Age-Related Decline in Striatal Dopamine Transporter Binding with Iodine-123-β-CIT SPECT

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The effect of age on human striatal dopamine (DA) transporters was investigated with SPECT using the ligand [^{123}I]2β-carbomethoxy-3β-(4-iodophenyl)tropane ([^{123}I]β-CIT). **Methods:** Iodine-123-β-CIT binding in the striatum was examined in 28 healthy human subjects (14 men, 14 women) who ranged in age from 18 to 83 yr. Following injection with [^{123}I]β-CIT (mean ± s.d. = 9.9 ± 1.2 mCi), subjects were scanned with the brain-dedicated CERASPECT camera. A reconstructed transaxial slice 13.3-mm thick at the level of maximal striatal activity was used to determine tracer uptake in striatal and occipital regions of interest. The stability of regional uptake on Day 2 (approximately 18–24 hr postinjection) permitted estimation of the specific-to-nondisplaceable equilibrium partition coefficient: V_3'', calculated as (striatal − occipital)/occipital uptake at equilibrium. **Results:** Values of V_3'' ranged from 3.6 to 11.4 for this sample (6.7 ± 1.9). V_3'' showed a significant inverse correlation with age ($r = -0.73$, $n = 28$, $p < 0.0001$). Linear regression analysis revealed that V_3'' declined by 51% over the age range studied or approximately 8% per decade. **Conclusion:** These findings confirm postmortem reports of dopamine transporter loss with aging. In vivo methodologies may permit the age-related degeneration of dopamine nerve terminals to be studied in relation to the cognitive and motor deficits that occur in normal aging.

Key Words: iodine-123-β-CIT; dopamine transporter; single-photon emission computed tomography

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The frequency of extrapyramidal symptoms in the elderly (*1–4*) suggests that the nigrostriatal dopaminergic system degenerates with advancing age. In recent years, both presynaptic and postsynaptic elements of this neural system have been probed for age-related changes. Most studies of receptor binding with aging have been conducted with receptors whose location is primarily postsynaptic. In vitro studies of the effect of aging on the two dopamine receptor subtypes in striatum have been somewhat contradictory. D2 receptor levels have most often been reported to decrease with age (*5–11*). D1 receptor levels have also generally been found to decline (*5,7,10*), although some studies have found no change (*6,12*), and one has found an increase with age (*8*). In vitro studies of presynaptic elements have also shown deterioration with age. The number of neurons in the substantia nigra (*13*) and striatal dopamine content (*14,15*) demonstrate an age-dependent reduction. The concentration of striatal dopamine transporters shows a similar decline (*12,16,17*). Dopamine transporter mRNA content in substantia nigra decreases even more precipitously, with an abrupt decline after age 57 (*18*).

With the advent of functional neuroimaging, it has become possible to study the aging of neural systems in living subjects. The nigrostriatal dopamine system has received considerable attention in this regard. PET studies have corroborated a number of age-related changes in postsynaptic elements, including a decline in D1 (*19*) and D2 (*20,21*) receptor density.

The function of the presynaptic part of this system has also been visualized with PET ligands such as 6-[^{18}F]fluoro-L-3,4-dihydroxyphenylalanine ([^{18}F]FDOPA), which provides a measure of activity of the synthetic enzyme aromatic amino acid decarboxylase (*22,23*). Fluorine-18-FDOPA PET studies of aging, however, have been contradictory because both reduced (*24*) and unchanged (*25,26*) striatal [^{18}F]FDOPA uptake with age have been reported. Quantitation of imaging results with [^{18}F]FDOPA remains problematic, in part because of the presence of a peripheral radiolabeled metabolite in the striatum and the relatively low target-to-background activity in the brain (<2:1) (*27*). Despite these relative limitations, PET studies with [^{18}F]FDOPA have provided valuable insights into the pathophysiology of parkinsonian syndromes (*23*).

An alternative approach to measurement of enzymatic activity is the quantitation of presynaptic receptor sites located on the terminals of dopamine neuronal projections from substantia nigra to striatum. The dopamine transporter is an example of such a marker and functions to remove dopamine from the synapse back into the terminal for storage or metabolism. Several PET and SPECT probes of the dopamine transporter have been developed. Cocaine analogs have been particularly promising because

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of their high affinity and low nonspecific binding (*28–30*). Few data exist on the change of dopamine transporters with age in the living human brain. Tedroff et al. (*31*) studied seven healthy volunteers aged 24–81 yr with [^{11}C]nomifensine and PET and found a marked age-related decline in striatal to cerebellar radioactivity ratios.

The potent cocaine analog 2β-carbomethoxy-3β-(4-iodophenyl)tropane (β-CIT; also referred to as RTI-55) has high affinity for the dopamine (*32,33*) and serotonin (5-HT) (*34*) transporters, low nonspecific binding and slow brain kinetics. Iodine-123-β-CIT has been shown to be a promising SPECT radiotracer for visualization of dopamine and 5-HT transporters (*35,36*). In humans and nonhuman primates, the tracer concentrates in striatal and midbrain regions (*37–39*). Pharmacological characterization of the regional uptake in primates has demonstrated that the striatal activity is associated with dopamine transporters, while the midbrain activity is associated with 5-HT transporters (*37*). Iodine-123-β-CIT has shown promise as a ligand to diagnose and evaluate idiopathic Parkinson's disease (*38*). An understanding of the effects of normal aging on [^{123}I]β-CIT binding, however, is essential to the full interpretation of these results.

Our initial investigation of [^{123}I]β-CIT in healthy subjects aimed to derive an adequate measure of dopamine transporter density that would not be affected by regional cerebral blood flow or peripheral tracer clearance. In a study of five healthy subjects (*40*), we compared graphical, kinetic and equilibrium methods to analyze SPECT [^{123}I]β-CIT binding to dopamine transporters in vivo. We arrived at an equilibrium analysis on Day 2 (from approximately 18 to 24 hr postinjection) as the method of choice for clinical studies since it does not require multiple scans or the measurement of arterial plasma tracer concentrations.

This study used the equilibrium model to examine the effects of age on dopamine transporter binding with [^{123}I]β-CIT. In the course of that analysis, we attempted to revalidate, in a larger healthy sample, the assumption of stable regional uptake required for the equilibrium model. We further investigated any possible age-related bias in the stability of regional uptake and in the estimation of our outcome measure, V_3''.

MATERIALS AND METHODS

Subjects

The study population consisted of 28 healthy volunteers (14 men, 14 women) ranging in age from 18 to 83 (48 ± 20) yr. All subjects, except one, were right-handed. Subjects underwent a clinical examination by a research psychiatrist in order to exclude any neurological or psychiatric disease, alcohol or substance abuse. Screening procedures included a physical and neurological examination, EKG, serum chemistries, thyroid function studies, CBC, urinalysis and urine toxicology screen. Female subjects with childbearing potential were required to have a negative pregnancy test (serum at screening; urine immediately prior to tracer injection). Those subjects older than 65 yr of age (n = 7) were also assessed with the Folstein Mini-Mental State Examination (MMSE) (*43*) and brain MRI. MMSE scores for these subjects ranged from 28 to 30 (29.6 ± 0.8), indicating no significant cognitive impairment. All brain MR scans were read by a neuroradiologist and were considered normal with respect to subject age. Five subjects were taking prescribed medication. Two subjects, a 73-yr-old man and a 77-yr-old woman, had mild hypertension (medications: hydrochlorothiazide and enalapril, respectively). One subject, a 63-yr-old woman, had degenerative arthritis (medication: diclofenac). None of these drugs is known to affect the brain's dopaminergic system. Two female subjects (aged 23 and 27) were taking oral contraceptives (norethindrone/ethinyl estradiol).

All subjects gave written informed consent to the research protocol as approved by the institutional human investigation committee. Subjects received 0.6 g of potassium iodide (SSKI solution) 24 hr before imaging.

Radiopharmaceutical Preparation and Purity

Iodine-123-β-CIT was prepared from the corresponding trimethylstannyl precursor and high-radionuclidic purity [^{123}I]NaI (Nordion International, Ltd., Vancouver, Canada) as previously described (*41*). Radiochemical purity was determined by high-performance liquid chromatography (HPLC) and specific activity was measured by comparing UV absorbance of the labeled product with a standard curve made from authentic β-CIT. Iodine-123-β-CIT was obtained in an average radiochemical yield of 77.6% ± 10.4% (n = 28, with this and subsequent values expressed as mean ± s.d.), radiochemical purity of 97.4% ± 1.5%, and specific activity >5,000 Ci/mmole. Sterility was confirmed by lack of growth in two media: trypticase and fluid thioglycollate at 35°C for 2 wk (*42*). Apyrogenicity was confirmed by the LAL test (Endosafe, Charleston, NC).

Data Acquisition

SPECT data were acquired with the multislice brain dedicated CERASPECT device [Digital Scintigraphics, Waltham, MA; (*44*)] with a resolution of 7–8 mm FWHM in all three axes. Four fiducial markers filled with 10 μCi of $Na^{99m}TcO_4$ were attached to the skin along the canthomeatal plane for identification during image analysis.

Iodine-123-β-CIT (9.9 ± 1.2 mCi) was injected as a single bolus over 30 sec at approximately 3:00 PM (Day 1). Scans were acquired on the day after injection (Day 2) according to the following protocol: three scans of 12 to 20 min each at approximately 18, 21 and 24 hr postinjection. Four subjects instead were scanned hourly from 18 to 24 hr postinjection, and one subject had two scans at 18 and 24 hr postinjection. These different acquisition protocols were evenly sampled during the 18–24-hr interval. The temporal midpoint (T_{mid}) of all SPECT acquisitions for each subject ranged from 1213 to 1357 min postinjection (1281 ± 28) and showed no age-related bias ($r = -0.032$; $p = 0.87$).

Image Analysis

Images were reconstructed from photopeak counts (159 ± 16 keV) using a Butterworth filter (cutoff = 1 cm, power factor = 10) and displayed as a 64 × 64 × 32 matrix (pixel size = 3.3 mm × 3.3 mm, slice thickness = 3.3 mm, voxel volume = 36.7 mm^3). Subsequent image analysis was performed by an operator who was unaware of subject demographics. SPECT data were reoriented to correct for deviations from the canthomeatal plane as identified by the four fiducial markers. The four slices corresponding to the highest striatal uptake were digitally summed, yielding a final slice of 13.3 mm thick. Attenuation correction was performed using a

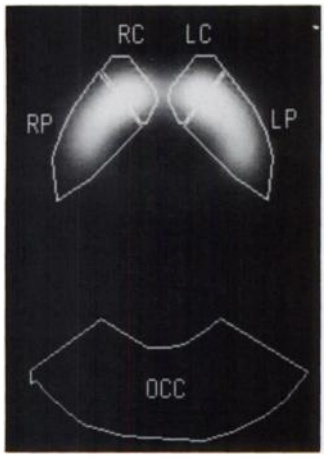

FIGURE 1. Regions of interest positioned on the [^{123}I]β-CIT SPECT image (displayed in gray scale) at the level of highest striatal uptake. RC = right caudate, LC = left caudate, RP = right putamen, LP = left putamen, OCC = occipital cortex. Left and right caudate and putamen were combined into a single striatal ROI for data analysis.

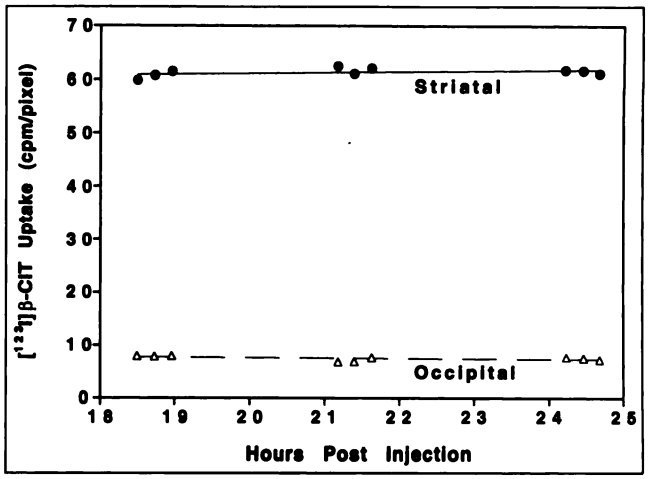

FIGURE 2. Brain regional uptake (cpm/pixel) from approximately 18–24 hr postinjection of 10 mCi [^{123}I]β-CIT in a 32-yr-old healthy woman. Linear regression analysis revealed that striatal uptake (closed circles) was stable and increased at 0.23%/hr. Occipital uptake (open triangles) was stable and declined at −0.84%/hr. These rates of change are close to the mean values for the sample.

Chang zero order method (attenuation coefficient $\mu = 0.15\ cm^{-1}$) within an ellipse drawn around the skull as identified by the markers. Five standard regions of interest (ROIs) of preset area and shape (Fig. 1) were visually positioned on this summed slice: left and right caudate (254 mm^2 each), left and right putamen (682 mm^2 each) and occipital cortex (2543 mm^2).

Count densities for all five ROIs were then decay-corrected for the time of injection and divided by the scan duration to generate ROI activity in counts per minute per pixel. Average striatal uptake was then computed by dividing total counts per minute by total pixels for left and right caudate and putamen. No attempt was made to correct for partial volume effects.

Equilibrium Model

As noted above, the equilibrium analysis (*40*) is based on the stability of regional tracer uptake on Day 2 (from about 18 to 24 hr postinjection). That analysis presumes a three-compartment model comprising the arterial plasma compartment (C_a), the nondisplaceable compartment (C_2) and the specific compartment (C_3). The equilibrium distribution volume of a compartment i (V_i) is defined as the equilibrium ratio of the tracer concentration in that comparment to the free arterial concentration: $V_i = C_i/(f_1C_a)$, where f_1 = free fraction of parent tracer in plasma. V_3 is equal to the binding potential (BP) which is the ratio of receptor density (B_{max}) to the dissociation constant (K_D) (*45*). We have defined V_3'' (*46*) (also termed the specific-to-nondisplaceable equilibrium partition coefficient) as $V_3/V_2 = BP/V_2 = B_{max}/(K_D*V_2)$. V_3'' is thus proportional to B_{max}, assuming that both K_D and V_2 are relatively invariant in the population.

Under equilibrium conditions, the outcome measure V_3'' can be estimated by C_3/C_2. By assuming that the occipital region is devoid of dopamine transporters (*12,47*), this parameter can be computed without conversion of SPECT counts per minute to absolute units of radioactivity and without measurement of arterial plasma tracer concentrations as $[(cpm/pixel)_{striatal} - (cpm/pixel)_{occipital}]/(cpm/pixel)_{occipital}$.

Statistical Analysis

The Day 2 equilibrium analysis relies on several assumptions (*40*), most importantly, the stability over time of striatal and occipital uptake on Day 2. The present study therefore included a revalidation of that assumption in a larger healthy subject sample. In these subjects for each SPECT acquisition, the striatal, occipital (nondisplaceable) and specific (striatal–occipital) uptake was computed in decay-corrected cpm/pixel. To examine the stability of these three parameters and V_3'' on Day 2, a linear regression slope was computed for each parameter over 18–24 hr in all 28 subjects. To detect any systematic age-related bias, the relationship between the regression slope and age was examined for each parameter by Pearson's product moment correlation coefficient (r).

The equilibrium model was then applied to the present healthy subject sample to study the effects of age on [^{123}I]β-CIT binding to dopamine transporters (V_3''). An average value of V_3'' was calculated for all SPECT acquisitions throughout the 18–24 hr postinjection interval for each subject. The relationship between V_3'' and subject age was examined by linear regression analysis and Pearson's product moment correlation coefficient (r).

To test whether the addition of gender (with or without the interaction of age and gender) significantly improved the prediction of V_3'' after controlling for the contribution of age, we also used multiple linear regression analysis and partial F tests (*48*) with the models:

$$V_3'' = \beta_0 + \beta_1 age + \beta_2 gender + E,$$

$$V_3'' = \beta_0 + \beta_1 age + \beta_2 gender + \beta_3 age * gender + E.$$

RESULTS

Stability of Regional Tracer Uptake

In the present sample of 28 healthy subjects, as in our previous report with [^{123}I]β-CIT (*40*), regional tracer uptake showed remarkable stability on Day 2 (Figs. 2 and 3). Striatal uptake changed by an average of 0.29% ± 0.79%/hr and occipital uptake by −1.19% ± 1.68%/hr. Specific uptake changed by 0.54% ± 0.90%/hr and V_3'' by 1.72% ± 1.96%/hr. The occipital washout rate of 1.19% ± 1.68%/hr fell clearly within the limits we previously outlined (<5%/hr) (*40*) for accurately estimating V_3'' using the ratio of specific-to-nondisplaceable uptake.

The relationship between the regression slopes for regional tracer uptake and the outcome measure, V_3'', versus subject age is displayed in Figure 3. The rates of change

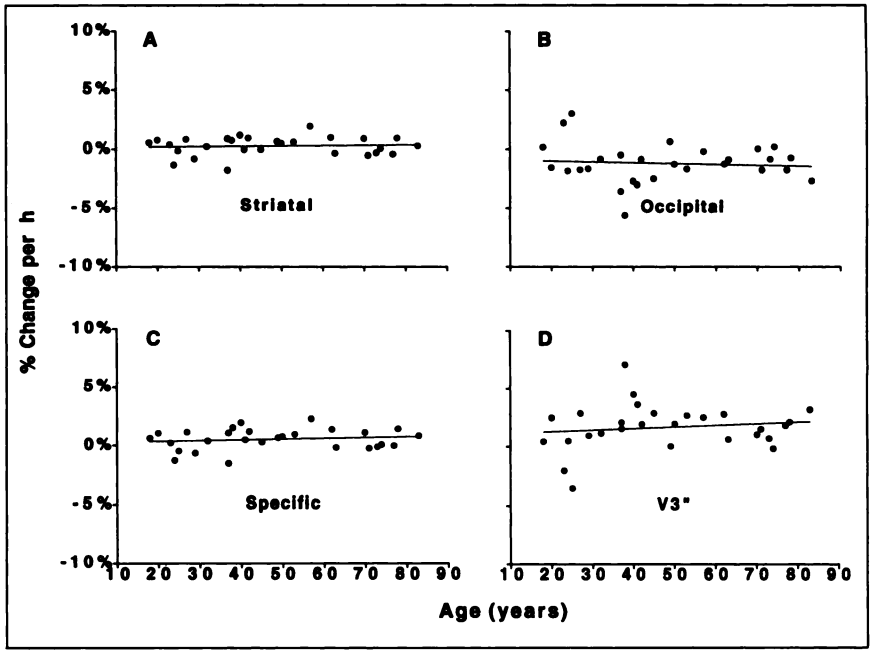

FIGURE 3. Rate of change (%/hr) in striatal (A), occipital (B) and specific (C) uptake of [^{123}I]β-CIT and in V_3'' (D) expressed as a function of subject age. Each dot denotes an individual healthy subject (n = 28). Values are obtained by linear regression and expressed as the percentage of mean regional uptake (cpm/pixel) between 18 and 24 hr postinjection. The rate of change in each parameter is unrelated to subject age (striatal: r = 0.08, n = 28, p = 0.69; occipital: r = −0.09, n = 28, p = 0.64; specific: r = 0.15, n = 28, p = 0.44; V_3'': r = 0.15, n = 28, p = 0.44).

(%/hr) in striatal uptake, occipital uptake, specific uptake and V_3'' were all unrelated to age.

Effect of Age on Iodine-123-β-CIT Binding

The values of V_3'' (averaged for all SPECT acquisitions from 18 to 24 hr postinjection) ranged from 3.6 to 11.4 (6.7 ± 1.9) for this healthy subject sample. There was an age-dependent decline in V_3'' values (r = −0.73, n = 28, p < 0.0001) (Fig. 4). Linear regression analysis revealed that V_3'' declined by 51% over the age range of 18–83 yr or approximately 8% per decade. Figure 5 displays typical SPECT images in young and old healthy subjects.

The addition of gender (with or without the interaction of age and gender) did not significantly improve the prediction of V_3'' after controlling for the contribution of age: $F_{1,24}$(gender|age) = 0.05, NS; $F_{2,24}$(gender, age * gender|age) = 0.34, ns (notation from Kleinbaum et al. (*48*)).

DISCUSSION

These data demonstrate that dopamine transporter binding as measured by [^{123}I]β-CIT and SPECT is inversely correlated with age. The outcome measure V_3'' declined by 51% over the age range studied, or approximately 8% per decade.

Our results provide in vivo corroboration for previous postmortem reports. Three studies have examined the effects of age on dopamine transporter density in putamen (*12,17*) or caudate nuclei (*16*) using in vitro homogenate binding with the dopamine transporter radioligand [^{3}H]GBR-12935. All three reports found highly significant inverse correlations between dopamine transporter density

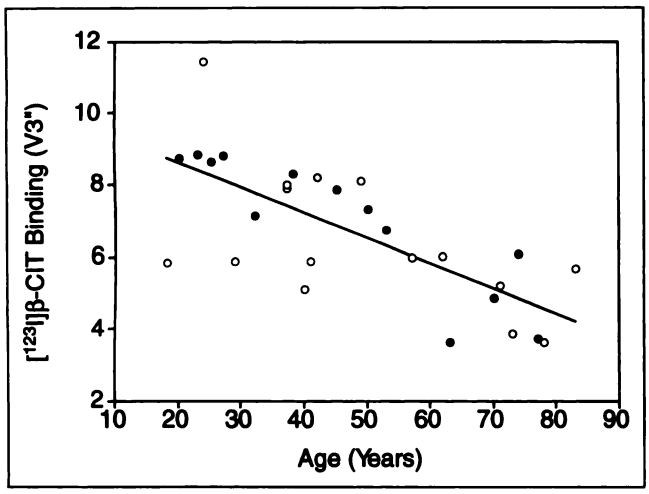

FIGURE 4. Striatal [^{123}I]β-CIT binding (V_3'') versus age in 28 healthy volunteers. V_3'' = −0.070 × age + 10.031; r = −0.73; p < 0.0001 (Pearson's test). Each dot denotes an individual subject; closed circles represent females and open circles represent males.

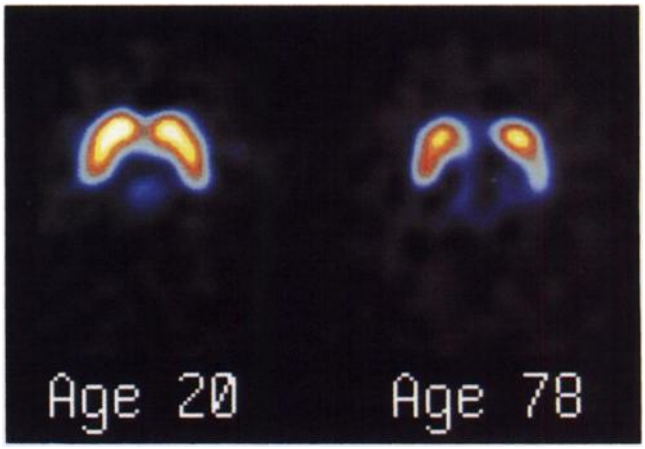

FIGURE 5. Images of striatal [^{123}I]β-CIT uptake in two subjects aged 20 and 78 yr, respectively. Data were acquired 24 hr following injection of approximately 10 mCi of isotope. Both images are normalized to the minimum and maximum counts per pixel values for the younger subject. The older subject shows a 56% reduction in dopamine transporter binding (V_3'') relative to the younger one.

(B_{max}) and age. De Keyser et al. (*12*) examined postmortem putamen from 32 subjects aged 19–88 yr and found an approximately 65% decline over that age range. Allard et al. (*17*) also examined putamen from 20 subjects aged 19–100 yr and reported a 70% decline over the adult age range of 20–80 yr. Zelnik et al. (*16*) examined the caudate nuclei of 19 subjects aged 0–82 yr and found roughly a 75% decline over the age studied. When extrapolated to the age range of our subject sample, these studies reflect approximately a 60%–70% decline or about 10% per decade.

Assumptions Underlying V_3''

Whether the specific-to-nondisplaceable uptake ratio (C_3/C_2) 18–24 hr postinjection provides an adequate estimation of V_3'' rests on several assumptions (*40*). These include that a state of equilibrium indeed exists on Day 2, as reflected by stable tracer uptake in striatal and occipital ROIs. The present results suggest even greater regional stability than we originally reported in five healthy subjects (*40*). In those subjects (4 of whom are included in the present sample of 28), striatal and occipital uptake changed by an average of 0.5%/hr and −1.5%/hr, respectively, as compared to 0.3%/hr and −1.2%/hr in the present study.

In particular, the slow occipital washout rate supports the validity of C_3/C_2 on Day 2 as an estimate of V_3''. We previously reported (*40*) that simulations performed using a terminal half-life of 50 hr (corresponding to an occipital washout rate of 1%/hr) showed that the C_3/C_2 ratio measured between 1200 and 1440 min was within 6% of the true V_3''. The error was less than 20% for half-lives longer than 10 hr (5%/hr occipital washout) and increased to unacceptable levels for half-lives shorter than 10 hr. In the present sample, only 1 of 28 subjects had an occipital washout rate >5%/hr (5.6%/hr).

Equally critical for the present study was the absence of an age-related bias in the estimation of V_3'', i.e., in the presence or absence of equilibrium. In this subject sample, the linear regression slopes versus time of striatal uptake, occipital uptake, specific uptake and V_3'' were all unrelated to age. Thus, it is unlikely that the time interval chosen (18–24 hr postinjection) represented a different phase of the time-activity curve (i.e., preequilibrium versus postequilibrium) for young versus old subjects.

The use of V_3'' as an outcome measure (proportional to B_{max}) rests on the assumption that K_D and V_2 are relatively invariant in the population and, specifically, do not vary as a function of age. The aforementioned in vitro homogenate binding studies with [^{3}H]GBR-12935 (*12,16,17*) all reported low intersubject variability and no significant age-related change in K_D. Assumptions regarding the low intersubject variability of V_2 remain unproven. Further studies are needed with plasma tracer concentrations on Day 2 in order to provide validation.

Interpretation of Age Effect

Since β-CIT has high affinity for 5-HT as well as dopamine transporters (*34*), a confounding serotonergic contribution must be considered in the present study. It is unlikely, however, that an age-related decline in [^{123}I]β-CIT striatal binding reflects a significant loss of 5-HT transporters as well as dopamine transporters. We have previously demonstrated that [^{123}I]β-CIT uptake in striatum is associated almost exclusively with dopamine transporters in primates (*37*). Moreover, any small contribution to striatal [^{123}I]β-CIT uptake by 5-HT transporters is likely to bias *against* an aging effect, since 5-HT transporter density is preserved with age in most brain regions (*49–50*), including neostriatum (*50*).

Another potential confounding factor in our data is a change in endogenous dopamine levels with age. It is theoretically possible that [^{123}I]β-CIT binding could be interfered with by competition with endogenous dopamine. This possibility is not, however, a likely explanation for the present findings, because the decline in striatal dopamine content with age (*14*) would actually tend to increase [^{123}I]β-CIT binding. Moreover, in our recent studies with this tracer in nonhuman primates (*37*), infusion of L-DOPA (50 mg/kg intravenously) failed to displace striatal [^{123}I]β-CIT binding, suggesting that changes in intrasynaptic dopamine concentration with age would have little effect on the outcome measures used in the present study.

Whether the reduction in [^{123}I]β-CIT binding with age represents a loss of transporters per unit volume or simply an age-related shrinkage in the whole corpus striatum is unclear. A recent study with quantitative MRI has demonstrated that striatal volume declines with age (*51*). Given that our analysis uses standard ROIs of preset area and shape, it may be sensitive to such changes in striatal volume. Moreover, this problem is not fully solved by the use of variable ROIs to outline the anatomical borders of striatum (*25,26*), since smaller structures are still susceptible to greater partial volume effects (*52*). Future studies with MRI coregistration, segmentation and image-blurring (*53*) may control for these effects. The impact of aging, however, may be better appreciated by an outcome measure that is sensitive to reduction in transporter number apart from density. Thus, a fixed ROI may, in fact, be preferable.

The extent to which the density of dopamine transporters reflects that of dopamine nerve terminals is also uncertain. The transporter-terminal relationship may be altered with age, as dopamine transporters are known to be modulated by levels of synaptic dopamine. Chronic drug administration studies in rodents have shown that reserpine decreased dopamine transporter levels (*54*). Conceivably, as the loss of dopamine neurons reaches a critical level, the remaining cells maintain synaptic dopamine levels in part by decreasing the available dopamine transporters per terminal. Compensatory mechanisms linked to dopamine cell loss in Parkinson's disease and animal models of parkinsonism have been described (*55*).

That dopamine transporters decline with age, however, probably reflects a loss of nerve terminals rather than "downregulation" of transporters. Nigrostriatal dopamine cell loss with age closely mirrors the decline in dopamine transporter binding with [^{123}I]β-CIT reported here. McGeer et al. (*13*) found that the number of dopamine cell

bodies in the pars compacta of the substantia nigra (SNc) shows roughly a 50% reduction over the age range of the present study. Complicating this issue is the uncertain relationship between dopamine cell bodies in the SNc and terminals in striatum. One report (*56*) found no change in the relative number of tyrosine hydroxylase (TH) containing nerve terminals in caudate with age, suggesting that the remaining dopamine neurons compensate to maintain caudate levels of TH-containing nerve terminals. That study was based on only six subjects (including one over age 43) and warrants replication in a larger sample.

Yet another uncertainty is whether an age-related decline in [^{123}I]β-CIT uptake in the striatum reflects a generalized loss of dopamine transporters with aging. Dopamine transporters are also located in the midbrain and in cortical and limbic areas (*57,58*). The present study focused on [^{123}I]β-CIT uptake in the striatum, in part because this region has the most robust signal of any area in the brain. Extrastriatal dopamine transporters are difficult to quantitate with current SPECT methods because some regions (e.g., substantia nigra) are too small for the limited resolution of present imaging devices, whereas other regions (e.g., cortical and limbic areas) have low ratios of specific-to-nonspecific uptake. Alterations of dopamine transporter levels in striatum with age may reflect those in extrastriatal regions, although this possibility cannot be assessed directly with presently available SPECT instrumentation.

The absence of a gender effect on [^{123}I]β-CIT binding in the present dataset should be taken with caution. Autoradiographic studies in rodents with [^{3}H]GBR-12935 demonstrate higher dopamine transporter density in intact women than in men or ovariectomized females (*59*). Chronic estradiol and/or progesterone treatment increased dopamine transporter density in ovariectomized females (*60*); whereas acute estradiol but not progesterone treatment had a similar effect (*61*). Dopamine transporter density has also been shown to fluctuate during the female estrous cycle (*59*). Factors in the present study mitigating against a gender effect or an interaction of age and gender include the small sample size, the high outlying value of V_3'' for an individual 24-yr-old man, the absence of controls for the menstrual cycle and the inclusion of two women taking oral contraceptives. Future studies should address this issue more rigorously, given inter alia a possible neuroprotective role of female sex hormones in Parkinson's disease (*62*).

CONCLUSION

Our findings help to explain the decreased efficacy with age of psychostimulant drugs interacting with the dopamine transporter (*63–67*). They certainly demonstrate the necessity of careful age-matching when [^{123}I]β-CIT and other dopamine transporter ligands are used in the diagnosis of Parkinson's disease (*38*). Future studies with SPECT and PET should examine whether changes in the nigrostriatal dopamine system with normal aging have functional significance, specifically, whether they are related to cognitive and motor deficits that occur in the elderly. In vivo methodologies are uniquely poised to study dopamine neurochemistry in relation to behavioral changes in aging.

NOTE ADDED IN PROOF

While this paper was under review, Volkow et al. (*68*) reported a similar decrease in dopamine transporters with age using [^{11}C]cocaine and PET.

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