Quantitation of Neuroreceptors: A Need to Increase Imaging Resolution?

TO THE EDITOR: van Dyck et al. (*1*) recently reported estimations of the specific-to-nondisplaceable partition coefficient V_3'' of [^{123}I]β-CIT calculated as (striatal–occipital)/occipital uptake at tracer equilibrium (18–24 hr after injection of tracer). The measurements were performed with the multislice brain dedicated GERASPECT device with a spatial resolution of 7–8 mm FWHM. The images shown, however, in their article (Figs. 1 and 5) correspond roughly to image resolution of 12–14 mm (not 7–8 mm). Imaging resolution depends on many things such as collimator used, reconstruction filtering, scatter, etc. and is not the same as the spatial FWHM resolution of the scanner. Numerical values of V_3'' are fully dependent on imaging resolution as well as on reconstruction errors of the low count density reference region (occipital) and on the regions of interest drawn. Numerical values of V_3'' from 4 to 12 are reported even in age-matched healthy control subjects. Similar numerical values of the other parameters of [^{123}I]β-CIT vary 100%–300% (*2,3*). What does this mean? The values between laboratories are not comparable.

The answer to this problem is better SPECT. With essentially all [^{123}I]β-CIT is in the striatum at 18–24 hr after injection of tracer, one might be able to increase imaging resolution by 5–6 mm. A choice of the proper reconstruction filter is important. Figure 1 shows a 2.8-mm-thick transaxial slice of a 37-yr-old healthy male imaged with the Siemens MultiSPECT 3 gamma camera with fan-beam collimators by using a Butterworth filter (order = 8) with two cutoff frequencies. The dose used was 185 MBq (5 mCi). The quality difference of these two images is impressive. In addition, the numerical value of V_3'' with the softer filter is 30% less than that of the harder one. The tracer [^{123}I]β-CIT is satisfactory and quantitation is easy, but overall results depend on the excellence of the SPECT system used. This is a sensitive and specific tracer to image patients with Parkinson's disease. It demonstrates the loss of presynaptic nerve endings in the striatum in relation to the severity of parkinsonian disability and is helpful in the early diagnosis and follow-up of Parkinson's disease. Hopefully, we will not distort its use with faulty quantitation.

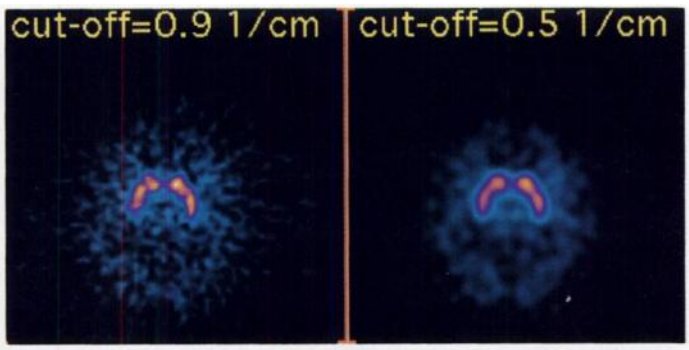

FIGURE 1. Transaxial slice of striatal [^{123}I]β-CIT uptake in a 37-yr-old healthy man using two cutoff SPECT filtering frequencies. The filter used was a Butterworth with an order of 8. There is a 30% difference in semiquantitation of (striatum-occipital)/occipital ratio between these two images.

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REPLY: Dr. Kuikka raises timely questions for an era of increasing multicenter imaging studies (*1*). He is certainly correct that estimations of the specific-to-nondisplaceable partition coefficient V_3'' with [^{123}I]β-CIT SPECT depend on image resolution which, in turn, depends on choice of collimator, reconstruction filter, etc. He also notes that values of V_3'' and other outcome measures with [^{123}I]β-CIT vary substantially between laboratories. We should caution, however, that the studies he cites use somewhat different outcome ratios [i.e., basal ganglia/white matter (*2*) or basal ganglia/cerebellum (*3*) than that used by our group [(striatum-occipital)/occipital (*4*)]. We should further point out that, even using identical parameters, some biological variability is to be expected. Within our program, differences of twofold or greater are observed in V_3'' between healthy subjects of the same age (*4*), which are consistent with in vitro dopamine transporter binding studies (*5–7*).

To be sure, some of the variability between laboratories in outcome measures with [^{123}I]β-CIT accrues from the factors detailed by Dr. Kuikka. His example illustrates this point well: V_3'' is altered 30% by a change in Butterworth filter cutoff frequency. However, we do not believe that interlaboratory comparability will be achieved solely by increasing (or otherwise standardizing) image resolution. Enhanced resolution may come at the cost of decreased sensitivity (in the case of increased collimator resolution) or increased noise (in the case of "harder" filtering). Moreover, increased resolution alone will not address the many other issues necessary to achieve interlaboratory comparability, including camera sensitivity, attenuation and scatter corrections and regions of interest. As formal multicenter trials are organized with [^{123}I]β-CIT and other neuroligands, they will need to employ either identical imaging equipment and reconstruction algorithms or, more realistically, phantom-derived conversion factors.

We should note, finally, that the problems raised by Dr. Kuikka need not compromise the validity of results *within* a given laboratory, provided that they affect the outcome measure in a linear manner. For our studies with [^{123}I]β-CIT, we have performed phantom studies to verify linearity between known activity (across a physiologic range, including activity levels representative of the low-count occipital region) and reconstructed counts. These studies have used the same camera, collimation and filtering as utilized in our human studies. Therefore, when we report a decline in V_3'' by 51% from age 18 to 83 (*4*), we expect that this result could be reproduced by another laboratory using different imaging equipment and reconstruction algorithms.

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TABLE 1
Radiochemical Purity of Fractionated MAG3 and MIBI Kits Relative to Storage Time

Storage time (days)		Kit			
		MAG3 1	MAG3 2	MIBI 1	MIBI 2
0	RP (%) ± s.d.	98.1 ± 0.79	97.2 ± 0.79	98.1 ± 0.71	97.9 ± 0.54
Immediate use	n	17	10	8	4
	n below RP limit	0	0	0	0
1–6	RP (%) ± s.d.	98.1 ± 1.43	63.6 ± 47.8	97.9 ± 0.71	94.9 ± 10.1
	n	10	11	5	6
	n below RP limit	0	3	0	1
Longer than 6	RP (%) ± s.d.	98.6 ± 1.39	23.9 ± 40.0	96.9 ± 2.51	96.1 ± 6.0
	n	11	10	4	5
	n below RP limit	0	8	0	1

Numbers 1 and 2 refer to generator eluate. n = number of preparations.

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Problems with Fractionated Cold Kits

TO THE EDITOR: Cost-effective preparation of ^{99m}Tc radiopharmaceuticals by fractionated use of expensive cold kits has been explored extensively in recent years and has been discussed at a Society of Nuclear Medicine annual meeting (*1–3*). Methods have been published for HMPAO (*4,5,6*), MIBI (*2,7*), MAG3 (*7,8*) and ECD (*3*). In general, the kits are reconstituted with saline, divided into several fractions, transferred to sterile vials and stored at −10 to −70°C. In our department, we have used this approach for MAG3 and MIBI. By changing our generator system from TECEGEN S™ (CIS/Behring, Marburg, Germany; reference activity 8 and 20 GBq, generator 1) to ULTRATECHNEKOW™ (Mallinckrodt, Petten, The Netherlands; reference activity 12.9 and 10.6 GBq, generator 2), we found an unexpected high rate of an unacceptable low radiochemical purity (RP) of longer-stored kits, which had never been observed before.

MAG3 kits were reconstituted in 10 ml nonbacteriostatic, low-dissolved-oxygen (LDO) saline (i.e., nitrogen purged for 15 min) and split into four fractions, 2.5 ml each. The fractions were stored in a freezer at −10°C for up to 30 days. For labeling, the kits were thawed at room temperature and, after the addition of 740-1110 MBq ^{99m}Tc-pertechnetate (generator in growth less than 24 hr, eluate not older than 2 hr) in 1.5 ml (final volume 4 ml), boiled for 10 min. Determination of RP was performed using the SEPPAK method recommended by the producer of the United States kit; the RP limit for the European kit is 96%. MIBI kits were dissolved in 3 ml LDO saline and split into three fractions, 1 ml each, and stored up to 7 days. After thawing the kit at room temperature, labeling was performed by adding 3–4 GBq [^{99m}Tc]pertechnetate (generator in growth less than 24 hr, eluate not older than 2 hr) in 1.0 ml (final volume 2 ml) and boiling for 10 min. Radiochemical purity was determined using the recommended TLC method (Baker flex Aluminia foils/Ethanol), with a RP limit of 90%.

Using the eluate from generator 1, an excellent RP of 98.3% ± 1.02% (mean ± SD; n = 38) for ^{99m}Tc-MAG3 and 97.8% ± 1.3% (mean ± s.d.; n = 17) for ^{99m}Tc-MIBI was found in all preparations independent of the storage time. For up to 30 days of storage no RP decrease was found. Labeling performed with eluate from generator 2, however, resulted in an unacceptable low RP for MAG3 kits stored more than 1 wk. Of 10 MAG3 kits, only two had a RP greater than 96% (mean = 23.9%). Also, at shorter storage times, 3 of 11 preparations failed to yield the required RP limits (mean = 63.6%). The use of fractionated MIBI kits labeled with eluate from generator 2 resulted in a higher stability, but two preparations stored for longer times failed to give the required RP. The results are summarized in Table 1.

Many arguments could be found to explain a reduced stability of kits for ^{99m}Tc labeling using a different generator eluate. It may be due, for instance, to a higher amount of dissolved oxygen in the eluate oxidizing the tin(II) in the kit. Differences in the generator technology of the two described generators may also play a role. Another reason for the observed low RP levels in the fractionated kits is that storage can significantly reduce the amount of tin(II). This also explains why fractionated MAG3 kits caused more problems than fractionated MIBI kits, as the theoretical amount of tin(II) in the former (10 μg as SnC12.2H2O) is much lower than that in the latter (25 μg as SnC12.2H2O). Although the problem can be eliminated by adding additional amounts of tin(II), as has been described for HMPAO (*4*), MIBI (*2*) and ECD (*3*), or by changing the storage conditions (*7*), we want to stress that kit fractionation must only be performed under a strict quality control program, which should include RP determination before application and sterility testing. Small changes in the original tested protocol such as changing the generator, using different vials for storage or changing volumes or activity can lead to unexpected results and to an unsafe product. Also, keeping in mind legal considerations, it should be emphasized that cold-kit fractionation must remain in the hands of individuals with proven radiopharmacy experience.

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