INTERFERENCE OF SCALP AND SKULL WITH EXTERNAL MEASUREMENTS OF BRAIN ISOTOPE CONTENT: PART 1. ISOTOPE CONTENT OF SCALP AND SKULL

William H. Oldendorf and Youichi Iisaka

Wadsworth Hospital, Veterans Administration Center
and UCLA Center for the Health Sciences, Los Angeles, California

Although interference from overlying tissues complicates all organ counting, it is to be expected that superimposed scalp and skull tissue isotope will be a particularly important source of error in quantitative studies of brain because of the very low concentration most tracers achieve in brain tissue due to the blood-brain barrier. Experimental animal and radiographic human studies were undertaken to define the magnitude of scalp and skull isotope content relative to brain for two brain-scanning agents, representative of small and large labeled molecules. Such an estimate requires a knowledge of the tissue isotope concentrations and the relative weights of the cranial tissue compartments. Tissue volumes and weights of brain, scalp and skull were estimated from postmortem radiography in ten humans. The ^{99m}Tc-pertechnetate and ^{131}I-IHSA content of rabbit whole blood, scalp, skull, brain and muscle were defined as a function of time. In a separate study blood volumes of the animal tissues were calculated using ^{125}I-IHSA, and its contribution to each total tissue count was estimated.

From these animal and human data, the extent of scalp and skull interference with external-brain isotope measurements was estimated for clinical studies using collimation designed for total cranial counting. However, the basic data is applicable to other counting geometries. A technique for minimizing the scalp and skull isotope contribution is presented.

METHODS

Radiographic estimation of human cranial tissue volumes. To estimate the volumes of the cranial cavity, calvarial skull and scalp in humans, radiographs were taken of ten unembalmed male human heads postmortem. Separate radiographic exposure techniques for skull and scalp detail were used with a 180-cm tube distance. Correction for linear magnification was made.

An estimate of the skull and scalp volume covering the hemispheres, rostral to a plane passing through the orbital roofs and internal occipital protuberance, was made by considering the skull and scalp to be hemispheric surfaces whose mean radii and thicknesses were calculated from radiographic measurements at the reference points indicated in Fig. 1. The volume of cranial cavity rostral to this plane was estimated by calculating the hemispheric volume using the mean interior radius of the skull. This approximates the brain volume rostral to the floor of the anterior and middle cranial fossae and the tentorium cerebelli.

The estimated volumes will be only approximate because of the irregular shape of the skull, but they will provide some basis for estimating the absolute isotope content of these cranial tissues. Some of the inferior portion of the temporal lobes will be excluded from these estimates.

Isotope distribution in animal tissues. New Zealand white rabbits (3–4 kg) were given 100–200 μCi of ^{99m}Tc-pertechnetate or 20 μCi of ^{131}I-IHSA intravenously.* At various intervals afterwards the animals were decapitated after pentobarbital anesthesia. The radioactivity per unit weight of tissue was determined in whole blood, calvarial scalp, lumbar skin, lumbar paraspinous muscle, brain and calvarial skull. Cerebrospinal fluid (CSF) was obtained by cisternal
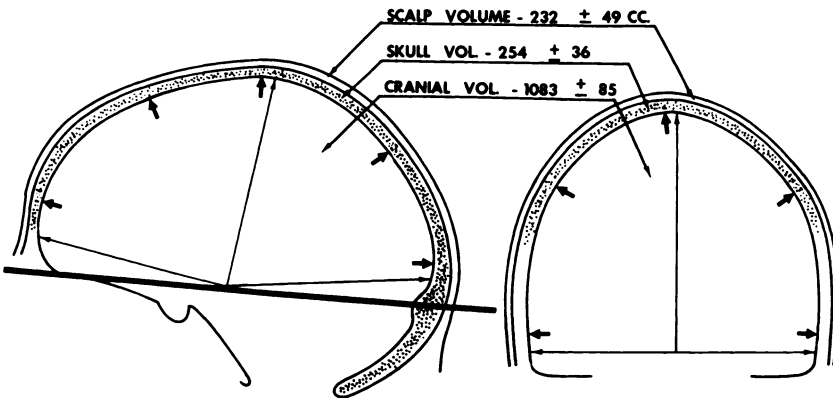
FIG. 1. Cross-section shows points (heavy arrows) at which skull and scalp thickness were measured on radiographs. Only tissues rostral to heavy line were considered.

puncture just before decapitation. The hair was clipped from the scalp and skin specimens.

Correction for tissue blood content was obtained in ten animals by intravenous injection of ^{125}I-IHSA made 1 min before decapitation as discussed by Matthews (1). It was assumed that at the end of 1 min (at decapitation) the radiolabel was confined to the blood pool of the tissues and that reasonably complete intravascular mixing had occurred. The ^{125}I-IHSA content of whole blood, scalp, lumbar skin, lumbar paraspinous muscle, brain and calvarial skull was determined and used to calculate the percent blood volume in each tissue specimen.

The counts of ^{125}I-IHSA per gram of tissue divided by the counts per gram of blood multiplied by 100 was taken as the percentage of tissue which was blood. The percentage is assumed to remain constant throughout any one experiment. The contribution of blood isotope to the tissue count will remain constant if the blood concentration remains constant. There is a small error resulting from the use of a plasma label such as IHSA since this will define the plasma content of the tissue. Because the plasma volume of a tissue does not accurately represent its whole-blood volume due to the lowered microcirculatory hematocrit, the estimation of tissue blood volume from tissue blood-plasma content relative to a whole-blood specimen will result in a somewhat high estimation of tissue blood content. To correct for this would require equal labeling of red cells and plasma, and this was not done here.

RESULTS

The tissue volumes of human scalp, skull and cranial cavity determined postmortem by radiographic measurements are shown in Fig. 1. The percentage of total cranial tissue volume and weight made up of scalp, skull and brain is given in Table 1. The averages for skull and scalp thicknesses are shown in Table 2.

TABLE 1. PERCENTAGE OF CRANIAL TISSUE VOLUMES AND WEIGHTS ESTIMATED RADIOGRAPHICALLY IN MAN

Cranial tissue compartments	Estimated volume of cranial tissues (cc)	Percentage of cranial volume	Estimated weight of cranial tissues (gm)	Percentage weight of total cranial tissues
Calvarial scalp	232 ± 50	15.0	232	13.0
Calvarial skull	254 ± 36	16.0	457	25.8
Cranial cavity	1,083 ± 85	69.0	1,083	61.1
Total	1,569 ± 65	100.0	1,772	100.0

Tissue weights were estimated from tissue volumes assuming a soft-tissue density of 1 and a bone density of 1.8. Skull probably has a density somewhat less than this because of its numerous diploe.

TABLE 2. RADIOGRAPHIC ESTIMATION OF CRANIAL SKULL AND SCALP VOLUMES IN MAN

	Average thickness (T) of:		Average radius (r) of:		Volume (V) of:	
	skull (mm)	scalp (mm)	skull (cm)	scalp (cm)	skull (cm³)	scalp (cm³)
Average*	5.96	4.85	8.26	8.83	254	232
Standard deviation	±0.69	±1.06	±0.18	±0.15	±36	±49

* Each average represents results of ten studies as shown in Fig. 1.

The percent isotope content of ^{99m}Tc-pertechnetate and ^{131}I-IHSA in scalp, skull, brain, lumbar skin, muscle and CSF relative to blood as a function of time after injection is shown in Fig. 2A and 2B and in Table 3. The ratio of isotope concentrations in scalp and skull to brain is indicated in Fig. 3 and Table 4.

No correction for blood content has been made in Figs. 2 and 3 and Tables 3 and 4. Since these

TABLE 3. PERCENTAGE TISSUE RADIOACTIVITY RELATIVE TO WHOLE BLOOD OF 99mTc-PERTECHNETATE*
AND 131I-IHSA† (TISSUE/BLOOD × 100)

	99mTc-pertechnetate				
Tissue	5 min	1 hr	2 hr	6 hr	24 hr
Whole blood	100.0	100.0	100.0	100.0	100.0
Scalp	34.2 ± 7.3	58.7 ± 6.5	46.4 ± 7.8	54.3 ± 12.0	69.1 ± 7.2
Lumbar skin	27.7 ± 7.5	53.8 ± 7.0	43.0 ± 8.9	47.6 ± 3.6	51.3 ± 10.2
Muscle	5.5 ± 1.0	8.1 ± 0.6	6.3 ± 1.3	7.8 ± 1.6	6.9 ± 2.5
Skull	14.0 ± 5.1	31.7 ± 2.1	28.7 ± 6.5	25.4 ± 8.3	34.3 ± 6.8
Brain	2.2 ± 0.3	3.7 ± 0.8	3.3 ± 0.4	4.6 ± 1.9	4.8 ± 1.3
CSF	0.17 ± 0.05	0.51 ± 0.05	0.46 ± 0.17	0.52 ± 0.21	0.37 ± 0.34

	131I-IHSA				
Tissue	10 min	1 hr	2 hr	24 hr	48 hr
Whole blood	100.0	100.0	100.0	100.0	100.0
Scalp	5.6 ± 1.2	8.3 ± 3.7	11.9 ± 4.5	24.8 ± 4.4	32.2 ± 3.7
Lumbar skin	3.1 ± 0.1	4.3 ± 0.1	—	18.1 ± 5.3	27.2 ± 9.9
Muscle	2.2 ± 1.4	2.5 ± 1.0	1.8 ± 0.7	4.8 ± 1.8	5.4 ± 1.0
Skull	(6.0 ± 1.0)	—	—	14.0 ± 4.9	20.3 ± 1.2
Brain	1.7 ± 0.3	2.5 ± 0.2	2.3 ± 0.1	2.5 ± 0.6	2.1 ± 0.3
CSF	0.05 ± 0.04	0.16 ± 0.50	1.05 ± 0.13	0.73 ± 0.16	0.80 ± 0.17

* Each figure represents an average of five animals. Isotope was injected (100–200 μCi), the animals decapitated at various time intervals thereafter and the activity/unit weight of tissue determined.

† Each figure represents an average of five animals. Isotope was injected (20 μCi), the animals decapitated at various time intervals thereafter and activity/unit weight of tissue determined.

TABLE 4. RATIO OF TISSUE COUNT/BRAIN COUNT OF 99mTc-PERTECHNETATE AND 131I-IHSA

	99mTc-pertechnetate				
Tissue	5 min	1 hr	2 hr	6 hr	24 hr
Scalp	16.5 ± 2.0	16.3 ± 2.3	13.9 ± 2.0	13.2 ± 3.2	13.1 ± 1.8
Skull	6.0 ± 1.9	8.8 ± 1.1	8.8 ± 1.6	6.8 ± 3.6	6.5 ± 1.4
Muscle	2.7 ± 0.2	2.3 ± 0.4	1.9 ± 0.3	1.9 ± 0.5	1.2 ± 0.4

	131I-IHSA				
Tissue	10 min	1 hr	2 hr	24 hr	48 hr
Scalp	3.3 ± 1.1	2.7 ± 1.2	5.1 ± 1.8	10.4 ± 1.5	15.5 ± 2.8
Skull	3.7 ± 0.6	—	—	6.4 ± 2.7	10.3 ± 0.5
Muscle	1.3 ± 0.8	0.9 ± 0.5	0.8 ± 0.3	1.8 ± 0.7	2.6 ± 0.5

TABLE 5. TISSUE BLOOD CONTENT OF 125I-IHSA*

Tissue	Scalp	Skin	Muscle	Skull	Brain
% blood volume	1.56 ± 0.13	0.92 ± 0.07	0.56 ± 0.04	6.00 ± 0.33	1.80 ± 0.09

* Ten animals: 10 μCi/injection. Specimens obtained immediately after decapitation 1 min after injection.

tissues were obtained after decapitation, some of the blood present during life will have been lost. This method of termination was elected, however, because time of death could be determined precisely, and the circumstances of death would be uniform from animal to animal. Percentage of blood volume contributed to each tissue using 125I-IHSA is shown in Table 5.

DISCUSSION

The human brain seems the most ideally suited of all the major body organs for studying gamma emitters by external measurement. The brain is situated well away from other major organs, allowing considerable isolation using collimated detectors. Moreover, it is the only organ that can be surrounded in isolation by multiple detectors so that absolute de-
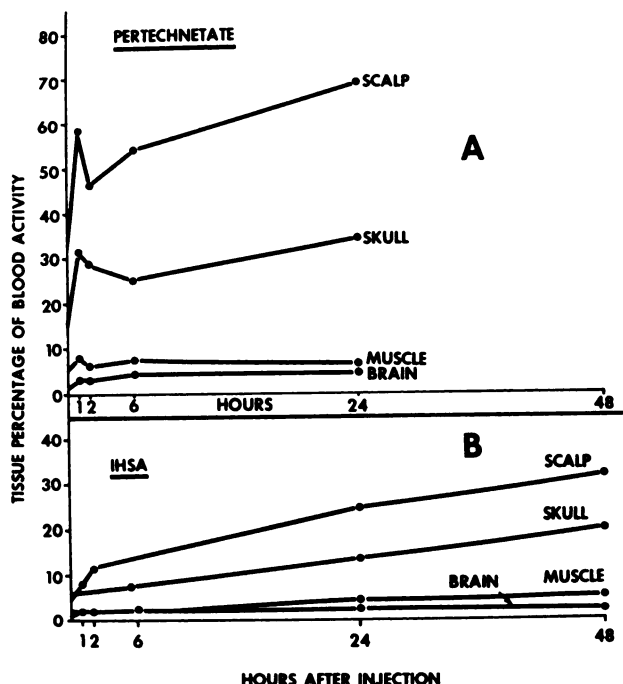
FIG. 2. Concentrations of ⁹⁹ᵐTc-pertechnetate and ¹³¹I-IHSA relative to blood in various cranial tissues after single intravenous injection. No correction is made for blood content of isotope in various tissues.
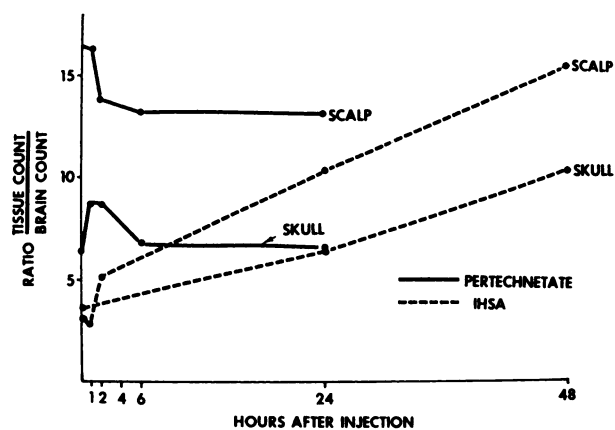

FIG. 3. Ratio of tissue concentration relative to brain after single injection of ⁹⁹ᵐTc-pertechnetate and ¹³¹I-IHSA. No correction is made for blood content of isotope in tissues. If correction for blood content had been made, ratios would be considerably greater since most of brain content is present in blood of brain.

tector counting efficiency can be established. Attempts at quantification of other total organs are more complex. The liver (2), for example, cannot be isolated from other nearby organs, nor can its position relative to the detector be predicted.

Despite its apparently advantageous location, the brain is surrounded by a 3–7-mm layer of non-neural soft tissue composed of scalp and a small amount of muscle as well as a layer of bone. Table 1

and Fig. 1 indicate that the soft tissue covering the cerebrum constitutes about 232 cc while the skull makes up about 254 cc. Assuming a soft-tissue density of approximately 1.0 and a bone density of approximately 1.8 (3), this represents about 232 gm of soft tissue and 457 gm of skull. The isotope content in brain is seen against a background count originating in these superficial tissues.

The degree of interference of these superficially-originating counts with the count from brain depends on the relative isotope concentrations in scalp, skull and brain. The total weight of scalp and cranial skull (689 gm) is nearly as great as the underlying supratentorial brain (1,083 gm). Therefore if the isotope concentrations achieved in these superficial tissues approaches that in brain, this superficial source of count becomes formidable. In our work the ratio of animal scalp tissue concentration to brain concentration never was less than about 3 (IHSA at 10 min after injection) and later was much higher (Fig. 3 and Table 4). Because of these low relative concentrations in brain, the isotope in the overlying tissues becomes of great significance.

The percentage of total cranial count originating in supratentorial brain will be

$$100 \times \frac{\text{Brain wt} \times C}{(\text{Brain wt} \times C) + (\text{Skull wt} \times C) + (\text{Scalp wt} \times C)}$$

in which C is the concentration of label per unit weight (wt) of tissue. Given approximate minimum 10-min (Table 4) scalp and skull ¹³¹I-IHSA concentrations (3.3 and 3.7 × brain), the percentage of the total count from the cranial portion of the head of man which originates in brain is calculated as 30.6%. This percentage assumes 1,083 gm of supratentorial brain, 232 gm of scalp and 457 gm of skull. Using the 5-min tissue-to-brain ratios (Table 4) achieved with ⁹⁹ᵐTc-pertechnetate (16.5 and 6.0 for scalp and skull) the percentage which is brain is only 14.1%.

These percentages (30.6 and 14.1) are valid only when the detection system sees the entire head rostral to the floor of the cranial cavity with uniform counting efficiency. With other detection geometries such as focused collimators, the influence of the tissues external to brain usually will be somewhat less.

It is clear from these calculations that the scalp and skull constitute serious sources of interference with attempts to quantify brain content of isotopes by external counting. Several studies have attempted to use such counting of brain regionally (4–6) and of the total cranium (7,8). In these studies the scalp and skull will produce substantially more

180

counts than the underlying brain. External counting of this isotope *in situ* will be complicated by the shielding effect of skull (9) and the inverse-square effect which will further emphasize the surface count. Absolute quantification of the brain isotope content will be complicated even further by internal absorption of gamma radiation by brain tissue. This latter factor will emphasize the count originating near the surface of the cerebral hemispheres and will be minimized by using higher-energy gamma emitters.

If the entire head rostral to the floor of the cranial cavity is counted, the scalp and skull surrounding the vertex will contribute to the total count. If the collimation were restricted to a horizontal slice perhaps 5 cm thick (Fig. 4), this vertex scalp and skull would not be included but very little brain would be excluded. This would improve the brain-to-nonbrain tissue count ratio. This kind of collimation would also improve the reproducibility of counts because the count will be less a function of how far into the detector field the head is positioned (Fig. 4). If, for example, a patient with a constant cranial content of ^{131}I-IHSA is repeatedly counted in the cranial detector with complete repositioning between counts, a variability of several percent above counting statistical variations is experienced.

From Table 5 it is clear that blood is a major source of counts from all the cranial tissues immediately after injection. This will be particularly true for IHSA which equilibrates with extracellular fluid much more slowly than pertechnetate. This is easily corrected in total cranial counting by counting serial blood specimens (7). Assuming uniformity of blood isotope distribution, a correction for blood isotope can be made by assuming that the cranial blood volume remains constant in any one subject during one test. One can calculate net tissue uptake by subtracting the figures for tissue blood volumes of Table 5 from the figures for total tissue uptake relative to blood in Table 3.

The scalp contains approximately 15 times the ^{99m}Tc-pertechnetate concentration as brain during the first 24 hr. Correcting for blood content raises this ratio to about 23. Skull contains about six times as much pertechnetate as brain at 5 min (Table 4). The blood of scalp, skull and cerebral cortex and the scalp and skull content of isotope described here adequately explain the "halo" seen about the cranial cavity in high-resolution scanning particularly when using ^{99m}Tc-pertechnetate. The halo about the cranial cavity is particularly prominent because of the elongated acceptance field of most focused collimators which causes a considerable thickness of scalp and skull to be counted when the collimator sees these tissues tangentially.
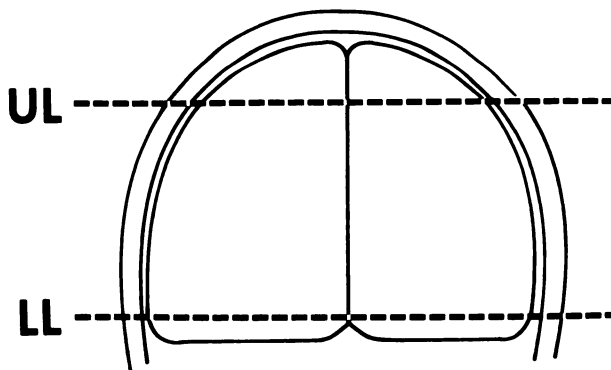

FIG. 4. If all of tissues rostral to floor of cranial cavity (LL) are counted, scalp and skull overlying superior surface of brain will contribute to count, thereby reducing brain-to-non-neural tissue ratio seen by detectors. By shielding out region of head near vertex (above UL) so that flat, horizontal slab of tissue perhaps 5 cm thickness is seen by detector, one gets a greater relative representation of brain. Shielding would also make positioning of head less critical.

The high ^{99m}Tc content of scalp and lumbar skin found here (Table 2) is in keeping with results previously reported in which the skin of the rat contained about one-third of the administered pertechnetate 35 min after injection (10). The high ^{99m}Tc content of skin has also been described elsewhere (11,12). The large lumbar skin and scalp ^{99m}Tc-pertechnetate content found here is compatible with a largely extracellular distribution. The volume of skin extracellular space (ECS) is large. An inulin space of 35.4% in dog skin has been reported (13), and a sulfate space of 70.6% has been described in chicken skin (14). Extracellular pertechnetate equilibrates rapidly with plasma while the IHSA equilibrates more slowly. Tissue extracellular IHSA will never come into equilibrium with plasma and is therefore a poor extracellular compartment label. Nevertheless, even at 10 min after injection, the scalp concentration of IHSA is already three times brain. It is unlikely that this represents entirely the tissue blood content; it suggests that the IHSA has already come into partial equilibrium with scalp extracellular space or capillary wall intracellular space, perhaps being incorporated into pinocytotic vesicles.

The scalp and skull are particularly serious sources of interference in brain isotope counting because the brain concentration of most externally introduced solutes remains low. This is largely because of the extreme impermeability of brain capillaries to most nonlipid soluble substances. A further factor which keeps the concentration of brain extracellular foreign solutes low is the sink action of cerebrospinal fluid (CSF) (15,16), which removes these foreign substances from brain parenchyma and transports them by bulk flow back into blood. The CSF is best

considered an extension of brain ECS. Low capillary permeability and sink action of cerebrospinal fluid are two of the mechanisms which the brain uses to maintain an ultrastability of its extracellular fluid. Since any of the labeled compounds we are likely to use in brain scanning will be "foreign" in the sense that the brain has no reason to want them normally in its extracellular fluid, we can anticipate that these labels will achieve very low isotope concentrations in brain tissue. This would not be the case with lipid soluble tracers or with metabolites such as glucose and amino acids labeled in a way that maintains their chemical identity. Specific carrier systems are present in brain capillaries to transport these metabolites (17,18) and high brain-tissue concentrations of these substances can be achieved.

In studies of the passage of a nondiffusible intravenous bolus of gamma emitter through the brain blood pool (19–21), scalp and skull interference will be minimal since, of all the blood passing through the cranial region of the head, most will pass through brain and relatively little through scalp and skull. The magnitude of the extent of equilibration with scalp ECS within the brief span of bolus passage is unknown. Tracers of low molecular weight such as iodohippurate (MW 304 in solution) may come rapidly into equilibrium with scalp ECS, and this could be nearly complete within the few seconds required for bolus passage through the head (22). Since the scalp ECS evidently is much larger than the scalp blood-plasma compartment, most of the tracer would be in the scalp ECS at equilibrium. This probably would be an advantage in brain blood-pool measurements by the first derivative method (23) since this scalp extracellular isotope would not contribute significantly to the negative peak of the first derivative.* The deposition of a large proportion of the scalp blood-flow tracer in the scalp ECS would thus minimize scalp blood-flow interference with measurements of brain blood flow.

---

* The cranial counting-rate curve following an intravenous rapid injection probably will remain at a higher plateau level when one uses iodohippurate or pertechnetate than when one uses a less-diffusible tracer such as IHSA because of partial equilibration of plasma small molecule tracers with scalp ECS. The IHSA would not achieve any measurable equilibrium with scalp ECS within the few seconds required for bolus transit and would remain intravascular. In the brain blood pool both of these tracers will remain intravascular. Thus, if plasma iodohippurate or pertechnetate come into equilibrium with scalp ECS during bolus passage, they would wash out of this extracellular compartment approximately monoexponentially and much more slowly than the intravascular brain bolus would leave the head. This would alter the rate of fall of total cranial counting rate very little. The entrance rate of the bolus into the head, however, would be slightly distorted. If this reasoning could be experimentally documented, it would suggest that the first derivative pool-transit-time method largely excludes non-neural tissues.

Clinical studies of blood-brain barrier by total brain counting would be of considerable interest if a correction could be made for scalp and skull uptake. One possible method for achieving this would be to use a double-tracer technique in which ^{131}I-IHSA and ^{125}I-IHSA were given simultaneously. The ^{125}I gamma emissions will be 60–80% absorbed by skull (9). Its soft-tissue half-thickness (2 cm) and low concentration in brain will confine its counting largely to the scalp and outer surface of skull. The count derived from ^{131}I-IHSA will be representative of all cranial tissues. Subtracting the ^{125}I-IHSA count should allow a reasonably complete correction for tissues superficial to brain.

In semiquantitative routine clinical brain scanning where only gross regional differences are sought, the interference from scalp and skull isotope is not prohibitive because it constitutes uniform background. It can be subtracted from the scan image electronically during scan production or mentally during scan interpretation. The double-tracer technique noted above might be used in regional scanning to subtract this superficially originating background and thereby let one show more subtle regional differences in brain isotope. A suitable combination of isotopes for regional brain scanning might use ^{123}I- and ^{125}I-labeled compounds (or perhaps free I$^-$) having a disappearance from blood similar to pertechnetate which might also be used for subtraction during routine technetium scanning.

SUMMARY

Tissue volumes and weights of brain, scalp and skull were estimated from postmortem radiography in ten subjects. The isotope distribution in animal tissues for ^{99m}Tc-pertechnetate and ^{131}I-IHSA were defined as a function of time. The brain isotope concentration was low relative to other tissues. Blood volumes were calculated using ^{125}I-IHSA in the animal tissues. The percentage of total cranial counts originating in brain were calculated for a total cranial head counting geometry. Restriction of collimation in total-head counting to exclude the vertex scalp and skull would improve the brain-to-nonbrain tissue count ratio. Isotope data described here suggest an explanation for the "halo" seen on ^{99m}Tc head scans. A difference was shown between calvarial scalp and lumbar skin isotope content for both ^{99m}Tc-pertechnetate and ^{131}I-IHSA. Brain mechanisms for keeping most "foreign" or externally introduced solutes low in concentration are briefly discussed. The effect of scalp and skull on external counting of a nondiffusible bolus passage is discussed in relation to the first derivative method of measuring brain blood pool transit time. A tracer technique is sug-

gested which would allow correction for the superficially originating count in scalp and skull.

REFERENCES

1. MATTHEWS, C. M. E.: The albumin pool. In *Compartments, Pools, and Spaces in Medical Physiology*. Eds. P. E. Bergner and C. C. Lushbaugh. USAEC Conf. 661010, 1967, p. 229.

2. OSBORN, S. B., ROBERTS, C. N. AND WALSHE, J. M.: Uptake of radiocopper by the liver. A study of patients with Wilson's disease and various control groups. *Clin. Sci.* 24:13, 1963.

3. *Clinical Dosimetry*, NBS Handbook 87, Washington, D.C., U.S. Government Printing Office, 1963.

4. WHITTIER, J. R., JACOBSON, L. E. AND DHRYMIOTIS, A.: Age differences in routine scans of human brain with Arsenic-74. *J. Nucl. Med.* 5:134, 1964.

5. TAKAHASHI, M., NOFAL, N. M. AND BEIERWALTES, W. H.: Correlation of brain scan image and area counting after scanning with tumor pathology. *J. Nucl. Med.* 7:32, 1966.

6. TAUXE, W. N.: 100-level smoothed scintiscans processed and produced by a digital computer. *J. Nucl. Med.* 9:58, 1968.

7. OLDENDORF, W. H. AND KITANO, M.: Clinical measurement of brain uptake of radioisotope. *Arch. Neurol.* 9:574, 1963.

8. OLDENDORF, W. H. AND KITANO, M.: Increased brain radiocopper uptake in Wilson's disease. *Arch. Neurol.* 13: 533, 1965.

9. OLDENDORF, W. H. AND IISAKA, Y.: Interference of skull and scalp with external measurements of brain isotope content. Part 2. Absorption by skull of gamma radiation originating in brain. *J. Nucl. Med.* 10:184, 1969.

10. MATTHEWS, C. M. E. AND MALLARD, J. R.: Distribution of ⁹⁹ᵐTc and tumor/brain concentrations in rats. *J. Nucl. Med.* 6:404, 1965.

11. HARPER, P. V.: Personal communication. 1968.

12. McAFEE, J. G., FUEGER, C. F., STERN, H. S., WAGNER, H. N., JR. AND MIGITA, T.: Tc⁹⁹ᵐ pertechnetate for brain scanning. *J. Nucl. Med.* 5:811, 1964.

13. NICHOLS, G., JR., NICHOLS, N., WEIL, W. AND WALLACE, W. M.: The direct measurement of the extracellular phase of tissues. *J. Clin. Invest.* 32:1,299, 1953.

14. MELLICK, R. AND CAVANAGH, J. B.: Extracellular space of normal peripheral nerve and normal skin as measured by sulfate in the chicken. *Exper. Neurol.* 18:224, 1967.

15. DAVSON, H. AND BRADBURY, M.: The extracellular space of the brain. In *Biology of Neuroglia*. Eds. DeRobertis, E. D. P. and Carrea R. Elsevier Publishing Co., New York, 1965, p. 124.

16. OLDENDORF, W. H. AND DAVSON, H.: Brain extracellular space and the sink action of cerebrospinal fluid. *Arch. Neurol.* 17:196, 1967.

17. LAJTHA, A. AND TOTH, J.: Uptake and transport of amino acids by the brain. *J. Neurochem.* 8:216, 1961.

18. CRONE, C.: Facilitated transfer of glucose from blood into brain tissue. *J. Physiol.* 181:103, 1965.

19. EICHHORN, O.: Die Radiocirculographie. Ein Klinische Methode Zür Messung Hirndurchblutung. *Wein. Klin. Wschr.* 71:499, 1959.

20. LOVE, W. D., O'MEALLIE, L. P. AND BURCH, G. E.: Assessment of cerebral circulation by an external isotope technique. *J. Lab. Clin. Med.* 58:445, 1961.

21. OLDENDORF, W. H. AND KITANO, M.: Isotope study of brain blood turnover in vascular disease. *AMA Arch. Neurol.* 12:30, 1965.

22. RENKIN, E. M.: Transport of large molecules across capillary walls. *The Physiologist* 7:3, 1964.

23. OLDENDORF, W. H.: Measurement of the mean transit time of cerebral circulation by external detection of an intravenously injected radioisotope. *J. Nucl. Med.* 3:382, 1962.