5-[^{18}F]Fluoro-2-(1-methyl-1H-pyrrolo[2,3-b]pyridin-5-yl)-oxazolo[5,4-b]pyridine

[^{18}F]MK-3328

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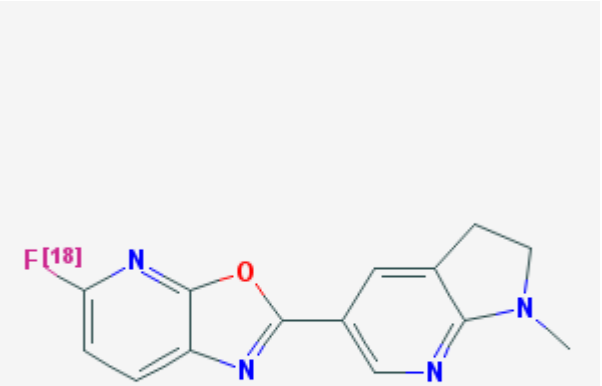
Chemical name:	5-[^{18}F]Fluoro-2-(1-methyl-1H-pyrrolo[2,3-b]pyridin-5-yl)-oxazolo[5,4-b]pyridine
Abbreviated name:	[^{18}F]MK-3328
Synonym:	
Agent Category:	Compound
Target:	β-amyloid plaques
Target Category:	Protein
Method of detection:	Positron emission tomography (PET)
Source of signal / contrast:	^{18}F
Activation:	No
Studies:	• *In vitro* • Non-human primates



Click on above structure for information in PubChem.

Background

[PubMed]

The gradual development and worsening of dementia is a characteristic feature of Alzheimer's disease (AD), and currently no cure is available for this ailment. Medications

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available to treat AD can only modify or delay the onset of this disease, and there are many ongoing preclinical and clinical studies to develop a suitable therapy to treat this condition (1). It is hypothesized (known as the amyloid cascade hypothesis) that accumulation of an amyloid precursor protein derivative, the β-amyloid protein (Aβ), in the brain is responsible for bringing about changes in the biochemical and cellular functions of the brain and leads to the progression and manifestation of AD (2). Therefore, the Aβ is believed to be a good biomarker for the detection of AD. Most earlier studies performed to investigate this disease used human post-mortem brains, and little is known about the early events that promote the development of AD or about how the formation and deposition of the Aβ contributes to the disease process (2). More recently, many noninvasive imaging probes that can be used with the various imaging modalities, have been developed and evaluated for the detection of early stage Aβ plaques; however, due to inadequate permeation across the blood–brain barrier and/or low binding to the Aβ aggregates, these agents were considered unsuitable for visualization of the Aβ plaques in the human brain (2).

In an effort to develop imaging compounds that can be used for the visualization of Aβ plaques, investigators have produced and evaluated radiolabeled compounds that can be used with positron emission tomography (PET) (3). Among these, the ^{11}C-labeled Pittsburg Compound-B ([^{11}C]PIB), which has been shown to bind specifically to Aβ deposits in post-mortem AD brains under *in vitro* conditions, is most commonly used to diagnose the condition, but the major limitation of using this probe is the short half-life of ^{11}C ($t_{1/2}$ = 20 min) and the requirement of an onsite cyclotron for the generation of this radionuclide (2, 3). AS a result, ^{11}C-labeled tracers are not the most suitable for commercialization (4), and [^{11}C]PIB is not approved by the United States Food and Drug Administration (FDA) as a diagnostic imaging agent for the detection of AD. ^{18}F-Labeled AV-45 ([^{18}F]AV-45), which has a $t_{1/2}$ = 110 min and has been developed, characterized, and evaluated for the detection of Aβ plaques, has been shown to be suitable for the identification of patients with AD, and the binding of this tracer in the brain of AD patients correlated well with the presence of Aβ plaques observed in this organ during post-mortem studies (5). However, [^{18}F]AV-45 is not approved by the FDA as yet for the detection of AD in patients. Although [^{18}F]AV-45 appears to be a promising agent for the noninvasive detection of Aβ plaques, it may not be suitable for the visualization of early-onset Aβ deposits because this tracer (and other similar agents) shows high uptake in the white matter of the brain (3, 5). In a continued effort to develop an imaging agent that is superior to those currently available for the detection of Aβ deposits, Hostetler et al. developed 5-[^{18}F]fluoro-2-(1-methyl-1H-pyrrolo[2,3-b]pyridin-5-yl)-oxazolo[5,4-b]pyridine ([^{18}F]MK-3328) and showed that it was a promising PET agent for the detection of Aβ plaques because it has a low *in vivo* uptake in the white matter and the cortical gray matter of the rhesus monkey brain (3).

Related Resource Links

Related chapters in MICAD

β-Amyloid (A4) precursor protein gene information (Gene ID 351)

Information in Online Mendelian Inheritance in Man (OMIM) Database

Clinical trials related to β-amyloid plaques

Amyloid inhibitors on U.S. Food and Drug Administration web site

Partial structure of β-amyloid protein (Protein Data Bank, NCBI)

Synthesis

[PubMed]

The synthesis of MK-3328 has been described by Harrison et al. (6), and its labeling with [^{18}F]fluoride is detailed by Hostetler et al. (3). The average yield of [^{18}F]MK-3328 was reported to be 14 ± 13%, with a specific activity of 91.5 ± 51.4 GBq/mmol (2,471 ± 1,389 Ci/mmol) and a radiochemical purity (RP) of >98% (n = 25 synthesis reactions).

For comparison purposes, some other ^{18}F-labeled compounds were synthesized and studied (3). These radiolabeled compounds are listed in Table 1 below:

Table 1: Comparison of radiolabeled compounds

Radiolabeled compound	Yield	Specific activity (per mmol)	Radiochemical purity	Number of synthesis performed
[^{18}F]-AD-278	40 ± 18%	38.2 GBq (1,032 Ci)	>99%	2
[^{18}F]AD-269	35 ± 18%	186 ± 108 GBq (5,023 ± 2,927 Ci)	>99%	4
[^{18}F]AD-265	10 ± 3%	Not reported	>98%	2
[^{18}F]AV-45	39%	36.8 GBq (994 Ci)	>98%	1

In Vitro Studies: Testing in Cells and Tissues

[PubMed]

The lipophilicity (Log D) of various Aβ ligands was determined as described by Hostetler et al. (3). The Log D values for the various Aβ binding compounds (Table 2 below) were higher (ranging from 2.91 for MK-3328 to 3.52 for AD-265) than that of PIB (Log D = 2.23).

Table 2: Lipophilicity and *in vitro* binding characteristics of various Aβ binding compounds

Compound	Lipophilicity (Log D)	Binding affinity (IC_{50} in nM)

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MK-3328	2.91	10.5 ± 1.3
AD-269	3.42	8.0 ± 3.7
AD-278	3.52	4.0 ± 3.7
AD-265	3.03	17.0*
PIB	2.23	2.3 ± 0.17

*Single determination.

In a competition binding assay using brain frontal cortex homogenates (controls) of AD patients ([$^3H_3$]DMAB was used as the binding ligand), the Log D values and IC_{50} concentrations of various nonradioactive Aβ binding compounds were compared (Table 2) (6). From this assay it was clear that all the compounds except PIB had a low affinity for the homogenates and that AD-265 was the least potent ligand. The absence of Aβ plaques in the homogenates was confirmed with immunostaining. This suggested that the low-affinity compounds (MK-3328, AD-269, and AD-278) showed no specific binding to the control brain homogenates. In addition, none of the compounds were determined to be unsuitable substrates for the P-glycoprotein transporter, and all of them had suitable cell permeability rates (for details, see Hostetler et al. (3).

Hostetler et al. compared the binding of [^{3}H]MK-3328 (5 nM) and [^{3}H]AD-269 (5 nM) to Aβ deposits in human AD donor brain slices with autoradiography of the brain slice exposure to the two labeled compounds (3). From the autoradiographic images it was clear that only [^{3}H]MK-3328 bound to the amyloid deposits in the frontal cortex of the diseased human brain. In a self-blocking experiment, nonradioactive MK-3328 was shown to completely block the binding of [^{3}H]MK-3328 to the Aβ plaques in the AD donor brain slices. In addition, [^{3}H]MK-3328 showed no binding in the cerebellum of an AD patient brain (the cerebellum of the AD patient brain is known to have very few Aβ plaque deposits). Similarly, [^{3}H]MK-3328 did not show any binding to the frontal cortex of a non-AD human donor brain. From this study the investigators concluded that the specific binding of [^{3}H]MK-3328 to the Aβ plaques was probably due to its lower lipophilicity (Log D = 2.91) compared with that of [^{3}H]AD-269 (Log D = 3.42).

Animal Studies

Rodents

[PubMed]

No publication is currently available.

Other Non-Primate Mammals

[PubMed]

No publication is currently available.

Non-Human Primates

[PubMed]

The *in vivo* evaluation of [18F]MK-3328 and the other Aβ PET tracers was not performed in transgenic rodents that produce Aβ plaques in their brains because probes that can be used for the imaging of AD in humans do not generate a specific signal in these animals (3). It has been shown that PIB does not have a high affinity for Aβ plaques in the chimpanzee or rhesus monkey brain (3). Therefore, the investigators studied the brain uptake and subsequent kinetics of [18F]MK-3328 and the other Aβ plaque-binding PET tracers to establish which one of these agents was the most suited for further studied. Because Aβ plaques are present mostly in the gray matter areas of the human AD brain, the investigators concluded that a comparative analysis of tracer uptake in the white and gray matter of the monkey brain could provide a clue to the selection of a suitable imaging agent for the imaging of Aβ deposits.

Harrison et al. studied the brain uptake and kinetics of [18F]MK-3328 in a healthy rhesus monkey (under anesthesia) with PET imaging (6). The route of administration and the amount of tracer injected into the monkey were not reported. From the PET images it was observed that the peak uptake values of the tracer in the cortex and the cerebellum of the brain were ~2.7 and ~3.0 standardized uptake value units, respectively, and in both areas of the brain the tracers were reduced to ~50% of the peak value at 45–90 min. The investigators concluded that, because the Aβ plaques in human AD brain are present throughout the brain but not in the cerebellum until the last stages of the disease, the cerebellum could be used as a suitable baseline reference region for imaging studies.

In another study, two rhesus monkeys (numbers R352 and R460) under anesthesia were used for the brain uptake and washout kinetics studies of the different Aβ deposit binding PET agents (3). The route of administration and the amount of tracer injected into each monkey were not reported. Baseline reference PET scans were obtained with [18F]MK-3328, [18F]AD-269, and [18F]AD-278 in both the monkeys; with [18F]AD-265 and [18F]AV-45, the scans were acquired from monkey R352 only. Time-activity curves generated from the PET images showed that peak uptake values of the labeled compounds in the cerebellum ranged from 2.4 to 3.2 SUV. In general, the peak uptake was lower in the white matter than in the gray matter (the cortex and cerebellum), but at later time points the white matter showed a higher retention of label compared with the cortical tissues. Similar kinetics of tracer uptake and washout were observed in the cerebellum and the cortex; both tissues showed a rapid washout of radioactivity. Using Logan graphical analysis with the cerebellum cortex as the reference point, the binding potentials (BP) of the various tracers were determined (Table 3) from the PET images as described by Hostetler et al. (3).

Table 3: Binding potential of various Aβ plaque PET imaging agents in cortex and white matter of monkeys

[^{18}F]-Tracer	Monkey R352		Monkey R460	
	Binding potential			
	Cortex	White matter	Cortex	White matter
[^{18}F]MK-3328	0.13	0.15	0.10	0.08
[^{18}F]AD-269	0.15	0.15	0.08	0.12
[^{18}F]AD-278	0.13	0.34	0.08	0.19
[^{18}F]AD-265	0.22	0.55	–	–
[^{18}F]AV-45	0.11	0.33	–	–

Among the different PET agents, the highest BP in the cortex and the white matter was observed with [^{18}F]AD-265. [^{18}F]AD-278 and [^{18}F]AV-45 showed a similar trend, with a low BP in the cortex but a high BP in the white matter. Compared with the other tracers, both [^{18}F]MK-3328 and [^{18}F]AD-269 had a lower BP in the cortex and the white matter, indicating that these tracers were probably most suitable for the imaging of Aβ plaques. The observations with [^{18}F]MK-3328 and [^{18}F]AD-269 were subsequently confirmed in a second monkey (monkey R460). Although [^{18}F]MK-3328 and [^{18}F]AD-269 had similar BP values, [^{18}F]AD-269 had a higher lipophilicity than [^{18}F]MK-3328 (see Table 2 above).

With results obtained from the *in vitro* and *in vivo* studies described above, the investigators concluded that [^{18}F]MK-3328 was superior to all the other Aβ PET agents evaluated in this study, but further evaluation is needed before [^{18}F]MK-3328 can be used for the detection of AD in humans (3).

Human Studies

[PubMed]

No publication is currently available.

Supplemental Information

[Disclaimers]

No information is currently available.

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