Nucl. Med. Biol. Vol. 16, No. 6, pp. 569–576, 1989
Int. J. Radiat. Appl. Instrum. Part B
Printed in Great Britain.

0883-2897/89 $3.00 + 0.00

Biodistribution, Dosimetry, Metabolism and Monkey PET Studies of [^{18}F]GBR 13119. Imaging the Dopamine Uptake System *In Vivo*

MICHAEL R. KILBOURN,* JAMES E. CAREY, ROBERT A. KOEPPE, MICHAEL S. HAKA, GARY D. HUTCHINS, PHIL S. SHERMAN and DAVID E. KUHL

Division of Nuclear Medicine, Department of Internal Medicine, University of Michigan Medical Center, Ann Arbor, MI 48109, U.S.A.

(*Received* 13 *January* 1989)

The *in vivo* charactersitics of a new radiotracer, [^{18}F]GBR 13119, have been examined. Full body biodistribution in rats has been determined and the expected human dosimetry calculated. Pharmacological specificity of *in vivo* regional brain distribution in rats was examined. Blockage of specific binding was accomplished by dopamine reuptake inhibitors but no effect was observed for pretreatment with serotonin or norepinephrine reuptake inhibitors. Preliminary examination of rat blood shows the presence of radiolabeled metabolites, which can be rapidly identified using bonded-phase (Sep-Pak) chromatography. Finally, the striatum of living primates has been imaged using PET and i.v. administration of [^{18}F]GBR 13119. These results represent the intermediate steps in the development of [^{18}F]GBR 13119 as a radiotracer for the study of the dopamine uptake system in man.

Introduction

As part of our program to develop new positron-emitter labeled radiopharmaceuticals for PET studies of neurotransmitter function in normal and diseased brain, we have prepared a ^{18}F labeled antagonist of the presynaptic dopamine uptake system. This radiotracer, [^{18}F]GBR 13119 (1-[(4-[^{18}F]fluorophenyl) (phenyl)methoxy)ethyl]-4-(3-phenylpropyl)-piperazine, (Fig. 1) (Kilbourn and Haka, 1988), shows a regional rat brain distribution determined by *ex vivo* analyses (Kilbourn, 1988) and *in vivo* quantitative autoradiography (Kilbourn *et al.*, 1988) that is consistent with the distribution of dopaminergic neurons. This regional distribution is furthermore not affected by dopamine receptor agents (spiperone, *cis*-flupenthixol) but is blocked by administration of competing ligands for the dopamine uptake system (nomifensine, mazindol). These studies have suggested there is successful binding of the radiotracer to the target system, the presynaptic dopamine uptake site.

The demonstration of regional and pharmacologically distinct uptake and retention of a radiotracer in rodent brain is but the first step in the development of a new radiopharmaceutical for human neurological PET studies. Prior to initiating human studies with [^{18}F]GBR 13119, we sought to (a) demonstrate the successful imaging of the dopamine uptake site in primate brain, (b) further evaluate the pharmacological specificity of [^{18}F]GBR 13119 binding in rodent brain, and (c) determine the biodistribution and examine the metabolism of [^{18}F]GBR 13119 in rodents for the purpose of estimating human dosimetry. This paper describes these intermediate steps in the development of [^{14}F]GBR 13119 as a radiopharmaceutical for the study of presynaptic dopamine uptake sites in the human brain using PET.

*Author for correspondence.

Experimental

Synthesis of [^{18}F]GBR 13119

No-carrier-added [^{18}F]GBR 13119 was prepared by slight modifications of the methods previously described (Kilbourn and Haka, 1988; Haka *et al.*, 1989). Radiochemical and chemical purity was >97% by TLC (silica gel, 95:5 chloroform:methanol) and HPLC (C_8 column, 70:25:5 acetonitrile:water:140 mM $HClO_4$). Specific activity determined from an analytical HPLC was

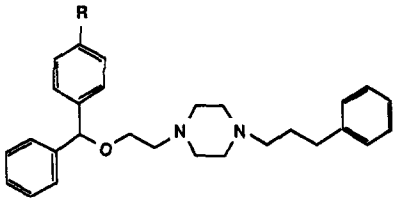

Fig. 1. Structures of GBR 12935 and [^{18}F]GBR 13119.

> 2000 Ci/mmol (limit of detection by u.v.). The product was prepared for injection by dissolution in 0.9% saline containing a trace of hydrochloric acid.

Biodistribution studies

Rats (Sprague–Dawley, 180–250 g) were injected with 10–50 μCi of [^{18}F]GBR 13119. At specified times the animals were killed by decapitation and the tissues dissected out, weighed, and counted for ^{18}F. Radioactivity remaining in the carcass was measured in a dose calibrator.

Pharmacological specificity

Studies of the pharmacological specificity of regional brain uptake of [^{18}F]GBR 13119 were performed in a manner analogous to that previously reported (Kilbourn, 1988). Animals (Sprague–Dawley rats, 150–200 g; 4–6 animals per study) were injected i.v. with large doses of potential blocking agents (5 mg/kg nisoxetine, 10 mg/kg fluoxetine, 10 mg/kg GBR 13119: all as salts dissolved in saline solution) 30 min prior to injection of 10–50 μCi of [^{18}F]GBR 13119. Control animals were injected with saline. After 1 h the animals were killed and the brain rapidly removed and dissected into specific areas of interest (Glowinski and Iversen, 1966). These tissue samples were then weighed and counted for ^{18}F.

Metabolism studies

[^{18}F]GBR 13119 (816 and 726 μCi) was injected via the femoral vein into two Sprague–Dawley rats (370 and 288 g). At 5, 30, 60 and 90 min blood samples (0.25 mL) were withdrawn via the tail vein. These blood samples were mixed with 1 mL of ethanol, vortexed briefly, and the mixture centrifuged. The supernatant was drawn off and diluted with 9 mL of water, and the mixture passed through a C-18 Sep-Pak (Waters Co). The solid phase was washed with 10 mL of 30% ethanol and 10 mL of 40% ethanol. The eluents and C-18 Sep-Pak were then counted for ^{18}F.

To determine recovery capabilities and loss of [^{18}F]GBR 13119 in the intermediate washings, control experiments were done using rat blood and authentic [^{18}F]GBR 13119 (>99% purity). Blood workup and Sep-Pak analysis were done by the same protocol as described above.

PET imaging studies

Studies were done in female pigtail monkeys (*Macaca nemistrina*) weighing 4–6 kg. The animals were anaesthetized with ketamine and administered atropine (0.04 mg/kg) and xylazine hydrochloride (1 mg/kg). Ketamine was repeated as necessary to maintain anaesthesia. Studies were done using the TCC 4600A PET scanner (three ring, five slice tomograph) operating in the high resolution mode (12 mm FWHM). Cerebral blood flow studies using i.v. injections of 3–5 mCi of ^{15}O labeled water were done prior to the [^{18}F]GBR 13119 study to aid in positioning of the animals and selection of the proper imaging plane through the striatum. The animals were then injected intravenously with from 1.35 to 6.2 mCi of no carrier added [^{18}F]GBR 13119 and sequentially imaged (1 min frames early, progressing to 30 min frames at longer time periods) for a total of 80–150 min. Studies repeated in the same animal were at least three weeks apart.

Results

Biodistribution of [^{18}F]GBR 13119. The full body distribution of [^{18}F]GBR 13119 is shown in Table 1. Radiotracer uptake is highest in the organs involved in metabolism (liver, kidneys). Low bone uptake indicates insignificant defluorination, as would be expected for an aryl fluoride. At three time points (5, 15 and 60 min) radioactivity in the remaining carcass was assayed to allow calculation of remainder of body radioactivity levels. Essentially all of the injected dose could be accounted for (91.1 ± 7% and 5 min, 99.6 ± 3.6 at 15 min, 97.8 ± 3.3 at 60 min).

Table 1. Biodistribution of [^{18}F]GBR 13119 in rats (data given as mean ± SD, $n = 5$)

	Time (min)					
	5	15	30	60	90	120
Brain	0.548 ± 0.111	0.443 ± 0.138	0.392 ± 0.084	0.239 ± 0.039	0.201 ± 0.043	0.146 ± 0.033
Heart	0.801 ± 0.098	0.411 ± 0.052	0.237 ± 0.079	0.178 ± 0.026	0.187 ± 0.04	0.138 ± 0.018
Kidney	4.38 ± 0.192	22.91 ± 0.207	2.05 ± 0.09	1.65 ± 0.042	1.23 ± 0.069	0.929 ± 0.141
Liver	28.97 ± 3.82	37.9 ± 3.19	36.86 ± 2.82	25.9 ± 1.62	27.2 ± 2.48	26.24 ± 5.63
Lung	6.14 ± 1.01	5.3 ± 0.75	3.68 ± 1.34	2.28 ± 0.83	2.55 ± 0.16	1.76 ± 0.68
Testes	0.258 ± 0.008	0.24 ± 0.013	0.316 ± 0.027	0.323 ± 0.005	0.391 ± 0.045	0.386 ± 0.05
Ovary	0.154 ± 0.047	0.178 ± 0.009	0.125 ± 0.007	0.092 ± 0.012	0.137 ± 0.001	0.094 ± 0.012
Rest of body	50.5 ± 5.2	51.4 ± 2.6		68.2 ± 3.35		
Total	91.1 ± 7.2	99.66 ± 3.67		97.85 ± 3.35		

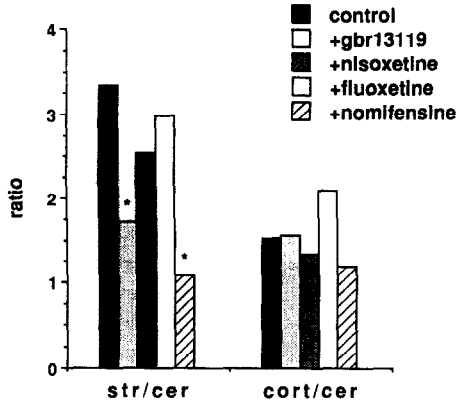

Fig. 2. Pharmacological specificity of striatal uptake and retention of [^{18}F]GBR 13119 in rat brain ($n = 22$ for controls: $n = 4$–6 for all other data points). Pretreatments were with 10 mg/kg GBR 13119, 5 mg/kg nisoxetine, 10 mg/kg fluoxetine or 8 mg/kg nomifensine (see text). Data are given as mean ± SD and were analyzed for statistical significance using a two-tailed Students t-test: $^*P < 0.05$.

Pharmacological specificity

The regional brain distribution [^{18}F]GBR 13119 in rats, and the effect of blocking doses of dopamine receptor agents and dopamine reuptake inhibitors, have been previously reported (Kilbourn, 1988). We have further examined the specificity of the [^{18}F]GBR 13119 brain uptake by attempting to block radiotracer accumulation and retention by preinjection with nisoxetine, a norepinephrine reuptake inhibitor; fluoxetine, a serotonin reuptake inhibitor; and GBR 13119. Nisotexine and fluoxetine have no observable effect on the striatum-to-cerebellum ratio (Fig. 2) or %ID/brain (data not shown) obtained with [^{18}F]GBR 13119. The target tissue uptake and retention is, however, blocked by administration of GBR 13119 (Fig. 2), as previously observed with nomifensine (data shown for comparison).

Dosimetry calculations

The expected absorbed doses to humans shown in Table 2 were calculated using the rat biodistribution data from Table 1, following the MIRD formalism (Loevinger and Berman, 1976). The percent administered dose per organ values were modified to reflect the different proportions of organ to total body mass in rat and man (Roedler, 1980). Residence times were obtained by integration under the organ time vs activity curves, with the effective half-life of [^{18}F]GBR 13119 assumed to be equal to the physical half-life of ^{18}F for times exceeding the last data point. Residence times were entered into the MIRDOSE2 program (Watson *et al.*, 1988) for the generation of absorbed doses to selected target organs per unit administered activity. No urinary excretion was observed over the sampling period and therefore the urinary bladder was not considered as a source organ.

Table 2. Calculated absorbed dose estimates to the adult from [^{18}F]GBR 13119

Target organ	Total dose (mGy/MBq)	(rad/mCi)
Brain	0.00419	0.0155
Heart wall	0.0159	0.0587
Kidney	0.0208	0.0767
Liver	0.252	0.193
Lungs	0.375	0.132
Ovaries	0.378	0.139
Red marrow	0.012	0.0443
Testes	0.00634	0.0234
Uterus	0.0115	0.0424
Total body	0.0113	0.0416

Metabolism studies

The distribution of radioactivity in the metabolism studies is shown in Table 3. In control experiments, recovery of authentic [^{18}F]GBR 13119 from blood is 85%. Extraction of radioactivity was similar (range 13–19%) in the rat blood samples used for the metabolite analyses. [^{18}F]GBR 13119 is very well retained on a C-18 Sep-Pak, with only a 3% breakthrough up to concentrations of 40% ethanol in water. The percentage of [^{18}F]GBR 13119 in whole blood was calculated using a correction for the retention of [^{18}F]GBR 13119 radioactivity in the cellular debris (15%) and the loss through the early washings (3% through the 40% ethanol wash).

PET imaging with [^{18}F]GBR 13119

An example of the imaging of [^{18}F]GBR 13119 in the striatum of primate brain is shown in Fig. 3, and the time vs activity curve for striatum and cerebellum one of the PET studies is shown in Fig. 4. Following i.v. injection of [^{18}F]GBR 13119, radioactivity uptake in the brain was rapid and reached maximum levels at 2–3 min. Radioactivity in both striatum and cerebellum decreased steadily thereafter, with a faster rate of washout from the cerebellum than from the striatum. At the end of the study (150 min) cerebellar levels had decreased to 34% of the peak value, and striatum levels to 59% of the peak values. The striatum-to-cerebellum ratio thus steadily increased from 1 to a value of 1.76 at the end of the study shown in Fig. 3, providing a clear image of the radiotracer retention in the striatum.

Table 3. Distribution of radioactivity upon C-18 Sep-Pak analysis of blood of rats ($n = 2$) injected intravenously with [^{18}F]GBR 13119. Data shown are average values. Percentage of authentic [^{18}F]GBR 13119 in blood samples was calculated using corrections for recovery of radioactivity from cellular debris and breakthrough into ethanol/water washes determined in control experiments (for details see Experimental)

	Time (min)				
	5	30	60	90	Control
	Percent of radioactivity				
Pellet	13	19	13	19	15
10% EtOH	12	12	6	6	1.5
30% EtOH	2	4	6	9	0
40% EtOH	2	1	1	2	1.5
C18	71	64	74	64	82
Total	100	100	100	100	100
% GBR 13119	82	76	79	77	100

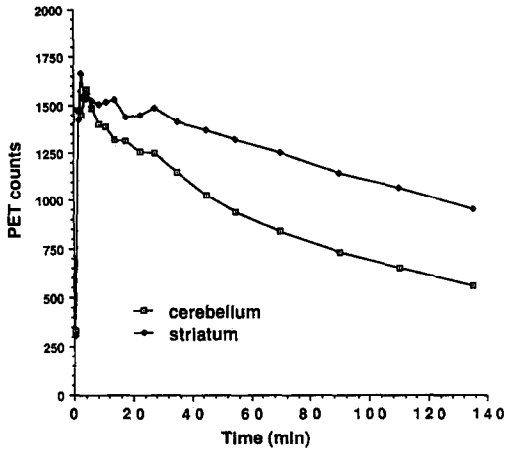

Fig. 4. Tissue–activity curves for regions of monkey brain following administration of [^{18}F]GBR 13119.

Discussion

The present studies into the pharamcological specificity of [^{18}F]GBR 13119 binding in rat brain have confirmed and extended our previous findings. Antagonists of the norepinephrine (nisoxetine; Wong and Bymaster, 1976) and serotonin (fluoxetine; Wong *et al.*, 1983) reuptake systems are ineffective in blocking [^{18}F]GBR 13119 uptake (Fig. 2), as expressed as striatum-to-cerebellum (target-to-nontarget) ratios. Selective uptake and retention is however blocked with known active dopamine reuptake blockers of this class of compounds (GBR 12935, GBR 13119) or of different chemical structure (nomifensine, mazindol) (Kilbourn, 1988). These results are consistent with selective binding of [^{18}F]GBR 13119 to the dopamine uptake site *in vivo* and have been confirmed using *in vivo* autoradiography with [^{18}F]GBR 13119 (Kilbourn *et al.*, 1988). The whole body distribution is consistent with a lipophilic radiotracer (early accumulation in lung, for example) that is extensively metabolized by the liver. The data in Table 1 suggest that the mode of excretion is not via the urine, and thus the bladder should not be the critical organ in the human dosimetry calculations. This was taken into account in the calculation of the dosimetry shown in Table 2. The estimated absorbed doses per unit administered activity are within the range delivered by established radiopharmaceuticals labeled with positron emitting radionuclides (ICRP, 1988). This will allow sufficient amounts of [^{18}F]GBR 13119 to be administered to patients without a high radiation burden.

The use of appropriately labeled compounds in this class of drugs, the aryl 1,4-dialkylpiperazines, have been suggested by ourselves and others (Chagroui *et al.*, 1987; Janowsky *et al.*, 1987) as potential candidates for PET imaging of the dopamine reuptake system in neurodegenerative diseases. A close structural congener, GBR 12909 (1-(2-(bis(4-fluorophenyl)methoxy)ethyl)-4-(3-phenylpropyl)piperazine), is currently in clinical trials for treatment of Parkinson's disease symptoms (Sogaard *et al.*, 1988). A marker for the presynaptic dopamine uptake system could serve to provide information, via PET, on dopaminergic cell density in neurodegenerative diseases, and thus be a valuable complement to studies employing 6-[^{18}F]fluorodopa (for measurement of dopamine synthesis) or radiolabeled dopamine D_2 receptor agents (butyrophenone neuroleptics, benzamides). Using PET and i.v. injection of [^{18}F]GBR 13119, we have now successfully demonstrated the *in vivo* uptake and retention of radioactivity in the striatum of living primates (Fig. 3). Brian uptake of this radiotracer peaked early and showed differential washout rates in striatum relative to all other tissue of the brain, leading to a striatum to cerebellum ratio of 1.76 at the conclusion of the study shown in Fig. 4. This result was reproduced with this monkey (striatum/cerebellum (s/c) ratio of 1.46 at 70 min; study terminated early) and was slightly higher in a second monkey (s/c ratio of 1.95 at 135 min). It must be noted that these are approximate values only, and should be similar or better in the human brain, as it is very difficult to obtain ROI data for a small monkey brain using a PET scanner with this spatial resolution; this problem in primate imaging has been noted and discussed by others (Aquilonius *et al.*, 1987; Mintun *et al.*, 1984; Hoffman *et al.*, 1979). No significant retention of radioactivity was noted in any other region of the brain, although the washout from brain tissue in general is fairly slow. We have not determined if this general brain uptake is due to specific binding to a class of homogeneously distributed sites; such a distribution is however consistent with non-specific binding simply due to the lipophilic nature of the tracer. Our PET imaging results are slightly different than those observed for ^{11}C labeled nomifensine, a dopamine uptake blocker previously used for PET studies in monkeys and man (Aquilonius *et al.*, 1987). Both radiotracers are rapidly taken up into brain tissue. Loss of radioactivity in both target (striatum) and non-target (cerebellum) tissue is more rapid with [^{11}C]nomifensine than with [^{18}F]GBR 13119 (65 vs 22% in striatum over 70 min). Striatum to cerebellum ratios, however, are similar at 50 min (1.53 ± 0.05 for [^{11}C]nomifensine and 1.51 (average, $n = 2$) for [^{18}F]GBR 13119), most likely due to the higher lipophilicity and higher non-specific binding of [^{18}F]GBR 13119. One advantage of the ^{18}F labeled ligand is that imaging at longer time points is feasible, and the striatum to cerebellum ratio continues to improve. From the data of Aquilonius *et al.* it is not evident that the striatum-to-cerebellum ratio would continue to improve with longer imaging times using [^{11}C]nomifensine (or a ^{18}F labeled derivative of nomifensine). Uptake and retention of [^{11}C]nomifensine was also seen in the thalamus, due to the high binding affinity of this drug for the norepinephrine reuptake system (Fielding and Szewczak, 1984). In

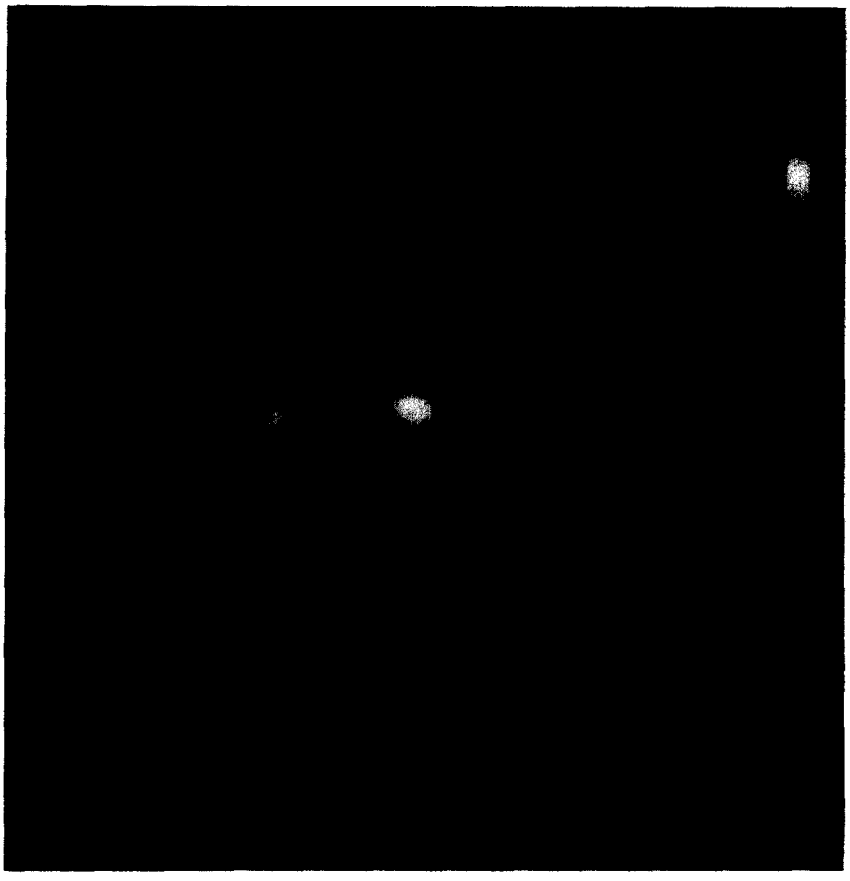

Fig. 3. PET image of [^{18}F]GBR 13119 distribution in monkey brain, showing localization in the striatal areas.

contrast, no uptake into thalamus was evident in our monkey studies with [^{18}F]GBR 13119, consistent with the low affinity of this class of compounds for the norepinephrine reuptake system (Van der Zee *et al.*, 1980).

The metabolism of radiotracers is a potential complicating factor in the construction of quantitative mathematical models of brain radiotracer pharmacokinetics. Uptake of metabolites into the target organ should be minimal, and it is necessary to determine the chemical identity of radioactivity in blood (radiotracer vs metabolites) to allow construction of a true input function for the mathematical model. Studies of the metabolism of the GBR class of compounds have not been published. Radiolabeled metabolites were not observed after *in vivo* injection of [^{3}H]GBR 12935 into mice and analysis of brain tissue (extraction of radioactivity and HPLC analysis) (Andersen *et al.*, 1987). We have examined whole blood for the presence of metabolites using the bonded-phase chromatography system approach successful with several other PET radiopharmaceuticals. There are definitely metabolites of [^{18}F]GBR 13119 in blood as soon as 5 min after i.v. injection into rats, amounting to 18% of the total radioactivity. This proportion of metabolites to authentic [^{18}F]GBR 13119 does not change greatly during the length of the study (90 min). This reasonably constant metabolite/radiotracer ratio contrasts with the increasing proportions of metabolites observed in blood after injection of many other radiopharmaceuticals (e.g. [^{18}F]spiperone (M. R. Kilbourn *et al.*, unpublished), [^{18}C]tropanyl benzilate (Mulholland *et al.*, 1988)). We have not identified the chemical form(s) of the metabolites. It is reasonable to expect that GBR 13119 is metabolized by N-dealkylation reactions involving the piperazine ring, or O-dealkylation involving the ether linkage. O-Dealkylation would produce 4-[^{18}F]fluorobenzhydrol, which should be conjugated and excreted through the urine, as has been previously reported with a structurally related compound cinnarizine (1-benzhydryl-4-cinnamylpiperazine) (Soudijn, 1968). The lack of urinary excretion of radioactivity in the rat suggests that, at least in this species, the major metabolism routes are more likely N-dealkylation reactions. Such reactions should produce hydrophilic metabolites that should not cross the blood–brain barrier. For these reasons we are optimistic that metabolites of [^{18}F]GBR 13119 will not be a problem in PET imaging; this will have to be conclusively proven using either analysis of brain tissue for metabolites of [^{18}F]GBR 13119 or the blood transfusion method (Welch *et al.*, 1988). As the metabolites of [^{18}F]GBR 13119 can be very simply separated by the Sep-Pak technique, determination of an accurate input function should not be a deterrant to quantitative mathematical modeling.

[^{18}F]GBR 13119 would thus appear a promising new radiotracer for the *in vivo* study of the dopamine uptake system with PET. The construction and validation of a tracer kinetic model of [^{18}F]GBR 13119 pharmacokinetics, and the application of this potential new radiopharmaceutical in MPTP-lesioned monkeys and in man, are underway.

Acknowledgements—This work was supported by a grant from the National Institutes of Health (NINCDS 2-PO1-NS15655) and a NIH Training Grant 1-T32-NS07222 (to M. S. H.), and a Grant from the Department of Energy (DE-AC02-76EV02031). The authors thank Dr David Robertson (Lilly) for samples of nisoxetine and fluoxetine, Dr P. Van der Zee (Gist-Brocades) for samples of GBR 12935 and GBR 13119, Dr Ed Domino for use of one of the monkeys, and Drs Anne Young and Jack Penney for helpful advice.

References

Andersen, P. H.; Jansen, J. A.; Nielsen, E. B. [^{3}H]GBR 12935 binding *in vivo* in mouse brain: labelling of a piperazine acceptor size. *Eur. J. Pharmacol.* 144:1–6; 1987.

Aquilonius, S.-M.; Bergstrom, K.; Eckernas, S.-A.; Hartvig, P.; Leenders, K. L.; Lundquist, H.; Antoni, G.; Gee, A.; Rimland, A.; Unlin, J.; Langstrom, B. *In vivo* evaluation of striatal dopamine reuptake sites using ^{11}C–nomifensine and positron emission tomography. *Acta Neurol. Scand.* 76:283–287; 1987.

Chagroui, A.; Bonnet, J.-J.; Protais, P.; Constentin, J. *In vivo* binding of [^{3}H]GBR 12783, a selective dopamine uptake inhibitor, in mouse striatum. *Neurosci. Lett.* 78:175–179; 1987.

Fielding, S.; Szewczak, M. R. Pharmacology of nomifensine: a review of animal studies. *J. Clin. Psychiat.* 45:12–20; 1984.

Glowinski, J.; Iversen, L. L. Regional studies of catecholamines in the rat brain—I. *J. Neurochem.* 13:655–669; 1960.

Haka, M. S.; Kilbourn, M. R.; Watkins, G. L. Arytrimethylammonium trifluoromethanesulfonates as precursors to aryl [^{18}P]fluorides: improved synthesis of [^{18}F]GBR 13119. *J. Labeled Compd. Radiopharm.* 1989; in press.

Hoffman, E. J.; Huang, S. C.; Phelps, M. E. Quantitation in positron emission computed tomography I. Effect of object size. *J. Comput. Asst. Tomogr.* 3:299; 1979.

ICRP. *Radiation Dose to Patients from Radiopharmaceuticals*, ICRP Publication 53. Oxford: Pergamon Press; 1988.

Janowsky, A.; Berger, P.; Vocci, F.; Labarca, R.; Skolnick, P.; Paul, S. M. Characterization of sodium-dependent [^{3}H]GBR 12935 binding in brain: radioligand for selective labelling of the dopamine transport complex. *J. Neurochem.* 46:1272–1276; 1986.

Kilbourn, M. R.; Haka, M. S. Synthesis of [^{18}F]GBR 13119, a presynaptic dopamine uptake antagonist. *Appl. Radiat. Isot.* 39:279–282; 1988.

Kilbourn, M. R. *In vivo* binding of [^{18}F]GBR 13119 to the brain dopamine uptake system. *Life Sci.* 42:1347–1353; 1988.

Kilbourn, M. R.; Ciliax, B. J.; Haka, M. S.; Penney, J. B. *In vivo* autoradiography of [^{18}F]GBR 13119 binding in rat brain. Society for Neuroscience Abstracts Abstr. No. 376.11, 1988.

Loevinger, R., Berman, M. *A Revised Scheme for Calculating Absorbed Dose from Biologically Distributed Radionuclides*, MIRD Pamphlet No. 1, revised. New York: Society of Nuclear Medicine; 1976.

Mintun, M. A.; Raichle, M. E.; Kilbourn, M. R.; Wooten, G. F.; Welch, M. J. A quantitative model for the *in vivo* assessment of drug binding sites with positron emission tomography. *Ann. Neurol.* 15:217–227; 1984.

Mulholland, G. K.; Otto, C. A.; Jewett, D. M.; Kilbourn, M. R.; Sherman, P. S.; Koeppe, R. A.; Wieland, D. M.; Frey, K. A.; Kuhl, D. E. Synthesis and preliminary evaluation of [C-11-(+)-2-tropanyl benzilate (C-11 TRB) as a ligand for the muscarinic recptor. *J. Nucl. Med.* 29:932; 1988.

Roedler, H. Accuracy of internal dose calculations with special consideration of radiopharmaceuticals biokinetics. *Radiopharmaceutical Dosimetry Symposium*, Oak Ridge National Laboratory, HHS-Publ (FDA) 81-8166; 1980: pp. 1–20.

Sogaard, U.; Michalow, J.; Butler, B.; Laursen, A. L.; Ingwersen, S. H.; Skrumager, B. K.; Rafaelson, J. O. GBR 12909; A clinical phase I study of a selective dopamine uptake blocker. *Psychopharmacology* 96:274; 1988.

Soudijn, W.; van Wijngaarden, I. The metabolism and excretion of cinnarizine by rats. *Life Sci.* 7:231–238; 1968.

Van der Zee, P.; Koger, H. S.; Gootjes, J.; Hespe, W. Aryl 1,4-dialk(en)ylpiperazines as selective and very potent inhibitors of dopamine uptake. *Eur. J. Med. Chem.* 15:363–370; 1980.

Watson, E.; Stabin, M.; Bolch, W. MIRDOSE2. Oak Ridge Associated Universities, Oak Ridge, Tenn.; 1988.

Welch, M. J.; Katzenellenbogen, J. A.; Mathias, C. J.; Brodack, J. W; Carlson, K. E.; Chi, D. Y.; Dence, C. S.; Kilbourn, M. R.; Perlmutter, J. S.; Raichle, M. E.; Ter-Pogossian, M. M. N-(3-[^{18}F]fluoropropyl)-spiperone: the preferred ^{18}F labeled spiperone analog for positron emission tomographic studies of the dopamine receptor. *Nucl. Med. Biol.* 15:83–97; 1988.

Wong, D. T.; Bymaster, F. P.; Reid, L. R.; Threlkeld, P. G. Fluoxetine and two other serotonin uptake inhibitors without affinity for neuronal receptors. *Biochem. Pharmacol.* 32:1287–1293; 1983.

Wong, D. T.; Horng, J. S.; Bymaster, F. P. DL-N-Methyl-3-(*o*-methoxyphenoxy)-3-phenylpropyalmine hydrochloride, Lilly 94939, a potent inhibitor for uptake of norepinephrine into rat brain synaptosomes and heart. *Life Sci.* 17:755–760; 1976.