Imaging Histamine H1 Receptors in the Living Human Brain with Carbon-11-Pyrilamine

Victor L. Villemagne, Robert F. Dannals, Patricia M. Sánchez-Roa, Hayden T. Ravert, Silvia Vazquez, Alan A. Wilson, T.K. Natarajan, Dean F. Wong, Kazuhiko Yanai, and Henry N. Wagner, Jr.

Division of Nuclear Medicine, The Johns Hopkins Medical Institutions, Baltimore, Maryland

The brain distribution and kinetics of the H1 receptor antagonist, carbon-11-pyrilamine (^{11}C-pyrilamine) were examined in vivo in two baboons and one human by positron emission tomography. After i.v. administration of the tracer, brain activity peaked within 20 min after injection and subsequently decreased, reflecting reversible binding to the receptor. Pretreatment with 1 mg/kg diphenhydramine reduced the brain activity at 70 min by 33%, 29%, 26%, and 23% of the control values in frontal cortex, temporal cortex, hippocampus, and cerebellum, respectively. Coinjection of 1 and 5 mg/kg cold pyrilamine reduced the activity at 70 min by 40%, 36%, 34%, and 30% in frontal, temporal, hippocampus and cerebellum, respectively. The in vivo specific binding to the H1 receptors in different brain regions at 70 min after injection correlated with the in vitro H1 histamine receptors distribution in human brain tissue obtained at autopsy, with high values in the frontal and temporal cortex and low values in cerebellum and brain stem. In the healthy human volunteer study, the value of washout of radioactivity increased by about 50% after injection of 0.7 mg/kg diphenhydramine.

J Nucl Med 1991; 32:308–311

Histamine was first isolated and characterized by Barger and Dale (*1*). Bovet and Staub (*2*) discovered histamine blocking activity during laboratory experiments with guinea pigs. These experiments led to the synthesis of the first anti-histaminic drugs in the 1940s. Ash and Schild (*3*) found that these anti-histaminics blocked only some of the effects of histamine; thus, they proposed a subtype of receptors, the H1. Black and colleagues (*4*) described the "pyrilamine insensitive" or H2 receptor subtype and Arrang and collaborators (*5*) proposed the presynaptic H3 receptor that controls both the release and synthesis of histamine (*6, 7*). Further studies have shown that the H1 subtype acts through an increase in the Ca^{++} influx which stimulates the formation of cGMP (*8*); while the H2 subtype acts through cAMP (*9*). The presynaptic H3 subtype is thought to be related to restricted influx of Ca^{++} (*6,7*).

Histamine was proposed as a brain neurotransmitter by Snyder and Taylor (*10*) and Schwartz (*11*). Central histamine H1 receptors are believed to be involved in arousal, locomotor activity, appetite control, cardiovascular regulation, and thermo-regulation. They may be altered in some disease states (*12*).

Pyrilamine (also known as mepyramine) is a potent and selective histamine H1 antagonist. Tritium-labeled pyrilamine has been widely used in brain receptor binding studies in vitro (*13–16*), autoradiography (*17*) and in vivo experiments (*18–20*).

Recently, pyrilamine has been labeled with the positron emitter carbon-11 (^{11}C) [half-life 20.4 min] (*21*). In vivo studies in mice have demonstrated specificity and saturability of binding (*21*).

In the present report, we describe the in vivo distribution and kinetics of ^{11}C-pyrilamine binding to brain histamine H1 receptors in baboons and a human volunteer, measured by positron emission tomography (PET), together with evidence of specific binding, saturability, and appropriate regional distribution.

MATERIALS AND METHODS

For the initial PET studies, two 32-kg male baboons (Papio Anubis) were anesthetized with 8–10 mg/kg alfadolone and alfaxolone acetate i.m. and intubated. Anesthesia was maintained throughout the study by a continuous i.v. infusion drip of 6–9 mg/kg/hr alfadolone and alfaxolone acetate.

The animals were comfortably restrained to the PET bed using a stereotactic headholder, which allowed reproducible positioning between studies. Arterial blood pressure, pulse, and oxygen saturation were continuously monitored during the studies. Blood oxygen saturation was always maintained above 85%.

Carbon-11-pyrilamine synthesis involved the N-alkylation of the appropriate precursor with ^{11}C-labeled methyl iodide obtained from high-specific activity ^{11}C-labeled carbon dioxide produced in a Scanditronix AB RNP-16 biomedical cyclotron, as previously described (*21*).

After i.v. injection of 20 mCi of high-specific activity ^{11}C-pyrilamine (average specific activity was 2116 ± 439 mCi/μmole; corresponding to 0.29 nmole/kg), sequential quantitative tomographic slices of the brain passing through cerebel-

Received Feb. 20, 1990; revision accepted Aug. 14, 1990

For reprints contact: Henry N. Wagner, Jr., MD, Division of Nuclear Medicine, The Johns Hopkins Medical Institutions, 615 N. Wolfe St., Room 2001, Baltimore, MD 21205.

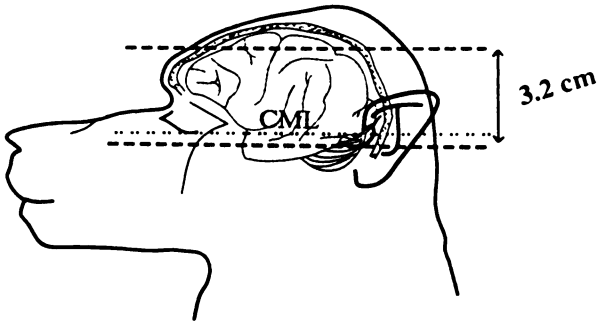

FIGURE 1
Two different imaging planes at which the scans were simultaneously obtained are shown in this drawing. The lower plane is located about 7 mm below the canto-meatal line (CML = the dotted line).

lum, temporal, and frontal lobes, were obtained in the NeuroEcat PET scanner over a period of 70 min. The scanner has three rings of detectors spaced 3.2 cm apart (inplane resolution of 8 mm FWHM). This allowed the simultaneous acquisition within the baboon brain of two imaging planes. The baboons were positioned so that the lowest plane was located about 7 mm below the canto-meatal line (Fig. 1). Arterial blood samples were drawn throughout the study.

For the competition studies, 1 mg/kg of diphenhydramine (Benadryl®) (*22*), a selective H1 antagonist, was injected intravenously 5 min prior to the administration of the radiotracer.

For the saturation studies, either 1 or 5 mg/kg of cold pyrilamine were coinjected with the radiotracer; this yielded injected specific activities of 0.15 (3.5 μmole/kg) and 0.03 mCi/μmole (17.5 μmole/kg), respectively.

Images were reconstructed, corrected for attenuation and decay, and normalized for injected dose. Regions of interest (3 × 3 pixels) were placed over the different areas of the baboon brain using a baboon PET atlas (*23*) and were averaged for the different lobes. Time-activity curves were generated from each lobe. The 1- and 5-mg/kg studies were used to establish the nonspecific binding. The in vivo specific binding to the baboon brain was compared with the known in vitro specific binding regional distribution of histamine H1 receptors in the human brain.

For the human study, a 29-yr-old healthy male volunteer was injected with 15 mCi of high-specific activity ^{11}C-pyrilamine. The tomograms were aligned using marks over an individual thermoplastic mask based on a previous MRI study. Three simultaneous slices, 3.2 cm apart, were obtained for over a period of 90 min. At 45 min postinjection, diphenhydramine (0.7 mg/kg) was injected intravenously to displace the ^{11}C-pyrilamine. Arterialized blood samples were drawn throughout the study.

RESULTS

In the baboon studies, brain radioactivity peaked within 20 min after injection and subsequently decreased, reflecting reversible binding to the receptor. The highest brain uptake at 70 min (expressed as % ID/l) was found in the cortical areas, with intermediate levels in the hippocampus, and lowest uptake in cerebellum and brain stem. The dissociation and clearance was slower in the cortical areas (frontal $t_{1/2}$ ~120 min) than in the cerebellum ($t_{1/2}$ ~80 min) and brain stem.

Pretreatment with 1 mg/kg diphenhydramine 5 min before the injection of the radiotracer reduced the brain radioactivity at 70 min by 33%, 29%, 26%, and 23% of the control values in the frontal cortex, temporal cortex, hippocampus, and cerebellum, respectively (Fig. 2), providing evidence of binding of ^{11}C-pyrilamine to the histamine H1 receptor subtype.

Coinjection of 1 mg/kg cold pyrilamine reduced the radioactivity at 70 min by 40, 36, 34, and 30% in the frontal cortex, temporal cortex, hippocampus, and cerebellum, respectively (Fig. 2). The degree of blockade obtained at 70 min with coinjection of 5 mg/kg of cold pyrilamine was similar to that obtained with 1 mg/kg (Fig. 3). These findings are in good agreement with previous in vivo mice studies with either ^{3}H or ^{11}C-labeled pyrilamine, where 1 mg/kg cold pyrilamine completely saturated the specific binding (*19,21*).

The in vivo specific binding, obtained by subtracting the values of the 1 mg/kg cold pyrilamine study from those of the control, in several regions of the baboon brain correlated well with the in vitro H1 histamine receptor distribution from human brain tissue obtained at autopsy (r = 0.88) (Fig. 4), i.e., high values in the frontal and temporal cortex, intermediate values in the hippocampus, and low values in cerebellum and brain stem (*16*).

In the human study, there was less accumulation of radioactivity in the sensory-motor area than in the other cortical areas (Fig. 5). The highest brain uptake at 40 min after injection (expressed as % ID/l) was found in the cortical areas, intermediate in the hippocampus, and lowest in cerebellum and brain stem. Radioactivity peaked between 10–15 min postinjection. The dissociation was slower in the cortical areas (frontal $t_{1/2}$ ~70

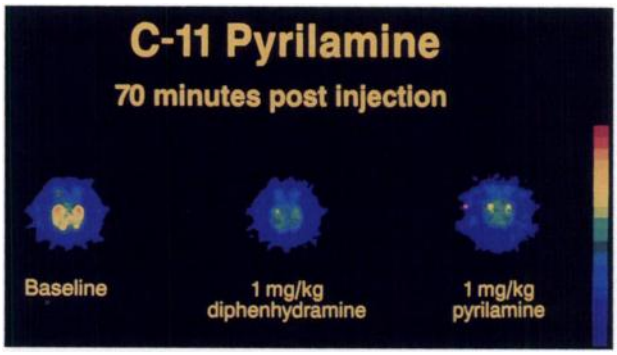

FIGURE 2
The first PET image corresponds to the control baboon PET study about 70 min postinjection at the lower imaging plane. The second image corresponds to the 1 mg/kg diphenhydramine study. The third shows the 1 mg/kg "cold" pyrilamine blocking study. There was a progression in the decrease of binding in all brain areas, but there was no decrease in the activity in the surrounding soft tissue.

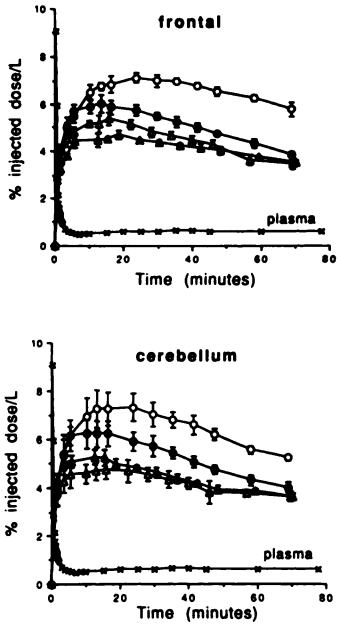

FIGURE 3
Baboon PET study time activity curves from the frontal cortex, cerebellum, and plasma are shown. Note that in the control study (○) the radioactivity peaks around twenty minutes followed by a slow dissociation. The administration of 1 mg/kg diphenhydramine (●) markedly decreased the ^{11}C-pyrilamine binding, showing specificity of the binding of ^{11}C-pyrilamine to histamine H1 receptor subtype. Coinjection of both 1 (▲) and 5 (△) mg/kg "cold" pyrilamine decrease the binding to the same degree, showing saturability of the binding.

min) than in the cerebellum ($t_{½}$ ~50 min), but faster than in the baboon. The administration of 0.7 mg/kg diphenhydramine at 45 min after injection caused increased dissociation. The half-life of clearance decreased to 30 and 25 min in the frontal and cerebellum, respectively (Fig. 6). The regional radioactivity distribution at 40 min correlated well with the in vitro H1 histamine

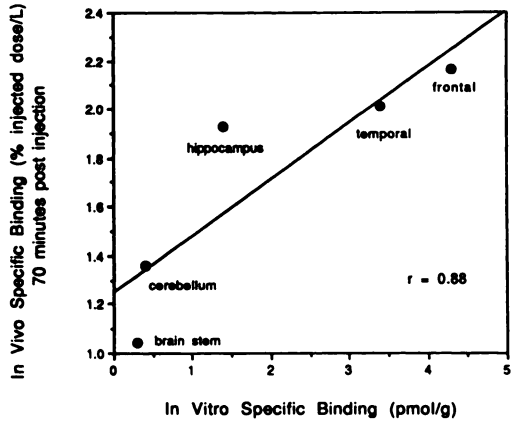

FIGURE 4
The in vivo specific binding regional distribution at 70 min postinjection (mpi) in the baboon is compared to the known in vitro regional distribution in the human brain.

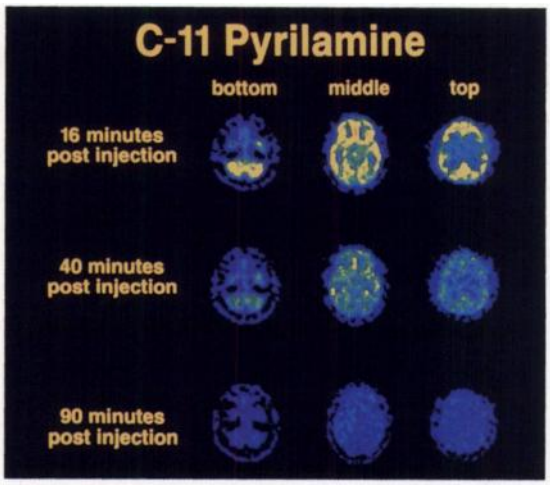

FIGURE 5
PET images from the human study. The upper row of images correspond to the 16-min scan, the middle row correspond to the 40-min scan, and the lower row to the 90-min scan (45 min after the administration of diphenhydramine).

receptor distribution from human brain tissue obtained at autopsy (r = 0.91).

DISCUSSION

We observed the highest specific binding in the frontal cortex in both the baboon and the volunteer. There are considerable differences in the in vitro histamine

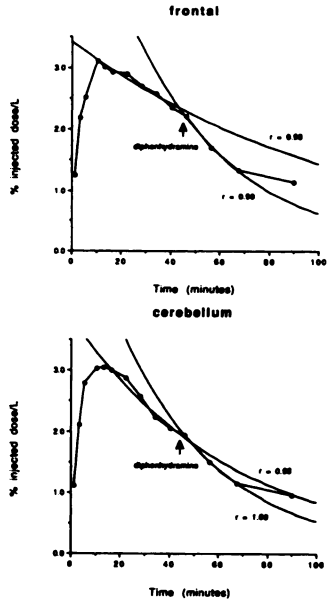

FIGURE 6
Time-activity curves from the frontal cortex and cerebellum in the human study. The arrows indicate the time at which 0.7 mg/kg of diphenhydramine were injected.

H1 receptor densities in different species. In humans, the highest concentration is observed in the frontal cortex, while in the guinea pig and the rat, the highest specific binding is found in the cerebellum and hypothalamus, respectively (*16*).

Diphenhydramine was used to block H1 receptors because it is a well-characterized commercially available drug, that produces very mild pharmacologic effects. We observed a high level of blockade of central histamine H1 receptors in vivo after administration of a therapeutic dose of diphenhydramine. Despite a 15-fold lower affinity of diphenhydramine compared to pyrilamine in vitro (*16*), we found that the same dose of each by weight, produced similar degree of blockade of ^{11}C-pyrilamine binding in vivo.

Carbon-11-pyrilamine binding to histamine H1 receptors is saturable, displaceable, and the in vivo regional distribution correlates with the known in vitro distribution of histamine H1 receptors. The reversible kinetics of the binding allows attainment of equilibrium during the scanning period, thus facilitating the quantification of receptor densities using high- and low-specific activities, or by studies in the presence of the unlabeled inhibitor diphenhydramine. Both methods have been used for other reversible binding ligands such as ^{11}C-N-methylspiperone binding to S2 receptors (*24*).

These results demonstrate the feasibility of histamine H1 receptor imaging in vivo with PET, which makes it possible to study the role of histamine H1 receptors in the living human brain in health and disease, as well as to measure the in vivo kinetics and biodistribution of new drugs, allowing a better understanding of their pharmacologic activity. One direction of future work is the evaluation of "non-sedative" H1 antihistaminics and the investigation of the sedative characteristics of new psychotropic drugs, including antidepressants and neuroleptics.

ACKNOWLEDGMENTS

The authors would like to thank Mr. David Clough for his assistance with PET data acquisition and analysis and Mr. Robert Smoot for technical assistance with cyclotron operation and radiochemistry. This work was supported in part by USPHS grant numbers NS-15080 and CA-32845.

REFERENCES

1. Barger G, Dale HH. The presence in ergot and physiological activity of β-imidazolylethylamine. *J Physiol* 1910;40:38–40.
2. Bovet D, Staub AM. Action protectrice des éthers phénoliques au cours de l'intoxication histaminique. *C R Soc Biol (Paris)* 1937;124:547–549.
3. Ash ASF, Schild HO. Receptors mediating some actions of histamine. *Br J Pharmac Chemother* 1966;27:427–439.
4. Black JW, Duncan WAM, Durant CJ, Ganelin CR, Parsons EM. Definition and antagonism of histamine H2-receptors. *Nature* 1972;236:385–390.
5. Arrang JM, Garbarg M, Schwartz JC. Auto-inhibition of brain histamine release mediated by a novel class (H3) of histamine receptor. *Nature* 1983;302:832–837.
6. Arrang JM, Garbarg M, Schwartz JC. Autoregulation of histamine release in brain by presynaptic H3-receptors. *Neuroscience* 1985;15:553–562.
7. Arrang JM, Garbarg M, Schwartz JC. Autoinhibition of histamine synthesis mediated by presynaptic H3-receptors. *Neuroscience* 1987;23:149–157.
8. Richelson E. Histamine H1 receptor-mediated guanosine 3′,5′-monophosphate formation by cultured mouse neuroblastoma cells. *Science* 1978;201:69–71.
9. Baudry M, Martres MP, Schwartz JC. H1 and H2 receptors in the histamine-induced accumulation of cyclic AMP in guinea pig slices. *Nature* 1975;253:362–364.
10. Snyder SH, Taylor KM. Histamine in the brain: a neurotransmitter? In: Snyder SH, ed. *Perspectives in neuropharmacology.* New York: Oxford University Press; 1972;43:73.
11. Schwartz JC. Histamine as a transmitter in brain. *Life Sci* 1975;17:503–518.
12. Taylor JE, Richelson E. Histamine receptors. In: Yamamura HI, Enna SJ, eds. *Neurotransmitter receptors Part 2: biogenic amines.* New York: Chapman and Hall; 1980:71–100.
13. Hill SJ, Emson PC, Young JM. The binding of [^{3}H]mepyramine to histamine H1 receptors in guinea-pig brain. *J Neurochem* 1978;31:997–1004.
14. Tran VT, Chang RSL, Snyder SH. Histamine H1 receptors identified in mammalian brain membranes with [^{3}H]mepyramine. *Proc Natl Acad Sci USA* 1978;75:6290–6294.
15. Chang RSL, Tran VT, Snyder SH. Histamine H1 receptors in brain labeled with ^{3}H-mepyramine. *Eur J Pharmacol* 1978;48:463–464.
16. Chang RSL, Tran VT, Snyder SH. Heterogeneity of histamine H1-receptors: species variations in [^{3}H]mepyramine binding of brain membranes. *J Neurochem* 1979;1653–1663.
17. Palacios JM, Wamsley JK, Kuhar MJ. The distribution of histamine H1-receptors in the rat brain: an autoradiographic study. *Neuroscience* 1981;6:15–37.
18. Quach TT, Duchemin AM, Rose C, Schwartz JC. In vivo occupation of cerebral histamine H1-receptors evaluated with [^{3}H]mepyramine may predict sedative properties of psychotropic drugs. *Eur J Pharmacol* 1979;60:391–392.
19. Quach TT, Duchemin AM, Rose C, Schwartz JC. Labeling of histamine H1 receptors in the brain of the living mouse. *Neurosci Lett* 1980;17:49–54.
20. Diffley D, Tran VT, Snyder SH. Histamine H1-receptors labeled in vivo: antidepressant and antihistamine interactions. *Eur J Pharm* 1980;64:177–181.
21. Yanai K, Dannals RF, Wilson AA, et al. (N-methyl-[^{11}C]) pyrilamine, a radiotracer for histamine H1 receptors: radiochemical synthesis and biodistribution study in mice. *Appl Radiat Isot, Int J Rad Appl Instrum Part B* 1988;15:605–610.
22. Kanba S, Richelson E. Histamine H1 receptors in human brain labeled with [^{3}H]doxepin. *Brain Res* 1984;304:1–7.
23. Riche D, Hantraye P, Guibert B, et al. Anatomical atlas of the baboon's brain in the orbito-meatal plane used in experimental positron emission tomography. *Brain Res Bull* 1988;20:283–301.
24. Wong DF, Gjedde A, Dannals RF, et al. Quantification of dopamine and serotonin receptors in living human brain: equilibrium binding kinetics using two methods of inhibition [Abstract]. *J Cereb Blood Flow Metab* 1989;9:S247.