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In Vivo Measurement of the Volume of Distribution of Water in Cerebral Grey Matter: Effects on the Calculation of Regional Cerebral Blood Flow

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Summary: The present study was undertaken to determine the apparent value for the volume of distribution of water to be used in the dynamic/integral technique for generating functional CBF images. A value of 0.86 resulted in only a minor loss of accuracy compared to the more accurate (but time-consuming) dynamic only analysis, which incorporated the regionally fitted estimates of the volume of distribution of water. In contrast to the traditionally used in vitro value of 0.95, the value of 0.86 allows for the inclusion of a significant part of the washout phase in the integral analysis, thereby producing statistically improved CBF images. **Key Words:** Volume of distribution of water—Cerebral blood flow—Oxygen-15 labelled CO_2—Grey matter—Positron emission tomography.

Recently, a new method has been described for the measurement of CBF using inhalation of $C^{15}O_2$ and positron emission tomography (PET) (Lammertsma et al., 1990). This method involves two separate steps in generating reproducible functional CBF images, which are required in the analysis of activation or stimulation studies.

First, the dynamic PET frames collected during and after inhalation of $C^{15}O_2$ are analyzed using a nonlinear regression technique to obtain accurate estimates of delay and dispersion of the arterial blood curve (Lammertsma et al., 1989). Second, using these values of delay and dispersion, a pixel-by-pixel integral analysis is then applied to the inhalation (buildup) phase.

The accuracy of functional CBF images is somewhat compromised by the need to assume, in the final integral analysis, a value for the volume of distribution of water (V_d) in brain tissue relative to that in blood. It has been shown that the value of V_d obtained from the initial dynamic analysis applied to a whole brain region of interest (ROI) is not suitable because of tissue heterogeneity effects (Iida et al., 1989; Lammertsma et al., 1990). Therefore, based on in vitro water content data, a value of 0.95 is usually taken (Herscovitch and Raichle, 1985). Since this in vitro value could be incorrect for application in in vivo studies, only the buildup phase of tracer administration is used in the integral analysis. This reduces potential errors in the calculation of CBF, which can arise when the washout phase with its dependency on V_d is included.

However, it has been shown that calculated values for white matter CBF are insensitive to the actual value of V_d used (Lammertsma et al., 1990). The accuracy of the integral calculation could thus be improved by using V_d for grey matter. This would allow the washout phase to be included in the integral analysis, resulting in CBF images with improved statistical quality.

Although values for both grey and white matter V_d have been published previously (Huang et al., 1982,1983; Depresseux, 1983; Gambhir et al., 1987; Iida et al., 1989; Lammertsma et al., 1989,1990), the reported values have not been consistent. The purpose of the present study was to obtain, through kinetic analysis, a value for grey matter V_d that

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Addreviations used: PET, positron emission tomography; ROI, region of interest.

would allow for the inclusion of a 1 min washout period into the integral analysis. The capability to apply this integral analysis to an extended data collection period (including the washout phase) is important, because it would allow optimization in terms of the signal to noise ratio. Preliminary simulation studies have indicated that a 1 min washout period might be optimal for a 2 min inhalation period.

METHODS

Data from six normal male subjects with a mean (±SD) age of 36 ± 14 years were analyzed. No subject had a significant medical history and all were found to be normal on neurological examination. For each subject, a transmission scan for subsequent attenuation correction of the emission data was collected first, followed by six repeat $C^{15}O_2$ runs. The scanner, scanning protocol, and blood monitoring system were identical to those described previously (Lammertsma et al., 1990). For the purpose of the present study, only the first and last of these runs, collected under resting baseline conditions with eyes closed and ears unplugged, were analyzed. The other four runs were performed as part of an ongoing activation/stimulation programme and will be reported elsewhere.

All subjects gave their informed consent prior to scanning. Permission to administer the radioactive tracers was obtained from the United Kingdom Administration of Radioactive Substances Advisory Committee.

Using the integral $C^{15}O_2$ image (sum of all frames collected over the two min inhalation period), for each study, one large grey matter ROI was defined by including those pixels in all 15 planes that were >70% of the global maximum (over all planes) pixel value. This (grey matter) ROI was projected onto the dynamic $C^{15}O_2$ images and the resulting time–activity curves were fitted for CBF and V_d, and delay and dispersion of the input function, as described previously (Lammertsma et al., 1990). In the routine application of the dynamic/integral method, delay and dispersion of the input function are obtained using a whole brain ROI (Lammertsma et al., 1990). To check the validity of the latter approach, the grey matter time–activity curves were refitted with delay and dispersion fixed to values obtained from the standard global (whole brain ROI) analysis. All time–activity curves were then refitted with V_d fixed to the mean value obtained from the above analysis both with and without inclusion of the washout phase.

Finally, previously reported data (Lammertsma et al., 1990) were reanalyzed with V_d fixed to the value obtained from the above analysis. The CBF values obtained in this manner for both the buildup and the combined buildup and washout periods were compared to the values obtained with fitted values of V_d and with V_d fixed to 0.95.

RESULTS

In Table 1, results of grey matter CBF and V_d are given for the total of 12 baseline runs from the six normal subjects. It can be seen that there was no significant difference in CBF and V_d values if either delay and dispersion of the input function were obtained from the fit itself or if global (whole brain ROI) values for these parameters were used. In both cases, mean V_d was 0.86, with only a small variation between studies (standard deviation = 0.04).

In Table 2, fitted CBF values are given using the mean grey matter V_d of 0.86, obtained from the studies shown in Table 1, for both a 2.5 min (buildup only) and a 3.5 min (buildup and washout) study duration. Fixing V_d to a value of 0.86 resulted in CBF values that were on average the same as

TABLE 1. *Grey matter CBF and V_d*

	Method A[a]		Method B[b]	
Study no.[c]	CBF (ml ml^{-1} min^{-1})	V_d (ml ml^{-1})	CBF (ml ml^{-1} min^{-1})	V_d (ml ml^{-1})
1079a	0.71 ± 0.01	0.83 ± 0.02	0.73 ± 0.02	0.82 ± 0.02
1079b	0.85 ± 0.03	0.83 ± 0.02	0.89 ± 0.03	0.81 ± 0.02
1083a	0.72 ± 0.01	0.89 ± 0.02	0.72 ± 0.01	0.89 ± 0.02
1083b	0.76 ± 0.02	0.87 ± 0.02	0.75 ± 0.01	0.88 ± 0.02
1112a	0.71 ± 0.01	0.89 ± 0.01	0.70 ± 0.01	0.90 ± 0.01
1112b	0.69 ± 0.01	0.88 ± 0.01	0.69 ± 0.01	0.88 ± 0.01
1119a	0.65 ± 0.01	0.91 ± 0.01	0.66 ± 0.01	0.91 ± 0.01
1119b	0.74 ± 0.01	0.79 ± 0.01	0.72 ± 0.01	0.79 ± 0.01
1131a	0.65 ± 0.01	0.85 ± 0.01	0.64 ± 0.01	0.86 ± 0.01
1131b	0.64 ± 0.01	0.87 ± 0.01	0.60 ± 0.01	0.90 ± 0.01
1207a	0.84 ± 0.03	0.81 ± 0.02	0.80 ± 0.02	0.83 ± 0.02
1207b	0.78 ± 0.02	0.86 ± 0.02	0.77 ± 0.02	0.86 ± 0.02
Mean	0.73	0.86	0.72	0.86
SD	0.07	0.04	0.08	0.04

[a] Delay and dispersion obtained from grey matter ROI.
[b] Delay and dispersion obtained from whole brain ROI.
[c] a is first and b is last (baseline) run in series of six.

TABLE 2. *Grey matter CBF for fixed V_d (=0.86)*[a]

Study no.[b]	Inhalation period only, CBF (ml ml^{-1} min^{-1})	Inhalation period + 1 min washout phase, CBF (ml ml^{-1} min^{-1})
1079a	0.73 ± 0.01	0.71 ± 0.01
1079b	0.88 ± 0.01	0.84 ± 0.02
1083a	0.75 ± 0.01	0.74 ± 0.01
1083b	0.77 ± 0.01	0.76 ± 0.01
1112a	0.72 ± 0.01	0.72 ± 0.01
1112b	0.70 ± 0.01	0.70 ± 0.01
1119a	0.68 ± 0.01	0.69 ± 0.01
1119b	0.70 ± 0.01	0.67 ± 0.01
1131a	0.64 ± 0.01	0.64 ± 0.01
1131b	0.62 ± 0.01	0.62 ± 0.01
1207a	0.80 ± 0.01	0.77 ± 0.01
1207b	0.78 ± 0.01	0.77 ± 0.01
Mean	0.73	0.72
SD	0.07	0.06

[a] Delay and dispersion obtained from whole brain ROI.
[b] a is first and b is last (baseline) run in series of six.

those obtained with individually fitted values for V_d (Table 1), both with and without inclusion of the washout phase.

In Table 3, fitted CBF values for an independent group of normal subjects are given using the same fixed V_d of 0.86. These results are compared with a previous analysis of these data (Lammertsma et al., 1990) using both a fixed V_d of 0.95 and free (fitted) V_d. Using a V_d of 0.86 resulted in CBF values that, for grey matter, were closer to those obtained with free V_d than the previous analysis with V_d fixed to 0.95 (for the inhalation period, 4 vs. 9% underestimation if V_d is fixed to 0.86 and 0.95, respectively). This difference is even more pronounced if the 1 min washout period was included in the analysis (6 vs. 13%, respectively).

TABLE 3. *Comparison of CBF values*

Method	Insular grey	Frontal grey	White
Inhalation period only			
V_d fixed[a,b]	0.76 ± 0.08	0.68 ± 0.10	0.16 ± 0.02
V_d = 0.95[a]	0.70 ± 0.06	0.62 ± 0.08	0.15 ± 0.01
V_d = 0.86	0.74 ± 0.07	0.65 ± 0.08	0.16 ± 0.01
Inhalation period + 1 min washout phase			
V_d free[a,c]	0.75 ± 0.07	0.67 ± 0.09	0.16 ± 0.02
V_d = 0.95[a]	0.66 ± 0.07	0.58 ± 0.06	0.16 ± 0.01
V_d = 0.86	0.71 ± 0.07	0.62 ± 0.07	0.16 ± 0.01

Mean CBF (±SD) in ml ml^{-1} min^{-1} for 14 runs.
[a] Data from Lammertsma et al. (1990).
[b] V_d individually fixed to the value obtained from the corresponding fit of inhalation + washout phase with free V_d.
[c] Fitted V_d values were 0.82 ± 0.10 and 0.80 ± 0.08 ml ml^{-1} for insular and frontal grey matter, respectively. For white matter, no reliable fitted estimates of V_d could be obtained. See Lammertsma et al. (1990) for further discussion.

DISCUSSION

It is well recognized that measurements of CBF with $H_2{}^{15}O$, either administered by intravenous injection or via inhalation of $C^{15}O_2$, are most accurate if a dynamic scanning sequence is utilized (Lammertsma and Mazoyer, 1990). This allows for corrections to be made for delay and dispersion of the arterial whole blood input function (Lammertsma et al., 1989). In addition, it avoids the assumption of a constant, possibly incorrect, value for the volume of distribution of water V_d. However, at present, noise considerations and computational time effectively limit this approach to selected ROIs. For many applications (e.g., activation studies), the availability of a functional CBF image is important.

Recently, a combined dynamic/integral approach has been described (Lammertsma et al., 1990). In this method, corrections for delay and dispersion of the input functions were obtained from the dynamic analysis and subsequently used in an integral analysis resulting in a functional CBF image. However, in the final integral pixel-by-pixel calculation, a value for V_d had to be assumed, since the value obtained from the (global) dynamic analysis was strongly dependent on tissue heterogeneity effects (Iida et al., 1989; Lammertsma et al., 1990). This dependency might also, at least in part, explain the variability in reported V_d values (Huang et al., 1982,1983; Depresseux, 1983; Gambhir et al., 1987; Iida et al., 1989; Lammertsma et al., 1989,1990). As a result of the sensitivity to tissue heterogeneity, these reported values will, even if corrections for delay and dispersion of the input function are made, depend both on ROI size and spatial resolution of the PET scanner.

To reduce the vulnerability of the CBF calculations to the assumed value of V_d, the integral calculation in the above-mentioned dynamic/integral method was only applied to the buildup phase during a slow administration of the tracer. A drawback of this procedure is the somewhat inefficient use of administered radioactivity due to the omission of the washout phase in the calculation of the functional CBF image.

The noise level in the functional CBF image could simply be reduced by including the washout phase in the integral analysis. However, in order not to compromise accuracy, this would require a realistic value for V_d.

It has been demonstrated that, due to its longer transit time, the calculation of white matter V_d is insensitive to the actual assumed value of V_d (Lammertsma et al., 1990). In addition, for activation

studies, especially grey matter structures are of primary interest. Therefore, potentially, the dynamic/integral method could be improved by fixing V_d in the integral analysis to a previously measured functional value for grey matter rather than a calculated value based on in vitro water content data.

In the present study, a representative global grey matter ROI was defined by thresholding the integral buildup image, thus incorporating all pixels that were within 30% of the study maximum. This resulted in time–activity curves with good statistical properties.

To exclude errors due to possible variations in arrival times within the brain, these grey matter time–activity curves were fitted in two different ways. First, delay and dispersion of the input function were included as parameters of the fit. Second, they were fixed to the values obtained from the usual whole brain analysis, as described previously (Lammertsma et al., 1990). It is reassuring to note that both fitting procedures resulted in identical CBF and V_d values (Table 1), providing further evidence that global analysis of delay and dispersion is a valid approach.

It is of interest to note that the variability of V_d was less than that of CBF. From Table 1, it can be seen that the coefficient of variation (COV) for CBF was 11%, while that for V_d was <5%. This finding suggests that the intersubject variation of water partitioning in grey matter (V_d) is small. This was confirmed by refitting the data with V_d fixed to the mean value of 0.86, giving CBF values (Table 2) that were not significantly different from those obtained with free V_d, even if the washout phase was included in the analysis with fixed V_d.

Previously, it had been shown (Lammertsma et al., 1990) that fixing V_d to a value of 0.95 resulted in significantly lower CBF values than those obtained with free V_d. In addition, in this case, the CBF results were dependent on the actual data interval used for analysis. Analysis of the same data with a fixed V_d of 0.86 resulted in a significant reduction of both the underestimation in CBF and the time dependency (Table 3). It should be realized that, in the latter analysis, ROIs were defined according to anatomical structure and not by thresholding. Therefore, it is likely that the degree of tissue heterogeneity for these ROIs (Table 3) is quite different from those used in Tables 1 and 2. From the analysis of several grey matter structures, Iida et al. (1989) concluded that tissue heterogeneity was probably most pronounced in the frontal cortex. From Table 3, it follows that even for this region the underestimation in CBF is small (4% for the inhalation period only, 7% when the washout phase is included).

Although fixing V_d to a value of 0.86 results in CBF values that are slightly less accurate than those obtained with free V_d, they are superior over those obtained with V_d fixed to 0.95 (Table 3). For activation and stimulation studies where a functional CBF image is required, fixing V_d to 0.86 seems to be a reasonable compromise. It allows for inclusion of the washout phase in the integral analysis, hence in improving the statistical quality of the functional CBF images with minimal loss of accuracy due to tissue heterogeneity effects.

CONCLUSIONS

The dynamic/integral technique provides a means for generating reproducible functional CBF images with inherent corrections for the delay and the dispersion of the arterial whole blood input function. However, the final integral analysis requires an assumption on the volume of distribution of water. To minimize the effects of errors in this assumption, only the buildup phase was used in its initial implementation.

To include the washout phase in the integral analysis, it is necessary to fix the volume of distribution to such a value that calculated CBF is least dependent on study duration. In the present study, it was found that a value of 0.86 provides a reasonable compromise between accuracy and precision. This value allows the efficient calculation of functional CBF images with only a small reduction in accuracy. At the same time, the study duration can be increased, which results in better statistical quality.

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