

Starting around 6–8 days, the relaxation times of the hematoma begin to increase, particularly T2, which crosses over the normal brain value. Thus both T1- and T2-weighted MR images at this stage display a high-intensity signal. While these relatively late changes were not the main focus of our study, and no explanation is offered for them, it is highly unlikely that they are related to methemoglobin formation. By this time, the transformation to methemoglobin has either stopped or is no longer a factor in the image appearance. Finally, at 2 months, a low-intensity band is evident on the MR images, corresponding to a bright band on the CT image that suggests calcification. The concomitant MR effect at this stage may be due to calcium deposition as well as to long-term residue of iron, either as hemosiderin or protein-bound iron.

Our results should be applicable to all of the current high-field scanners because both diamagnetic and paramagnetic contributions to relaxation of hemoglobin are approximately independent of field for field strengths above 0.25 T. At much lower fields, for example, 0.02 T, the situation is different [4]. Here the nonparamagnetic relaxing effect of hemoglobin, caused by its thermal motion, is much greater, leading to shorter relaxation times. The paramagnetic contribution is also greater at these low fields, but since the relaxation times are shorter to begin with, the change in image appearance may not be as striking.

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