Brain Water Alterations After Unilateral Nephrectomy

DAVID C. ANDERSON, M.D., MARGARET M. JORDAN, B. CHEM., RONALD L. JACOBSON, PH.D., TÖRU HAYAKAWA, M.D., AND ARTHUR G. WALTZ, M.D.

SUMMARY White and regional gray matter distributions of water, blood flow, and the protein tracer pertechnetate were measured in five normal squirrel monkeys. A second group of five monkeys, which had undergone unilateral nephrectomy six months previously, were found at the time of study to have blood pressures similar to those of the control animals but increased brain water and altered distribution of blood flow which was increased in white matter. No alteration of capillary permeability to the protein tracer attended these changes, which appeared to be influenced by blood pressure. Nephrectomy without hypertension influences brain water content, perhaps because of an effect on cerebral resistance vessels. In hypertensive encephalopathy renal lesions, as well as intraluminal pressure changes, may be related to cerebral edema.

THE PATHOGENESIS of hypertensive encephalopathy has been subject to research attention for some time and has recently stirred considerable controversy. Most investigators are in agreement that excess brain water is a critical characteristic of clinical and experimental hypertensive encephalopathy. Keen interest in the behavior of the so-called "blood-brain barrier" has characterized most studies, which have been designed to demonstrate the escape of a surplus of brain water to the plasma-brain interface as demonstrated by such tracer leakage. Attention has been directed to the course of events leading to injury of the barrier to tracer.

The current study was undertaken with the objective of determining the relationships among water, tracer leakage, and blood flow in identical focal areas of brain in an animal model of chronic hypertension. Despite unilateral nephrectomy and wrapping of the remaining kidney, at the time of sacrifice, the experimental animals did not have blood pressures different on the average from those of control animals. The results, however, demonstrate the presence of increased brain water in the monkeys with nephrectomy and suggest that other factors besides increased intraluminal pressure may be important in experimental and clinical hypertensive encephalopathy. Furthermore, the data suggest that the basic assumption concerning water distribution and its relationship to tracer barriers may be incomplete since these data show increased brain water without evident barrier leakage of the protein tracer.

Methods

Five adult male squirrel monkeys (Saimiri sciureus) weighing 780-900 gm underwent unilateral right nephrectomy under sodium pentobarbital anesthesia (25 mg/kg) approximately six months prior to investigation. The remain-

The experiments were performed in the Cerebrovascular Clinical Research Center, Dept. of Neurology, University of Minnesota, Minneapolis, Minn. Investigation was supported by USPHS Grant NS-3364.

Dr. Hayakawa's present address is: Dept. of Neurosurgery, Osaka U., Osaka, Japan.

Dr. Waltz's present address is: Dept. of Neurology, Pacific Medical Center, San Francisco CA 94120.

Reprint requests to Dr. Anderson, Dept. of Neurology, Hennepin County Medical Center, 701 Park Ave. S., Minneapolis, MN 55415.
ten areas (bilateral frontal, parietal, temporal, and occipital cortical gray matter and parietal white matter) for a total of 120 samples from each animal. Water, technetium-99m, and 14C-antipyrine were measured in each of the samples.

Following determination of the wet weight of a sample, the activity of technetium-99m was measured in each sample and in the blood using a well counter. A brain/blood ratio for technetium-99m was calculated for each sample. The samples were then dried to constant weight by heating at 60°C for three days and exposure to a vacuum of less than 0.1 mm Hg for four hours. The difference between the wet and dry weights of the samples was assumed to be due to water loss. Water content was therefore measured gravimetrically and expressed as percent water using the formula: 

\[
\text{Percent Water} = \frac{\text{Wet Weight} - \text{Dry Weight}}{\text{Wet Weight}} \times 100
\]

Finally, the samples were dissolved in a tissue solubilizer (NCS, Amersham-Searle), and the activity of 14C-antipyrine was measured in a liquid scintillation counter. The timed serial arterial blood samples obtained during antipyre infusion were similarly prepared and counted, yielding an arterial curve for each monkey.

Cerebral blood flow for each monkey was calculated from these data using a modified technique of Reivich. In two monkeys, both controls, cerebral blood flow estimates were invalidated by technical difficulties so that only percent water and technetium-99m brain/blood ratios were available. Water content, pertechnetate, and flow data were available for all samples of the eight other monkeys. Statistical methods included separate analyses of variance for each parameter.

As indicated, blood pressure was monitored continuously over the period of preparation of each animal. A value for mean arterial blood pressure was obtained visually from the polygraph record of each monkey by five individuals and the estimates were averaged. Heart and kidney(s) of four nephrectomized and two control monkeys were removed for histopathological study.

### Results

Worthy of comment was the uniformity of all measured parameters within the sampled areas and within the nephrectomized and control groups. Variability, expressed as standard deviations (table 1), was small, reflecting the diffuse nature of the observed differences.

#### Blood Pressure

No significant fluctuations of blood pressure were observed in individual monkeys. Mean arterial blood pressures were not different in the two groups, ranging from 116 to 166 (mean 146) in the five control monkeys and 124 to 164 (mean 149) in the nephrectomized animals (table 2).

#### Water Content

Significantly greater water content was found in the white matter and all the gray matter regions sampled in the nephrectomized animals, compared with the corresponding

### Table 1. Average Water, Pertechnetate and Blood Flow in White and Regional Gray Matter of Nephrectomized and Control Groups

<table>
<thead>
<tr>
<th></th>
<th>Frontal</th>
<th>Parietal</th>
<th>Occipital</th>
<th>Temporal</th>
<th>White</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nephrectomized Monkeys</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Water (%</td>
<td>78.98</td>
<td>76.30</td>
<td>77.95</td>
<td>75.38</td>
<td>65.49</td>
</tr>
<tr>
<td>Pertechnetate (Brain/Blood Ratio)</td>
<td>0.055</td>
<td>0.054</td>
<td>0.058</td>
<td>0.049</td>
<td>0.049</td>
</tr>
<tr>
<td>Blood Flow (ml/gm/min)</td>
<td>1.12</td>
<td>0.96</td>
<td>0.73</td>
<td>0.79</td>
<td>0.38</td>
</tr>
<tr>
<td>Blood Flow (nep/serene)</td>
<td>0.11</td>
<td>0.12</td>
<td>0.08</td>
<td>0.16</td>
<td>0.09</td>
</tr>
<tr>
<td>Control Monkeys</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Water (%)</td>
<td>77.20</td>
<td>76.30</td>
<td>76.21</td>
<td>75.77</td>
<td>62.85</td>
</tr>
<tr>
<td>Pertechnetate (Brain/Blood Ratio)</td>
<td>0.065</td>
<td>0.051</td>
<td>0.060</td>
<td>0.047</td>
<td>0.044</td>
</tr>
<tr>
<td>Blood Flow (ml/gm/min)</td>
<td>1.41</td>
<td>1.28</td>
<td>1.03</td>
<td>1.04</td>
<td>0.31</td>
</tr>
<tr>
<td>Blood Flow (nep/serene)</td>
<td>0.25</td>
<td>0.15</td>
<td>0.18</td>
<td>0.16</td>
<td>0.07</td>
</tr>
</tbody>
</table>

*Pooled Sampling Error

### Table 2. Mean Water, Pertechnetate and Blood Flow in White and Gray Matter of Individual Monkeys

<table>
<thead>
<tr>
<th>Blood Pressure (mmHg)</th>
<th>Water (%)</th>
<th>Pertechnetate (Brain/Blood Ratio)</th>
<th>Blood Flow (ml/gm/min)</th>
<th>White Matter</th>
<th>Pertechnetate (Brain/Blood Ratio)</th>
<th>Blood Flow (ml/gm/min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nephrectomized Monkeys</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>124</td>
<td>77.83</td>
<td>0.050</td>
<td>0.59</td>
<td>63.62</td>
<td>0.044</td>
<td>0.27</td>
</tr>
<tr>
<td>161</td>
<td>79.10</td>
<td>0.056</td>
<td>1.18</td>
<td>67.64</td>
<td>0.055</td>
<td>0.47</td>
</tr>
<tr>
<td>159</td>
<td>76.17</td>
<td>0.054</td>
<td>0.83</td>
<td>62.43</td>
<td>0.040</td>
<td>0.28</td>
</tr>
<tr>
<td>164</td>
<td>79.52</td>
<td>0.059</td>
<td>0.92</td>
<td>67.03</td>
<td>0.058</td>
<td>0.45</td>
</tr>
<tr>
<td>158</td>
<td>78.54</td>
<td>0.050</td>
<td>0.97</td>
<td>66.71</td>
<td>0.045</td>
<td>0.41</td>
</tr>
<tr>
<td>Mean</td>
<td>78.63</td>
<td>0.054</td>
<td>0.90</td>
<td>65.49</td>
<td>0.049</td>
<td>0.38</td>
</tr>
<tr>
<td>S.D.</td>
<td>0.68</td>
<td>0.004</td>
<td>0.22</td>
<td>2.31</td>
<td>0.008</td>
<td>0.10</td>
</tr>
<tr>
<td>Control Monkeys</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>146</td>
<td>75.91</td>
<td>0.050</td>
<td>—</td>
<td>63.55</td>
<td>0.039</td>
<td>—</td>
</tr>
<tr>
<td>166</td>
<td>76.49</td>
<td>0.056</td>
<td>1.12</td>
<td>63.17</td>
<td>0.050</td>
<td>0.30</td>
</tr>
<tr>
<td>116</td>
<td>76.03</td>
<td>0.046</td>
<td>—</td>
<td>61.74</td>
<td>0.038</td>
<td>—</td>
</tr>
<tr>
<td>143</td>
<td>76.88</td>
<td>0.045</td>
<td>0.97</td>
<td>62.96</td>
<td>0.036</td>
<td>0.30</td>
</tr>
<tr>
<td>161</td>
<td>76.80</td>
<td>0.067</td>
<td>1.48</td>
<td>62.82</td>
<td>0.057</td>
<td>0.31</td>
</tr>
<tr>
<td>Mean</td>
<td>76.82</td>
<td>0.053</td>
<td>1.19</td>
<td>62.85</td>
<td>0.044</td>
<td>0.30</td>
</tr>
<tr>
<td>S.D.</td>
<td>0.78</td>
<td>0.009</td>
<td>0.26</td>
<td>0.68</td>
<td>0.009</td>
<td>0.01</td>
</tr>
</tbody>
</table>

--- Not Available

*Means Statistically Different from that of Controls (p < 0.05)
regions in the controls (table 1, fig. 1). The difference in water content between the two groups, expressed as percent water, was greater in white matter (65.49 - 62.85 = 2.64%) than gray matter (78.63 - 76.82 = 1.81%) (table 2). If the assumption is made that increased water content in the nephrectomized group represents an increment of water rather than a loss of solid constituents, then the associated relative volume increase (x) may be calculated by the formula: x = \[(\text{percentage dry weight control}) - \text{percentage dry weight nephrectomized})\] \times 100. Using the mean percentage dry weights for gray and white matter in the two groups, the calculated volumetric increments due to water increase in the nephrectomized group were 8.5 and 7.6 percent respectively. Intraregional sample variability was small in both groups (table 1), indicating a homogeneity of regional water distribution in the normal animals and a uniformity of the increment in the nephrectomized group.

In all animals the well-recognized difference between gray and white matter water content was confirmed. Additionally, regional (lobar) gray matter differences in water content were demonstrated in both control and nephrectomized animals. Frontal and temporal water content was significantly greater (p < 0.05) than parietal and occipital content in both groups (fig. 1).

Technetium-99m Brain/Blood Ratio

No significant differences between the control and nephrectomized monkeys were demonstrated for white or gray matter technetium-99m brain/blood ratios (table 2). In both groups the white matter tissue pertechnetate was lower than that in gray matter, and the differences were statistically significant (p < 0.05). Moreover, regional (lobar) gray matter differences in technetium-99m brain/blood ratio were found and were similar in the two groups (fig. 1). Tissue pertechnetate was slightly but significantly higher in the occipital lobes (p < 0.05), lower in the temporal lobes (p < 0.05), and intermediate in the frontal and parietal gray matter. Again, intraregional sample variability for tissue pertechnetate was small in both groups (table 1).

Blood Flow

Cerebral blood flow tended to be lower in the nephrectomized monkeys compared with the controls in all gray matter regions (table 1). However, while the expected difference between gray and white matter blood flow was substantiated in all animals, a trend for flow to be relatively higher in the white matter of the monkeys of the nephrectomized group was found (table 2). The ratio of blood flow in white matter relative to that in gray matter in each animal, [(mean white matter flow)/(mean gray matter flow)], reflects the difference between the two groups. In the nephrectomized animals the ratio averaged 0.42, compared with the average control ratio of 0.26. While the distribution of flow between gray and white matter was altered in the nephrectomized animals, within gray matter a similar relative interregional (lobar) flow distribution was found in the two groups (fig. 1). Frontal and parietal gray matter flow was significantly higher (p < 0.05) than temporal and occipital flow in both groups. Intraregional sample variability was small in both groups (table 1), suggesting that the uniformity of flow was not influenced by nephrectomy.

Water, Pertechnetate And Flow Correlations

Correlations between tissue water and pertechnetate, water and flow, and pertechnetate and flow were calculated within each region among monkeys. The results provide white and regional gray matter correlations between each pair of the three parameters in the nephrectomized and control groups (table 3). No consistent gray matter trends were demonstrated; in frontal gray matter only, water and pertechnetate were correlated in nephrectomized animals, while a negative relationship between water and flow was suggested in the control monkeys. In white matter, interestingly, a correlation between flow and water was demonstrated in the nephrectomized but not the control monkeys.

**Table 3 Water, Pertechnetate and Blood Flow Correlations in White and Regional Gray Matter Among Monkeys of Nephrectomized and Control Groups**

<table>
<thead>
<tr>
<th>Group</th>
<th>n</th>
<th>Gray Matter</th>
<th>White Matter</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Frontal</td>
<td>Parietal</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Water</td>
<td>Pertechnetate</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.9±</td>
<td>0.4±</td>
</tr>
<tr>
<td></td>
<td></td>
<td>-0.8</td>
<td>-0.2</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.6±</td>
<td>0.8±</td>
</tr>
<tr>
<td></td>
<td></td>
<td>-1.0±</td>
<td>0.0</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.4±</td>
<td>0.6±</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.1</td>
<td>1.0</td>
</tr>
</tbody>
</table>

*Significantly different from zero (p<0.05)

n = Nephrectomized

c = Control
Blood Pressure Correlations

In both gray and white matter, mean water content was directly related to mean arterial blood pressure in the nephrectomized group (figs. 2 and 3). In contrast, no relationship between the two parameters was evident in gray or white matter of the control monkeys. Relationships between mean arterial blood pressure and tissue pertechnetate and between blood pressure and blood flow appeared to be similar in the two groups in both gray and white matter.

Histopathology

The hearts of nephrectomized monkeys demonstrated no microscopic evidence of sustained hypertension. The remaining cellophane-wrapped kidneys of the nephrectomized animals were found to have thickened fibrotic capsules with modest infiltration of inflammatory cells and occasional giant cells. Hemosiderin deposits were present in the perinephric fat. A few scattered foci of chronic interstitial inflammation were found in the kidneys of animals of both groups.

Discussion

Clinical encephalopathy characterized by diffuse and multifocal cerebral symptoms and signs, accompanied by evidence of increased intracranial pressure (papilledema and elevation of manometric cerebrospinal fluid pressure), is seen in the setting of rapidly developing hypertension (e.g., eclampsia) or exacerbation of more longstanding hypertension. In patients dying during a hypertensive crisis, generalized edema, with or without areas of focal encephalomalacia and blood extravasation, has been described pathologically. The roles of edema and increased intracranial pressure in clinical dysfunction are not clearly understood. Moreover, the characteristics and origin of hypertension-related surplus brain water have not been fully elucidated. That chronic maladjustment of water distribution may exist in hypertension has been suggested by both human pathological studies and some experimental models.

In a series of autopsy specimens from patients with chronic hypertension in life, Adachi found water content, measured gravimetrically, to be increased, especially in white matter. Pathological findings in chronic hypertension also suggest longstanding edema of similar distribution, as described by Feigin.

Experimentally, Byrom demonstrated presence of excess water in relationship to areas of focal extravasation of trypan blue in nephrectomized rats made acutely more hypertensive with pressor agents. However, he also described in the same model, a diffuse water increment unrelated to tracer-identified vascular leakage. Meinig found a similar diffuse increase in water content confined primarily to white matter following induction of hypertension in dogs. The increase in water content depended on the duration of sustained pressure elevation and did not correspond to areas of Evan's blue dye extravasation, which occurred cortically.

Rosenblum found increased brain water in chronically hypertensive nephrectomized mice treated with DOCA and salt. The excess fluid appeared to be located in both gray and
chronic hypertension has been produced, usually by various pharmacologically-induced pres-encephalopathy have been observed to develop either spontane-ously or with further pharmacologically-induced pres-encephalopathy have been observed to develop either spon-

increase of brain water in monkeys with nephrectomies despite without renal lesions.
cumulation have been described in models of acute hyper-
tension, usually pharmacologically produced. 8-17,19,22

Experimental hypertension is therefore difficult to separate to that described in autopsied hypertensive human brains have, for the most part, however, been those in which chronic hypertension has been produced, usually by various renal manipulations. 8, 8, 13 Signs consistent with encephalopathy have been observed to develop either spontane-ously or with further pharmacologically-induced pres-

Barriers to solute and colloid. The "breakthrough" theory7,14-22 suggests that hypertension produces vasodilatation because intraluminal pressures exceed those that can be compen-sated for by autoregulatory mechanisms, resulting in a loss of integrity of the "blood-brain barrier" and edema. Both concepts imply that fluid ingress to the parenchyma follows, causally and temporally, an acute disruption of vascular barriers to solute and colloid.

Models demonstrating generalized brain swelling similar to that described in autopsied hypertensive human brains have, for the most part, however, been those in which chronic hypertension has been produced, usually by various renal manipulations. 6, 8, 13 Signs consistent with encephalopathy have been observed to develop either spontane-ously or with further pharmacologically-induced pres-

Difficult to reconcile with these considerations are reports that the brains of animals with experimental uremia26 and autopsied normotensive uremic human brains are not edematous.29

The second possibility, that cerebral regulatory vessels fail to dampen central arterial pressure for protection of capillaries following nephrectomy, would be consistent with the results. Such a pathophysiological mechanism would result in increased intraluminal hydrostatic pressure at the capillary level. The result would represent an abortive "breakthrough" phenomenon stopping short of endothelial disruption and solute permeability alteration. The data suggest that distribution of blood flow was altered in the nephrectomized animals with a relatively (and absolutely) greater flow in white matter. The apparent maldistribution of flow may be a manifestation of a primary effect of nephrectomy on cerebral resistance vessels.

Finally, the excess cerebral water might be attributed to a failure of membrane homeostasis in the brain, rather than a lowering of the vascular threshold for transudation, resulting in diffuse intracellular accumulation of water.24,25 Such a mechanism, designated "cytotoxic" edema by Klatzow, 6 would not involve enhancement of transendothelial passage of solutes or colloids. Effects would be most pronounced in gray matter. The direct relationship of water content with mean arterial blood pressure suggested by the study would not be consistent with this mechanism. The data are in best agreement with the second postulated mechanism above.
Acknowledgment

The authors gratefully acknowledge the technical contribution of Mr. Terry Hansen and the expert assistance of Dr. Angeline Mastri who examined the pathological material. We are indebted to Ms. Marilyn Sullivan who typed the manuscript.

References

Brain water alterations after unilateral nephrectomy. A study of regional circulatory factors in squirrel monkeys.

D C Anderson, M M Jordan, R L Jacobson, T Hayakawa and A G Waltz

*Stroke*. 1977;8:462-467
doi: 10.1161/01.STR.8.4.462

*Stroke* is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 1977 American Heart Association, Inc. All rights reserved.
Print ISSN: 0039-2499. Online ISSN: 1524-4628

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://stroke.ahajournals.org/content/8/4/462