RADIOLABELLING OF A NEW GROUP OF TRACERS FOR PET IMAGING OF CB1 CANNABINOID RECEPTORS

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Introduction: Two different cannabinoid receptors have been characterized: CB1 which is mainly located in the neural tissue and in the peripheral tissues and CB2 which is mainly found in the peripheral tissues. Many papers have been published stating that endocannabinoids and cannabinoids play an essential role in many central and peripheral disorders, for example in neuropsychiatric or in mood and anxiety disorders.

The best known agonist for CB receptors is the Δ^9-tetrahydrocannabinol (Δ^9-THC) which originates from the hemp plant, *cannabis sativa*. This compound has been used for medicinal and intoxicating purposes since very early times. The plant and its extracts are best known as cannabis, hashish or marijuana. The pleasurable subjective effects of marijuana smoking include euphoria, feelings of tranquillity and altered perceptions.

Some early attempts to localize the CB receptors using radiolabelled agonists and antagonists were carried out by S. John Gatley at Brookhaven National Laboratory, NY in the mid-90s. Baboon SPECT studies, mouse brain dissection studies and *ex vivo* autoradiography in rat brain demonstrated rapid passage of [^{123}I]AM281 into the brain after intravenous injection, appropriate regional brain specificity of binding and reduction of binding after treatment with SR141716A.

From these early compounds many other new ones were developed. Instead of a pyrazole system like in AM281, AM4975 employs an indole system as its main functional group.

Methods: $^{11}CH_3I$ was produced in the 6" grid target at the EBCO cyclotron with the GE Box in line. The $^{11}CH_3I$ was condensed using dry ice/acetonitrile cooling into the reaction vessel containing 1 mg precursor AM4964 in 0.3 ml DMF/DMSO 3:1 vv. The mixture was then heated for 5 min at 100°C in an oil bath.

The mixture was diluted with 1 ml of the HPLC solvent and then injected onto the column. After having collected the product, the solvent was evaporated under vacuum. The specific activity of the product was determined by comparing the mass of the product (obtained from the standard curve) and the total radioactivity collected in the product peak.

[^{11}C]AM4975 was injected intravenously into an adult female baboon (*papio anubis)*. The baboon was anaesthetized with ketamine and isoflurane prior to examination and was put on a respirator for the length of the scans. Right before the injection of the radiotracer, a transmission scan of the baboon's brain was performed.

Results: The overall synthesis time was 80 min including a 20 min cyclotron irradiation. Beam time for the baboon runs was always 20 min and yielded into an average activity of 2.8 mCi of final product in MIV. The average total mass was around 9.9 nmol and the specific activity at EOB was 6.2 mCi. The average radiochemical yield was 40.7%. [^{11}C]AM4975 shows a fast uptake into the brain. Several brain structures show a high uptake of AM4975. Only 2.04% of the labelled compound stayed unbound from plasma proteins. Besides the normal baboon study, a blocking study using AM281 injected 5 min prior to injection of the radiotracer was performed. Figure 1 shows an overview with the different axis. Figure 2 shows the time/activity curve for [^{11}C]AM4975.

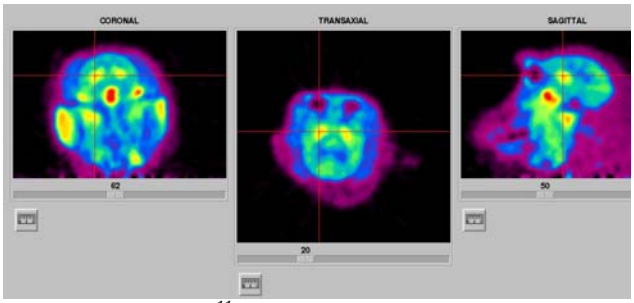

Fig. 1: PET scan of [^{11}C]AM4975 in a female baboon showing specific uptake.

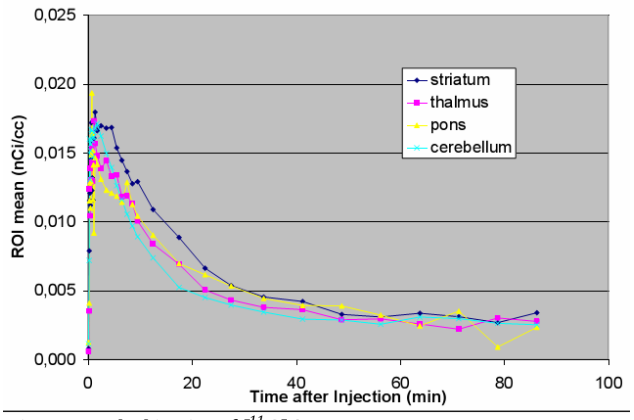

Fig. 2: Uptake kinetics of [^{11}C]AM4975.

Conclusions: AM4975 is easily methylated according to the developed procedure. Although more than half of the activity stuck to the Millipore in the beginning, this problem could be solved by using some ethanol in the end to rinse the Millipore. The yield was always enough to perform a baboon study. PET images show a rapid uptake and clearance from the baboon brain including the cerebellum, pons, putamen and the thalamus.

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