Entwicklung neuer Übergangsmetall-vermittelter Radiofluorierungsmethoden zur Herstellung von PET-Tracern

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Kurzzusammenfassung

Der Stellenwert der Positronen Emissions Tomographie (PET) in der klinischen Diagnostik hat in den letzten Jahrzehnten stetig zugenommen. Dies ist vor allem darauf zurückzuführen, dass sie in der Lage ist, biochemische Prozesse *in vivo* auf molekularer Ebene und in Echtzeit darstellen und anschließend quantitativ bewerten zu können. Um allerdings das Potential dieser Technik vollständig ausschöpfen zu können, ist die Bereitstellung eines gro0en Spektrums an PET-Tracern erforderlich. Eine Vielzahl potentieller PET-Sonden wurde identifiziert und beschrieben, die sich für die molekulare Bildgebung eignen. Dazu gehört eine nicht unerhebliche Zahl an aromatischen Aminosäuren und Aminen, wie beispielsweise 6-[^{18}F]Fluor-L-3,4-dihydroxyphenylalanin (6-[^{18}F]FDOPA) oder 6-[^{18}F]Fluordopamin (6-[^{18}F]FDA). Da ihre Herstellung aber schwierig oder teilweise gar nicht möglich ist, müssen Anstrengungen unternommen werden, um Radiofluorierungsverfahren zu entwickeln, die ihre einfache Herstellung ermöglichen. Die bisher bekannten ^{18}F-Markierungsmethoden, die über Isotopenaustausch oder elektrophile Substitution ablaufen, ermöglichten zwar prinzipiell die Synthese dieser radiofluorierten Aminosäuren, jedoch lediglich in geringen radiochemischen Ausbeuten und mit hohem Trägergehalt. Deshalb rückte in den letzten Jahren vor allem die Entwicklung neuer Methoden zur trägerarmen Synthese von radiofluorierten, elektronenreichen Aromaten, die auf nucleophilen aromatischen Substitutionsreaktionen beruhen, in den Fokus der radiochemischen Forschung. Diese sind insbesondere auch für die Radiosynthese von aromatischen Aminosäuren von besonders großem Interesse. Übergangsmetall-vermittelte Strategien erwiesen sich als besonders vielversprechend für diesen Zweck und sind daher Gegenstand dieser kumulativen Dissertation.

Die Weiterentwicklung der Ni-vermittelten Radiofluorierung ermöglichte die Herstellung von 6-[^{18}F]FDOPA, 6-[^{18}F]FDA und 6-[^{18}F]Fluor-L-*m*-tyrosin (6-[^{18}F]FMT) in geringen Ausbeuten. Das mit dieser Methode hergestellte 6-[^{18}F]FDOPA mit hoher molarer Aktivität wurde in einem Parkinson Modell der Ratte mit 6-[^{18}F]FDOPA mit geringer molarer Aktivität (elektrophil produziert) verglichen. Überraschenderweise konnten bei der Auswertung der PET-Bilder weder visuell noch in den „Zeit-Aktivitäts-Kurven“ signifikante Unterschiede zwischen den beiden Spezies gefunden werden.

Durch die Übertragung der „minimalistischen“ Methode, mit der es möglich ist, auf Phasentransferkatalysator und azeotrope Trocknung zu verzichten, auf die Cu-vermittelte Radiofluorierung von Mesitylaryliodoniumsalzen konnten erstmals elektronenreiche Aromaten in guten radiochemischen Umsätzen mit ^{18}F markiert werden. Des Weiteren wurden die klinisch

relevanten Tracer 6-[^{18}F]Fluordopamin, 4-[^{18}F]Fluorphenylalanin (4-[^{18}F]FPhe) und [^{18}F]DAA1106 in guten radiochemischen Ausbeuten hergestellt.

Außerdem ist es gelungen, die „minimalistische“ Cu-vermittelte Radiofluorierung mit Hilfe der Modellverbindung 4-[^{18}F]Fluoranisol auf ein kommerzielles Synthesemodul zu übertragen. Die daraus abgeleitete Methode eignete sich auch zur routinemäßigen Herstellung von 4-[^{18}F]FPhe und [^{18}F]DAA1106, was eine der Hauptvoraussetzungen für die Anwendung der PET-Tracer in der klinischen Praxis darstellte. Dadurch konnte auch in ersten präklinischen *in vivo* Untersuchungen das Potential von [^{18}F]DAA1106 zur Visualisierung von neuroinflammatorischen Prozessen nachgewiesen werden.

Ein weiteres wichtiges Ergebnis dieser Arbeit war der Nachweis, dass durch die Zugabe von Alkoholen die radiochemischen Ausbeuten der Cu-vermittelten Radiofluorierung von Arylboronsäuren, Arylboronsäurepinacolestern und Arylstannanen deutlich erhöht werden konnten. So konnte ein breites Spektrum an aromatischen Substraten mit unterschiedlichen elektronischen Eigenschaften mit Hilfe dieser Methode effizient radiofluoriert werden. Darunter befanden sich auch ungeschützte Indole, Phenole und Aniline, die auf anderem Wege gar nicht oder nur sehr schwer mit ^{18}F markierbar sind. Darüber hinaus konnte die Leistungsfähigkeit dieser Methode durch die Produktion klinisch relevanter Verbindungen wie [^{18}F]DAA1106, 6-[^{18}F]FDA und 6-[^{18}F]FDOPA eindrucksvoll demonstriert werden.

Abstract

In the last decades, positron emission tomography (PET) has become an important tool in clinical diagnostics, especially due to its unique ability to visualize biochemical processes *in vivo* at the molecular level in real time. A plethora of potential, innovative molecular probes have been identified and described, including a not inconsiderable number of aromatic amino acids and amines such as 6-[^{18}F]fluoro-L-3,4-dihydroxyphenylalanine (6-[^{18}F]FDOPA) and 6-[^{18}F]fluorodopamine (6-[^{18}F]FDA). Current methods for ^{18}F-labeling of aromatic compounds often suffer from several disadvantages like low radiochemical yields and high carrier content. Recently, the development of efficient methods for the synthesis of no carrier added (n.c.a.), electron-rich aromatics have been shifted into the focus of radiochemical research. Transition metal mediated strategies have proved to be very promising for this purpose and are therefore the main subject of this cumulative dissertation.

Further developments in Ni-mediated radiofluorination enabled the synthesis of n.c.a. 6-[^{18}F]FDOPA, 6-[^{18}F]FDA and 6-[^{18}F]fluor-L-*m*-tyrosine (6-[^{18}F]FMT). Despite low yields, n.c.a. 6-[^{18}F]FDOPA was evaluated in a preclinical model of Parkonson's disease. It was used to compare its biodistribution with that of carrier-added (c.a.) 6-[^{18}F]FDOPA. Surprisingly, biodistribution and time activity curves of n.c.a and c.a. 6-[^{18}F]FDOPA were very similar leading to the conclusion that the carrier-amount of 6-[^{18}F]FDOPA does not significantly affect imaging properties of the probe.

Transferring the "minimalistic" protocol, which obviates use of transfer catalyst and azeotropic drying, to the Cu-mediated radiofluorination of (mesityl)(aryl)iodonium salts allowed to prepare ^{18}F-labeled electron-rich aromatics in high radiochemical conversions. In addition, the preparation of the clinically relevant tracers 6-[^{18}F]FDA, 4-[^{18}F]fluorophenylalanine (4-[^{18}F]FPhe) and [^{18}F]DAA1106 was accomplished in good radiochemical yields. The successful transfer of the "minimalistic" Cu-mediated radiofluorination to a commercial syntheses modul demonstrates the suitability of this method for the routine production of PET tracers such as 4-[^{18}F]FPhe and [^{18}F]DAA1106. In addition the potential of [^{18}F]DAA1106 for the visualization of neuroinflammation was demonstrated in first *in vivo* experiments.

In another series of experiments, the beneficial effect of alcohols on Cu-mediated radiofluorination of arylboronic acids, arylboronic acid pinacol esters and arylstannanes was shown. A wide range of substrates could be radiofluorinated, regardless of their electronic properties. Impressivley, ^{18}F-labeled indoles, phenols and anilines were dircetly prepared from their corresponding unprotected precursors. These approach allowed additionally the

production of clinically relevant tracers like [^{18}F]DAA1106, 6-[^{18}F]FDA and 6-[^{18}F]FDOPA demonstrating the versatility of this novel radiolabeling protocol.

Inhaltsverzeichnis

1 Einleitung

Molekulare Bildgebungsverfahren sind aus der modernen Diagnostik nicht mehr wegzudenken, da sie es ermöglichen, die mit verschiedensten Pathologien assoziierten biochemischen Prozesse auf molekularer Ebene zu visualisieren. Unter den zur Verfügung stehenden Techniken ist die Positronen-Emissions-Tomographie das mit Abstand wichtigste Verfahren. Während klassische Bildgebungsverfahren wie die Computer-Tomographie (CT) oder die Magnet-Resonanz-Tomographie (MRT) lediglich morphologische Veränderungen, deren Ursprung häufig in molekularen Prozessen liegt, darstellen können, ist die Positronen-Emissions-Tomographie (PET) in der Lage, genau diese biochemischen Prozesse auf molekularer Ebene in Echtzeit zu erfassen.

Seit ihrer ersten klinischen Anwendung vor rund 60 Jahren hat sich die PET stark weiterentwickelt. Heute steht eine Vielzahl von Tracern für unterschiedlichste Fragestellungen für verschiedene Fachgebiete der Medizin zur Verfügung.[1] Am weitesten verbreitet ist ihre Anwendung in der Onkologie, in der sie zu Diagnostik von Tumoren, sowie zur Therapieüberwachung eingesetzt wird.[2] Darüber hinaus wird sie auch zunehmend in der Neurologie zur Differenzialdiagnostik von neurodegenerativen Erkrankungen eingesetzt.[3] Trotz dieser enormen Fortschritte konnte das diagnostische Potential vieler ^{18}F-markierter Verbindungen bisher noch nicht vollständig ausgeschöpft werden, da es an geeigneten Syntheseverfahren mangelt. Besonders die Radiofluorierung von elektronenreichen aromatischen Systemen erwies sich in der Vergangenheit als äußerst herausfordernd. Bis vor wenigen Jahren wurden derartige Verbindungen, wie beispielsweise 6-[^{18}F]Fluor-L-3,4-dihydroxyphenylalanin (6-[^{18}F]FDOPA), routinemäßig über elektrophile Synthesewege hergestellt.[4] Aufgrund der geringen Targetausbeuten bei der Herstellung von [^{18}F]F_2, lassen sich Tracer auf diesem Weg nur in geringen Aktivitäten synthetisieren. Darüber hinaus führt der Einsatz von [^{19}F]F_2 dazu, dass nur niedrige molare Aktivitäten erreicht werden können.

Vor diesem Hintergrund ist es ein zentrale Herausforderung der radiochemischen Forschung, effiziente Syntheseverfahren zur Radiofluorierung von aromatischen Verbindungen zu entwickeln. Insbesondere Übergangsmetall-vermittelte Strategien, die erst vor kurzem für die organisch präparative Chemie entwickelt wurden, stehen im Fokus aktueller Untersuchungen. Sie haben das Potential, eine Vielzahl der früher schlecht zugänglichen bzw. unzugänglichen

PET-Tracer für medizinische Anwendungen verfügbar zu machen. Daher standen diese Methoden auch im Fokus der hier vorliegenden Arbeit

1.1 Grundlagen der Positronen-Emissions-Tomographie (PET)

Die PET ist ein minimal-invasives Verfahren zur *in vivo* Bildgebung von biochemischen Prozessen auf molekularer Ebene. Hierzu wird das räumliche und zeitliche Verteilungsmusters eines, mit einem β^+-Strahler markierten Radiopharmakons (Tracer) durch Detektion der, aus der Positronenannihilation resultierenden, antiparallenen γ-Photonen, gemessen. Mit Hilfe dieser Daten können dann Schnittbilder mit der Radionuklidverteilung des untersuchten Körpers rekonstruiert werden.[5]

Ausgangspunkt dieses diagnostischen Verfahrens ist ein mit einem β^+-Emitter markiertes Radiopharmakon, welches dem Patienten appliziert wird. Anschließend verteilt es sich im Körper und reichert sich durch spezifische Wechselwirkungen an seinem molekularen Target an. Solche Targets lassen sich in Rezeptoren, metabolische Prozesse oder Siganltransduktionswege klassifizieren. Durch pathophysiologische Prozesse werden sie über- oder unterexprimiert, was sich mit PET Sonden sensitiv erfassen und visualisieren lässt.

Die signalgebende Einheit wird durch den β^+-Emitter dargestellt. Das Radionuklid unterliegt hierbei einem spontanen β^+-Zerfall, wobei sich ein Proton (p) in ein Neutron (n) umwandelt. Des Weiteren wird ein Positron (β^+) und ein Neutrino (ν_e) ausgesendet.

$$ {}^{1}_{1}p = {}^{1}_{0}n + e^+ + \nu_e \qquad \text{Gl. 1.0} $$

Das so entstandene Positron wird im menschlichen Gewebe abgebremst und legt hierbei eine Wegstrecke von ca. 1–2 mm zurück bis es sich mit seinem Antiteilchen, einem Elektron, verbindet..[6] Es kommt zur Annihilation beider Teilchen, wobei durch die Umwandlung der Masse der beiden Teilchen zwei hochenergetische Gammaphotonen (γ-Quanten) in einem Winkel von 180° ausgesandt werden. Ihre Energie von jeweils 511 keV entspricht der Ruheenergie von Elektron und Positron.[7].

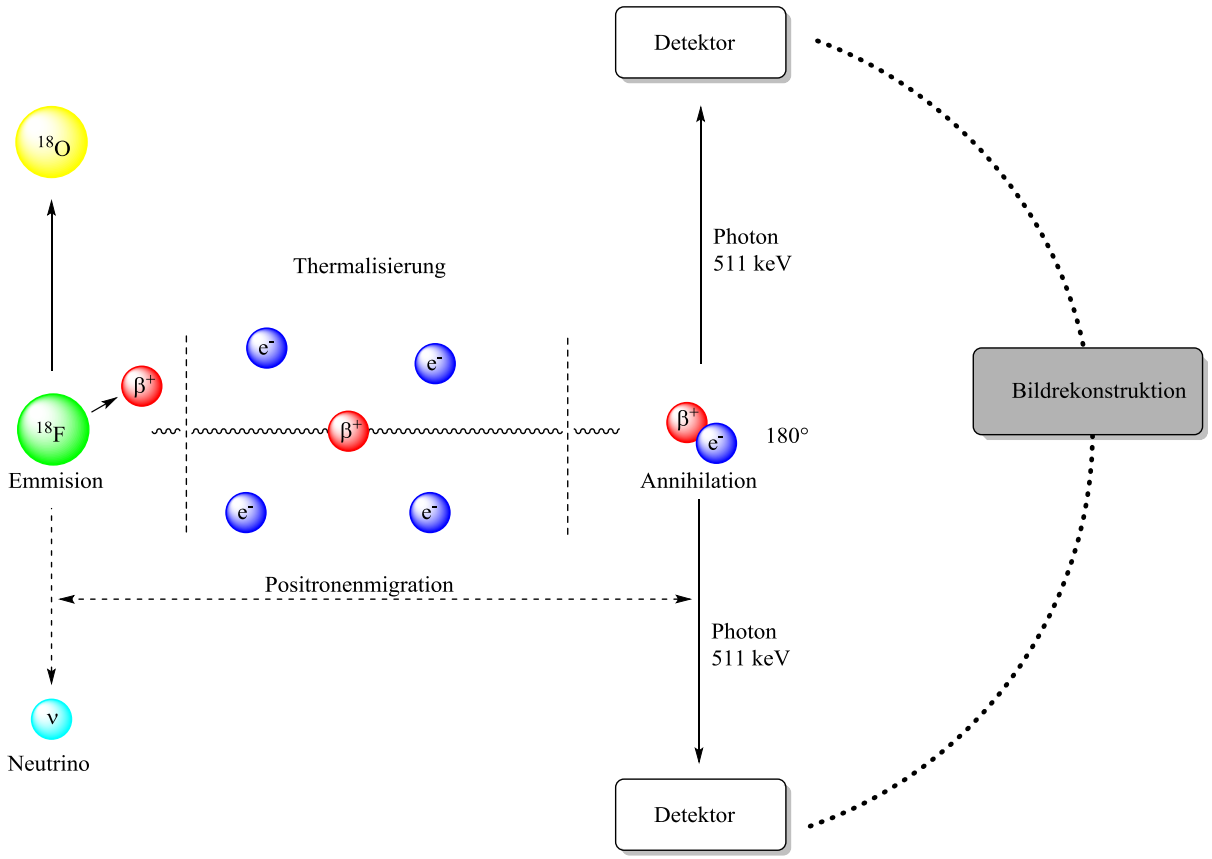

Abbildung 1.0: Schematische Darstellung der Funktionsweise der Positronen-Emissions-Tomographie.

Da γ-Quanten eine sehr geringe Wechselwirkungswahrscheinlichkeit mit dem umliegenden Gewebe haben, verlassen sie den Körper nahezu ungehindert. Zu ihrer Erfassung besitzt ein Positronen-Emissions-Tomograph eine große Anzahl von Detektoren, die ringförmig um das zu untersuchende Objekt angebracht sind. Um ausschließlich γ-Quanten, die von einem β^+-Zerfall stammen, detektieren zu können, ist eine Koinzidenz-Schaltung notwendig. Nur Zerfallsereignisse, die von jedem der zwei gegenüberliegenden Detektoren innerhalb eines Koinzidenzintervalls durch die eintreffenden γ-Quanten gleichzeitig detektiert wurden, werden registriert. Mittels Ausleseelektronik und Bildbearbeitungssoftware wird aus diesen Daten ein Bild rekonstruiert, das die räumliche Verteilung des Radiotracers wiedergibt.[1, 5a]

1.2 ^{18}F als geeignetes PET-Nuklid

Die Eignung eines Nuklids für die PET ist an einige Bedingungen geknüpft. Neben physikalischen Größen wie Halbwertzeit und Positronenenergie, sind auch die Strahlenbelastung für den Patienten oder die Verfügbarkeit der Radionuklide zu berücksichtigen.[6a] So setzt beispielsweise die Verwendung der in der Nuklearmedizin gängigen Nuklide ^{11}C, ^{13}N und ^{15}O auf Grund ihrer geringen Halbwertszeiten (siehe Tabelle

1.1) ein hauseigenes Zyklotron voraus. Bedingt durch seine längere Halbwertzeit von 109,7 min ist im Falle von ^{18}F die, zur Kostenreduktion führende, Radiopharmaka-Versorgung mehrerer klinischer Einrichtungen durch eine Produktionsstätte möglich.[8]

Tabelle 1.1: Radionuklide für die PET.[9]

Radionuklid	Halbwertszeit $t_{1/2}$ [min]	$E\beta^+max$ [MeV]	Kernreaktion	Targetfüllung
^{11}C	20,4	0,96	$^{14}N(p,\alpha)^{11}C$	$N_2(O_2)$
^{13}N	10,0	1,2	$^{16}O(p,\alpha)^{13}N$	$H_2{}^{16}O$
^{15}O	2,0	1,7	$^{14}N(d,n)^{15}O$ $^{15}N(p,n)^{15}O$	$N_2(O_2)$ $^{15}N_2(O_2)$
^{18}F	109,6	0,64	$^{18}O(p,n)^{18}F$ $^{20}Ne(d,\alpha)^{18}F$	$H_2{}^{18}O$ $Ne(F_2)$

Darüber hinaus ermöglicht die Halbwertzeit von ^{18}F auch die Durchführung zeitaufwendiger Synthesen komplexer Moleküle.[10]

In der pharmazeutischen Chemie finden Fluor und fluorierte Substituenten bereits länger breite Anwendung. Ein bekannter Effekt der Einführung von Fluor oder fluorierten Substituenten ist die Beeinflussung der metabolischen Stabilität.[11] So ist bekannt, dass durch die Substitution eines Wasserstoffs an einem Aromaten durch Fluor die oxidative Verstoffwechselung häufig deutlich verlangsamt wird. Darüber hinaus lässt sich auch die Lipophilie einer Verbindung beeinflussen, was einen modulierenden Einfluss auf die Verteilung des Wirkstoffs und seine Aufnahme ins Gewebe haben kann. Fluorsubstituenten können auch, durch Veränderung der Basizität bzw. Azidität, Einfluss auf die Bindungsaffinität einer Verbindung zu einem bestimmten Rezeptor haben.[12]

Darüber hinaus hat Fluor einen, dem des Wasserstoffs annähernd gleichenden Van-der-Waals Radius (1,2 Å zu 1,35 Å) und die Bindungslängen einer C-F- und C-OH-Bindungen stimmen nahezu überein.[13] Aus diesen Gründen eignet sich ^{18}F sehr gut für die Synthese sogenannter „Analogtracer“. Hinter diesem Begriff verbirgt sich das Konzept, köpereigenen Stoffen sterisch und chemisch ähnliche Tracer zu entwickeln, die, trotz Einführung von Fremdelementen, wie zum Beispiel ^{18}F, ein analoges Bindungsverhalten und ähnliche pharmakologische Eigenschaften aufweisen.[14] Darüber hinaus sind mit ^{18}F, im Vergleich zu ^{11}C, deutlich höhere molare Aktivitäten erreichbar, was insbesondere bei rezeptorspezifischen Tracern von großer Bedeutung ist.

Eine Vielzahl von Kernreaktionen zur Gewinnung von ^{18}F sind bereits beschrieben worden (Tabelle 1.2).

Tabelle 1.2: Kernreaktionen zur Produktion von ^{18}F

Kernreaktion	Target	Chem. Form des ^{18}F	Mol. Aktivität [Ci/mmol]
$^{18}O(p,n)^{18}F$	$H_2{}^{18}O$	$^{18}F^-_{(aq)}$	10^5
$^{16}O(^3He,p)^{18}F$	H_2O	$^{18}F^-_{(aq)}$	10^5
$^{20}Ne(d,\alpha)^{18}F$	$Ne(0{,}1\%\ F_2)$	$[^{18}F]F_2$	1–10
$^{18}O(p,n)^{18}F$	$^{18}O_2(0{,}1\%\ F_2)$	$[^{18}F]F_2$	1–50

Für die routinemäßige Herstellung von ^{18}F sind vor allem zwei Kernreaktionen von großer Bedeutung. $[^{18}F]F_2$ wird über die Kernreaktion $^{20}Ne(d,\alpha)^{18}F$ gewonnen. Bei diesem Verfahren ist es allerdings unumgänglich, dem Targetgas 0,1% nicht radioaktives F_2 zuzufügen, wodurch das hergestellte ^{18}F mit dem natürlichen Isotop ^{19}F geträgert (carrier-added. c.a.) wird. Mittlerweile wird nicht geträgertes (non-carrier-added, n.c.a.) $^{18}F^-$ fast ausschließlich über die Kernreaktion $^{18}O(p,n)^{18}F$ hergestellt. Bedingt durch den hohen Wirkungsquerschnitt dieser Kernreaktion sind sowohl hohe Targetausbeuten als auch hohe molare Aktivitäten erreichbar.[15]

1.3 Strategien zur ^{18}F-Markierung

Es gibt verschiedene Möglichkeiten, ^{18}F in Moleküle einzuführen. Man unterscheidet dabei zwei unterschiedliche Synthesestrategien für die ^{18}F-Markierung organischer Verbindungen:[16]

- Indirekte Fluorierungen über prosthetische Gruppen ermöglichen den Zugang zu, mit ^{18}F markierten, komplexen und empfindlichen Biomolekülen, indem im ersten Syntheseschritt ^{18}F-markierte Bausteine dargestellt werden, welche daraufhin über funktionelle Gruppen an das Zielmolekül gekoppelt werden.
- Direkte Fluorierungsverfahren lassen sich ihrerseits nochmals in nucleophile und elektrophile Fluorierungen unterteilen. Beiden gemein ist, dass ^{18}F in einem Schritt direkt in das Zielmolekül eingeführt wird. Da jedoch relativ harsche Reaktionsbedingungen erforderlich sind, eignet sich diese Methode nicht für die Markierung von empfindlichen Biomolekülen.

In den Anfängen der Entwicklung von ^{18}F-markierten Tracern für die PET spielten elektrophile Synthesestrategien eine wichtige Rolle. Viele der heute etablierten Radiopharmaka wurden zunächst über eine elektrophile Syntheseroute hergestellt. Dennoch haben sie heute, vor allem aufgrund der geringen Ausbeuten und einer häufig auftretenden großen Zahl an Nebenprodukten, an Bedeutung verloren. Theoretisch sind mit dieser Methode radiochemische Ausbeuten von maximal 50% möglich, da bei der Herstellung von [^{18}F]F_2, wie bereits zuvor beschrieben, ^{19}F hinzugefügt werden muss und aus statistischen Gründen daher ^{18}F mit ^{19}F konkurriert. Dadurch kann statistisch nur maximal 50% des Zielmoleküls radiofluoriert werden. Zudem wird durch den Zusatz von Fluorgas Träger eingeführt, so dass nur sehr geringe molare Aktivitäten erreicht werden können.[17]

Daher, und auf Grund der höheren Startaktivität, sind heute nucleophile aromatische und aliphatische Substitutionsreaktionen die Methode der Wahl. Hierzu wird über die in Abschnitt 1.2 beschriebene Kernreaktion n.c.a. [^{18}F]Fluorid aus [^{18}O]H_2O hergestellt. Das vom Zyklotron in wässriger Lösung erhaltene [^{18}F]Fluorid ist unter diesen Bedingungen stark solvatisiert, wodurch seine Nucleophilie stark verringert ist. Zur Steigerung der Reaktivität ist es daher erforderlich, Wasser zu entfernen, was üblicherweise durch folgendes Verfahren erreicht wird:

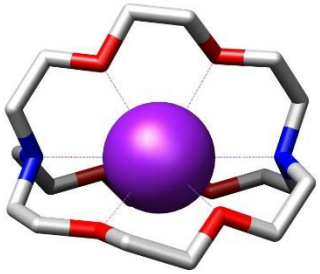

Abbildung 1.1 Komplexierung eines Kaliumkations durch Kryptofix-2.2.2[19b]

Zunächst wird $^{18}F^-$ auf einem Anionentauscher fixiert und anschließend mit einer organisch-wässrigen (MeCN/H_2O, 50:50 v/v) Lösung mit K_2CO_3 eluiert. Da die Kalium-Fluor-Bindung die Reaktivität von $^{18}F^-$ immer noch verringert, gibt man Kryptanden wie Kryptofix-222 zu, die starke Komplexe mit dem Kaliumkation bilden. Nach Entfernung des Wassers durch mehrfache azeotrope Trocknung liegt ^{18}F in aprotisch polaren Lösungsmitteln als frei zugängliches, hoch nucleophiles Fluoridion vor.[18] Alternativ zu K_2CO_3 wird auch Tetrabutylammoniumhydrogencarbonat zur Elution verwendet, wodurch sich *in situ* [^{18}F]Tetrabutylammoniumfluorid bildet. Auf diese Weise kann auf den Einsatz von Kryptanden verzichtet werden.[19]

1.4 Radiofluorierung ohne azeotrope Trocknung und Zusatz von Kryptanden („minimalistischer“ Ansatz)

In der Vergangenheit wurde eine Vielzahl von Methoden zur Aktivierung von [^{18}F]Fluorid entwickelt, die auf die aufwendige Trocknungsprozedur verzichten können. Allerdings gehen diese alle mit mehr oder weniger großen Problemen einher.

Lemaire *et al.* setzten zu diesem Zweck Phosphazen-Superbasen, wie P_2Et, in einem Gemisch aus Acetonitril und Wasser ein.[20] Dies ermöglicht eine quantitative Elution von [^{18}F]Fluorid von der Anionenaustauschersäule. Anschließend wird das Eluat mit einem geeigneten Vorläufer, unter Zugabe von 2-*tert*-Butyl-1,1,3,3-tetramethylguanidin (BTMG), zu den entsprechenden radiofluorierten Verbindungen umgesetzt. Allerdings ist die Substratauswahl für dieses Verfahren relativ begrenzt, da der Großteil der, in der Radiochemie eingesetzten, Markierungsvorläufer unter diesen extrem basischen Reaktionsbedingungen nicht stabil ist. Des Weiteren ist die für diese Methode erforderliche, hohe Menge an toxischen Verbindungen wie P_2Et und BTMG als sehr problematisch für die weitere Verwendung als Radiopharmakon anzusehen.

Ähnlich beschränkt in der Wahl der Vorläufer ist die kryptandfreie Radiofluorierung von Diaryliodoniumtosylaten, wie sie von Chun *et al.* beschrieben wurde.[21] Die Verwendung eines wässrig-organischen Reaktionsmediums mit einem Wasseranteil von bis zu 28% ermöglicht die Synthese der ^{18}F-markierten Verbindungen direkt aus bestrahltem [^{18}O]Wasser. Allerdings ist diese Methode ausschließlich mit sehr wenigen, stark aktivierten Vorläufern durchführbar. Des Weiteren ermöglicht der geringe Wasseranteil, der für die Reaktion gerade noch akzeptabel ist, nur die Synthese von sehr geringen Aktivitätsmengen.

Andere Ansätze, die es ermöglichen, auf die azeotrope Trocknung zu verzichten, weisen meist die Problematik auf, hohe Vorläufermengen für akzeptable radiochemische Ausbeuten zu benötigen.[22]

Einen anderen Ansatz verfolgten Richarz, *et al*. Sie konnten zeigen, dass sich auf der Anionenaustauschersäule fixiertes $^{18}F^-$ mit, in MeOH oder EtOH gelösten Onium-Salzen nahezu quantitativ eluieren lässt.[23] Durch den niedrigen Siedepunkt kann MeOH innerhalb von 2–3 min vollständig entfernt werden. Daraufhin wird ein geeignetes, aprotisches Lösungsmittel zugegeben und das Reaktionsgemisch erhitzt. Durch Verzicht auf eine azeotrope Trocknung ergibt sich eine Zeitersparnis von 20–25 min. Auch die Adsorption von $^{18}F^-$ auf den Gefäßwänden wird durch diesen Prozess erheblich verringert. Außerdem konnte gezeigt werden, dass bereits eine Vorläufermenge von 0,1 mg ausreicht, um über 85% der Aktivität von der Ionenaustauschersäule zu eluieren. Dies stellt, verglichen mit den bisher für die Elution benötigten großen Vorläufermengen, eine deutliche Verbesserung dar.

1.5 Metallkatalysierte Radiofluorierungsstrategien

Metallkatalysierte Reaktionen wurden in der Radiochemie zunächst für die Synthese von radiomarkierten prosthetischen Gruppen verwendet. So beschrieb Marrière *et al.* die Synthese

eines radiomarkierten (-)-Cytisin-Analogs.[24] Stannylcytisin wurde über eine STILLE-Kupplung mit 4-[^{18}F]Fluorbrombenzaldehyd zum gewünschten Produkt umgesetzt. Wüst *et al.* beschrieben, ausgehend von 4-[^{18}F]Fluoriodbenzol, die ebenfalls über eine STILLE-Kupplung ablaufende Synthese von 5-(4´-[^{18}F]Fluorphenyl)-uridin[25] sowie von zwei potentiellen Cyclooxygenase-2 (COX-2) Inhibitoren.[26] Unter Verwendung des Markierungsbausteins 4-[^{18}F]Fluoriodbenzol gelang des Weiteren die Übertragung der SONOGASHIRA-Kreuzkupplung in die Radiochemie.[27] Steiniger *et al.* nutzte diese prosthetische Gruppe zur Synthese von ^{18}F-markierten Biphenylen über die SUZUKI-Kreuzkupplung.[28] Die Verwendung von [^{18}F]Fluoriodbenzol als Markierungsbaustein für die Kreuzkupplung bringt jedoch einige Nachteile mit sich. Bei der Synthese, ausgehend von Diioddiphenyliodonium triflat, tritt stets als Nebenprodukt Diiodbenzol auf. Dieses muss zunächst mittels HPLC abgetrennt werden, da es sonst in Konkurrenz zu [^{18}F]Fluoriodbenzol in den Kreuzkupplungen reagiert.

Eine erste direkte Markierung beschrieben Hollingworth *et al.* Sie setzten, unter Pd-Katalyse, [^{18}F]Tetrabutylammoniumfluorid mit einem Überschuss von Cinnamylmethylcarbonat zum entsprechenden ^{18}F-markierten Cinnamylfluorid um.[29] Jedoch ist diese Methode auf die Einführung von ^{18}F in allylische Positionen beschränkt.

1.6 Metall-vermittelte aromatische Radiofluorierung

Durch die in Abschnitt 1.4 beschriebenen Methoden und nicht zuletzt durch die Einführung der Radiofluorierung unter „minimalistischen“ Bedingungen sind eine Vielzahl von ^{18}F-markierten Verbindungen über die nucleophile aromatische Substitution (S_NAr) zugänglich. Die Einführung von $^{18}F^-$ in elektronenreiche aromatische Systeme stellte jedoch weiterhin eine große Herausforderung dar. Radiomarkierte aromatische Aminosäuren, wie 6-[^{18}F]Fluor-L-3,4-dihydroxyphenylalanin (6-[^{18}F]FDOPA) oder 6-[^{18}F]Fluor-L-*m*-tyrosin ([^{18}F]FMT), sind als Tracer für die PET von besonderem Interesse. Als elektronenreiche aromatische Verbindungen waren sie bisher allerdings nur schwer zugänglich und wurden meistens über elektrophile Radiofluorierungsverfahren hergestellt.[4b]

1.6.1 Ni-vermittelte Radiofluorierung

Lee *et al.* beschrieben erstmalig eine Übergangsmetall-vermittelte direkte ^{18}F-Markierung von elektronenreichen aromatischen Systemen. Unter Verwendung von zwei speziellen Pd-Komplexen war es möglich, elektronenreiche Aromaten zu radiofluorieren.[30] Ein, als elektrophiles Fluorierungsreagenz fungierender, Pd(IV)-Komplex bindet zunächst das Fluorid und überträgt dieses auf einen Pd(II)-aryl-Komplex. Dieser Komplex enthält bereits den zu markierenden Aromaten in Form eines Liganden. Durch die darauf folgende reduktive

Eliminierung entsteht die ^{18}F-markierte Verbindung. Aufgrund der hohen Empfindlichkeit der Pd-Komplexe gegenüber Luft und Feuchtigkeit und der schwierigen Zugänglichkeit der benötigten Pd-Komplexe ist die Synthese allerdings sehr komplex und nicht für die routinemäßige Herstellung von Radiopharmaka geeignet.

Als Weiterentwicklung der Pd-vermittelten oxidativen Fluorierung wurde von derselben Arbeitsgruppe 2012 ein Ni-vermitteltes Verfahren veröffentlicht. Bei diesem Verfahren wird der zu fluorierende Aromat an einen Ni-Komplex koordiniert und eine hypervalente Iodverbindung als Oxidationsmittel zugesetzt. Diese Methode weist gegenüber der Pd-vermittelten Synthese zwei signifikante Vorteile auf. Die Ni-vermittelte Variante ist, im Gegensatz zum zweistufigen Pd-Mechanismus, einstufig. Dadurch konnte die Reaktionszeit, die in der Radiochemie eine wichtige Rolle spielt, deutlich verkürzt werden. Des Weiteren konnte durch den Einsatz des Nickelkomplexes auf einen, der äußerst feuchtigkeitsempfindlichen Pd-Komplexe verzichtet werden. Die Rolle des Palladium(IV)komplexes nimmt hier die hypervalente Iodverbindung 1,1‘-(Phenyl-λ^3-iodandiyl)bis(4-methoxypyridinium)bis (trifluormethansulfonat) **A**, ein, die jedoch weiterhin eine Handhabung unter inerten Bedingungen voraussetzt. Sie dient als Oxidationsmittel und bildet in Anwesenheit von 18-Krone-6 die elektrophile Fluorierungsspezies [^{18}F]**B**. Von dort wird Fluor in einem Transmetallierungsschritt auf **C** übertragen und es bildet sich der oktaedrische Ni(IV)-Komplex [^{18}F]**D**. Durch die nachfolgende reduktive C-F Eliminierung wird das Markierungsprodukt erhalten (Abb. 1.2).

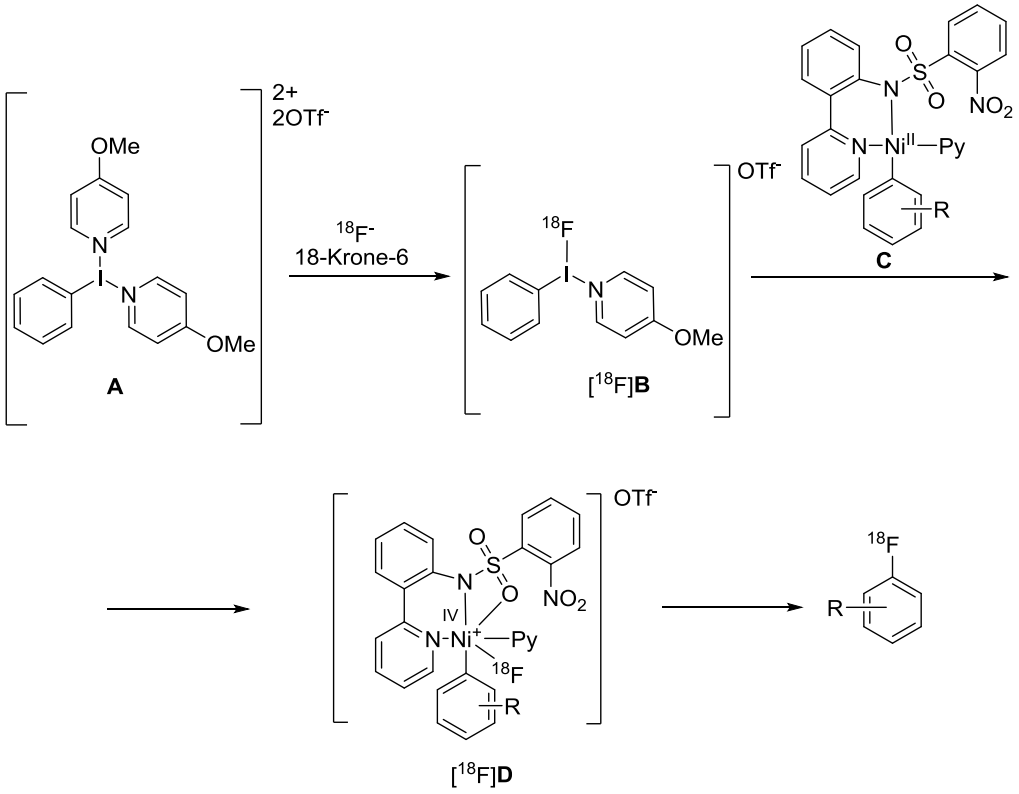

Abbildung 1.2 Ni-vermittelte Radiofluorierung von Aromaten.

Gemäß Lee *et al.* toleriert die Ni-vermittelte Radiofluorierung einen Wasseranteil von bis zu 1% und wurde direkt mit bestrahltem [^{18}O]H_2O durchgeführt werden.

Auf Grund der kleinen maximalen Anfangsaktivität, resultierend aus dem geringen tolerablen Wasseranteil, musste bei der späteren Synthese von Radiotracern für die PET wie beispielsweise [^{18}F]MDL100907 und [^{18}F]5-Fluoruracil, das Syntheseprotokoll modifiziert werden. Um größere Aktivitätsmengen einsetzen zu können, musste $^{18}F^-$ auf einem Anionentauscher fixiert, mit Kaliumcarbonat eluiert und anschließend azeotrop getrocknet werden. Die beiden klinisch relevanten Verbindungen konnten in sehr geringen radiochemischen Ausbeuten* (RCA) von 3% für [^{18}F]MDL100907 und 0,9% im Falle von [^{18}F]5-Fluoruracil erhalten werden.[31]

Ein Bestandteil der vorliegenden Arbeit war es, diese Radiofluorierungsmethode im Hinblick auf den Einsatz in der Praxis zu untersuchen. Unter diesen optimierten Reaktionsbedingungen sollten dann die klinisch relevanten Tracer 6-[^{18}F]FDOPA, 6-[^{18}F]Fluordopamin (6-[^{18}F]FDA) und 6-[^{18}F]FMT in hohen Aktivitätsdosen hergestellt werden, wie sie für eine präklinische Evaluierung und spätere klinische Anwendung notwendig sind. Darüber hinaus sollte das, über

* Radiochemische Ausbeuten beziehen sich immer auf das isolierte und radiochemisch reine Produkt. Die Aktivität wurde vor und nach der Synthese gemessen.

diese Methode hergestellte, n.c.a. 6-[^{18}F]FDOPA bezüglich seiner *in vivo* Eigenschaften für die Bildgebung untersucht und mit „elektrophilem“ c.a. 6-[^{18}F]FDOPA verglichen werden.

1.6.2 Cu-vermittelte Radiofluorierung von Mesitylaryliodonium-Salzen

Mit der Cu-vermittelten Fluorierung von Mesitylaryliodonium Tetrafluorboraten stellten Ichiishi *et al.* 2013 eine, gegenüber der Pd- und Ni-vermittelten Methodik, erheblich vereinfachte Fluorierungsmethode vor. Das Iodonium-Substrat wird in Anwesenheit von $Cu(OTf)_2$ mit KF in DMF bei 60 °C umgesetzt. Mit dieser Methode war es möglich, verschiedene Aromaten unabhängig von ihren elektronischen Eigenschaften zu fluorieren. Dabei wurden Ausbeuten von bis zu 85% erreicht.[32]

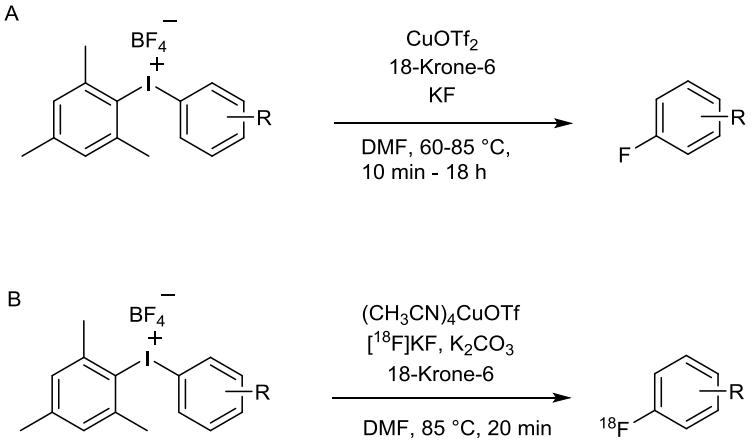

Abbildung 1.3 Cu-vermittelte Fluorierung (A) und Radiofluorierung (B) von (Aryl)(mesityl)liodonium tetrafluorborat.

Bestandteil der vorliegenden Arbeit war der Transfer dieses Verfahrens in die Radiochemie und dessen Optimierung für die Herstellung von Radiotracern für die PET. Während dieser Untersuchungen wurde von den oben genannten Autoren die Übertragung der Fluorierungsmethode in die Radiochemie veröffentlicht. Anstelle des, für die organisch präparativen Synthesen genutzten, gegenüber Wasser sensitiven, $Cu(OTf)_2$, wurde von Ichiishi *et al*. für diese Art von Radiosynthesen das weitaus weniger empfindliches $(CH_3CN)_4CuOTf$ als Cu-Quelle verwendet.[33] Den Autoren gelang es so, eine kleine Auswahl an Vorläufern in moderaten bis guten radiochemischen Umsätzen† (RCU) von 14–79% mit ^{18}F zu markieren.

Auch wenn diese Veröffentlichung mitten in den laufenden Arbeiten zur Übertragung der organisch-präparativen Synthese in die Radiochemie erschien, blieb die anfängliche Aufgabenstellung einer einfachen ^{18}F-Markierung von elektronenreichen Aromaten in hohen Dosen, bestehen. Wie bei der „minimalistischen“ Radiofluorierung wird bei der Cu-vermittelte

† Radiochemische Umsatz bezieht sich auf den Anteil an $^{18}F^-$, das in das ^{18}F-markierte Produkt überführt und analytisch mittels HPLC oder DC aus dem Reaktionsgemisch bestimmt wurde.

Radiofluorierung von (Aryl)(mesityl)iodoniumsalzen ein Oniumsalz-Vorläufer verwendet, der sich potentiell zu direkten Elution von $^{18}F^-$ eignet. Daher sollte das „minimalistische“ Protokoll auf die Cu-vermittelte Radiofluorierung übertragen werden um, unter Verzicht auf K_2CO_3, 18-Krone-6 und eine azeotrope Trocknung, PET-Tracer in hohen Aktivitätsdosen zu erhalten.

1.6.3 Cu-vermittelte Radiofluorierung von Phenylboronsäuren, Phenylboronsäurepinacolestern und Phenylstannanen

Teare *et al.* beschrieben 2010 eine Ag-vermittelte Radiofluorierung von Arylstannanen mittels [^{18}F]Selectfluor.[34] Bei Raumtemperatur konnten innerhalb von 20 min radiochemische Umsätze von bis zu 89% erreicht werden. 2013 gelang Stenhagen *et al.* unter ähnlichen Reaktionsbedingungen die ^{18}F-Markierung von Arylboronsäureestern.[35] Im Gegensatz zu den Stannanen konnten diese nicht direkt radiofluoriert werden, sondern mussten zunächst in eine Ag(I)-Verbindung überführt werden. Anschließend wurde dieser Komplex erfolgreich mit [^{18}F]Selectfluor umgesetzt. Bei Raumtemperatur und einer Reaktionszeit von 20 min für den Radiomarkierungsschritt ließen sich radiochemische Umsätze von bis zu 50% erreichen. Vergleicht man die Synthese von 6-[^{18}F]FDOPA ausgehend von dem Boronsäureester-Vorläufer mit der Synthese über den Stannan-Vorläufer, so wurde durch Erstere, trotz einer deutlich aufwendigeren Synthese, 6-[^{18}F]FDOPA in etwas höheren radiochemischen Ausbeuten (19% gegenüber 12%) isoliert. In beiden Fällen konnten allerdings nur geringe molare Aktivitäten erreicht werden (2,6 und 3,4 GBq/µmol).

Unter Cu-vermittelten Reaktionsbedingungen gelang es Tredwell *et al.* 2014 erstmalig, Arylboronsäurepinacolester, mit $^{18}F^-$ zu markieren.[36] In ersten Experimenten diente $Cu(OTf)_2$ als Kupferquelle. Im Verlauf weiterer Untersuchungen zeigte sich der positive Effekt von Pyridin als Co-Ligand auf die Produktbildung. Dementsprechend wurde im weiteren Verlauf $[Cu(OTf)_2(Py)_4]$ als Kupferquelle verwendet. Durch Verwendung dieses Komplexes konnten die radiochemischen Umsätze deutlich gesteigert werden. Ferner zeigte sich, dass dieser Komplex im Gegensatz zu $Cu(OTf)_2$ unter Raumluft stabil ist und somit keine aufwendige Handhabung unter Schutzgasatmosphäre erfordert.

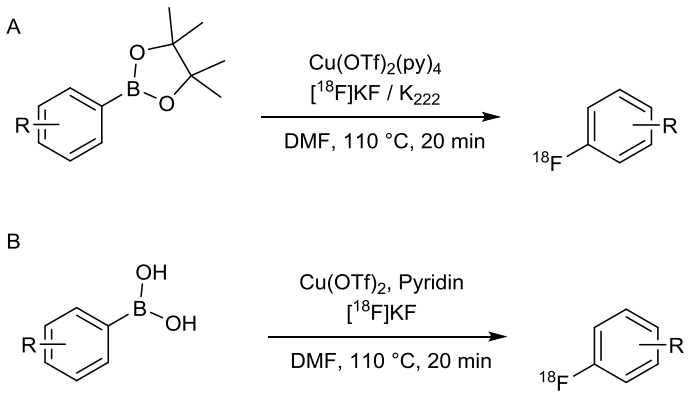

Abbildung 1.4 Cu-vermittelte Radiofluorierung von Arylboronsäuren und –boronsäurepinakolestern.

Darüber hinaus konnten die Autoren zeigen, dass für eine erfolgreiche Reaktionsführung die Anwesenheit von Luftsauerstoff zwingend erforderlich ist. Unter optimierten Bedingungen konnte eine Reihe von Modellverbindungen in radiochemischen Umsätzen von 5–74% synthetisiert werden. Auf Grund der trägerarmen Bedingungen wurden molare Aktivitäten von 112 GBq/µmol erreicht.

In einer Fortentwicklung beschrieben 2015 Mossine *et al.* die Cu-vermittelte Radiofluorierung von Arylboronsäuren, die, im Vergleich zu Boronsäurepinacolestern, einfacher zugänglich sind.[37] Unter optimierten Reaktionsbedingungen konnte eine Auswahl von aromatischen Modellverbindungen mit elektronenziehenden, -neutralen und –schiebende Substituenten in moderaten bis guten radiochemischen Umsätzen von 12–73% mit ^{18}F markiert werden. Als klinisch relevantes Anwendungsbeispiel gelang es des Weiteren [^{18}F]FPEB, einen Radiotracer zur Visualisierung von metabotropischen Glutamatrezeptoren vom Subtyp 5,[38] zu synthetisieren. In manuellen Synthesen konnte die Zielverbindung in radiochemischen Umsätzen von bis zu 8%, in automatisierten von bis zu 4% synthetisiert werden (isolierte radiochemische Ausbeuten wurden nicht veröffentlicht). Durch die Verwendung von $Cu(OTf)_2$ an Stelle des Pyridin-Komplexes ergeben sich hier allerdings wieder, die oben beschriebenen, Probleme in der Handhabung. Ferner zeigten die Autoren, dass die Zugabe von Pyridin essentiell für die erfolgreiche Radiomarkierung mittels $Cu(OTf)_2$ ist.

2016 beschrieben Gamache *et al.* die nucleophile Fluorierung von Arylstannanen, ausgehend von Triphenyldifluorsilikat.[39] Arylstannane sind in der Radiochemie bereits lange als Vorläufer etabliert und wurden bisher beispielsweise für die elektrophile ^{18}F-Markierung oder Radioiodierung verwendet.[4b, 40] Daher sind Stannan-Vorläufer für eine Vielzahl von Tracern kommerziell erhältlich. Die Autoren zeigten, dass sich Arylstannane bei Raumtemperatur und in Anwesenheit von $Cu(OTf)_2$ innerhalb von 3 h in Ausbeuten von bis zu 76% fluorieren lassen. Mechanistisch leiten sie die Reaktion von dem, von Ye *et al.* 2013, vorgeschlagenen

Mechanismus für die Cu-vermittelte Fluorierung von Aryltrifluorboraten mit KF her.[41] Demzufolge kommen $Cu(OTf)_2$ zwei entscheidende Rollen zu. Zum einen dient es als Vermittler für die C–F Kupplung, zum anderen als Oxidationsmittel, um ein postuliertes Cu(III)-Intermediat zu bilden. Vorgeschlagen wurde ein Mechanismus über die Transmetallierung eines Cu(OTf)(F)-Komplexes. Dieser wird durch $Cu(OTf)_2$ zu einer hoch reaktiven Cu(III)-Spezies oxidiert. Durch reduktive Eliminierung wird aus diesem Cu(III)-Komplex das Fluorierungsprodukt gebildet (Abb 1.5).

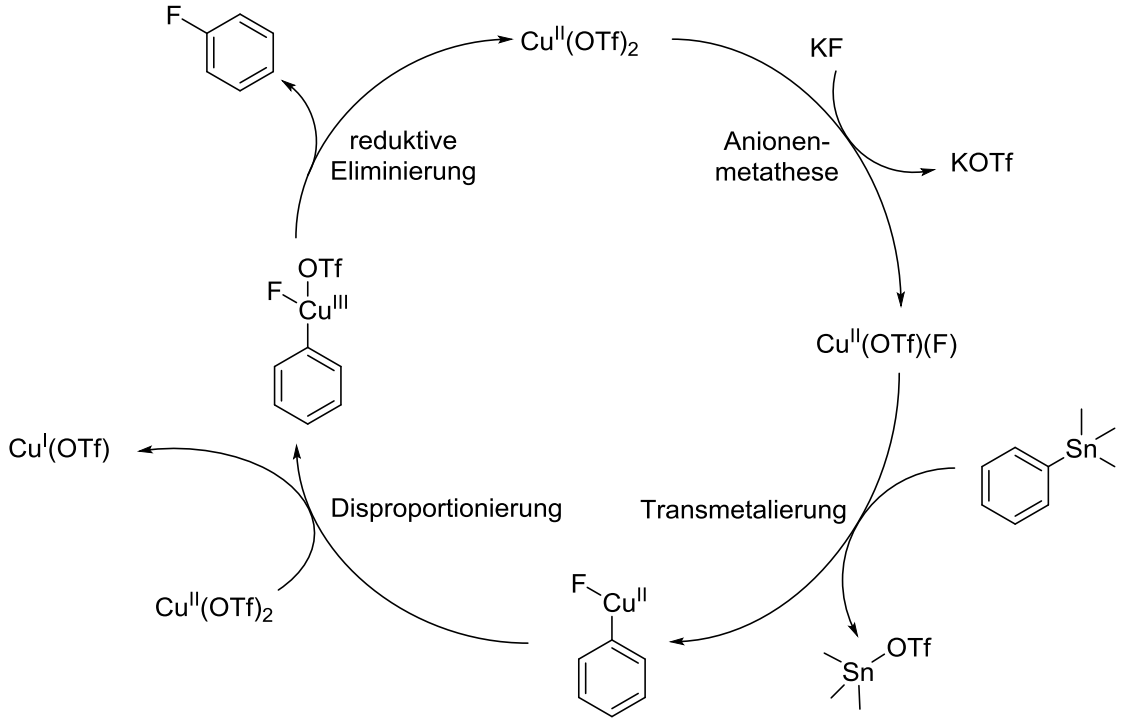

Abbildung 1.5 Postulierter Mechanismus der Cu-vermittelten Fluorierung von Arylstannanen.

Als weiteres Schlüsselelement wird der stabilisierende Einfluss von MeCN auf die Cu-Intermediate hervorgehoben. Durch die Zugabe von 10% MeCN konnten die Ausbeuten signifikant gesteigert werden.

Ebenfalls 2016 beschrieben Makaravage *et al.* die erfolgreiche radiochemische Anwendung der Cu-vermittelten Fluorierung von Arylstannanen.[42] Dabei war ihr Ausgangspunkt die von Ye *et al.* beschriebene Optimierung der Radiofluorierung von Aryltrifluorboraten. Unter optimierten Reaktionsbedingungen gelang ihnen die Fluorierung mit KF in MeCN innerhalb von 15 min. Zur Übertragung dieser Methode in die ^{18}F-Chemie mussten drei Modifikationen vorgenommen werden. Es wurde ein Amid-basiertes Lösungsmittel (Dimethylformamid (DMF) oder Dimethylacetamid (DMA)) verwendet, die Reaktionstemperatur musste auf 140 °C erhöht werden und, wie schon bei den Boronsäure-Vorläufern, wurde auch hier Pyridin als Cu-Ligand hinzugefügt. Unter diesen Bedingungen konnten verschiedene

Modellverbindungen in radiochemischen Umsätzen von 7–64% mit ^{18}F markiert werden. Abschließend wurde [^{18}F]MPPF, ein spezifisch bindender Ligand für den $5HT_{1A}$-Rezeptor,[43] in einer radiochemischen Ausbeute von 13% isoliert.

Bei allen drei, der hier beschriebenen Cu-vermittelten Radiofluorierungsstrategien ist eine azeotrope Trocknung zwingend erforderlich. Diese geht, wie schon in den vorherigen Abschnitten beschrieben, mit hohen Aktivitätsverlusten einher. So beschreiben Mossine *et al*, dass nach der azeotropen Trocknung nur noch 20–50% des eingesetzten $^{18}F^-$ gelöst werden konnte. Dies zeigt sich auch in der deutlichen Diskrepanz zwischen radiochemischen Umsätzen und radiochemischen Ausbeuten der isolierten Produkte. Trotz ^{18}F-Inkorporationsraten von bis zu 74%, konnte keine der Verbindungen in höheren Ausbeuten als 13% isoliert werden. Daher war es Bestandteil der vorliegenden Arbeit, die hier beschriebenen Cu-vermittelten Radiofluorierungsverfahren zu verfeinern, sodass Tracer für die PET in hohen radiochemischen Ausbeuten und Dosen hergestellt werden können. Da es sich bei den hier verwendeten Vorläufer nicht um Oniumsalze handelt, eignen sich diese Methoden nicht für die Anwendung der „minimalistischen“ Methode. Um dennoch auf die azeotrope Trocknung verzichten zu können, lag der Fokus auf der Entwicklung einer neuen Elutionsmethode, die es ermöglicht, das eluierte $^{18}F^-$ ohne weitere Aufbereitung für die Radiofluorierung zu verwenden.

2 Abdrucke der, zur kumulativen Promotion eingereichten Publikationen

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1) B. D. Zlatopolskiy, J. Zischler, E. A. Urusova, H. Endepols, E. Kordys, H. Frauendorf, F. M. Mottaghy, B. Neumaier: A Practical One-Pot Synthesis of Positron Emission Tomography (PET) Tracers via Nickel-Mediated Radiofluorination, *ChemistryOpen*, **2015**, *4*, 457–462. © 2015 The Authors. Published by Wiley-VCH Verlag GmbH & Co. KGaA.

2) B. D. Zlatopolskiy, J. Zischler, P. Krapf, F. Zarrad, E. A. Urusova, E. Kordys, H. Endepols, B. Neumaier: Copper-Mediated Aromatic Radiofluorination Revisited: Efficient Production of PET Tracers on a Preparative Scale. *Chemistry – A European Journal*, **2015**, *21*, 5972–5979. Copyright Wiley-VCH Verlag GmbH & Co. KGaA. Reproduced with permission.

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A Practical One-Pot Synthesis of Positron Emission Tomography (PET) Tracers via Nickel-Mediated Radiofluorination

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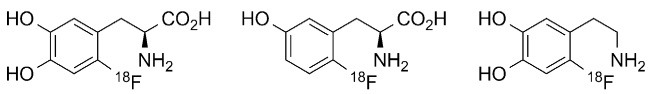
Recently a novel method for the preparation of ^{18}F-labeled arenes via oxidative [^{18}F]fluorination of easily accessible and sufficiently stable nickel complexes with [^{18}F]fluoride under exceptionally mild reaction conditions was published. The suitability of this procedure for the routine preparation of clinically relevant positron emission tomography (PET) tracers, 6-[^{18}F]fluorodopamine (6-[^{18}F]FDA), 6-[^{18}F]fluoro-L-DOPA (6-[^{18}F]FDOPA) and 6-[^{18}F]fluoro-*m*-tyrosine (6-[^{18}F]FMT), was evaluated. The originally published base-free method was inoperative. However, a "low base" protocol afforded protected radiolabeled intermediates in radiochemical conversions (RCCs) of 5–18%. The subsequent deprotection step proceeded almost quantitatively (>95%). The simple one-pot two-step procedure allowed the preparation of clinical doses of 6-[^{18}F]FDA and 6-[^{18}F]FDOPA within 50 min (12 and 7% radiochemical yield, respectively). In an unilateral rat model of Parkinson's disease, 6-[^{18}F]FDOPA with high specific activity (175 GBq μmol^{-1}) prepared using the described nickel-mediated radiofluorination was compared to 6-[^{18}F]FDOPA with low specific activity (30 MBq μmol^{-1}) produced via conventional electrophilic radiofluorination. Unexpectedly both tracer variants displayed very similar in vivo properties with respect to signal-to-noise ratio and brain distribution, and consequently, the quality of the obtained PET images was almost identical.

Introduction

Radiofluorinated aromatic amines and amino acids are important diagnostic tools in modern positron emission tomography (PET) imaging (Figure 1). 6-[^{18}F]Fluoro-3,4-L-dihydroxyphenylalanine (6-[^{18}F]FDOPA, [^{18}F]**1a**) is applied in clinical practice as a biomarker of catecholamine synthesis, storage and metabolism, and enables visualization of neuroendocrine tumors as well as the measurement of integrity and function of the nigrostriatal dopaminergic system.[1] 6-[^{18}F]Fluoro-L-*m*-tyrosine (6-[^{18}F]FMT, [^{18}F]**1b**), a close structural and functional analogue of 6-[^{18}F]FDOPA with improved biodistribution properties, is also used in the clinic for the same purposes.[2]

In addition to 6-[^{18}F]FDOPA and 6-[^{18}F]FMT, 6-[^{18}F]FDA ([^{18}F]**1c**) has also gained importance for the detection of neuroendocrine tumors such as pheochromocytomas and paragangliomas.[3] However, until quite recently, widespread application of these compounds has been hampered by a paucity of effective production routes, since incorporation of [^{18}F]fluoride into electron-rich aromatic systems is challenging.



6-[^{18}F]FDOPA ([^{18}F]**1a**) 6-[^{18}F]FMT ([^{18}F]**1b**) 6-[^{18}F]FDA ([^{18}F]**1c**)

Figure 1. Structures of 6-[^{18}F]FDOPA ([^{18}F]**1a**), 6-[^{18}F]FMT ([^{18}F]**1b**), and 6-[^{18}F]FDA ([^{18}F]**1c**).

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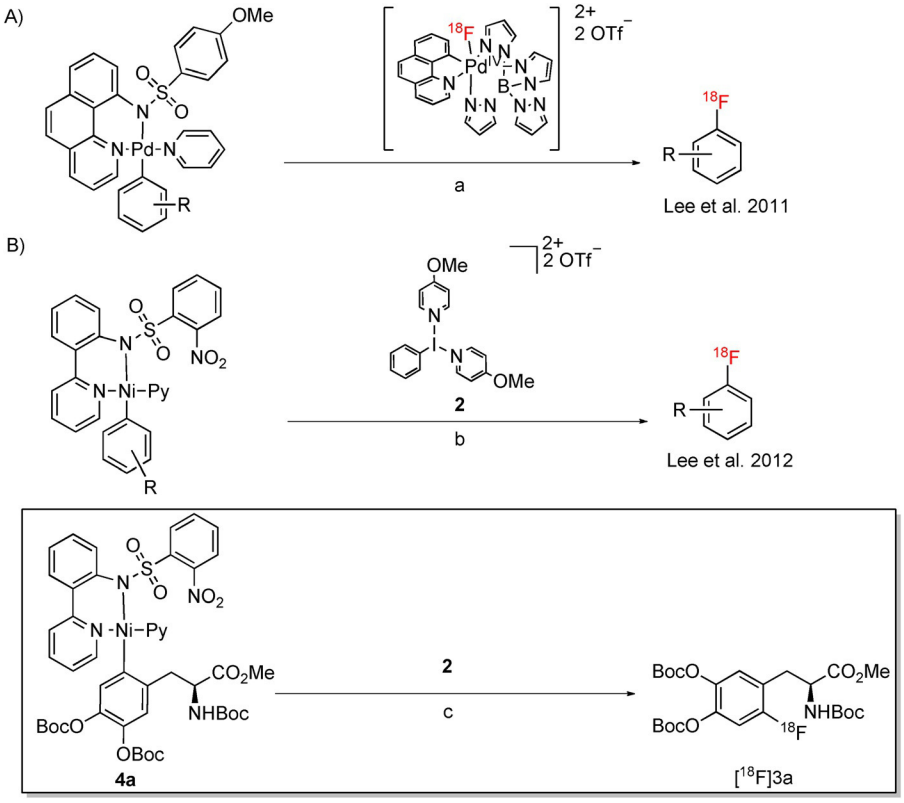
Electrophilic fluorodestannylation using $[^{18}F]F_2$ or $[^{18}F]CH_3COOF$ is routinely used for the preparation of these tracers.[4] However, this method suffers from several disadvantages. $[^{18}F]F_2$ gas target handling is difficult owing to the toxicity and corrosive character of fluorine. Moreover, $[^{19}F]F_2$ has to be applied as a carrier gas for ^{18}F to compete with surface adsorption enabling complete recovery of radioactivity from the target. As a consequence, specific activity (SA) of $[^{18}F]F_2$ does not exceed 350–500 MBq μmol^{-1}.[5] Accordingly, production of tracers with high SA via electrophilic radiofluorination is impossible. In order to obtain radiotracers with high SA and avoid F_2 target gas handling, radiosyntheses should start from $[^{18}F]$fluoride. Wagner et al.[6] proposed a synthesis route for 6-$[^{18}F]$FDOPA via isotopic $^{18/19}F$ exchange using a formyl-activated precursor followed by Baeyer–Villiger oxidation and final hydrolysis of the radiolabeled intermediate. 6-$[^{18}F]$FMT and several other aromatic amino acids were also successfully prepared using this synthetic route.[7]



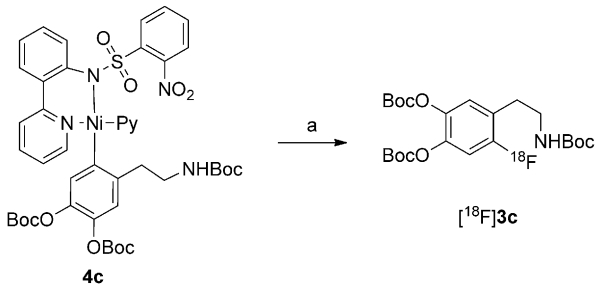
Scheme 1. Palladium- and nickel-mediated preparation of $[^{18}F]$fluoroarenes according to Lee et al. (A and B, respectively).[11] *Reagents and conditions*: a) acetone, 85 °C, 10 min; b) aq $[^{18}F]$fluoride, 18-crown-6, MeCN, RT, < 1 min; c) aq $[^{18}F]$fluoride, 18-crown-6, MeCN, RT, < 1 min, RCC = 15 ± 7 %; RCC = radiochemical conversion.[12]

Various multistep radiosyntheses of no-carrier-added (n.c.a.) 6-$[^{18}F]$FDOPA using nucleophilic $[^{18}F]$fluoride have been reported. All these procedures start with the preparation of a suitably protected 6-$[^{18}F]$fluoro-3,4-dihydroxybenzaldehyde (usually, 6-$[^{18}F]$fluoroveratraldehyde).[8] The latter is converted into the corresponding benzyl bromide or iodide, which is subsequently used for the alkylation of the chiral glycine equivalent or achiral glycine derivative with or without a chiral phase transfer catalyst.[9] Finally, acidolytic cleavage of protecting groups (typically under harsh reaction conditions) followed by HPLC or if necessary chiral HPLC purification is carried out. 6-$[^{18}F]$FDOPA is obtained in moderate radiochemical yields (RCYs), with SA > 37 GBq μmol^{-1} and high enantiomeric purity. Several procedures for the preparation of 6-$[^{18}F]$FDA from $[^{18}F]$fluoride have also been reported.[4,10] Unfortunately, similarly to those for 6-$[^{18}F]$FDOPA and 6-$[^{18}F]$FMT, the majority of these syntheses consist of numerous laborious reaction and operation steps, and are therefore difficult to implement in routine radiopharmaceutical production. Accordingly, convenient preparation procedures for the preparation of $[^{18}F]$**1a–c** via operationally simple nucleophilic ^{18}F-labeling of electron-rich arenes are highly sought after.

Recently Lee et al. reported a radiofluorination procedure using a palladium-based fluoride-derived electrophilic radiofluorination reagent for the synthesis of ^{18}F-labeled aromatic compounds (Scheme 1 A).[11a] It comprises the reaction of a radiofluorinated Pd^{IV} complex prepared in advance with an appropriate arylpalladium(II) complex yielding, after reductive elimination, the corresponding ^{18}F-labeled arene. A further development of this method involves radiolabeling of an arylnickel(II) complex with a radiofluorination agent generated in situ from a hypervalent iodine oxidant [bis(onio)-substituted aryliodine(III), **2**] and $[^{18}F]$fluoride (Scheme 1 B).[11b] According to the latter procedure, radiolabeling was carried out in the presence of only 1 mg of the corresponding nickel complex at room temperature under ambient atmosphere within less than 1 min to yield radiolabeled arenes with > 37 GBq μmol^{-1} SA in fair to moderate radiochemical conversions (RCCs).[12] According to this publication, neither base nor azeotropic drying were necessary. This method was applied to prepare protected 6-$[^{18}F]$FDOPA ($[^{18}F]$**3a**) from the corresponding nickel complex (**4a**) in 15 % RCC (Scheme 1 B). The exceptional operational simplicity, very mild reaction conditions and short reaction time prompted us to study the applicability of this method for the preparation of clinically relevant doses of $[^{18}F]$**1a–c**. Finally, high and low SA preparations of 6-$[^{18}F]$FDOPA obtained via nickel-mediated and conventional electrophilic radiofluorination, respectively, were compared in a rat model of hemi-Parkinson's disease.

Results and Discussion

At the outset, we tried to prepare protected 6-[^{18}F]FDA ([^{18}F]**1c**) from corresponding nickel complex **4c** (Scheme 2).



Scheme 2. Radiosynthesis of protected [^{18}F]FDA ([^{18}F]**3c**). *Reagents and conditions*: a) [^{18}F]fluoride, base, 18-crown-6, oxidant **2**, MeCN, RT, 1—20 min.

Unexpectedly, under base-free conditions originally reported by Lee et al.,[11b] no product formation was observed; only [^{18}F]F$^-$ was detected in the reaction mixture. Similarly, if azeotropically dried [^{18}F]KF/18-crown-6 prepared according to the conventional nucleophilic radiofluorination protocol[11b] using potassium carbonate (2.5–3.2 mg) was applied, no ^{18}F-incorporation took place. We assumed that oxidant **2** is prone to decomposition under strongly basic conditions. Consequently, the applicability of our "low base" protocol initially developed for copper-mediated radiofluorination was tested.[13]

^{18}F$^-$ was trapped on an anion-exchange resin and then eluted with a methanolic solution of potassium carbonate into a reaction vial containing 18-crown-6. A small amount of potassium carbonate (0.16 mg) was sufficient to recover ^{18}F$^-$ quantitatively (>98%). Since low-boiling methanol could be completely removed at 70–80 °C within 2–3 min, the time of drying of [^{18}F]KF/18-crown-6 complex was significantly decreased. The residue was taken up in acetonitrile and added to nickel complex **4c** and oxidant **2.** The resulting mixture was stirred at ambient temperature under inert conditions. Application of **4c** (1 mg) according to the literature[11b] afforded ^{18}F-labeled protected 6-FDA [^{18}F]**3c** in only 1–4% RCC after 1 min. In contrast, if a greater amount of nickel complex **4c** (5–10 mg) was utilized, [^{18}F]**3c** was formed in 12±4% RCC within 5 min. At elevated temperatures (>30 °C), no ^{18}F incorporation was observed. Use of other bases such as cesium carbonate and potassium bicarbonate also provided [^{18}F]**3c**, but in lower RCCs (5±2 and 7±3%, respectively) (Figure 2).

In contrast to the literature,[11b] formation of [^{18}F]**3c** took place only in the presence of a base. This finding prompted us to optimize this radiolabeling procedure using nickel complex **4c** as a model substrate with respect to other reaction parameters such as oxidant/precursor ratio, reaction time and solvent.

The oxidant/precursor ratio strongly affected the formation of [^{18}F]**3c** (Figure 3). Application of an excess of radiolabeling precursor **4c** afforded only traces of [^{18}F]**3c**. If equimolar amounts of oxidizing agent **2** and nickel complex **4c** were

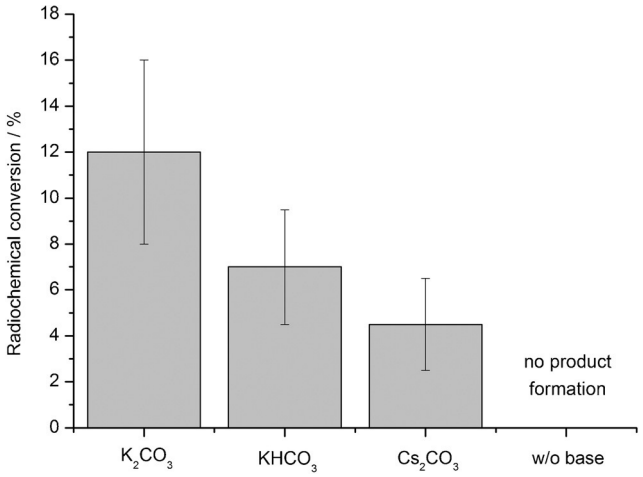


Figure 2. Influence of base on radiochemical conversion (RCC) of [^{18}F]**3c**. The appropriate [^{18}F]fluoride salt/18-crown-6 complex (50–500 MBq) was prepared using the corresponding base (1.16 μmol) in MeOH (200 μL). MeOH was evaporated, and the residue taken up in MeCN (900 μL). The solution was added to nickel complex **4c** (5 μmol) and **2** (1 equiv), and the reaction mixture was stirred for 5 min at RT. Thereafter, water (5 mL) was added, the mixture was vigorously stirred for 1 min and then analyzed by radio-HPLC. Values represent the mean ± standard deviation (SD) of at least three experiments.

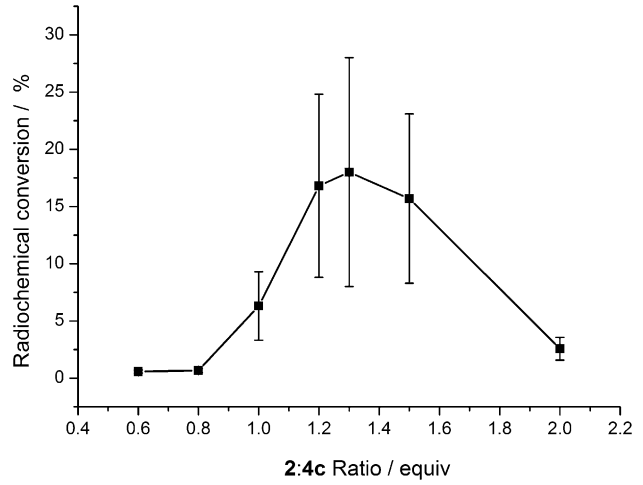


Figure 3. Radiochemical conversion (RCC) of [^{18}F]**3c** as a function of the oxidant/precursor ratio. A solution of [^{18}F]KF/18-crown-6 (50–500 MBq) in MeCN (900 μL) was added to nickel complex **4c** (5 μmol) and **2** (0.6–2.0 equiv), and the reaction mixture was stirred for 5 min at RT. Thereafter, water (5 mL) was added, the mixture was vigorously stirred for 1 min and then analyzed by radio-HPLC. Values represent the mean ± standard deviation (SD) of at least three experiments.

used, the RCC value of [^{18}F]**3c** amounted to 6±3%. If the oxidant to nickel complex molar ratio was 1.3, a maximal RCC value of 18±10% was obtained. Above a ratio of 1.5, a considerable decrease in RCC values was observed.

Radiofluorination of **4c** was tested in different solvents (Figure 4). The highest RCC values were achieved in anhydrous acetonitrile. In contrast to literature reports,[11b] the presence of a low amount of water (1%) in the reaction mixture caused a significant decrease in RCC values. In sulfolane, diglyme and

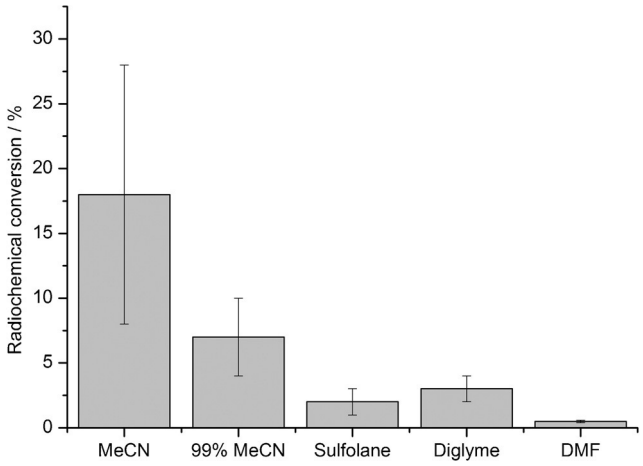


Figure 4. Radiochemical conversion (RCC) of [^{18}F]**3c** in different solvents. A solution of [^{18}F]KF/18-crown-6 (50–500 MBq) in the corresponding solvent (900 μL) was added to nickel complex **4c** (5 μmol) and **2** (6.5 μmol), and the reaction mixture stirred for 5 min at RT. Thereafter, water (5 mL) was added, the mixture was vigorously stirred for 1 min and then analyzed by radio-HPLC. Values represent the mean ± standard deviation (SD) of at least three experiments.

dimethylformamide (DMF), formation of [^{18}F]**3c** was significantly lower in comparison to that observed in acetonitrile. Dependency of RCC of **4c** on reaction time was also determined (Figure 5). Under optimized conditions, reaction times of 1 and 5 min afforded [^{18}F]**3c** in RCCs of 7 ± 3% and 18 ± 10%, respectively. A further extension of the reaction time did not increase the RCC.

During our experiments, we noticed the extreme moisture sensitivity of the oxidizing agent **2**.[14] This was, for example, the reason for the high statistical deviations of RCC values that

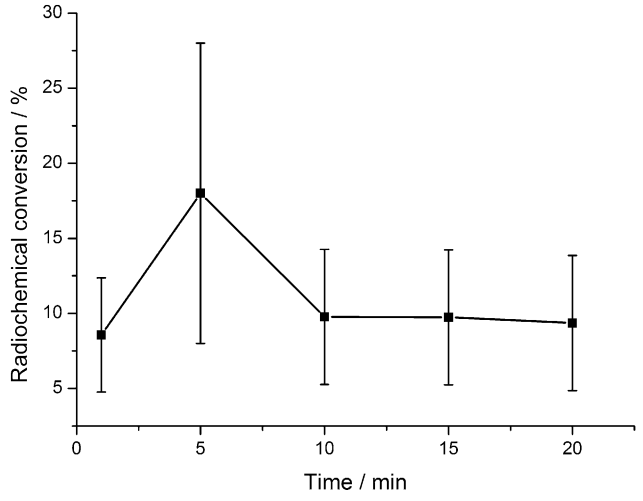


Figure 5. Dependence of radiochemical conversion (RCC) of [^{18}F]**4c** on reaction time. A solution of [^{18}F]KF/18-crown-6 (50–500 MBq) in MeCN (900 μL) was added to nickel complex **4c** (5 μmol) and **2** (1.3 equiv), and the mixture was stirred for the corresponding time at RT. Thereafter, water (5 mL) was added, the mixture was vigorously stirred for 1 min and then analyzed by radio-HPLC. Values represent the mean ± standard deviation (SD) of at least three experiments.

were observed in the optimization experiments. In an attempt to overcome this problem, we prepared several hypervalent iodine compounds, such as **5**,[15] and iodosylarenes **6a,b**[16] as less moisture-sensitive alternatives to **2** (Figure 6). However, application of these compounds as oxidants did not afford incorporation of ^{18}F.

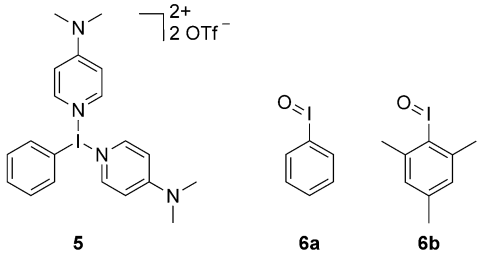


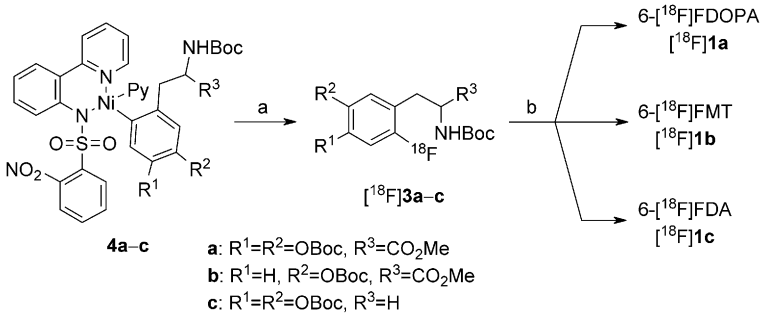
Figure 6. Hypervalent iodine oxidants tested as possible alternatives to **2**.

With optimized reaction conditions for radiolabeling of **4c** in hand, we tested ^{18}F-radiofluorination of nickel complexes **4a**[11b] and **4b**. Accordingly, protected derivatives of 6-[^{18}F]FDOPA and 6-[^{18}F]FMT, [^{18}F]**3a** and [^{18}F]**3b**, were prepared in 13 ± 2% and 9 ± 1% RCC, respectively. The next preparation step should comprise the deprotection of the corresponding ^{18}F-labeled intermediates, [^{18}F]**3a–c**, to the desired PET tracers. Initially, trifluoroacetic acid (TFA) at 80 and 130 °C was tested as a deprotection agent. Under these conditions, complete decomposition of radiolabeled intermediates was observed with ^{18}F$^-$ as the only detectable radioactive product. In contrast, quantitative deprotection of [^{18}F]**3c** was accomplished using 12 M aq HCl at 130 °C after just 5 min. In the case of [^{18}F]**3a** and [^{18}F]**3b**, cleavage of the methyl ester group required a slightly extended deprotection time of 10 min.

Once the protocol for the radiosynthesis of [^{18}F]**3a–c** via nickel-mediated radiofluorination had been established, we tried to implement a scale-up synthesis procedure to obtain the corresponding PET tracers in amounts sufficient for biological evaluation. After accomplishment of the radiofluorination step, acetonitrile was evaporated under a gentle stream of argon, and the residue was redissolved in 12 M aq HCl. After hydrolysis had been completed, the bulk of HCl was removed by coevaporation with acetone. The crude products were purified by HPLC to give the corresponding PET tracers as ready-to-use solutions in a 4% ethanolic phosphate buffer. Application of a one-pot protocol allowed us to minimize loss of radioactivity due to surface adsorption and to decrease radiosynthesis time significantly.

Using this procedure, [^{18}F]**1a**, [^{18}F]**1b**[17] and [^{18}F]**1c** were isolated in RCYs of 7 ± 1, 5 ± 1 and 12 ± 2%, respectively, with excellent radiochemical and chemical purity (Scheme 3). Specific activities of [^{18}F]**1a** and [^{18}F]**1c** were determined to 175 and 60 GBq μmol^{-1}, respectively.[18]

Finally, we studied, whether the specific activity of 6-[^{18}F]FDOPA affects PET imaging of dopaminergic activity in brain. For this purpose, n.c.a. 6-[^{18}F]FDOPA produced via nickel-mediated radiofluorination and carried-added (c.a.) 6-



Scheme 3. Preparation of 6-[^{18}F]FDOPA ([^{18}F]**1 a**), 6-[^{18}F]FMT ([^{18}F]**1 b**) and 6-[^{18}F]FDOPA ([^{18}F]**1 c**) via radiofluorination of nickel complexes **4 a–c**. *Reagents and conditions*: a) [^{18}F]KF/18-crown-6, oxidant **2**, MeCN, RT, 5 min; b) 12 M aq HCl, 130 °C, 5–10 min. [^{18}F]**1 a**, RCY = 7%, 220 MBq from 6.3 GBq $^{18}F^-$, SA = 175 GBq μmol^{-1}; [^{18}F]**1 b**, RCY = 5%; [^{18}F]**1 c**, RCY = 12%, 250 MBq from 4 GBq $^{18}F^-$, SA = 60 GBq μmol^{-1}; RCY = radiochemical yield;[12] SA = specific activity.

striatum with similar kinetics (time activity curves in Figure 7). Mean striatal standardized uptake values (SUVs) (%ID/g) were 0.22 and 0.21 for n.c.a. and c.a. 6-[^{18}F]FDOPA, respectively. In addition, the striatum-to-cerebellum ratio was determined to be independent of the specific activity of the tracer. In both cases, the dopaminergic lesion was clearly visible with an identical ipsi-to-contralateral striatal ratio.

This somewhat unexpected result could be explained by the high capacity and/or high competitiveness of the main biochemical processes responsible for the accumulation of 6-[^{18}F]FDOPA and its metabolites in dopaminergic neurons. In this case, competition between 6-[^{18}F]FDOPA and 6-FDOPA should be negligible compared with that between 6-[^{18}F]FDOPA and other competitors.

6-[^{18}F]FDOPA metabolism in brain comprises five main steps. Initially, the radiotracer is taken up across the blood–brain barrier (BBB) mainly via L-type amino acid transporter (LAT-1).[21] LAT-1 is also responsible for brain uptake of proteinogenic neutral amino acids like Phe, Tyr, Val, Ile, Leu, and Trp.[22] Therefore, the competition between 6-[^{18}F]FDOPA and other amino acids is significantly higher relative to that with 6-FDOPA. In striatal neurons, 6-[^{18}F]FDOPA is decarboxylated into 6-[^{18}F]FDA.[23] This step is catalyzed by aromatic L-amino acid decarboxylase (AADC) with very broad substrate specificity and high capacity.[24] 6-[^{18}F]FDA is transported into the vesicles by the vesicular monoamine transporter type 2 (VMAT2).[23] Since VMAT2 is responsible for detoxification of toxic amines like 6-FDA,[25] its transport capacity should be high. Subsequently, stored 6-[^{18}F]FDA is transported to the synaptic cleft by exocytosis and taken up there again by dopamine transporters (DAT). Finally, cytosolic 6-[^{18}F]FDA is metabolized to 6-[^{18}F]fluoro-3,4-dihydroxyphenylacetic acid (6-[^{18}F]FDOPAC), 6-[^{18}F]fluoro-3-methoxytyramine (6-[^{18}F]FMTA) and 6-[^{18}F]fluorohomovanilic acid (6-[^{18}F]FHVA) by monoamine oxidase (MAO) and catechol-*O*-methyltransferase (COMT).[26] These enzymes also exhibit broad substrate specificity and high capacity. The acidic metabolites, 6-[^{18}F]FDOPAC and 6-[^{18}F]FHVA, are rapidly cleared from brain.[27] Vesicular stored 6-[^{18}F]FDA as well as its cytosolic and extracellular metabolites are responsible for more than 90% of the striatal PET signal generated by 6-[^{18}F]FDOPA in brain at 10–90 min p.i.[26]

[^{18}F]FDOPA prepared via conventional electrophilic radiofluorination were compared in an unilateral rat model of hemi-Parkinson's disease.[19] Accordingly, loss of dopaminergic midbrain neurons was induced by stereotaxic injection of neurotoxic 6-hydroxydopamine (6-OHDA) into the medial forebrain bundle.[20] Six weeks after 6-OHDA injection, rats pretreated with carbidopa were injected into the lateral tail vein with 56–74 MBq of n.c.a 6-[^{18}F]FDOPA and measured. Eight weeks after 6-OHDA injection, the same animals were measured using c.a 6-[^{18}F]FDOPA (67–72 MBq) (Figure 7).

Specific activity of the c.a. tracer (30 MBq μmol^{-1}) was more than three orders of magnitude lower than that of the n.c.a. compound. Both tracer variants were taken up by the intact

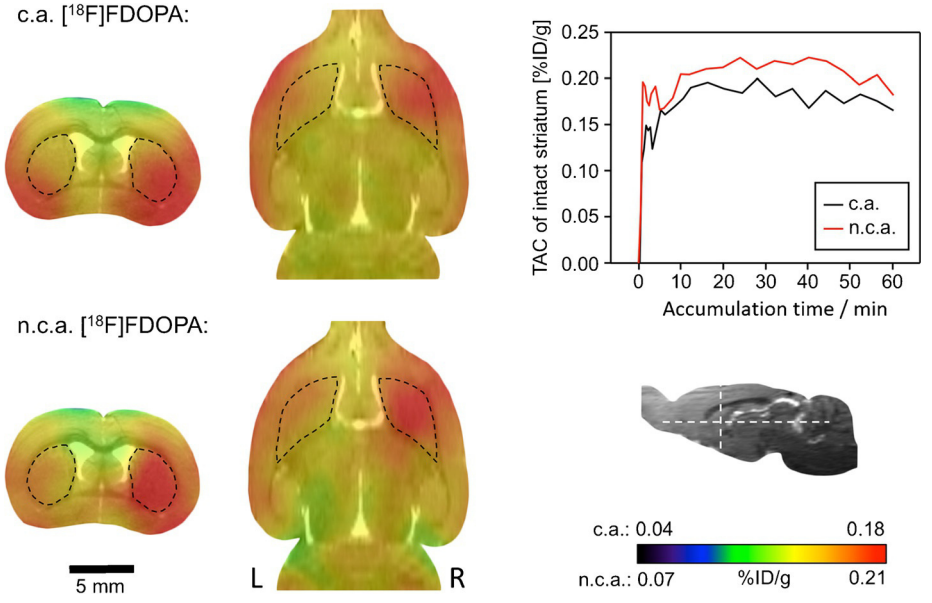


Figure 7. No-carrier-added (n.c.a.) versus carrier-added (c.a.) 6-[^{18}F]FDOPA: magnetic resonance/positron emission tomography (MR/PET) imaging in a rat model of Parkinson's disease. Two transverse (left column) and two horizontal images (middle column) from the same animal with left side lesion induced by the injection of 6-hydroxydopamine (6-OHDA) are shown. Left and right striatum are indicated by dashed black outlines. 6-OHDA-Induced lesion is visible as a reduction of the PET signal in the left striatum. Section levels are indicated by white dashed lines in the insert (bottom right). The time activity curves (TAC; top right) for both images were taken from the intact right striatum.

Conclusions

Nickel-mediated radiofluorination under optimized "low base" conditions enabled fast access to n.c.a. 6-[^{18}F]FDA, 6-[^{18}F]FDOPA, and 6-[^{18}F]FMT via a one-pot two-step procedure. Owing to the simplicity, this procedure should be well suited for automation given that a less moisture-sensitive alternative for oxidant **2** will be found. Furthermore, it was shown that, in a Parkinson's rat model, biodistribution and, consequently, imaging properties of 6-[^{18}F]FDOPA were independent from specific activity.[28]

Keywords: [^{18}F]fluoride · nucleophilic aromatic substitution · positron emission tomography (PET) · radiopharmaceuticals · radiosynthesis

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[12] Radiochemical conversion (RCC), determined by radio-HPLC, refers to the amount of radiofluoride reacted with the labeling precursor to give the desired ^{18}F-labeled compound. Radiochemical yield (RCY) refers to the isolated yield of the radiochemically and chemically pure radiolabeled compound; all RCYs reported here are corrected for decay.

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[14] To avoid decomposition, compound **2** should be handled and stored (for not longer than two months) in a glove box under strict exclusion of moisture.

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[17] In this work, racemic 6-[^{18}F]fluoro-*m*-tyrosine (6-[^{18}F]FMT) was prepared.

[18] Complete experimental details for the preparation of the reference compounds and precursors for radiofluorination, as well as radiochemical and biological experiments, together with HPLC chromatograms, NMR spectra, and details of the specific activity calculations are available on the WWW under http://dx.doi.org/10.1002/open.201500056.

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ChemistryOPEN

Supporting Information

A Practical One-Pot Synthesis of Positron Emission Tomography (PET) Tracers via Nickel-Mediated Radiofluorination

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Supporting Information

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Materials and Methods

General: ^{1}H-NMR spectra: Bruker Avance II 300 (300 MHz) and Bruker Avance II+ 600 (600 MHz). ^{1}H chemical shifts are reported in ppm relative to residual peaks of deuterated solvents. Higher-order NMR spectra were approximately interpreted as first-order spectra, where possible. The observed signal multiplicities are characterized as follows: s = singlet, d = doublet, t = triplet, q = quartet, quin = quintet, m = multiplet, and br = broad. Coupling constants (*J*) were reported in Hertz (Hz). ^{13}C-NMR spectra [additional APT (Attached ProtonTest)]: Bruker Avance II 300 (75.5 MHz) and Bruker Avance II+ 600 (125.9 MHz). ^{13}C chemical shifts are reported relative to residual peaks of deuterated solvents. Low resolution ESI-MS: Finnigan LCQ. High resolution ESI-MS: Bruker APEX IV 7T FTICR MS. TLC: Merck precoated sheets, 0.25 mm Sil G/UV$_{254}$. The chromatograms were viewed under UV light and/or by treatment with phosphomolybdic acid (10%

in ethanol). Column chromatography: Merck silica gel, grade 60, 230–400 mesh. Solvent proportions are indicated in a volume:volume ratio. All reactions were carried out with magnetic stirring unless otherwise stated and, in the case of air- or moisture-sensitive substrates and/or reagents, were handled in flame-dried glassware under argon or nitrogen. Organic extracts were dried with anhydrous $MgSO_4$. Oxidant **2** was handled and stored in a glove box under nitrogen at < 0.1 ppm RH.

2,[1] **4a**,[2] 6-bromo-*m*-tyrosine,[3] 6-bromodopamine,[4] 4-(chloromethyl)-5-fluoroveratrol,[5] 4-(cyanomethyl)-5-fluoroveratrol,[6] 2-(2-pyridinyl)phenyl-2-nitrobenzenesulfonamide silver(I) salt,[2] 1,1′-(phenyl-λ_3-iodandiyl)bis(4-dimethylaminopyridinium) bistrifluormethansulfonate,[7] iodosobenzene (**7a**)[8] and iodosomesytilene (**7b**)[9] were prepared according to literature.

StrataX cartridges were obtained from Phenomenex (Aschaffenburg, Germany) and Sep-Pak Accell Plus QMA carbonate plus light cartridges, 46 mg sorbent per cartridge from Waters GmbH (Eschborn, Germany).

HPLC analyses and purifications were carried out on Dionex Ultimate 3000 System with Ultimate 3000 Diode Array Detector coupled in series with Berthold NaI detector. Unless stated, a Chromolith® SpeedROD RP-18e column (Merck, Darmstadt Germany), 50×4.6 mm, was used for analyses and purifications of radiofluorinated products.

UV and radioactivity detections were connected in series, giving a time delay of 0.5–0.9 min depending on a flow rate. ^{18}F-labeled compounds were identified by spiking of the reaction mixture with unlabeled standards using HPLC.

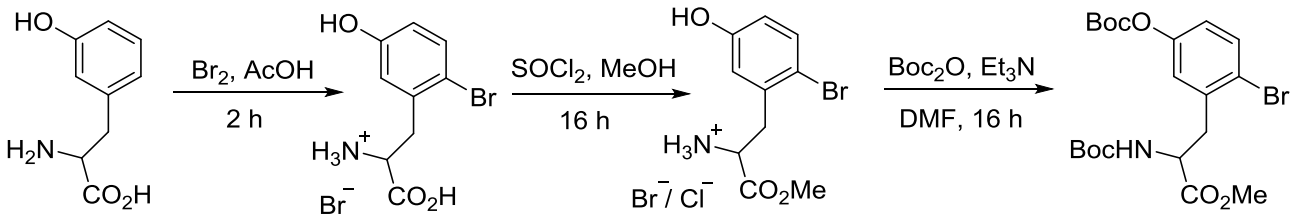
Radiolabeled products were analyzed using two sets of conditions. Conditions A (for protected intermediates [^{18}F]**3a-c**): column: (Chromolith SpeedROD®, 50×4.6 mm (Merck Milipore); gradient: 0–3 min: 20% MeCN, 3–4 min: 20→99% MeCN, 4–7 min: 99% MeCN, 7–8 min:

99→20% MeCN; flow rate: 1.5 mL/min. Conditions B (for PET-tracers [^{18}F]**1a-c**): column: Synergy 4 µm Hydro-RP (150×4.6 mm, Phenomenex); 4% EtOH in 0.02 M sodium phosphate buffer (pH 2.5); flow rate: 1.5 mL/min. [^{18}F]**1a-c** were isolated by semipreparative HPLC using conditions B.

[^{18}F]Fluoride was produced via the ^{18}O(p,n)^{18}F reaction by bombardment of enriched [^{18}O]water with 16.5 MeV protons using a MC16 cyclotron (Scanditronix, Uppsala, Sweden). All isolated radiochemical yields are decay-corrected. Unless otherwise indicated, all radiochemical experiments were carried out at least in triplicates.

Before radiosyntheses [^{18}F]fluoride was preprocessed as follows. [^{18}F]Fluoride in [^{18}O]H_2O (0.05–40 GBq) was trapped on an anion-exchange resin (QMA cartridge), the resin was washed with MeOH (5 mL) and flushed with a gentle stream of Ar (1 min). [^{18}F]Fluoride was eluted into a conical vial (5 mL), containing 18-crown-6 (24 mg, 90 µmol), with K_2CO_3 (0.16 mg, 1.16 µmol in 10 µL H_2O) in MeOH (200 µL) followed by additional MeOH (700 µL). After evaporation of the solvent under a gentlet stream of Ar at 70–80 °C, MeCN (1 mL) was added to the reaction vial and evaporated under a gentle stream of Ar at 100 °C (three times). The residue was taken up in the corresponding solvent and used for further experiments. It should be noted, that the aqueous [^{18}F]fluoride was loaded onto the cartridge from the male side, whereas flushing, washing and $^{18}F^-$ elution were carried out from the female side of the cartridge. If the QMA cartridge had been loaded, flushed and eluted from the female side only, sometimes a significant amount of [^{18}F]fluoride remained on the resin (this is probably because QMA-light (46 mg) cartridges have a single frit on the male side but four on the female side).

Chemistry

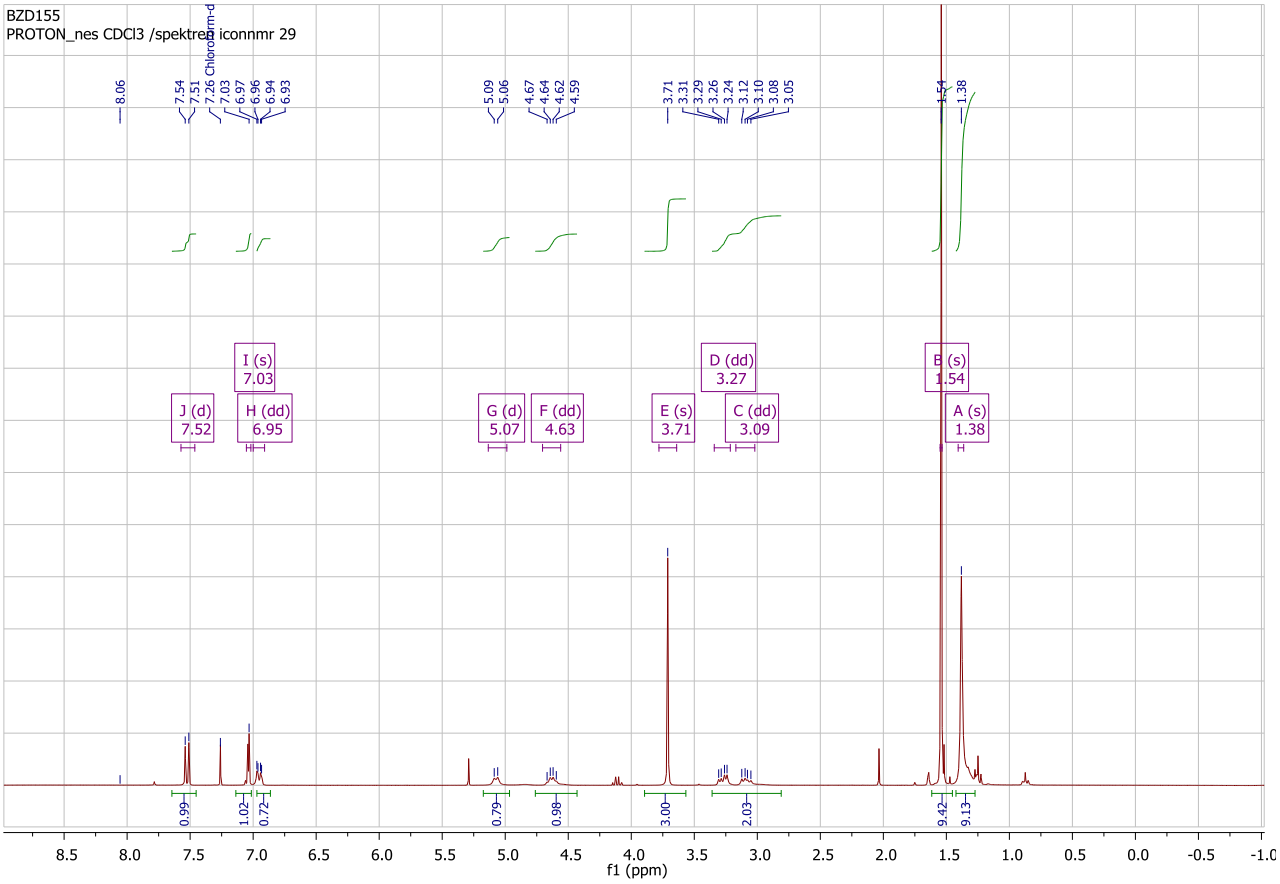


Methyl *N*,*O*-di-*tert*-butyloxycarbonyl-6-bromo-*m*-tyrosinate: A solution of Br_2 (0.91 mL, 2.42 g, 16.64 mmol) in AcOH (140 mL) was added dropwise within 45 min to a vigorously stirred suspension of *m*-tyrosine (3.2 g, 16.64 mmol) in AcOH (140 mL). The reaction mixture (the starting suspension was completely dissolved to give a clear solution; thereafter, a precipitataion of product was begun) was stirred for a further 2 h and filtered. A filter cake was washed with AcOH (3×50 mL) and transferred into a round flask. Toluene (50 mL) was added and evaporated under reduced pressure (three times) to give a crude 6-bromo-*m*-tyrosine hydrobromide[3] (2.0 g) as an off-white solid which was used for the next step without any further purification.

$SOCl_2$ (2.9 mL, 4.76 g, 40 mmol) was added dropwise within 30 min to an ice-cold solution of 6-bromo-*m*-tyrosine (1.95 g, max. 5.72 mmol) in anhydrous MeOH (30 mL) under Ar. Thereafter, the cooling bath was removed and the reaction mixture was stirred for a further 16 h and concentrated under reduced pressure. The residue was taken up in toluene (50 ml) and volatiles were removed under reduced pressure (two times) to give a crude methyl 6-bromo-*m*-tyrosinate hydrochloride/hydrobromide (1.7 g) as a rose solid which was directly used for the next step.

Et_3N (6.7 mL, 4.84 g, 47.87 mmol) was slowly added to a solution of methyl 6-bromo-*m*-tyrosinate hydrochloride (1.7 g, max. 5.47 mmol) and Boc_2O (6.7 g, 30.7 mmol) in anhydrous DMF (20 mL) under Ar. The reaction mixture was stirred for a further 16 h and concentrated under reduced

pressure. The residue was taken up in EtOAc (100 mL) and the resulting solution was washed with H_2O (7×40 mL), brine (2×30 mL), dried and concentrated under reduced pressure. The residue was purified by column chromatography (EtOAc:hexane = 1:3) affording the title compound (1.39 g, 17% on three steps) as a colorless viscous oil. R_f = 0.25, EtOAc:hexane = 1:3. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.52 (d, *J* = 8.7 Hz, 1H), 7.03 (s, 1H), 6.95 (dd, *J* = 8.7, 2.5 Hz, 1H), 5.07 (d, *J* = 8.2 Hz, 1H), 4.63 (dd, *J* = 14.3, 7.0 Hz, 1H), 3.71 (s, 3H), 3.27 (dd, *J* = 13.8, 7.0 Hz, 1H), 3.09 (dd, *J* = 13.8, 8.2 Hz, 1H), 1.54 (s, 3H), 1.38 (s, 3H). ^{13}C NMR (75 MHz, $CDCl_3$) δ 172.3, 155.1, 151.5, 150.4, 137.5, 133.6, 124.2, 121.8, 121.4, 84.0, 80.1, 53.4, 52.6, 38.9, 28.4, 27.8. MS (ESI): positive mode *m/z* = 971.2 ($[2M + Na]^+$), 498.3 ($[M + Na]^+$), 474.1 ($[M + H]^+$); MS (ESI): negative mode *m/z* = 472.1 ($[M - H]^-$); ESI HRMS: calcd for $C_{20}H_{28}NO_7BrNa^+$: 496.0937; found: 496.0941; calcd for $C_{20}H_{29}NO_7Br^+$: 474.1109; found: 474.1122; calcd for $C_{20}H_{27}NO_7Br^-$: 472.0958; found: 474.0976. Correct isotopic pattern.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4.64
4.62
4.59
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3.29
3.26
3.24
3.12
3.10
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3.05
1.54
1.38
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J (d) 7.52
H (dd) 6.95
G (d) 5.07
F (dd) 4.63
D (dd) 3.27
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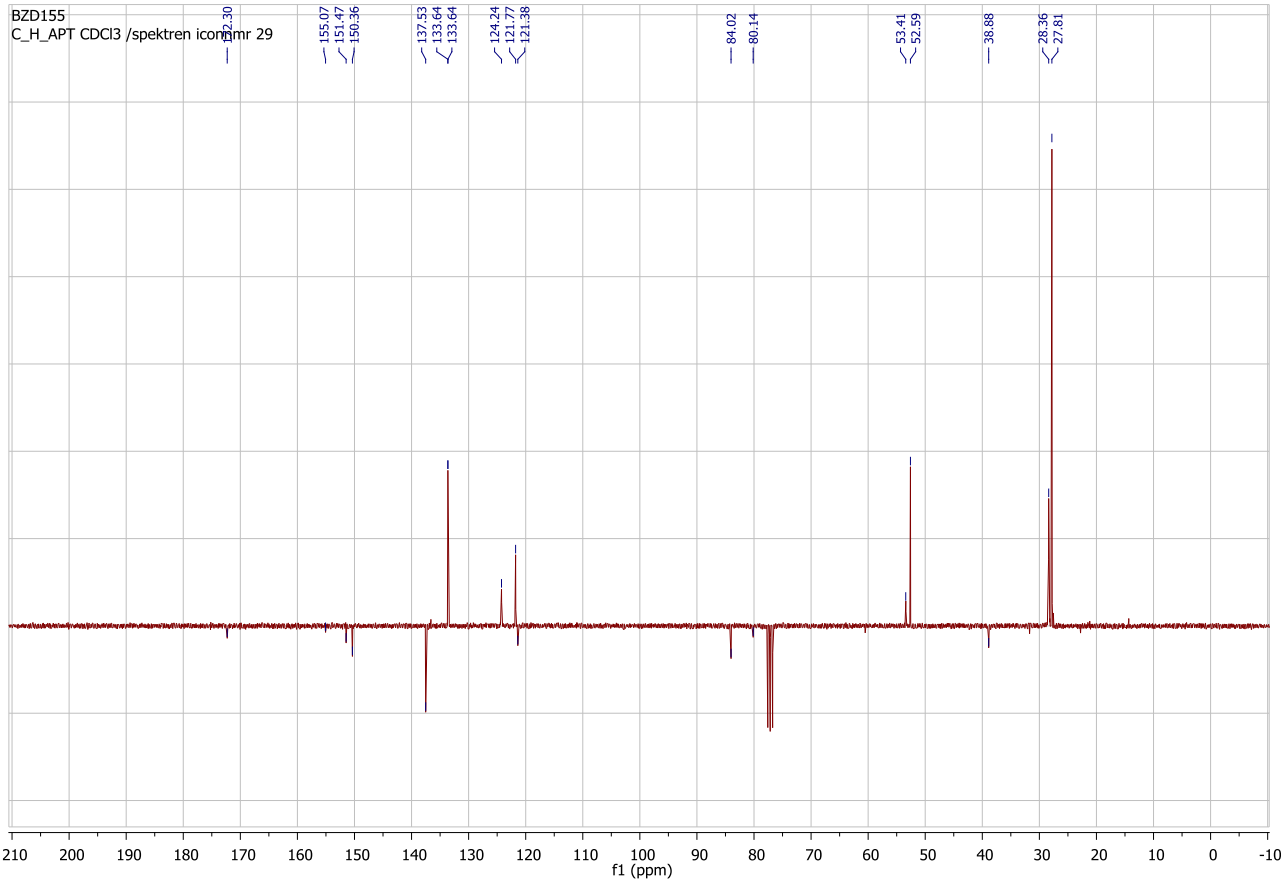
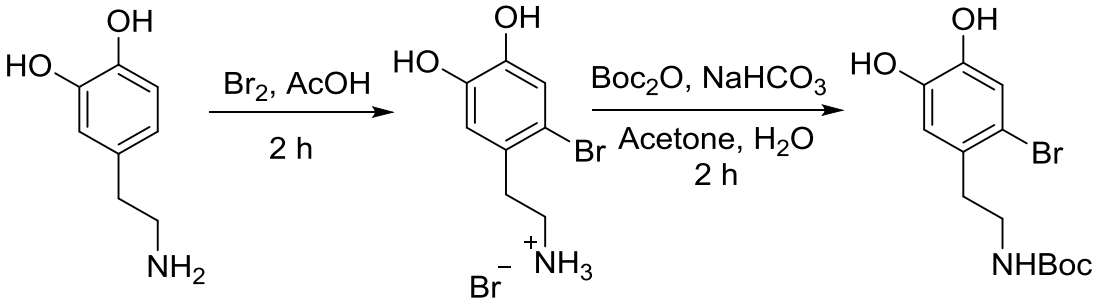


***N-tert*-Butyloxycarbonyl-6-bromodopamine:**



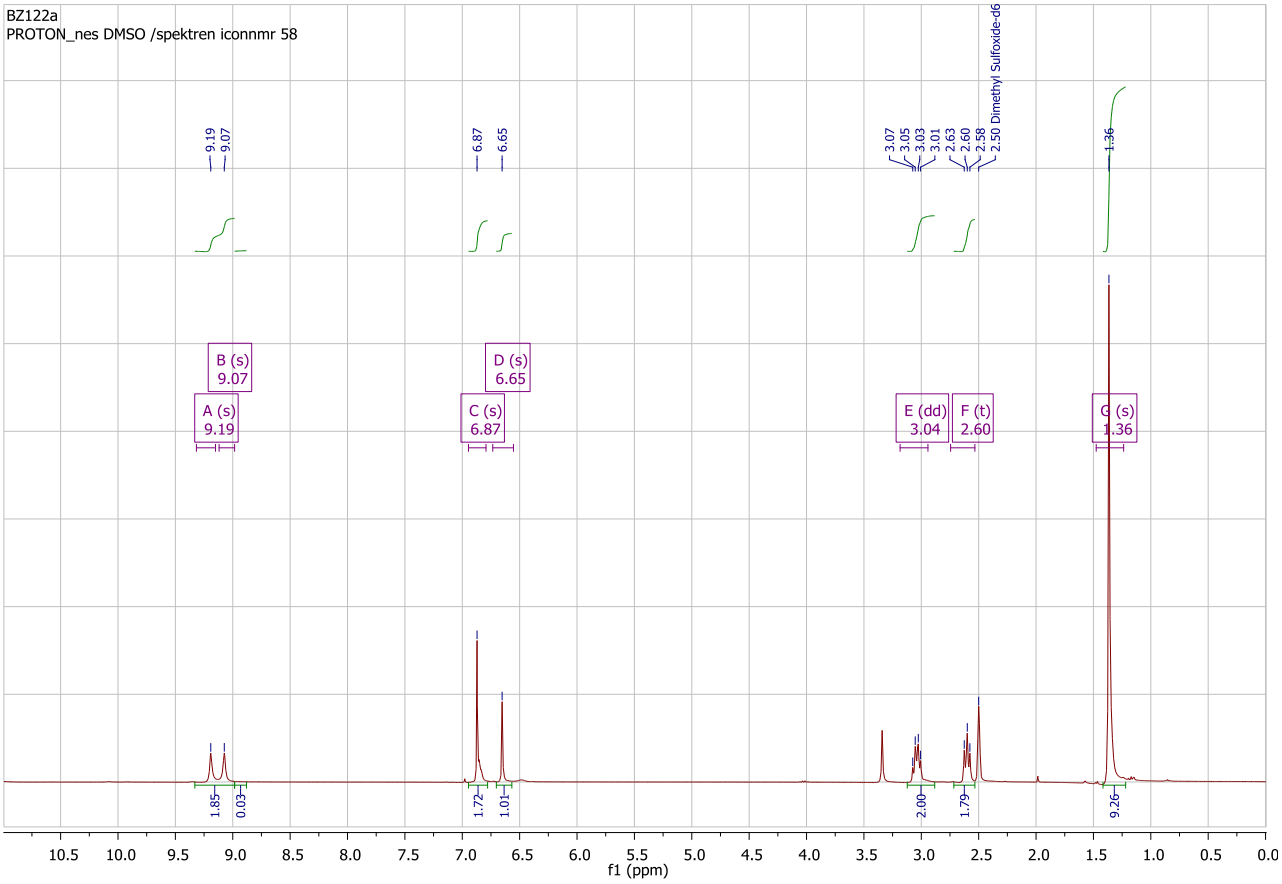
A solution of Br_2 (1.13 mL, 3.51 g, 21.99 mmol) in AcOH (25 mL) was added dropwise within 2 h to a vigorously stirred suspension of dopamine hydrochloride (4.17 g, 21.99 mmol) in AcOH (100 mL) and the reaction mixture was concentrated under reduced pressure. The dark-brown residue was taken up in toluene (50 mL) and volatiles were evaporated under reduced pressure (three times) to

give a crude 6-bromodopamine hydrobromide[4] which was directly used for the next step without any further purification.

Boc_2O (5.50 g, 25.2 mmol) in acetone (40 mL) was added to a solution of 6-bromodopamine hydrobromide (6.9 g, max. 21.99 mmol) and $NaHCO_3$ (5.54 g, 66.0 mmol) in H_2O (70 mL; degassed by passing of a gentle stream of argon for 1 h) under Ar and the reaction mixture was stirred for 2 h (if necessary additional acetone and/or H_2O was added to homogenize the mixture). Thereafter, the mixture was concentrated under reduced pressure. To remove the residual water the residue was taken up in toluene (50 ml) and volatiles were removed under reduced pressure (three times). The residue was purified by column chromatography (acetone:hexane = 1:2) to give after recrystallization from CH_2Cl_2/hexane the title compound (3.33 g, 46% on two steps) as a colorless solid.

R_f = 0.38, acetone:hexane = 1:2. ^{1}H NMR [300 MHz, $(CD_3)_2SO$] δ 9.19 (s, 1H), 9.07 (s, 1H), 6.87 (s, 2H), 6.65 (s, 1H), 3.04 (dd, *J* = 14.3, 6.8 Hz, 2H), 2.60 (t, *J* = 6.8 Hz, 2H), 1.36 (s, 9H).^{13}C NMR [75 MHz, $(CD_3)_2SO$] δ 155.5, 145.0, 144.9, 128.7, 118.8, 117.6, 111.3, 77.5, 40.4, 35.0, 28.3. MS (ESI): positive mode *m/z* = 687.1 ($[2M + Na]^+$), 354.0 ($[M + Na]^+$); MS (ESI): negative mode *m/z* = 663.1 ($[2M - H]^-$), 330.0 ($[M - H]^-$); ESI HRMS: calcd for $C_{13}H_{18}NO_4BrNa^+$: 354.03111; found: 354.0309; calcd for $C_{13}H_{17}NO_4Br^-$: 330.0346; found: 330.0345. Correct isotopic pattern.

NMR-Spectra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9.07
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6.65
3.07
3.05
3.03
3.01
2.63
2.60
2.58
2.50 Dimethyl Sulfoxide-d6
1.36
B (s)
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A (s)
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D (s)
6.65
C (s)
6.87
E (dd)
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F (t)
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G (s)
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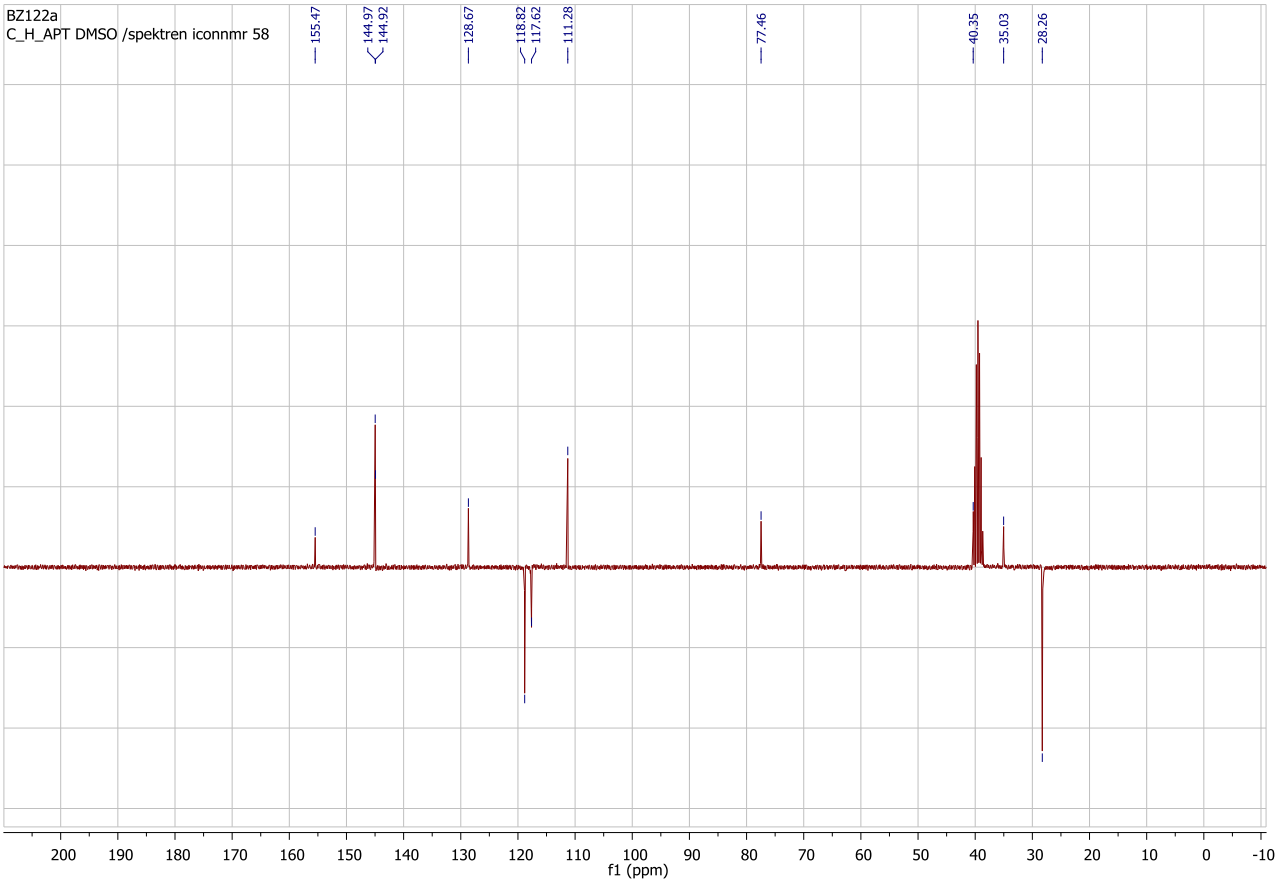
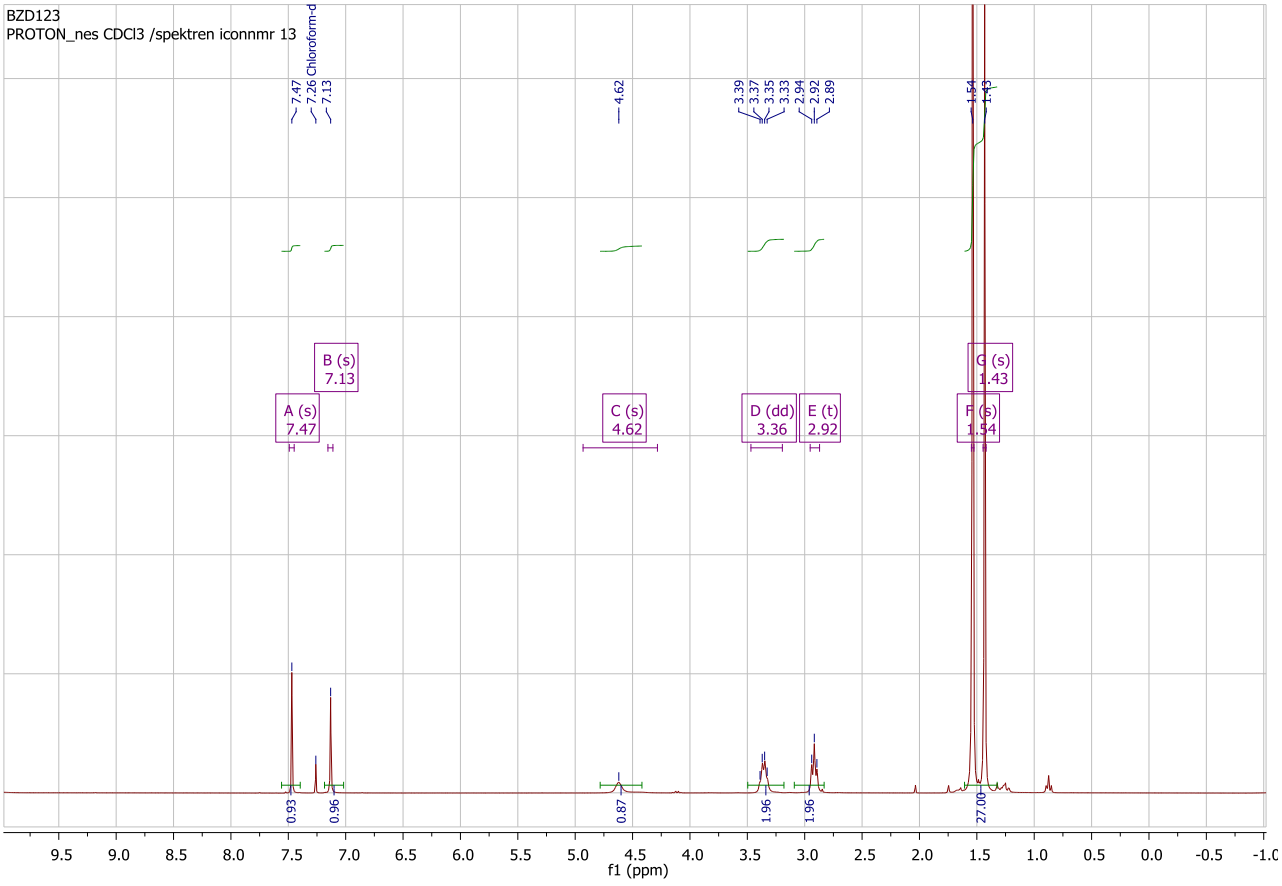
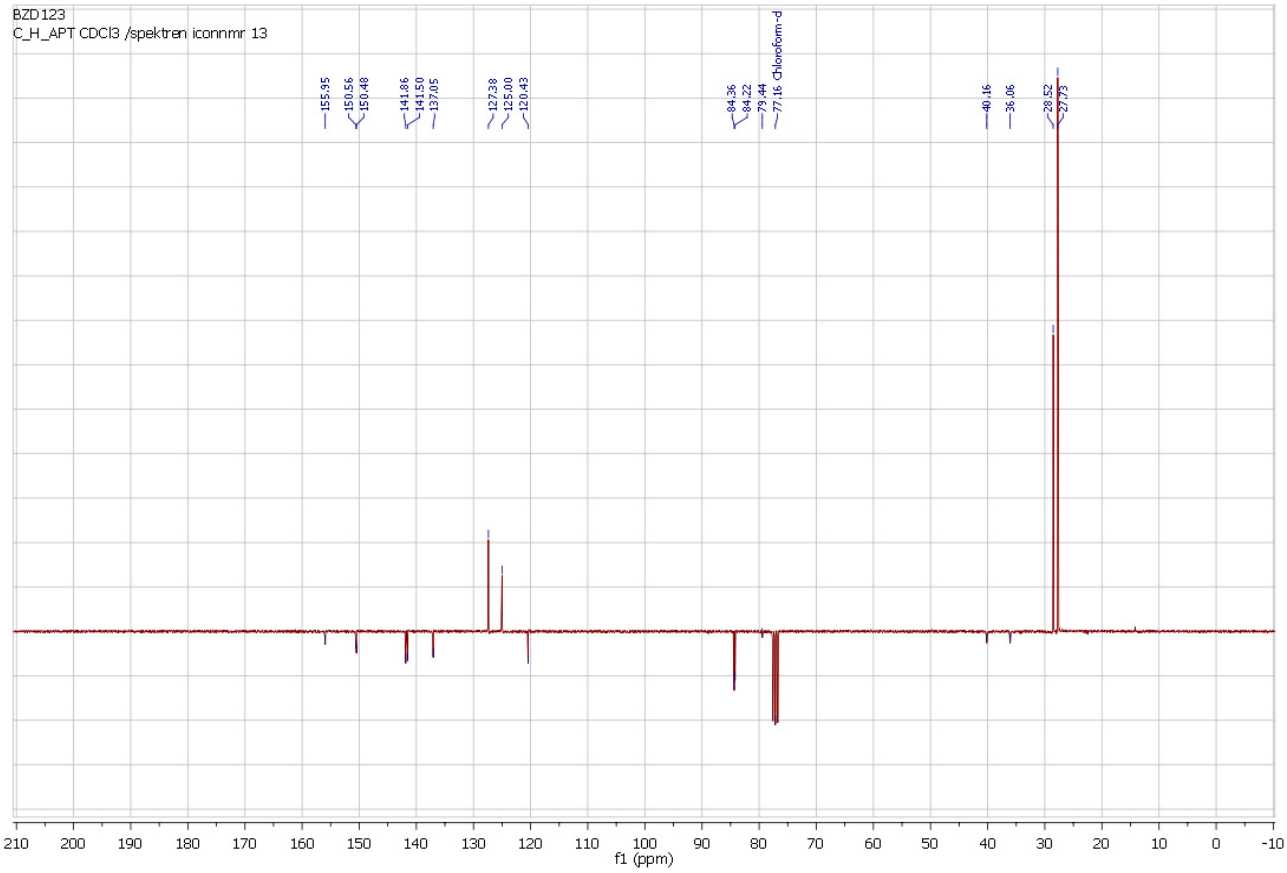
f1 (ppm)



***N,O,O*-tri-*tert*-Butyloxycarbonyl-6-bromodopamine:** Et_3N (3.4 mL, 2.46 g, 24.31 mmol) was slowly added to a solution of *N*-*tert*-butyloxycarbonyl-6-bromodopamine (3.23 g, 9.72 mmol) and Boc_2O (5.31 g, 24.31 mmol) in anhydrous DMF (30 mL) under Ar. The reaction mixture was stirred for a further 16 h and concentrated under reduced pressure. The residue was taken up in Et_2O (100 mL) and the resulting solution was washed with H_2O (8×40 mL), 1 M $NaHSO_4$ (3×30 mL), 5% $NaHCO_3$ (3×30 mL), brine (2×30 mL), dried and concentrated under reduced pressure. The residue was purified by column chromatography (EtOAc:hexane = 1:3) affording after recrystallization from Et_2O/hexane the title compound (3.83 g, 74%) as a colorless solid. R_f = 0.29, EtOAc:hexane = 1:3. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.47 (s, 1H), 7.13 (s, 1H), 4.62 (br, 1H), 3.36 (dd, *J* = 12.4, 6.5 Hz, 2H), 2.92 (t, *J* = 6.5 Hz, 2H), 1.54 (2×s, 2×9H), 1.43 (s, 9H). ^{13}C

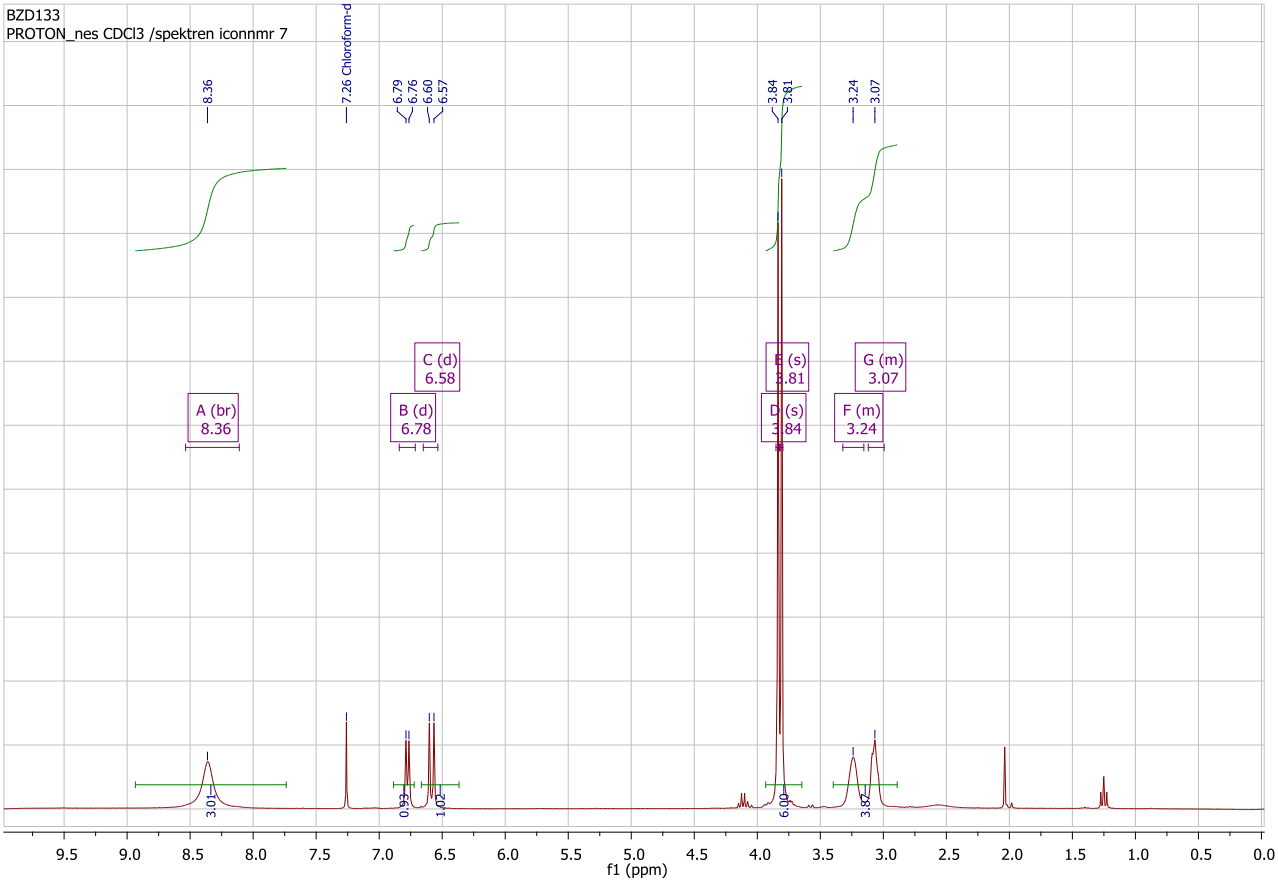
NMR (75 MHz, $CDCl_3$) δ 156.0, 150. 6, 150.5, 141.9, 141.5, 137.1, 127.4, 125.0, 120.4, 84.4, 84.2, 79.4, 40.2, 36.1, 28.5, 27.7 (×2). MS (ESI): positive mode *m/z* = 1087.3 ($[2M + Na]^+$), 570.1 ($[M + K]^+$), 554.1 ($[M + Na]^+$), 549.2 ($[M + NH_4]^+$), 532.2 ($[M + H]^+$); MS (ESI): negative mode *m/z* = 530.2 ($[M - H]^-$); ESI HRMS: calcd for $C_{23}H_{34}NO_8BrK^+$: 570.1099; found: 570.1096; calcd for $C_{23}H_{34}NO_8BrNa^+$: 556.1341; found: 556.1343; calcd for $C_{23}H_{33}NO_8Br^-$: 530.1395; found: 530.1383. Correct isotopic pattern.

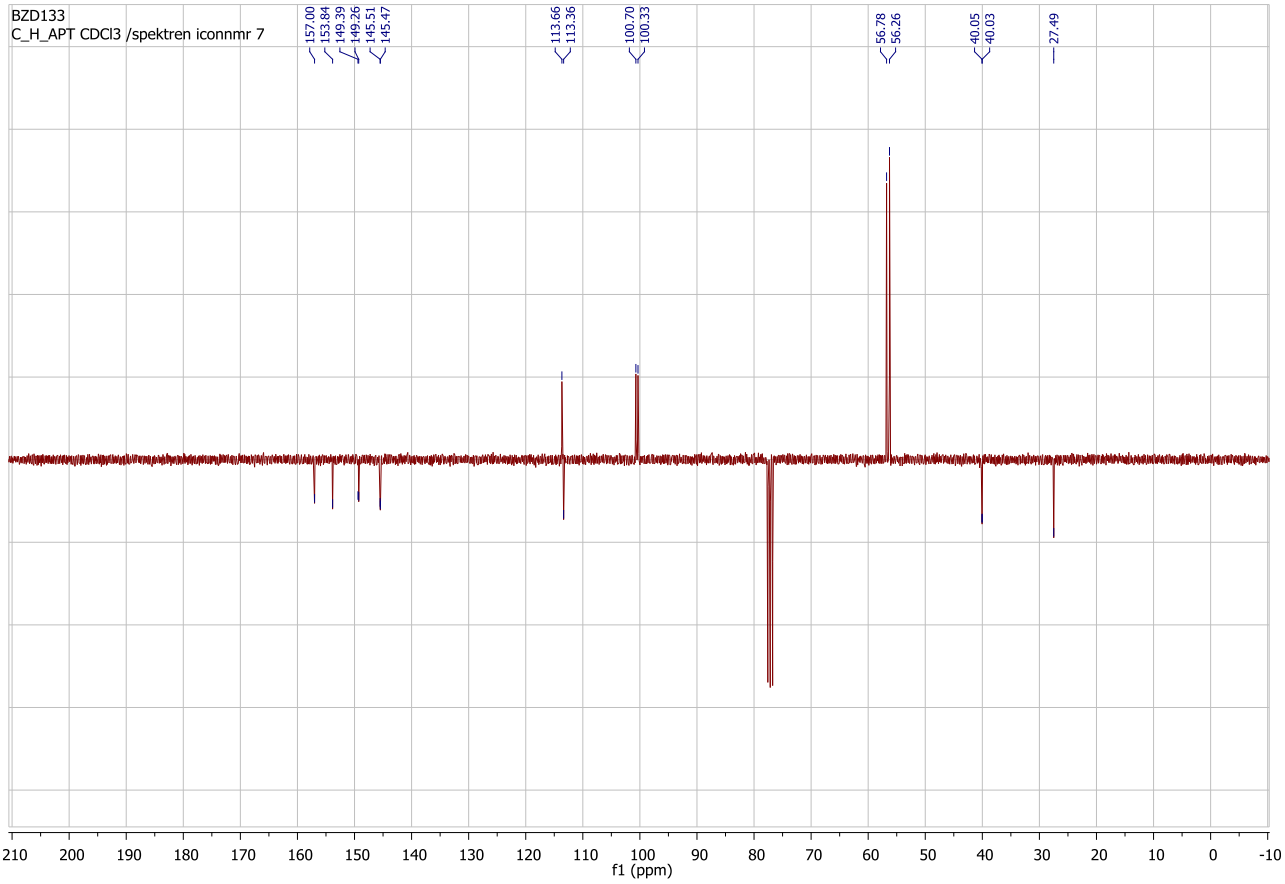




2-Fluoro-4,5-dimethoxyphenethylamine hydrochloride:[10] TFA (2.51 mL, 3.74 g, 32.79 mmol) was added dropwise to a stirred suspension of $NaBH_4$ (1.24 g, 32.79 mmol) in THF (30 mL) under Ar over 10 min. Afterwards, a solution of 2-fluoro-4,5-dimethoxybenzonitrile[6] (0.63 g, 3.22 mmol) in THF (10 mL) was added via a cannula and the reaction mixture was stirred for further 16 h. Thereafter, the mixture was ice-cooled and quenched by the careful addition of water. The resulting mixture was concentrated under reduced pressure to give the residue which was extracted with CH_2Cl_2 (5×30 mL). The extract was washed with H_2O (10 mL) and brine (2×20 mL), dried, filtered and concentrated under reduced pressure. The residue was dissolved in EtOAc (10 mL) and the resulting solution was treated with 2 M HCl in EtOAc (5 mL). The solution was evaporated to dryness under reduced pressure and the residual oil was taken in EtOAc (30 mL) and heated to reflux for a short time using a heat gun. After cooling to ambient temperature, the precipitate was

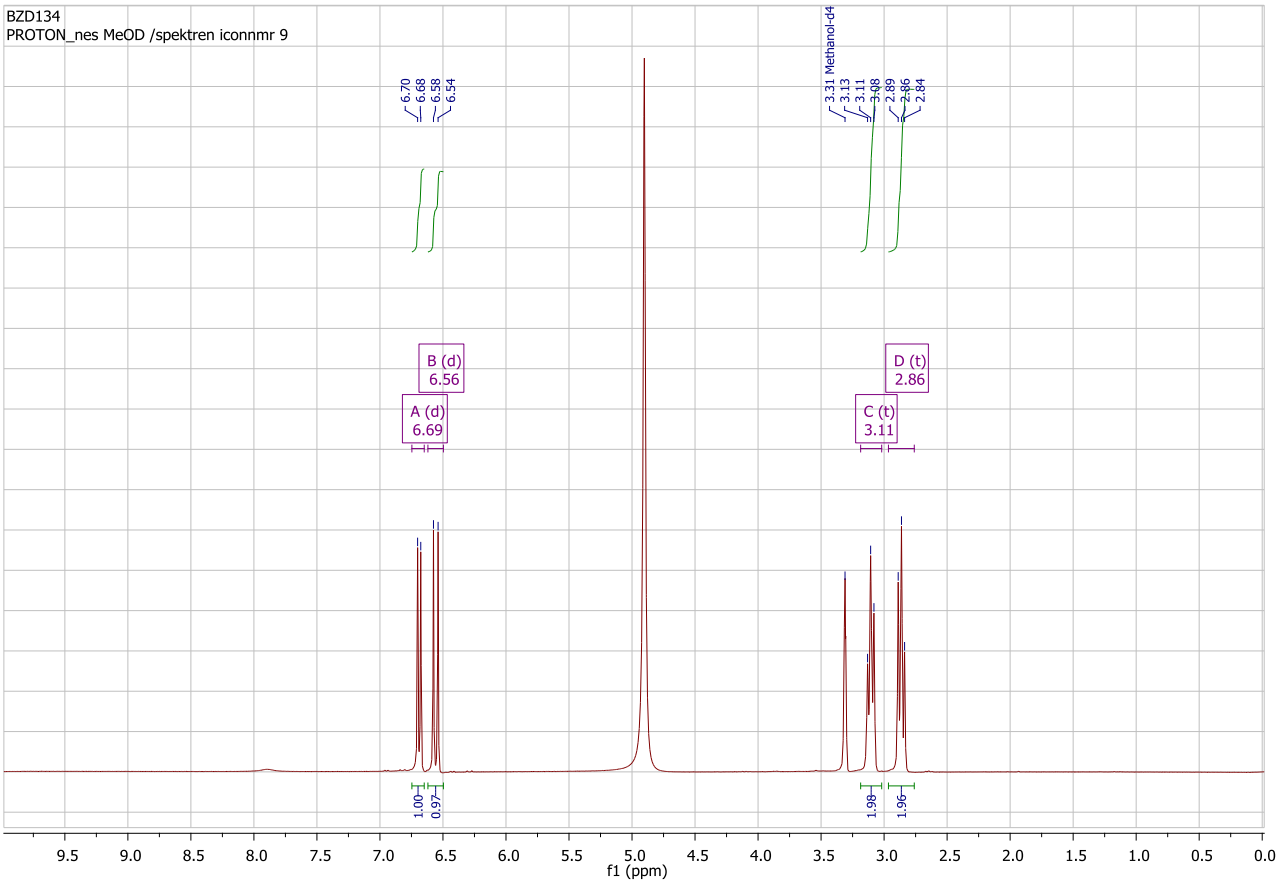
collected by filtration. The filter cake was washed with EtOAc (10 mL) and dried to give the title compound (0.45 g, 59%) as a rose solid. ^{1}H NMR (300 MHz, $CDCl_3$) 8.36 (br, 3H), 6.78 (d, J = 7.1 Hz, 1H), 6.58 (d, J = 11.1 Hz, 1H), 3.84 (s, 3H), 3.81 (s, 3H), 3.32–3.15 (m, 2H), 3.12–2.99 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$) δ 155.4 (d, J = 237 Hz), 149.3 (d, J = 9.8 Hz), 145.5 (d, J = 3.0 Hz), 113.7, 113.4, 100.5 (d, J = 27.8 Hz), 56.8, 56.3, 40.0 (d, J = 1.5 Hz), 27.5.

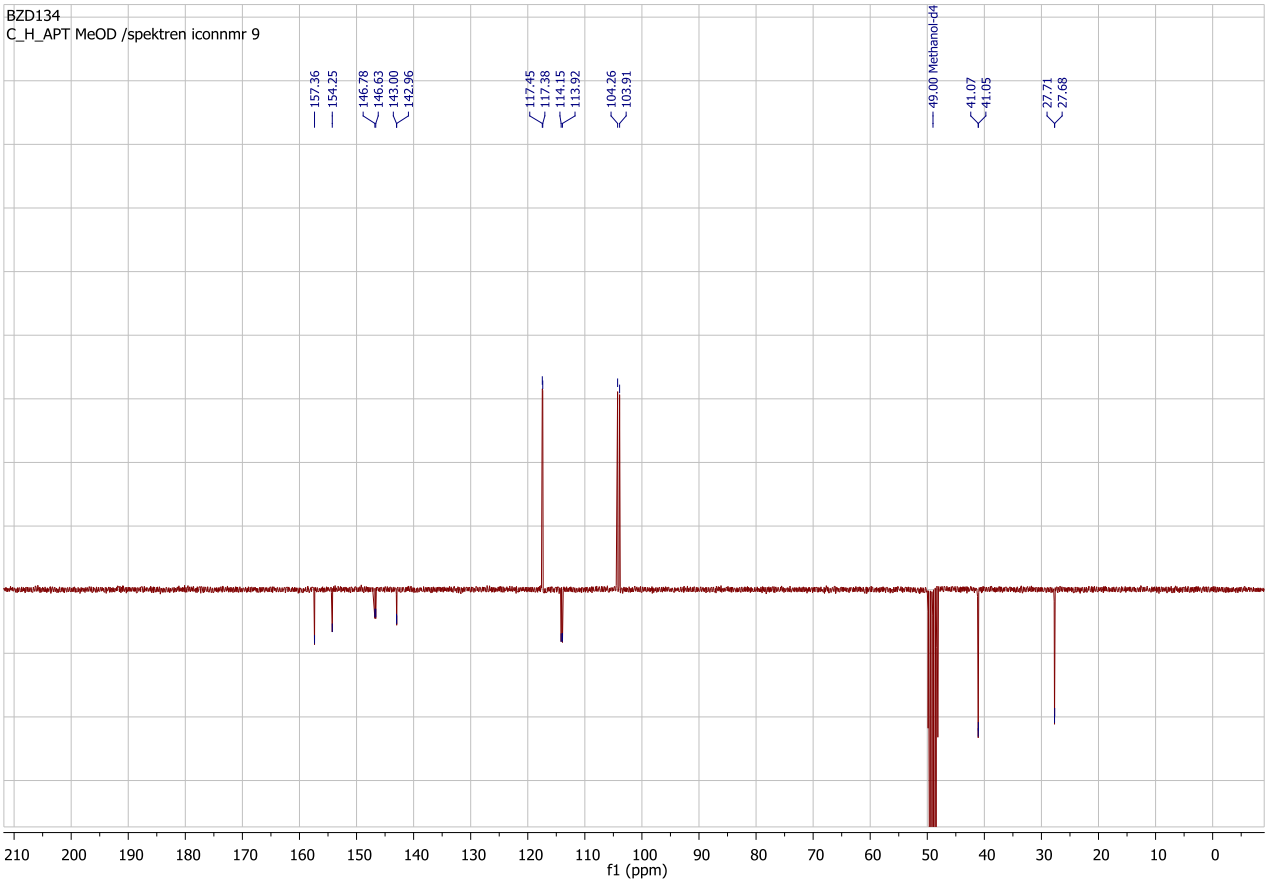




6-Fluorodopamine hydrobromide (1c·HBr):[11] A solution of 2-fluoro-4,5-dimethoxy-phenethylamine hydrochloride (0.42 g, 1.78 mmol) in 48% HBr (4 mL) was heated under Ar at 140 °C for 90 min. Thereafter, the reaction mixture was cooled to ambient temperature and placed in a refrigerator. After 3 h the precipitated solid was collected by filtration and washed with cold water (5 mL) to give the first crop of **3c**·HBr (110 mg, 25%) as an off-white solid. The mother liquor was concentrated under reduced pressure and the residue was triturated with Et_2O to give the second drop of **3c**·HBr (0.29 g, overall yield: 89%). ^{1}H NMR (CD_3OD, 300 MHz): δ 6.69 (d, *J* = 7.5 Hz, 1H), 6.56 (d, *J* = 11.0 Hz, 1H), 3.11 (t, *J* = 7.6 Hz, 2H), 2.86 (t, *J* = 7.6 Hz, 2H); ^{13}C NMR (CD_3OD, 75 MHz): δ 155.8 (d, *J* = 233.3 Hz), 146.7 (d, *J* = 12 Hz), 143.0 (d, *J* = 2.3 Hz),

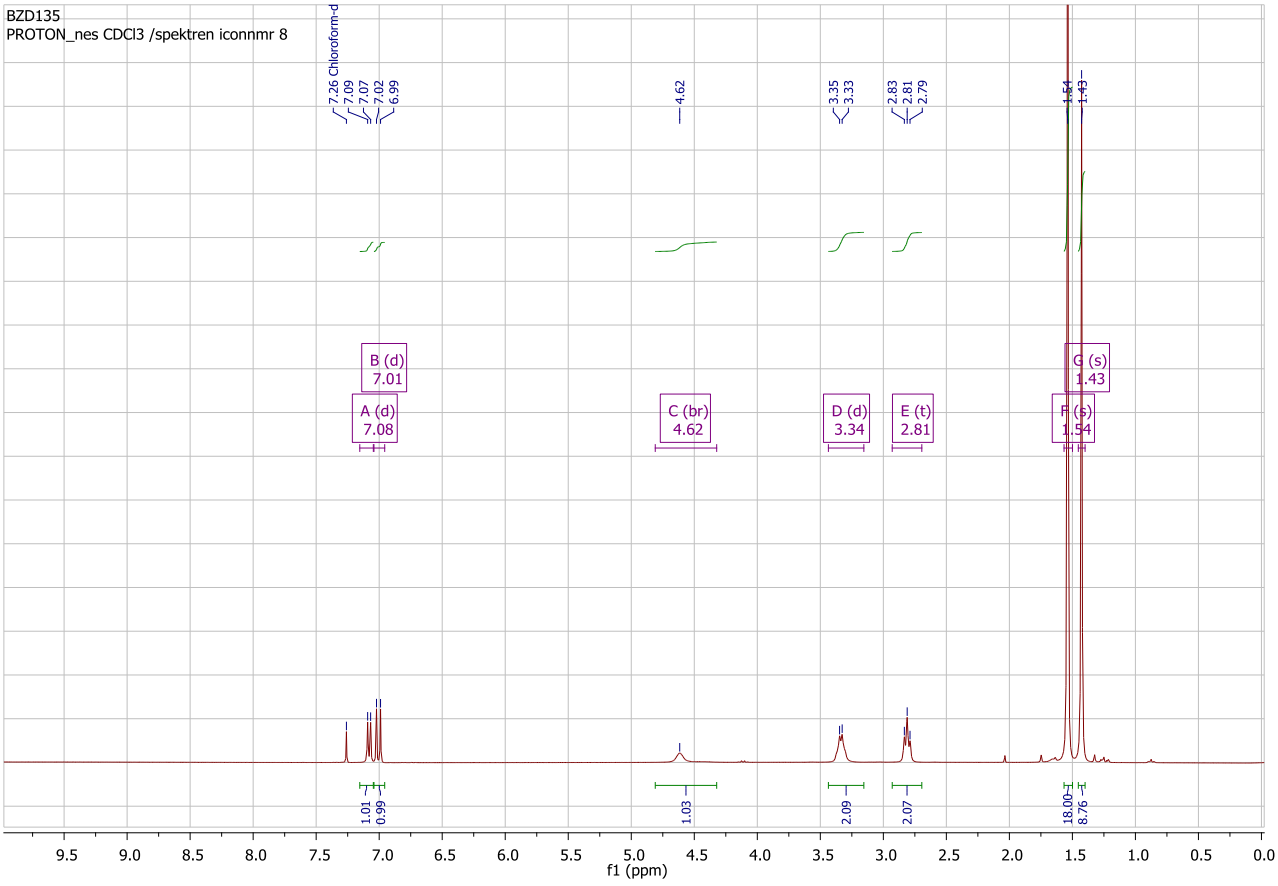
117.5 (d, *J* = 5.3 Hz), 114.0 (d, *J* = 17.3 Hz), 104.1 (d, *J* = 26.3 Hz), 41.1 (d, *J* = 1.5 Hz), 27.7 (d, *J* = 2.3 Hz).

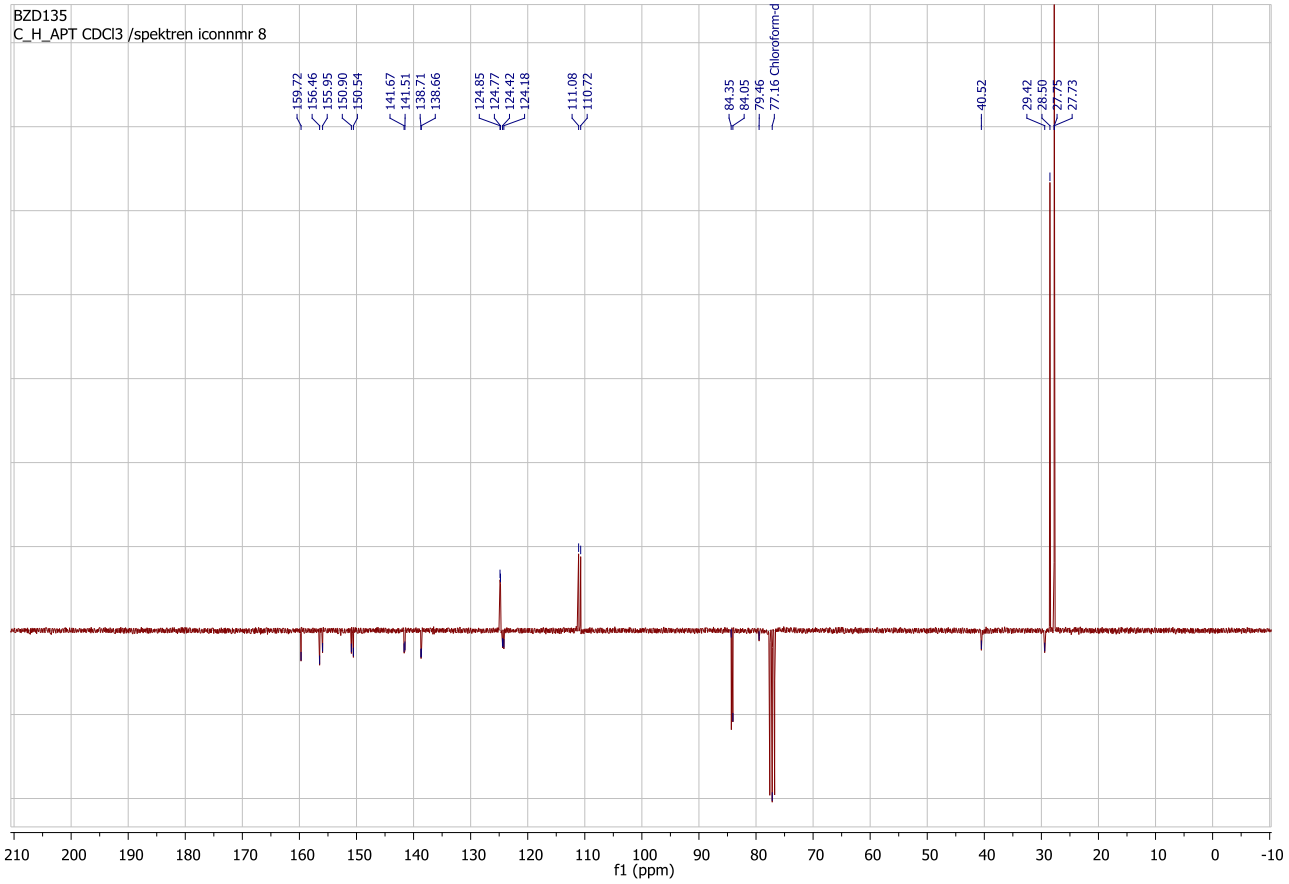




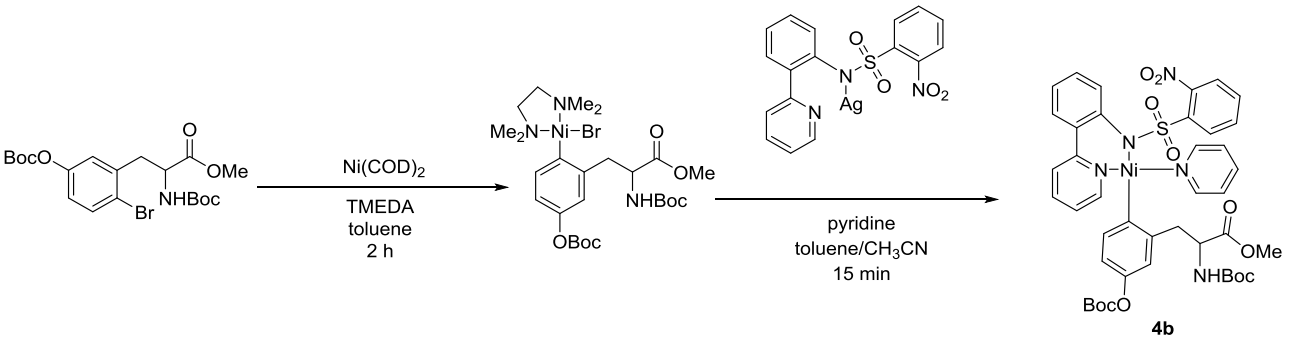
***N,O,O*-tri-*tert*-Butyloxycarbonyl-6-fluorodopamine (3c):** Et_3N (0.95 mL, 0.69 g, 6.82 mmol) was slowly added to a solution of **1c**·HBr (0.29 g, 1.15 mmol) and Boc_2O (1.6 g, 7.33 mmol) in anhydrous DMF (10 mL) under Ar. Thereafter, the reaction mixture was stirred for a further 16 h and concentrated under reduced pressure. The residue was taken up in Et_2O (100 mL) and the resulting solution was washed with H_2O (8×40 mL), 1 M $NaHSO_4$ (3×30 mL), 5% $NaHCO_3$ (3×30 mL), brine (2×30 mL), dried and concentrated under reduced pressure. The residue was purified by column chromatography (EtOAc:hexane = 1:3). The product fractions were concentrated under reduced pressure. The residual colorless oil was triturated with pentane to give **3c** (0.44 g, 62%) as a colorless solid. R_f = 0.24, EtOAc:hexane = 1:3. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.08 (d, *J* = 7.1 Hz, 1H), 7.01 (d, *J* = 9.6 Hz, 1H), 4.62 (br, 1H), 3.34 (q, *J* = 6.4 Hz, 2H), 2.81 (t, *J* =

6.7 Hz, 2H), 1.54 (s, 18H), 1.43 (s, 9H). ^{13}C NMR (75 MHz, $CDCl_3$) δ 158.1 (d, *J* = 244.5 Hz), 156.0, 150.9, 150.5, 141.6 (d, *J* = 12 Hz), 138.7 (d, *J* = 3.8 Hz), 124.8 (d, *J* = 6.0 Hz), 124.3 (d, *J* = 18 Hz), 111.1 (d, *J* = 27 Hz), 84.4, 84.1, 79.5, 40.5, 29.4, 28.5, 27.75, 27.73. MS (ESI): positive mode *m/z* = 494.2 ($[M + Na]^+$), 489.3 ($[M + NH_4]^+$); MS (ESI): negative mode *m/z* = 470.2 ($[M - H]^-$); ESI HRMS: calcd for $C_{23}H_{34}NO_8BrNa^+$: 556.1341; found: 556.1343; calcd for $C_{23}H_{34}NO_8Na^+$: 494.2161; found: 494.2158; calcd for $C_{23}H_{33}NO_8F^-$: 470.2196; found: 470.2177.





Ni-aryl complex 4b:

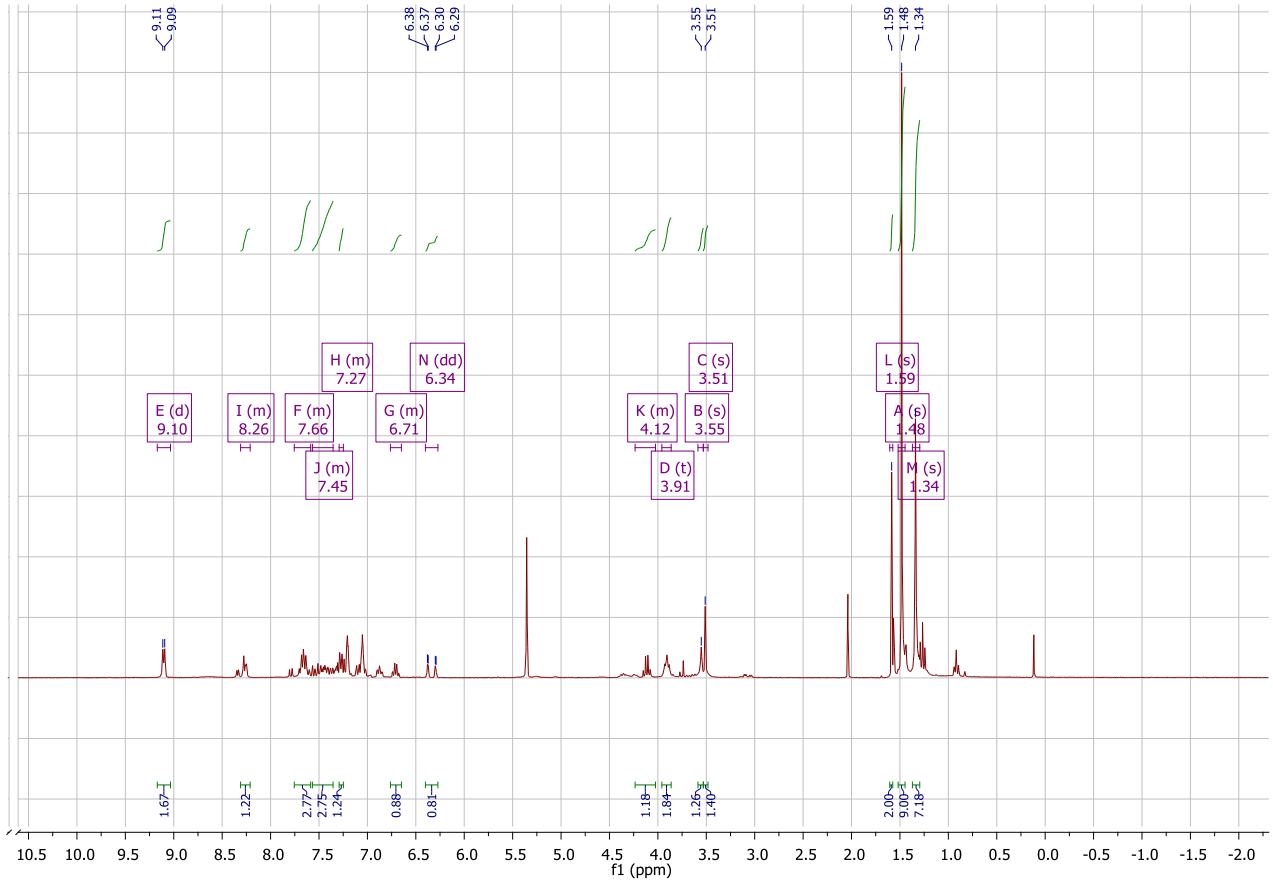


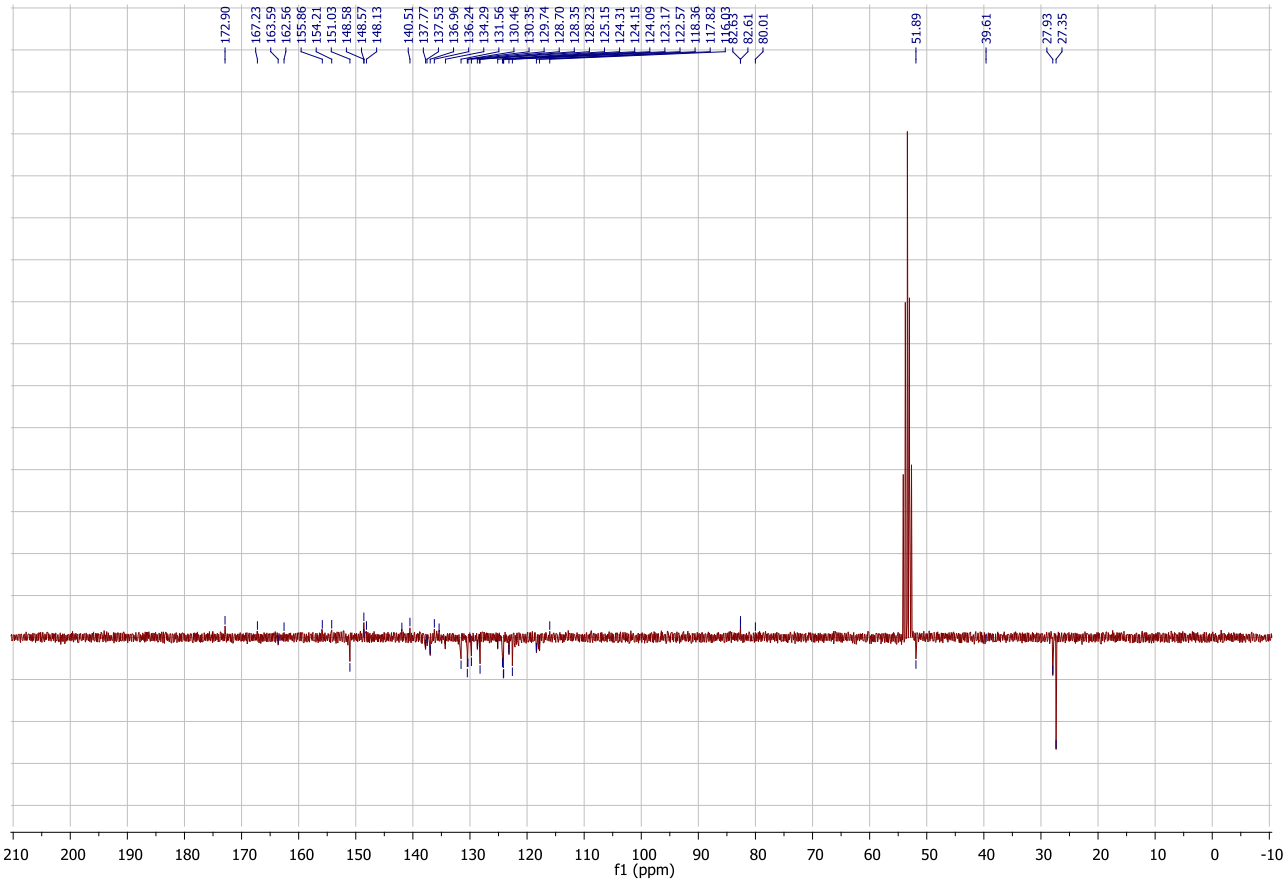
To a solution of TMEDA (0.386 mL, 0.297 g, 2.56 mmol) and methyl *N*,*O*-di-*tert*-butyloxycarbonyl-6-bromo-*m*-tyrosinate (1.3 g, 2.72 mmol) in toluene (8 mL) was added $Ni(COD)_2$ (0.72 g, 2.62mmol) and the mixture was stirred at room temperature for a further 2 h. The solution was concentrated under reduced pressure. Pentane (16 mL) was added to the residue and the formed

precipitate was filtered off, washed with pentane (3×5 mL) and dried *in vacuo* affording [Boc-6-*m*-Tyr(Boc)OMe]Ni(TMEDA)Br (1.04 g, 71%) as a yellow solid, which was directly used for the next step without any further purification.

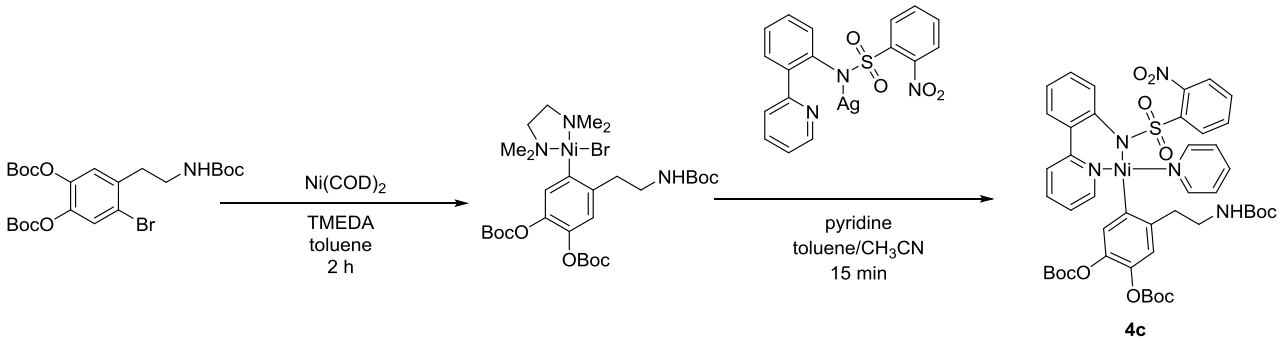
To 2-(2-pyridinyl)phenyl-2-nitrobenzenesulfonamide silver(I) (571 mg, 1.24 mmol) and nickel aryl bromide complex [Boc-6-*m*-Tyr(Boc)OMe]Ni(TMEDA)Br (715 mg, 1.28 mmol) was added a solution of pyridine (105 µL, 104 mg, 1.31 mmol) in toluene (8 mL) followed by acetonitrile (2.0 mL). After stirring for 15 min the mixture was filtered through a glass frit, and the filter cake was extracted with dichloromethane (3×5 mL). Combined filtrates were concentrated under reduced pressure. The residue was purified by column chromatography [hexane/EtOAc 1:6 (0.5% Et_3N)] and recrystallization from CH_2Cl_2/pentane to afford **4b** (0.43 g, 38%) as a yellow solid which contained about 30 mol. % (2.6 weight %) ethyl acetate. Broadness of signals in ^{1}H- and ^{13}C-NMR spectra was already observed previously for the similar nickel complex **4a**.[12] Conformational isomers were observed in the ^{1}H-spectra, which are possibly due to slow rotation about bonds as seen before for such complexes.[12]

^{1}H NMR (300 MHz, CD_2Cl_2) δ 9.10 (d, *J* = 6.0 Hz, 2H), 8.31 – 8.21 (m, 1H), 7.76 – 7.59 (m, 3H), 7.57 – 7.35 (m, 3H), 7.29 – 7.25 (m, 1H), 6.76 – 6.65 (m, 1H), 6.34 (dd, *J* = 24.4, 2.4 Hz, 1H), 4.12 (m, 1H), 3.91 (t, *J* = 6.6 Hz, 2H), 3.55, 3.51 (2×s, 3H), 1.48 (s, 9H); 1.34, 1.59 (2×s, 9H). ^{13}C NMR (75 MHz, CD_2Cl_2) δ 172.9, 167.2, 163.6, 162.6, 155.9, 154.2, 151.0, 148.6, 148.6, 148.1, 141.9, 140.5, 137.8, 137.5, 137.0, 136.2, 135.4, 134.3, 131.6, 130.5, 130.4, 130.0, 129.7, 128.7, 128.4, 128.2, 125.2, 124.3, 124.2, 124.1, 123.2, 122.6, 118.4, 117.8, 116.0, 82.6, 80.0, 51.9, 39.6, 27.9, 27.4. MS (ESI): positive mode *m/z* = 829.2 ([M – Py + Na]$^+$), 807.2 ([M – Py + H]$^+$); ESI HRMS: calcd for $C_{37}H_{40}N_4O_{11}SNiNa^+$: 829.1660; found: 829.1650. Correct isotopic pattern.


9.11
9.09
6.38
6.37
6.30
6.29
3.55
3.51
1.59
1.48
1.34
H (m)
7.27
N (dd)
6.34
C (s)
3.51
L (s)
1.59
E (d)
9.10
I (m)
8.26
F (m)
7.66
G (m)
6.71
K (m)
4.12
B (s)
3.55
A (s)
1.48
J (m)
7.45
D (t)
3.91
M (s)
1.34
1.67
1.22
2.77
2.75
1.24
0.89
0.81
1.18
1.84
1.26
1.40
2.00
9.00
7.18
10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0 -0.5 -1.0 -1.5 -2.0
f1 (ppm)



Ni-aryl complex 4c:

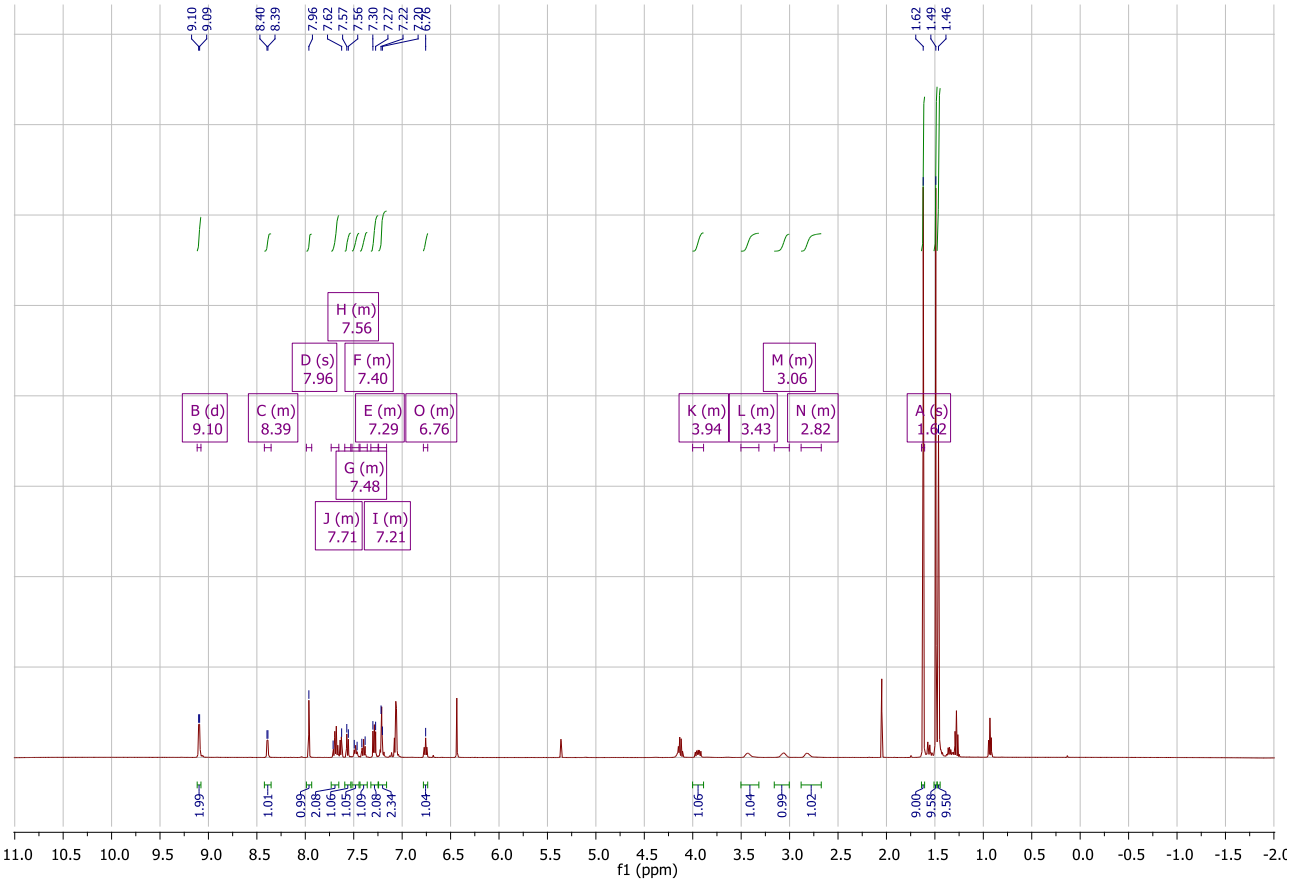


To a solution of TMEDA (0.291 mL, 0.224 g, 1.91 mmol) and *N*,*O*,*O*-tri-*tert*-butyloxycarbonyl-6-bromodopamine (1.01 g, 1.90mmol) in toluene (10 mL) was added $Ni(COD)_2$ (0.53 g, 1.93 mmol) and the mixture was stirred at room temperature for a further 2 h. The solution was concentrated under reduced pressure. Pentane (16 mL) was added to the residue and the formed precipitate was

filtered off, washed with pentane (3×5 mL) and dried *in vacuo* affording [Boc-6-DA(Boc)$_2$]Ni(TMEDA)Br (1.1 g, 82%) as a peach solid, which was directly used for the next step without any further purification.

To 2-(2-pyridinyl)phenyl-2-nitrobenzenesulfonamide silver(I) (0.53 g, 1.15 mmol) and nickel aryl bromide complex Boc-6-DA(Boc)$_2$]Ni(TMEDA)Br (0.8 mg, 1.13 mmol) was added a solution of pyridine (185 µL, 181 mg, 2.288 mmol) in toluene (12 mL) followed by addition of acetonitrile (3 mL). After stirring for 15 min the mixture was filtered through a glass frit, and the filter cake was extracted with dichloromethane (3×5 mL). Combined filtrates were concentrated under reduced pressure. The residue was purified by column chromatography [hexane/EtOAc 1:3 (0.5% Et_3N)] and recrystallization from CH_2Cl_2/pentane (two times) to afford **4c** (0.81 g, 70%) as a brown solid which contained 46 mol % (3.5 weight %) pentane and 35 mol % (3 weight %) ethyl acetate. Broadness of signals in ^{1}H- and ^{13}C-NMR spectra was already observed previously for the similar nickel complex **4a**.[12]

^{1}H NMR (600 MHz, CD_2Cl_2) δ 9.10 (d, *J* = 5.1 Hz, 2H), 8.42 – 8.35 (m, 1H), 7.96 (s, 1H), 7.73–7.65 (m, 2H), 7.59–7.53 (m, 1H), 7.52–7.44 (m, 1H), 7.44–7.36 (m, 1H), 7.32–7.25 (m, 2H), 7.24–7.16 (m, 2H), 6.78–6.74 (m, 1H), 4.00–3.89 (m, 1H), 3.50–3.32 (m, 1H), 3.16–3.00 (m, 1H), 2.88–2.67 (m, 1H), 1.62 (s, 9H), 1.49 (s, 9H), 1.46 (s, 9H). ^{13}C NMR (126 MHz, CD_2Cl_2) δ 155.7, 155.3, 153.4, 151.4, 151.2, 151.03, 151.00, 146.8, 141.8, 140.6, 138.8, 138.7, 137.8, 137.1, 136.1, 135.8, 131.7, 130.5, 130.0, 128.3, 128.2, 127.2, 124.5, 124.2, 123.0, 122.6, 122.3, 118.7, 83.1, 83.0, 78.6, 41.4, 39.1, 28.2, 27.5, 27.3. MS (ESI): positive mode *m/z* = 887.2 ([M – Py + Na]$^+$), 865.2 ([M – Py + H]$^+$); MS (ESI): negative mode *m/z* = 882.3 ([M – H + NH_3]$^-$), 863.2 ([M – H]$^-$); ESI HRMS: calcd for $C_{40}H_{46}N_4O_{12}NiSNa^+$: 887.2079; found: 887.2080; calcd for $C_{40}H_{50}N_5O_{12}NiS^+$: 882.2525; found: 882.2515; calcd for $C_{40}H_{47}N_4O_{12}NiS^+$: 865.2259; found: 865.2251; calcd for $C_{40}H_{45}N_4O_{12}NiS^-$: 863.2103; found: 863.2091. Correct isotopic pattern.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9.09
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7.56
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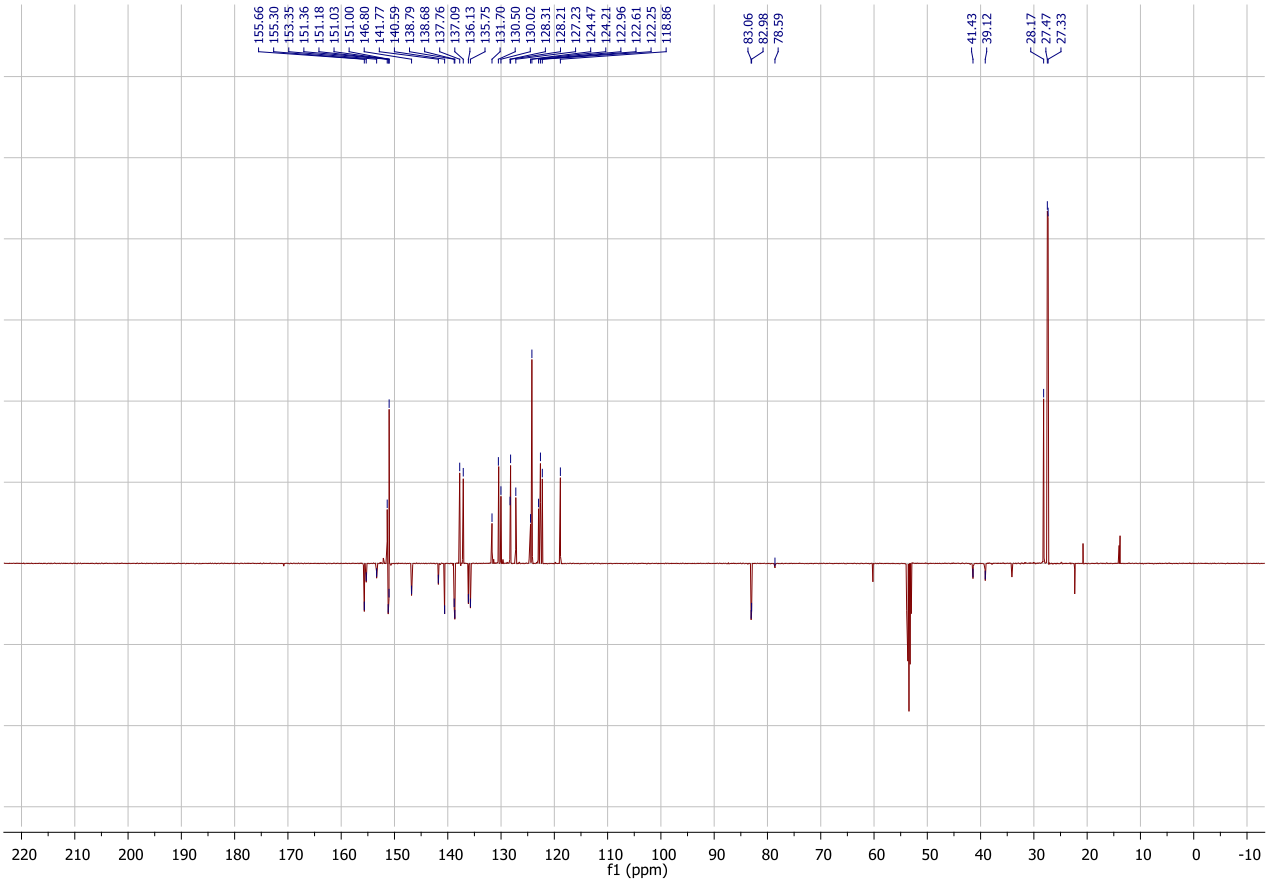
7.27
7.22
7.20
6.78
1.62
1.49
1.46
H (m)
7.56
D (s)
7.96
F (m)
7.40
M (m)
3.06
B (d)
9.10
C (m)
8.39
E (m)
7.29
O (m)
6.76
K (m)
3.94
L (m)
3.43
N (m)
2.82
A (s)
1.62
G (m)
7.48
J (m)
7.71
I (m)
7.21
1.99
1.01
0.99
2.08
1.06
1.05
1.09
2.08
2.34
1.04
1.06
1.04
0.99
1.02
9.00
9.58
9.50
11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0 -0.5 -1.0 -1.5 -2.0
f1 (ppm)



Radiochemistry

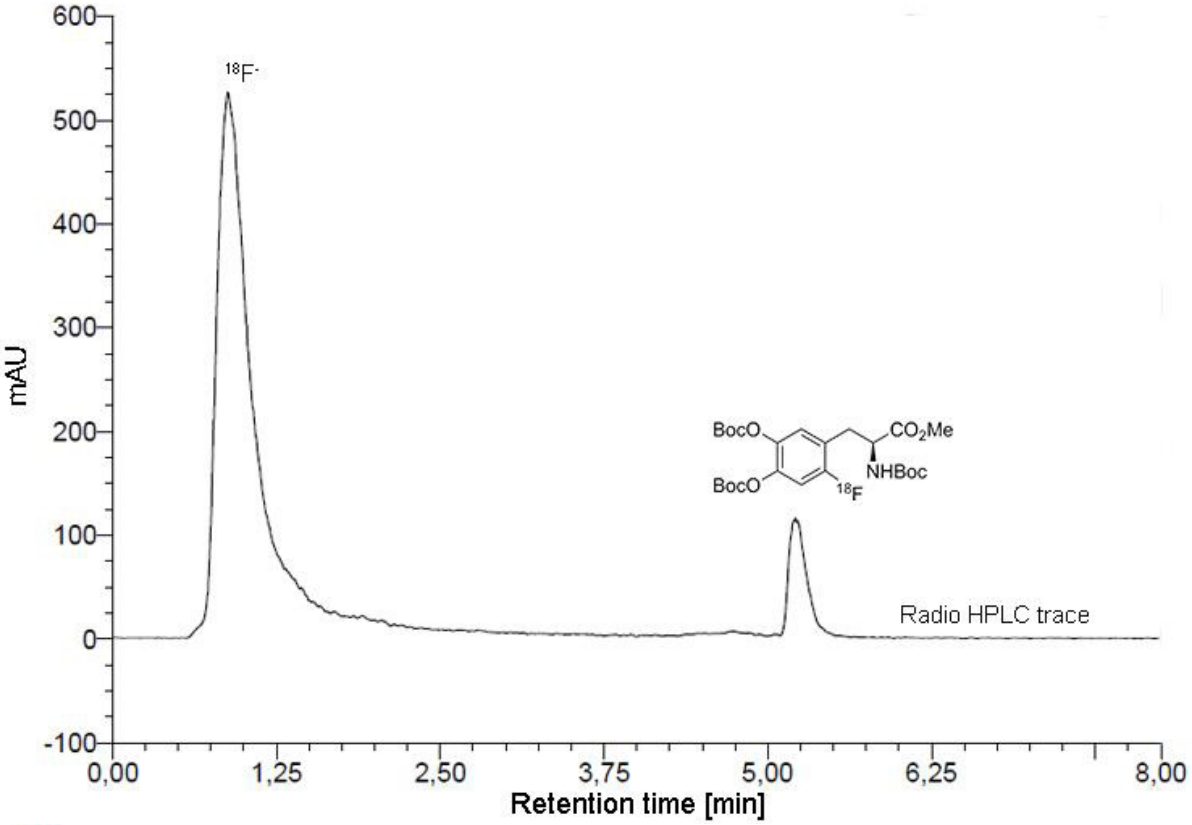
Synthesis of [^{18}F]3c – Optimization study – General procedure 1 (GP1): A solution of [^{18}F]KF/18-crown-6 (50–500 MBq; *vide supra*) in the corresponding solvent (900 µL) was added to a mixture of Ni-complex precursor **4c** and oxidant **2**, weighted in a glove box and the mixture was stirred for a given time at ambient temperature. Thereafter, water (5 mL) was added, the mixture was vigorously stirred for 1 min and analyzed by radio-HPLC (Conditions A: [^{18}F]**3c**: t_R=5.3 min).

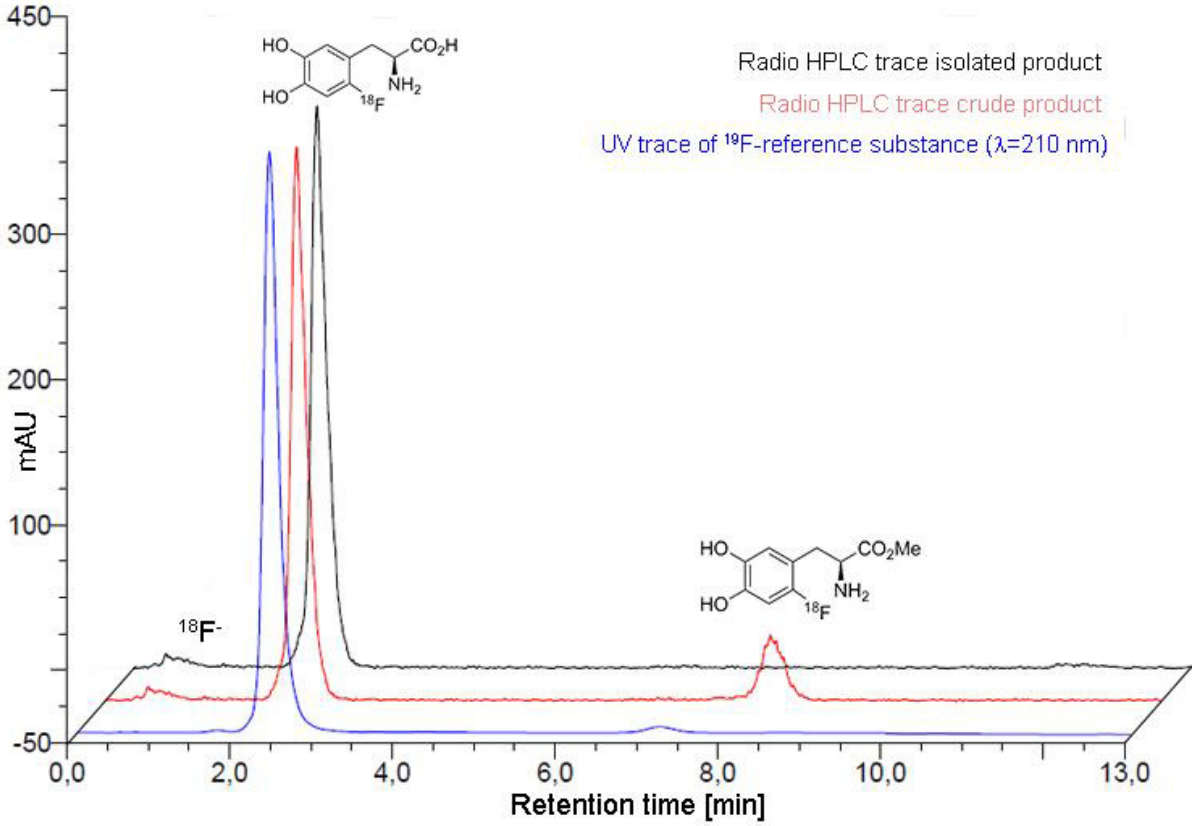
Synthesis of [^{18}F]1a-c with SPE purification of intermediates [^{18}F]3a-c – General Procedure 2 (GP2): A solution of [^{18}F]KF/18-crown-6 (50–500 MBq; *vide supra*) in MeCN (900 µL) was added to a mixture of the corresponding Ni-complex precursor **4a-c** (5 µmol) and oxidant **2** (4.7 mg, 6.5 µmol), weighted in a glove box and the mixture was stirred for 5 min at ambient temperature.

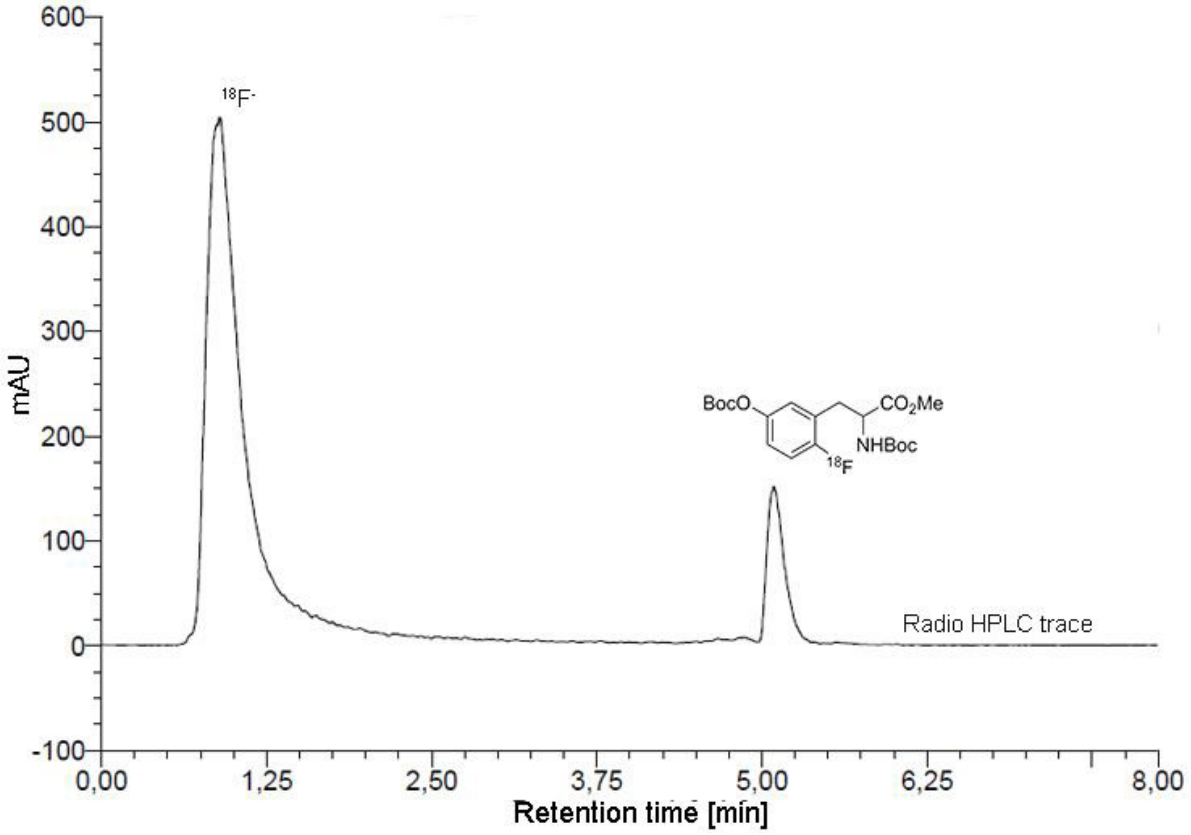
Thereafter, water (5 mL) was added; the mixture was vigorously stirred for 1 min and passed through a polymer-RP cartridge (StrataX, Phenomenex). The cartridge was washed with 40% MeCN (2 mL) and the partially purified radiolabeled intermediates [^{18}F]**3a-c** were eluted with MeCN (0.5 mL). MeCN was evaporated under a gentle stream of argon. The residue was taken up in 37% HCl (200 µL) and the reaction mixture was stirred at 130° C for 10 min. All volatiles were removed under a gentle stream of argon and the residue was dissolved in 0.02 M sodium phosphate buffer (500 µL, pH 2.5). PET-tracers [^{18}F]**1a-c** were isolated by semi-preparative HPLC (system B: [^{18}F]**1a**: t_R=3.2 min; [^{18}F]**1b**: t_R=5.4 min; [^{18}F]**1c:** t_R=3.6 min) and obtained as ready to use solutions.

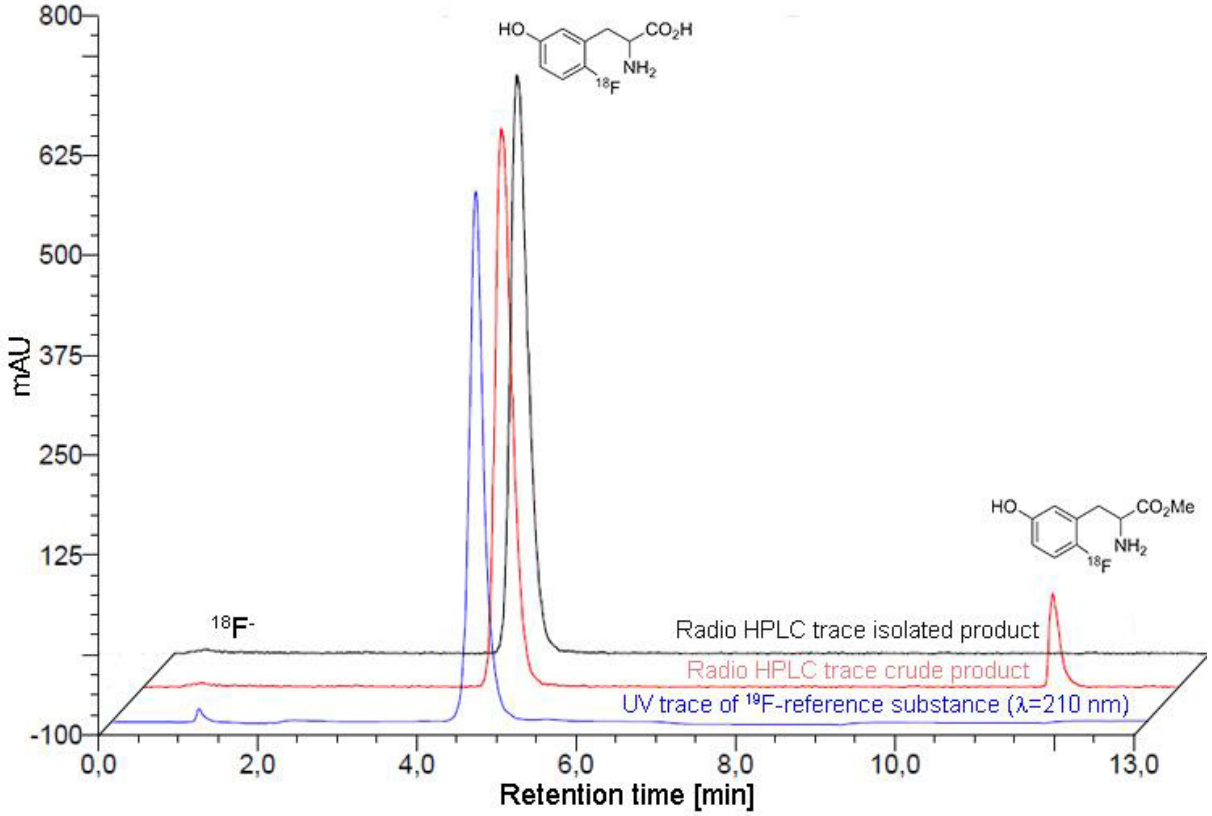
One-pot synthesis of [^{18}F]1a-c – General Procedure 3 (GP3): A solution of [^{18}F]KF/18-crown-6 (0.05–40 GBq; *vide supra*) in MeCN (900 µL) was added to a mixture of the corresponding Ni-complex precursor **4a-c** (5 µmol) and oxidant **2** (4.7 mg, 6.5 µmol), weighted in a glove box and the mixture was stirred for 5 min at ambient temperature. The reaction mixture was concentrated under reduced pressure. The residue was taken up in 37% HCl (200 µL) and the reaction mixture was stirred at 130° C for 10 min. Acetone (1 mL) was added and all volatiles were removed under a gentle stream of argon (two times). The residue was dissolved in 0.02 M sodium phosphate buffer (500 µL, pH 2.5). PET-tracers [^{18}F]**1a-c** were isolated by semi-preparative HPLC (system B) and obtained as ready to use solutions.

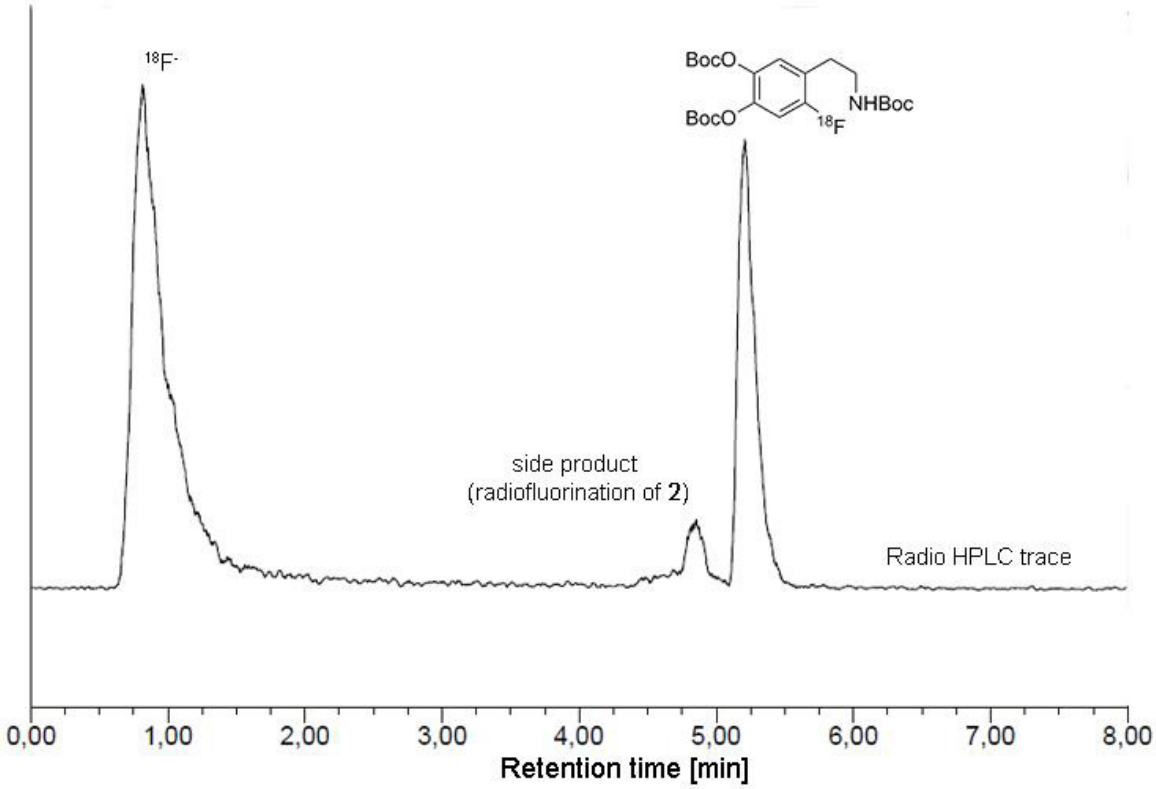
HPLC-chromatograms

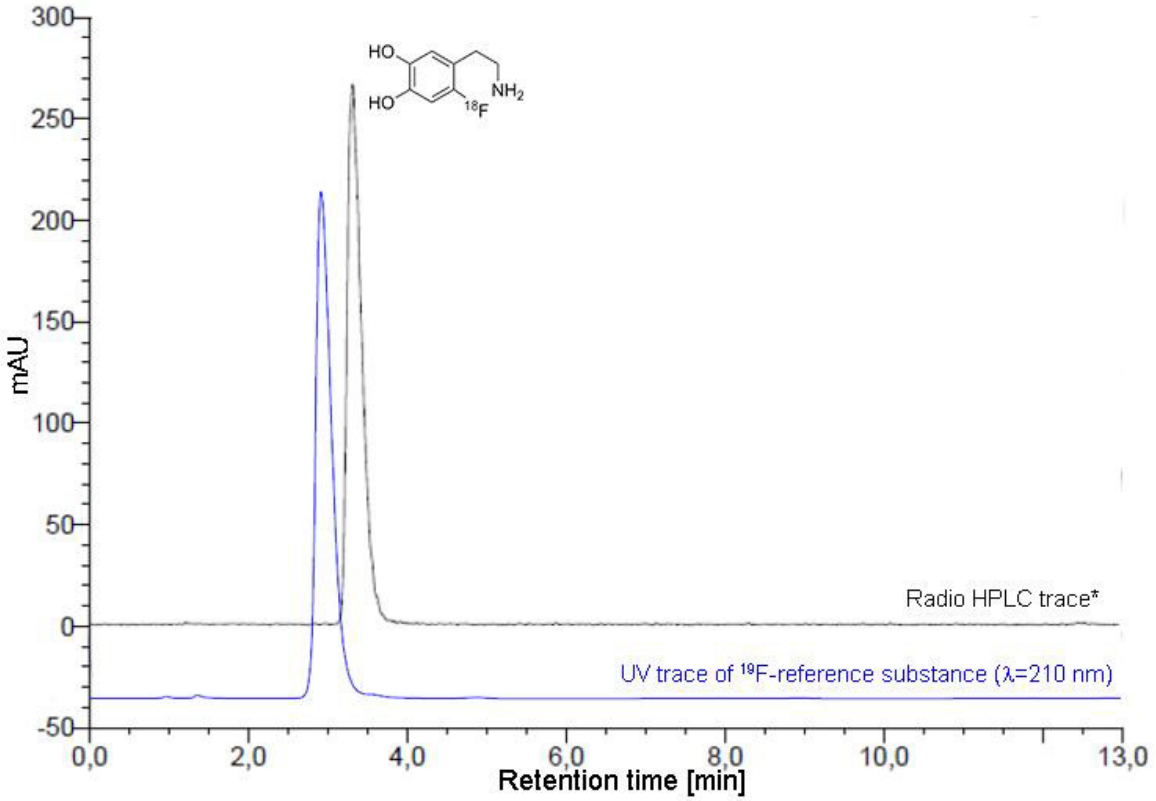





600
500
400
300
200
100
0
-100
mAU
$^{18}F^-$
BocO
CO_2Me
NHBoc
^{18}F
Radio HPLC trace
0,00
1,25
2,50
3,75
5,00
6,25
8,00
Retention time [min]


800
625
500
375
250
125
-100
mAU
HO
CO_2H
NH_2
^{18}F
HO
CO_2Me
NH_2
^{18}F
$^{18}F^-$
Radio HPLC trace isolated product
Radio HPLC trace crude product
UV trace of ^{19}F-reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
13,0
Retention time [min]

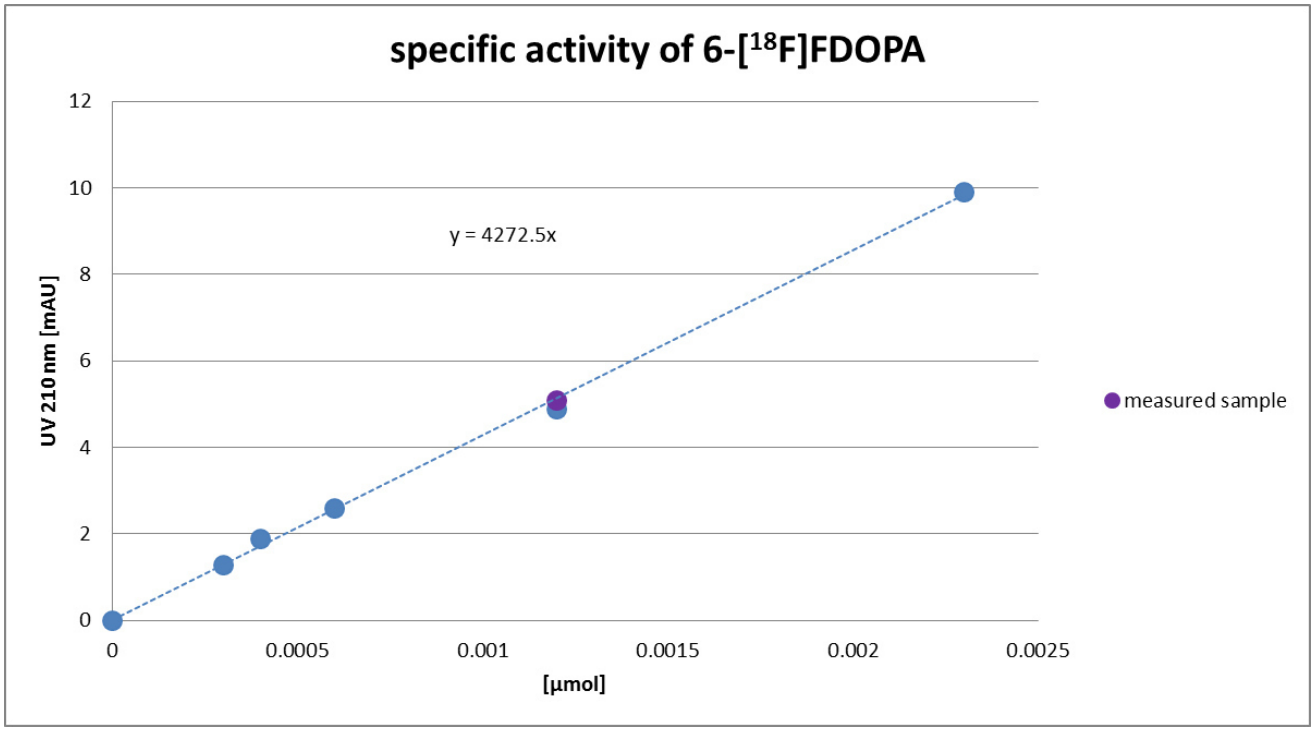


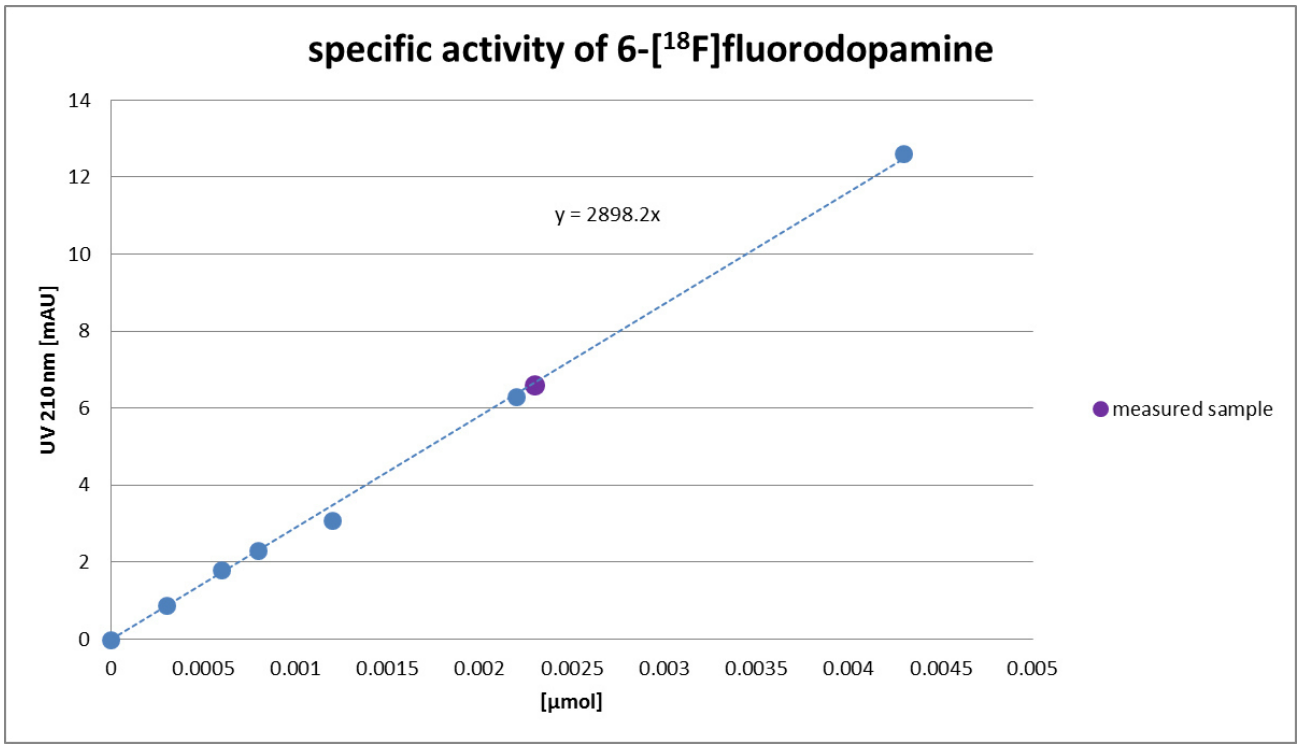


* Chromatograms of the isolated and the crude product are identically.

Specific activity calculation

The specific activities (GBq/µmol) were calculated by dividing the radioactivity of the ^{18}F-labeled product by the amount of the unlabeled tracer determined from the peak area in the UV-HPLC chromatograms (λ=210 nm). The amounts of unlabeled compounds were determined from the UV-absorbance/concentration calibration curve. The solutions of 6-[^{18}F]FDOPA an 6-[^{18}F]FDA (all synthesis started from 7 GBq of $^{18}F^-$) obtained after HPLC purification were concentrated under reduced pressure, the residues were redissolved in 4% EtOH in 0.02 M sodium phosphate buffer (300 µL, pH 2.5) and the resulting solutions were completely injected. The specific activity of 6-[^{18}F]FDOPA was 175 GBq/µmol. The specific activity of 6-[^{18}F]FDA was 60 GBq/µmol.





In vivo evaluation

All experiments were carried out in accordance with the EU directive 2010/63/EU for animal experiments and the German Animal Welfare Act (TierSchG, 2006) and approved by regional authorities (LANUV NRW).

Rat model of hemi-Parkinson's disease: Adult male Long Evans rats (Janvier, France) were used. Rats were anesthetized (initial dosage: 5% isoflurane in O_2/N_2O (3:7), then reduction to 2.5%) and fixed in a stereotaxic frame. Body temperature was monitored rectally and was held constant at 37 °C using a heating pad. After removing skin and periosteuma, a small hole (approx. 1 mm in diameter) was drilled in the skull 1.2 mm lateral and 4.4 mm posterior from bregma, and the cannula of a Hamilton syringe was inserted 8 mm deep, measured from the level of the dura mater. 6-Hydroxydopamine hydrobromide (6-OHDA, Sigma Aldrich, contains ascorbic acid as stabilizer) (21 µg) in isotonic saline (3 µL) was slowly injected, and the cannula was left in place for 10 min. After

retraction of the cannula, the burr hole was closed with bone wax and the skin wound was sutured. Finally, carprofen for analgesia (Rimadyl®) (3 mg) was subcutaneously injected.

µPET-Imaging: PET measurements were carried out 6–8 weeks after 6-OHDA injection. Prior to the PET measurement animals were anesthetized (initial dosage: 5% isoflurane in O_2/N_2O (3:7), then reduction to 2%), and a catheter for tracer-injection was inserted into the lateral tail vein. Rats were placed on an animal holder (medres GmbH, Cologne, Germany), and fixed with a tooth bar in a respiratory mask. Dynamic PET scans in list mode were performed using a Focus 220 micro PET scanner (CTI-Siemens, Erlangen, Germany) with a resolution at the center of field of view of 1.4 mm. Data acquisition started immediately after intravenous injection of 6-[^{18}F]FDOPA [56–93 MBq in 0.5 mL of 4% EtOH 0.02 M sodium phosphate buffer (pH 2.5–4)] and was carried out for 60 min. Thereafter a 10 min transmission scan was acquired using a ^{57}Co point source. Breathing rate was monitored with a Dasy Lab system 9.0 (DasyLab, Mönchengladbach, Germany) and kept around 60/min by adjusting the isoflurane concentration (1.5–2.5%). Body temperature was maintained at 37 °C by a feedback-controlled system (medres GmbH, Cologne, Germany). Following Fourier rebinning, data were reconstructed using an iterative OSEM3D/MAP procedure[13] including attenuation correction in two different ways: 1) 24 frames (6×30 s, 3×1 min, 3×2 min, 12×4 min) for compilation of striatal time activity curves. 2) 2 frames (2×30 min) for other VOI analyses. Resulting voxel sizes were always 0.38×0.38×0.82 mm.

MR-Imaging: To rule out gross structural brain anomalies and to provide individual templates for co-registration of the PET images, T2-weighted structural MR images were acquired. MRI scans were performed in an 11.7-T BioSpec animal scanner (Bruker BioSpin®) using a quadrature receive-only rat brain surface coil (Bruker BioSpin®) in combination with an actively decoupled, transmit-only quadrature resonator with 72 mm inner diameter (Bruker Biospin), fitting into the

BFG-150/90-S14 combined gradient and shim set of 90 mm inner diameter (Resonance Research Inc., Billerica, MA, USA) with a maximum gradient strength of 745 mT/m. A T2-weighted sequence, rapid acquisition with relaxation enhancement (RARE) was used: RARE factor = 8, repetition time/effective echo time = 6500/32.5 ms, averages = 2, matrix size = 256×256, FOV = 3.2×3.2 cm^2, 58 slices, slice thickness = 0.5 mm, interslice spacing = 0.5 mm. The inhalation anesthesia procedure was the same as that used for the μPET scan. During scanning, body temperature was recorded and maintained at 37° using feedback water control (medres GmbH, Cologne, Germany); physiological parameters, in particular respiration rate, were monitored using DASYLab 9.0 (DasyLab, Moenchengladbach, Germany).

Imaging Data Analysis: MR- [Gauss filtered (2 mm FWHM)] and μPET-Images were manually co-registered using VINCI 4.04 software.[14] SUVs (%ID/g) were calculated by dividing the concentration of [^{18}F]FDOPA in the region of interest by the total injected dose (ID) and multiplying by 100.

To obtain time activity curves, an elliptical 10 mm^3 volume of interest (VOI) was placed over the intact striatum. Mean SUV values were extracted from each of the 24 frames, decay corrected, and plotted over time.

For other VOI analyses, the frame covering 30–60 min post injection was used. Two elliptical VOIs (17 mm^3 each) were placed in the left and right striatum, respectively, and a 90 mm^3 VOI was used for the cerebellum. Mean SUV values were used to calculate striatum-to-cerebellum and ipsi-to-contralateral striatal ratios.

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Radiopharmaceuticals

Copper-Mediated Aromatic Radiofluorination Revisited: Efficient Production of PET Tracers on a Preparative Scale

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Abstract: Two novel methods for copper-mediated aromatic nucleophilic radiofluorination were recently reported. Evaluation of these methods reveals that, although both are efficient in small-scale experiments, they are inoperative for the production of positron emission tomography (PET) tracers. Since high base content turned out to be responsible for low radiochemical conversions, a "low base" protocol has been developed which affords ^{18}F-labeled arenes from diaryliodonium salts and aryl pinacol boronates in reasonable yields. Furthermore, implementation of our "minimalist" approach to the copper-mediated [^{18}F]-fluorination of (mesityl)(aryl)iodonium salts allows the preparation of ^{18}F-labeled arenes in excellent RCCs. The novel radiofluorination method circumvents time-consuming azeotropic drying and avoids the utilization of base and other additives, such as cryptands. Furthermore, this procedure enables the production of clinically relevant PET tracers; [^{18}F]FDA, 4-[^{18}F]FPhe, and [^{18}F]DAA1106 are obtained in good isolated radiochemical yields. Additionally, [^{18}F]DAA1106 has been evaluated in a rat stroke model and demonstrates excellent potential for visualization of translocator protein 18 kDa overexpression associated with neuroinflammation after ischemic stroke.

Introduction

Positron emission tomography (PET) is one of the leading imaging modalities in clinical diagnostics. This technique allows the visualization of different physiological and pathological processes at the molecular level with high resolution and extraordinary sensitivity, using probes labeled with suitable β^+-emitting radioisotopes. Among these radionuclides, ^{18}F holds a dominant position, owing to its almost ideal decay properties with respect to half-life (109.8 min) and decay energy (630 keV). The efficacy of PET examinations is strongly dependent on the development of PET tracers that selectively address targets of high relevance. For this purpose, development of novel high-yielding and simple radiofluorination techniques is essential. However, in comparison to fluorination methods, these techniques are still rather rare in radiochemistry. This is mainly due to the relatively short half-life and the tiny amounts of ^{18}F used for tracer preparation. Moreover, during radiosyntheses with ^{18}F radiation, safety issues have to be considered. Despite this, in recent years several novel fluorination techniques have been successfully applied for the preparation of radiofluorinated compounds.[1] Pioneering approaches involving transition metal-mediated [^{18}F]-fluorination[2] were used for the efficient preparation of ^{18}F-labeled arenes and heteroarenes, including electron-rich ones, from nucleophilic [^{18}F]-fluoride. The original procedure reported by Lee et al. (Scheme 1 A)[3] comprises the reaction of a radiofluorinated Pd^{IV} complex prepared in advance with an appropriate Pd^{II}–aryl compound, yielding the corresponding ^{18}F-labeled arene after reductive elimination (Scheme 1 A).[3,4] A further development of this method involves radiolabeling of an arylnickel(II) complex with a radiofluorination agent generated in situ from a hypervalent iodine oxidant [bis(onio)-substituted aryliodine(III)] and [^{18}F]-fluoride (Scheme 1 B).[5] These works are of fundamental importance for the development of radiochemistry.

Both methods, however, are rather impractical and difficult to perform in automated synthesis modules due to the high moisture sensitivity of the involved reaction components (radiofluorinated Pd^{IV} complex and hypervalent iodine oxidant). These difficulties have so far precluded widespread application of either procedure.

Recently, two methods for the ^{18}F-labeling of arenes based on copper-mediated aromatic nucleophilic radiofluorination were reported. The first, developed by Sanford, Scott, and coworkers (Scheme 2 A),[6] enables the preparation of diverse radiofluorinated arenes via sterically-controlled Cu-mediated [^{18}F]-fluorination of (mesityl)(aryl)iodonium salts using [^{18}F]KF/18-

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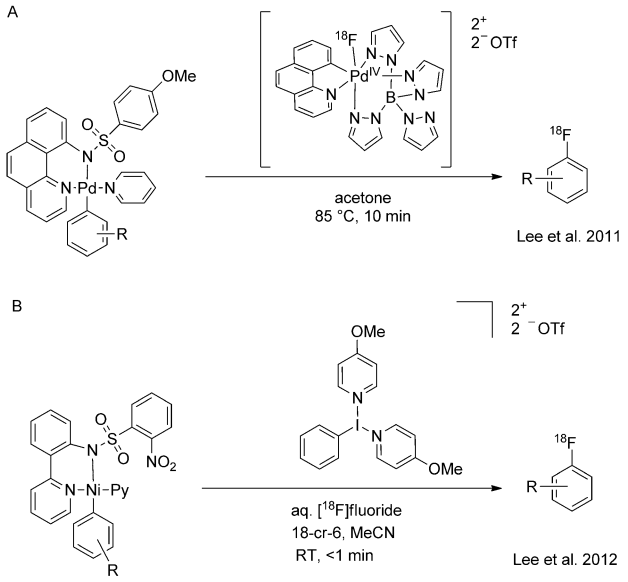
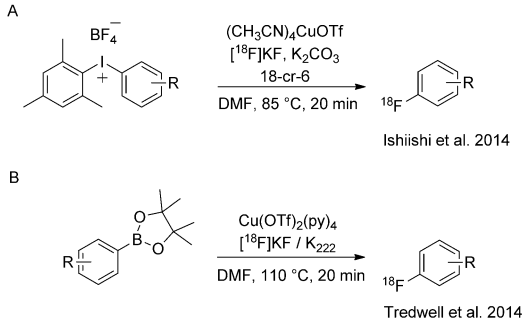


Scheme 1. Pd- and Ni-mediated preparation of [^{18}F]fluoroarenes according to Ritter and co-workers (A[3] and B,[5a] respectively).



Scheme 2. Copper-mediated ^{18}F-labeling of diaryliodonium salts (A) and pinacolyl arylboronates (B).

crown-6. For uncatalyzed fluorination of diaryliodonium salts, *ortho* substituents direct the addition of fluoride to the more sterically crowded ring.[7] In contrast, if copper catalysts are used, the mesityl group directs oxidative addition and fluorination to the sterically less crowded aryl group on iodine, regardless of its electronic properties. Most importantly, this method allows preparation of fluoroarenes with electron-donating substituents in high yields and with excellent selectivity. These compounds are difficult to access via nucleophilic fluorination reactions under commonly used non-catalytic conditions.

In the second method, proposed by Gouverneur and co-workers (Scheme 2B), [^{18}F]arenes are obtained from pinacolyl arylboronates (arylBPin) upon treatment with [^{18}F]KF/K_{222} and $Cu(OTf)_2(py)_4$.[8] They adapted the method of $Cu(OTf)_2$-mediated fluorination of aryltrifluoroborates and arylBPin with KF, originally reported by Ye et al. for the preparation of [^{18}F]arenes.[9]

Both procedures were used for the radiofluorination of various substrates containing electron-donating, -withdrawing and -neutral substituents. No special precautions were necessary and radiofluorinations were carried out under air. The complexes used as copper sources ($(MeCN)_4CuOTf$ and $Cu(OTf)_2(py)_4$ for the Sanford/Scott and Gouverneur protocols, respectively) have the advantage of being bench stable and commercially available.

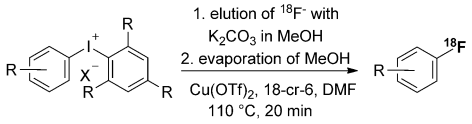
Our studies revealed that the pioneering methods for the copper-catalyzed preparation of [^{18}F]arenes, though efficient in small-scale experiments, were not well-suited for PET tracer production on a practical scale. The reason for the failure of the two methods on preparative-scale application was elucidated and prompted us to develop novel radiofluorination procedures to overcome this shortcoming.

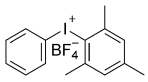
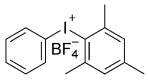
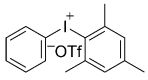
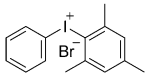
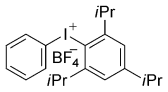
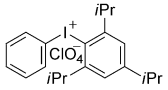
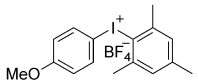
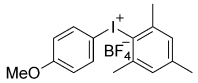
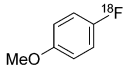
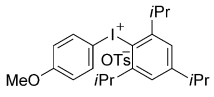
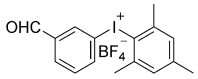
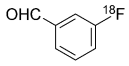
Results and Discussion

Initially, we studied the $Cu(OTf)_2$-mediated fluorination of mesityl-substituted diaryliodonium salts with [^{18}F]KF/18-crown-6 for ^{18}F-labeling.[10] (Mesityl)(aryl)iodonium precursors of 3-[^{18}F]fluorobenzene, [^{18}F]fluorobenzaldehyde (3-[^{18}F]FBA), and 4-[^{18}F]fluoroanisole were selected as models of electron-neutral, -rich, and -deficient arene substrates, respectively (Table 1). After some experimental refinements, the following procedure was applied. $^{18}F^-$ was trapped on an anion-exchange resin and then eluted with a methanolic solution of K_2CO_3 into a reaction vial containing 18-crown-6. A small amount of K_2CO_3 (0.5 mg instead of usually used 2.8–3.5 mg) was sufficient to recover $^{18}F^-$ quantitatively (>98%). Low-boiling methanol was completely removed at 70–80 °C within 2–3 min without the need for azeotropic drying. Radioactivity loss during the evaporation step was negligible (<2%). The residue was taken up in a solution of the corresponding iodonium precursor and $Cu(OTf)_2$ in DMF and heated for 20 min at 85 °C min under Ar. Afterwards, an excess of water was added to quench the reaction and completely solubilize surface-adsorbed $^{18}F^-$. Subsequently, radiochemical conversions (RCCs)[1b] were determined via radio-HPLC. [^{18}F]Fluorobenzene, 4-[^{18}F]fluoroanisole and 3-[^{18}F]FBA, were successfully prepared from the respective (mesityl)(aryl)iodonium tetrafluoroborate precursors in moderate RCCs of 41, 29, and 50%, respectively (Table 1, entries 1, 7, and 10). The selectivity of radiofluorination was excellent in all experiments. [^{18}F]Fluoride and traces of [^{18}F]fluoromesitylene (<1%) were the only radioactive impurities. A prolonged reaction time of 30 min resulted in a higher RCC of 64% in the case of [^{18}F]fluorobenzene but not for 4-[^{18}F]fluoroanisole (Table 1, entries 2 and 8, respectively). Additionally, (2,4,6-triisopropylphenyl)(aryl)iodonium salt precursors also afforded radiofluorinated arenes, although in lower RCCs (Table 1, entries 5, 6, and 9). In addition to tetrafluoroborates, other (mesityl)(aryl)iodonium salts such as perchlorates, tosylates, bromides and triflates were tested. Perchlorate and triflate salts were found to be suitable substrates for radiofluorination (Table 1, entries 3 and 6).[11] Tosylate or bromide, in contrast, afforded only low RCCs (Table 1, entries 4 and 9).

While our own studies were in progress, the radiolabeling protocols of Sanford/Scott[6] and Gouverneur[8] were published. We therefore carried out experiments to compare our procedure (Table 1) with the published ones (Table 2 and Table 3).

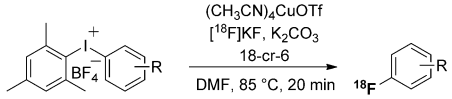
Table 1. $Cu(OTf)_2$-mediated one-pot radiofluorination of (mesityl)(aryl)iodonium salts using K_2CO_3 in MeOH for elution of $^{18}F^-$.[a]

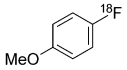
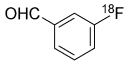


Entry	Precursor	Product	RCC [%]
1			41
2[b]			64
3			35
4			0
5			36
6			35
7			29
8[b]			29
9			13
10			50

[a] $^{18}F^-$ (100–750 MBq) was eluted from an anion exchange resin with K_2CO_3 (0.5 mg, 4 μmol; in 1 μL H_2O) in MeOH (500 μL) and methanol was evaporated under He flow at 70–80 °C. The residue was taken up in DMF (200 μL) containing $[Ar_2I]X$ (12 μmol), $Cu(OTf)_2$ (4 mg, 12 μmol) and 18-crown-6 (8.8 mg, 30 μmol). The resulting solution was heated at 85 °C for 20 min under Ar. Afterwards, the reaction mixture was cooled to room temperature and diluted with water (2 mL). [b] Reaction time: 30 min. All syntheses were carried out manually. RCCs were determined by radio-HPLC. Each experiment was carried out at least in triplicate. The standard deviation of RCC did not exceed 10% of its mean value.

Table 2. $(MeCN)_4CuOTf$-mediated radiofluorination of (mesityl)(aryl)iodonium salts: "High base" vs. "low base" protocol.[a]



Entry	Product	Aliquot[b] RCC [%]	One-Pot RCC [%] "High base"[c,d]	One-Pot RCC [%] "Low base"[e]
1		56	1 (5)	28
2		84	< 0.5 (3)	56
3		36	5 (1)	26

[a] All syntheses were carried out manually. RCCs were determined by radio-HPLC. Each experiment was carried out at least in triplicate. The standard deviation of RCC did not exceed 10% of its mean value. [b] Syntheses of the radiolabeled compounds were carried out according to the literature.[13] Thereafter, the reaction mixture was cooled down to ambient temperature and diluted with H_2O (2 mL). [c] Syntheses of the radiolabeled compounds were carried out according to the literature.[14] After that, the reaction mixture was cooled down to ambient temperature and diluted with H_2O (2 mL). [d] RCCs of [^{18}F]fluoromesitylene are given in parentheses. [e] $^{18}F^-$ (100–750 MBq) was eluted from an anion exchange resin with K_2CO_3 (0.11 mg, 0.8 μmol) in water (200 μL). The resin was washed with a solution of 18-crown-6 (0.47 mg, 1.8 μmol) in MeCN (1 mL). The combined fractions were concentrated under reduced pressure at 95 °C and the residue was azeotropically dried with MeCN (2× 1 mL) under reduced pressure and air. The residue was taken up in a solution of $(MeCN)_4CuOTf$ (4.2 mg, 12 μmol) in DMF (300 μL) and heated at 85 °C for 20 min under air. The reaction mixture was cooled to room temperature and diluted with water (2 mL).

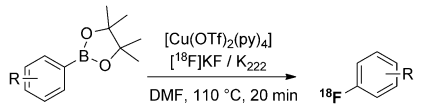
In accordance with the Sanford/Scott protocol, the corresponding $[Ar–I–Mes]^+BF_4^-$ was treated with [^{18}F]KF/18-crown-6 and $(MeCN)_4CuOTf$ in DMF at 85 °C for 20 min under air.[12] Using small aliquots of [^{18}F]KF/18-crown-6 (10–30 MBq) in DMF obtained via redissolution of dried [^{18}F]KF/18-crown-6, high and selective ^{18}F-incorporation was observed (Table 2) affording [^{18}F]fluorobenzene, 4-[^{18}F]fluoroanisole and 3-[^{18}F]FBA in RCCs of 56, 84 and 36%, respectively. However, unacceptably high losses of ^{18}F activity due to adsorption onto vessel walls were already observed before radiolabeling had been carried out. Accordingly, only 20–50% of azeotropically dried [^{18}F]KF/18-crown-6 could be resolubilized in DMF. In an attempt to circumvent this problem, a solution of the precursor and $(MeCN)_4CuOTf$ in DMF was added directly to the vial containing dried [^{18}F]KF/18-crown-6 (conventional protocol using 3 mg K_2CO_3 and 15 mg K_{222} in MeCN).[6] After addition, the mixture was heated at 85 °C for 20 min (one-pot radiosynthesis under "high base" conditions). Unexpectedly, ^{18}F-labeled model arenes were obtained in only 0.5–5% RCCs. The concurrent formation of [^{18}F]fluoromesitylene (up to 5%) was observed, indicating a loss of selectivity. We assumed that $(MeCN)_4CuOTf$ is unstable under basic conditions. Therefore, $^{18}F^-$ was eluted with essentially the same amount of K_2CO_3 as that used in the experiments with small aliquots of [^{18}F]KF/18-crown-6 (i.e., 0.1 instead of 3.5 mg K_2CO_3; "low base" conditions). Radioactivity recovery from the anion exchange resin was about 88% and radioactivity loss during azeotropic drying did not exceed 3% (decay-corrected; 12% not corrected for decay). Under "low base" conditions, acceptable RCCs of 28, 56, and 26% were

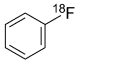
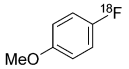
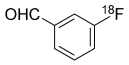
achieved for [^{18}F]fluorobenzene, 4-[^{18}F]fluoroanisole, and 3-[^{18}F]FBA, respectively.

In the next set of experiments, the respective arylBPin radiofluorination precursor, $Cu(OTf)_2(py)_4$ and [^{18}F]KF/K_{222} in DMF were heated at 110 °C for 20 min under air, as described by Gouverneur and co-workers (Table 3).[8] As with the Sanford/Scott approach, this procedure afforded [^{18}F]fluorobenzene, 4-[^{18}F]fluoroanisole, and 3-[^{18}F]FBA in good RCCs of 67, 66 and 76% if small aliquots of previously dried [^{18}F]KF/K_{222} (ca. 30 MBq) were used. In this case, about 20–25% of the radioactivity could not be resolubilized and remained on the vessel walls after azeotropic drying. To avoid such high activity losses a one-pot radiofluorination procedure was tested. Under "high base" conditions, one-pot radiosyntheses provided radiofluorinated arenes in only 5–7% RCCs.

In contrast, "low base" (0.06 mg K_2CO_3 instead of 2.8 mg) conditions afforded ^{18}F-labeled model compounds in 41–64% RCCs.

Table 3. $Cu(OTf)_2(py)_4$-mediated radiofluorination of pinacolyl arylboronates: "High base" vs. "low base" protocols.[a]



Entry	Product	Aliquot[b] RCC [%]	One-Pot RCC [%] "High base"[c]	One-Pot RCC [%] "Low base"[d]
1		67	5	64
2		66	7	42
3		76	7	41

[a] All syntheses were carried out manually. RCCs were determined by radio-HPLC. Each experiment was carried out at least in triplicate. The standard deviation of RCC did not exceed 10% of its mean value. [b] Syntheses of the radiolabeled compounds were carried out according to the literature.[1c] Thereafter, the reaction mixture was cooled down to ambient temperature and diluted with H_2O (2 mL). [c] Syntheses of the radiolabeled compounds were carried out according to the literature.[8] Briefly, [^{18}F]fluoride (95–170 MBq) was eluted from an anion-exchange resin with a solution of K_{222} (13 mg) and K_2CO_3 (2.8 mg) in 80% MeCN (1 mL). The solvent was evaporated under reduced pressure at 95 °C. The residue was azeotropically dried with MeCN (2×1 mL). Thereafter, solutions of $Cu(OTf)_2(py)_4$ (15 mg, 22 μmol) and the corresponding arylboronic acid pinacol ester (60 μmol) in DMF (each in 150 μL) were added and the reaction vial was purged with air. The reaction mixture was heated at 110 °C for 20 min, cooled to room temperature and diluted with water (2 mL). [d] $^{18}F^-$ (100–750 MBq) was eluted from an anion exchange resin with a solution of K_2CO_3 (0.06 mg, 0.43 μmol) and K2.2.2 (0.27 mg, 0.72 μmol) in 80% MeCN (1 mL). The solvent was evaporated under reduced pressure at 95 °C and the residue was azeotropically dried with MeCN (1 mL; two times) under air. A solution of $Cu(OTf)_2(py)_4$ (3.6 mg, 5.3 μmol) in DMF (150 μL) followed by a solution of arylboronic acid pinacol ester (60 μmol) in DMF/MeCN (5:1; 180 μL) were added to the residue and the reaction mixture was heated at 110 °C for 20 min under air. The reaction mixture was cooled to room temperature and diluted with water (2 mL).

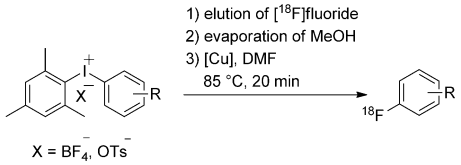
Once the counterproductive role of the base had been ascertained, we studied whether the "minimalist" approach could be applied in copper-mediated aromatic nucleophilic radiofluorination.[16] According to this radiolabeling protocol, [^{18}F]fluoride is directly eluted from the anion-exchange resin with alcoholic solutions of the labeling precursors bearing a quaternary ammonium, diaryliodonium, or triarylsulfonium functionality. After alcohol removal, the resulting [^{18}F]fluoride onium salt is then simply heated in a suitable solvent. This method eliminates not only the need for time-consuming azeotropic drying steps but also, most importantly with respect to copper-mediated aromatic ^{18}F-labeling, the necessity for a base or any other additives. Under "minimalist" conditions, the copper mediated radiofluorination was carried out as follows.$^{18}F^-$ was eluted from the solid support with a solution of a (mesityl)(aryl)iodonium salt precursor in MeOH almost quantitatively (>97% recovery). After methanol removal, the respective copper compound in DMF was added to the residue and the resulting solution was heated at 85 °C for 20 min before the reaction was quenched with an excess of water. Gratifyingly, with $(MeCN)_4CuOTf$, $(t\text{BuCN})_2CuOTf$ or $Cu(OTf)_2$, excellent RCCs of all three ^{18}F-labeled model compounds were achieved from the corresponding tetrafluoroborates (Table 4). Whereas radiofluorinations with $(MeCN)_4CuOTf$ and $(t\text{BuCN})_2CuOTf$ could be carried out under air, radiolabeling with $Cu(OTf)_2$ required inert conditions. In the case of [^{18}F]fluorobenzene and 4-[^{18}F]fluoroanisole, an excellent radiofluorination selectivity was observed. For 3-[^{18}F]FBA some erosion of selectivity (RCC of [^{18}F]fluoromesitylene = 5%), as well as the formation of several additional unidentified side-products (overall <7%) took place. The three copper salts afforded comparable RCCs of radiofluorinated arenes. Advantageously DMF solutions of $(MeCN)_4CuOTf$ remained stable under ambient conditions for a prolonged period of time (at least 6 h). (4-MeOPh–I–Mes)$^+X^-$ salts with different counterions $X^- = OTs$, ClO_4, Br and OTf were tested as radiolabeling precursors. (4-MeOPh–I–Mes)$^+OTs^-$ afforded 4-[^{18}F]fluoroanisole in slightly higher RCC than that of the corresponding tetrafluoroborate precursor (Table 4, entry 4). The remaining iodonium salts were not suitable for this labeling procedure.

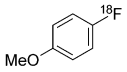
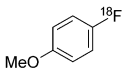
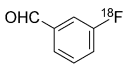
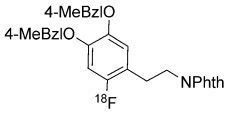
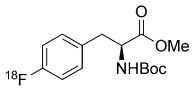
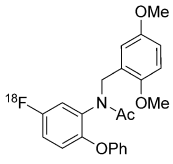
In the next step, the feasibility of the novel radiofluorination procedure for the preparative-scale production of ^{18}F-labeled arenes was evaluated. Accordingly, 4-[^{18}F]fluoroanisole was prepared from the corresponding iodonium tetrafluoroborate precursor starting from 12 GBq of $^{18}F^-$. Radiochemical yield (RCY) of the isolated labeled product amounted to 59% within 45 min.

With an efficient radiofluorination procedure in hand, we turned to the preparation of clinically relevant PET tracers.

6-[^{18}F]Fluorodopamine (6-[^{18}F]FDA, [^{18}F]-**4**) targets specifically catecholamine synthesis, storage, and secretion pathways.[18] Although originally developed as a sympathoneural imaging agent, 6-[^{18}F]FDA has been confirmed as an excellent tracer for the diagnosis and localization of chromaffin tumors, neuroblastomas, ganglioneuromas, and metastatic pheochromocytomas.[13,19]

Table 4. Copper-mediated aromatic nucleophilic radiofluorination under "minimalist" conditions.[a]



Entry	Product	[(MeCN)$_4$CuOTf] RCC [%]	Cu(OTf)$_2$ RCC [%]	[(*t*BuCN)$_2$CuOTf] RCC [%]
1		90	68	90
2		86	72	77
3[b]		90	–	–
4		78	69	85
5	 [18F]-**1**	71–94	83	–
6	 [18F]-**2**	81–92	–	–
7	 [18F]-**3**	93	92	–

4-MeBzl = 4-methylbenzyl group;[17] NPhth = *N*-phthalimido group. [a] [18F]Fluoride (0.1–10 GBq) was eluted from an anion exchange resin with precursor (12 μmol) in MeOH (500 μL). Methanol was evaporated under He flow at 70–80 °C, The residue was dissolved in a solution of a copper salt (12 μmol) in DMF (300 μL) and the resulting solution was heated at 85 °C for 20 min. The reaction mixture was diluted with water (2 mL). RCCs were determined by radio-HPLC. Radiofluorinations with (MeCN)$_4$CuOTf and (*t*BuCN)$_2$CuOTf were carried out under air. Radiofluorinations with Cu(OTf)$_2$ were carried out under inert conditions. All syntheses were carried out manually. Each experiment was carried out at least in triplicate. The standard deviation of RCC usually did not exceed 20% of its mean value. Where the standard deviation of RCCs exceeded 20% of its mean value, the range of RCCs is given. [b] Iodonium tosylate precursor was used.

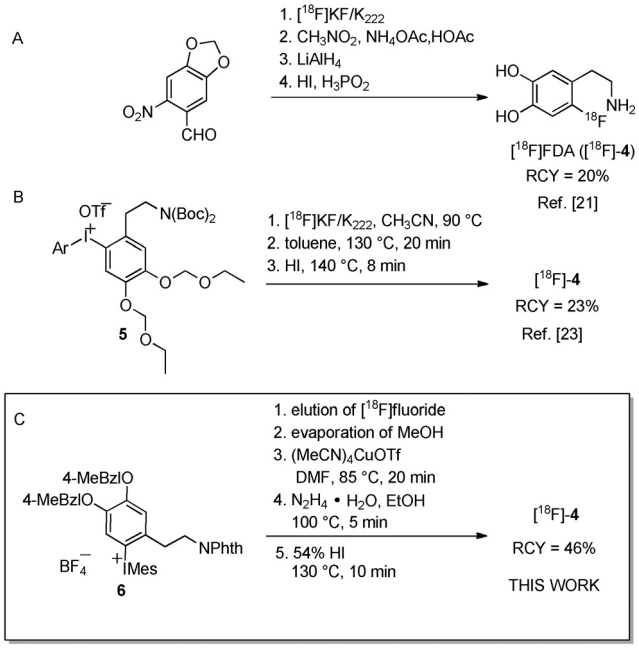
Several procedures for the preparation of 6-[18F]FDA have been reported.[20] The first radiosynthesis of [18F]-**4** via aromatic nucleophilic radiofluorination, published by Ding et al.,[21] consisted of four cumbersome reaction steps (Scheme 3A). Quite recently, the synthesis of 6-[18F]FDA using the respective (4-anisyl)(aryl)iodonium precursor **5** and [18F]KF/K$_{222}$ in toluene[22] was reported (Scheme 3B).[23] After deprotection of the radiolabeled intermediate with HI, 6-[18F]FDA was isolated in 23% RCY over two steps.

Using our novel procedure, the iodonium salt precursor **6** afforded 71–94% RCC of protected 6-[18F]fluorodopamine [18F]-**1** (Scheme 3C; Table 4, entry 5). Since deprotection with HI at 130 °C was incomplete, even after 20 min (55% conversion), the radiolabeled intermediate was first treated with hydrazine hydrate in EtOH to quantitatively remove the *N*-phthalimido group. Thereafter, the *O*-4-methylbenzyl groups[17] were cleaved with HI to give 6-[18F]FDA. Starting from 2.4 GBq of [18F]fluoride, a dose of 550 MBq of 6-[18F]FDA (46% RCY) was obtained as a ready-to-use solution in 4% ethanolic phosphate buffer within a total synthesis time of 130 min. Optimization of the deprotection chemistry is under way and will be reported in due course.

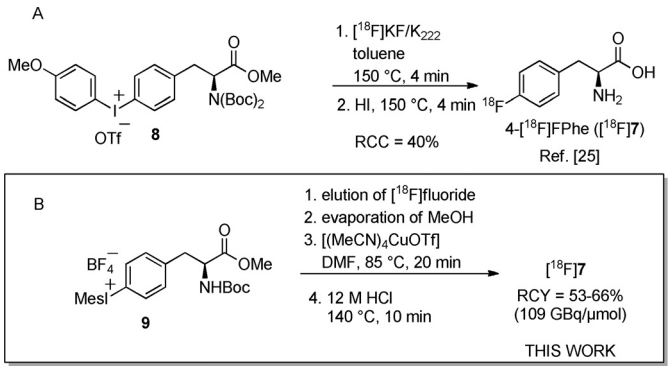
4-[18F]Fluoro-L-phenylalanine (4-[18F]FPhe, [18F]-**7**) is well-suited for the visualization of increased protein synthesis rate, especially in peripheral and cerebral tumors. However, examples of clinical applications of 4-[18F]FPhe are rather rare owing to its poor accessibility via nucleophilic radiofluorination.[14,24] Last year a novel preparation method for [18F]-**7** was presented by Neumann et al.[24,25] According to this protocol, 4-[18F]FPhe was prepared in 20% RCY with >37 GBq μmol^{-1} specific activity, from the respective (4-anisyl)(aryl)iodonium salt precursor **8** (Scheme 4A).

(MeCN)$_4$CuOTf-mediated radiofluorination of iodonium salt **9** under "minimalist" conditions afforded the radiofluorinated intermediate [18F]-**2** in 81–92% RCC (Scheme 4B, Table 4, entry 6). Deprotection of the labeled intermediate was accomplished with 12 N HCl at 140 °C for 10 min. Finally, HPLC purification afforded [18F]-**7** in a 4% ethanolic phosphate buffer solution. Enantiomerically pure 4-[18F]FPhe (> 98% *ee*; (*R*)-isomer was not observed) was isolated in 53–66% RCY in two steps with a specific activity of 109 GBq μmol^{-1} (14 GBq 4-[18F]FPhe were obtained from 49.5 GBq of $^{18}F^-$ within 117 min). Thus, RCY and specific activity of [18F]-**7** were within the same range as those achieved for 4-(2-[18F]fluoroethyl)-L-tyrosine ([18F]FET).[26] [18F]FET represents a very popular surrogate tracer for phenylalanine and is widely used, especially for diagnosis of brain tumors.[27] However, in contrast to 4-[18F]FPhe, protein incorporation of [18F]FET is very low.[25b] Tumor retention of 4-[18F]FPhe should be higher than that of [18F]FET, which could make 4-[18F]FPhe well-suited for imaging of brain tumors. In our opinion, this justifies its future preclinical and, probably, clinical evaluation.

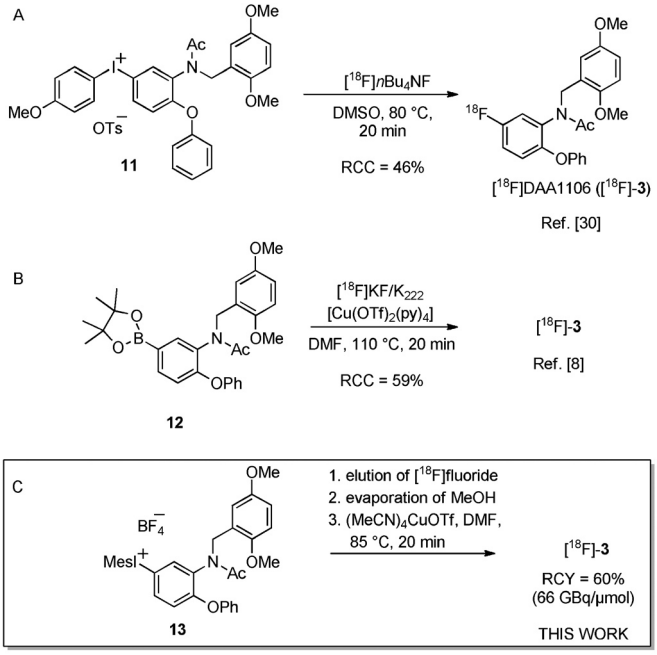
DAA1106 (**3**; Scheme 5) is a very potent and selective agonist at the translocator protein 18 kDa (TSPO). TSPO is a biomarker of brain inflammation and reactive gliosis, which are characteristic for a number of neurodegenerative and psychiatric diseases, stroke and brain tumors.[28] [11C]DAA1106 is intensely used for monitoring of these pathologies.[29] However, the short half-life of ^{11}C ($t_{1/2} = 20$ min) precludes commercialization and hampers its broad application. Suzuki et al.,[30] as well as Gouverneur and co-workers,[8] showed that ^{18}F-labeling of the corresponding (4-anisyl)(aryl)iodonium salt **10** or arylBPin **11** precursor, afforded [18F]DAA1106 in 46 and 59% RCC, respectively. However, [18F]DAA1106 has not yet been isolated or biologically tested.



Scheme 3. Preparation of 6-[^{18}F]fluorodopamine (6-[^{18}F]FDA, [^{18}F]-**4**).



Scheme 4. Radiosynthesis of 4-[^{18}F]fluoro-L-phenylalanine (4-[^{18}F]FPhe, [^{18}F]-**7**).



Scheme 5. Preparation of [^{18}F]DAA1106 ([^{18}F]-**3**).

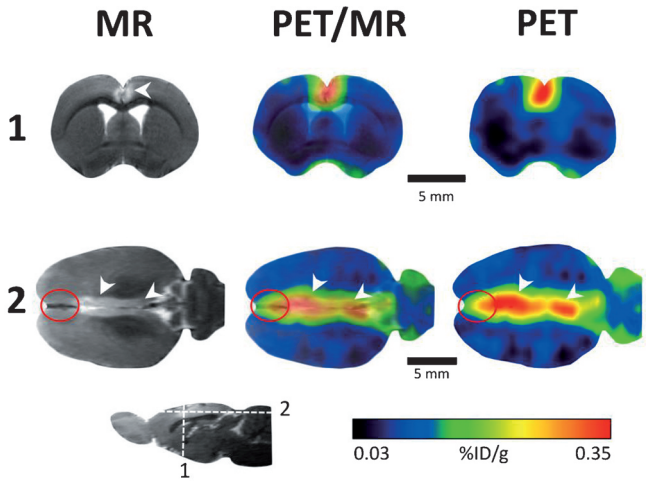


Figure 1. [^{18}F]DAA1106 MR-PET six days after anterior cerebral artery occlusion, showing transverse (1) and horizontal images (2), with the ischemic lesion in the anterior cingulate cortex visible as hyperintensity (white arrowheads in MR- and PET-MR-images). The peri-infarct zone is highlighted by red circles. Section levels are indicated by white dashed lines in the insert (bottom left).

Under our standard "minimalist" conditions, precursor **12** was efficiently radiofluorinated to give [^{18}F]DAA1106 in 89–96% RCC (Scheme 5C). The RCY of the tracer with a specific activity of 66 GBq µmol^{-1} after HPLC isolation and formulation as a solution in EtOH amounted to 60% (1.2 GBq were obtained from 2.4 GBq of $^{18}F^-$ in a total synthesis time of 45 min).[31]

Additionally, the amount of Cu in the final product solution of 4-[^{18}F]fluoroanisole, 4-[^{18}F]FPhe, and [^{18}F]DAA1106 was determined by using inductively coupled plasma mass spectrometry (ICP-MS). The Cu content varied from 0.6–1.4 ppm. This is significantly below any level of concern according to the ICH Guideline of Elemental Impurities (Q3D).[32]

Visualization of neuroinflammation by [^{18}F]DAA1106-PET was studied in a rat stroke model (Figure 1). Ischemic stroke was induced by occlusion of the anterior cerebral artery (ACA) using stereotaxic injection of the vasoconstrictor endothelin-1.[33] ACA occlusion resulted in an ischemic lesion in the anterior cingulate cortex. At day 6 after induction of stroke, the inflammatory processes in the ischemic core and the surrounding tissue were studied using [^{18}F]DAA1106-PET. PET images were projected on a structural MR image from the same animal, acquired on the following day. The [^{18}F]DAA1106 PET signal overlapped with the ischemic lesion, visible as a hyperintensity in the MR image (Figure 1, white arrowheads). The [^{18}F]DAA1106 PET image displayed the ischemic lesion at most section levels with excellent signal-to-noise ratio. A mismatch with the MR image was observed in the most rostral part (Figure 1, red circles), where the radioactive signal extended beyond the in-

farcted core. This was due to inflammatory processes in the peri-infarct zone after stroke.[34]

Conclusion

We have established a robust "low base" protocol for the efficient copper-mediated preparation of [^{18}F]arenes from diaryliodonium salts and aryl pinacol boronates. This method overcomes the problem of insufficient stability of copper catalysts like $(MeCN)_4CuOTf$ and $Cu(OTf)_2(py)_4$ under the basic conditions used in the previously reported procedures. Whereas these "high" base radiofluorination procedures afforded very low RCCs on a preparative scale, our approach enables the preparation of radiolabeled arenes in reasonable yields. Furthermore, we have developed a novel radiofluorination method that combines the advantages of our previously reported "minimalist" approach with the exceptional capabilities of copper-mediated aromatic nucleophilic radiofluorination. The versatility and scope of the novel radiofluorination procedure was demonstrated by the preparation of three ^{18}F-labeled model arenes and by the production of clinical doses of three PET tracers, [^{18}F]FDA, 4-[^{18}F]FPhe, and [^{18}F]DAA1106, in good to excellent radiochemical yields. Furthermore, this novel method circumvents time-consuming azeotropic drying and avoids the application of base and other additives, such as cryptands. Additionally, [^{18}F]DAA1106 was evaluated in a rat stroke model and demonstrated excellent potential for visualization of TSPO overexpression associated with neuroinflammation after ischemic stroke.

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Keywords: copper · fluorination · imaging agents · positron emission tomography · radiopharmaceuticals

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Supporting Information

Copper-Mediated Aromatic Radiofluorination Revisited: Efficient Production of PET Tracers on a Preparative Scale

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Supporting Information

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Materials and Methods

General: ^{1}H-NMR spectra: Bruker Avance II 300 (300 MHz) and Bruker Avance II+ 600 (600 MHz). ^{1}H chemical shifts are reported in ppm relative to residual peaks of deuterated solvents. Higher-order NMR spectra were approximately interpreted as first-order spectra, where possible. The observed signal multiplicities are characterized as follows: s = singlet, d = doublet, t = triplet, q = quartet, quin = quintet, m = multiplet, and br = broad. Coupling constants (*J*) were reported in Hertz (Hz). ^{13}C-NMR spectra [additional APT (Attached Proton Test)]: Bruker Avance II 300 (75.5 MHz) and Bruker Avance II+ 600 (125.9 MHz). ^{13}C chemical shifts are reported relative to residual peaks of deuterated solvents. Low resolution ESI-MS: Finnigan LCQ. High resolution ESI-MS: Bruker APEX IV 7T FTICR MS. TLC: Merck precoated sheets, 0.25 mm Sil G/UV$_{254}$. The chromatograms were viewed under UV light and/or by treatment with phosphomolybdic acid (10% in ethanol). Column chromatography: Merck silica gel, grade 60, 230–400 mesh. Solvent proportions are indicated in a volume/volume ratio. All reactions were carried out with magnetic stirring unless otherwise stated and, in the case of air- or moisture-sensitive substrates and/or

reagents, were handled in flame-dried glassware under argon or nitrogen. Organic extracts were dried with anhydrous $MgSO_4$.

(Diacetoxyiodo)-2,4,6-triisopropylbenzene,[1] (phenyl)(mesityl)iodonium tetrafluoroborate,[2] (phenyl)(mesityl)iodonium triflate,[3] (phenyl)(mesityl)iodonium perchlorate,[4] (phenyl)(mesityl)iodonium bromide,[5] (phenyl)(1,3,5-triisopropylphenyl)iodonium triflate,[3] (4-methoxyphenyl)(mesityl)iodonium tetrafluoroborate,[2] (4-methoxyphenyl)(mesityl)iodonium tosylate,[4] (4-methoxyphenyl)(1,3,5-triisopropylphenyl)iodonium tosylate,[6] and (3-formylphenyl)-(mesityl)iodonium tetrafluoroborate[7] were prepared according to the literature or modified literature procedures.[8] *N*-(3,4-dihydroxyphenethyl)phthalimide,[9] methyl *N*-*tert*-butyloxycarbonyl-L-(4-trimethylstannylphenyl)alaninate,[10] methyl *N*-*tert*-butyloxycarbonyl-(4-fluorophenyl)alaninate,[11] *N*-(5-bromo-2-phenoxyphenyl)-*N*-(2,5-dimethoxybenzyl)acetamide,[12] *N*-(5-fluoro-2-phenoxyphenyl)-*N*-(2,5-dimethoxybenzyl)acetamide (DAA1106, **8**),[12-13] $Cu(OTf)_2(Py)_4$[14] and $(t\text{BuCN})_2CuOTf$[15] were prepared according to the literature.

StrataX cartridges were obtained from Phenomenex (Aschaffenburg, Germany) and Sep-Pak Accell Plus QMA carbonate plus light cartridges, 46 mg sorbent per cartridge from Waters GmbH (Eschborn, Germany).

All radiosyntheses were carried out manually according to the radiation protection rules of our institute using appropriate measures and shielding. For all radiosyntheses 3 mL V-vials (Wheaton, NJ) were used.

A CRC-55tR" dose calibrator (Capintec, NJ) was used for activity measurements. For all radio-HPLC analyses (determination of RCCs), activity of the sample was measured before injection. All radioactive fractions were collected and measured again. Radioactivity recovery was > 90%. Radioactivity in the reaction vial was measured before and after the reaction mixture was heated. The difference between both values (corrected for decay) did not exceed 5-7%. Before opening, reaction vials containing volatile [^{18}F]fluorobenzene and 4-[^{18}F]fluoroanisole were cooled in an ice

bath. HPLC analyses and purifications were carried out on a Dionex Ultimate 3000 System with Ultimate 3000 diode array detector coupled in series with a Berthold NaI detector. Unless otherwise stated, a Chromolith® SpeedROD RP-18e column (Merck, Darmstadt Germany), 50×4.6 mm, was used for analyses and purifications of radiofluorinated products. Aqueous MeCN and EtOH solutions were used as a mobile phase.

UV and radioactivity detectors were connected in series, giving a time delay of 0.5–0.9 min depending on the flow rate. ^{18}F-Labeled compounds were identified by spiking of the reaction mixture with unlabeled standards using HPLC.

Radiolabeled products were analyzed using the following methods. Method A: column: Chromolith SpeedROD®, 50×4.6 mm (Merck Millipore); gradient: 0–2 min: 5% MeCN, 2–2.5 min: 5→20% MeCN, 2.5–6 min: 20% MeCN, 6–7 min: 20→70% MeCN, 7–9 min: 70% MeCN, flow rate: 3.0 mL/min. Method B: column: Chromolith SpeedROD®, 50×4.6 mm (Merck Millipore); gradient: 0–2 min: 5% MeCN, 2–2.5 min: 5→70% MeCN, 2.5–7 min: 70% MeCN, flow rate: 3.0 mL/min.

[^{18}F]Fluoride was produced via the ^{18}O(p,n)^{18}F reaction by bombardment of enriched [^{18}O]water with 16.5 MeV protons using a MC16 cyclotron (Scanditronix, Uppsala, Sweden) at the Max Plnack Institute for Metabolism Research. All isolated radiochemical yields were not corrected for decay. Unless otherwise indicated, all radiochemical experiments were carried out at least in triplicate.

Before radiosyntheses [^{18}F]fluoride was preprocessed as follows. Aqueous [^{18}F]fluoride (0.05–49.5 GBq) was loaded onto an anion-exchange resin (QMA cartridge). It should be noted, that aqueous [^{18}F]fluoride was loaded onto the cartridge from the male side, whereas flushing, washing and $^{18}F^-$ elution were carried out from the female side of the cartridge. If the QMA cartridge had been loaded, flushed and eluted from the female side only, sometimes a significant amount of [^{18}F]fluoride remained on the resin (this is probably because QMA-light (46 mg) cartridges have a

single frit on the male side but four on the female side). If a solution of K_2CO_3 was used for $^{18}F^-$ elution, the resin was flushed with air (> 10 mL) and [^{18}F]fluoride was eluted into a reaction vial. If a solution of diaryliodonium salt precursor was used for a $^{18}F^-$ elution, the resin was washed with MeOH (1 mL) and [^{18}F]fluoride was eluted into a reaction vial.

Syntheses of ^{18}F-labeled arenes from (mesityl)(aryl)iodonium tetrafluoroborate precursors using small aliquots of $^{18}F^-$[16] as well as one-pot syntheses of ^{18}F-labeled compounds under "high base" conditions[17] were carried out according to the literature. ^{18}F-Labeled arenes from pinacolyl arylboronates (arylBPin) using small aliquots of $^{18}F^-$[18] and one-pot syntheses under "high base" conditions[19] were also carried out according to the literature. Afterwards, the reaction mixtures were cooled to room temperature, water (2 mL) was added and the mixtures were shaken vigorously for 30 s. Thereafter, RCCs were determined by radio-HPLC.

$Cu(OTf)_2$ and (tBuCN)$_2$CuOTf were handled and stored in a glove box. $(MeCN)_4CuOTf$ and $Cu(OTf)_2(Py)_4$ as pure substance were handled and stored for a short period of time (1–3 h) under ambient air and stored for a long period of time under Ar. Solutions of $Cu(OTf)_2$ and (tBuCN)$_2$CuOTf in DMF should be prepared immediately before use. Solutions of $(MeCN)_4CuOTf$ in DMF could be used for at least 6 h. Radiosyntheses with $Cu(OTf)_2$ were carried out under Ar. Radiosyntheses with $(MeCN)_4CuOTf$, (tBuCN)$_2$CuOTf and $Cu(OTf)_2(Py)_4$ were carried out under synthetic air (Linde Industriegase, Pullach, Germany).

All animal experiments were carried out in accordance with the EU directive 2010/63/EU for animal experiments and the German Animal Welfare Act (TierSchG, 2006), and were approved by regional authorities (LANUV NRW).

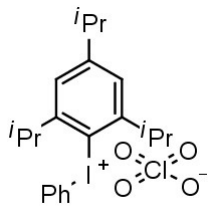
Chemistry

(Phenyl)(1,3,5-triisopropylphenyl)iodonium bromide: $BF_3 \cdot Et_2O$ (0.46 mL, 0.52g, 3.63 mmol) was added dropwise to an ice-cold solution of phenylboronic acid (0.177 g, 1.45 mmol) in anhydrous CH_2Cl_2 (10 mL) under Ar and the resulting mixture was stirred for 10 min. Thereafter, (diacetoxyiodo)-2,4,6-triisopropylbenzene,[1] (0.65 g, 1.45 mmol) was added, the cooling bath was removed and the mixture was stirred for a further 2 hours. The solvent was removed under reduced pressure. The residue was taken in CH_2Cl_2 (25 mL) and stirred vigorously with saturated NaBr (15 mL) for 15 min. The solution was repeatedly treated with saturated NaBr (4×15 mL), dried, filtered and concentrated under reduced pressure. The residue was recrystallized from CH_2Cl_2/Et_2O to give the desired iodonium salt (0.42 g, 59%) as a colorless solid (about 90% purity according to the ^{1}H-NMR spectrum). ^{1}H NMR (300 MHz, $CDCl_3$): ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.87–7.77 (m, 2 H), 7.45–7.22 (m, 3 H), 7.16–7.05 (m, 2 H), 3.47 (dt, *J* = 13.4, 6.7 Hz, 2 H), 2.92 (dt, *J* = 13.8, 6.9 Hz, 1 H), 1.31–1.19 (m, 18 H). ^{13}C NMR (75 MHz, $CDCl_3$) δ 153.7, 151.2, 132.3 131.2, 130.1, 126.9, 124.3, 120.0, 39.0, 34.1, 24.3, 23.7. MS (ESI): positive mode *m/z* = 407.1 ($[M]^+$); ESI HRMS: calcd for $C_{21}H_{28}I^+$: 407.1230; found: 407.1231.

(Phenyl)(1,3,5-triisopropylphenyl)iodonium tetrafluoroborate: $AgBF_4$ (82 mg, 0.42 mmol) was added to a solution of (phenyl)(1,3,5-triisopropylphenyl)iodonium bromide (0.172 g, 0.35 mmol) in MeOH (10 mL). After vigorous shaking of the mixture for 2 min while shielded from light, the precipitate of silver bromide was centrifuged off (4000 rpm, 10 min). The supernatant was concentrated under reduced pressure and the residue was recrystallized from CH_2Cl_2/Et_2O to give the desired iodonium salt (0.14 g, 80%) as a colorless solid. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.74–7.63 (m, 2 H), 7.60–7.52 (m, 1 H), 7.50–7.41 (m, *J* = 7.6 Hz, 2 H), 7.20 (s, 2 H), 3.27 (dt, *J* = 13.4, 6.7 Hz, 2 H), 2.98 (dt, *J* = 13.8, 6.9 Hz, 1 H), 1.29 (d, *J* = 6.9 Hz, 6 H), 1.25 (d, *J* = 6.7 Hz, 12 H). ^{19}F NMR (282 MHz, $CDCl_3$) δ -147.54. ^{13}C

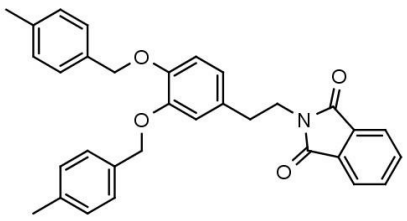
NMR (75 MHz, $CDCl_3$) δ 155.9, 152.6, 132.58, 132.55, 132.1, 125.5, 119.4, 111.9, 39.7, 34.2, 24.2, 23.6. MS (ESI): positive mode *m/z* = 407.1 ($[M]^+$); ESI HRMS: calcd for $C_{21}H_{28}I^+$: 407.1230; found: 407.1231.

(Phenyl)(1,3,5-triisopropylphenyl)iodonium perchlorate: $AgClO_4$ (75 mg, 0.36 mmol) was added



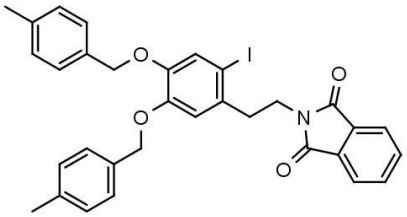
to a solution of (phenyl)(1,3,5-triisopropylphenyl)iodonium bromide (0.15 g, 0.31 mmol) in MeOH (10 mL). After vigorous shaking of the mixture for 2 min while shielded from light, the precipitate of silver bromide was centrifuged off (4000 rpm, 10 min). The supernatant was concentrated under reduced pressure and the residue was recrystallized from acetone/Et_2O to give the desired iodonium salt (0.12 g, 77%) as a colorless solid. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.71 (dd, *J* = 8.6, 1.1 Hz, 2 H), 7.63–7.36 (m, 3 H), 7.19 (s, 2 H), 3.38–3.21 (m, 2 H), 2.97 (dt, *J* = 13.8, 6.9 Hz, 1 H), 1.28 (d, *J* = 6.9 Hz, 6 H), 1.26 (d, *J* = 6.8 Hz, 12 H). ^{13}C NMR (75 MHz, $CDCl_3$) δ 155.7, 152.4, 132.8, 132.5, 132.0, 125.40, 120.40, 112.4, 39.7, 34.1, 24.3, 23.6. MS (ESI): positive mode *m/z* = 407.1 ($[M]^+$); MS (ESI): negative mode *m/z* = 98.9 ($[ClO_4]^-$); ESI HRMS: calcd for $C_{21}H_{28}I^+$: 407.1230; found: 407.1234.

***N*-[3,4-di(4-methylbenzyloxy)phenethyl]phthalimide:** A suspension of K_2CO_3 (2.15 g, 15.56



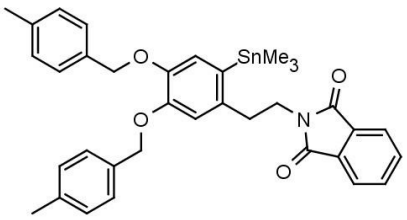
mmol) in a solution of *N*-(3,4-dihydroxyphenethyl)phthalimide,[9] (2 g, 7.05 mmol) and 4-methylbenzyl bromide (2.9 g, 15.67 mmol) in anhydrous DMF (30 mL) was stirred at 50 °C for 48 h. The reaction mixture was concentrated under reduced pressure. The residue was taken up in Et_2O and H_2O (100 mL of each), the organic fraction was washed with H_2O (8×50 mL), brine (2×20 mL), dried and concentrated under reduced pressure. The residue was purified by column chromatography (EtOAc:hexane = 3:10) to give the title compound (2.4 g) as a colorless solid. R_f = 0.27, EtOAc:hexane = 3:10. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.90–

7.79 (m, 2 H), 7.76–7.66 (m, 2 H), 7.38–7.28 (m, 4 H), 7.17 (dd, J = 7.7, 3.4 Hz, 4 H), 6.86 (d, J = 7.9 Hz, 2 H), 6.77 (d, J = 8.1 Hz, 1 H), 5.08 (s, 2 H), 5.05 (s, 2 H), 3.95–3.82 (m, 2 H), 2.97–2.84 (m, 2 H), 2.37 (s, 6 H). ^{13}C NMR (75 MHz, $CDCl_3$) δ 168.1, 149.1, 147.8, 137.4, 137.3, 134.4, 134.3, 133.9, 132.1, 131.3, 129.05, 129.04, 127.5, 127.4, 123.2, 121.7, 115.8, 115.50, 71.34, 71.26, 39.3, 34.0, 21.17, 21.16. MS (ESI): positive mode m/z = 1000.5 ($[2M + NH_4]^+$), 514.2 ($[M + Na]^+$), 509.3 ($[M + NH_4]^+$), 492.2 ($[M + H]^+$); ESI HRMS: calcd for $C_{32}H_{29}NO_4Na^+$: 514.1989; found: 514.1986; calcd for $C_{32}H_{33}N_2O_4^+$: 509.2435; found: 509.2439.

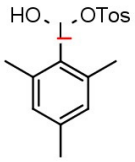


***N*-[3,4-Di(4-methylbenzyloxy)-6-iodophenethyl]phthalimide:** Adopted from the method of X. Su et al.[20] A solution of I_2 (1.03 g, 4.06 mmol) in CH_2Cl_2 (50 mL) was added dropwise to a vigorously stirred solution of *N*-[3,4-di(4-methylbenzyloxy)phenethyl]phthalimide (2 g, 4.06 mmol) and silver trifluoroacetate (0.9 g, 4.07 mmol) in CH_2Cl_2 (50 mL). After the addition was complete the resulting suspension was stirred for a further 15 min, washed with 10% $NaHSO_3$ (3×40 mL), H_2O (2×40 mL), brine (2×20 mL) and filtered through Celite®. The resulting solution was dried and concentrated under reduced pressure. The residue was recrystallized from CH_2Cl_2/pentane to give the title compound (2.25 g, 90%) as a colorless solid. R_f = 0.17, EtOAc:hexane = 1:6. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.90–7.79 (m, 2 H), 7.77–7.66 (m, 2 H), 7.38–7.28 (m, 2 H), 7.30 – 7.22 (m, 3 H), 7.16 (t, J = 7.4 Hz, 4 H), 6.82 (s, 1 H), 5.03 (s, 2 H), 4.92 (s, 2 H), 3.90 (t, J = 7.4 Hz, 2 H), 3.03 (t, J = 7.4 Hz, 2 H), 2.37 (s, 3 H), 2.36 (s, 3 H). ^{13}C NMR (75 MHz, $CDCl_3$) δ 168.1, 149.4, 148.5, 137.62, 137.57, 133.90, 133.87, 133.71, 133.66, 132.1, 129.13, 129.10, 127.50, 127.47, 125.5, 123.2, 116.4, 89.0, 71.4, 71.3, 38.7, 37.9, 21.2 (× 2). MS (ESI): positive mode m/z = 1252.3 ($[2M + NH_4]^+$), 640.1 ($[M + Na]^+$), 635.2 ($[M + NH_4]^+$); ESI HRMS: calcd for $C_{32}H_{28}INO_4Na^+$: 640.0955; found: 640.0949; calcd for $C_{32}H_{32}IN_2O_4^+$: 635.1401; found: 635.1394.

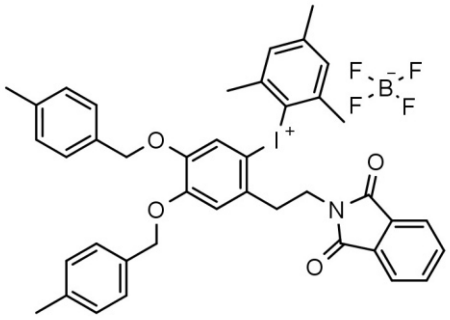
***N*-[3,4-Di(4-methylbenzyloxy)-6-trimethylstannylphenethyl]phthalimide:** Sn_2Me_6 (1.22 g, 3.73 mmol) was added to a solution of *N*-[3,4-di(4-methylbenzyloxy)-6-iodophenethyl]phthalimide (1 g, 1.62 mmol) and $Pd(PPh_3)_4$ (0.37 g, 0.32 mmol) in anhydrous toluene (20 mL) in a glove box. The reaction mixture was stirred at 110 °C for 3 h, cooled to ambient temperature, filtered through Celite® and concentrated under reduced pressure. The residue was purified by column chromatography (EtOAc:hexane = 3:10) to give the title compound (0.85 g, 80%) as a colorless solid. R_f = 0.38, EtOAc:hexane = 3:10. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.87 (dd, *J* = 5.5, 2.9 Hz, 2 H), 7.73 (dd, *J* = 5.5, 3.0 Hz, 2 H), 7.35 (dd, *J* = 8.0, 3.8 Hz, 4 H), 7.18 (dd, *J* = 7.8, 3.7 Hz, 4 H), 6.98 (s, 2 H), 5.11 (s, 2 H), 5.06 (s, 2 H), 3.87–3.78 (m, 2 H), 3.08–2.80 (m, 2 H), 2.37 (s, 6 H), 0.50–0.20 (s, 9 H). ^{13}C NMR (75 MHz, $CDCl_3$) δ 168.1, 149.7, 147.3, 138.2, 137.40, 137.36, 134.6, 134.2, 133.9, 133.6, 132.2, 129.09, 129.06, 127.57, 127.54, 123.3, 123.2, 115.7, 71.7, 71.0, 39.8, 37.0, 21.19, 21.17, –8.1. MS (ESI): positive mode *m/z* = 1326.4 ($[2M + NH_4]^+$), 678.2 ($[M + Na]^+$), 656.2 ($[M + H]^+$); ESI HRMS: calcd for $C_{35}H_{37}SnNO_4Na^+$: 678.1644; found: 678.1653 (correct isotopic pattern); calcd for $C_{35}H_{38}SnNO_4^+$: 678.1644; found: 678.1653 (correct isotopic pattern).



Hydroxy{[(4-methylphenyl)sulfonyl]oxy}(2,4,6-trimethylphenyl)-λ$_3$–iodine:[21] $TosOH·H_2O$ (1.05 g, 0.55 mmol) was added to an ice-cold solution of (diacetoxyiodo)-2,4,6-trimethylbenzene (2 g, 5.49 mmol) in MeCN (5 mL) and the reaction mixture was stirred at the same temperature for 15 min. The resulting yellow solution was concentrated under reduced pressure. The residue was recrystallized two times from CH_2Cl_2/Et_2O to give the title compound (1.7 g, 70%) as a colorless solid. The spectral data were in accordance with the literature.[21]



{2-[2-(1,3-Dioxoisoindol-2-yl)ethyl]-4,5-bis[(4-methylphenyl)methoxy]phenyl}(2,4,6-trimethyl-phenyl)iodonium tetrafluoroborate: A solution of hydroxy{[(4-methylphenyl)sulfonyl]oxy}-(2,4,6-trimethylphenyl)-λ_3–iodine (0.49 g, 1.13 mmol) in CH_2Cl_2 (10 mL) was added to a solution of *N*-[3,4-di(4-methylbenzyloxy)-6-trimethylstannylphenethyl]phthalimide (0.73 g, 1.12 mmol) in CH_2Cl_2 (10 mL) and the reaction mixture was stirred for 90 min. After that the mixture was concentrated under reduced pressure. The residue was purified by column chromatography (CH_2Cl_2:MeOH = 10:1) to give {2-[2-(1,3-dioxoisoindol-2-yl)ethyl]-4,5-bis[(4-methylphenyl)methoxy]phenyl}(2,4,6-trimethylphenyl)iodonium tosylate (0.61 g, 56%; contained about 8.5% hexane according to ^{1}H-NMR-spectrum) as a yellow viscous oil. R_f = 0.35, CH_2Cl_2:MeOH = 10:1. ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.83 (dd, *J* = 5.4, 3.1 Hz, 2 H), 7.73 (dd, *J* = 5.5, 3.0 Hz, 2 H), 7.57 (d, *J* = 8.1 Hz, 2 H), 7.18 (dd, *J* = 29.6, 8.0 Hz, 4 H), 7.10 (q, *J* = 8.2 Hz, 4 H), 7.03 (d, *J* = 7.9 Hz, 2 H), 6.96 (d, *J* = 18.4 Hz, 3 H), 6.84 (s, 1 H), 4.92 (s, 2 H), 4.88 (s, 2 H), 3.81 (t, *J* = 7.0 Hz, 2 H), 3.06 (t, *J* = 7.1 Hz, 2 H), 2.50 (s, 6 H), 2.36 (s, 3 H), 2.35 (s, 3 H), 2.32 (s, 3 H), 2.30 (s, 3 H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 167.8, 151.9, 149.0, 143.4, 143.0, 142.1, 139.1, 138.1, 137.8, 134.2, 133.5, 132.8, 132.7, 131.7, 130.2, 129.31, 129.26, 128.3, 127.4, 127.0, 125.9, 123.4, 122.8, 120.6, 115.5, 105.8, 71.2, 70.9, 37.5, 36.1, 26.9 (×2), 21.3, 21.2, 21.0. MS (ESI): positive mode *m/z* = 736.7 ($[M]^+$); MS (ESI): negative mode *m/z* = 171.0 ($[TosO]^-$); ESI HRMS: calcd for $C_{41}H_{39}INO_4^+$: 736.1918; found: 736.1914.



To a solution of tosylate salt (0.59 g, 0.59 mmol; 92% purity) in CH_2Cl_2 (20 mL) saturated $NaBF_4$ (10 mL) was added and the mixture was vigorously stirred for 10 min. The aqueous solution and precipitate were separated off, saturated $NaBF_4$ (10 mL) was added and the mixture was vigorously

stirred for 10 min (×5). The organic fraction was dried and concentrated under reduced pressure to give the title compound (0.47 g, 96%) as a faint yellow foam. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.86–7.77 (m, 2 H), 7.77–7.67 (m, 2 H), 7.20 (dd, *J* = 22.7, 7.9 Hz, 4 H), 7.12 (s, 4 H), 7.04 (d, *J* = 6.2 Hz, 3 H), 6.93 (s, 1 H), 5.00 (s, 2 H), 4.93 (s, 2 H), 3.83 (t, *J* = 6.9 Hz, 2 H), 3.07 (t, *J* = 6.9 Hz, 2 H), 2.48 (s, 6 H), 2.35 (s, 9 H). ^{19}F NMR (282 MHz, $CDCl_3$) δ –148.80. ^{13}C NMR (75 MHz, $CDCl_3$) δ 167. 9, 152.7, 149.4, 144.5, 142.7, 138.2, 137.8, 134.3, 134.0, 132.6, 132. 5, 131.6, 130.8, 129.34, 129.28, 127.5, 127.0, 123.5, 120.7, 118.9, 115.8, 102.0, 71.4, 71.1, 37.5, 36.5, 26.8 (×2), 21.2, 21.0. MS (ESI): positive mode *m/z* = 736.2 ($[M]^+$); MS (ESI): negative mode *m/z* = 87.0 ($[BF_4]^-$); ESI HRMS: calcd for $C_{41}H_{39}INO_4^+$: 736.1918; found: 736.1921.

{4-[(2*S*)-2-[(*tert*-Butoxycarbonyl)amino]-3-methoxy-3-oxopropyl]phenyl}(2,4,6-trimethylphenyl)iodonium tetrafluoroborate: A solution of hydroxy{[(4-methylphenyl)sulfonyl]oxy}(2,4,6-trimethylphenyl)-λ$_3$–iodine (0.68 g, 1.57 mmol) in CH_2Cl_2 (20 mL) was added to a solution of methyl *N*-*tert*-butyloxycarbonyl-L-(4-trimethylstannylphenyl)alaninate[10] (0.66 g, 1.49 mmol) in CH_2Cl_2 (60 mL) and the reaction mixture was stirred for 24 h. After that the mixture was concentrated under reduced pressure. The residue was purified by column chromatography (CH_2Cl_2:MeOH = 8:1) affording a colorless foam which was triturated with Et_2O to give {4-[(2*S*)-2-[(*tert*-butoxycarbonyl)amino]-3-methoxy-3-oxopropyl]phenyl}(2,4,6-trimethylphenyl)iodonium tosylate (0.52 g, 53%) as a colorless solid which was directly used for the next step. R_f = 0.38, MeOH:CH_2Cl_2 = 1:8.

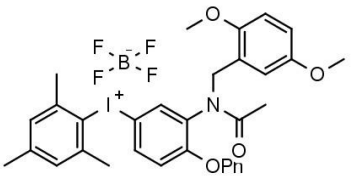
To a solution of tosylate salt (0.52 g, 0.79 mmol) in CH_2Cl_2 (20 mL) saturated $NaBF_4$ (10 mL) was added and the mixture was vigorously stirred for 10 min. The aqueous solution and precipitate were

separated off (if necessary to separate organic and aqueous phases the mixture was centrifuged at 4000 rpm for 10 min), saturated $NaBF_4$ (10 mL) was added and the mixture was vigorously stirred for 10 min (×12). The organic fraction was dried and concentrated under reduced pressure. The residue was recrystallized from CH_2Cl_2/Et_2O and EtOAc/Et_2O to give the title compound (0.36 g, 39% over two steps) as a colorless foam. The mother liqueur was concentrated under reduced pressure and the residue was recrystallized from EtOAc/Et_2O to give a second crop of the title compound (60 mg, overall 43% over two steps). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.64 (d, *J* = 8.4 Hz, 2 H), 7.23 (d, *J* = 8.2 Hz, 2 H), 7.12 (s, 2 H), 5.08 (d, *J* = 8.1 Hz, 1 H), 4.53 (m, 1 H), 3.70 (s, 3 H), 3.24–3.08 (m, 1 H), 3.06–2.90 (m, 1 H), 2.62 (s, 6 H), 2.36 (s, 3 H), 1.32 (s, 9 H). ^{19}F NMR (376 MHz, $CDCl_3$) δ –147.33. ^{13}C NMR (101 MHz, $CDCl_3$) δ 171.6, 155.0, 144. 8, 142.8, 141.5, 133.5, 133.1, 130.5, 119.1, 108.7, 80.2, 54.1, 52.5, 38.0, 28.2, 27.1, 21.1. MS (ESI): positive mode *m/z* = 524.4 ($[M]^+$); MS (ESI): negative mode *m/z* = 87.0 ($[BF_4]^-$); ESI HRMS: calcd for $C_{24}H_{31}INO_4^+$: 524.1292; found: 524.1292.

***N*-(5-Trimethylstannyl-2-phenoxyphenyl)-*N*-(2,5-dimethoxybenzyl)acetamide:** Sn_2Me_6 (1.6 g, 4.88 mmol) was added to a solution of *N*-(5-bromo-2-phenoxyphenyl)-*N*-(2,5-dimethoxybenzyl)acetamide[12] (1.25 g, 2.74 mmol) and $Pd(PPh_3)_4$ (0.45 g, 0.39 mmol) in anhydrous toluene (20 mL) in a glove box. The reaction mixture was stirred at 110 °C for 16 h, cooled to ambient temperature, filtered through Celite® and concentrated under reduced pressure. The residue was purified by column chromatography (EtOAc:hexane = 3:10) affording the title compound (0.85 g, 80%) as a colorless viscous oil. R_f = 0.12, EtOAc:hexane = 3:10. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.38–7.23 (m, 3 H), 7.20–7.05 (m, 1 H), 7.05–6.60 (m, 7 H), 5.21 (d, *J* = 14.4 Hz, 1 H), 4.63 (d, *J* = 14.4 Hz, 1 H), 3.67 (s, 3 H), 3.50 (s, 3 H), 1.96 (s, 3 H), 0.21 (s, 9 H). ^{13}C NMR (75 MHz, $CDCl_3$) δ 170.9, 156.1, 153.7, 153.5, 151.9, 137.5, 136.2, 136.1, 132.9, 129.7, 126.7, 123.8, 119.2, 118.1, 116.5, 113.4, 111.4, 55.8,

55.6, 45.8, 22.3, –9.5. MS (ESI): positive mode *m/z* = 564.1 ([M + Na]$^+$), 542.2 ([M + H]$^+$); ESI HRMS: calcd for $C_{26}H_{32}SnNO_4^+$: 542.1353; found: 542.1337 (correct isotopic pattern).

(3-{*N*-[(2,5-Dimethoxyphenyl)methyl]acetamido}-4-phenoxyphenyl)(2,4,6-trimethylphenyl)-



iodonium tetrafluoroborate: A solution of hydroxy{[(4-methylphenyl)sulfonyl]oxy}(2,4,6- trimethylphenyl)-λ_3–iodine (0.82 g, 1.89 mmol) in CH_2Cl_2 (20 mL) was added to a solution of *N*-(5-trimethylstannyl-2-phenoxyphenyl)-*N*-(2,5-dimethoxybenzyl)acetamide (1.02 g, 1.89 mmol) in CH_2Cl_2 (40 mL) and the reaction mixture was stirred for 3 h. After that the mixture was concentrated under reduced pressure. The residue was purified by column chromatography (CH_2Cl_2:MeOH = 10:1) to give (3-{*N*-[(2,5-dimethoxyphenyl)methyl]acetamido}-4-phenoxyphenyl)(2,4,6-trimethylphenyl)iodonium tosylate (0.78 g, 52%) as a yellow foam which was directly used for the next step. R_f = 0.45, MeOH:CH_2Cl_2 = 1:10.

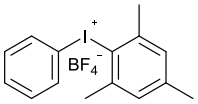
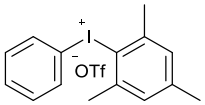
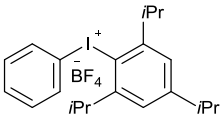
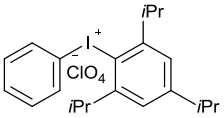
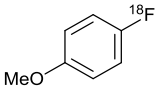
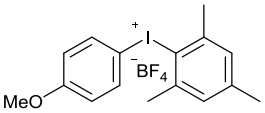
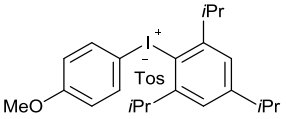
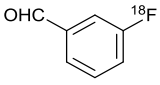
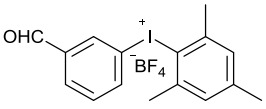
To a solution of tosylate salt (0.72 g, 0.91 mmol) in CH_2Cl_2 (20 mL) saturated $NaBF_4$ (10 mL) was added and the mixture was vigorously stirred for 10 min. The aqueous solution and precipitate were separated off (if necessary to separate organic and aqueous phases the mixture was centrifuged at 4000 rpm for 10 min), saturated $NaBF_4$ (10 mL) was added and the mixture was vigorously stirred for 10 min (×12). The organic fraction was dried, filtered through Celite® and concentrated under reduced pressure. The residue was recrystallized from CH_2Cl_2/Et_2O to give the title compound (0.69 g, 52% over two steps) as a faint yellow solid. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.67 (d, *J* = 7.2 Hz, 1 H), 7.39 (t, *J* = 7.8 Hz, 2 H), 7.26–7.18 (m, 2 H), 7.16 (s, 2 H), 7.07 (s, 2 H), 6.90 (d, *J* = 7.9 Hz, 2 H), 6.85–6.45 (m, 4 H), 5.13 (d, *J* = 14.2 Hz, 1 H), 4.63 (d, *J* = 14.3 Hz, 1 H), 3.68 (s, 3 H), 3.41 (s, 3 H), 2.51 (s, 6 H), 2.36 (s, 3 H), 1.87 (s, 3H). ^{19}F NMR (282 MHz, $CDCl_3$) δ –146.90. ^{13}C NMR

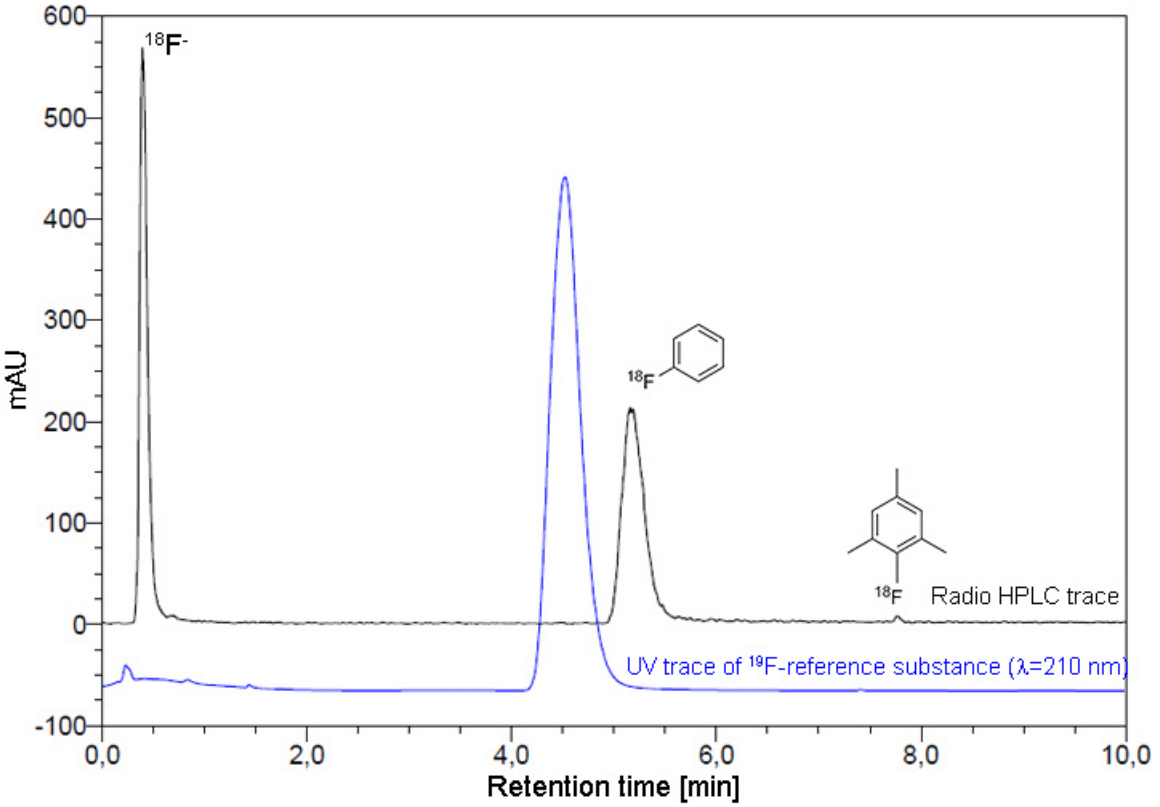
(75 MHz, $CDCl_3$) δ 170.4, 157.9, 153.9, 153.5, 151.7, 144.6, 142.5, 135.2, 134.9, 130.41, 130.36, 125.7, 125.5, 120.3 (×2), 119.8, 119.5, 116.4, 114.0, 111.5, 100.2, 55.74, 55.66, 45.4, 26. 9, 22.1, 21.1. MS (ESI): positive mode *m/z* = 622.2 ($[M]^+$); MS (ESI): negative mode *m/z* = 708.0 ($[M + BF_4 - H]^-$), 87.0 ($[BF_4]^-$); ESI HRMS: calcd for $C_{32}H_{33}INO_4^+$: 622.1449; found: 622.1449.

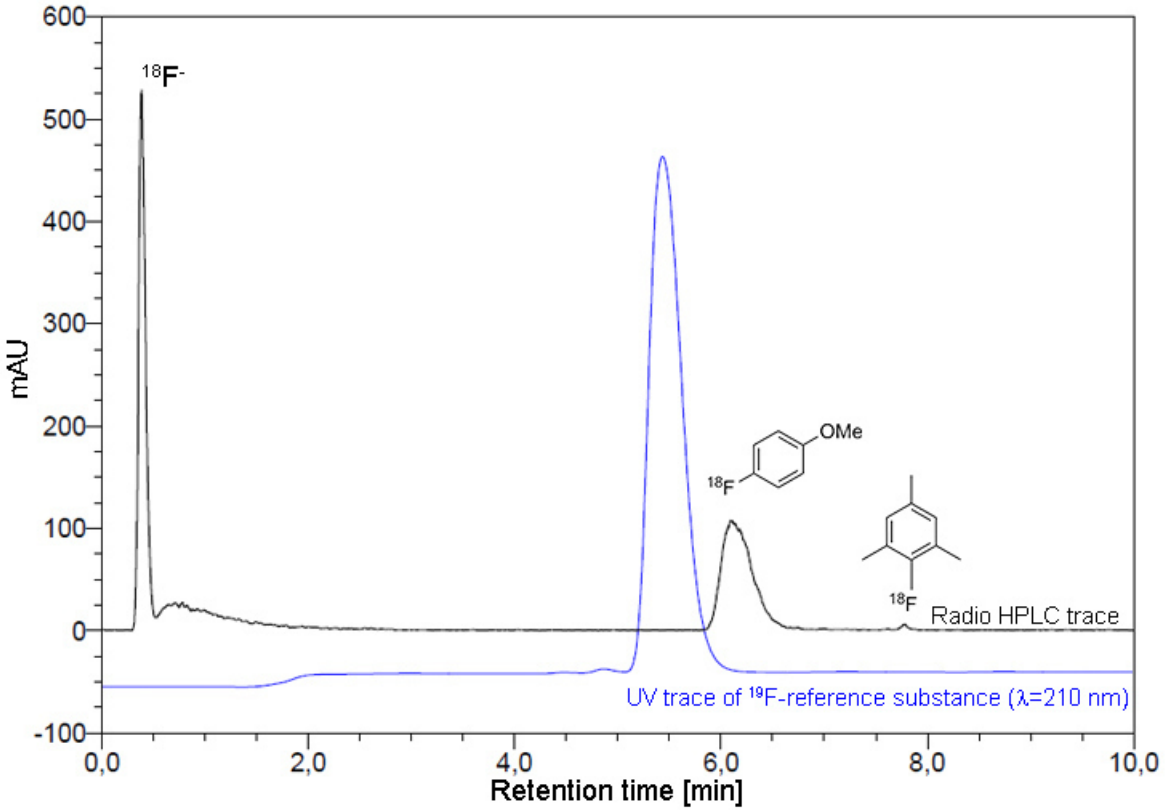
Radiochemistry

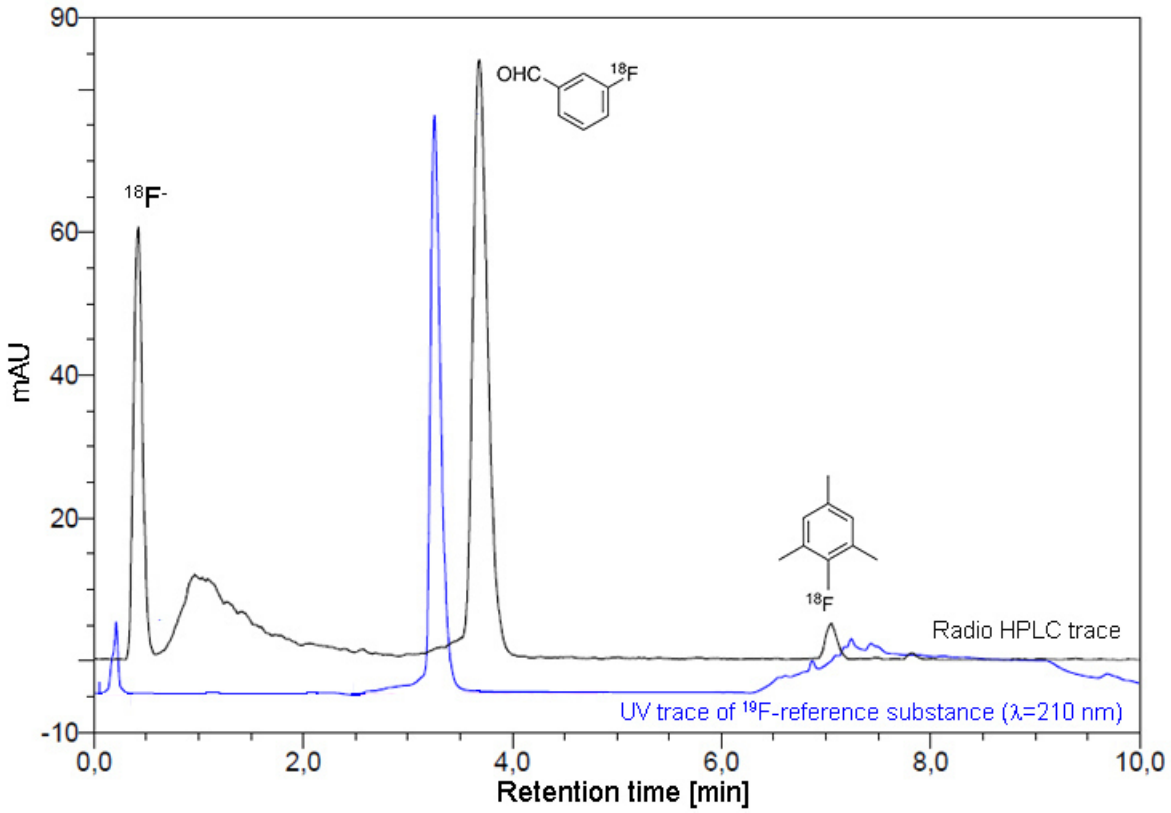
$Cu(OTf)_2$-Mediated synthesis of ^{18}F-labeled arenes from (mesityl)(aryl)iodonium precursors using K_2CO_3 in MeOH for [^{18}F]fluoride elution. General procedure 1 – (GP1): [^{18}F]Fluoride was eluted from a QMA cartridge with K_2CO_3 (0,5 mg, 4 µmol) in MeOH (0.5 mL), methanol was evaporated under He flow at 70–80 °C, the residue was taken up in DMF (200 µL) containing the corresponding iodonium salt precursor (12 µmol), $Cu(OTf)_2$ (4 mg, 12 µmol) and 18-crown-6 (8.8 mg, 30 µmol) and, unless otherwise stated, the resulting solution was heated at 85 °C for 20 min. Afterwards the reaction mixture was cooled to room temperature, water (2 mL) was added and the reaction mixture was shaken vigorously for 30 s. Thereafter, RCC was determined by radio-HPLC (Method A: [^{18}F]fluorobenzene t_R=5.2 min; [^{18}F]fluoroanisole t_R=6.1 min; 3-[^{18}F]FBA t_R=3.8 min).

Table 1 $Cu(OTf)_2$-Mediated synthesis of ^{18}F-labeled arenes from (mesityl)(aryl)iodonium precursors using K_2CO_3 in MeOH for [^{18}F]fluoride elution

entry	product	Precursor	Reaction time [min]	Temperature [°C]	RCC [%]
1			10	85	15
			20	85	41
			30	85	64
			20	100	45
			20	120	24
			20	85	35
			20	85	36
			20	85	35
2			20	85	29
			30	85	29
			40	85	32
			20	100	18
			20	85	13
3			20	85	50


600
500
400
300
200
100
0
-100
mAU
$^{18}F^-$
^{18}F
^{18}F
Radio HPLC trace
UV trace of ^{19}F-reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]

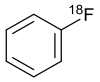
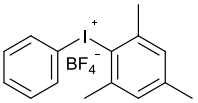
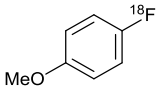
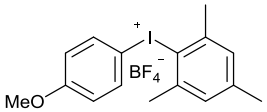
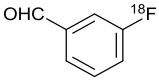
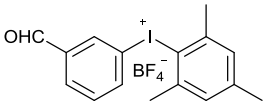

600
500
400
300
200
100
0
-100
mAU
$^{18}F^-$
OMe
^{18}F
^{18}F
Radio HPLC trace
UV trace of ^{19}F-reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]

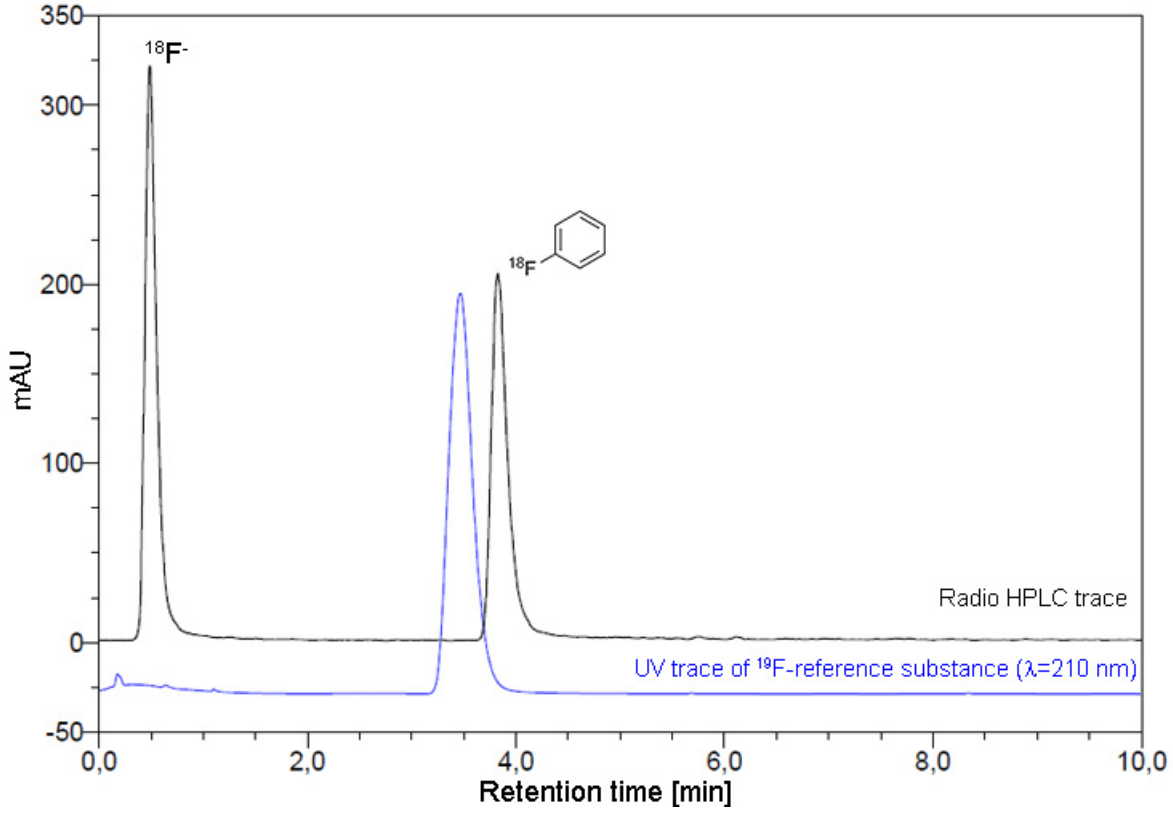


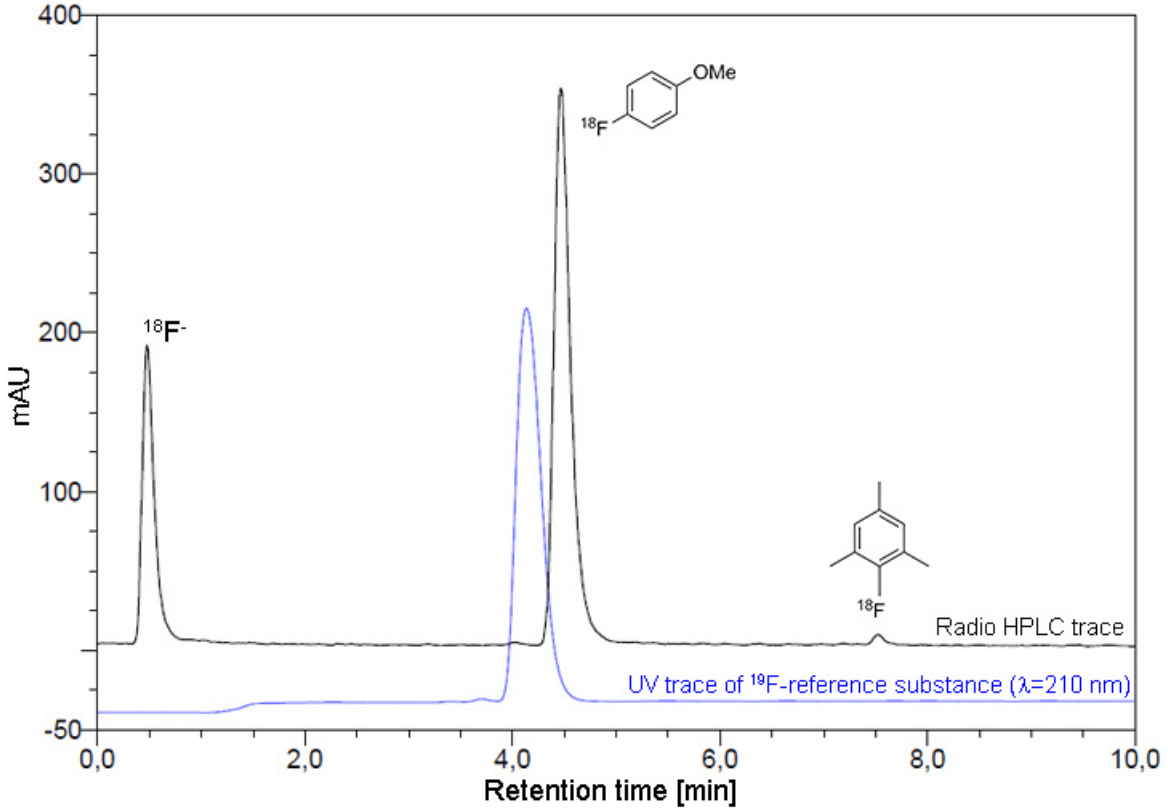
One-pot synthesis of ^{18}F-labeled compounds from (mesityl)(aryl)iodonium tetrafluoroborate precursors under "low base" conditions. General procedure 2 – (GP2):

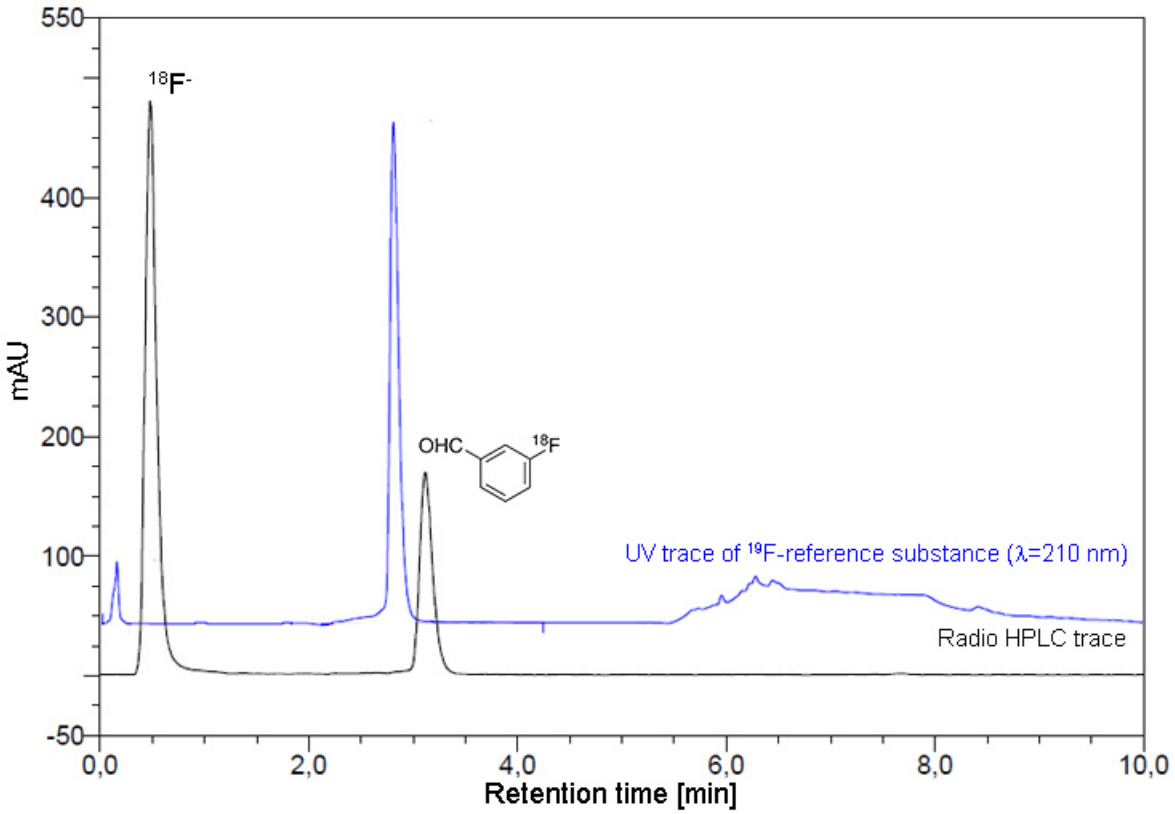
[^{18}F]Fluoride was eluted from an anion exchange resin with a solution of K_2CO_3 (0.11 mg, 0.8 µmol) in water (200 µL). The cartridge was washed further with a solution of 18-crown-6 (0.47 mg, 1.8 µmol) in MeCN (1 mL). The combined fractions were concentrated under reduced pressure at 95 °C and the residue was azeotropically dried with MeCN (1 mL; two times) under air. The residue was taken up in a solution of $(MeCN)_4CuOTf$ (4.2 mg, 12 µmol) in DMF (300 µL) and heated at 85 °C for 20 min under air. The reaction mixture was cooled to room temperature, diluted with water (2 mL) and shaken vigorously for 30 s. Thereafter, RCC was determined by radio-HPLC. (Method A: [^{18}F]fluorobenzene t_R=5.2 min; [^{18}F]fluoroanisole t_R=6.1 min; 3-[^{18}F]FBA t_R=3.8 min).

Table 2 One-pot synthesis of ^{18}F-labeled compounds from (mesityl)(aryl)iodonium tetrafluoroborate precursors under “low base” conditions.

Entry	Product	Precursor	RCC [%]
1			28
2			56
3			26

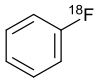
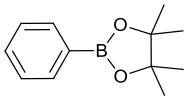
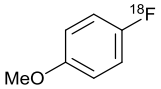
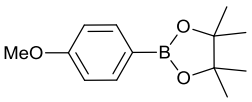
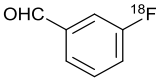
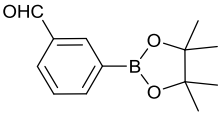

350
300
200
100
0
-50
mAU
$^{18}F^-$
^{18}F
Radio HPLC trace
UV trace of ^{19}F-reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]

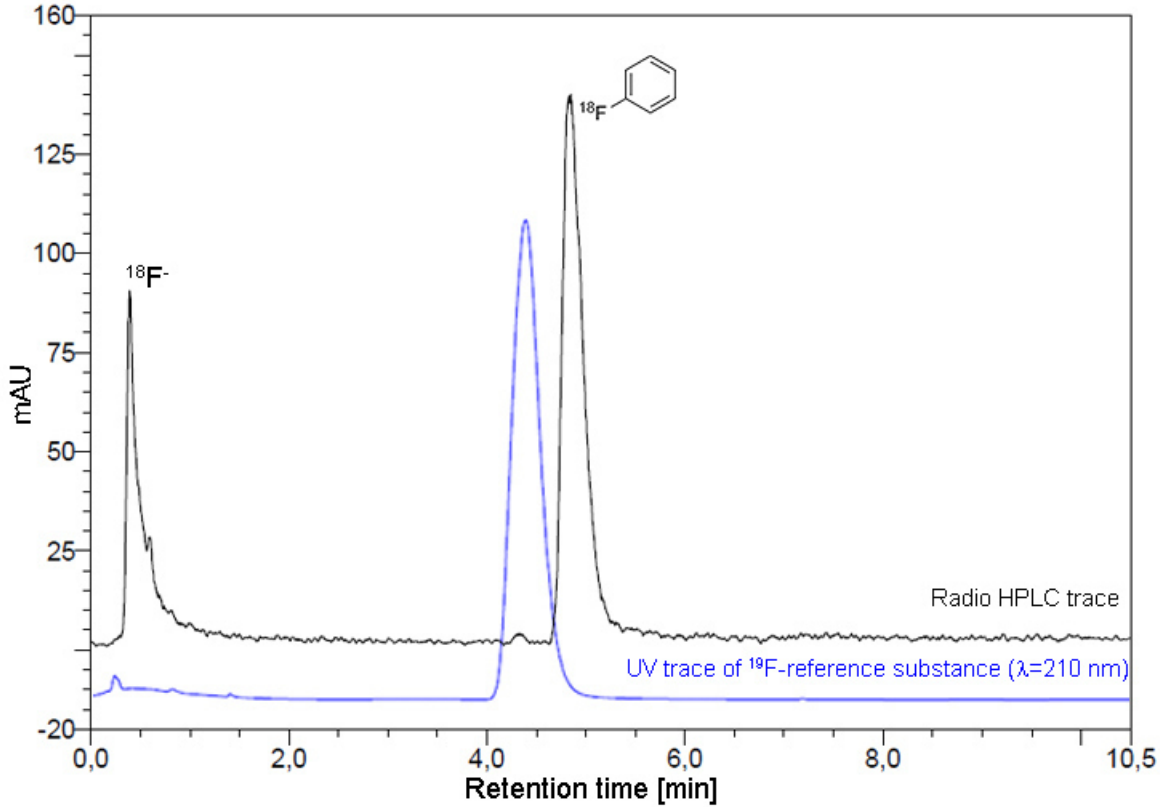

400
300
200
100
-50
mAU
$^{18}F^-$
OMe
^{18}F
^{18}F
Radio HPLC trace
UV trace of ^{19}F-reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]

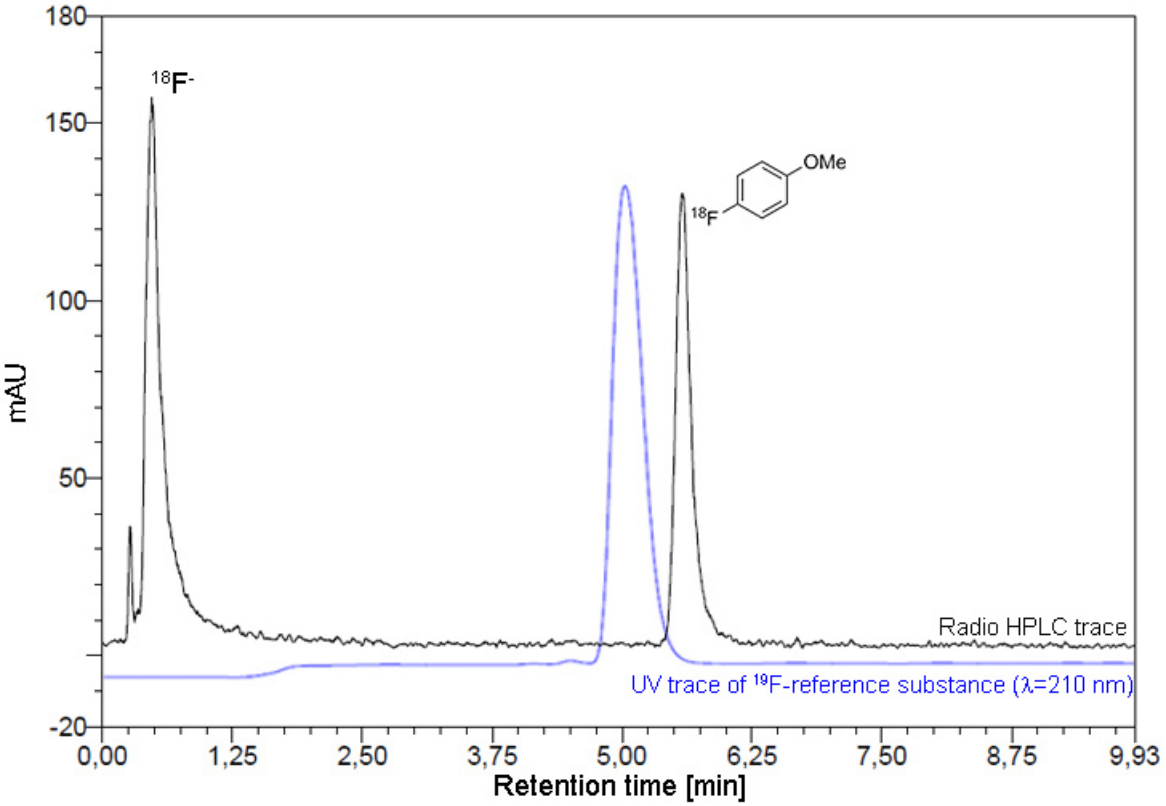


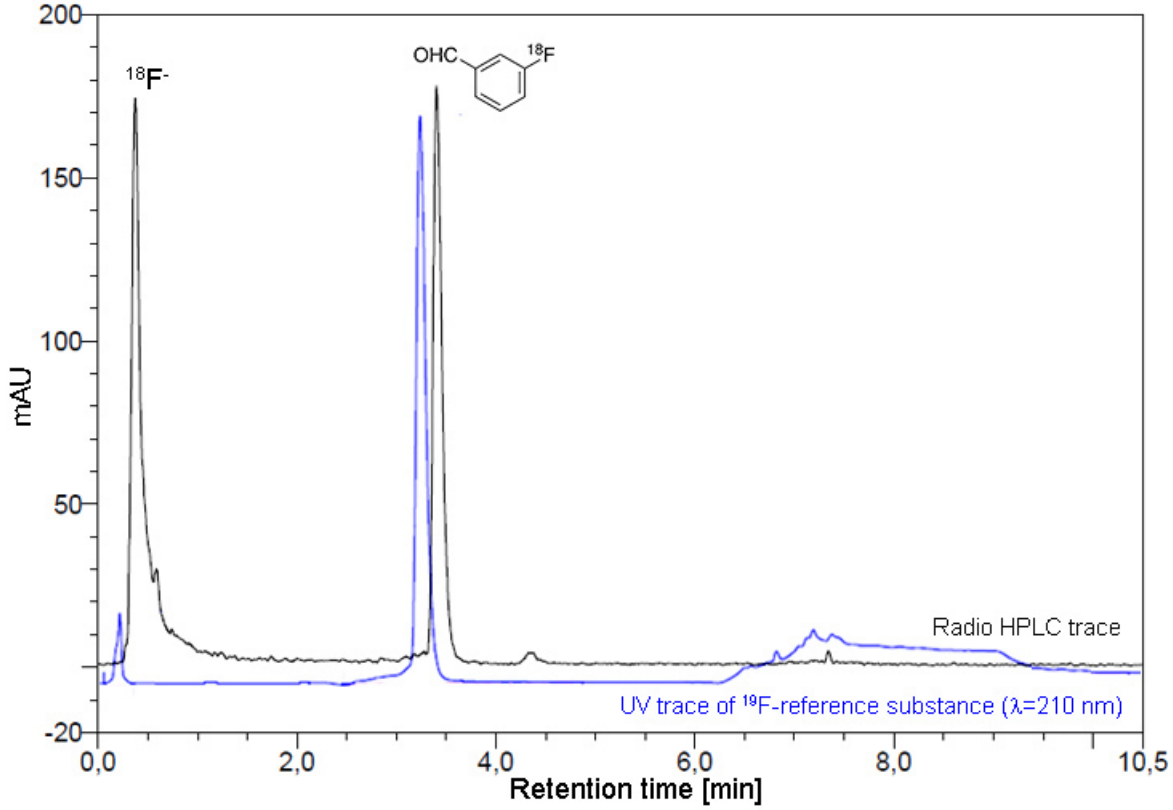
One-pot synthesis of ^{18}F-labeled compounds from pinacolyl arylboronates (arylBPin) under "low base" conditions. General procedure 3 – (GP3): $^{18}F^-$ (100–750 MBq) was eluted from a QMA cartridge with a solution of K_2CO_3 (0.06 mg, 0.43 µmol) and K2.2.2. (0.27 mg, 0.72 µmol) in 80% MeCN (1 mL). The solvent was evaporated under reduced pressure at 95 °C and the residue was azeotropically dried with MeCN (1 mL; two times) under air. A solution of $Cu(OTf)_2(py)_4$ (3.6 mg, 5.3 µmol) in DMF (150 µL) followed by a solution of arylboronic acid pinacol ester (60 µmol) in DMF:MeCN (5:1) (180 µL) were added to the residue and the reaction mixture was heated at 110 °C for 20 min under air. The reaction mixture was cooled to room temperature, diluted with water (2 mL) and shaken vigorously for 30 s. Thereafter, RCC was determined by radio-HPLC. (Method A: [^{18}F]fluorobenzene t_R=5.2 min; [^{18}F]fluoroanisole t_R=6.1 min; 3-[^{18}F]FBA t_R=3.8 min).

Table 3 One-pot synthesis of ^{18}F-labeled compounds from pinacolyl arylboronates (arylBPin) under “low base” conditions.

Entry	Product	Precursor	RCC [%]
1			64
2			42
3			41

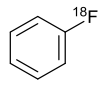
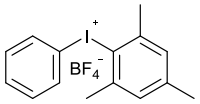
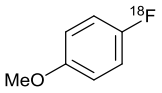
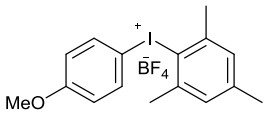
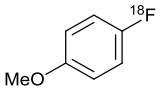
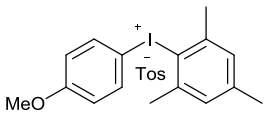
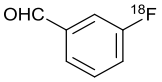
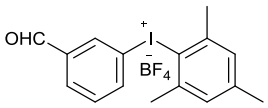
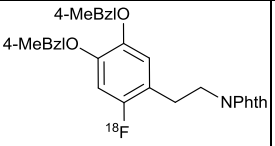
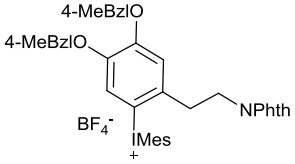
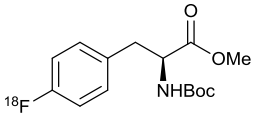
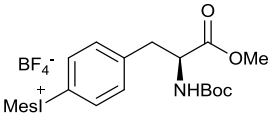
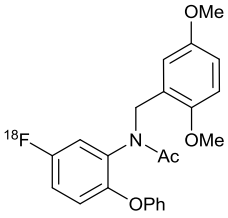
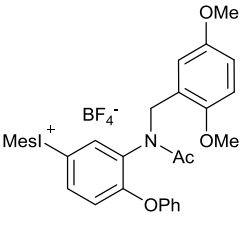

160
125
100
75
50
25
-20
mAU
$^{18}F^-$
^{18}F
Radio HPLC trace
UV trace of ^{19}F-reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,5
Retention time [min]

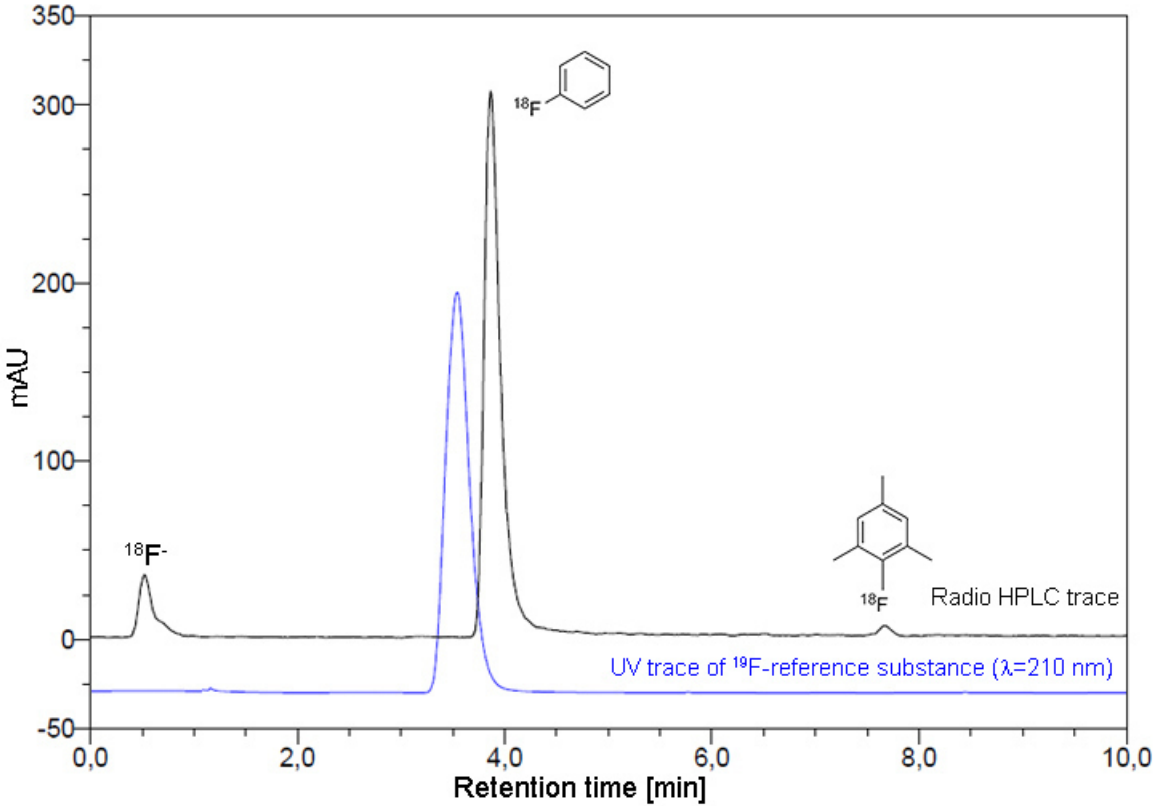

180
150
100
50
-20
mAU
$^{18}F^-$
OMe
^{18}F
Radio HPLC trace
UV trace of ^{19}F-reference substance (λ=210 nm)
0,00
1,25
2,50
3,75
5,00
6,25
7,50
8,75
9,93
Retention time [min]

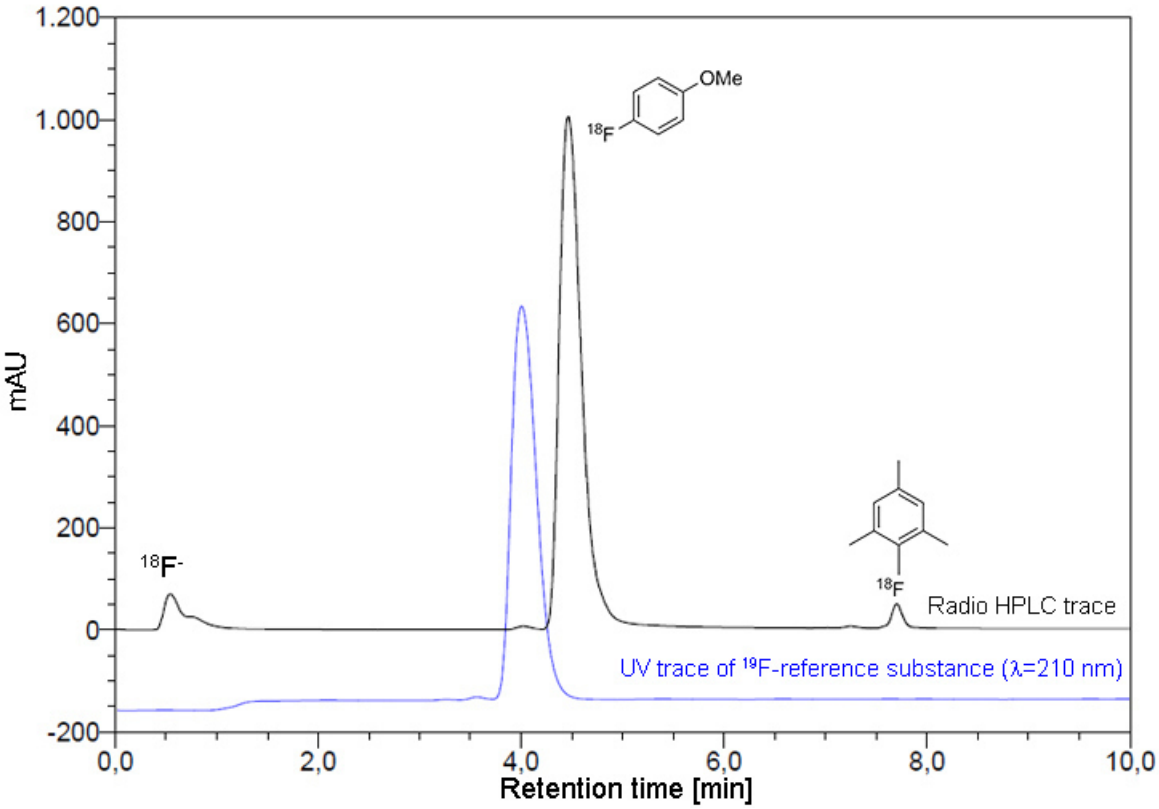


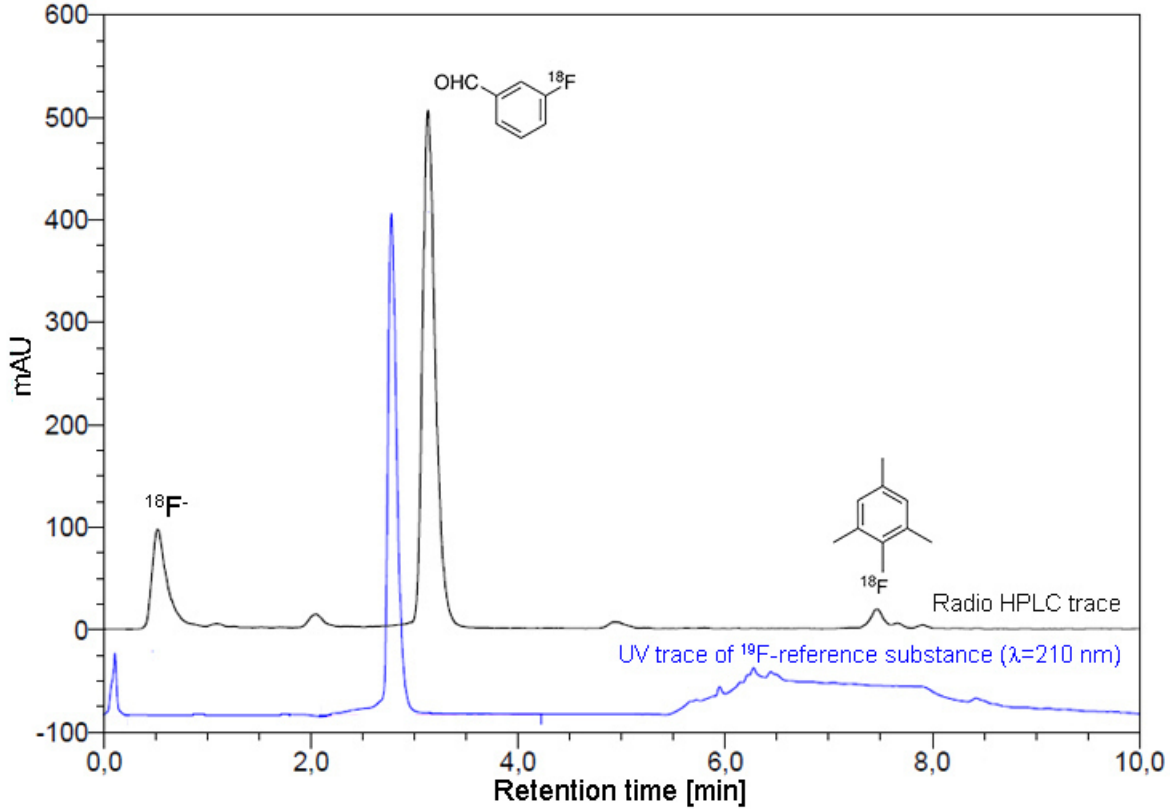
Synthesis of ^{18}F-labeled compounds from (mesityl)(aryl)iodonium precursors under "minimalist" conditions. General procedure 4 – (GP4): $^{18}F^-$ (0.1–10 GBq) was eluted from a QMA cartridge with precursor (12 µmol) in MeOH (500 µL). Methanol was evaporated under He flow at 70–80 °C. The residue was dissolved in a solution of the corresponding copper salt (12 µmol) in DMF (300 µL) and the resulting solution heated at 85 °C for 20 min. The reaction mixture was cooled to room temperature, diluted with water (2 mL) and shaken vigorously for 30 s. RCCs were determined by radio-HPLC. With $(MeCN)_4CuOTf$ the highest RCCs were obtained using DMF solutions prepared from the copper salt which had been stored in a closed vial under ambient air for 2–5 days. However, if $(MeCN)_4CuOTf$ was stored under ambient air for a longer period of time a significant decline of RCCs was observed. (Method A: [^{18}F]fluorobenzene t_R=5.2 min; [^{18}F]fluoroanisole t_R=6.1 min; 3-[^{18}F]FBA t_R=3.8 min; [^{18}F]**1** t_R=4 min; [^{18}F]**2** t_R=3.3 min; [^{18}F]**3** t_R=3.5 min).).

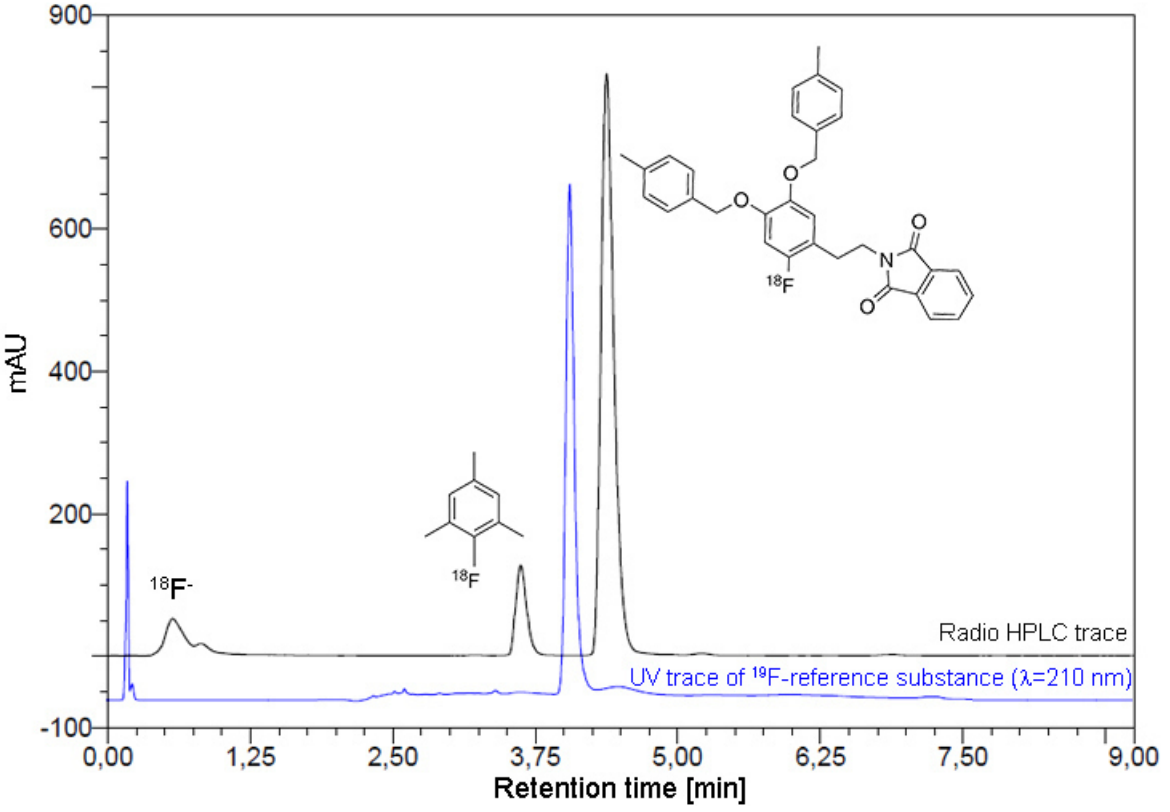
Table 4 Synthesis of ^{18}F-labeled compounds from (mesityl)(aryl)iodonium precursors under "minimalist" conditions.

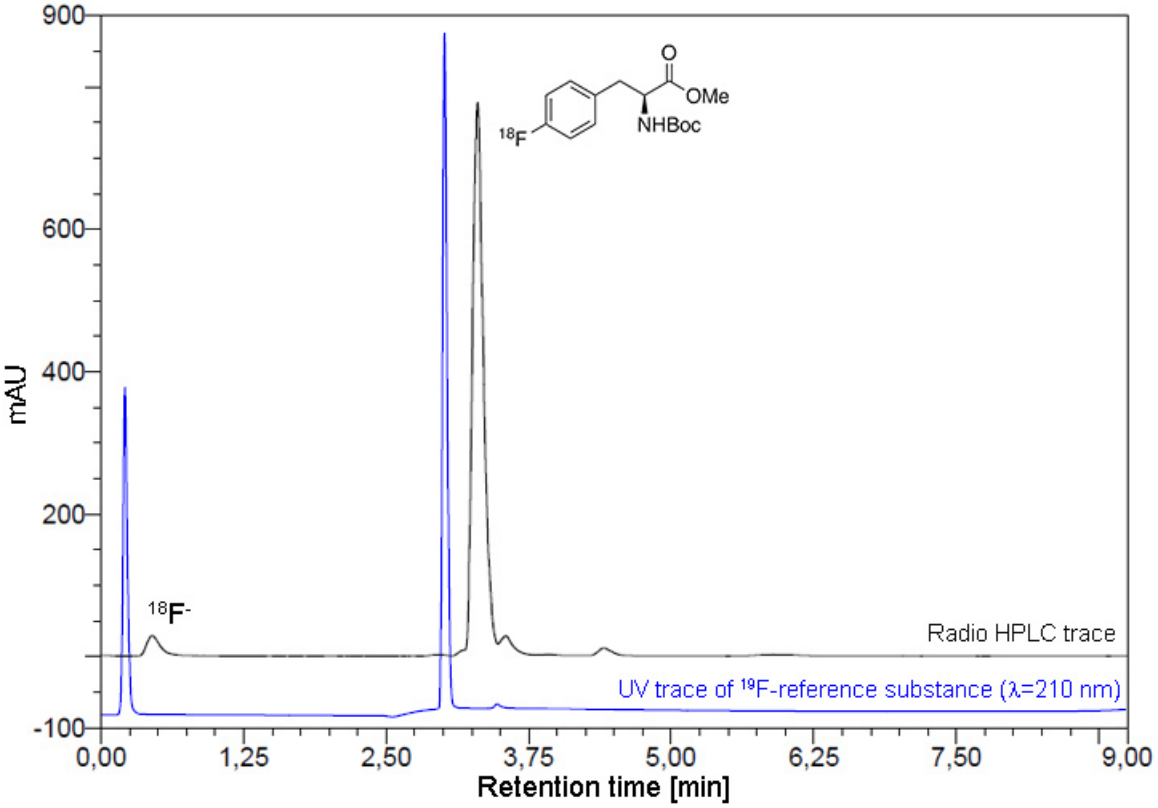
Entry	Product	Precursor	$(MeCN)_4CuOTf$ RCC [%]	$Cu(OTf)_2$ RCC [%]	$(^tBuCN)_2CuOTf$ RCC [%]
1			90	62	90
2			86	72	77
3			90	-	-
4			78	69	85
5	 [^{18}F]**1**	 **6**	71-94	83	-
6	 [^{18}F]**2**	 **9**	81-92	-	-
7	 [^{18}F]DAA1106 ([^{18}F]**3**)	 **12**	93	92	-

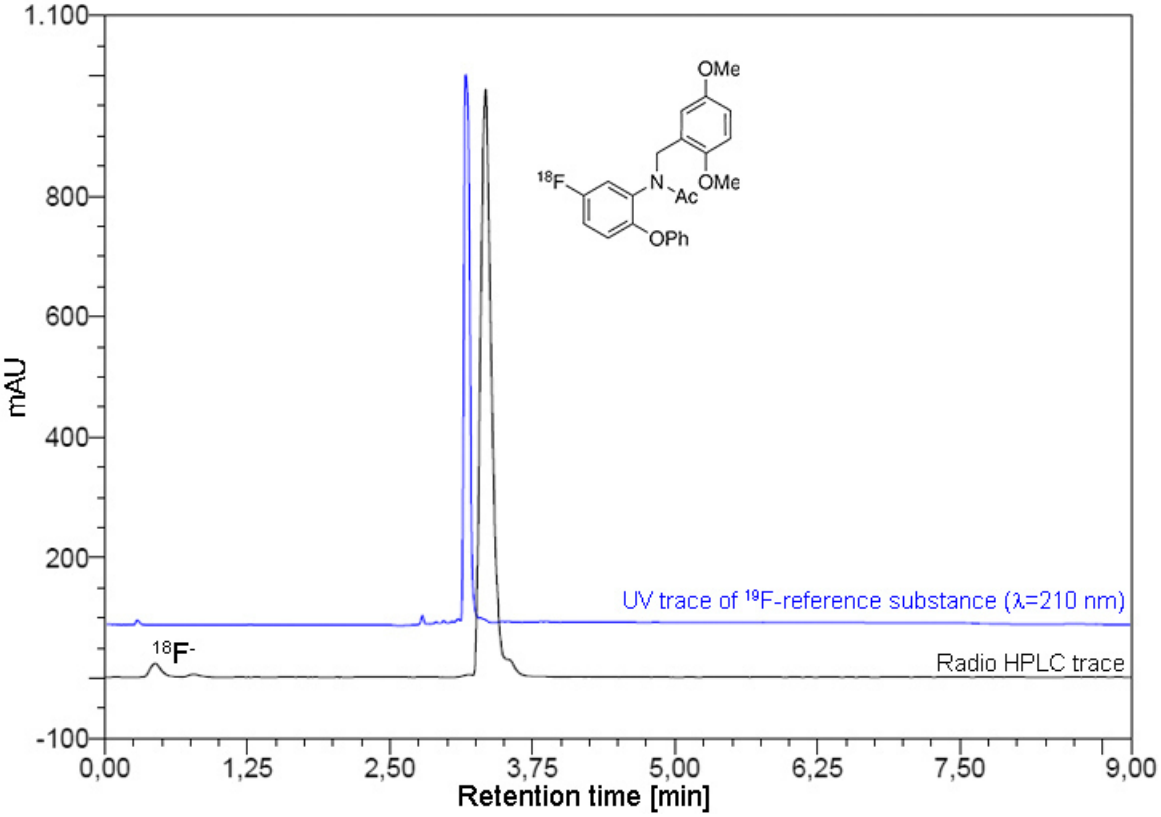

350
300
200
100
0
-50
mAU
^{18}F-
^{18}F
^{18}F
Radio HPLC trace
UV trace of ^{19}F-reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]


1.200
1.000
800
600
400
200
0
-200
mAU
OMe
^{18}F
^{18}F-
^{18}F
Radio HPLC trace
UV trace of ^{19}F-reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]

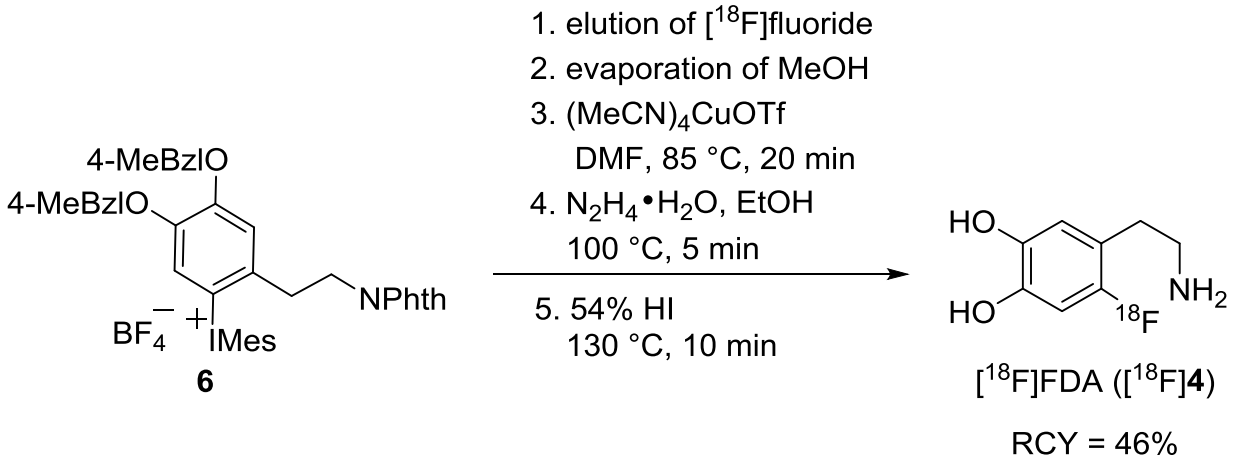

600
500
400
300
200
100
0
-100
mAU
OHC
18F
18F-
18F
Radio HPLC trace
UV trace of 19F-reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]


900
600
400
200
-100
mAU
O
O
N
O
O
18F
18F
18F-
Radio HPLC trace
UV trace of 19F-reference substance (λ=210 nm)
0,00
1,25
2,50
3,75
5,00
6,25
7,50
9,00
Retention time [min]


900
600
400
200
-100
mAU
0,00
1,25
2,50
3,75
5,00
6,25
7,50
9,00
Retention time [min]
O
OMe
NHBoc
18F
18F-
Radio HPLC trace
UV trace of 19F-reference substance (λ=210 nm)

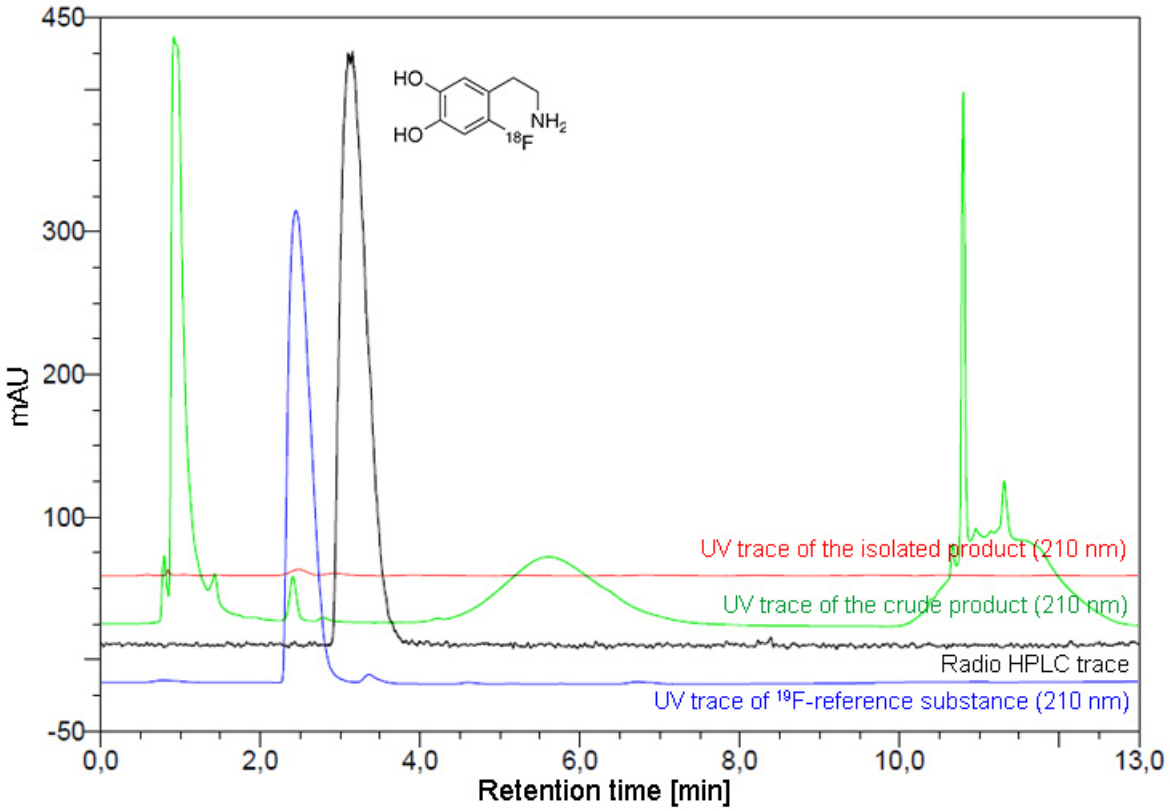

1.100
800
600
400
200
-100
mAU
0,00
1,25
2,50
3,75
5,00
6,25
7,50
9,00
Retention time [min]
OMe
18F
N
Ac
OMe
OPh
UV trace of 19F-reference substance (λ=210 nm)
18F-
Radio HPLC trace

Synthesis of [^{18}F]fluorodopamine ([^{18}F]FDA, [^{18}F]4) on a preparative scale:

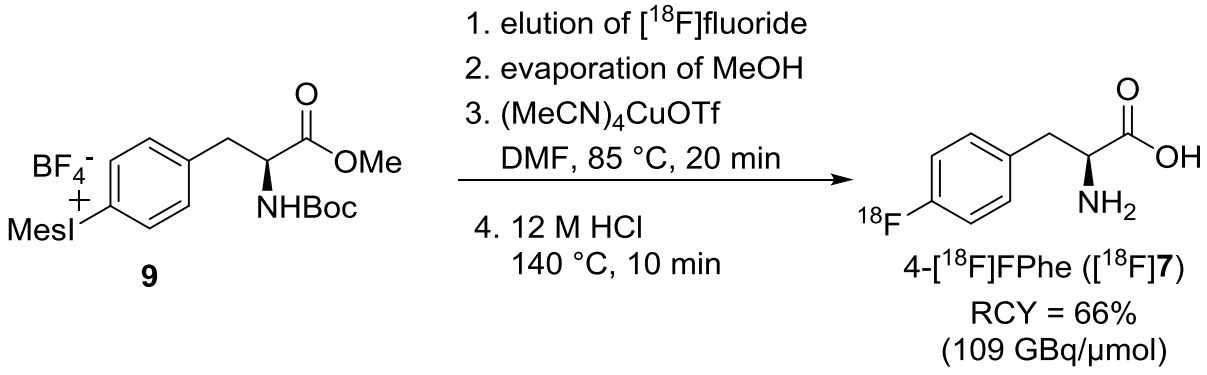


6 (17.6 mg, 12 µmol) was radiolabeled according to GP4 using [^{18}F]fluoride (2.4 GBq) and $(MeCN)_4Cu(OTf)_2$ (4.2 mg, 12 µmol) in DMF (300 µL). Thereafter, EtOH (2 mL) was added and the reaction mixture was concentrated to dryness at 100 °C under Ar. Afterwards, hydrazine monohydrate (10 µL, 0,2 mmol) in EtOH (100 µL) was added to the residue and the resulting solution was heated at 100 °C for 5 min. EtOH (2 mL) was added and the mixture was concentrated to dryness at 100 °C under Ar. The residue was taken up in 57% HI (200 µL) and heated at 140 °C for 10 min under Ar. Acetone (2 mL) was added and the mixture was concentrated to dryness at 80 °C under reduced pressure. The crude tracer was taken in 0.02 M NaH_2PO_4 (200 µL, pH 2.5) and purified by HPLC to give [^{18}F]FDA (550 MBq, 46%).

HPLC conditions for purification and quality control: column: Synergy 4µm Hydro-RP 150×4.6 mm (Phenomenex, Aschaffenburg, Germany); eluent: 4% EtOH in 0.02 M NaH_2PO_4 (pH 2.5); flow rate: 1.5 mL/min; t_R=3.5 min.



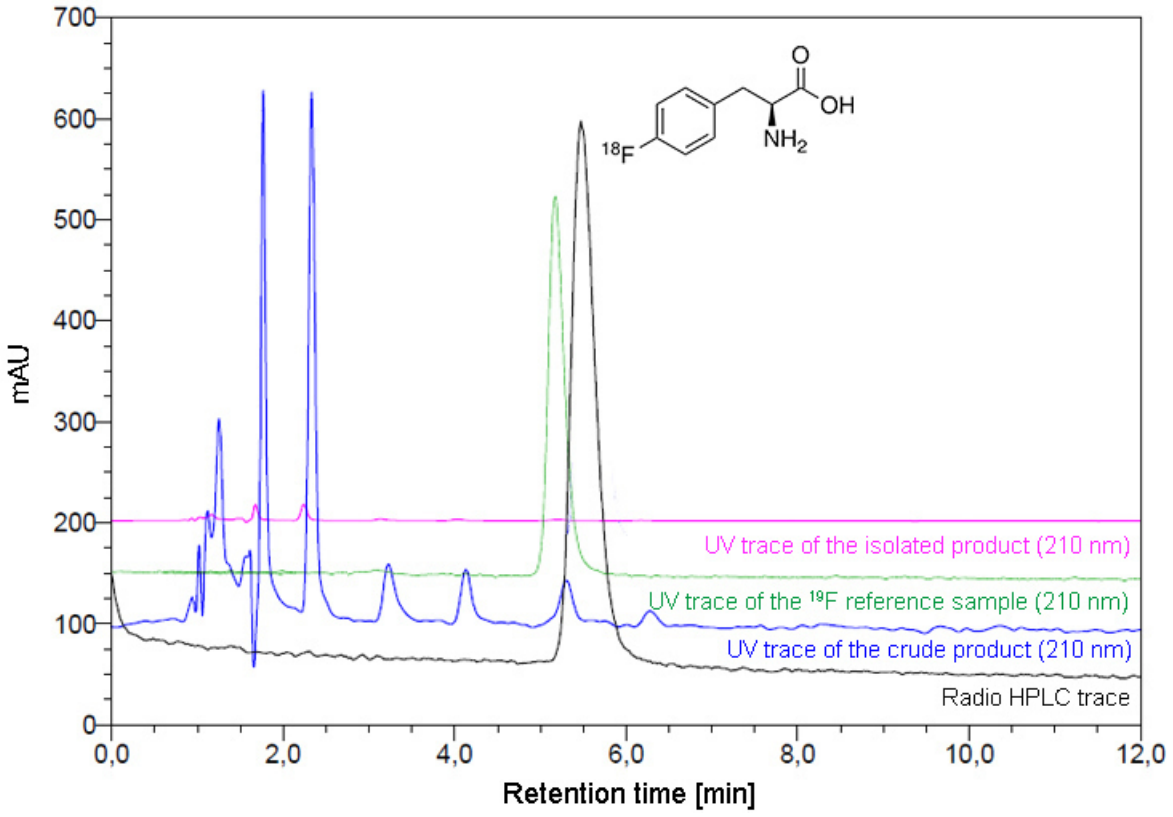
Synthesis of [^{18}F]fluorophenylalanine (4-[^{18}F]FPhe, [^{18}F]7) on a preparative scale:



9 (11.7 mg, 12 µmol) was radiolabeled according to GP4 using [^{18}F]fluoride (10 GBq) and $(MeCN)_4Cu(OTf)_2$ (4.2 mg, 12 µmol) in DMF (300 µL). Thereafter, EtOH (2 mL) was added and the reaction mixture was concentrated to dryness at 100 °C under Ar. The residue was taken up in 12 M HCl (200 µL) and the solution was heated at 140 °C for 10 min. Thereafter, acetone (2 mL) was added and the mixture was concentrated to dryness at 80 °C under reduced pressure. The crude

radiolabeled product was taken up in 4% EtOH in 0.02 M NaH_2PO_4 (500 µL, pH 2.5) and purified by HPLC to give 4-[^{18}F]FPhe (3.3 GBq, 66%).

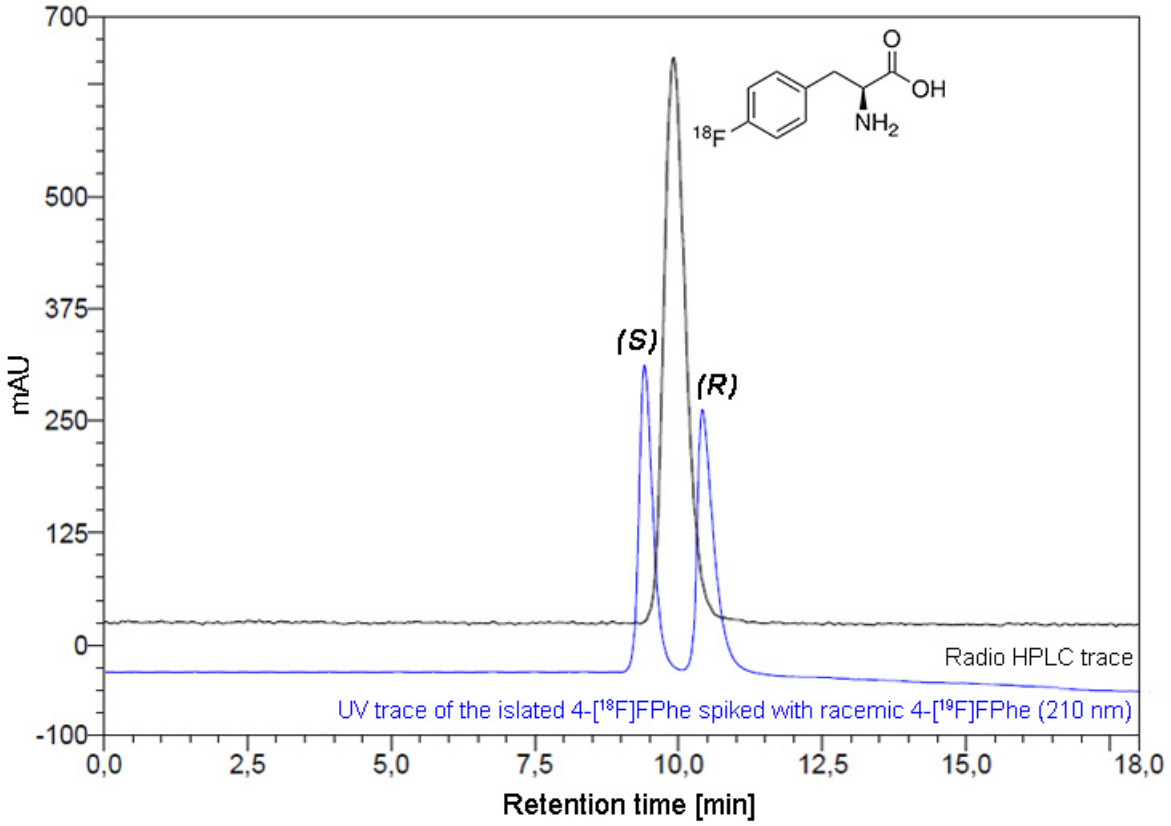
HPLC conditions for purification and quality control: column: Synergy 4µm Hydro-RP 150×4.6 mm; eluent: 4% EtOH in 0.02 M NaH_2PO_4 (pH 2.5); flow rate: 1.5 mL/min; t_R=3.0 min.



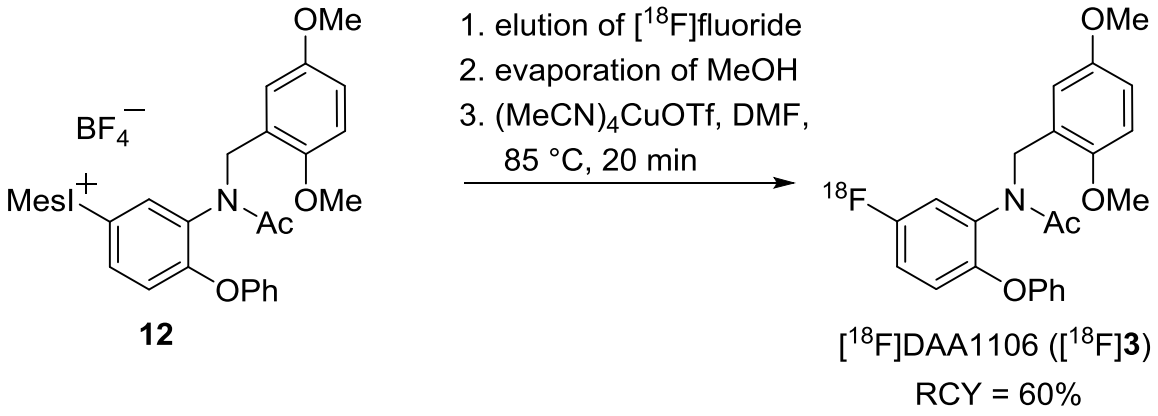
Determination of the enantiomeric purity of 4-[^{18}F]fluorophenylalanine (4-[^{18}F]FPhe)

An aliquot (20 µL) of the isolated 4-[^{18}F]fluorophenylalanine was analysed by HPLC.

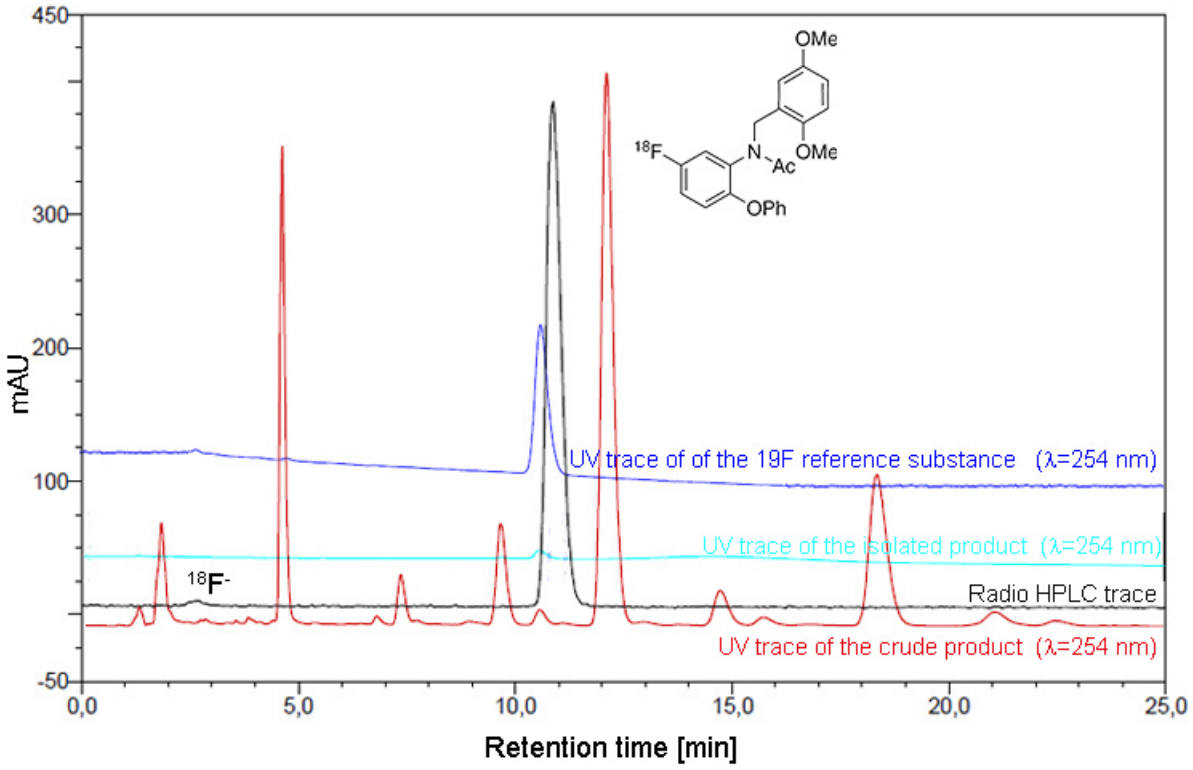
Conditions: column: Astec® CHIROBIOTIC® T 250×4.6 mm 5 µm (Supelco); eluent MeCN:H2O (v/v 80:20); flow rate: 1.5 mL/min; tR: 10 min.



Synthesis of [^{18}F]DAA1106 ([^{18}F]3) on a preparative scale:



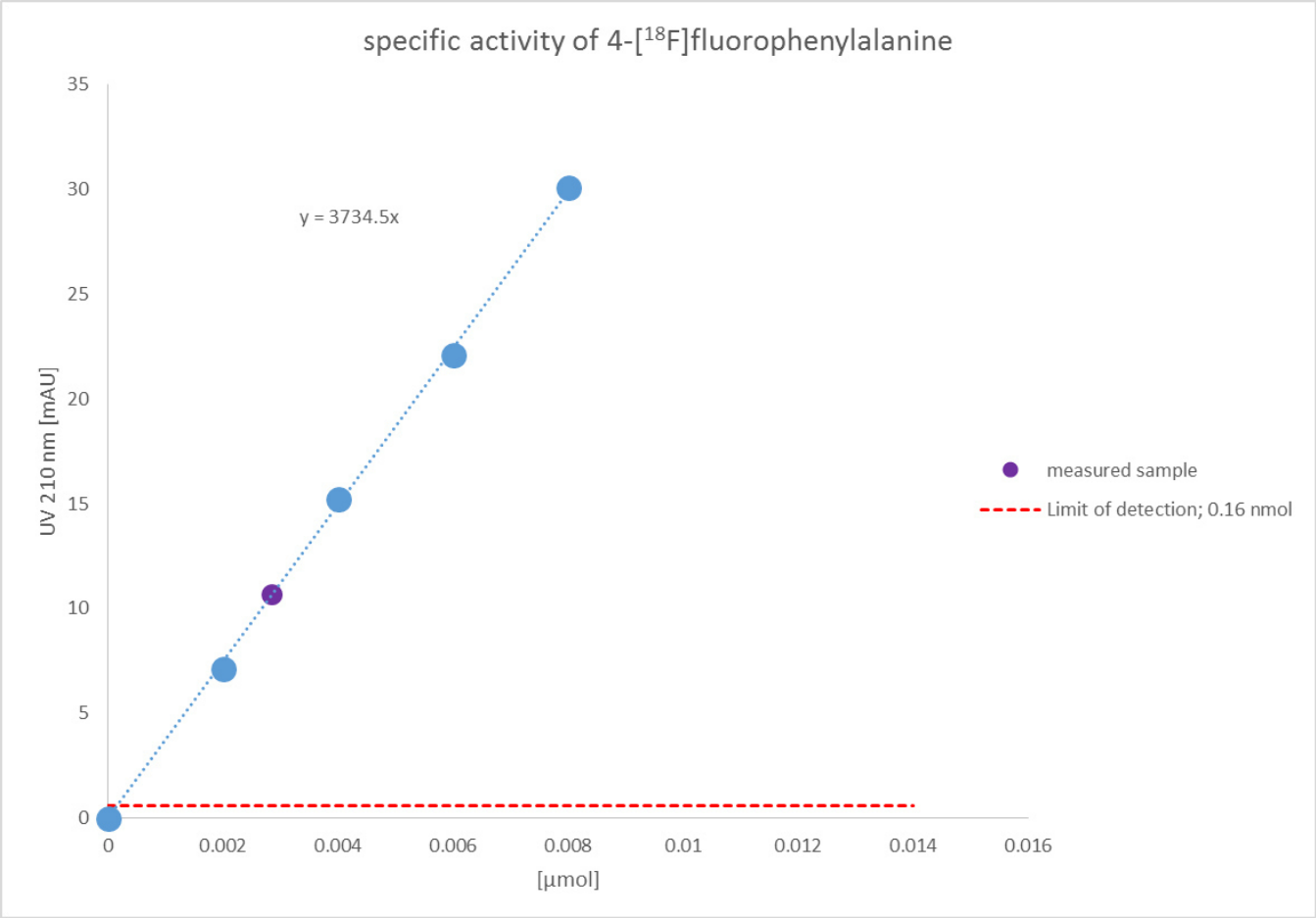
12 (15.1 mg, 12 µmol) was radiolabeled according to GP4 using [^{18}F]fluoride (2.4 GBq) and $(MeCN)_4Cu(OTf)_2$ (4.2 mg, 12 µmol) in DMF (300 µL). Thereafter, EtOH (2 mL) was added and the reaction mixture was concentrated to dryness at 100 °C under Ar. The reaction mixture was cooled to ambient temperature and diluted with H_2O (2 mL). [^{18}F]DAA1106 (60%, 1.2 GBq) was isolated by semi-preparative HPLC. The product containing fraction was diluted with H_2O (20 mL) and loaded onto a StrataX C18 cartridge (preconditioned with 1 mL EtOH followed by 20 mL water). The cartridge was washed with water (10 mL) and the product was eluted with EtOH (1 mL). HPLC conditions for purification and quality control: Method B: t_R=3.6 min.



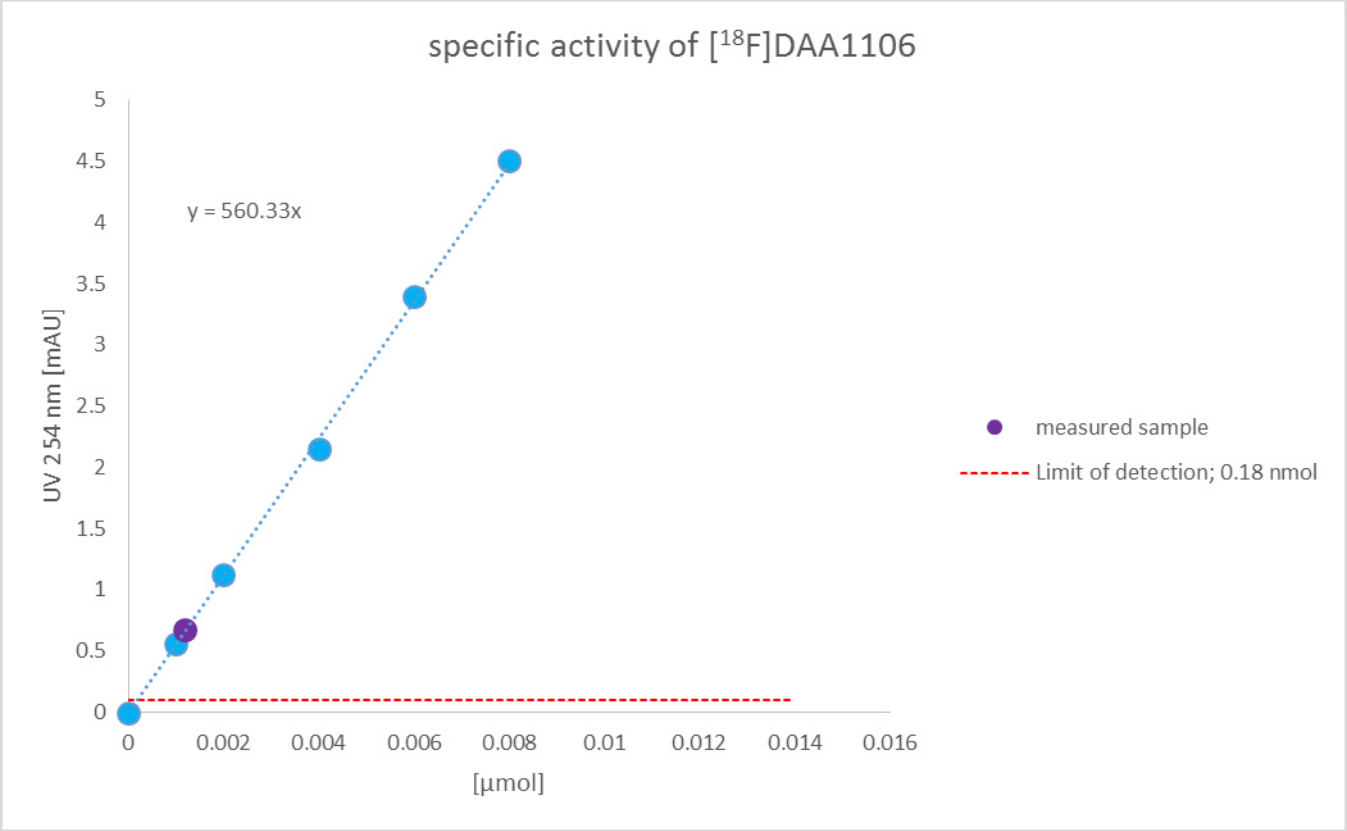
Specific activity calculation

The specific activities (GBq/μmol) were calculated by dividing the radioactivity of the radiolabeled product by the amount of the unlabeled tracer determined from the peak area in the UV-HPLC chromatograms (λ=210 and 254 nm for 4-[18F]FPhe and [^{18}F]DAA1106, respectively). The amounts of unlabeled compounds were determined from the UV-absorbance/concentration calibration curve. The solution of 4-[^{18}F]FPhe obtained after HPLC purification was concentrated under reduced pressure, the residue was redissolved in 4% EtOH in 0.02 m sodium phosphate buffer (300 μL, pH 2.5) and the resulting solution was completely injected. The solution of the HPLC-purified [^{18}F]DAA1106 (3–5 mL) was diluted with water (25 mL) and loaded onto a StrataX cartridge. The cartridge was washed with water (5 mL) and [^{18}F]DAA1106 was eluted with EtOH (200 μL). Water (250 μL) was added and the resulting solution was completely injected. The

specific activity of 4-[^{18}F]fluoro-L-phenylalanine was 109 GBq/µmol. The specific activity of [^{18}F]DAA1106 was 66 GBq/µmol.



Concentration/UV-absorbance calibration curve for 4-[^{18}F]fluoro-L-phenylalanine.



Concentration/UV-absorbance calibration curve for [^{18}F]DAA1106.

Rat stroke model: An adult male Long Evans rat was used for occlusion of the anterior cerebral artery (ACA).[22] The rat was anesthetized (initial dosage: 5% isoflurane in O_2/N_2O (3:7), then reduction to 2.5%) and fixed in a stereotaxic frame. Prior to surgery, 3 mg carprofen (Rimadyl®) was injected subcutaneously. After removing skin and periosteum, a small hole (approx. 1 mm in diameter) was drilled in the skull 1.5 mm anterior to bregma at midline, and the cannula of a Hamilton syringe was inserted 3.3 mm deep, measured from the level of the dura mater. 300 pmol Endothelin-1 (Sigma Aldrich®, St. Louis, USA) in 0.6 µl of 0.9% NaCl was slowly injected, and the cannula was left in place for 10 min. After retraction of the cannula, the burr hole was closed with bone wax and the skin wound was sutured.

µPET-Imaging: PET measurements were performed six days after ACA occlusion. Prior to the PET measurement, the animal was anesthetized (initial dosage: 5 % isoflurane in O_2/N_2O (3:7), then

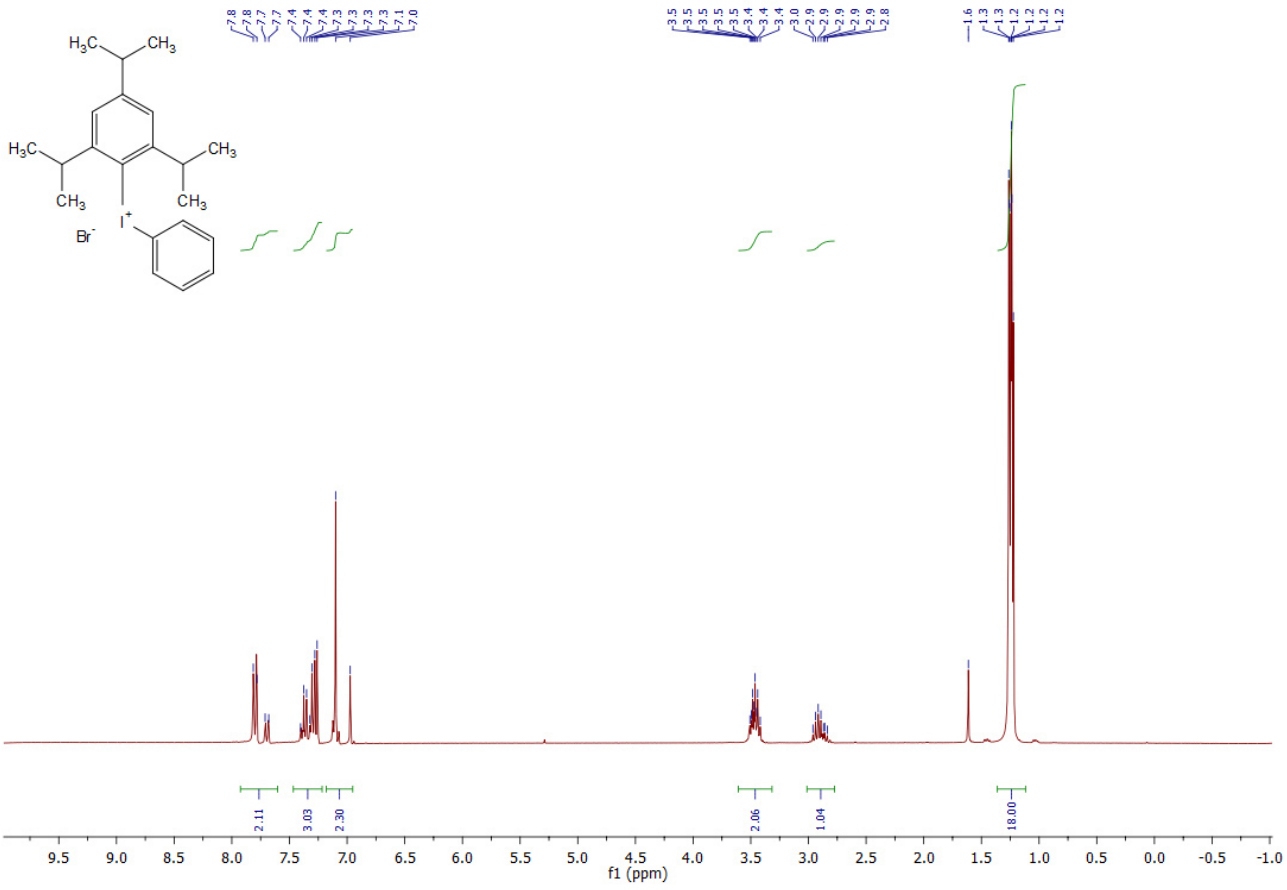
reduction to 2 %), and a catheter for tracer-injection was inserted into the lateral tail vein. The rat was placed on an animal holder (medres GmbH, Cologne, Germany), and fixed with a tooth bar in a respiratory mask. A dynamic PET scan in list mode was performed using a Focus 220 micro PET scanner (CTI-Siemens, Erlangen, Germany) with a resolution at the center of field of view of 1.4 mm. Data acquisition started immediately after intravenous injection of [^{18}F]DAA1106 (74 MBq in 0.5 mL EtOH:PEG-400:0.9% NaCl = 1:1:3) and was carried for 70 min. Thereafter a 10 min transmission scan was acquired using a ^{57}Co point source. Breathing rate was monitored with a Dasy Lab system 9.0 (DasyLab, Mönchengladbach, Germany) and kept around 60/min by adjusting the isoflurane concentration (1.5–2.5 %). Body temperature was maintained at 37°C by a feedback-controlled system (medres GmbH, Cologne, Germany). Following Fourier rebinning, data were reconstructed within one frame from 10 min to 70 min post injection, using an iterative OSEM3D/MAP procedure including attenuation correction. Resulting voxel sizes were 0.38×0.38×0.82 mm.

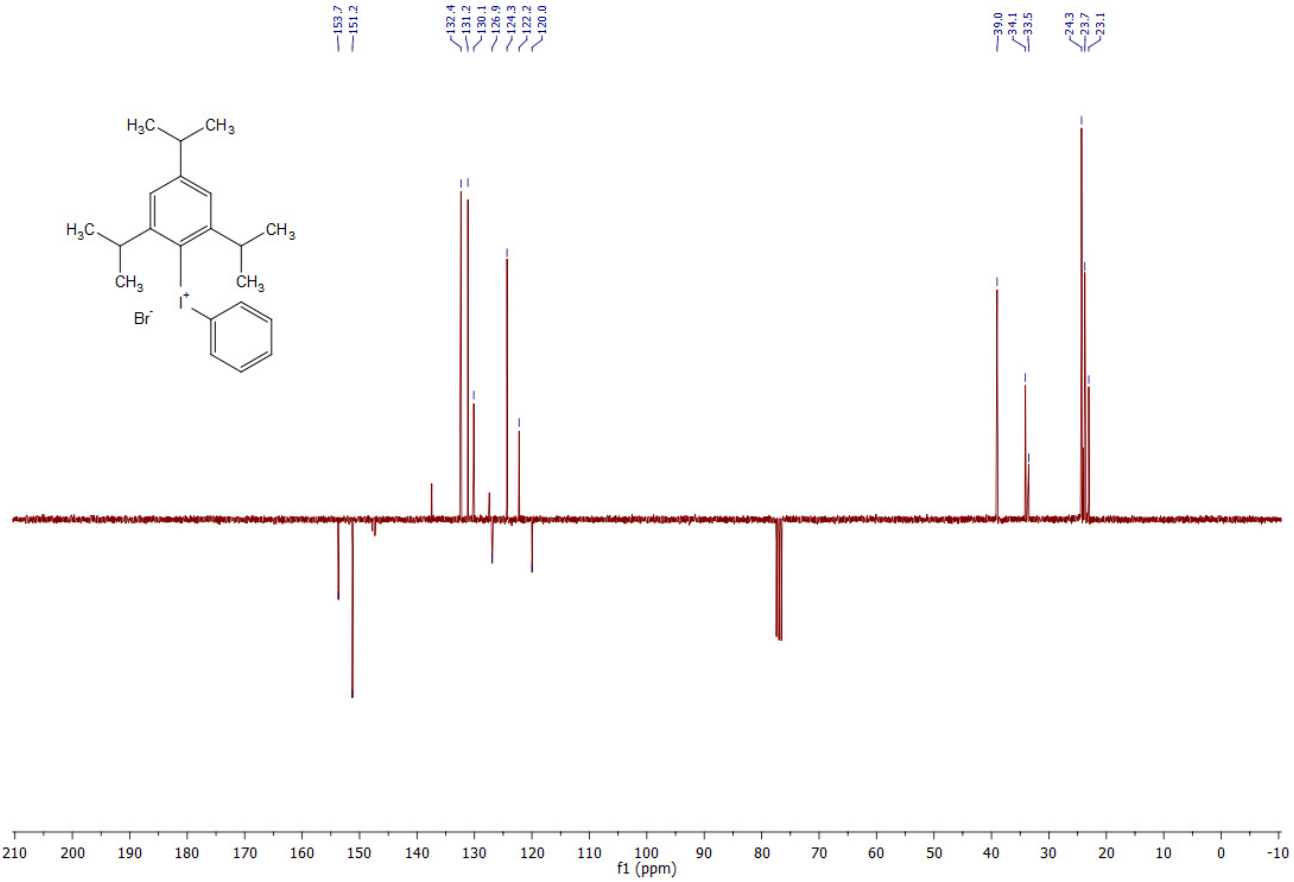
MR-Imaging: MRI measurements were performed seven days after ACA occlusion. To rule out gross structural brain anomalies and to provide individual templates for co-registration of the PET images, T2-weighted structural MR images were acquired. MRI scans were performed in an 11.7-T BioSpec animal scanner (Bruker BioSpin®) using a quadrature receive-only rat brain surface coil (Bruker BioSpin®) in combination with an actively decoupled, transmit-only quadrature resonator with 72 mm inner diameter (Bruker Biospin), fitting into the BFG-150/90-S14 combined gradient and shim set of 90 mm inner diameter (Resonance Research Inc., Billerica, MA, USA) with a maximum gradient strength of 745 mT/m. A T2-weighted sequence, rapid acquisition with relaxation enhancement (RARE) was used: RARE factor = 8, repetition time/effective echo time = 6500/32.5 ms, averages = 2, matrix size = 256 × 256, FOV = 3.2 × 3.2 cm^2, 58 slices, slice thickness = 0.5 mm, interslice spacing = 0.5 mm. Inhalation anesthesia procedure was the same as that used

for the μPET scan. During scanning, body temperature was recorded and maintained at 37° using feedback water control (medres GmbH, Cologne, Germany); physiological parameters, in particular respiration rate, were monitored using DASYLab 9.0 (DasyLab, Moenchengladbach, Germany).

Imaging Data Analysis: MR- and μPET-Images were manually coregistered with the help of VINCI 4.04.[23]

^{1}H-, ^{19}F- and APT-NMR Sprectra

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122.2
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39.0
34.1
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24.3
23.7
23.1
H3C
CH3
H3C
CH3
CH3
CH3
I+
Br-
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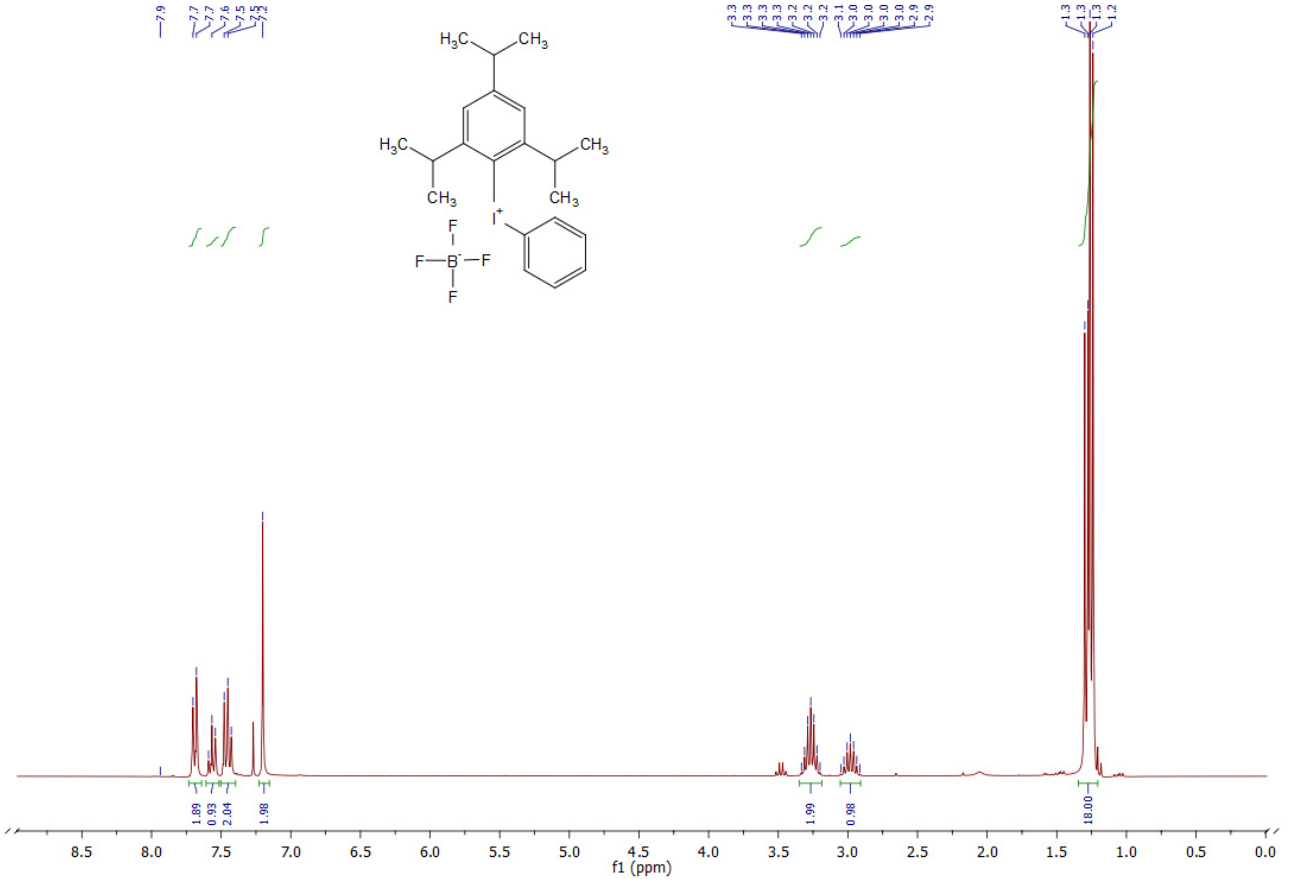
f1 (ppm)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1.2
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CH3
CH3
I+
F
F
B-
F
F
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0.93
2.04
1.98
1.99
0.98
18.00
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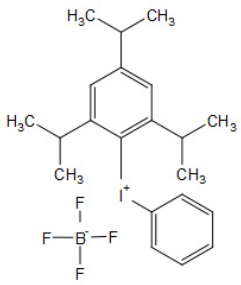
f1 (ppm)



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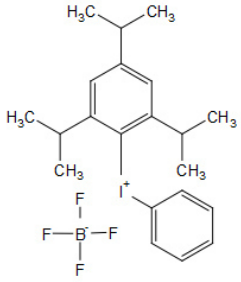
f1 (ppm)



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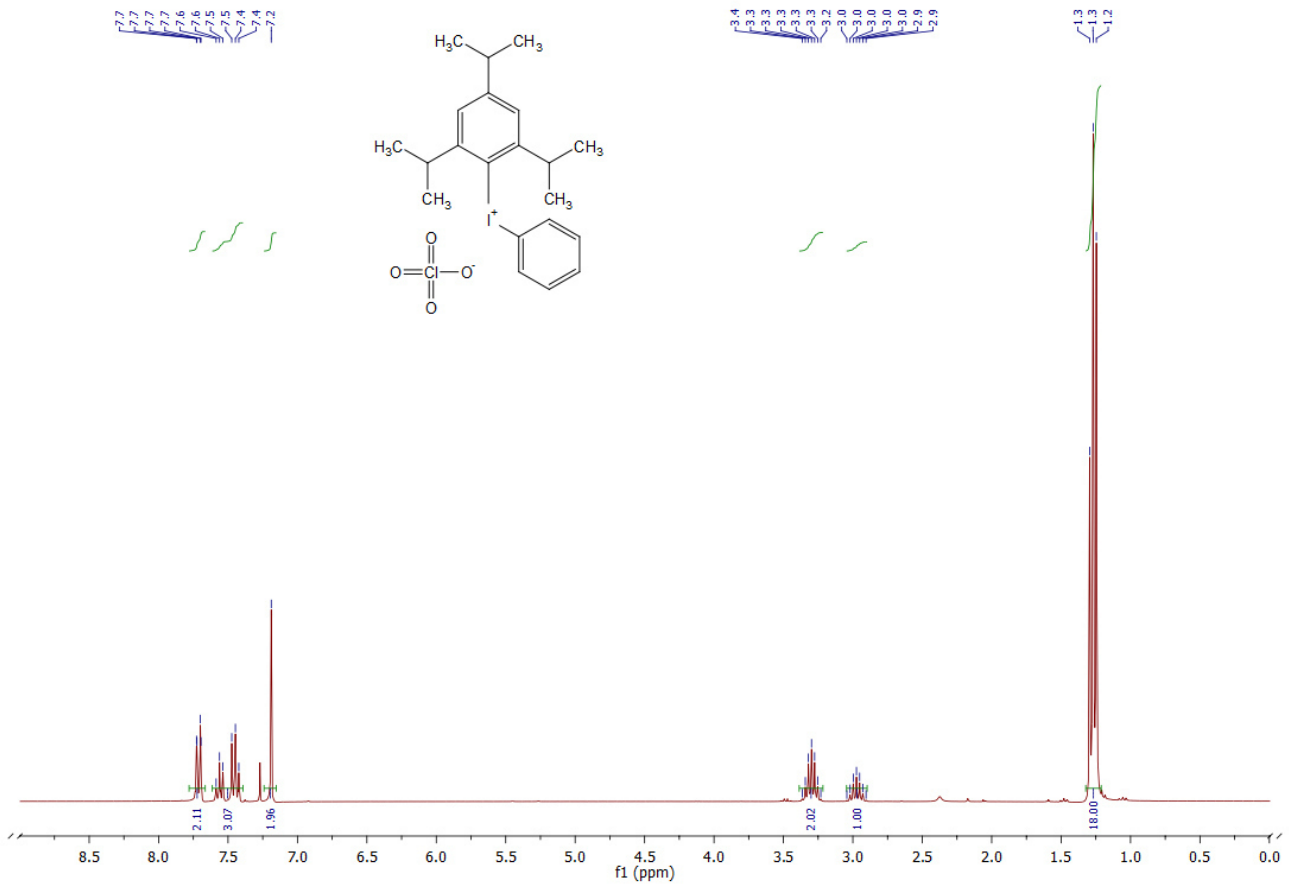
152.6
132.6
132.1
125.5
119.4
111.9
39.7
34.2
24.2
23.6
210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10
f1 (ppm)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CH3
H3C
CH3
CH3
CH3
I+
O
O=Cl—O-
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1.96
2.02
1.00
18.00
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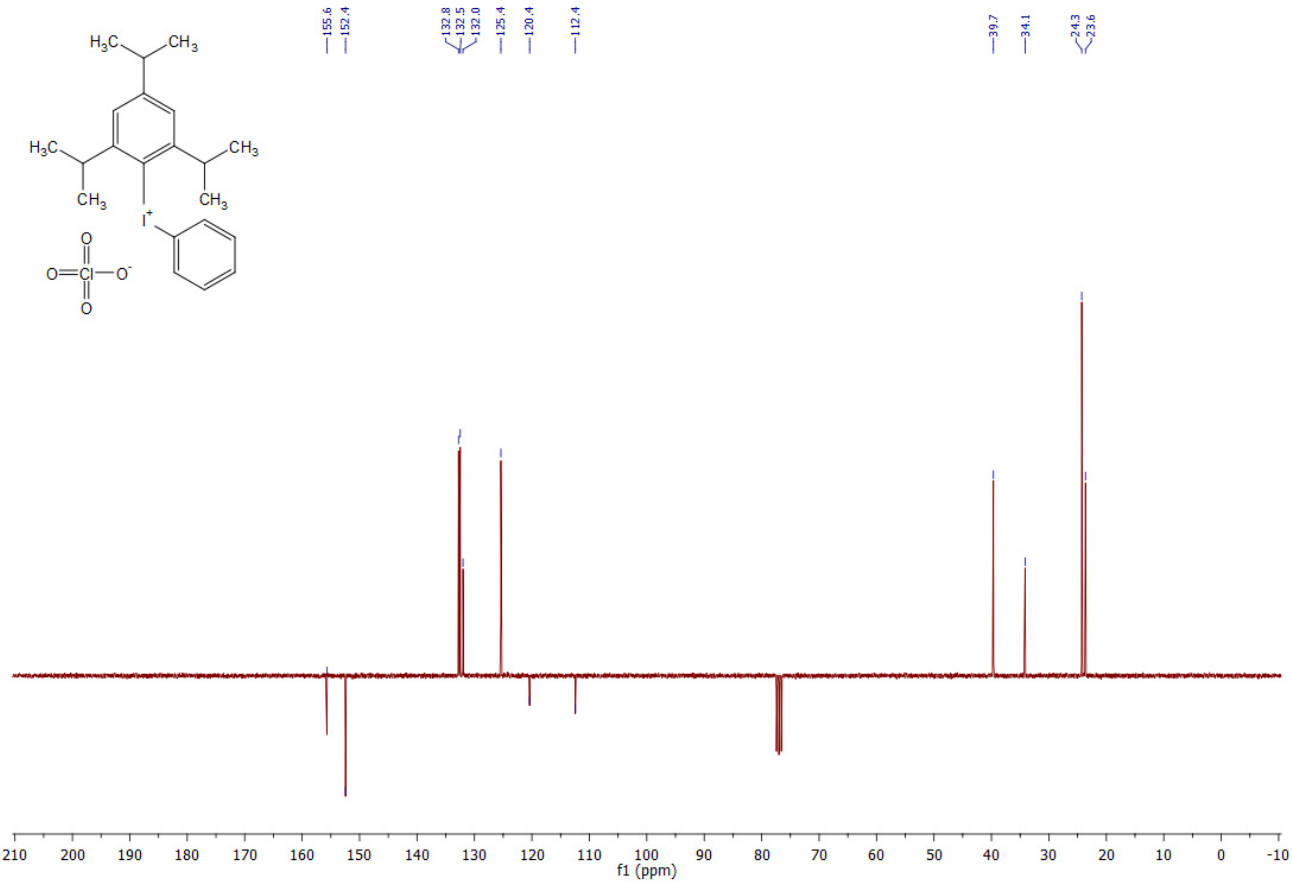
f1 (ppm)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34.1
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CH3
H3C
CH3
CH3
CH3
I+
O
O=Cl—O-
O
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200
190
180
170
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150
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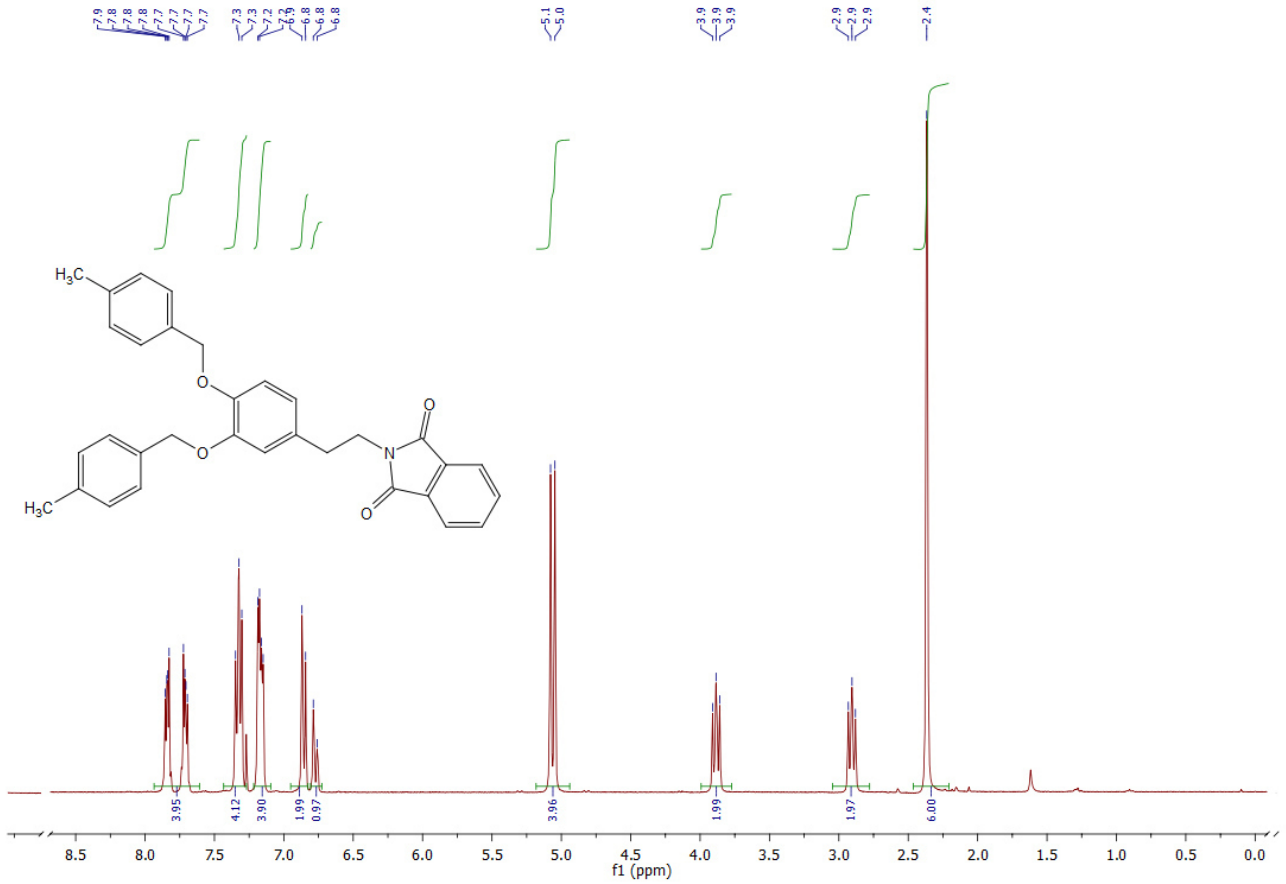
f1 (ppm)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2.9
2.4
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O
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N
O
O
H3C
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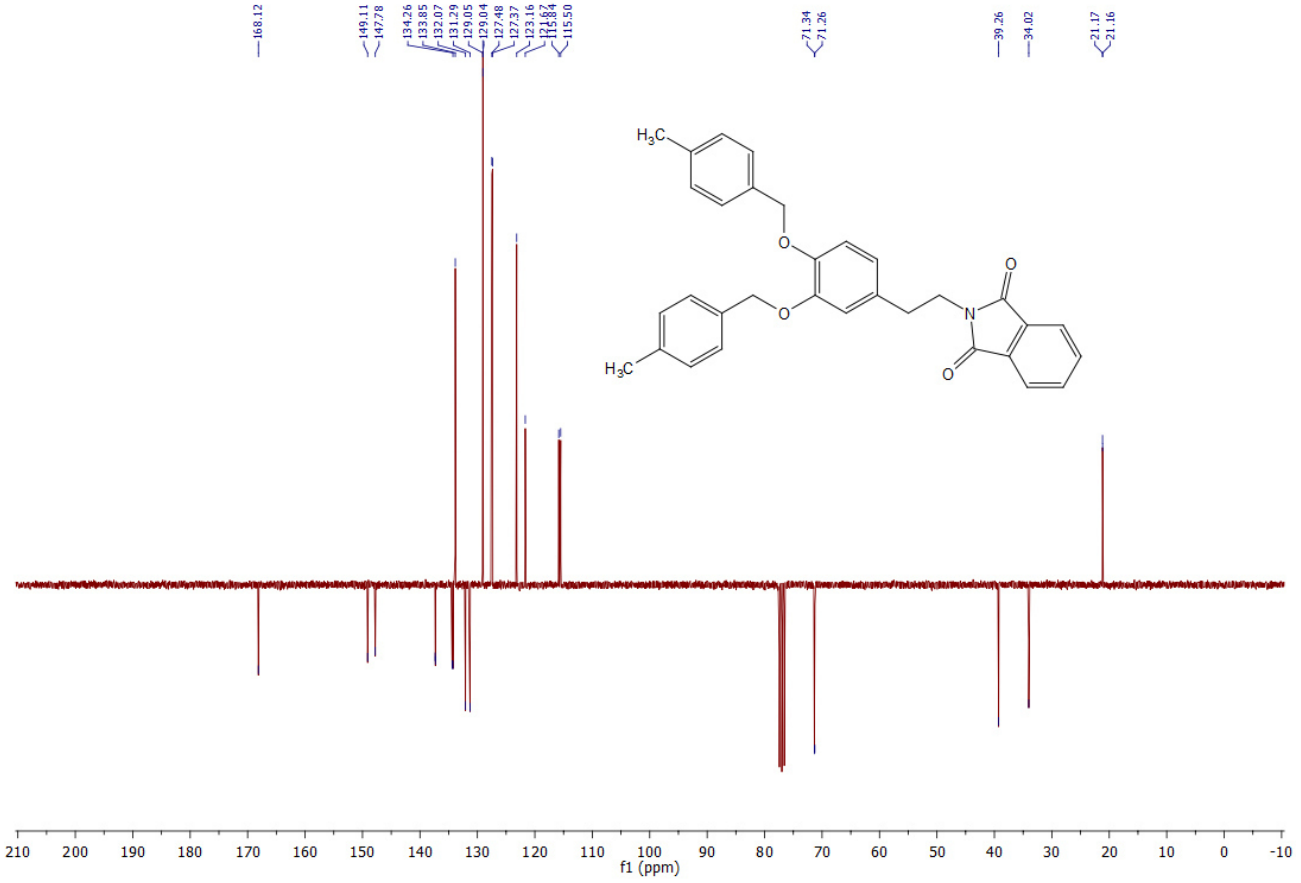
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115.50
71.34
71.26
39.26
34.02
21.17
21.16
H3C
O
O
N
O
O
H3C
210
200
190
180
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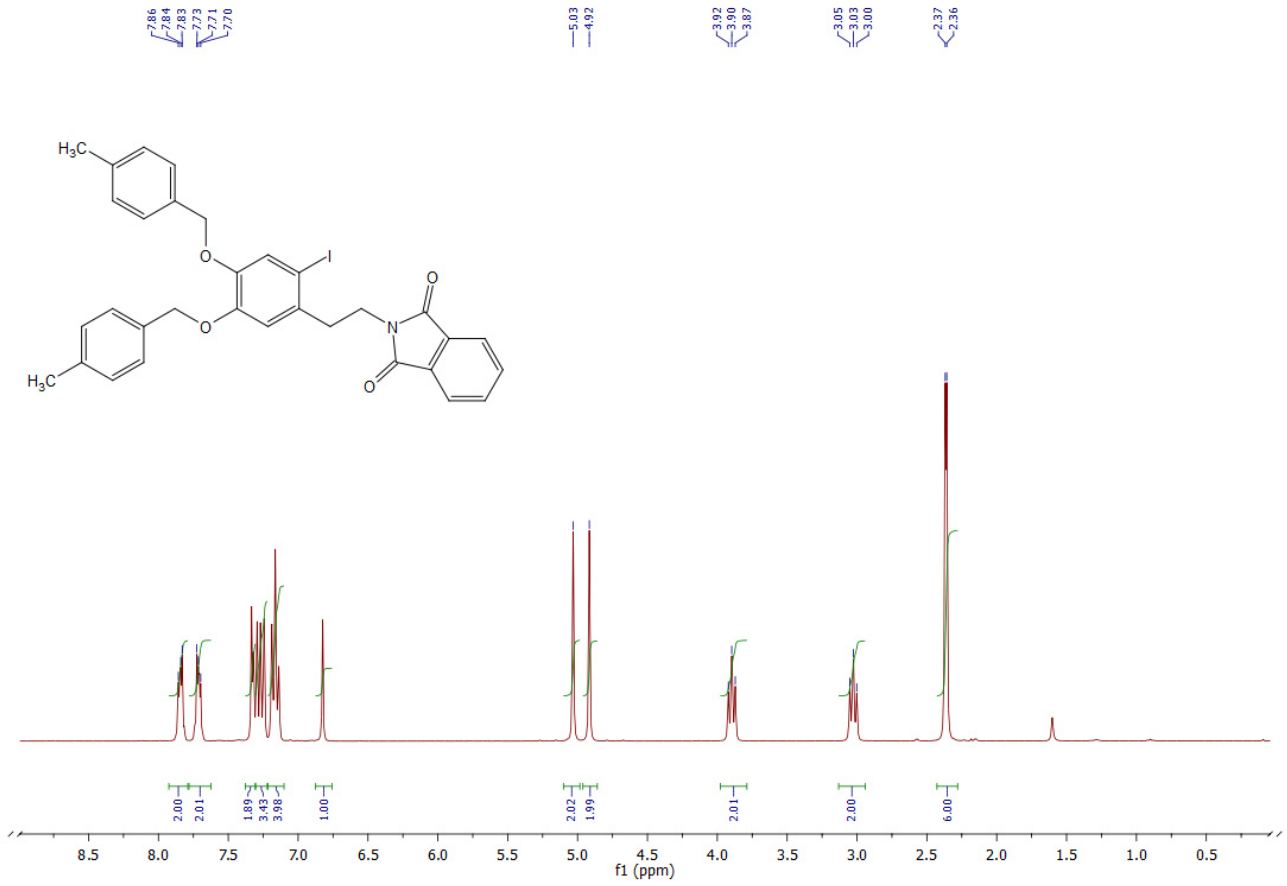
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3.00
2.37
2.36
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O
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3.98
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2.00
6.00
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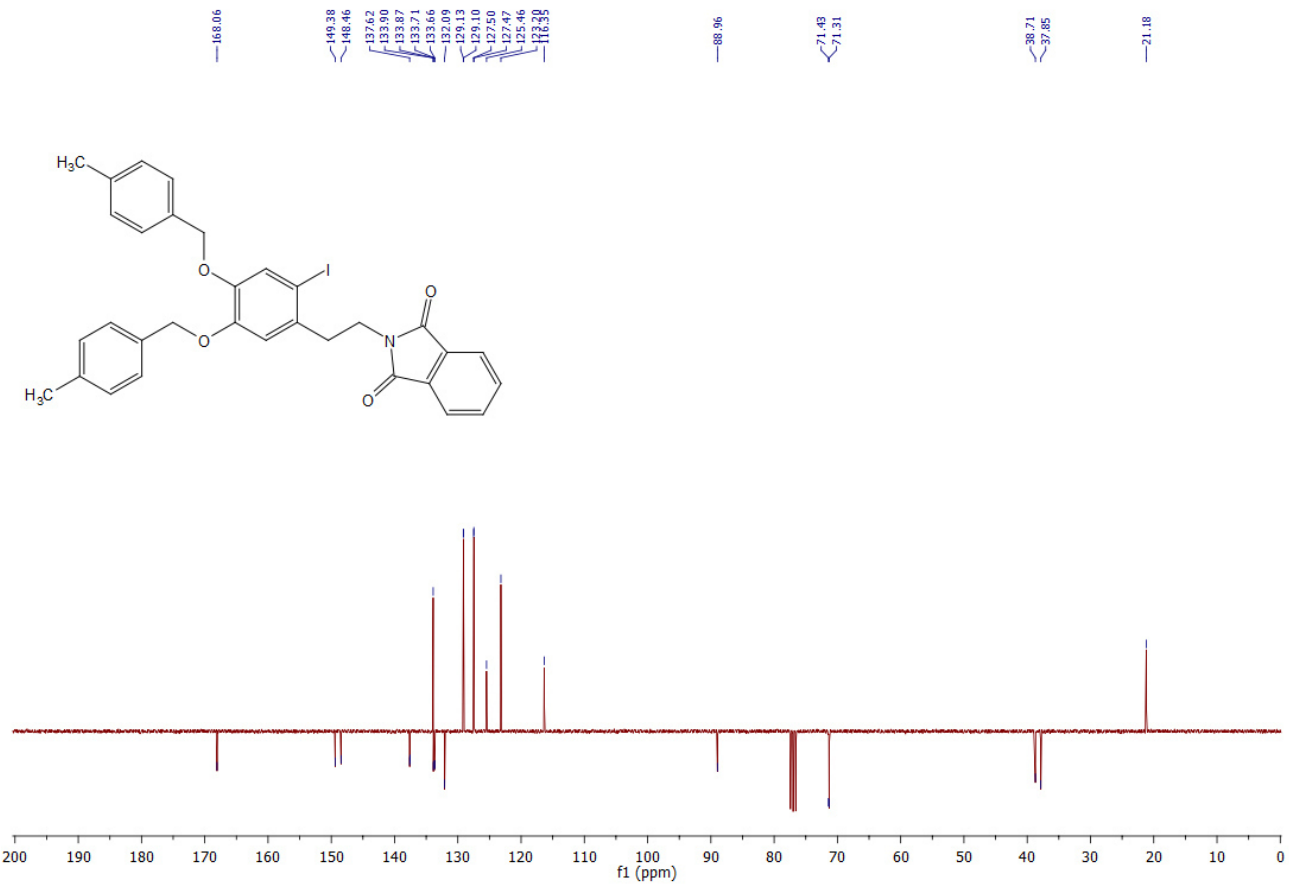
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123.20
116.35
88.96
71.43
71.31
38.71
37.85
21.18
H3C
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I
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O
N
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H3C
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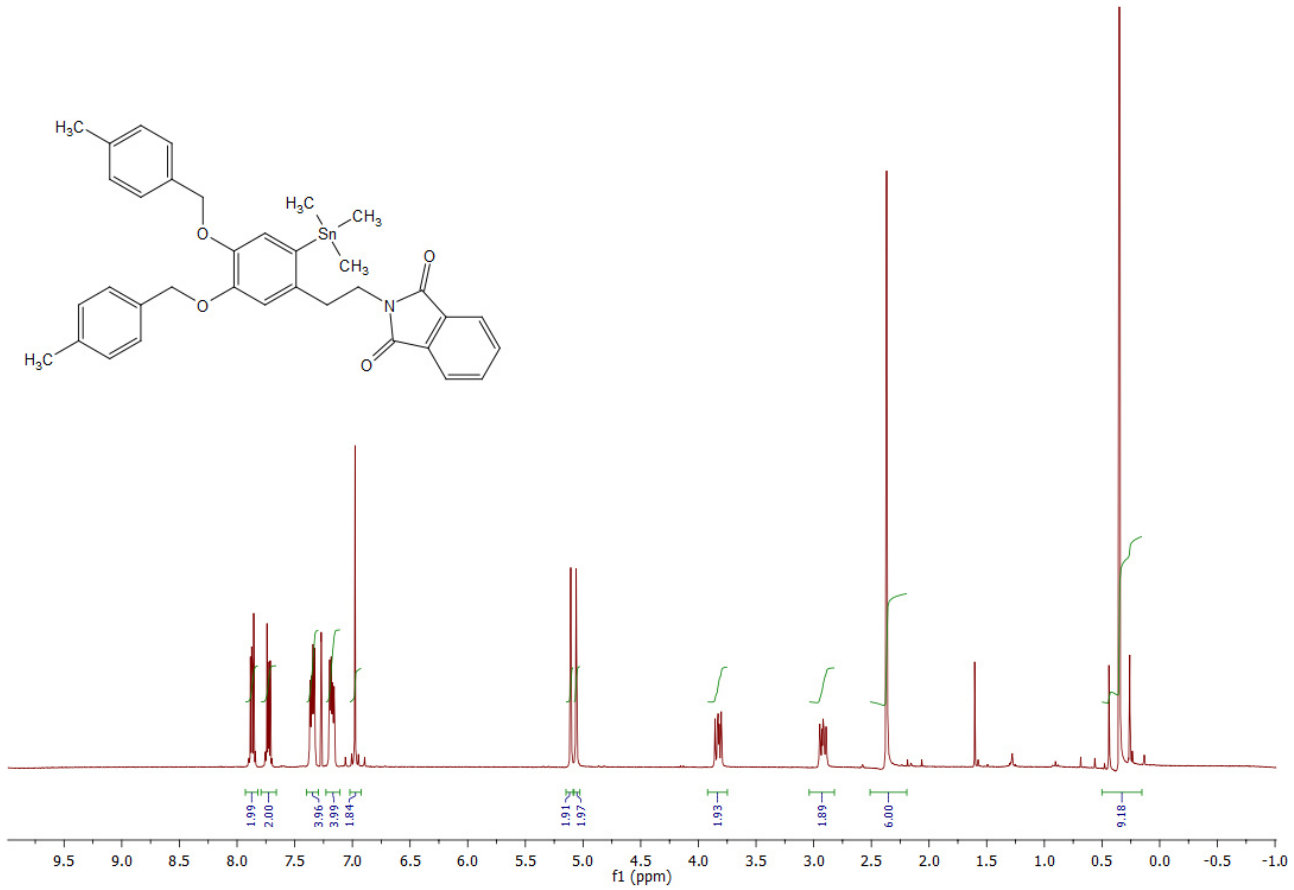

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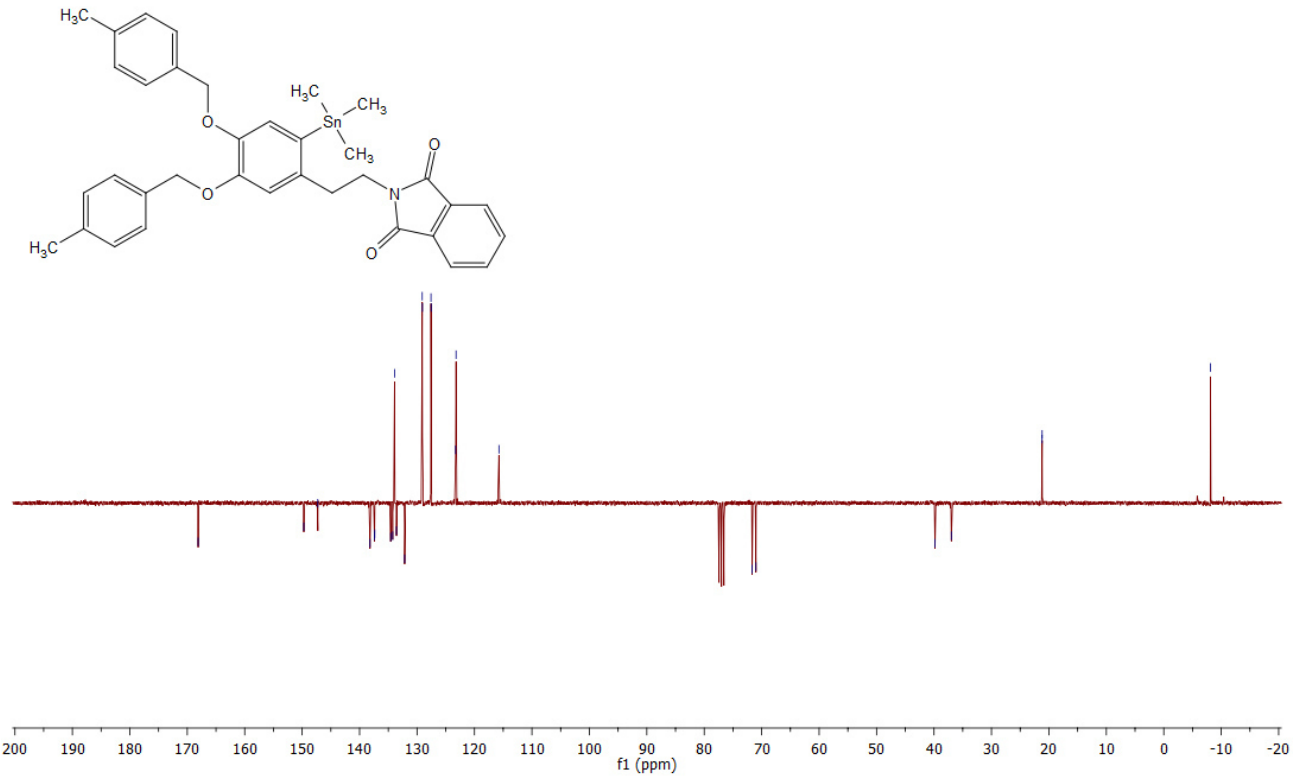

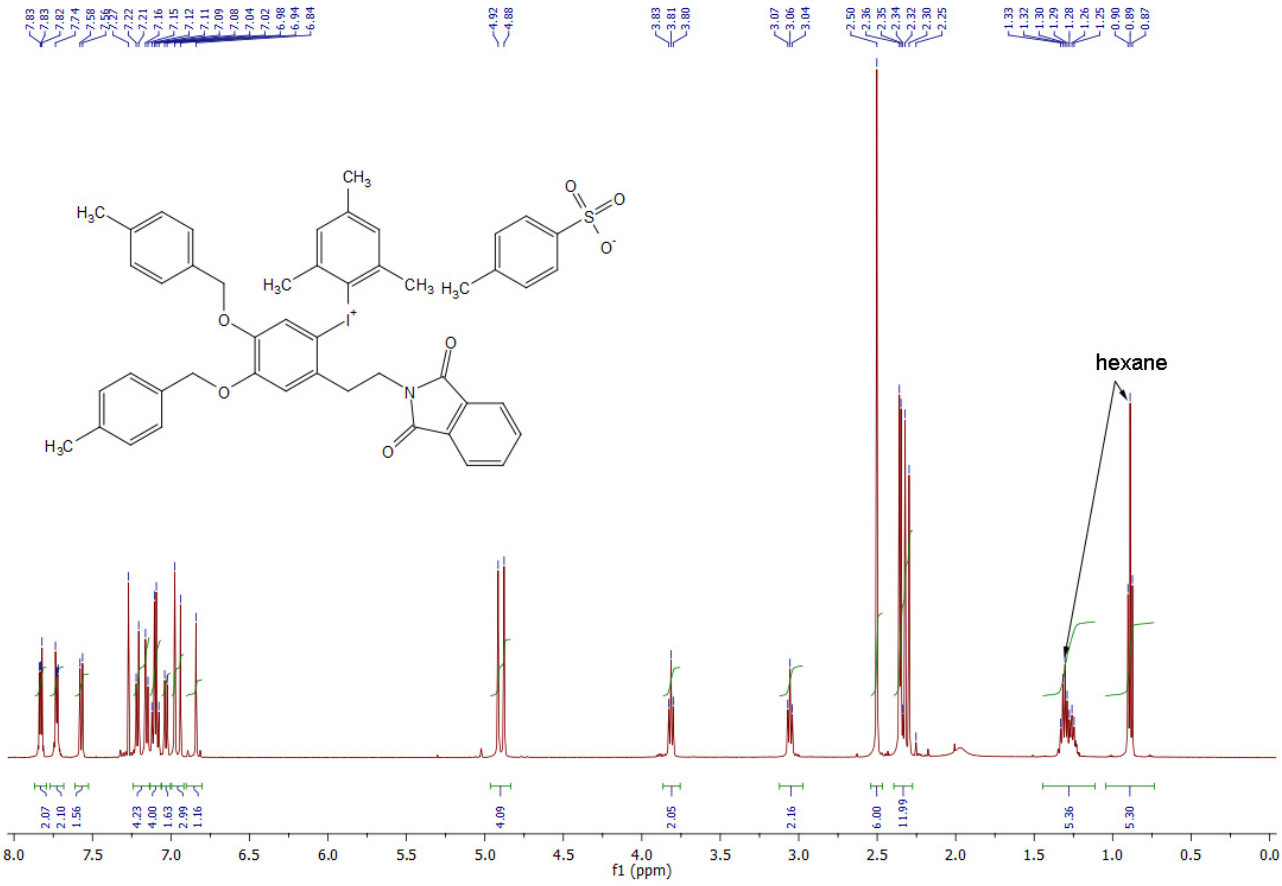
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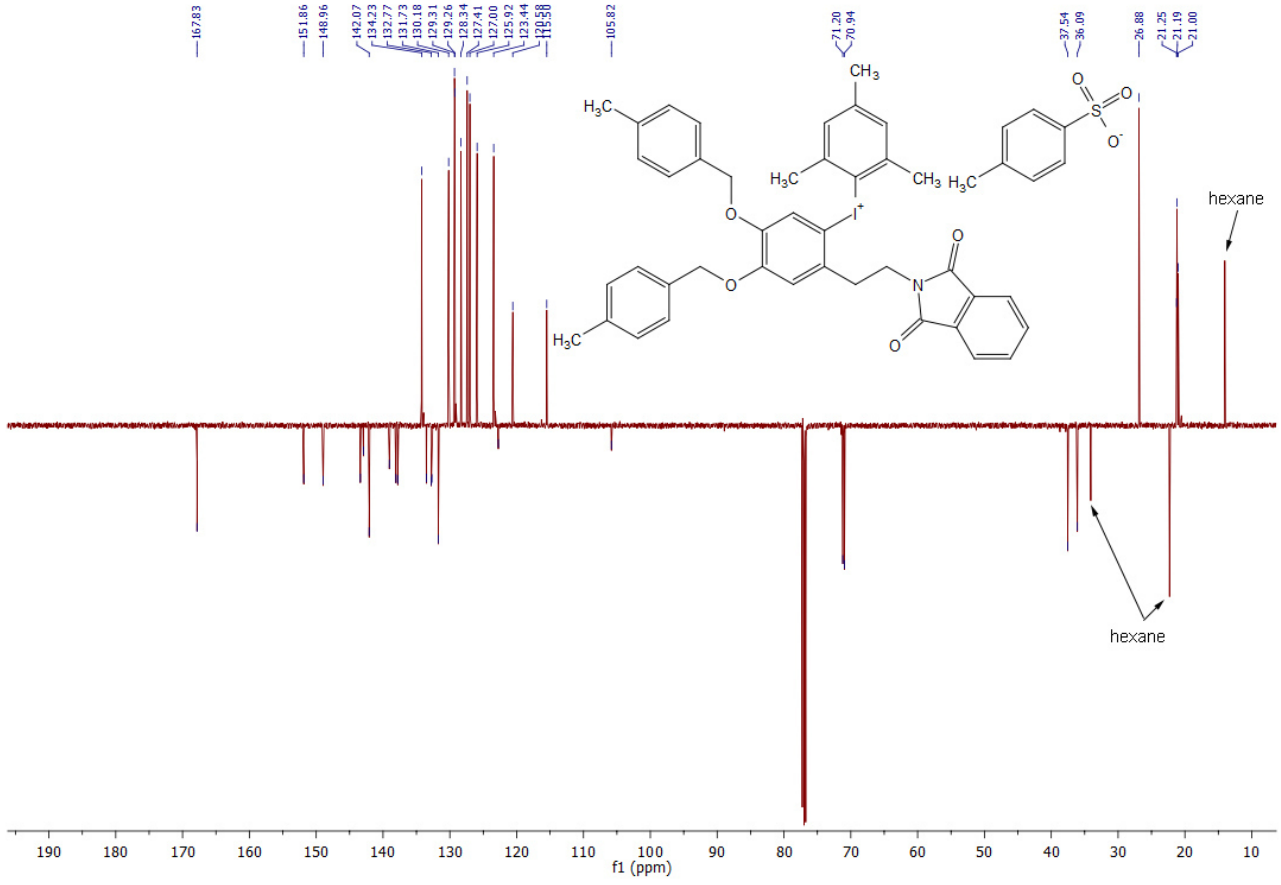
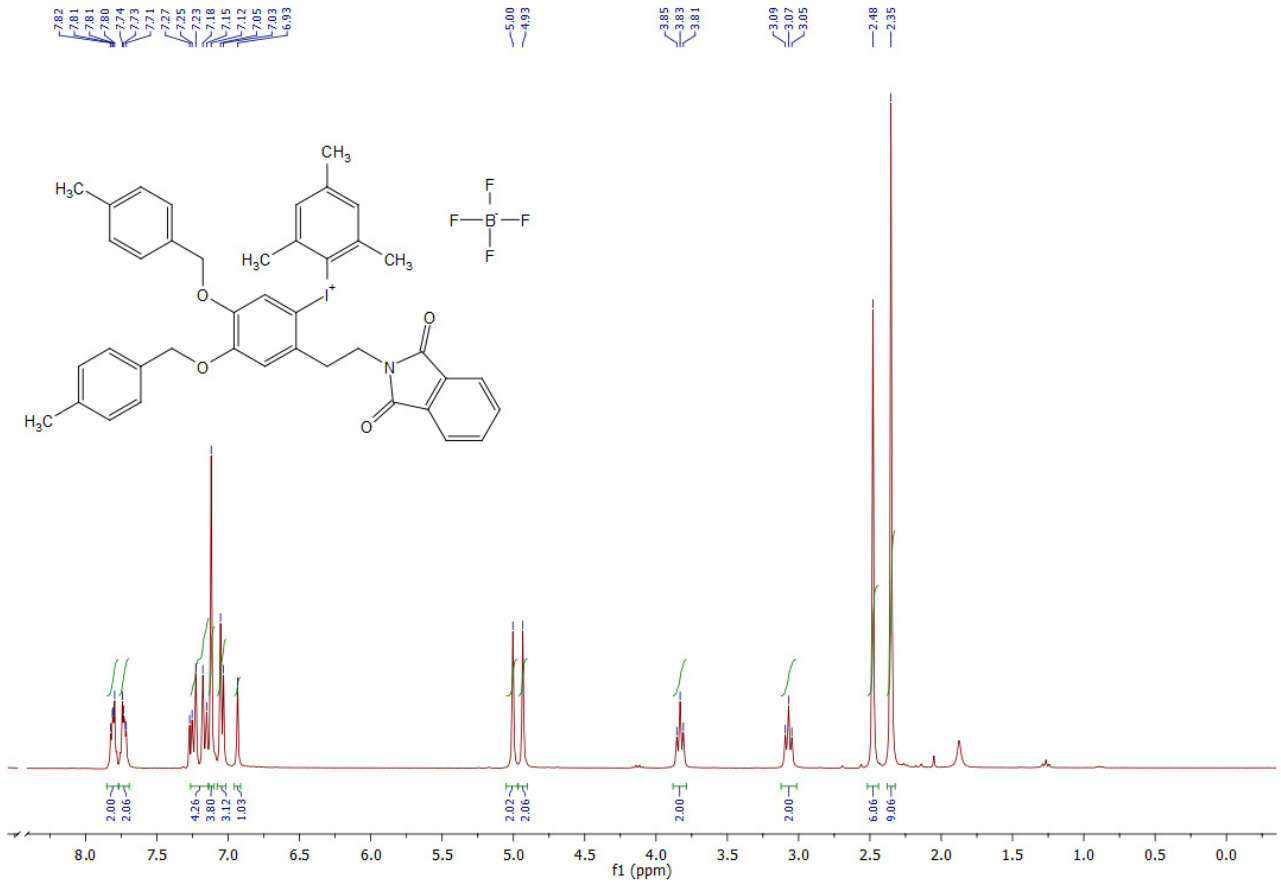

hexane
hexane
f1 (ppm)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7.05
7.03
6.93
5.00
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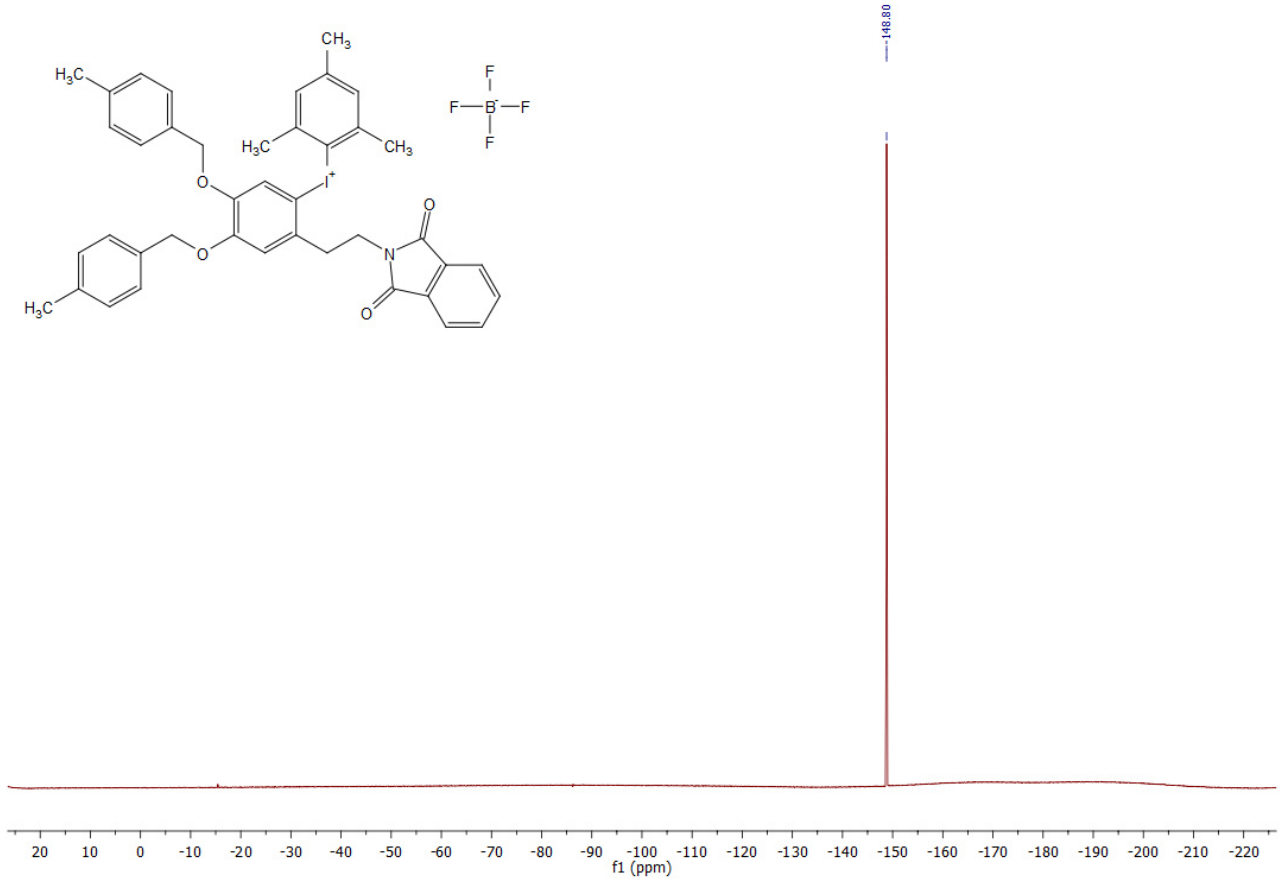
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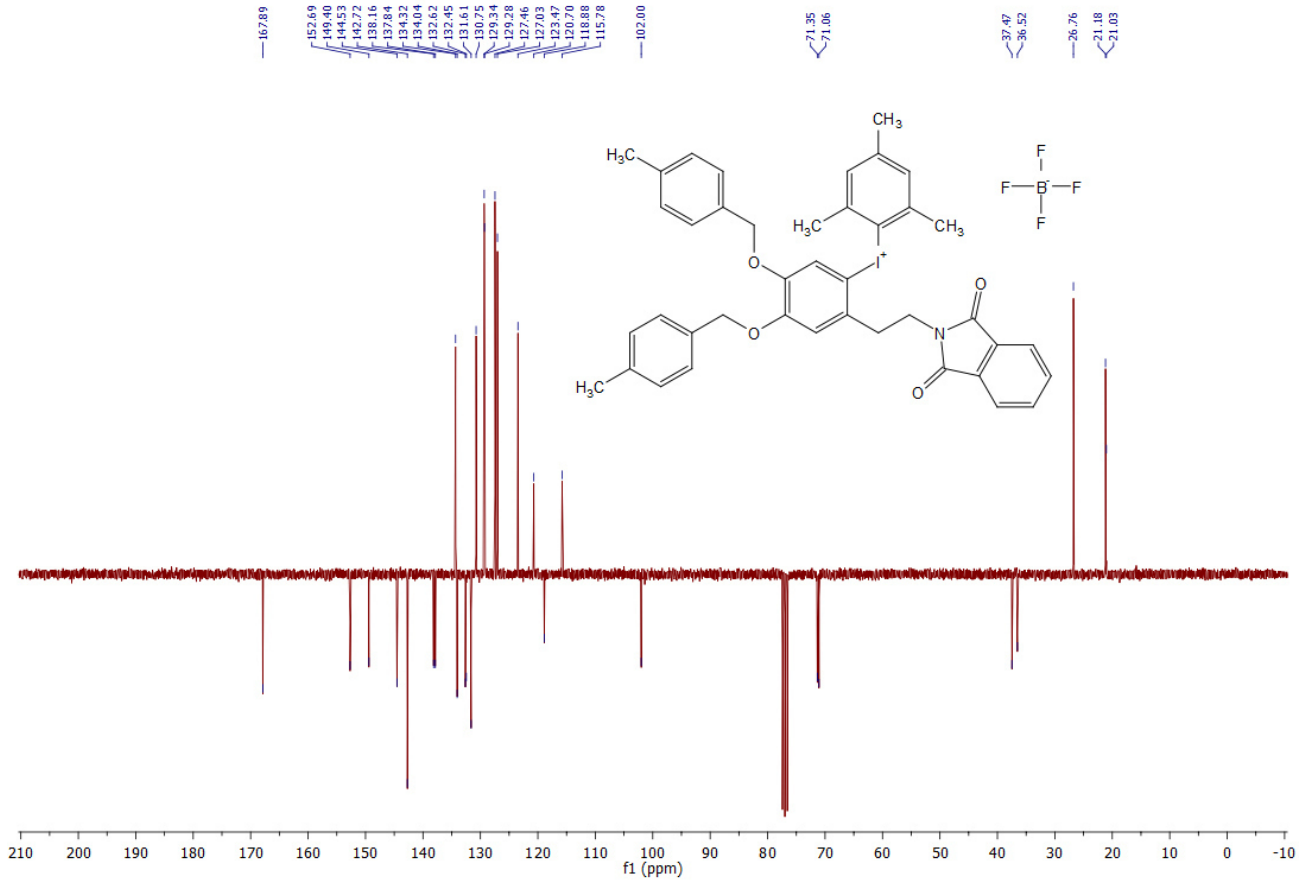
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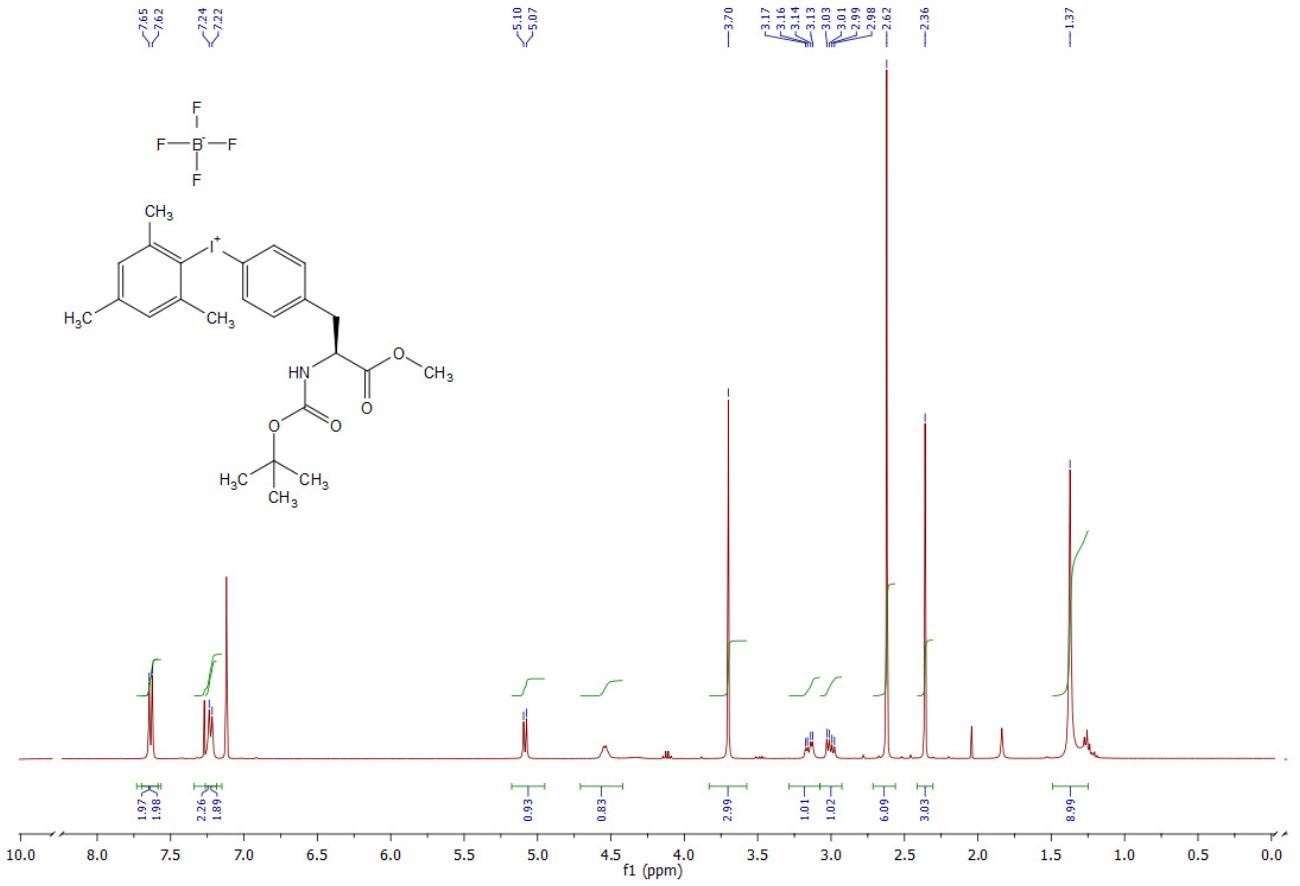
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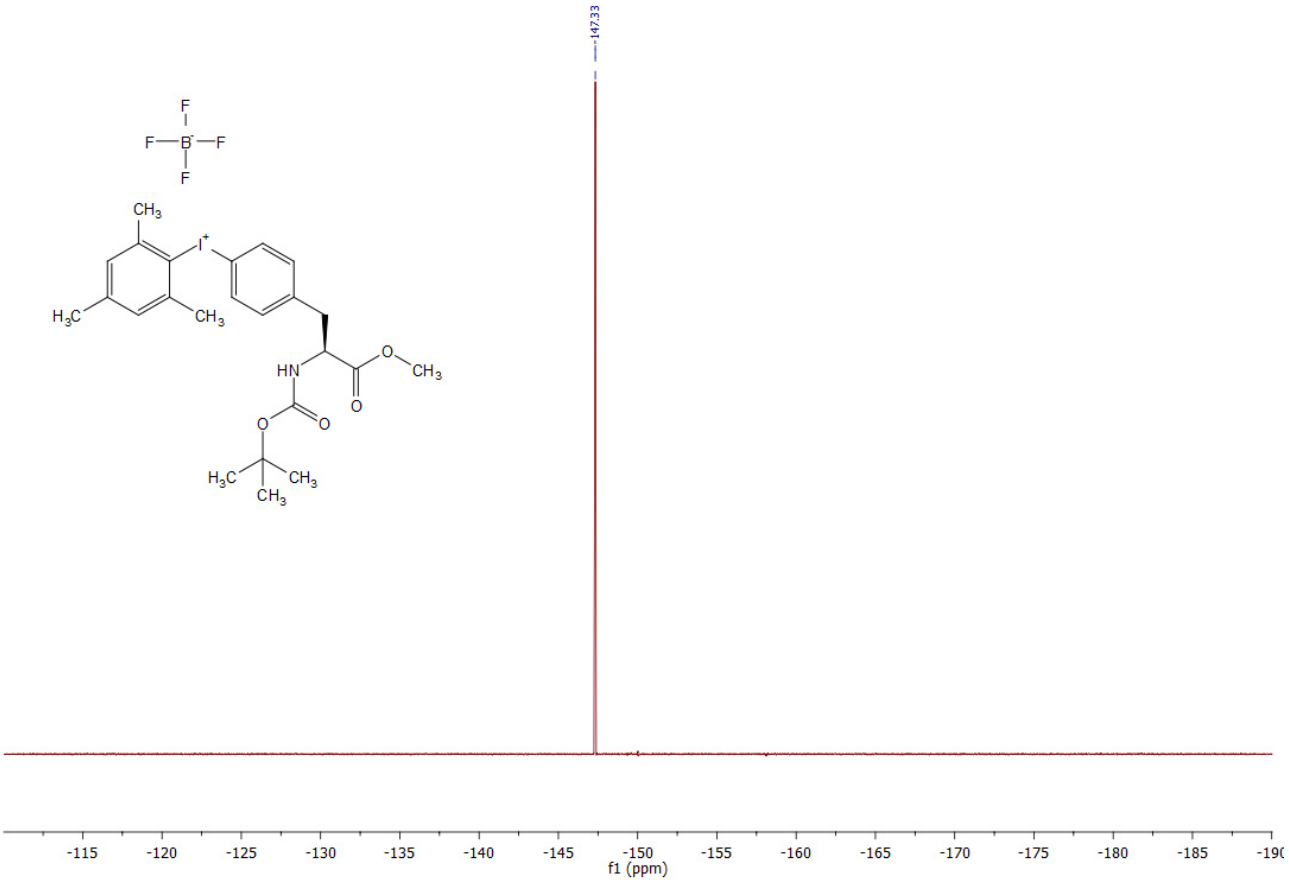
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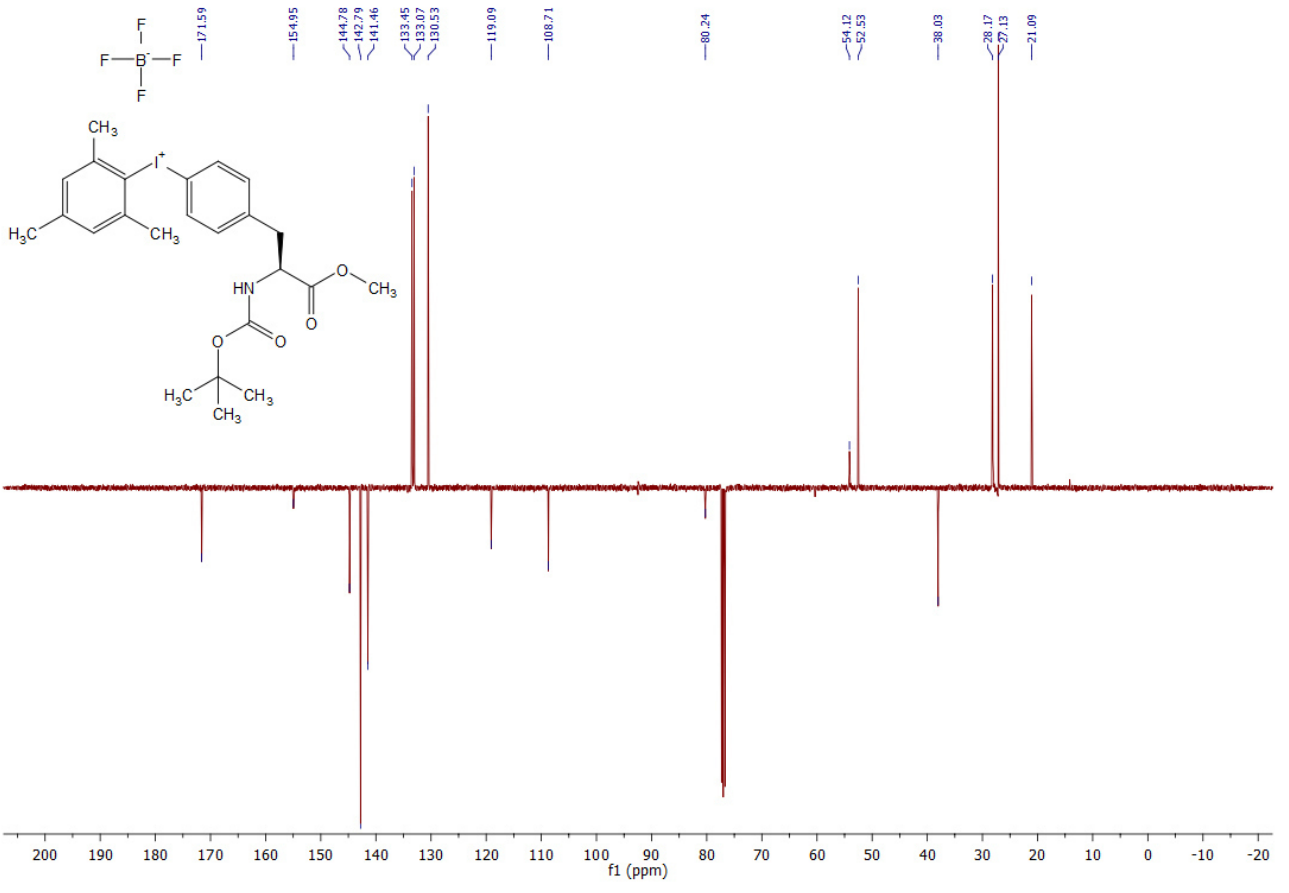
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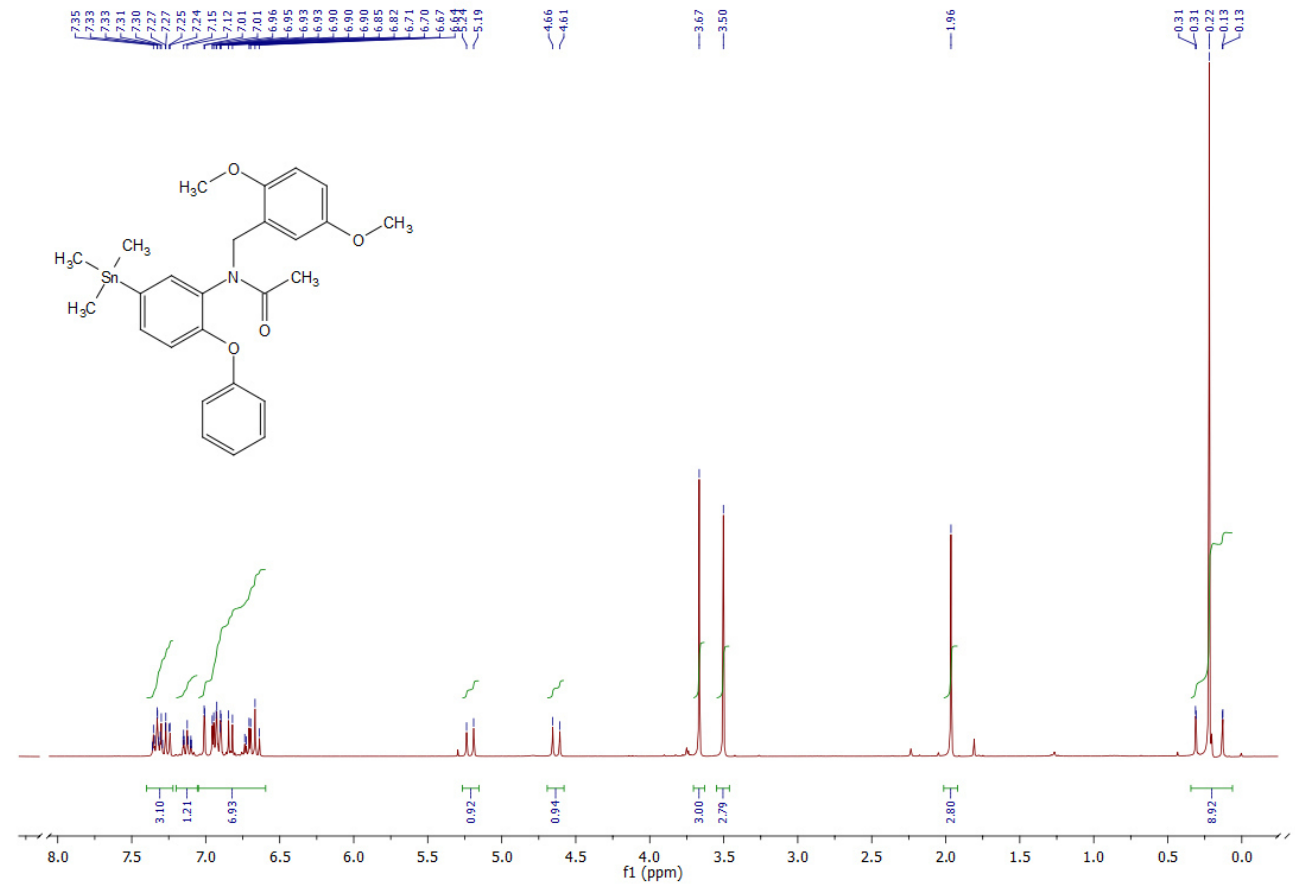
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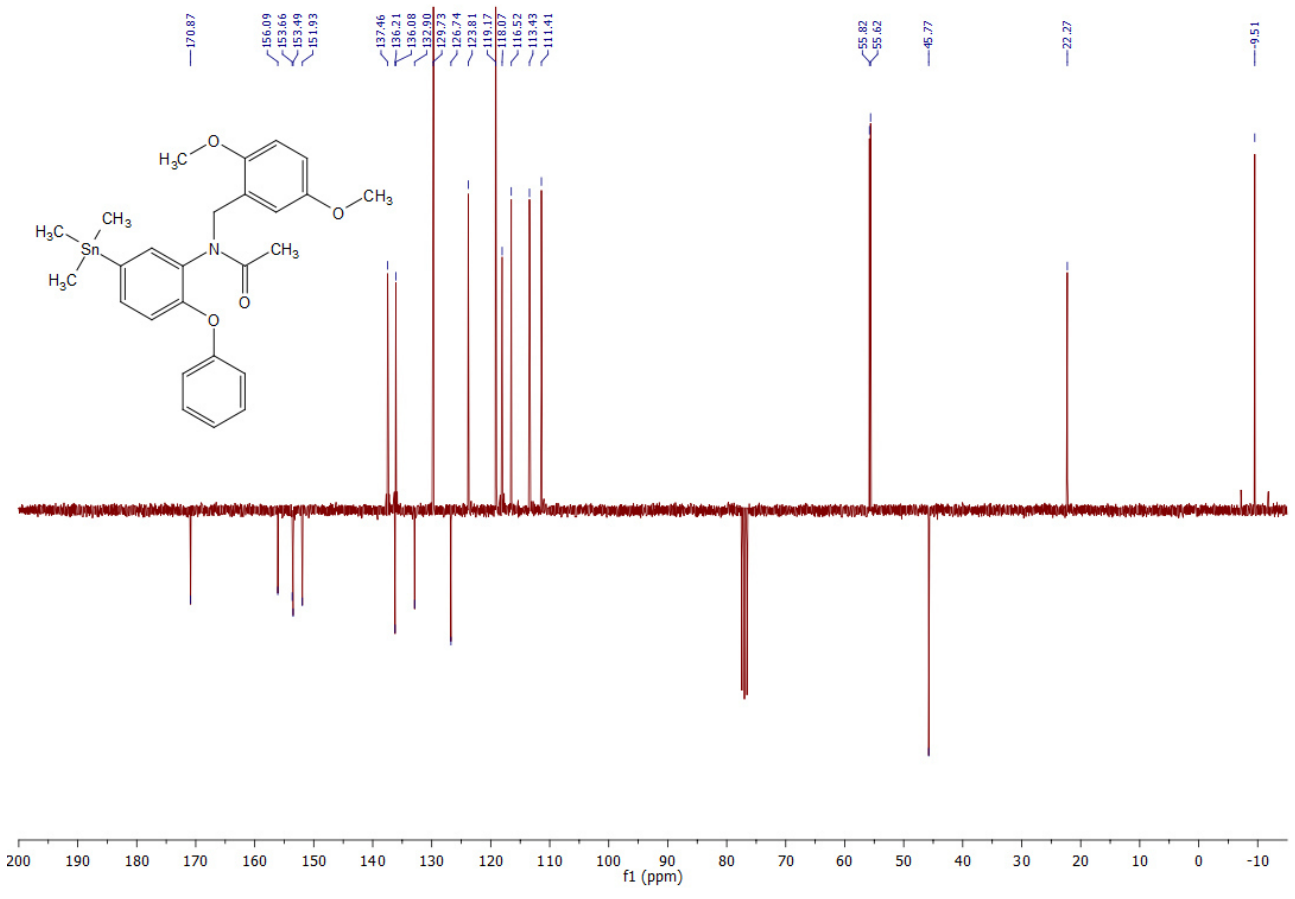
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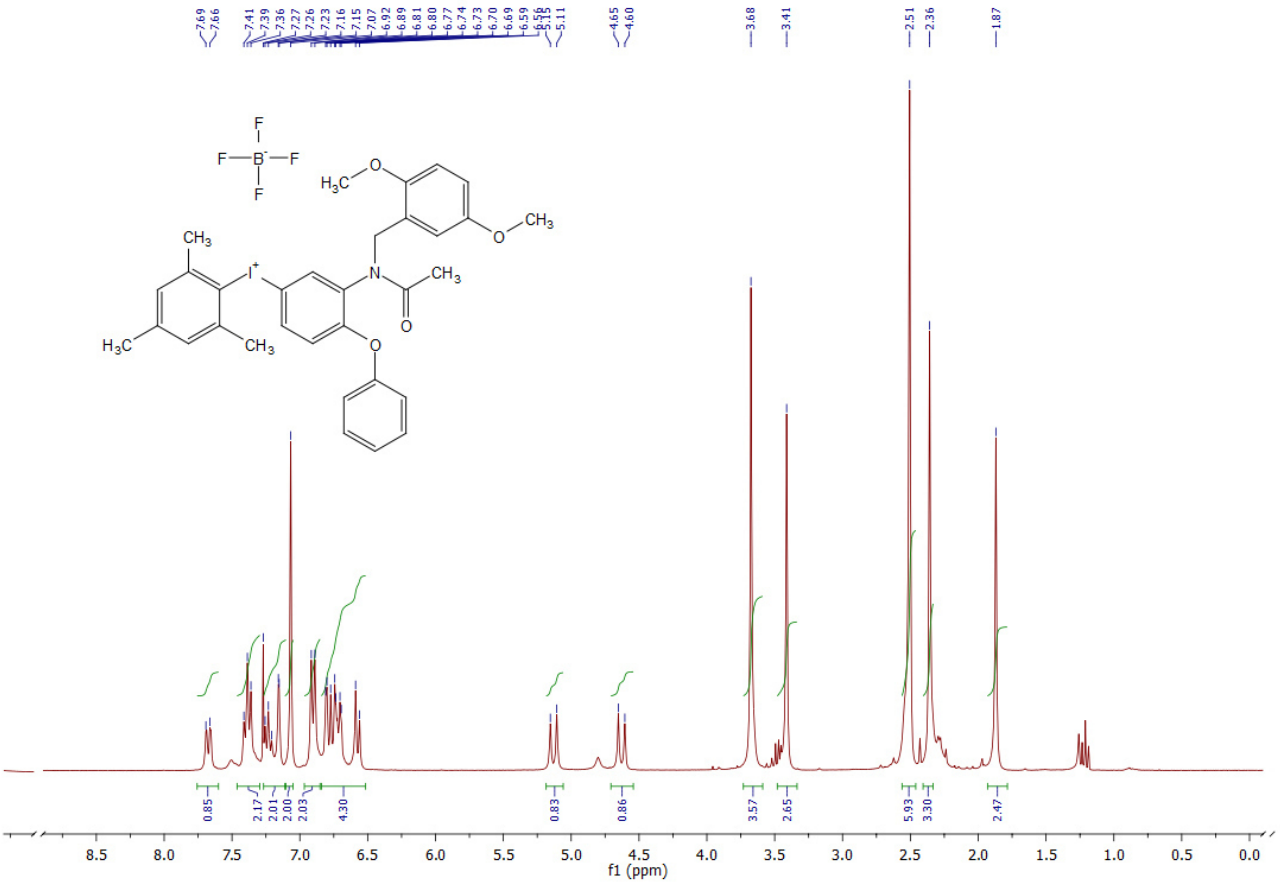
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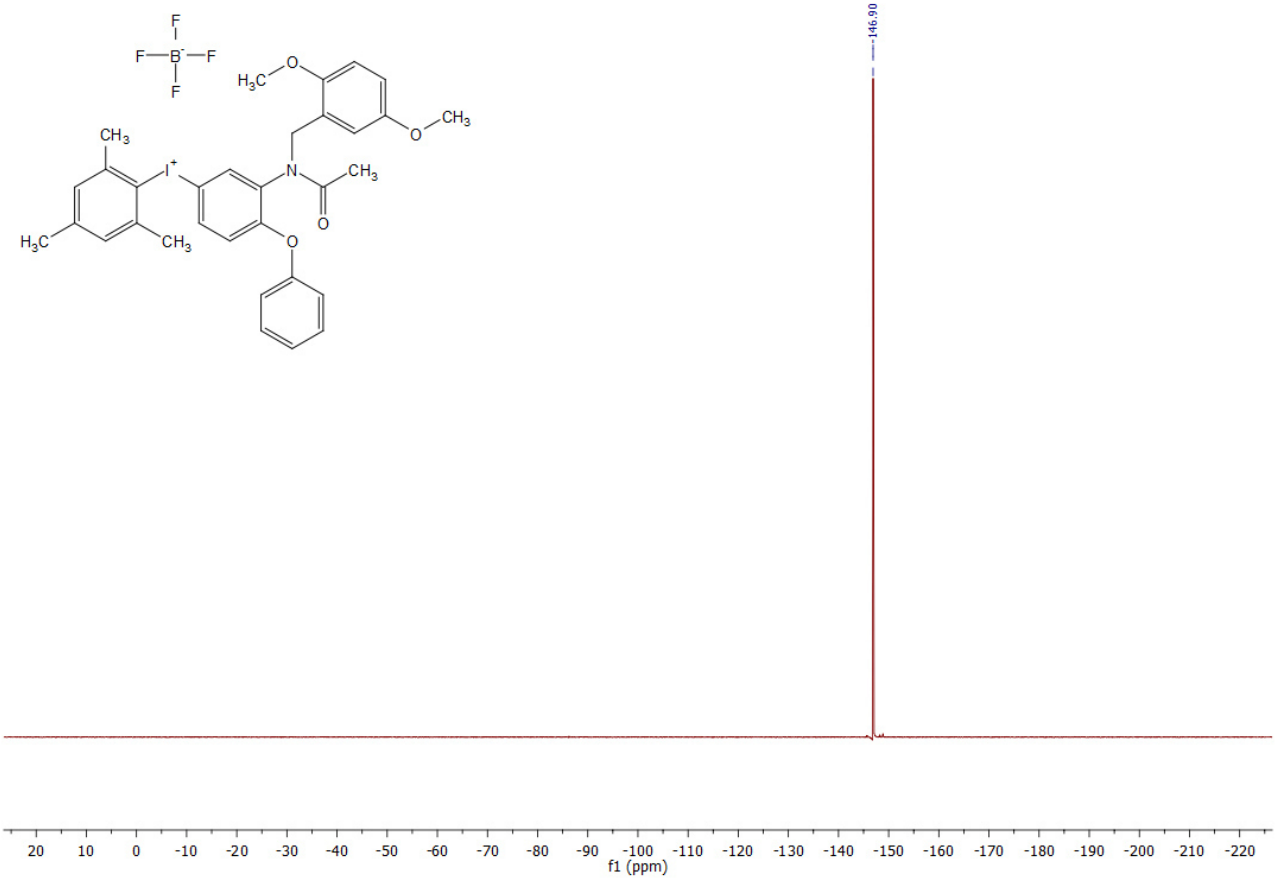
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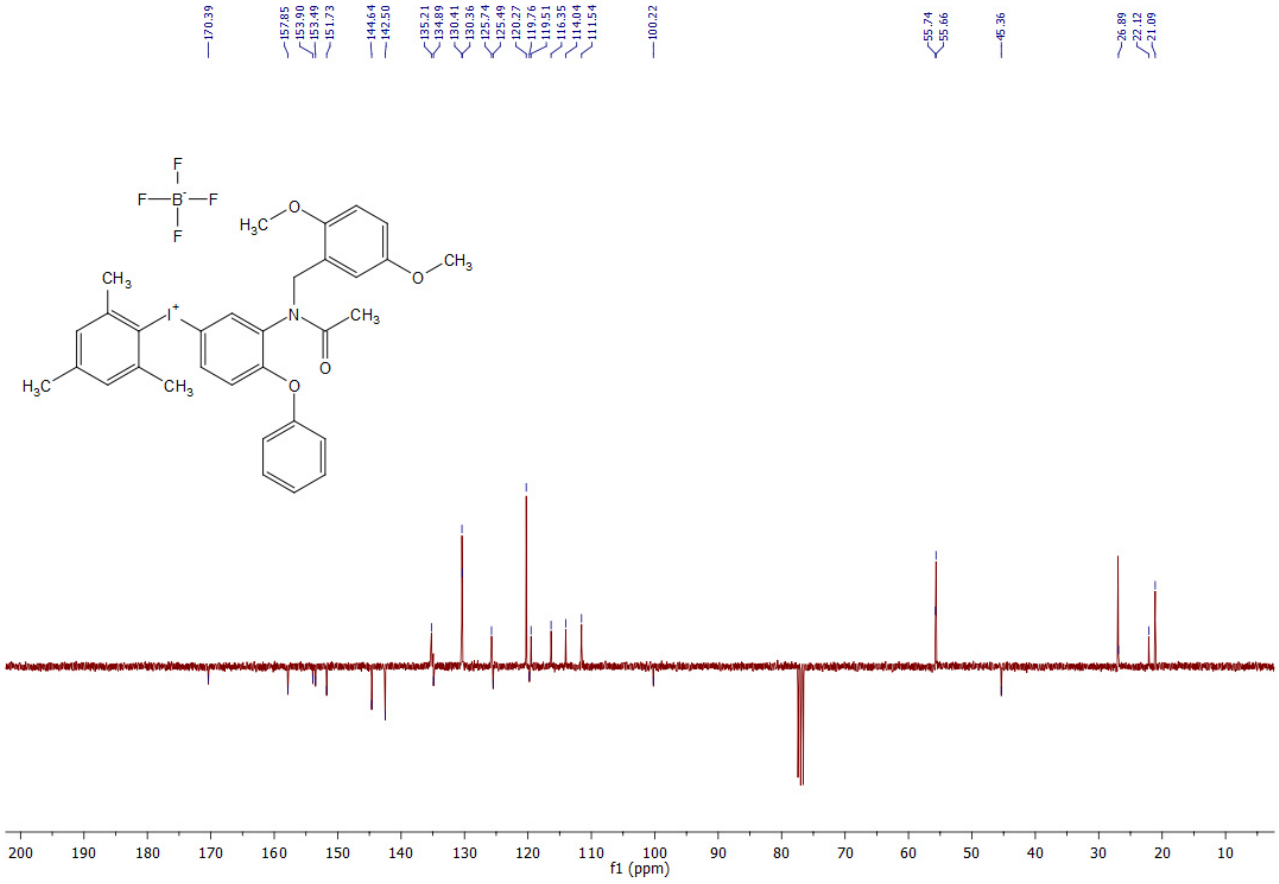
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Automated synthesis of 4-[^{18}F]fluoroanisole, [^{18}F]DAA1106 and 4-[^{18}F]FPhe using Cu-mediated radiofluorination under "minimalist" conditions

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HIGHLIGHTS

- Direct elution of [^{18}F]fluoride from an anion exchange resin with a iodonium salt precursor in MeOH.
- Neither azeotropic drying, nor base necessary.
- Purification of 4-[^{18}F]fluoroanisole, [^{18}F]DAA1106 and 4-[^{18}F]FPhe using solid phase extraction (SPE).
- Preparation of PET tracers in isolated radiochemical yields of 41–61% within 30–60 min.

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ABSTRACT

The application of the "minimalist" approach to Cu-mediated radiofluorination allows the efficient preparation of ^{18}F-labeled arenes regardless of their electronic properties. The implementation of this methodology on a commercially available synthesis module (hotbox^{three}, Scintomics, Germany) enabled the automated production of 4-[^{18}F]fluoroanisole as well as the clinically relevant PET-tracers, 4-[^{18}F]FPhe and [^{18}F]DAA1106, in radiochemical yields of 41–61% and radiochemical purities of $>95\%$ within 30–60 min. These results demonstrated the high efficacy and versatility of the developed method that will open up opportunities for a broad application of Cu-mediated radiofluorination in PET-chemistry.

1. Introduction

Radiofluorinated arenes are one of the most common motifs in the design of PET-ligands. Direct nucleophilic aromatic substitution with [^{18}F]fluoride on electron-deficient aromatic substrates enables a convenient access to n.c.a. ^{18}F-labeled compounds. However, until recently the preparation of radiofluorinated arenes labeled at unactivated and especially electron-rich positions using [^{18}F]F$^-$ was very challenging and required, e.g., the application of removable activating groups (Wagner et al., 2009). The utilisation of suitable aryliodonium (Pike and Aigbirhio, 1995; Ross et al., 2007) and diarylsulfonium (Mu et al., 2012; Sander et al., 2015) leaving groups allowed radiolabeling of electron-neutral and moderately electron-rich aromatics. Spirocyclic iodonium ylides (Rotstein et al., 2014; Satyamurthy and Barrio, 2010) were also successfully employed for the same purpose. However, although in many cases good to excellent HPLC or TLC radiochemical yields were originally reported, they usually did not take into account [^{18}F]fluoride adsorbed to the walls of the reaction vessel. Using these precursors, radiolabeled arenes could be prepared best only in fair to moderate isolated radiochemical yields {For the comparison of radiosyntheses of [^{18}F]FDA, 4-[^{18}F]FPhe and [^{18}F]DAA1106 via copper-mediated radiofluorination under "minimalist" conditions and using other preparation procedures, please, refer to: (Zlatopolskiy et al., 2015a)}. Furthermore, even

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from diaryliodonium and triarylsulfonium salts as well as iodonium ylides radiofluorinated electron-rich aromatic compounds like 6-[^{18}F]FMT and [^{18}F]fluoroindoles could be obtained only in low radiochemical yields. Extensive efforts to solve this problem ultimately led to the discovery of transition metal-mediated aromatic radiofluorination methodologies which could be efficiently used for the preparation of ^{18}F-labeled arenes and heteroarenes, including electron-rich ones, from nucleophilic [^{18}F]fluoride (Ichiishi et al., 2014; Tredwell et al., 2014a, 2014b; Zlatopolskiy et al., 2015b). Especially, the application of the "minimalist" approach (Richarz et al., 2014) to copper-mediated $^{18}F^-$ labeling (Zlatopolskiy et al., 2015a) allowed to produce ^{18}F-labeled arenes in good to excellent RCYs on a preparative scale regardless of their electronic properties. Additionally, this method allowed to circumvent time-consuming azeotropic drying and the application of base and other additives usually required to enhance ^{18}F nucleophilicity. Accordingly, [^{18}F]fluoride was directly eluted from the anion-exchange resin with a solution of the iodonium precursor in MeOH. After removal of MeOH the residual [^{18}F]fluoride iodonium salt was taken up in a solution of $(MeCN)_4CuOTf$ in DMF and heated under air.

The growing clinical impact of PET imaging results in an increasing demand for large-scale cGMP-compliant syntheses of various PET-tracers in a safe, operationally simple and reproducible manner. Therefore, novel preparation methodologies have to be transferred to automated synthesis modules in order to be applied for large-scale production of clinically relevant PET probes. Automation significantly reduces radiation exposure of the personnel and at the same time lowers costs of PET tracer production. However, automation suffers from several challenges, primarily owing to the loss of activity and/or solvents and chemicals during transfer steps and the inability to intervene during the synthesis. Furthermore, commercially available modules are not well-suited for the implementation of complex multistep preparation procedures and radiosyntheses which have to be carried out under controlled conditions (e.g., complete exclusion of oxygen and/or moisture). Moreover, they are usually incompatible with highly corrosive reagents and high pressures.

The exceptional operational simplicity and high reliability of copper-mediated radiofluorination under "minimalist" conditions prompted us to study amenability of this radiolabeling procedure to automation in commercially available synthesis modules and, consequently, its suitability for clinical PET applications.

2. Results

Initially, the preparation of 4-[^{18}F]fluoroanisole ([^{18}F]**2**) from (4-anisyl)(mesityl)iodonium tetrafluoroborate (**1**) (Zlatopolskiy et al. 2015a) (Fig. 1) was used as a model radiosynthesis to adapt the module setup and optimize the synthesis parameters for the automated Cu-mediated radiofluorination.

The first crucial step was the loading of $^{18}F^-$ in $H_2{}^{18}O$ on the QMA cartridge from the male side followed by washing and elution of $^{18}F^-$ with a solution of radiolabeling precursor 1 in MeOH from the female side of the cartridge (Fig. 2). This allowed

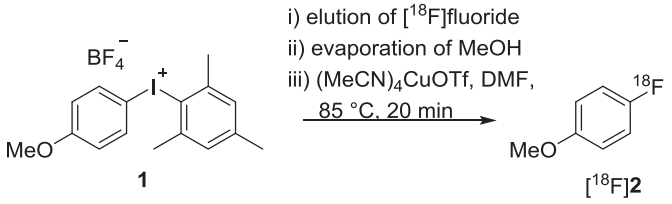


Fig. 1. Synthesis of 4-[^{18}F]fluoroanisole ([^{18}F]**2**).

recovering more than 90% of the $^{18}F^-$-activity. If loading and elution of $^{18}F^-$ was carried out from the same side of the cartridge, a significant amount of activity (up to 40–50%) remained on the anion exchange resin.

The second crucial step was the careful drying of the preformed iodonium [^{18}F]fluoride residue. Usually, MeOH can be removed already within 1–2 min. However, pilot experiments demonstrated a significant decrease of radiochemical yields (RCYs) in the presence of even traces of MeOH. Therefore, to ensure complete MeOH removal the evaporation time was extended to 5 min. The resulting iodonium [^{18}F]fluoride was taken up in a solution of $(MeCN)_4CuOTf$ in DMF and heated at 85 °C for 20 min. Finally, SPE purification afforded [^{18}F]**2** in 61% RCY and $>95\%$ radiochemical purity within 30 min (Fig. 3).

Once the protocol for the automated Cu-mediated radiofluorination under "minimalist" conditions had been established we focused on the production of clinically relevant PET-tracers.

DAA1106 (**4**) (Fig. 4) exhibits an exceptionally high affinity for the translocator protein 18 kDa (TSPO). TSPO is involved in various processes like neuroinflammation and reactive gliosis, associated with different neurodegenerative disorders and brain tumors (O'Brien et al., 2014; Rupprecht et al., 2010). ^{11}C-Labeled DAA1106 was already used to visualize different pathologies in patients (Maeda et al., 2004; Takano et al., 2010; Zhang et al., 2003). Recently, we prepared ^{18}F-labeled DAA1106 manually and evaluated its in vivo properties in a rat stroke model. In these experiments the tracer showed excellent potential to visualize neuroinflammation and microglia activation.

The automated preparation of [^{18}F]4 was carried out using the same configuration and parameters as for 4-[^{18}F]fluoroanisole. This preparation protocol afforded the desired radiotracer in 41% RCY and $>95\%$ RCP after HPLC purification within 30 min (Fig. 5). A specific activity of 66 GBq/μmol was determined for the HPLC-purified product (4.2 GBq). Alternatively, crude [^{18}F]**4** was purified by SPE affording the desired tracer in $>95\%$ RCP containing some non-radioactive impurities. The resulting ethanolic solution of the tracer was, after dilution with aqueous PEG400, directly used for preclinical in vivo experiments in rats. The application of SPE purified [^{18}F]4 did not engender any observable toxic side effects in animals (more than 30 animals were measured). Furthermore, in preliminary comparative PET studies the images revealed no difference in image quality between SPE- and HPLC-purified tracer in the same animal.

Finally, the amenability for automation of the more complex, two-step radiosynthesis of L-4-[^{18}F]fluorophenylalanine [4-[^{18}F]FPhe ([^{18}F]**7**)] was evaluated. This tracer could be potentially useful for the visualization of the protein synthesis, e.g. in brain tumors, owing to high amino acid transporters affinity and protein incorporation rate (Arnstein and Richmond, 1964). Especially, the high protein incorporation of 4-[^{18}F]FPhe could be of advantage in comparison to the widely used 4-(2-[^{18}F]fluoroethyl)-L-tyrosine ([^{18}F]FET). Tumor uptake of [^{18}F]FET is solely based on its intracellular transport via amino acid transporters (Coenen et al., 1989).

For the preparation 4-[^{18}F]FPhe radiofluorination precursor **5** (Zlatopolskiy et al., 2015a) was used (Fig. 6). Purification of the intermediate [^{18}F]6, was carried out using solid phase extraction. [^{18}F]6 was eluted from the cartridge into a second reaction vial (Fig. 7). After addition of 12 N HCl, deprotection of [^{18}F]**6** was accomplished at 140 °C for 10 min. Finally, enantiomerically pure 4-[^{18}F]FPhe ([^{18}F]**7**) [$>98\%$ ee; (*R*)-isomer was not observed; Fig. 8] was isolated by semi-preparative HPLC in an overall RCY of 56% and excellent RP of more than 95% as a ready-to-use solution in 4% ethanolic phosphate buffer within 60 min. Alternatively, 4-[^{18}F]FPhe was isolated by SPE using three cartridges in series. The crude mixture was diluted with H_2O and loaded on a HR-P M

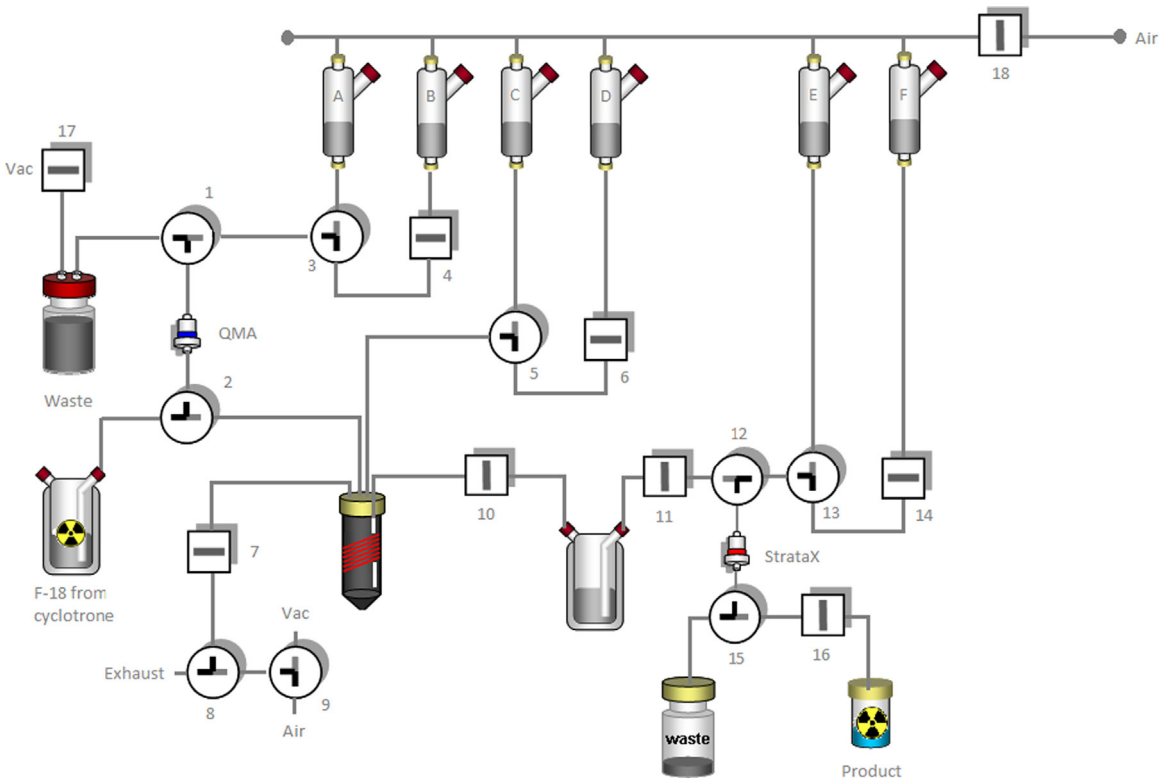


Fig. 2. Flow scheme for the automated radiosynthesis of 4-[^{18}F]fluoroanisole ([^{18}F]**2**) and [^{18}F]DAA1106 ([^{18}F]**4**) (Scintomics HotBox^{three}). A: MeOH (1 mL); B: Iodonium precursor **1** or **3** (21 μmol) in MeOH (500 μL); C: $(MeCN)_4CuOTf$ (12 μmol) in DMF (300 μL); D: H_2O (2 mL); E: H_2O (5 mL); F: EtOH (400 μL).

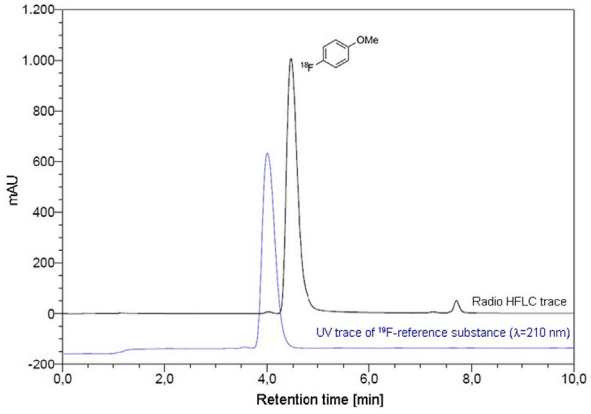


Fig. 3. Radio-HPLC chromatogram of SPE-purified 4-[^{18}F]fluoroanisole ([^{18}F]**2**).

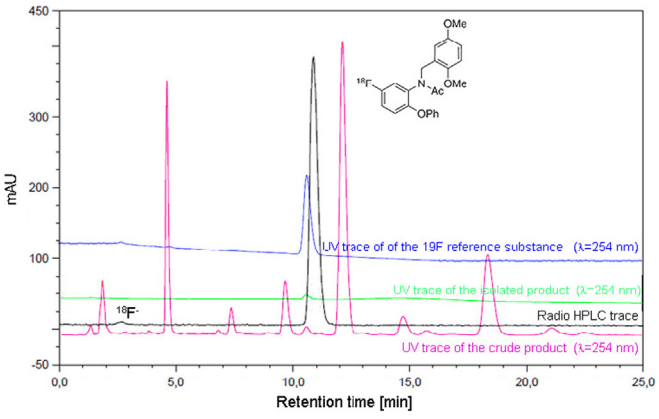
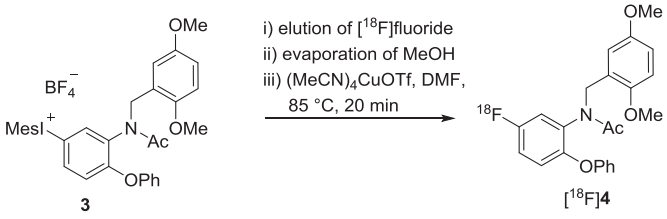


Fig. 5. Radio-HPLC chromatogram of [^{18}F]DAA1106 ([^{18}F]**4**) purified by HPLC.



3 [^{18}F]**4**

Fig. 4. Preparation of [^{18}F]DAA1106 ([^{18}F]**4**).

cartridge. After washing with H_2O the partially purified 4-[^{18}F]FPhe was eluted with 20% ethanolic 0.02 M NaH_2PO_4 (pH=2.5). To remove non-polar impurities the eluate was passed through a C18ec S followed by an Oasis WAX cartridge (Fig. 9). For SPE-purified [^{18}F]**7** (15 GBq) a specific activity of 86 GBq/μmol was determined.

The amount of Cu in the final solutions of purified [^{18}F]DAA1106 and 4-[^{18}F]FPhe was determined by using inductively coupled plasma mass spectrometry (ICP-MS). The Cu content varied from 0.5 to 3.5 ppm. This is far below any level of concern according to the ICH Guideline of Elemental Impurities (Q3D).

3. Conclusion

We have demonstrated that Cu-mediated nucleophilic aromatic radiofluorination under "minimalist" conditions can be easily transferred to a commercially available synthesis module (Table 1). The utility of the developed procedure was confirmed by a successful preparation of [^{18}F]DAA1106 and 4-[^{18}F]FPhe in good RCYs and excellent RCPs. The evident simplicity as well as the high

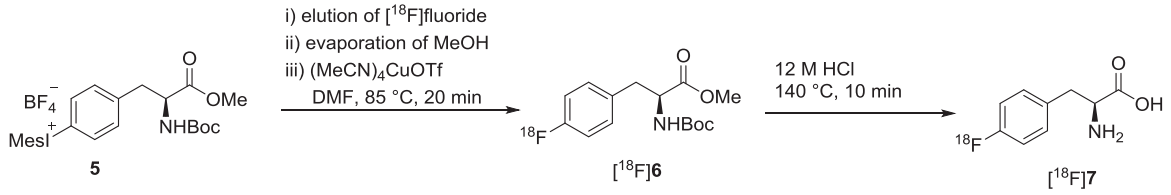


Fig. 6. Radiosynthesis of L–4-[^{18}F]fluorophenylalanine ([^{18}F]**7**).

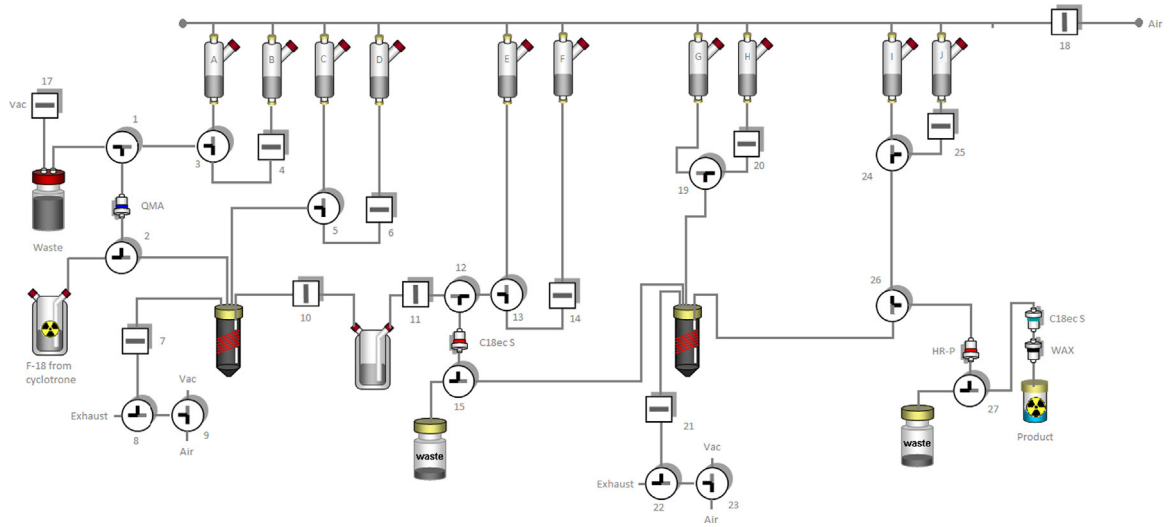


Fig. 7. Flow scheme for the automated radiosynthesis of 4-[^{18}F]FPhe ([^{18}F]**7**) (Scintomics HotBox^{three}). A: MeOH (1 mL); B: Iodonium precursor **5** (21 μmol) in MeOH (500 μL); C: $(MeCN)_4CuOTf$ (12 μmol) in DMF (300 μL); D: H_2O (2 mL); E: H_2O (5 mL); F: EtOH (400 μL); G: 12 M HCl (300 μL); H: H_2O (2 mL); I: H_2O (5 mL); J: 0.02 M NaH_2PO_4 solution (5 mL, pH 2.5, 20% EtOH).

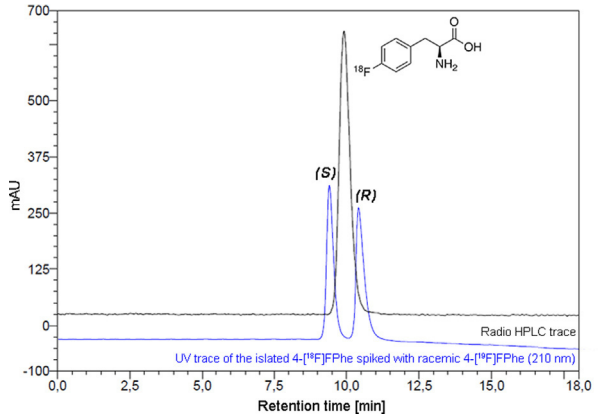


Fig. 8. Determination of the enantiomeric purity of 4-[^{18}F]FPhe. Conditions: column: Astec® CHIROBIOTIC® T 250 × 4.6 mm 5 μm (Supelco); eluent 20% MeCN; flow rate:1.5 mL/min; t_R:10 min.

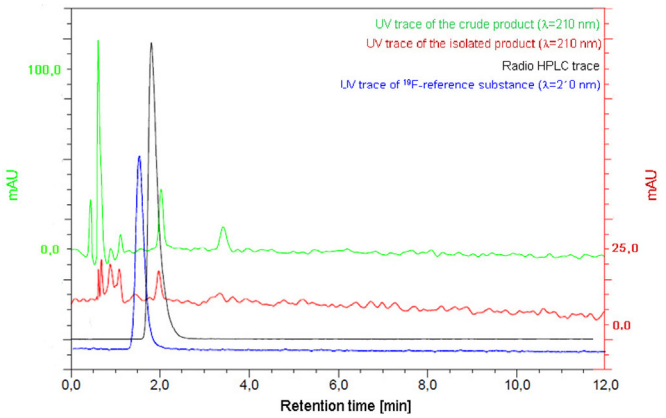


Fig. 9. HPLC chromatograms of 4-[^{18}F]FPhe ([^{18}F]**7**) isolated by SPE.

efficacy and versatility of the developed protocol enables a broad application of Cu-mediated radiofluorination in PET-chemistry in the very near future.

4. Experimental

4.1. Materials and methods

All radiosyntheses were carried out in a Scintomics HotBox^{three} module (Scintomics GmbH, Fürstenfeldbruck, Germany). All connections between the valves were made of PTFE tubing and PEEK

Table 1
Automated preparation of ^{18}F-labeled compounds using Cu-mediated radiofluorination under "minimalist" conditions.

Entry	Product	RCY ± SD [%] [1]	SA [GBq/μmol] (Product activity [GBq])
1	[^{18}F]**2**	61 ± 4	
2[2]	[^{18}F]**4**	41 ± 3	66 (4.2)
3[2]	[^{18}F]**7**	56 ± 5	109 (14)
4[3]	[^{18}F]**7**	42 ± 6	86 (15)

[1] Each experiment was carried out at least in triplicate. Purification of the products was carried out by [2] semi-preparative HPLC or [3] SPE.

fittings. The flow schemes for the preparation of [^{18}F]**2** and [^{18}F]**4** as well as [^{18}F]**7** are depicted in Figs. 2 and 7, respectively. Synthetic air (Westfalen AG, Muenster, Germany) was used as an operating gas.

4.2. Radiochemistry

4.2.1. Production of [^{18}F]fluoride and radiolabeling procedure

[^{18}F]Fluoride was produced via the ^{18}O(p,n)^{18}F reaction by bombardment of enriched [^{18}O]water with 16.5 MeV protons using a MC16 cyclotron (Scanditronix, Uppsala, Sweden) at the Max Plnack Institute for Metabolism Research. Aqueous [^{18}F]fluoride (25–37 GBq) was trapped on a SepPak Light Waters Accell™ Plus QMA cartridge (Waters GmbH, Eschborn, Germany), preconditioned with 1 mL of water. The cartridge was washed with 2 mL of MeOH and flushed with air (1 min). [^{18}F]Fluoride was eluted from the resin with the corresponding iodonium salt precursor (21 μmol) in MeOH (500 μL). Methanol was evaporated under air flow at 70–80 °C. The residue was dissolved in a solution of $(MeCN)_4CuOTf$ (12 μmol) in DMF (300 μL) and the reaction mixture was heated at 85 °C for 20 min. The resulting solution was cooled to room temperature and diluted with water (2 mL). The mixture was transferred into a vessel containing water (20 mL). Afterwards the radiolabeled product was trapped on a SPE cartridge (StrataX, Phenomenex), washed with water (5 mL) and eluted with EtOH (500 μL).

4.2.2. HPLC purification of [^{18}F]**4**

Column: Luna 5μ C18(2) 100 A, 250 × 4.6 mm (Phenomenex, Aschaffenburg, Germany); 60% MeCN, flow rate:1.5 mL/min (t_R = 10.8 min). Afterwards the product fraction was diluted with water to a total volume of 20 mL. The product was trapped on a C18ec cartridge and eluted with ethanol.

4.2.3. Deprotection of [^{18}F]**6**; isolation of 4-[^{18}F]Phe using HPLC

The protected intermediate [^{18}F]**6** was eluted from the C_{18}-cartridge with EtOH (500 μL) into a second reaction vessel. EtOH was evaporated and 12 M HCl (300 μL) was added. The resulting mixture was heated at 140 °C for 10 min. Acetone (1–2 mL) was added and the resulting solution was evaporated to dryness. The residue was taken up in a solution containing 4% EtOH in 0.02 M NaH_2PO_4 (500 μL, pH 2.5). 4-L-[^{18}F]fluorophenylalanine (4-[^{18}F]Phe) was isolated using HPLC. HPLC:column:Synergy 4 μm Hydro-RP 150 × 4.6 mm (Phenomenex, Aschaffenburg, Germany); eluent: 4% EtOH in 0.02 M NaH_2PO_4 (pH 2.5); flow rate:1.5 mL/min; t_R = 3.0 min.

4.2.4. SPE isolation of [^{18}F]**7**

Solution of [^{18}F]**7** in 12 M HCl (vide supra) was diluted with H_2O (10 mL) and passed through a Chromabond® HR-P cartridge (200 mg, Macherey-Nagel). The cartridge was washed with H_2O (10 mL) and the product eluted with a solution containing 20% EtOH in 0.02 M NaH_2PO_4 (pH 2.5) through a C18ec followed by an Oasis WAX cartridge.

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Radiolabeling

Alcohol-Enhanced Cu-Mediated Radiofluorination

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Abstract: The potential of many ^{18}F-labeled (hetero)aromatics for applications in positron emission tomography remains underexplored because convenient procedures for their radiosynthesis are lacking. Consequently, simple methods to prepare radiofluorinated (hetero)arenes are highly sought after. Herein, we report the beneficial effect of primary and secondary alcohols on Cu-mediated ^{18}F-labeling. This observation contradicts the assumption that such alcohols are inappropriate solvents for aromatic fluorination. Therefore, we developed a protocol for rapid radiolabeling of an extraordinarily broad scope of boronic and stannyl substrates under general reaction conditions. Notably, radiofluorinated indoles, phenols, and anilines were synthesized directly from the corresponding unprotected precursors. Furthermore, the novel method enabled the preparation of radiofluorinated tryptophans, [^{18}F]F-DPA, [^{18}F]DAA1106, 6-[^{18}F]FDA, and 6-[^{18}F]FDOPA.

Molecular imaging technologies are indispensable tools in modern medicine, enabling the noninvasive visualization of biochemical processes in vivo at a molecular level. Positron emission tomography (PET) and related hybrid methods (like PET/CT and PET/MR) are the most important imaging techniques with unique sensitivities that allow detection of cellular changes during disease development. PET delineates the biodistribution of molecular probes labeled with β^+-emitting nuclides by detecting antiparallel γ-photons originated from positron/electron annihilation. Among β^+-emitting nuclides for PET, fluorine-18 represents the most attractive one because of the easy accessibility in "no carrier added" (n.c.a.) form, convenient half-life (109.8 min) and low β^+-energy. However, the diagnostic potential of many ^{18}F-labeled PET tracers cannot be fully exploited because simple, efficient procedures for their preparation are lacking. Recently, several pioneering fluorination methods have been developed and transferred to PET chemistry.[1] Among them, transition-metal-mediated fluorinations,[2] and especially, Cu-mediated ones,[3] are exceptionally versatile routes to ^{18}F-labeled aromatics, regardless of their electronic properties (Figure 1).

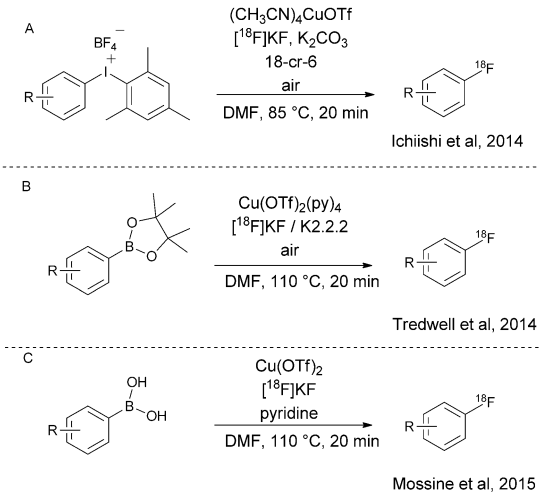

Figure 1. Copper-mediated ^{18}F-labeling of diaryliodonium salts (A), pinacol arylboronates (B), and boronic acids (C).

In particular, Cu-mediated radiofluorination of (aryl)(mesityl)iodonium salts under "minimalist" conditions[4] allows a simplified preparation of radiotracers that are not easily accessible by conventional radiochemistry. Azeotropic drying, bases or other additives are not required. Consequently, only precursor, [^{18}F]fluoride and a copper catalyst are used (Figure 2).[5]

In contrast, reported protocols for the Cu-mediated radiofluorination of pinacol arylboronates and arylboronic acids require azeotropic drying, a base and additives, such as pyridine, 2.2.2-cryptand (K2.2.2), KOTf, and $K_2C_2O_4$.[3b,c] Furthermore, in some cases, unacceptably high losses of $^{18}F^-$ (up to 40–50%)[6] due to adsorption onto vessel walls occur. To overcome these shortcomings, we tried to refine the Cu-mediated radiofluorination of boronic substrates. We found that the application of primary and secondary alcohols as co-solvents substantially increased radiolabeling yields. Consequently, this observation was used to develop a convenient procedure for Cu-mediated radiofluorination of boronic and stannyl substrates.

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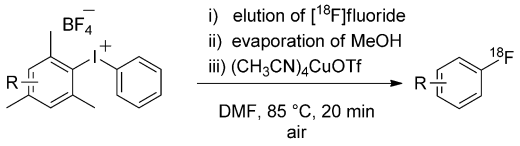

Figure 2. Copper-mediated ^{18}F-labeling of diaryliodonium salts under "minimalist" conditions.

Initially, to obviate azeotropic drying, we tested the elution of $^{18}F^-$ from an anion-exchange resin (QMA cartridge) using a solution of K_2CO_3, KOTf, K2.2.2, and pyridine in DMF. Before the elution the resin was washed with anhydrous DMF and dried in air to remove residual water after $^{18}F^-$ trapping. Less than 40% of $^{18}F^-$ was recovered from the QMA cartridge. Possibly, the poor wettability of the hydrophilic alkyltrimethylammonium phase with aprotic solvents led to poor $^{18}F^-$ recovery (Figure S1, Supporting Information).

In order to prevent resin collapse, we used hexane instead of DMF. Consequently, the resin was washed with hexane and dried with air before the elution step. Because hexane is immiscible with water, water in the pores of the hydrophilic stationary phase was not displaced. Indeed, if the cartridge was rinsed with hexane, the elution efficacy increased to 80%. We applied the resulting eluate for the Cu-mediated radiofluorination of pinacol phenylboronate (PhBPin). However, only traces of [^{18}F]fluorobenzene ([^{18}F]**1**) were observed in the reaction mixture, demonstrating the high sensitivity of the reaction towards water.

The application of alcohols for water removal was investigated.[7] Alcohols are protic solvents and are capable of completely displacing H_2O from the resin pores. Simultaneously they can prevent resin collapse. By using alcohols, we achieved a moderate $^{18}F^-$ recovery (up to 55%).[8] Direct heating of the eluates containing traces of alcohols with PhBPin and $Cu(py)_4(OTf)_2$ afforded [^{18}F]**1** in unexpectedly high radiochemical conversions (RCCs)[9] of 70–80%. For comparison, use of pure dimethylacetamide (DMA) or DMF as reaction solvent afforded [^{18}F]**1** in $\leq$65% RCC. These radiolabeling experiments were carried out according to Zlatopolskiy et al.[5b] and Preshlock et al.[3c]

Intrigued, we studied the influence of different alcohols on Cu-mediated radiofluorination systematically. $^{18}F^-$ was eluted from the anion exchange resin with a solution of Et_4NHCO_3 (2.7 mg) in the corresponding alcohol (0.4 mL; see Table 1) into a vial containing a solution of PhBPin and $Cu(py)_4(OTf)_2$ (60 and 26.5 μmol, respectively) in DMA (0.8 mL; final concentration of alcohol 33%). Subsequently, the resulting solutions were directly heated at 110 °C for 20 min in an air atmosphere. For practical reasons we changed from the elution with K_2CO_3/K2.2.2/KOTf/Py to the elution with Et_4NHCO_3 since solutions of K_2CO_3/K2.2.2/KOTf/Py have only very limited shelf-life and are relatively cumbersome to prepare. At the same time the elution efficacy and RCCs of the radiofluorination step with Et_4NHCO_3 was comparable or even better as those with K_2CO_3/K2.2.2/KOTf/Py.

Table 1. ^{18}F-Recovery and ^{18}F-incorporation (RCC) using different alcohols as co-solvents.[10]

	^{18}F recovery [%] (n ≥ 3)	RCC ± SD from PhBPin [%] (n ≥ 3)	RCC ± SD from $PhB(OH)_2$ [%] (n ≥ 3)
H_2O	99 ± 1	0	0
MeOH	97 ± 1	40 ± 3	67 ± 3
EtOH	91 ± 2	59 ± 2	90 ± 4
n-PrOH	70 ± 4	82 ± 3	96 ± 2
iso-PrOH	71 ± 2	62 ± 4	86 ± 2
*n*BuOH	80-95*	94 ± 2	98 ± 4
*i*BuOH	84 ± 6	89 ± 4	93 ± 3
*t*BuOH	35 ± 4	29 ± 3	46 ± 4
2-BuOH	43 ± 5	82 ± 1	85 ± 5
1-OctOH	46 ± 3	50 ± 5	90 ± 4
DMA w/o alcohol		82 ± 5	13 ± 2

^{18}F-Incorporation yields of 90–99% were observed using *n*BuOH and *i*BuOH, whereas *n*-PrOH and 2-BuOH delivered RCCs of >80%. Noteworthy, *n*BuOH enabled the most efficient elution of $^{18}F^-$ [radioactivity recovery amounted to 80–95% ($n > 100$)].[7]

Next, we evaluated the alcohol-enhanced, Cu-mediated ^{18}F-fluorination of $PhB(OH)_2$ as a model arylboronic acid (Figure 5).

Incorporation yields of ≥85% for most alcohols were observed. Only MeOH and *t*BuOH afforded [^{18}F]**1** in lower RCCs of 67 and 46%, respectively. In the absence of alcohols DMA or DMF afforded RCCs of only ≤12%.

Interestingly, our results contradict previous observations concerning the deleterious effect of protic solvents, including alcohols, on S_NAr fluorination, usually explained by extensive hydrogen bonding, which reduces the nucleophilicity of fluoride.[11] Unsurprisingly, to date, no transition-metal-catalyzed fluorinations in alcoholic media have been reported. Hence, we studied the tolerance of the radiofluorination reaction with respect to the alcohol content (Figure 3).

In 20–60% *n*BuOH in DMA, a near quantitative (>96%) formation of [^{18}F]**1** from PhBPin occurred. Even in 70–80% *n*BuOH, RCCs of >80% were observed. However, in pure *n*BuOH, ^{18}F-incorporation did not exceed 8%. Radiolabeling of $PhB(OH)_2$ was even more tolerant towards alcohols. With this substrate we tested environmentally benign EtOH, which furnished RCCs of >90% at 10–50% alcohol content. A further increase of the alcohol content resulted in a decrease of ^{18}F-incorporation to 75, 70, and 66% in 60, 80, and 90% ethanolic solutions, respectively. Nevertheless, radiolabeling even in pure EtOH still afforded [^{18}F]**1** in a RCC of 45%. The kind of aprotic solvent had a significant influence on RCCs. Thus, using pinacol 4-methoxyphenylboronate as a model substrate, DMA and *N*-methyl-2-pyrrolidone (NMP) afforded the highest RCCs of 4-[^{18}F]fluoroanisole ([^{18}F]**2**) of 87 and 77%, respectively.[7] DMF and sulfolane yielded radiolabeled arenes in moderate RCCs of 50%. ^{18}F-Incorporation in DMSO and acetonitrile was rather low (11 and 3%, respectively).

Additionally, radiolabeling kinetics was studied. At 110 °C incorporation yields of >80% and >90% were observed already after 5 and, 10 min, respectively.[10]

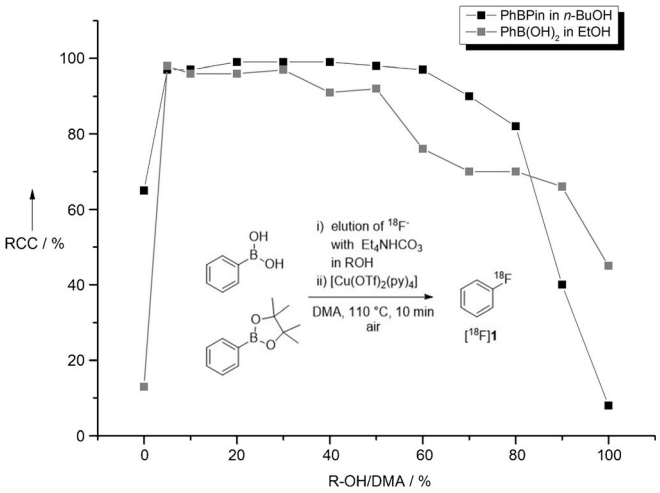

Figure 3. Dependency of RCC on alcohol content.[10].

To demonstrate the versatility of the procedure, we radiofluorinated a series of (hetero)arylboronic substrates (Figure 5). Different electron-rich, -deficient, and -neutral ^{18}F-labeled arenes were prepared in RCCs exceeding or averaging 80% (Figure 4). The introduction of *O*- and *N*-containing substituents into the *ortho*-position ([^{18}F]**4**, [^{18}F]**10**) resulted in lower RCCs, presumably due to unfavorable interactions with the leaving groups, impeding transmetalation. Remarkably, radiofluorinated phenols ([^{18}F]**8**, [^{18}F]**9**) could be directly prepared from the corresponding unprotected boronic substrates in 80–97% RCCs. For comparison, without alcohol, RCCs of only 7–15% were achieved.[3b,d,e] Furthermore, *n*BuOH afforded 2- and 3-[^{18}F]fluoroanilines ([^{18}F]**10**, [^{18}F]**11**) from the corresponding unprotected precursors, albeit in lower RCCs of up to 10 and 21%, respectively.[12] In the presence of *n*BuOH, highly electron rich unprotected 4-, 5-, and 6-indole boronic acids and pinacol boronates ([^{18}F]**12**–**14**) underwent near-quantitative (>96%) radiofluorination. Radiolabeling of indoles is of significant interest given their importance as cycloxygenase-2, cannabinoid receptor type 2, β-adrenoreceptor, and 5-hydroxytryptamine receptor type 3 ligands.[12] The metabolism of tryptophan, a proteinogenic amino acid with an indole ring in the side chain, is significantly altered in numerous neurological disorders and tumors.[13] The applicability of our method to prepare radiofluorinated tryptophans was confirmed by the preparation of protected radiolabeled 4- and 6-fluorotryptophans ([^{18}F]**15**, [^{18}F]**16**) in RCCs of 92 and 99%, respectively.[14]

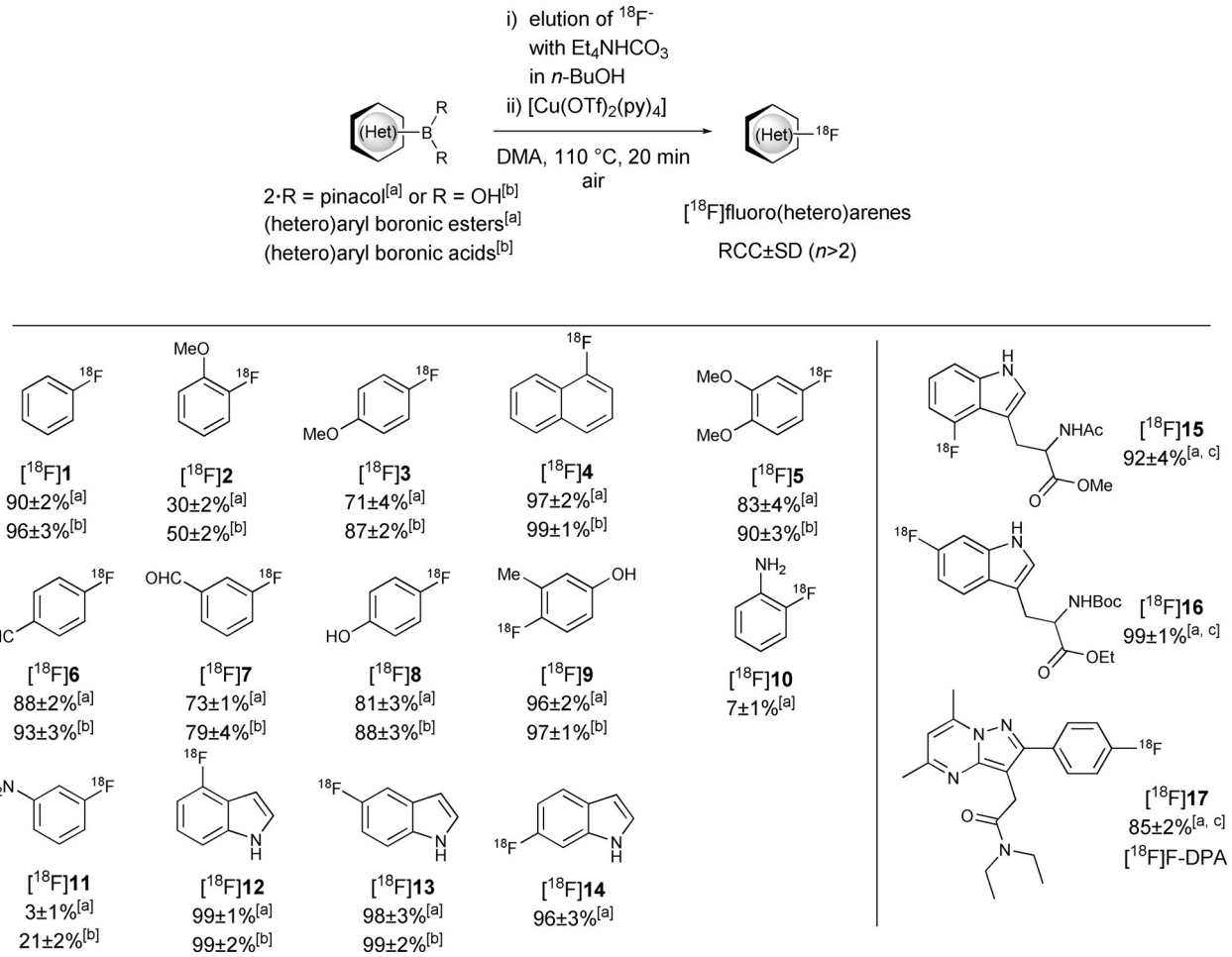

Figure 4. Substrate scope for the *n*BuOH-enhanced copper-mediated radiofluorination of pinacol boronates and boronic acids.[c]Et_4NHCO_3 was used for elution of $^{18}F^-$.[10] RCC ± SD [%].

The method was also used to prepare clinically relevant PET tracers. F-DPA is a very potent agonist at the 18 kDa mitochondrial translocator protein (TSPO), a biomarker of brain inflammation and reactive gliosis, characteristic of some neurodegenerative and psychiatric diseases, stroke and brain tumors.[15] To date, [^{18}F]F-DPA (**17**) is accessible only via electrophilic radiofluorination.[16] Preparation of this probe via conventional nucleophilic ^{18}F-fluorination of the respective iodonium, trimethylammonium, and nitro precursors resulted in a maximum ^{18}F-incorporation of 3%.[17] In contrast, using *n*BuOH-enhanced Cu-mediated fluorination, [^{18}F]F-DPA was obtained in 85% RCC. Afterwards, we produced 6-[^{18}F]fluoro-3,4-dihydroxyphenethylamine (6-[^{18}F]FDA) and 6-[^{18}F]fluoro-L-3,4-dihydroxyphenylalanine (6-[^{18}F]FDOPA). 6-[^{18}F]FDA is widely used to image chromaffin tumors.[18] 6-[^{18}F]FDOPA is the most popular PET tracer for visualizing the central dopaminergic system and is also used for diagnosis of neuroendocrine tumors.[19] For *N*,*N*,*O*,*O*-Tetra-Boc-protected substrates **18b** and **21b**, ^{18}F-incorporation amounted to 87 and 68%, respectively. During ^{18}F-incorporation, almost complete (>80%) cleavage of one of the *N*-Boc protection groups and partial cleavage of one of the *O*-Boc groups (overall >35%) occurred. Surprisingly, mono-*N*-Boc-protected precursors also yielded the desired radiolabeled products in reasonable RCCs (78 and 30% for [^{18}F]**19** and [^{18}F]**22**, respectively) (Figure 5). If radiolabeling of mono-*N*-protected 6-[^{18}F]FDOPA and 6-[^{18}F]fluoro-*m*-tyrosine (6-[^{18}F]FMT) precursors was carried out in pure aprotic solvents only ≤5% of the radiofluorinated amino acids were observed in the reaction mixture, possibly due to concurrent intramolecular Chan–Lam coupling, that is formation of the respective indolines instead of fluorination.[3d,e] In alcoholic media, this side reaction was efficiently suppressed. During radiofluorination of **18a** and **18b** we observed the formation of two identical radioactive products. Based on the retention time, the major product was identified as *N*,*O*,*O*-tri-Boc-protected dopamine [^{18}F]**19a**, indicating that a partial deprotection took place. In the case of the radiolabeling of **18b**, traces (<1%) of a more hydrophobic product, presumably, *N*,*N*,*O*,*O*-tetra-Boc-protected intermediate [^{18}F]**19** were also detected. We assumed that the minor product peak in HPLC chromatogram should correspond to partially deprotected intermediates (*N*,3-*O*-diBoc and *N*,4-*O*-di-Boc-protected dopamines). After subsequent treatment with HCl only [^{18}F]fluoride and 6-[^{18}F]FDA could be detected in the crude reaction mixture strongly supporting our assumption. Similarly, during ^{18}F-labeling of **21a** and **21b** the formation of the two identical radiolabeled products was observed. Additionally, in the case of the radiolabeling of **21b**, the more hydrophobic product, presumably, Boc$_2$-6-[^{18}F]FDOPA(Boc)$_2$-O*t*Bu ([^{18}F]**22**) (12%) was detected. Also in this case $^{18}F^-$ and 6-[^{18}F]FDOPA were the only radioactive products observed in the reaction mixture after the deprotection step. After the radiolabeling step, remaining pinacol boronate precursors were transformed into the corresponding trifluoroboronates. The latter were separated from the radiolabeled intermediates [^{18}F]**19** and [^{18}F]**22** by solid-phase extraction. Finally, hydrolysis of [^{18}F]**19** and [^{18}F]**22** with 12 M HCl at 110 °C for 5 min, followed by HPLC purification, afforded 6-[^{18}F]FDA and enantiomerically pure 6-[^{18}F]FDOPA in a ready-to-use form. The unoptimized radiochemical yields were 35 and 40% over the two steps for 6-[^{18}F]FDA and 6-[^{18}F]FDOPA, respectively. The specific activities amounted to 39 and 37 GBq μmol^{-} for 6-[^{18}F]FDA (260 MBq) and 6-[^{18}F]FDOPA (278 MBq), respectively The amounts of Cu determined in the final solutions by ICP-MS amounted to 6.54±0.08 and 5.92±0.03 μg/preparation for 6-[^{18}F]FDA and 6-[^{18}F]FDOPA, respectively.[20]

Trialkylstannanes are popular substrates for electrophilic radiofluorination with [^{18}F]F_2 and the preparation of radioiodinated probes. Some of them are commercially available, for example, precursors for 6-[^{18}F]FDOPA, 6-[^{18}F]FMT, 2-[^{18}F]fluoro-L-tyrosine (2-[^{18}F]FTyr), and 5-[I*]iodo-2′-desoxyuridine (5-[*I]IdU).

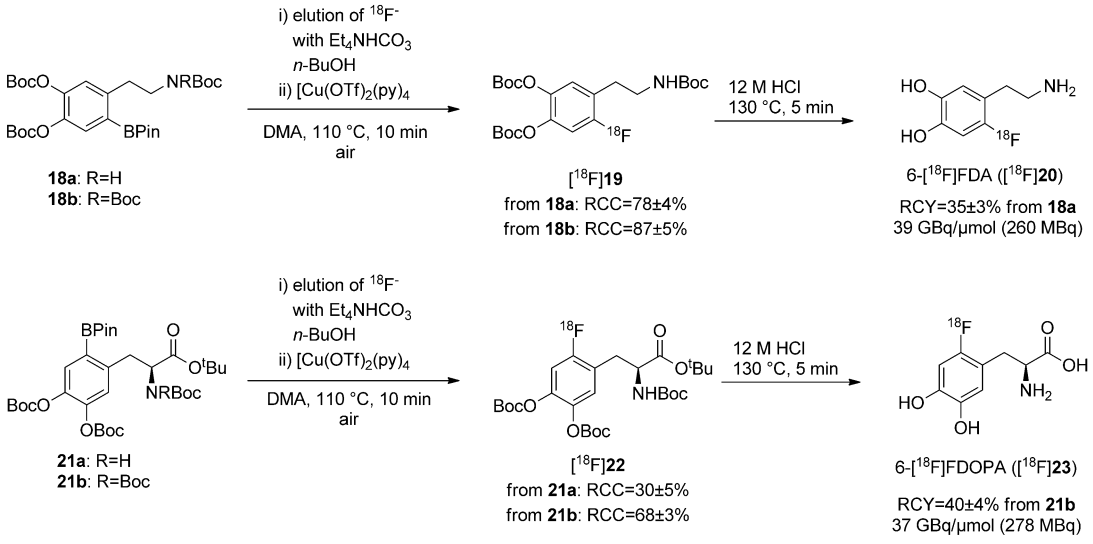

Figure 5. Preparation of 6-[^{18}F]FDA ([^{18}F]**20**) and 6-[^{18}F]FDOPA ([^{18}F]**23**).[10] [a] During radiolabeling of **18b** and **21b**, concurrent cleavage (>80%) of one of the *N*-Boc protection groups and partial cleavage of one of the *O*-Boc groups (overall >35%) occurred. Partial deprotection of [^{18}F]**19a** and [^{18}F]**22a** was also observed.

Quite recently, Mossine et al. reported the Cu-mediated [^{18}F]fluorination of these substrates with nucleophilic [^{18}F]fluoride.[21] We examined, whether trialkylstannanes could be radiolabeled using the novel ^{18}F-fluorination procedure. $PhSnMe_3$ (**24**) was efficiently radiolabeled to yield [^{18}F]FPh in 66% RCC (Figure 6). Moreover, [^{18}F]DAA1106 ([^{18}F] **26**), a further

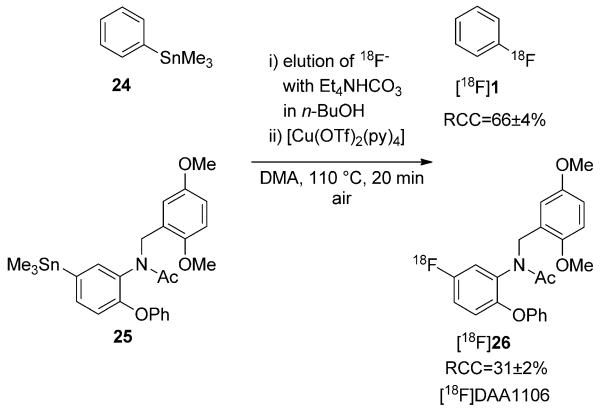

Figure 6. Radiofluorination of $SnMe_3$-substrates.[10].

promising PET probe for the visualization of translocator protein (TSPO) expression,[5b] was also prepared from the corresponding stannyl precursor **25** in an unoptimized RCC of 31%.[22] Finally, we briefly tested the applicability of the method for conventional fluorination. Tetramethylammonium fluoride as a fluoride source afforded 12% HPLC yield of 4-fluoroindole from indole-4-boronic acid under the same reaction conditions applied for ^{18}F-labeling.[23]

We have demonstrated the efficient application of alcohols as co-solvents for Cu-mediated ^{18}F-fluorination. Based on this finding, we developed a procedure for rapid radiolabeling of a broad range of substrates such as (hetero)arylboronic acids, pinacol boronates, and trialkylstannanes under general reaction conditions in many cases in yields of 80–99%. This procedure substantially simplifies the production of [^{18}F]fluorinated arenes by removing time-consuming azeotropic drying steps. Furthermore, it allows "last-stage" access to ^{18}F-fluorinated indoles, phenols, and anilines from unprotected precursors. The preparation of clinical doses of two PET-tracers, 6-[^{18}F]FDA and 6-[^{18}F]FDOPA has demonstrated the practicality of the method. The suitability of the protocol for commercially available synthesis modules is currently under investigation.

Acknowledgements

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Keywords: copper · fluorination · imaging agents · positron emission tomography · radiopharmaceuticals

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[6] See Reference [3b], Supporting Information Figures S9 and S10.

[7] Please, see Supporting Information for the complete details of optimisation studies.

[8] See Supporting Information, Table 1.

[9] Radiochemical conversion (RCC; ^{18}F-incorporation) refer to the amount of radiofluoride which is transformed to the desired ^{18}F-labeled compound, determined by radio-HPLC or TLC. Radiochemical yield (RCY) refers to the isolated yield of the radiochemically and chemically pure radiolabeled compound. All RCYs are corrected for decay.

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[14] We used the novel method for the preparation of 4-, 5-, 6- and 7-[^{18}F]fluorotryptophans (RCCs 90–99%). These probes were prepared as ready-to-use solutions in 40–50% isolated yields and with specific activities of 40–60 GBq μmol^{-} (for 1–2.5 GBq of tracer). Their biological evaluation is under evaluation and will be reported together with details of their preparation in due course. For recently published synthesis of 6-[^{18}F]fluorotryptophan, see: D. Schäfer, P. Weiß, J. Ermert, J. Castillo Meleán, F. Zarrad, B. Neumaier, *Eur. J. Org. Chem.* **2016**, 4621–4628.

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[22] In preliminary experiments 6-[^{18}F]FDOPA and 3-OMe-6-[^{18}F]FDOPA were prepared in 20–25 % RCYs starting from the corresponding tin precursors.

[23] No further attempts have been made to optimize this reaction.

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Supporting Information

Alcohol-Enhanced Cu-Mediated Radiofluorination

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Supporting Information

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Collapse of the QMA phase

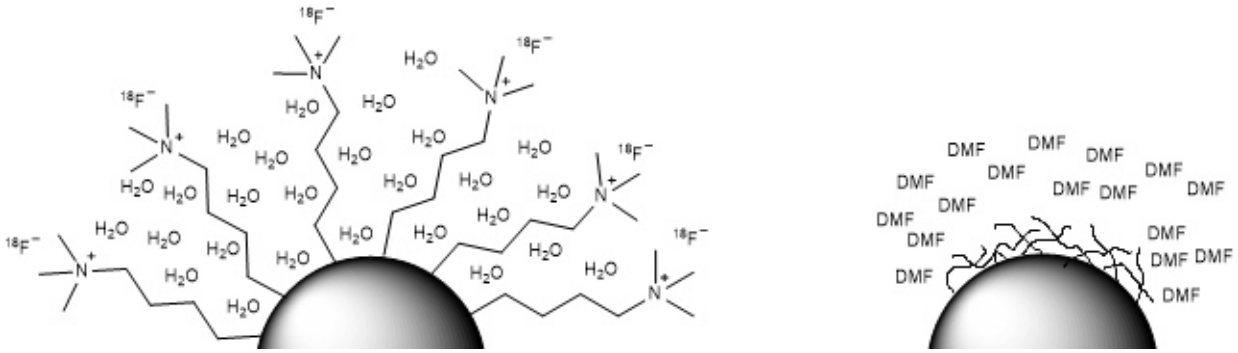


Figure S1. Assumed collapse of the anion exchange resin similar to the well-known collapse of the hydrophobic alkyl phases in highly aqueous media, observed in reversed phase HPLC.[1] Shown are the configuration of the Et_3N^+-derivatised side chains in aqueous solutions (A) and in 100% DMF (B).

Materials and Methods

General

^{1}H-NMR spectra: Bruker Avance II 300 (300 MHz), Bruker Avance (600 MHz) or Varian Inova (400 MHz) spectrometer using $CDCl_3$ as solvent. All shifts are given in ppm using the solvent residual signals as reference. ^{1}H chemical shifts are reported in ppm relative to residual peaks of deuterated solvents. Higher-order NMR spectra were approximately interpreted as first-order spectra, if possible. The observed signal multiplicities are characterized as follows: s = singlet, d = doublet, t = triplet, q = quartet, quin = quintet, m = multiplet, as well as br = broad. Coupling constants (*J*) were reported in hertz (Hz). ^{13}C-

NMR spectra [additional APT (Attached ProtonTest)]: Bruker Avance II 300 (75.5 MHz), Varian Inova (101 MHz) and Bruker Avance II 600 (125.9 MHz). ^{13}C chemical shifts are reported relative to residual peaks of deuterated solvents. Low resolution ESI-MS were measured on a Finnigan LCQ. High resolution ESI-MS spectra were obtained on a FTICR LTQ FT Ultra. Elemental analyses were obtained on a Vario EL cube. TLC: Merck precoated sheets, 0.25 mm Sil G/UV_{254}. The chromatograms were viewed under UV light and/or by treatment with phosphomolybdic acid (10% in ethanol). Column chromatography: Merck silica gel, grade 60, 230–400 mesh or Merck LiChroprep RP-C18, 25–40 μm. Automated flash chromatography was carried out using a Reveleris Flash Chromatography system on commercially available Flash Grace Reveleris™ cartridges (Grace, USA). Solvent proportions are indicated in a volume:volume ratio. All reactions were carried out with magnetic stirring, if not stated otherwise, and, if air or moisture sensitive, substrates and/or reagents were handled in flame-dried glassware under argon or nitrogen. Organic extracts were dried with anhydrous $MgSO_4$.

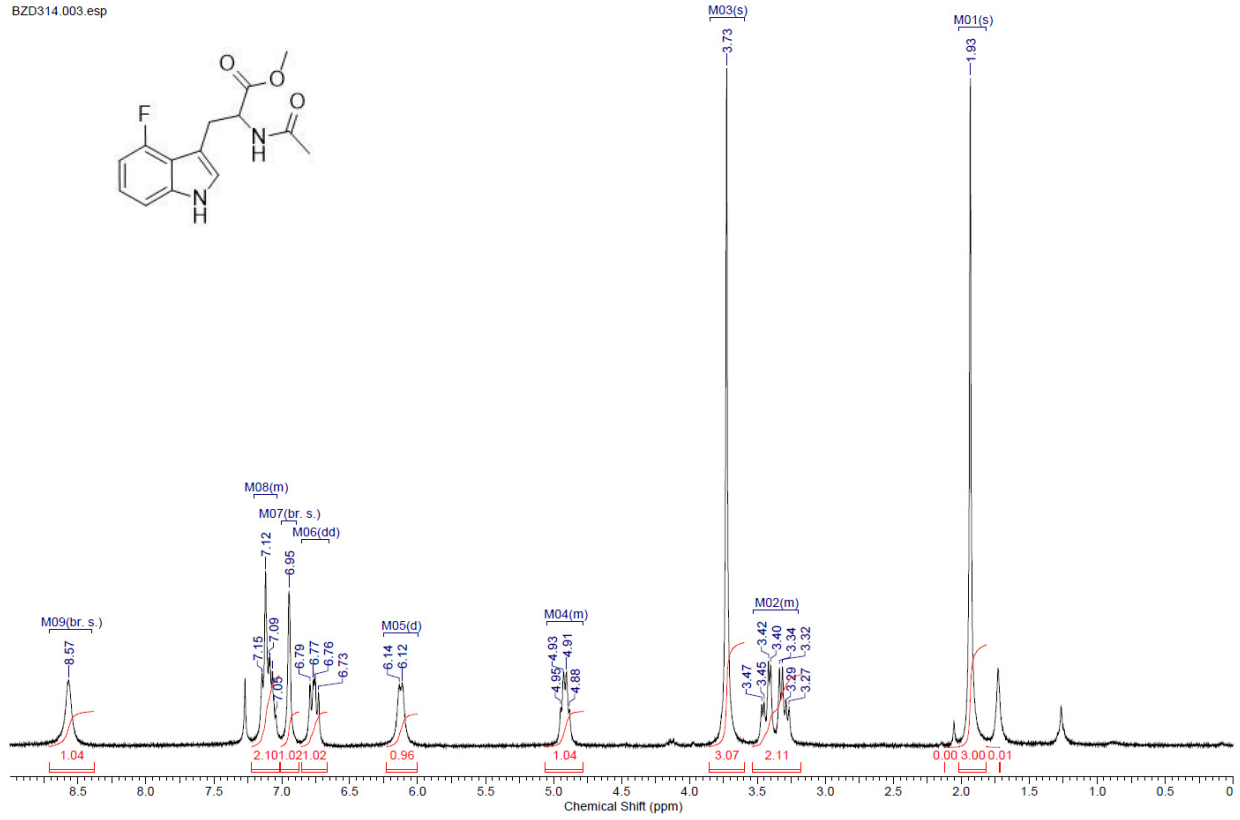
Before use $B_2(Pin)_2$ was purified by the crystallization from pentane and dried overnight at 40 °C and 15 mbar. KOAc was dried overnight at 300 °C.

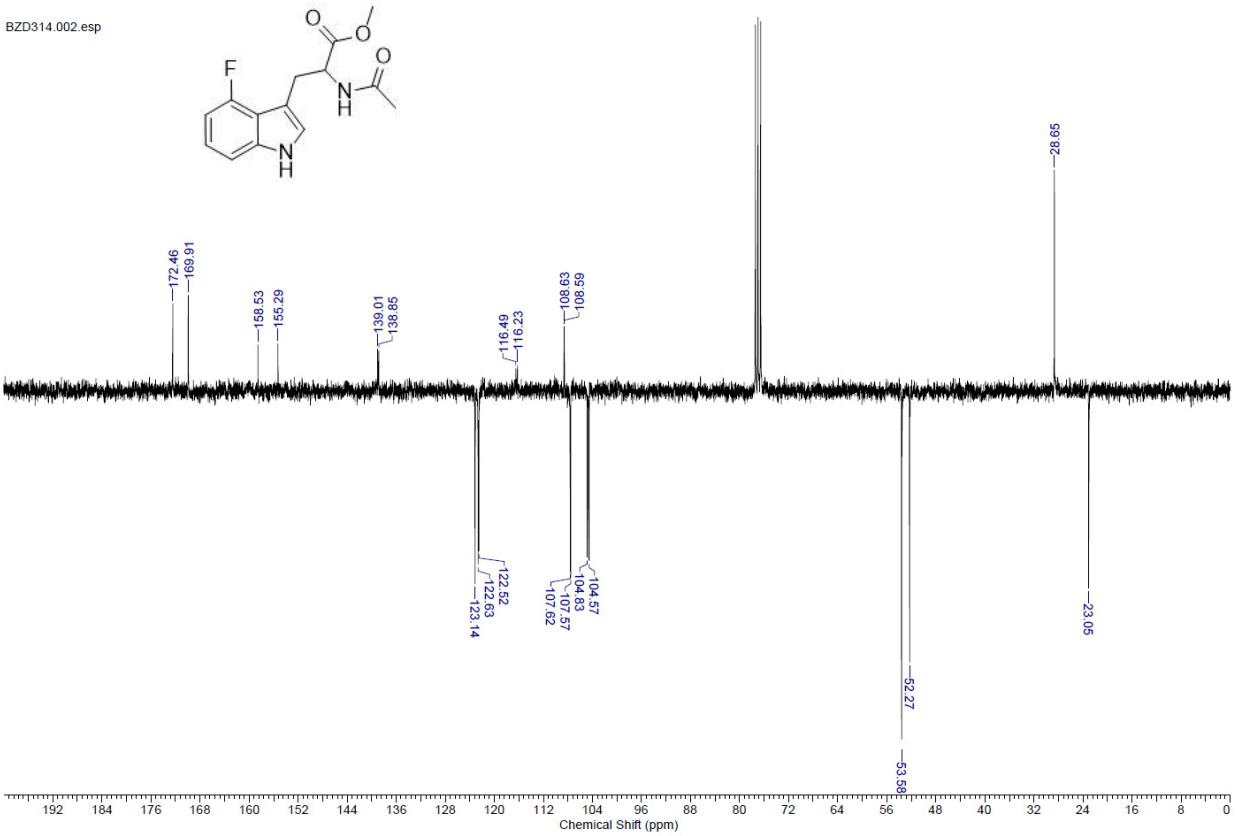
Chemistry

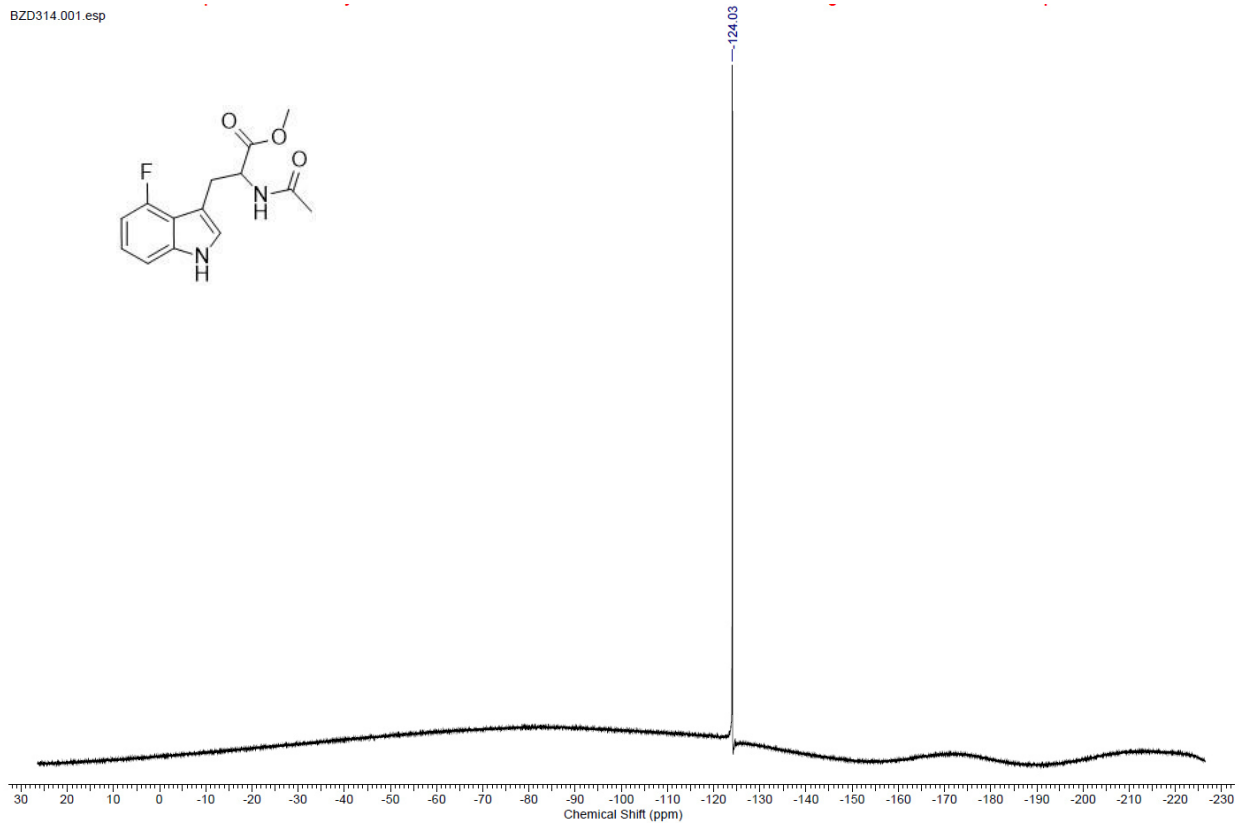
N-Acetyl-4-[4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-tryptophan methyl ester,[2] ethyl 2-(diphenylmethyleneamino)acrylate,[3] ethyl *N*-diphenylmethylene-3-trimethylammoniumalaninate iodide,[3] *N*,*N*-diethyl-2-(2-(4-iodophenyl)-5,7-dimethylpyrazolo[1,5-a]-pyrimidin-3-yl)acetamide,[4] *N*,*N*-diethyl-2-(2-(4-fluorophenyl)-5,7-dimethylpyrazolo[1,5-a]-pyrimidin-3-yl)acetamide,[4] N,O,O-tri-Boc-6-fluorodopamine[5] and 5-fluorodopamine[5] were prepared according to literature. According to HPLC analysis the purity of *N*,*N*-diethyl-2-(2-(4-fluorophenyl)-5,7-dimethylpyrazolo[1,5-a]-pyrimidin-3-yl)acetamide (**17**) amounted to 93%. No attempts to further purify this compound were made.

4,4,5,5-Tetramethyl-2-(2-methyl-4-hydroxyphenyl)-1,3,2-dioxaborolane:[6] Pinacol (1.23 g, 10.40 mmol) was added to a suspension of 4 Å molecular sieves (3 g) in a solution of 3-hydroxy-6-methylphenylboronic acid (1.5 g, 9.87 mmol) in anhydrous Et_2O (80mL) and the reaction mixture was stirred for 4 days. Afterwards, the mixture was filtered and concentrated under reduced pressure. The crude product was purified by column chromatography (EtOAc:hexane = 1:2.5) affording the title compound (2.11 g, 90%) as a colorless solid. R_f = 0.38, EtOAc:hexane = 1:2.5. The spectral data were in accordance with those reported in the literature.[6]

***N*-Acetyl 4-fluorotryptophan methyl ester (15):** 1 M $EtAlCl_2$ in hexane (7 mL) was added to an ice-cold solution of *N*-acetyl dehydroalanine methyl ester (0.5 g, 3.49 µmol) and 4-fluoroindole (0.71 g, 5.24 µmol) in anhydrous CH_2Cl_2 (25 mL). The cooling bath was removed and the reaction mixture was stirred for 3 hours. Afterwards, a saturated solution of $NaHCO_3$ (10 mL) and Et_2O (80 mL) were added. The organic layer was separated from the aqueous suspension, washed with brine (2×10 mL), dried and concentrated under reduced pressure. The residue was purified by column chromatography (EtOAc) to give the title compound (0.76 g, 78%) as a colorless solid. R_f = 0.26, EtOAc. ^{1}H NMR (300 MHz) δ 1.93 (s, 3 H), 3.37 (m, 2 H), 3.73 (s, 3 H), 4.92 (m, 1 H), 6.13 (d, *J*=6.25 Hz, 1 H), 6.76 (dd, *J*=11.03, 8.11 Hz, 1 H), 6.95 (s, 1 H), 7.12 (m, 2 H), 8.57 (br, 1 H). ^{19}F NMR (71.8 MHz) δ 124.03. ^{13}C NMR (75.5 MHz) δ 172.5, 169.9, 156.9 (d, *J*=245 Hz), 138.9 (d, *J*=12.1 Hz), 123.1, 122.6 (d, *J*=8.3 Hz), 116.4 (d, *J*=19.6 Hz), 108.6 (d, *J*=3 Hz), 107.6 (d, *J*=3.8 Hz), 104.7 (d, *J*=19.6 Hz), 53.6, 52.3, 28.7, 23.1. MS (ESI): positive mode m/z = 301.2 ($[M + Na]^+$); ESI HRMS: calcd for $C_{14}H_{15}O_3N_2FNa^+$: 301.0959; found: 301.0958.

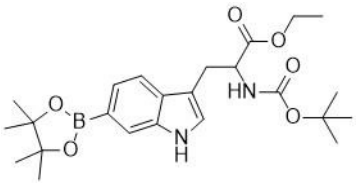






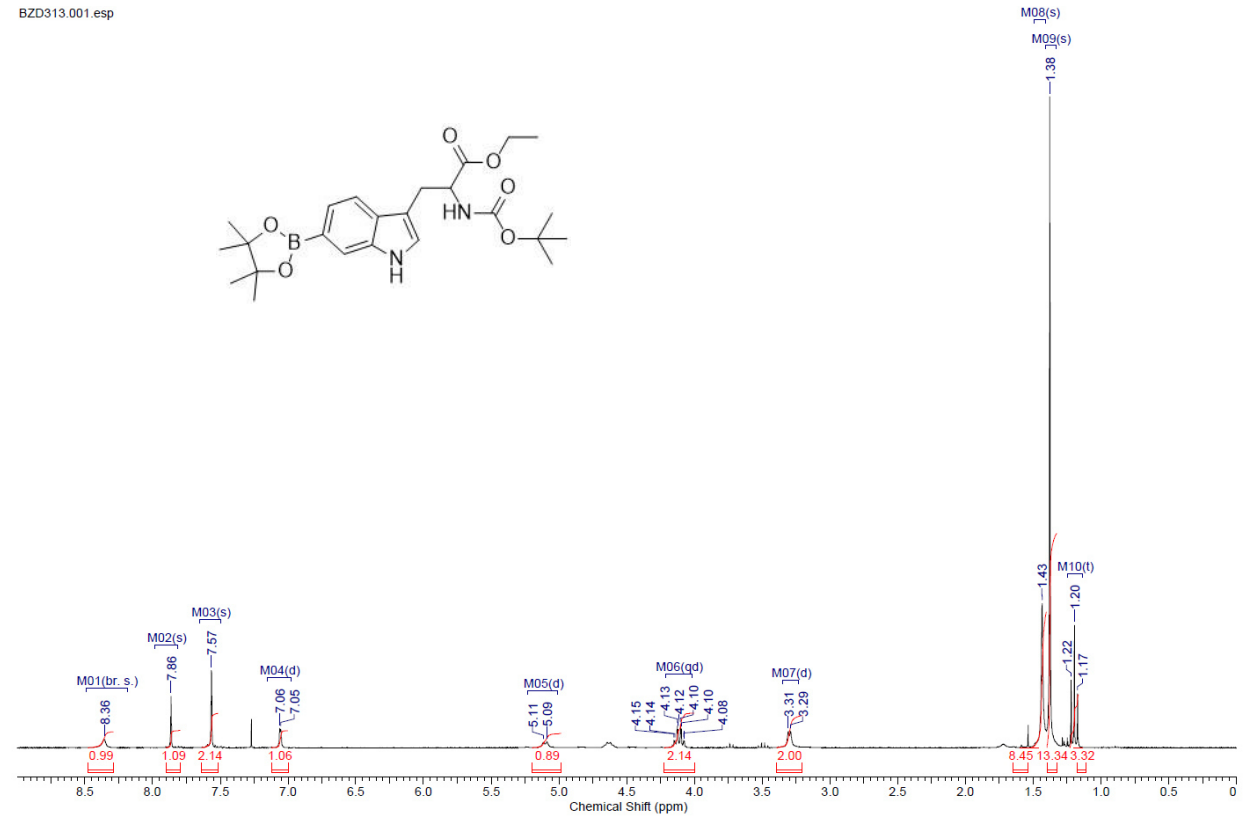
***N*-Boc-6-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)tryptophan ethyl ester:** 1 M $EtAlCl_2$ (5.7 mL) in hexane (7 mL) was added to an ice-cold solution of the freshly prepared ethyl 2-(diphenylmethyleneamino)acrylate[3] (0.8 g, 2.86 µmol) and 6-(4,4,5,5-tetramethyl-1,3,2-

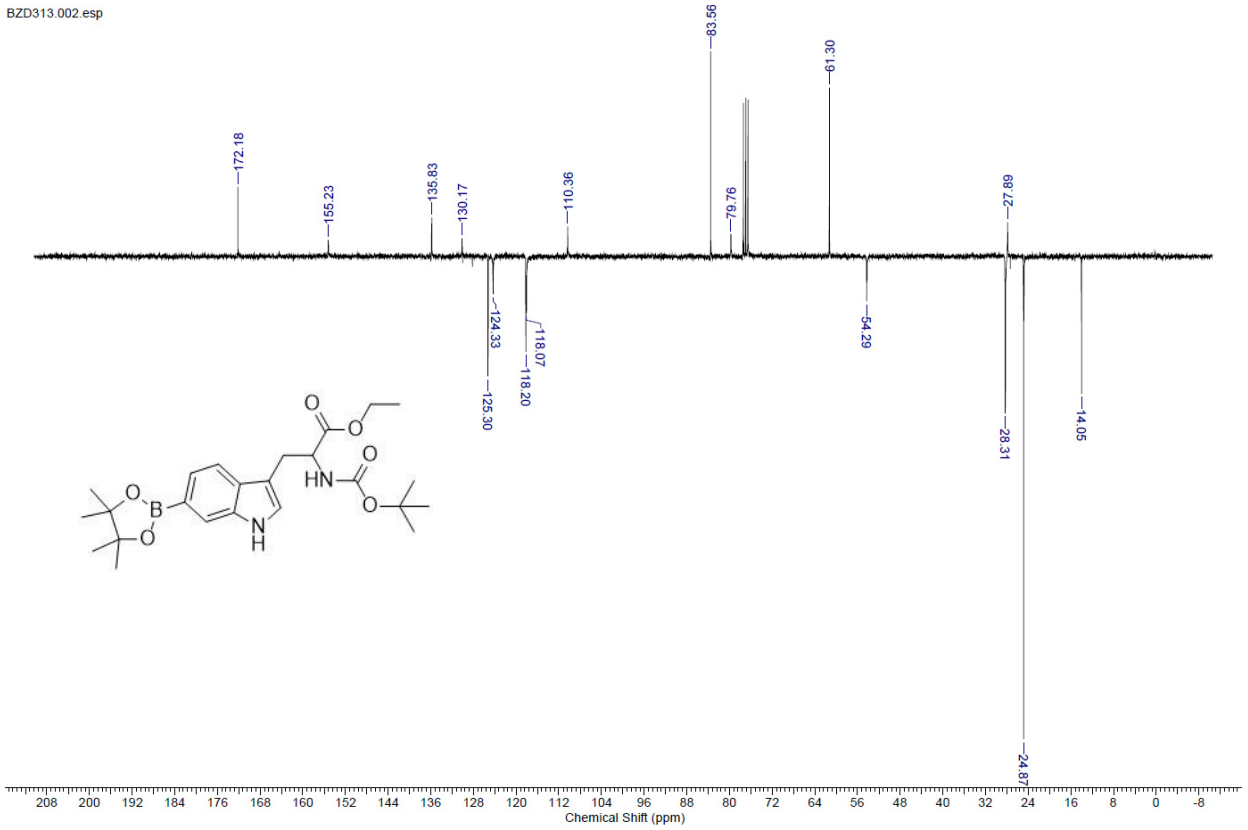
dioxaborolan-2-yl)-indole (1.044 g, 4.30 μmol) in anhydrous CH_2Cl_2 (10 mL) and the mixture was



stirred for 2 h. The cooling bath was removed and the reaction mixture was stirred for further 3 hours. Afterwards, a saturated solution of $NaHCO_3$ (10 mL) and Et_2O (80 mL) were added. The ethereal layer was separated from the aqueous suspension, washed with brine (2×10 mL), dried and concentrated under reduced pressure. The residue was purified by column chromatography (EtOAc:hexane=1:1.5; silica with 0.1% CaO) affording ethyl 2-(diphenylmethyleneamino)-3-[6-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-1*H*-indol-3-yl]propanoate (0.98 g, 65%) as a brown foam which was used for the next step without further purification. R_f = 0.55, EtOAc:hexane = 1:1.5.

1 M HCl (2.25 mL) was added to an ice-cold solution of ethyl 2-(diphenylmethyleneamino)-3-(6-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-1H-indol-3-yl)propanoate (0.98 g, 1.875 mmol) in THF (8 mL) and the bright yellow reaction mixture was incubated at –25 °C for 16 h. Thereafter, $NaHCO_3$ (0.32 g, 3.75 mmol) was added under vigorous stirring and the mixture was allowed to warm to ambient temperature. Boc_2O (0.944 g, 4.33 mmol) was added followed by the addition of EtOH and H_2O until a homogeneous solution was obtained. The reaction mixture was stirred for a further 3 h and partially concentrated under reduced pressure. The residue was taken up into EtOAc and H_2O (60 mL of each), the organic layer was separated and washed with H_2O (3×20 mL), brine (2×20 mL), dried and concentrated under reduced pressure. The oily residue was triturated first with pentane (3×20 mL), then with hexane (3×20 mL), and, finally, with Et_2O (30 mL) to give the title compound (0.55 g, 42% over three steps) as a colorless solid. R_f = 0.5, EtOAc:hexane = 1:1.5. ^{1}H NMR (300 MHz) δ 1.20 (t, *J*=7.15 Hz, 3 H), 1.38 (s, 4×3 H), 1.43 (s, 9 H), 3.30 (d, *J*=5.14 Hz, 2 H), 4.11 (qd, *J*=7.15, 1.71 Hz, 2 H), 5.10 (d, *J*=7.96 Hz, 1 H), 7.06 (d, *J*=2.22 Hz, 1 H), 7.57 (s, 2 H), 7.86 (s, 1 H), 8.36 (br, 1 H). ^{13}C NMR (75.5 MHz) δ 172.2, 155.2, 135.8, 130.2, 125.3, 124.3, 118.2, 118.1, 110.4, 83. 6, 79.8, 61.3, 54.3, 28.3, 27.9, 24.9, 14.1. C–B was not observed. MS (ESI): positive mode m/z = 481.3 ($[M + Na]^+$); ESI HRMS: calcd for $C_{24}H_{35}O_6N_2BNa^+$: 481.2480; found: 481.2479. Correct isotopic pattern.



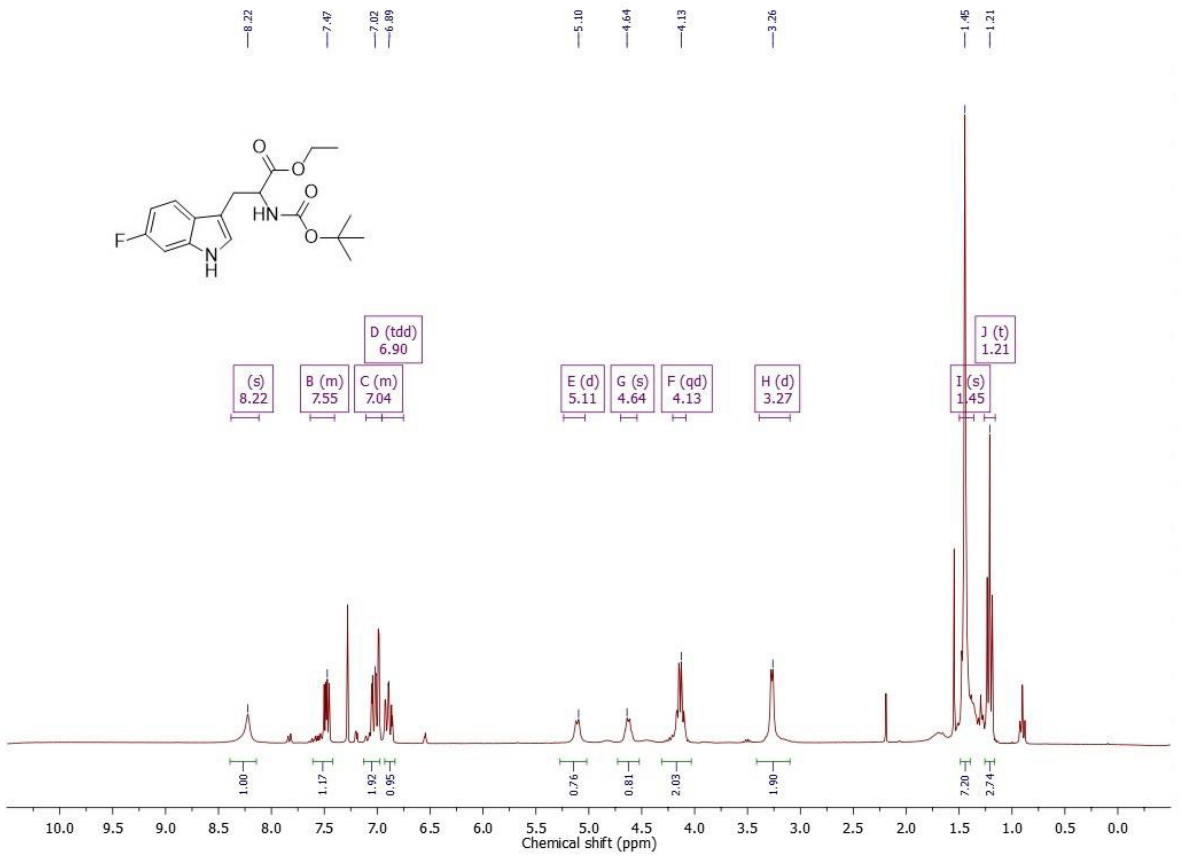


N-Boc-6-Fluorotryptophan ethyl ester (16): Freshly dried K_2CO_3 (0.27 g, 1.95 mmol) was added to a solution of ethyl *N*-diphenylmethylene-3-trimethylammoniumalaninate iodide[2] (0.45, 0.96 mmol) in anhydrous EtOH (10 mL) and the reaction mixture was stirred for 20 h. Pentane (30 mL) was added, the mixture was filtered and concentrated under reduced

pressure at 35 °C. The residue was taken up into pentane (30 mL), the precipitate was filtered off and the filtrate was concentrated under reduced pressure at 35 °C affording the crude ethyl 2-(diphenylmethyleneamino)acrylate[3] (0.23 g) as a yellow oil which was immediately used for the next step. R_f =0.4, EtOAc:hexane=1:5.

1 M $EtAlCl_2$ in hexane (1.7 mL) was added to an ice-cold solution of the crude ethyl 2-(diphenylmethyleneamino)acrylate[3] (0.23 g, max. 0.82 mmol) and 6-fluoroindole (0.17 g, 1.26 mmol) in anhydrous CH_2Cl_2 (10 mL) and the reaction mixture was stirred for 1 h. The cooling bath was removed and the mixture was stirred for a further 1 h. Afterwards, saturated $NaHCO_3$ (10 mL) and Et_2O (50 mL) were added, the ethereal layer was separated from the aqueous suspension, washed with saturated $NaHCO_3$ (10 mL), brine (2×10 mL), dried over Na_2SO_4 and concentrated under reduced pressure to give the mixture of *N*-diphenylmethylene-6-fluorotryptophan ethyl ester and 6-fluoroindole, which was directly used for the next step. *N*-Diphenylmethylene-6-fluorotryptophan ethyl ester: R_f =0.54, EtOAc:hexane=1:1.5; 6-fluorindole: R_f =0.7, EtOAc:hexane=1:1.5.

1 M HCl (1.2 mL) was added to a solution of the above mixture in Et_2O (10 mL) and the resulting biphasic mixture was vigorously stirred for 6 h. Thereafter, saturated $NaHCO_3$ (10 mL) followed by Boc_2O (0.53 g, 2.43 mmol) were added and the reaction mixture was stirred for further 3 h. The ethereal layer was separated, dried and concentrated under reduced pressure. The oily residue was sonicated with pentane (20 mL; three times), the resulting precipitate was filtered off and thoroughly washed with hexane:Et_2O=5:2 to give the title compound (0.2 g, 59% over four steps) as a colorless solid. According to HPLC analysis the purity of the title compound amounted to 90%. No attempts to further purify this compound were made. R_f =0.51, EtOAc:hexane=1:1.5. ^{1}H NMR (300 MHz) δ 1.21 (t, , 3H), 1.45 (s, 8H), 3.27 (d, *J*=4.9 Hz, 2H), 4.13 (qd, 2H), 4.64 (s, 1H), 5.11 (d, *J*=7.5 Hz, 1H), 6.90 (tdd, *J*=9.5, 6.3, 3.1 Hz, 1H), 7.11 (m, 2H), 7.63 (m, 1H), 8.22 (s, 1H). ^{13}C NMR (75,5 MHz) δ 172.20, 155.24, 132.43, 130.05, 128.27, 124.36, 122.93, 119.62, 119.48, 110.51, 79.87, 61.37, 54.21, 28.31, 28.03, 14.05. ^{19}F NMR (282 MHz) δ 121,18.


8.22
7.47
7.02
6.89
5.10
4.64
4.13
3.26
1.45
1.21
(s) 8.22
B (m) 7.55
C (m) 7.04
D (tdd) 6.90
E (d) 5.11
G (s) 4.64
F (qd) 4.13
H (d) 3.27
I (s) 1.45
J (t) 1.21
1.00
1.17
1.92
0.95
0.76
0.81
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1.90
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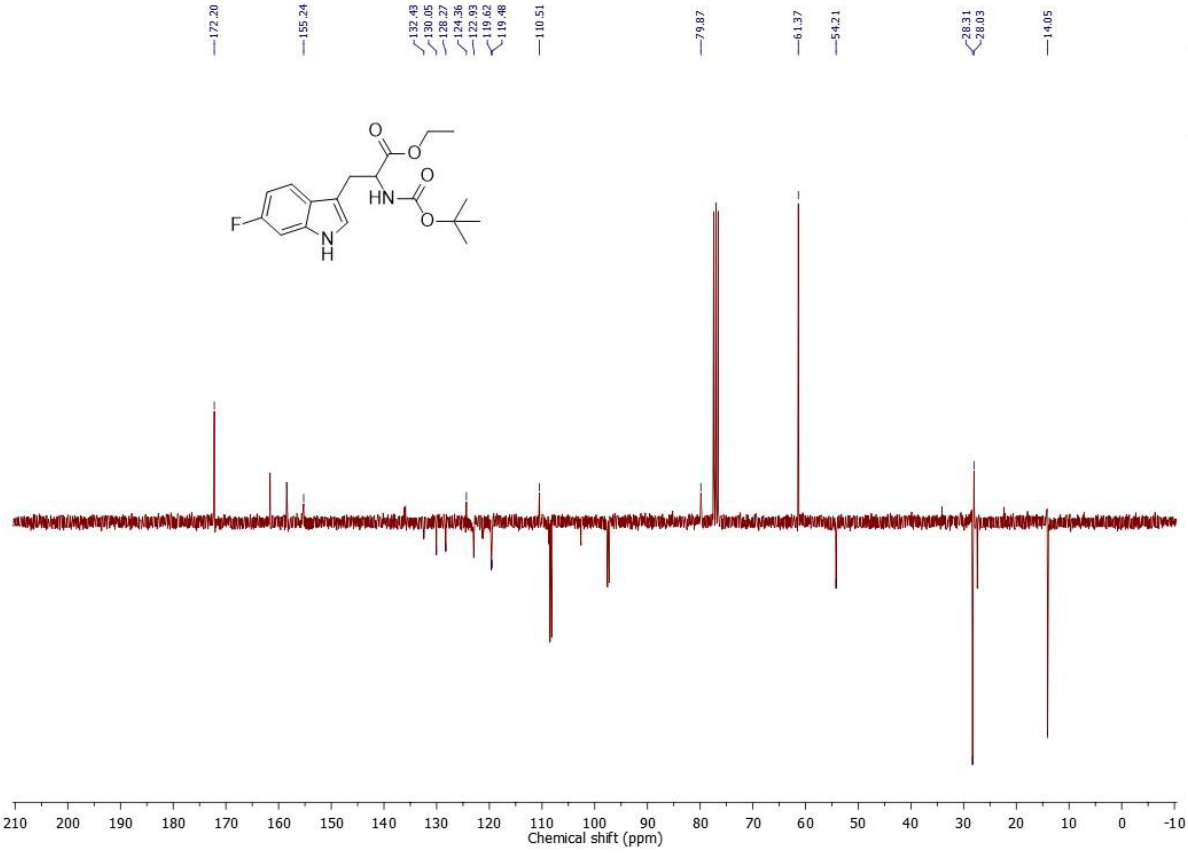
2.74
Chemical shift (ppm)


172.20
155.24
132.43
130.05
128.27
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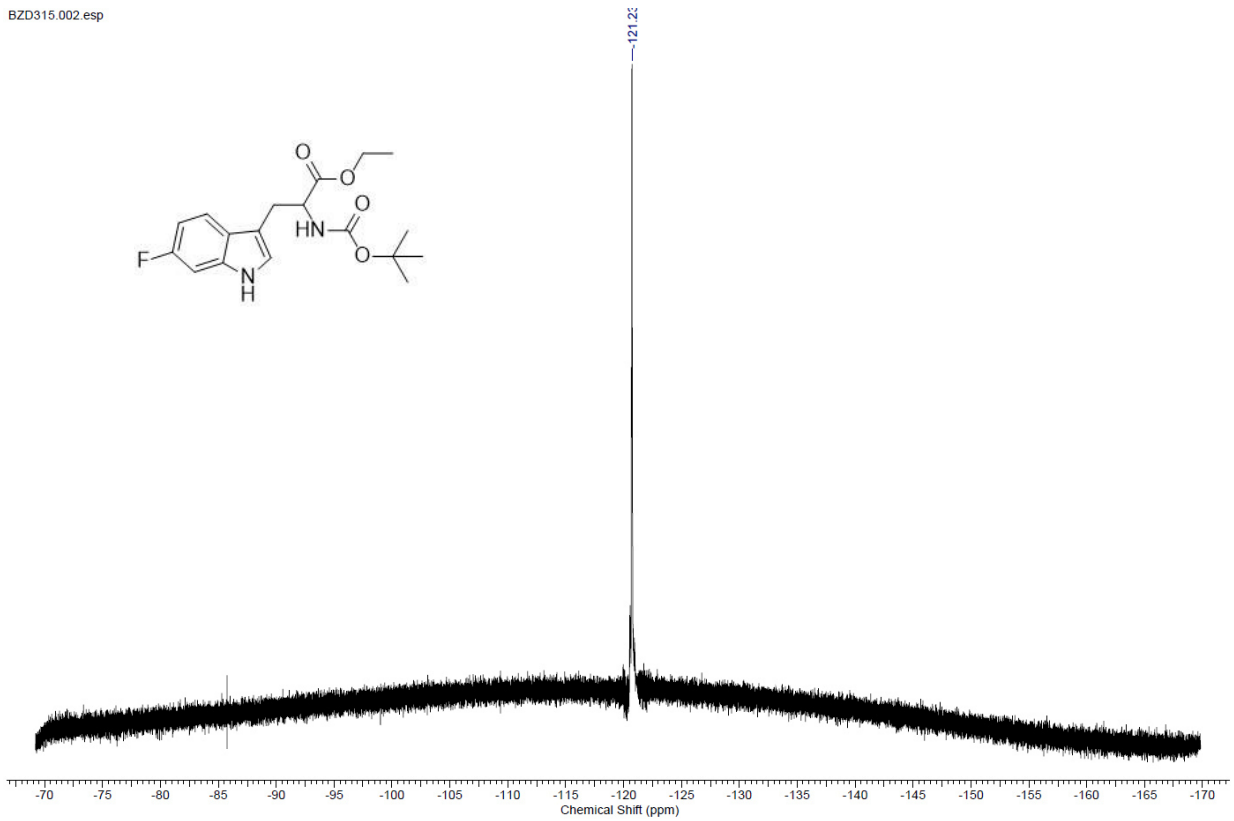
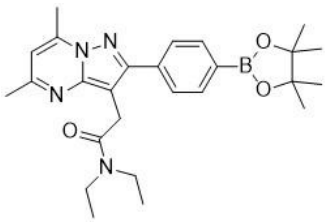
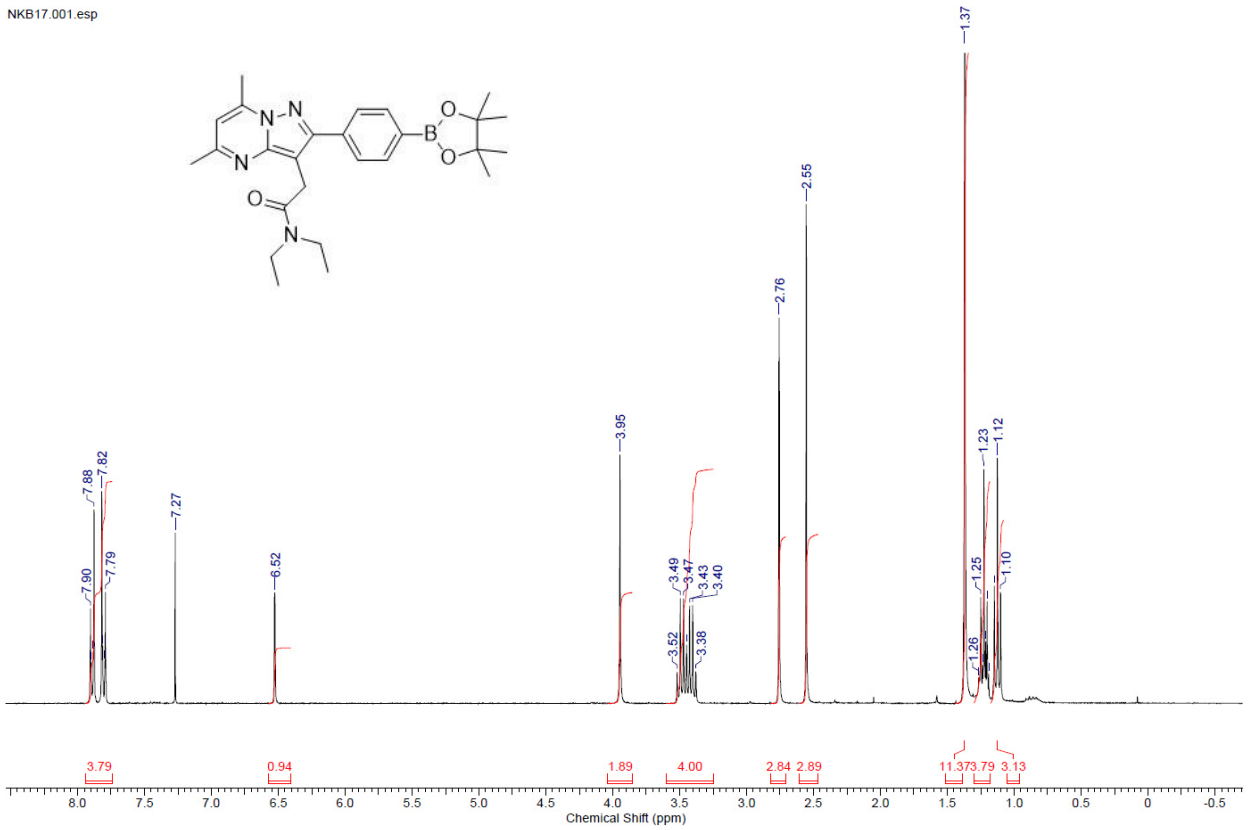
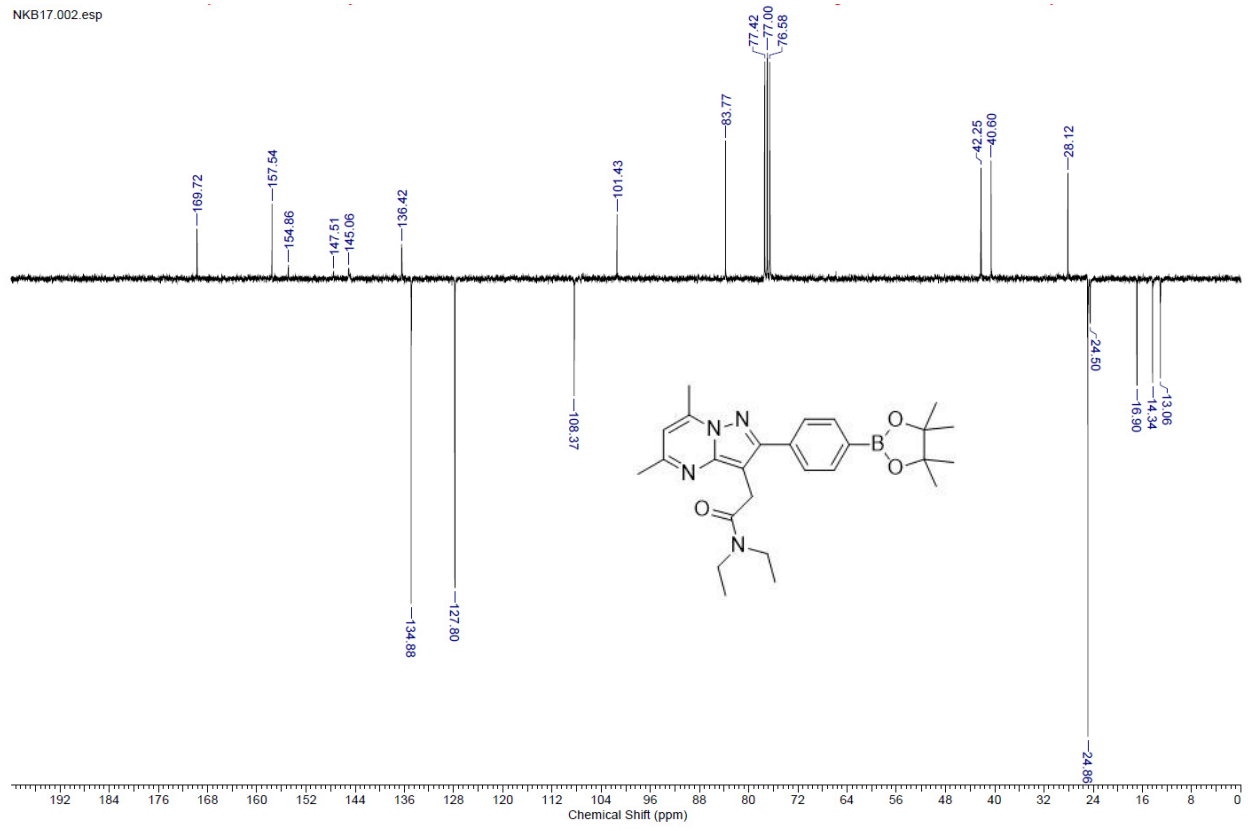
28.31
28.03
14.05
Chemical shift (ppm)



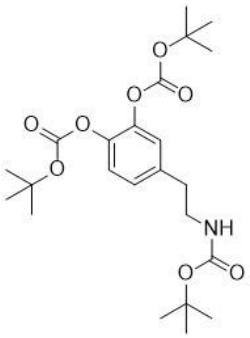
2-{5,7-Dimethyl-2-[4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl]pyrazol[1,5-a]pyrimidin-3-yl}-*N,N*-diethylacetamide: A solution of *N*,*N*-Diethyl-2-[2-(4-iodophenyl)-5,7-dimethylpyrazolo[1,5-a]-pyrimidin-3-yl]acetamide[4] (0.5 g, 1.1 mmol), $Pd_2(dba)_3$ (0.04 g, 0.04 mmol), XPhos (0.04 g, 0.09 mmol), $B_2(Pin)_2$ (0.33 g, 1.32 mmol) und KOAc (0.32 g, 3.3 mmol) was stirred for 24 h at 80 °C in anhydrous DMSO (7 mL). Afterwards, the reaction mixture was cooled to ambient temperature diluted with H_2O (6 mL) and extracted with EtOAc (20 mL). The EtOAc solution was washed with H_2O (20 mL), brine (20 mL), dried and concentrated under reduced pressure to yield the desired product (0.35 g, 70%) as a colorless solid. R_f = 0.42, $CHCl_3$/acetone 10:1. ^{1}H NMR (300 MHz) δ 1.14 (t, *J* = 7.08 Hz, 3 H), 1.24 (m, 3 H), 1.38 (s, 4×3 H), 2.57 (s, 3 H), 2.77 (d, *J* = 0.76 Hz), 3.47 (m, 4 H), 3.96 (s, 2 H), 6.54 (d, *J* = 0.76 Hz, 1 H), 7.86 (m, 4 H). ^{13}C NMR (75 MHz) δ 13.1, 14.4, 16.9 (×2), 24.9 (×4), 28.2, 40.6, 42.3, 83.8 (×2), 101.5, 108.4, 127.8 (×2), 134.9 (×2), 136.4, 157.6, 169.8. Signals from 4 aromatic quaternary carbons were not observed. ESI HRMS: calcd for $C_{26}H_{35}BO_3N_4Na^+$: 485.2694; found: 485.2694; calcd for $C_{26}H_{36}BO_3N_4^+$: 463.2875; found: 463.2876.



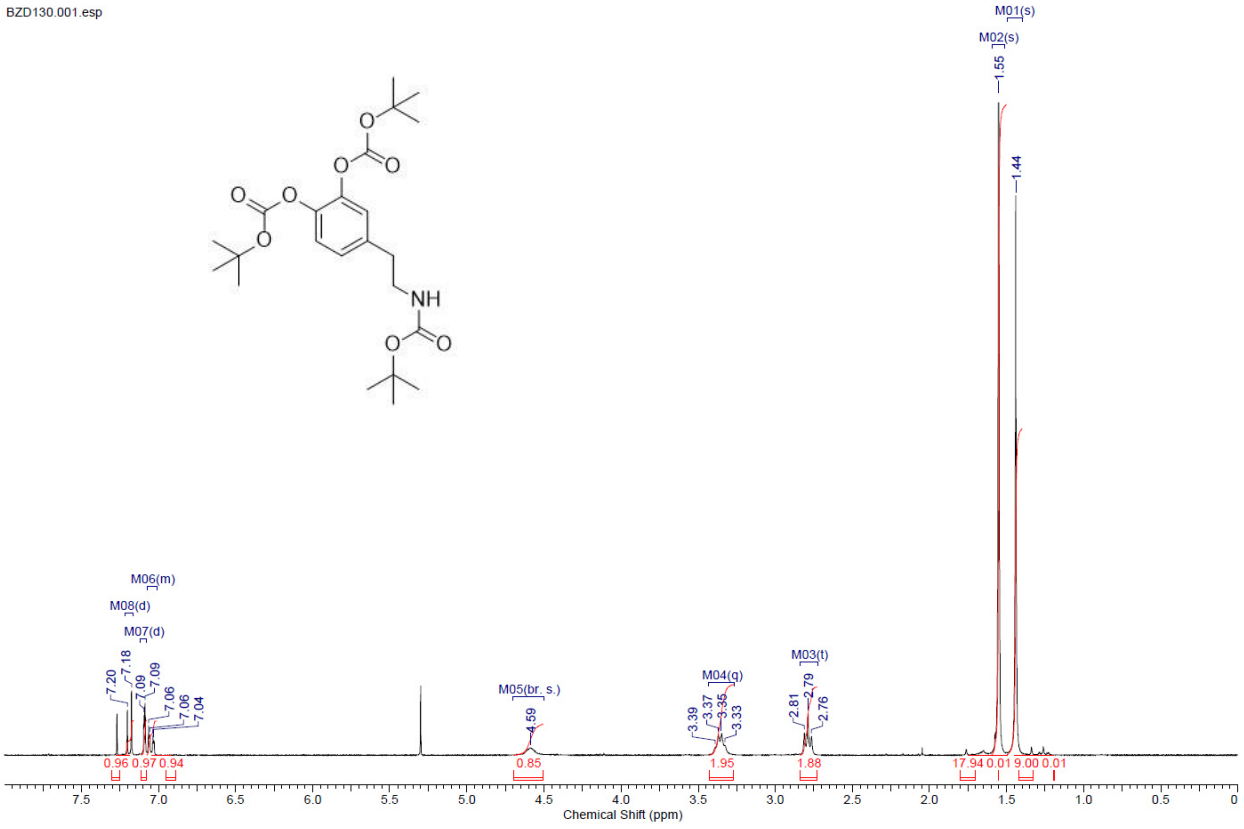


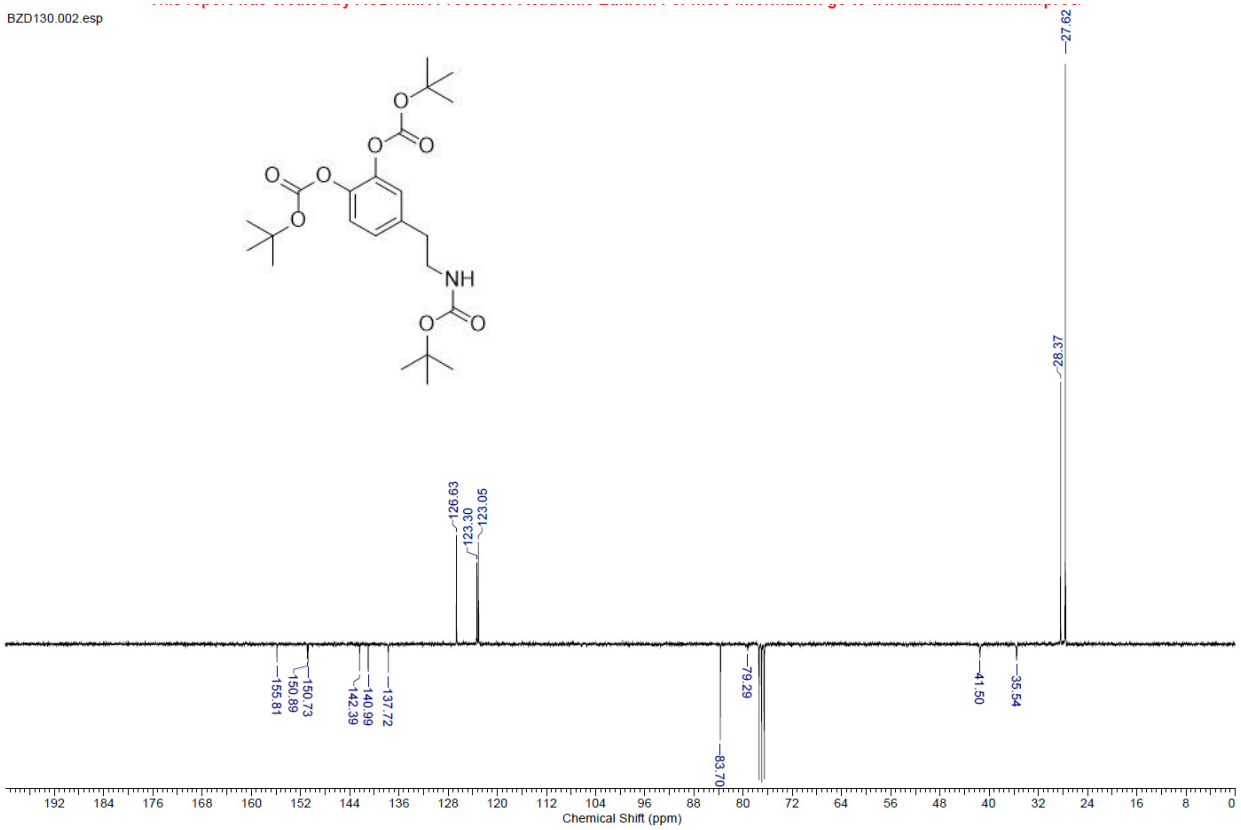


***N*,*O*,*O*-tri-Boc-dopamine:** Et_3N (10.70 mL, 7.74 g, 76.5 mmol) was slowly added to a solution of dopamine hydrochloride (3.17 g, 16.72 mmol) and Boc_2O (15.7 g, 71.93 mmol) in anhydrous DMF (40 mL) under Ar. The reaction mixture was stirred for a further 16 h and concentrated under reduced pressure. The residue was taken up into Et_2O (200 mL) and the resulting solution was

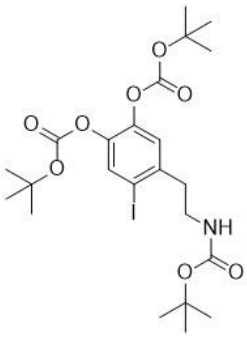


washed with H_2O (10×40 mL), 1 M $NaHSO_4$ (3×30 mL), 5% $NaHCO_3$ (3×30 mL), brine (2×30 mL), dried and concentrated under reduced pressure. The residue was purified by column chromatography (EtOAc:hexane = 1:3) affording the title compound (7.58 g, 100%) as a yellow oil which gradually solidified into a colorless solid. R_f = 0.27, EtOAc:hexane = 1:3. ^{1}H NMR (300 MHz) δ 1.44 (s, 9 H), 1.55 (s, 2×9 H), 2.79 (t, *J*=6.92 Hz, 2 H), 3.36 (q, *J*=6.63 Hz, 2 H), 4.59 (br, 1 H), 7.05 (m, 1 H) ,7.09 (d, *J*=1.92 Hz, 1 H), 7.19 (d, *J*=8.24 Hz, 1 H). ^{13}C NMR (75.5 MHz) δ 155.8, 150.9, 150.7, 142.4, 141.0, 137.7, 126.6, 123.3, 123.1, 83.70 (×2), 79.3, 41.5, 35.5, 28.4, 27.6 (×2). MS (ESI): positive mode m/z = 476.1 ($[M + Na]^+$); ESI HRMS: calcd for $C_{23}H_{35}O_8NNa^+$: 476.2255; found: 476.2254.

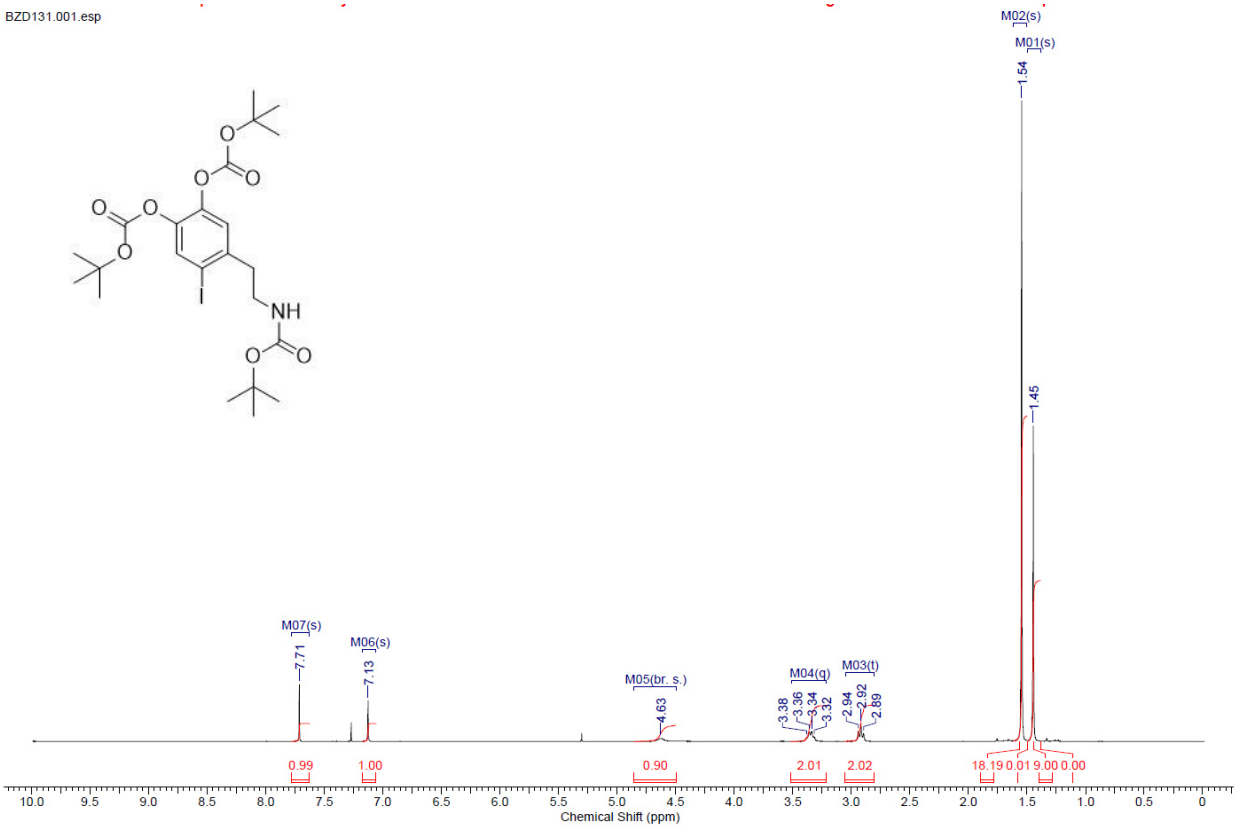


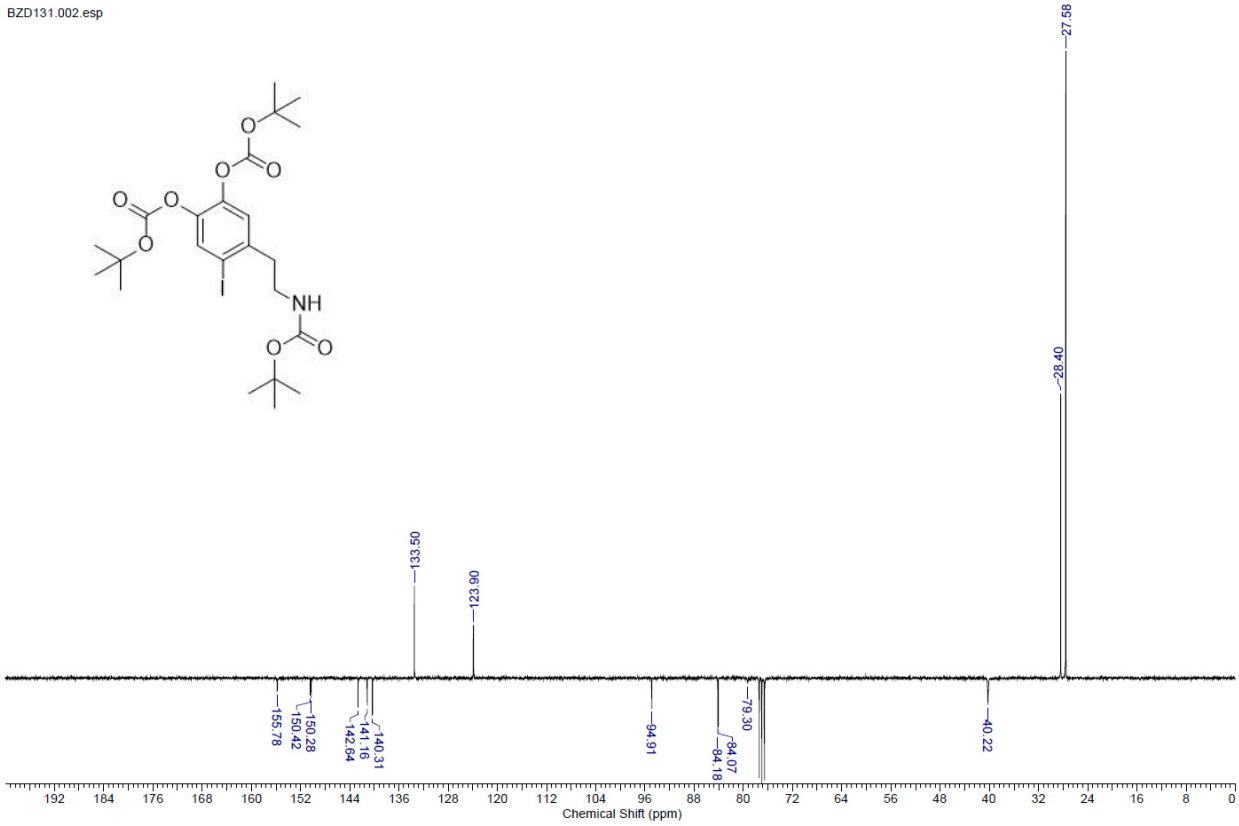


***N*,*O*,*O*-tri-Boc-6-iododopamine:** A solution of *N*,*O*,*O*-tri-Boc-dopamine (7.58 g, 16.72 mmol) in

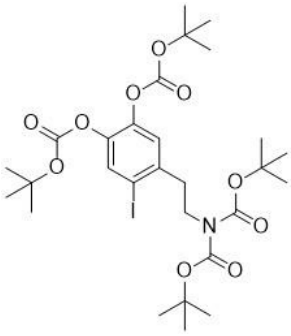


anhydrous CH_2Cl_2 (50 mL) was added to an ice-cold solution of iodine (5.52 g, 21.74 mmol) and [bis(trifluoroacetoxy)iodo]benzene (10.3 g, 23.95 mmol) in anhydrous CH_2Cl_2 (250 mL) and the reaction mixture was stirred for 1 h. The cooling bath was removed, the mixture was protected from light and stirred for further 16 h. Afterwards, the reaction mixture was washed with saturated $Na_2S_2O_3$ (3×100 mL), brine (2×30 mL), dried and concentrated under reduced pressure. The residue was purified by column chromatography (EtOAc:hexane = 1:3) yielding the title compound (5.66 g, 58% over two steps) as a colorless oil which gradually solidified into a colorless solid. Additionally, *N*,*O*,*O*-tri-Boc-dopamine (1.5 g) was recovered. R_f = 0.44, EtOAc:hexane = 1:3. ^{1}H NMR (300 MHz) δ 1.45 (s, 9 H), 1.54 (s, 2×9 H), 2.92 (t, *J*=6.96 Hz, 2 H), 3.35 (q, *J*=6.35 Hz, 2 H), 4.63 (br, 1 H), 7.13 (s, 1 H), 7.71 (s, 1 H). ^{13}C NMR (75.5 MHz) δ 155. 8, 150.4, 150.3, 142.6, 141.2, 140.3, 133.5, 123.9, 94.9, 84.2, 84.1, 79.3, 40.2 (×2), 28.4, 27.6.

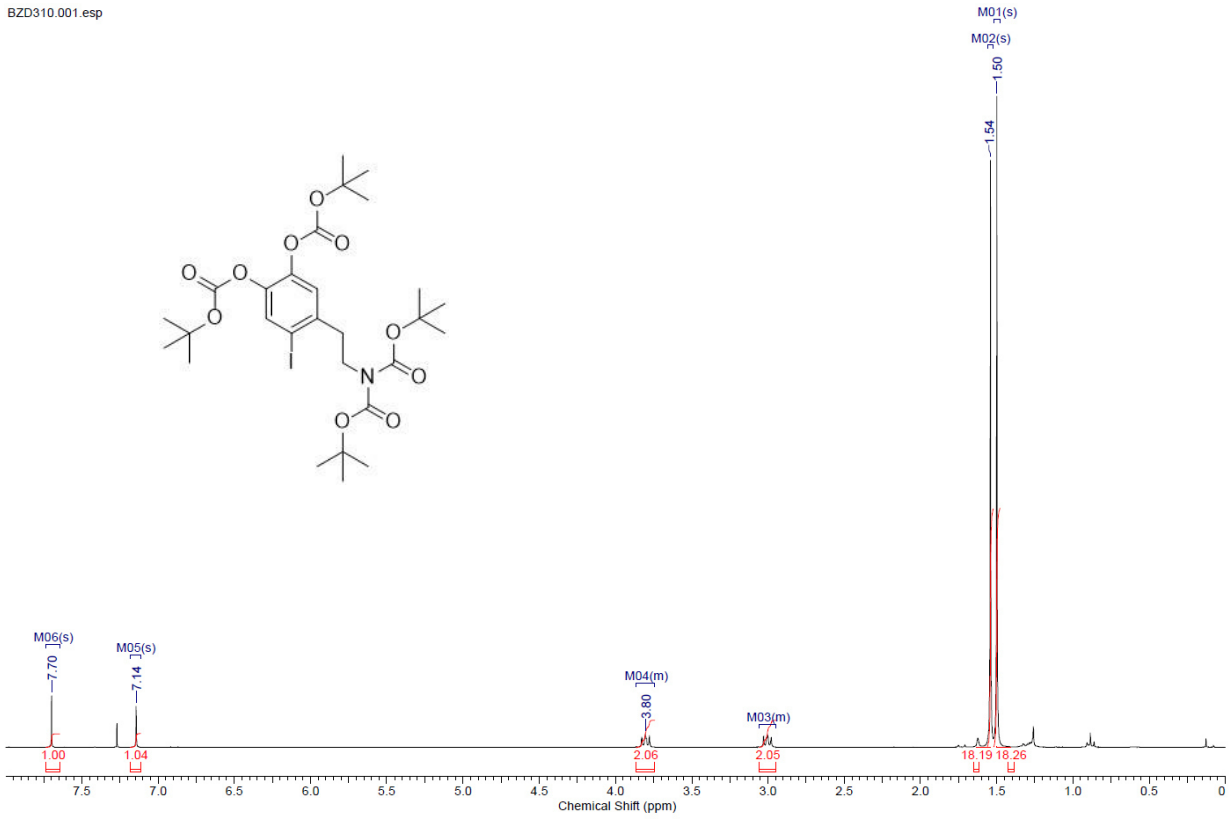


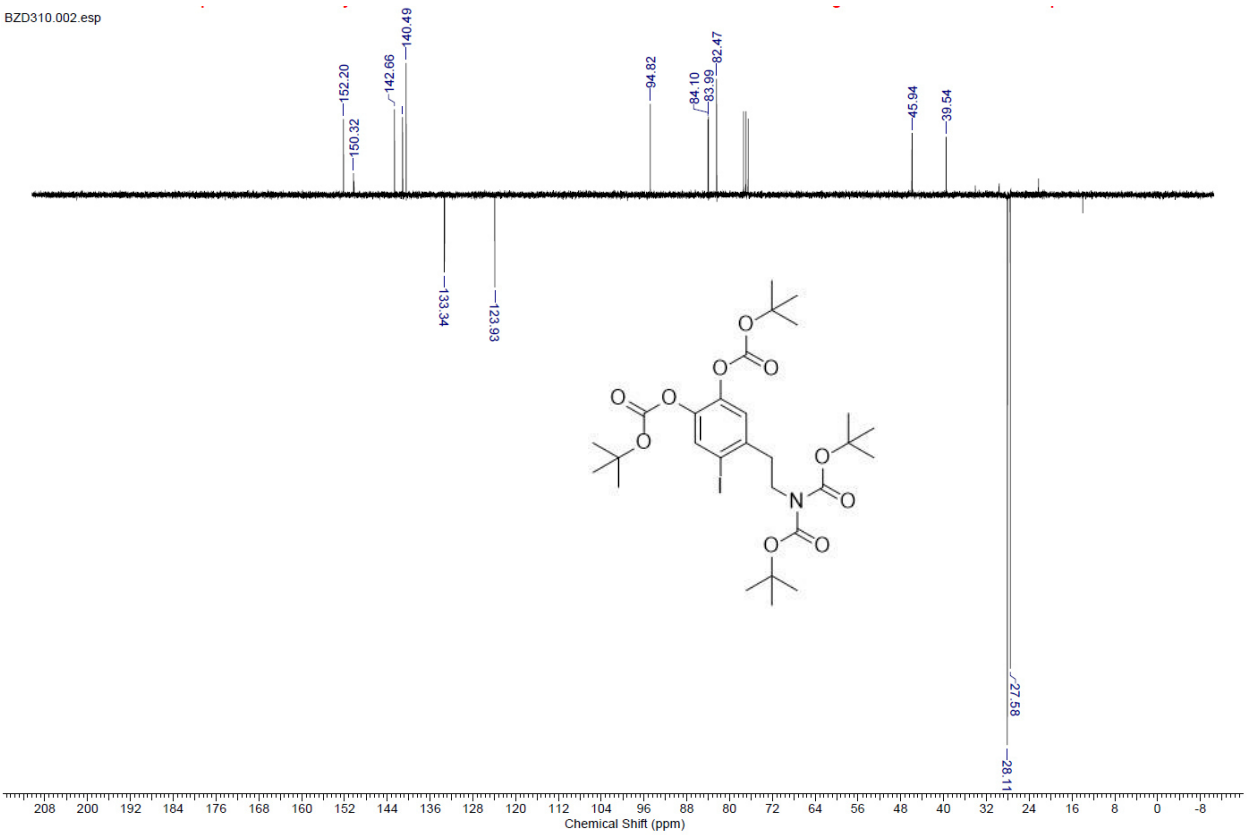


***N*,*N*,*O*,*O*-tetra-Boc-6-iododopamine:** Boc_2O (1.47 g, 6.73 mmol) was added to a solution of *N*,*O*,*O*-tri-Boc-6-iododopamine (7.58 g, 16.72 mmol) and DMAP (33 mg, 0.27 mmol) in anhydrous MeCN (10 mL) and the reaction mixture was stirred for 48 h. Subsequently, the mixture was concentrated under reduced pressure. The residue was taken up into Et_2O (50 mL) and the

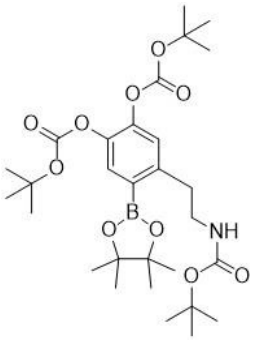


resulting solution was washed with 1 M $NaHSO_4$ (3×30 mL), H_2O (20 mL), 5% $NaHCO_3$ (3×30 mL), H_2O (3×20 mL), brine (2×30 mL), dried and concentrated under reduced pressure. The crude product was purified by column chromatography (EtOAc:hexane = 1:10) to give the title compound (1.45 g, 95%) as a light yellow oil. R_f = 0.51, EtOAc:hexane = 1:10. ^{1}H NMR (300 MHz) δ 1.50 (s, 2×9 H), 1.54 (s, 2×9 H), 3.01 (m, 2 H), 3.80 (m, 2 H), 7.14 (s, 1 H), 7.70 (s, 1 H). ^{13}C NMR (75.5 MHz) δ 152.2 (×2), 150.3, 150.3, 142.7, 141.2, 140.5, 133.3, 123.9, 94.8, 84.1, 84.0, 82.5 (×2), 45.9, 39.5, 28.1, 27.6. MS (ESI): positive mode m/z = 702.0 ([M + Na]$^+$); ESI HRMS: calcd for $C_{28}H_{42}O_{10}NINa^+$: 702.1746; found: 702.1748.

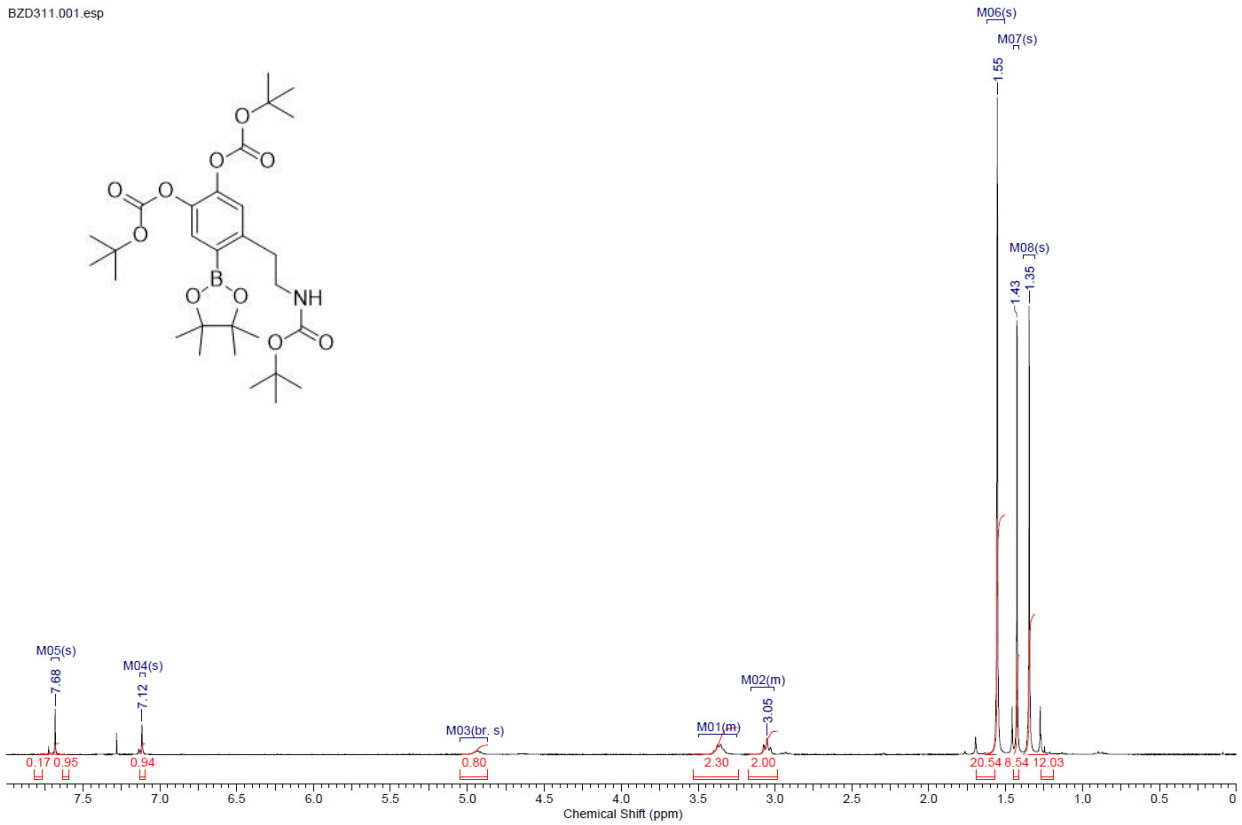


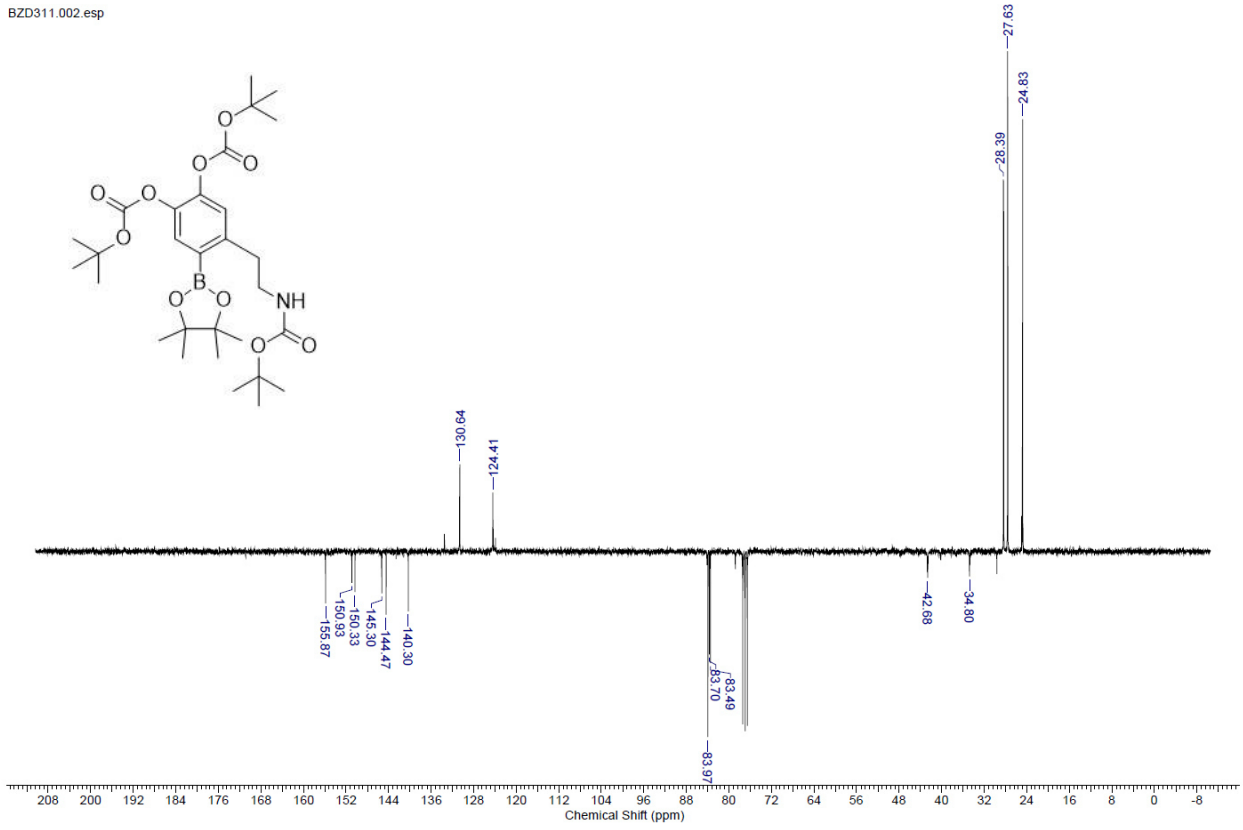


***N*,*O*,*O*-tri-Boc-6-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)dopamine (18a):** A solution of

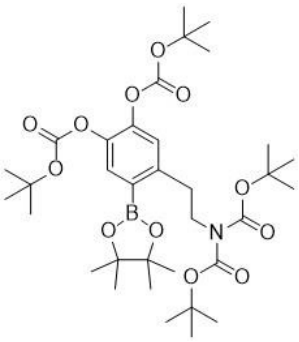


N,*O*,*O*-tri-Boc-6-iododopamine (1.3 g, 2.24 mmol), $B_2(Pin)_2$ (0.68 g, 2.69 mmol), $Pd(dppf)Cl_2$ (0.183 g, 0.224 mmol) and KOAc (0.64 g, 6.73 mmol) in anhydrous DMF (30 mL) was stirred at 80 °C for 3 h. The mixture was concentrated under reduced pressure. The residue was taken up into Et_2O (70 mL) and the resulting solution was washed with H_2O (10×40 mL), 5% $NaHCO_3$ (3×30 mL), brine (2×30 mL), dried and concentrated under reduced pressure. The residue was purified by column chromatography (EtOAc:hexane = 1:10; silica with 0.1% CaO; two times) yielding the title compound (0.42 g, 87% purity, 28%) as a colorless foam. The product containing, in accordance to the ^{1}H-NMR spectrum ca. 13% of *N*,*O*,*O*-tri-Boc-6-iododopamine was used for radiolabeling experiments without any further purification. R_f = 0.30, EtOAc:hexane = 1:10; developed twice. ^{1}H NMR (300 MHz) δ 1.35 (s, 4×3 H), 1.43 (s, 9 H), 1.55 (s, 2×9 H), 3.08 (m, 2 H), 3.38 (m, 2 H), 4.96 (br, 1 H), 7.12 (s, 1 H), 7.68 (s, 1 H). ^{13}C NMR (75.5 MHz) δ 155.9, 150.9, 150.3, 145.3, 144.5, 140.3, 130.6, 124.4, 84.0, 83.7, 83.5, 42.7, 34.8, 28.4, 27.63, 27.61, 24.8. C–B was not observed. MS (ESI): positive mode m/z = 602.2 ([M + Na]$^+$); ESI HRMS: calcd for $C_{29}H_{46}O_{10}NBNa^+$: 602.3107; found: 602.3107. Correct isotopic pattern.

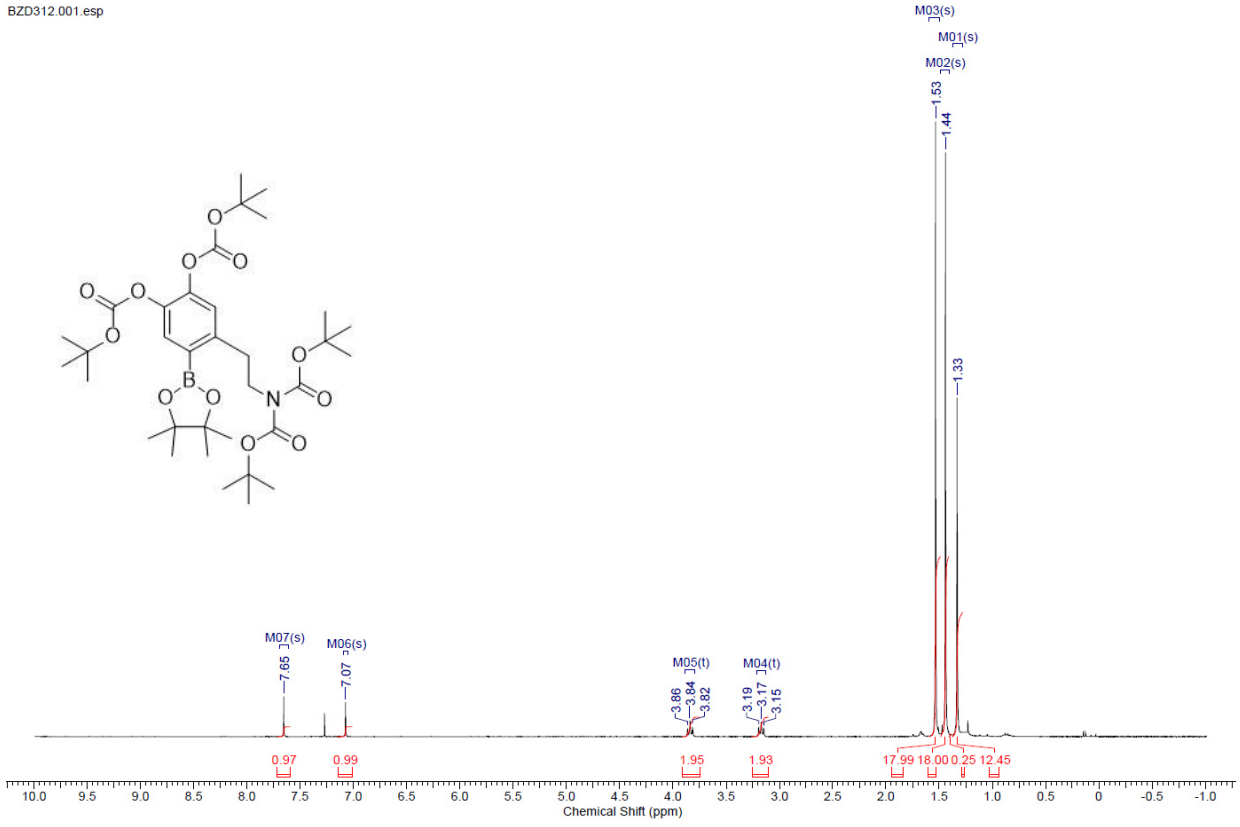


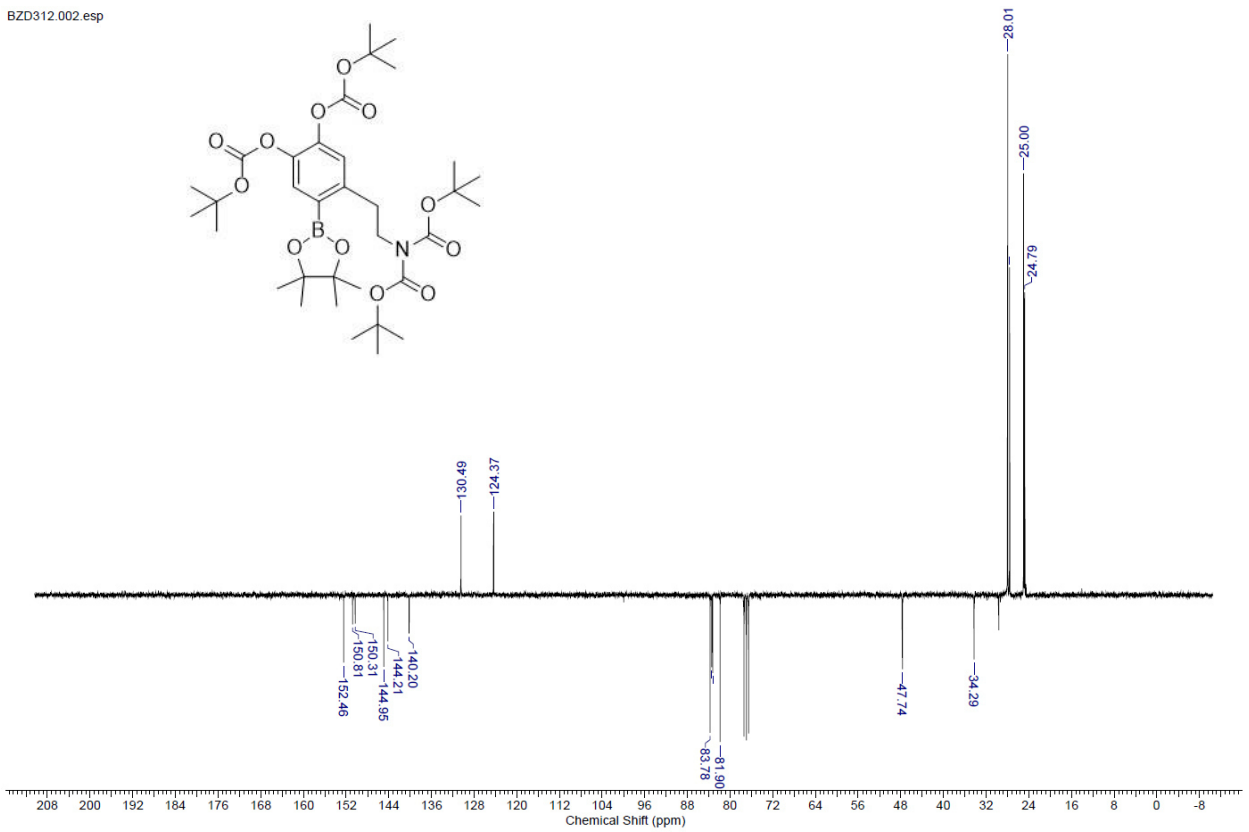


***N*,*N*,*O*,*O*-tetra-Boc-6-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)dopamine (18b):** A solution of *N*,*N*,*O*,*O*-tetra-Boc-6-iododopamine (0.76 g, 1.12 mmol), $B_2(Pin)_2$ (0.45 g, 1.79 mmol), Pd(dppf)Cl_2 (92 mg, 0.113 mmol) and KOAc (0.33 g, 3.36 mmol) in anhydrous DMF (30 mL) was stirred at 75 °C for 16 h. The mixture was concentrated under reduced pressure. The residue

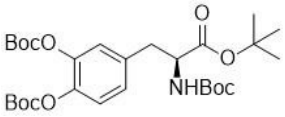


was taken up into pentane (70 mL) and H_2O (40 mL), the aqueous layer was separated and the pentane layer was washed with H_2O (5×40 mL), brine (2×30 mL), dried and concentrated under reduced pressure. The residue was purified by column chromatography (EtOAc:hexane = 1:13; silica with 0.1% CaO) yielding the title compound (0.46 g, 61%) as a colorless foam. R_f = 0.14, EtOAc:hexane = 1:13. ^{1}H NMR (300 MHz) δ 1.33 (s, 4×3 H), 1.44 (s, 2×9 H), 1.53 (s, 2×9 H), 3.17 (t, *J*=7.05 Hz, 2 H), 3.84 (t, *J*=7.05 Hz, 2 H), 7.07 (s, 1 H), 7.65 (s, 1 H). ^{13}C NMR (75.5 MHz) δ 152.5 (×2), 150.8, 150.3, 145.0, 144.2, 140.2, 130.5, 124.4, 83. 8, 83.5, 83.3, 81.9, 47.7, 34.3, 28.0, 27.6, 25.0, 24.8. C–B was not observed. MS (ESI): positive mode m/z = 702.3 ([M + Na]$^+$); ESI HRMS: calcd for $C_{34}H_{54}O_{12}NBNa^+$: 702.3631; found: 702.3627. Correct isotopic pattern.





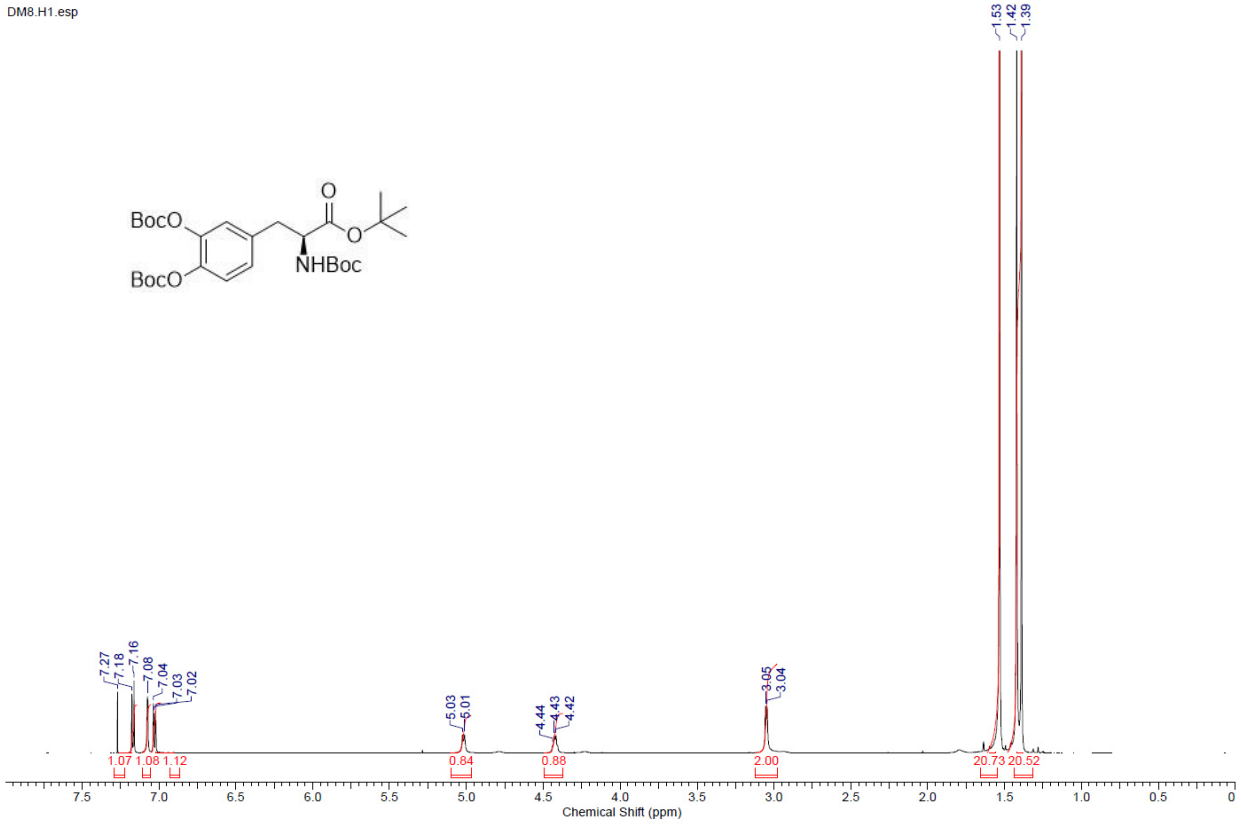
***N*,*O*,*O*-tri-Boc-DOPA-O*t*Bu:** 60% $HClO_4$ (0.83 mL, 7.61 mmol) was added slowly to an ice-cold

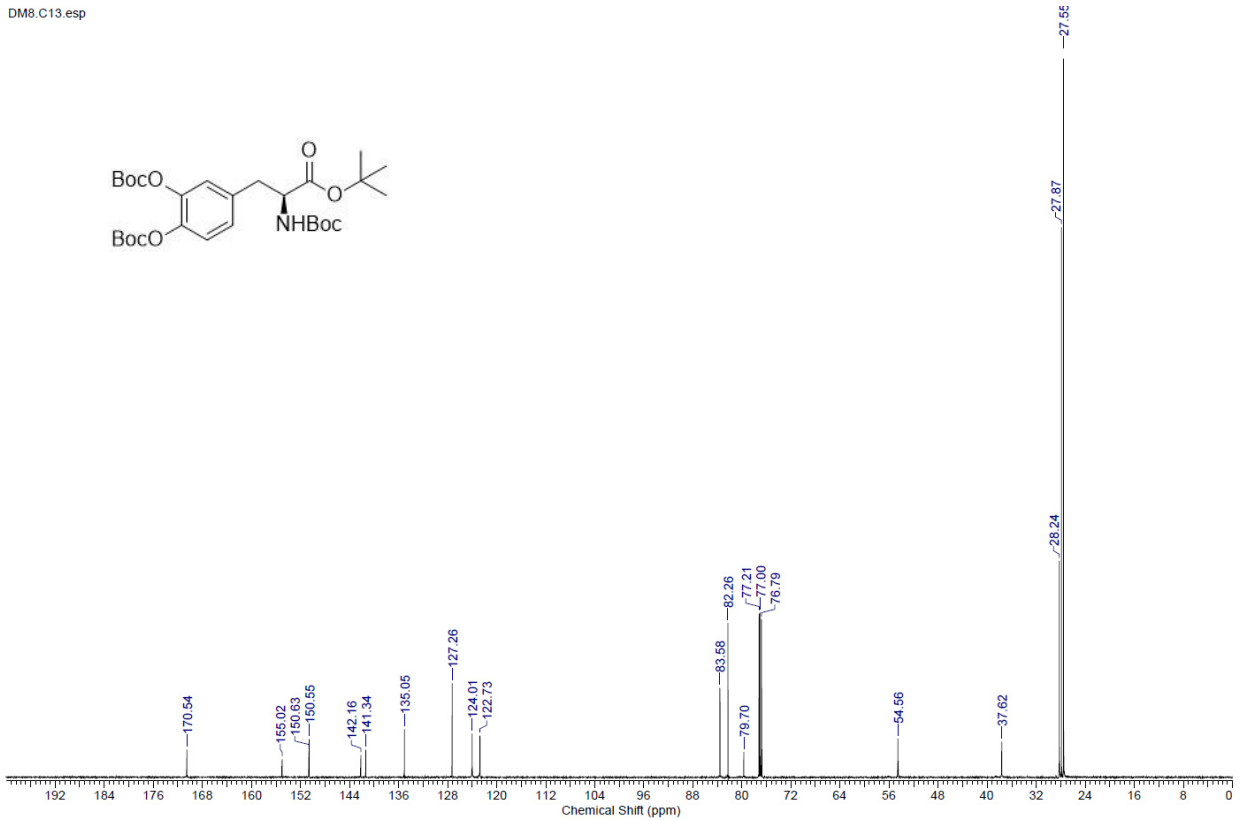


suspension of 3,4-dihydroxy-L-phenylalanine (DOPA) (1.00 g, 5.07 mmol) in AcO*t*Bu (12 mL). The cooling bath was removed and the reaction mixture was stirred for 24 h. Afterwards, the pH was adjusted to 8–9 using 10% K_2CO_3 and the mixture was extracted with EtOAc (×3). The organic layers were combined, dried and concentrated under reduced pressure yielding the crude H-DOPA-O*t*Bu (1.28 g) which was used in the next step without further purification.

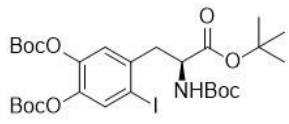
Boc_2O (3.98 g, 18.25 mmol) and Et_3N (2.53 mL, 1.84 g, 18.2 mmol) were added to an ice-cold solution of the crude H-DOPA-O*t*Bu (1.28 g, 5.07 mmol) in anhydrous DMF (25 mL). The cooling bath was removed and the reaction mixture was stirred for 24 h. Thereafter, the mixture was diluted with EtOAc (50 mL), washed with H_2O and brine, dried and concentrated under reduced pressure. The residue was purified by flash chromatography [5→10% EtOAc in petrol ether (0.1% triethylamine)] giving the title compound (1.41 g, 50% over two steps) as a highly viscous colorless oil. R_f = 0.35 (EtOAc:hexane = 1:4). ^{1}H-NMR (600 MHz) δ 7.16 (m, *J* = 8.3 Hz, 1 H), 7.07 (br, 1H), 7.02 (dd, *J* = 8.3, 1.8 Hz, 1 H), 5.01 (d, *J* = 7.7 Hz, 1 H), 4.41 (m, 1 H), 3.04 (m, 2 H), 1.52 (s, 2×9 H), 1.40 (m, 18 H). ^{13}C-NMR (151 MHz) δ 170.7, 155.2, 150.8, 150.7, 142.3, 141.5, 135.2, 127.4, 124.2, 122.9, 83.7, 82.4, 79.9, 54.7, 37.8, 28.4, 28.0, 27.7, 27.7. ESI HRMS: calcd for $C_{28}H_{43}O_{10}NNa^+$: 576.2779; found: 576.2775. Elemental analysis calcd (%) for

$C_{28}H_{43}NO_{10}$ (553.6): C 60.74%, H 7.83%, N 2.53%; found: C 60.45±0.14, H 7.87±0.06, N 2.58±0.05%.

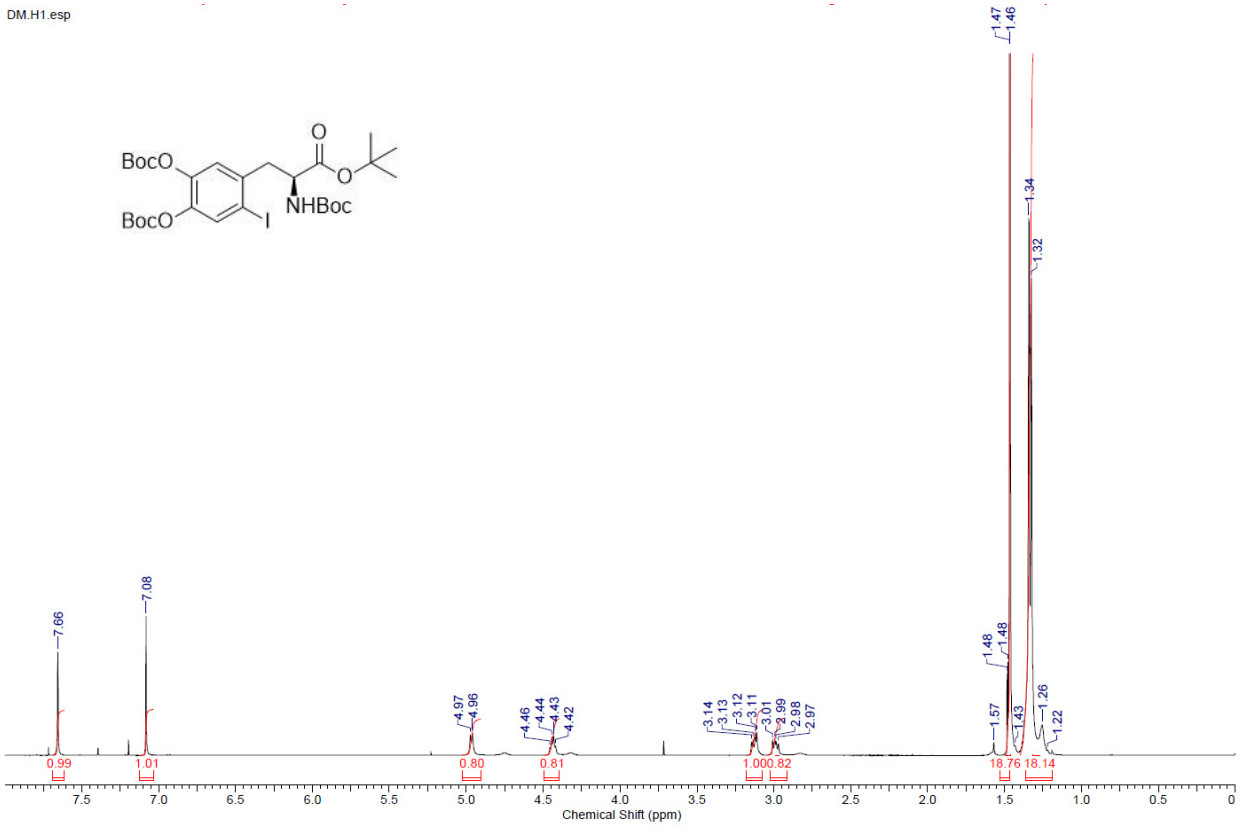


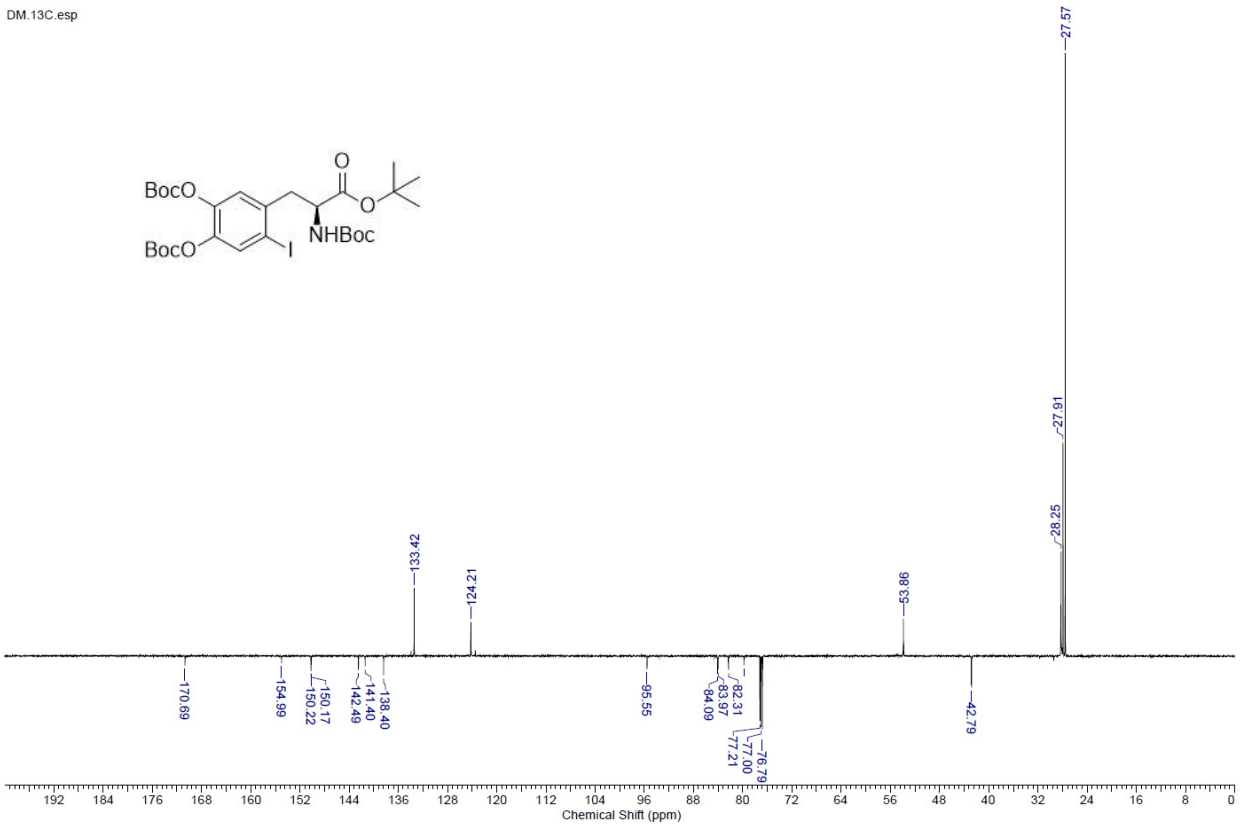


***N*,*O*,*O*-tri-Boc-6-iodo-DOPA-O*t*Bu:** Bis(trifluoroacetoxy)iodobenzene (1.89 g, 4.40 mmol) and I_2 (0.93 g, 3.67 mmol) were added to an ice-cold solution of *N*,*O*,*O*-tri-Boc-DOPA-O*t*Bu (2.03 g,

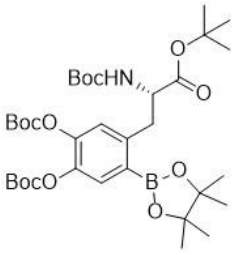


3.67 mmol) in anhydrous DCM (15 mL) and the reaction mixture was stirred for 15 min. Afterwards, the cooling bath was removed and the mixture was stirred for further 3 h. The reaction was washed with saturated Na_2SO_3 (×2), H_2O, brine, dried and concentrated under reduced pressure. The residue was purified by flash chromatography [5→10% EtOAc in petrol ether (0.1% triethylamine)] affording the title compound (1.31 g, 53%) as a highly viscous colorless oil. R_f = 0.39 (EtOAc:hexane = 1:4). ^{1}H NMR (600 MHz) δ 7.72 (s, 1 H), 7.15 (s, 1 H), 5.03 (d, J = 8.3 Hz, 1 H), 4.50 (m, 1 H), 3.19 (dd, J = 14.1, 6.0 Hz, 1 H), 3.05 (dd, J = 14.0, 8.5 Hz, 1 H), 1.53 (s, 2×9 H), 1.9 (s, 2×9 H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 172.0, 170.8, 155.2, 150.4, 150.3, 142.6, 141.6, 138.6, 134.1, 133.6, 124.4, 123.6, 95.7, 84.3, 84.1, 82.5, 79.9, 54.0, 43.0, 28.4, 28.1, 27.7. ESI HRMS: calcd for $C_{28}H_{47}O_{10}N_2^+$: 697.2192; found: 697.21795. Elemental analysis calcd (%) for $C_{28}H_{42}INO_{10}$ (679.5): C 49.49, H 6.24%, N 2.06%; found: C 49.49±0.24, H 6.22±0.04, N 1.81±0.01.

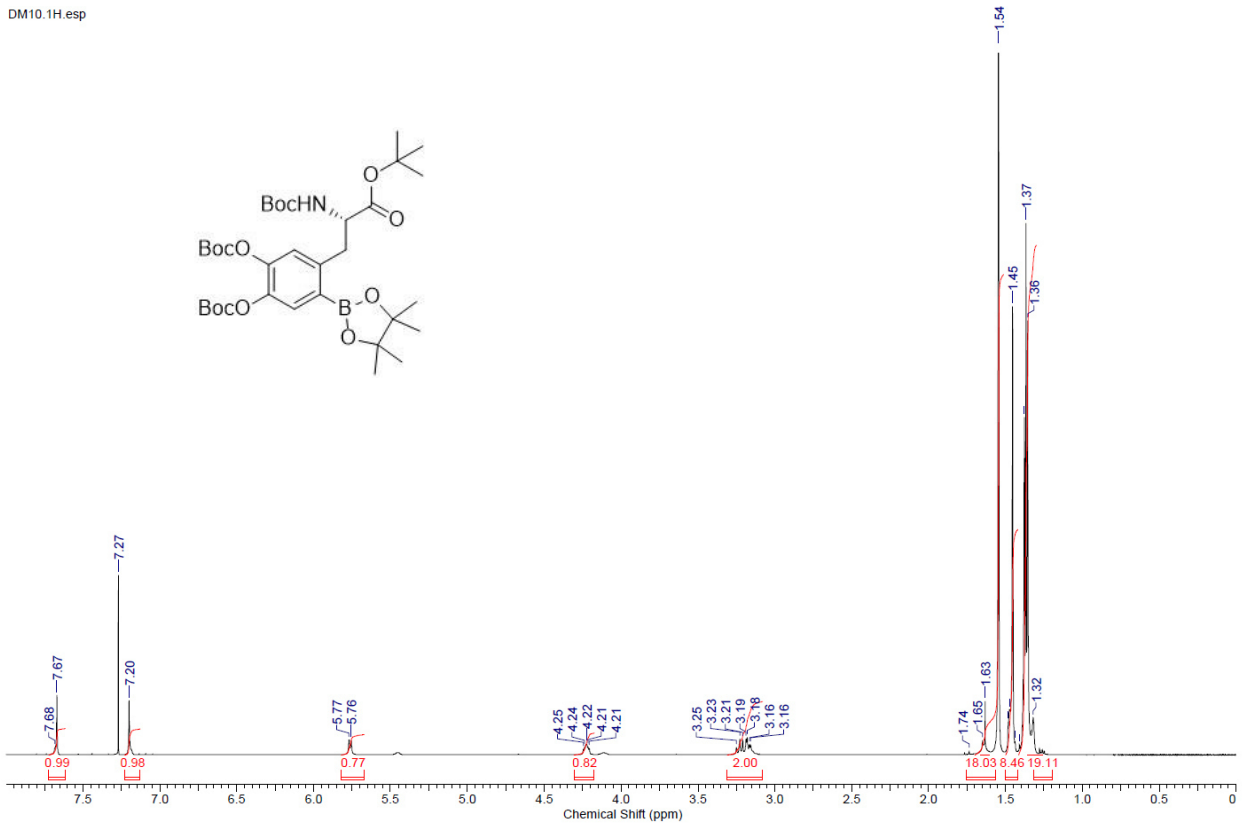


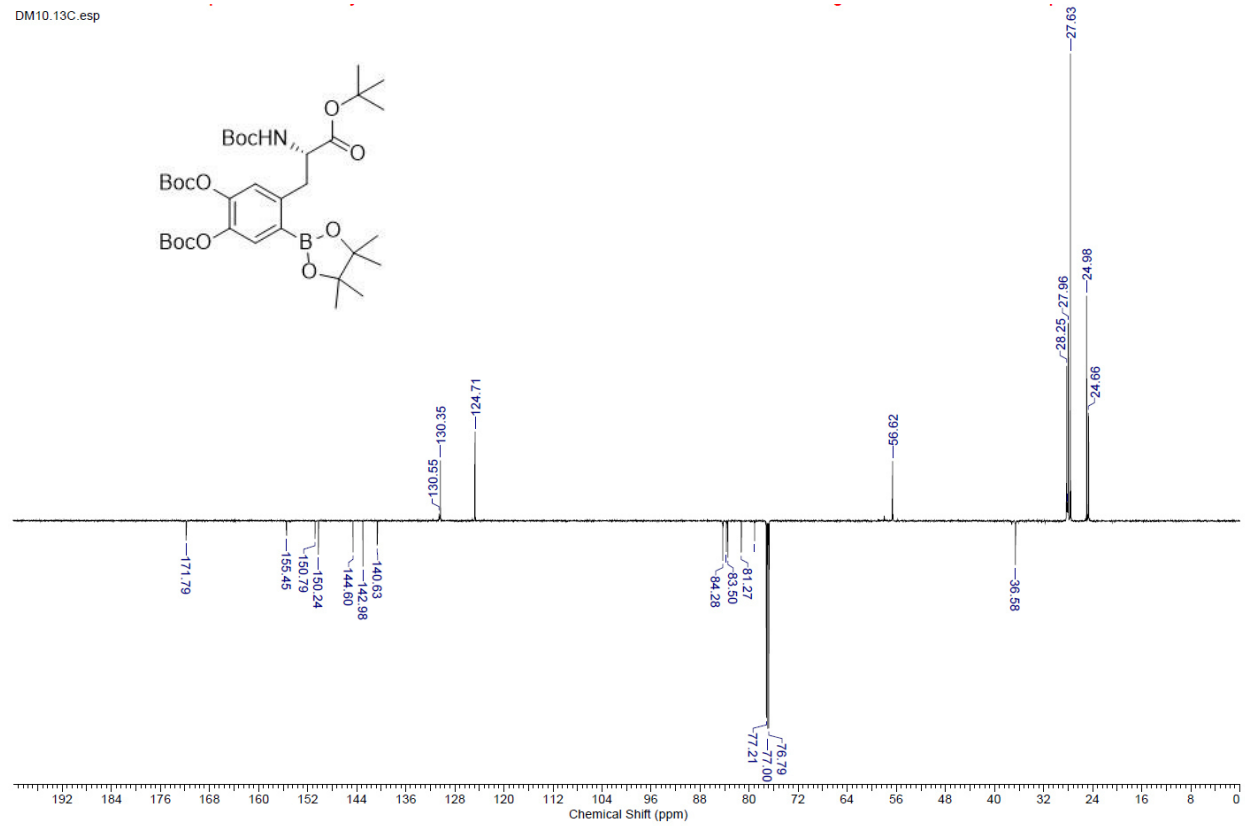


_N_,_O_,_O_-tri-Boc-6-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-DOPA-O*t*Bu (21[a]): A round

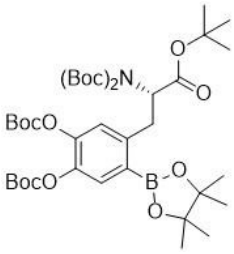


flask containing bis(pinacolato)diboron (0.29 g, 1.13 mmol), Pd(dppf)Cl_2×CH_2Cl_2 (80 mg,0.10 mmol) and KOAc (0.30 g, 3.09 mmol) was several times evacuated and backfilled with Ar. Thereafter, a solution of _N_,_O_,_O_-tri-Boc-6-iodo-DOPA-O*t*Bu (0.70 g, 1.03 mmol) in anhydrous DMF (10 mL) was added and the reaction mixture was stirred for 3 h at 80 °C. The mixture was cooled to ambient temperature, filtered and concentrated under reduced pressure. The residue was taken up into Et_2O (5 mL) and diluted with hexane (50 mL). The precipitate was removed by filtration through Celite® and the solution was concentrated under reduced pressure. The oily residue was purified by RP-flash chromatography (90% MeCN) affording the title compound (0.28 g, 40%) as a colorless foam. R_f = 0.34 (EtOAc:hexane = 1:4). ^{1}H NMR (600 MHz) δ 7.66 (s, 1 H), 7.19 (s, 1 H), 5.75 (d, J = 8.1 Hz, 1 H), 4.22 (m, 1 H), 3.19 (m, 2 H), 1.53 (s, 2×9 H), 1.44 (s, 9 H), 1.36 (5×s, 21 H). ^{13}C NMR (151 MHz) δ 171.9, 155.6, 150.9, 150.4, 144.8, 143.1, 140.8, 130.5, 124.9, 84.4, 83.9, 83.7, 81.4, 79.3, 56.8, 36.7, 28.4, 28.1, 27.8, 27.8, 25.1, 24.8. ESI HRMS: calcd for $C_{34}H_{54}O_{12}BNNa^+$: 702.3630; found: 702.3631. Elemental analysis calcd (%) for $C_{34}H_{54}BNO_{12}$ (679.6): C 60.09%, H 8.01%, N 2.06%; found: C 59.90±0.07%, H 8.00±0.04%, N 1.97±0.02%.

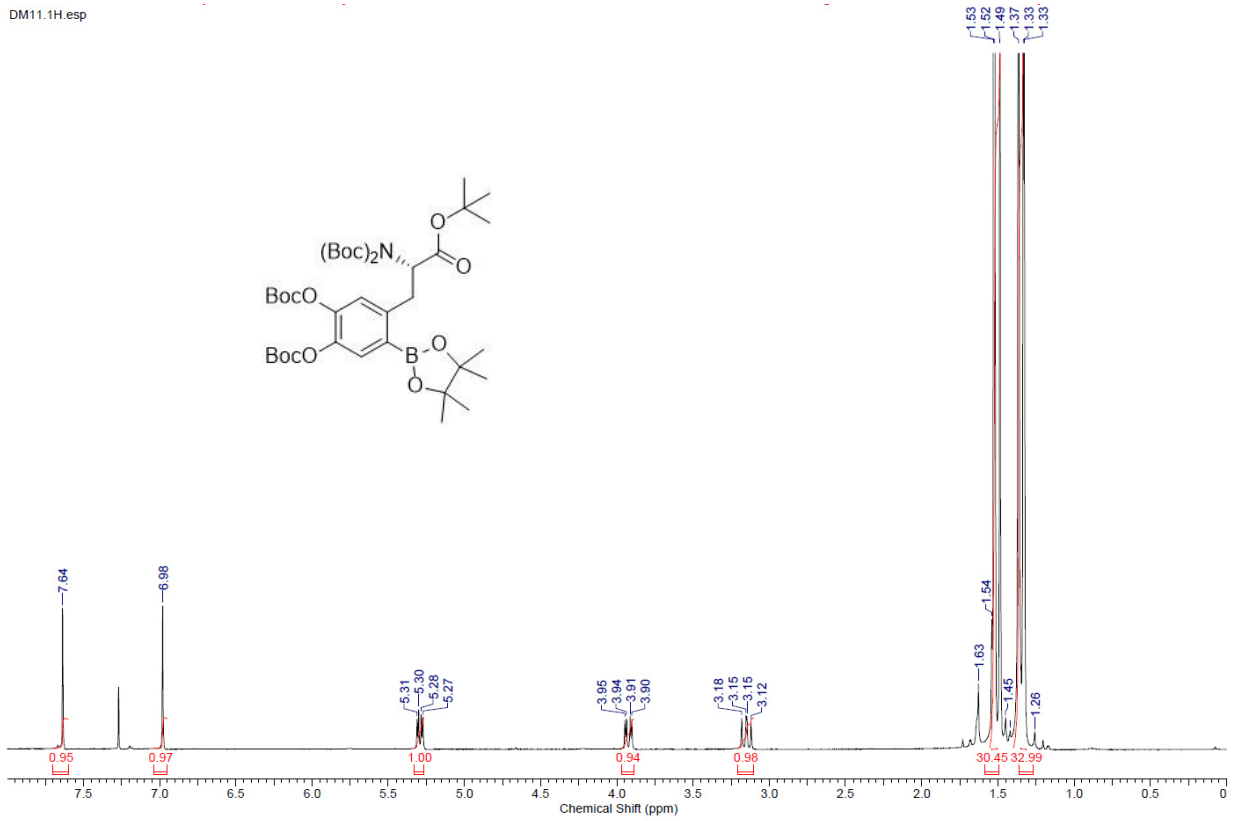


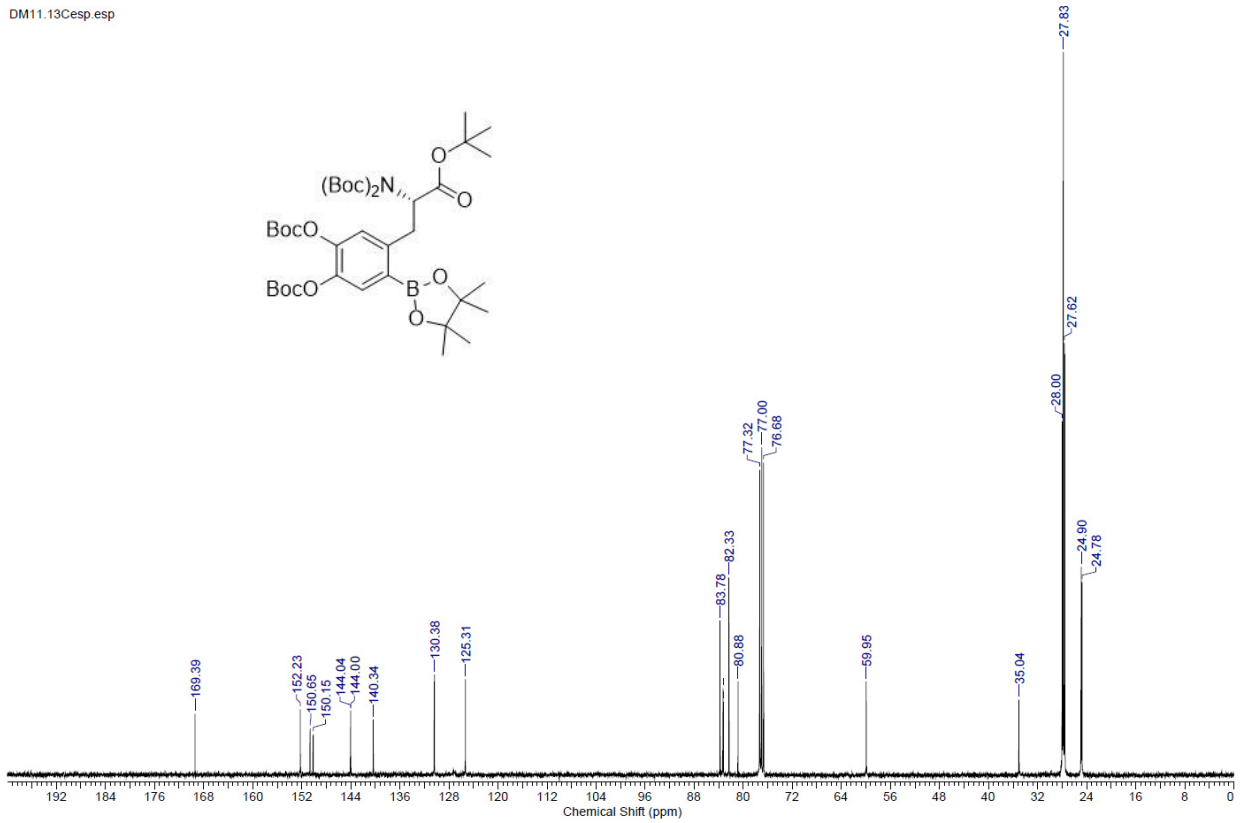


***N*,*N*,*O*,*O*-tetra-Boc-6-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-DOPA-O*t*Bu (21b)**: DMAP (10 mg, 0.10 mmol) was added to an ice-cold solution of *N*,*O*,*O*-tri-Boc-6-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-DOPA-O*t*Bu (0.35 g, 0.52 mmol), Boc_2O (0.34 g, 1.55 mmol) and Et_3N (0.21 mL, 1.55 mmol) in anhydrous MeCN (10 mL) and the reaction mixture was stirred for 15 min. Afterwards, the cooling bath was removed, the mixture was warmed up to 40

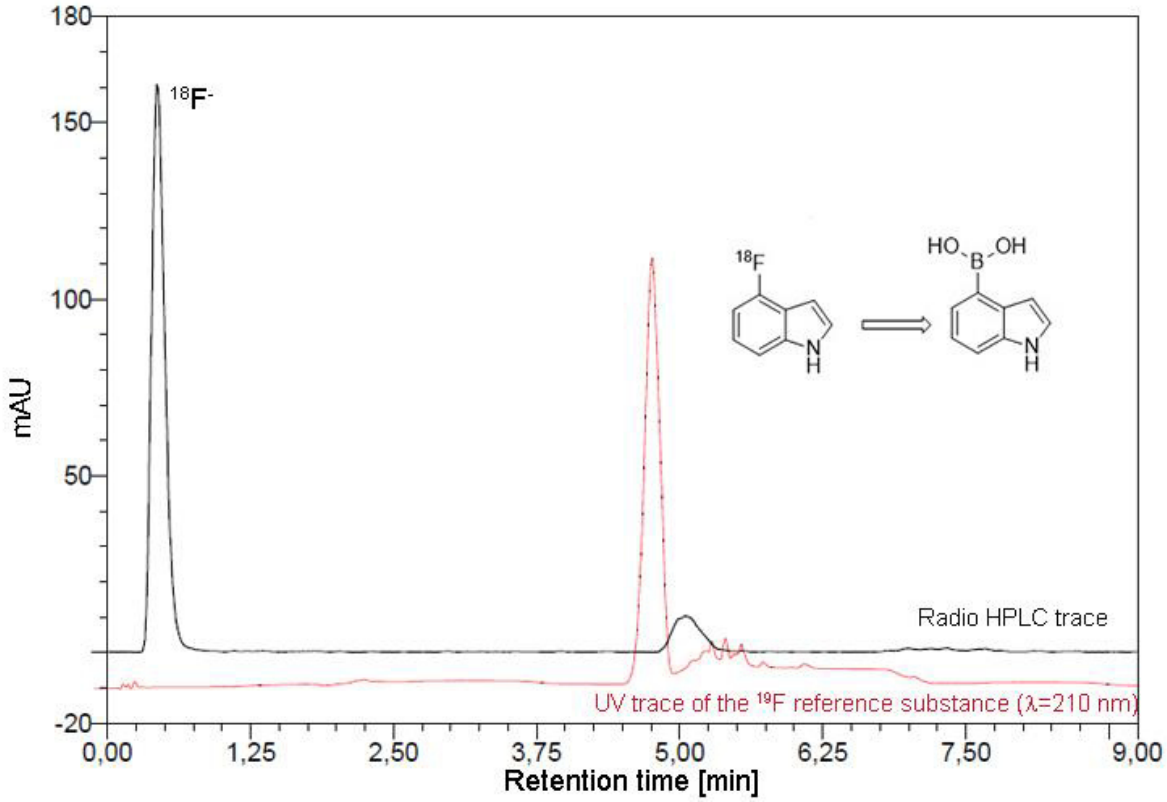


°C and stirred at this temperature for further 2 h. Thereafter, the reaction mixture was diluted with EtOAc, washed with saturated NH_4Cl, H_2O, and brine, dried and concentrated under reduced pressure. The residue was taken up into Et_2O, the resulting solution was filtered through a small pad of silica and concentrated under reduced pressure. The residue was purified by RP-flash chromatography (90→95% MeCN) yielding the title compound (0.17 g, 30%) as a colorless foam. R_f = 0.47 (EtOAc:hexane = 1:4). ^{1}H-NMR (400 MHz) δ 7.63 (s, 1 H), 6.97 (s, 1 H), 5.28 (dd, J = 11.1, 4.3 Hz, 1 H), 3.92 (dd, J = 13.6, 4.3 Hz, 1 H), 3.14 (dd, J = 13.4, 11.2 Hz, 1 H), 1.52 (s, 2×9 H), 1.48 (m, 9 H) 1.36 (s, 2×9 H), 1.32 (s, 4×3 H). ^{13}C-NMR (101 MHz) δ 169.6, 152.4, 150.8, 150.3, 144.2, 144.2, 140.5, 130.5, 127.3, 125.5, 83.9, 83.5, 83.4, 82.5, 81.0, 60.1, 35.2, 28.2, 28.0, 27.8, 27.7, 25.1, 24.9. ESI HRMS: calcd for $C_{39}H_{62}O_{14}BNNa^+$: 802.4156; found: 802.4150. Elemental analysis calcd (%) for $C_{39}H_{62}BNO_{14}$ (779.7): C 60.08%, H 8.01%, N 1.80%; found: C 59.93±0.12%, H 8.11±0.02%, N 1.74±0.01%.





Preparation of 4-Fluoroindole: Tetramethylammonium fluoride (1 mg, 10.7 µmol) was taken up intoto *i*-PrOH (1 mL) and the resulting solution was concentrated to dryness at 600 mbar and 100 °C. Thereafter, the residue was taken up into a solution of $^{18}F^-$ (50 MBq) obtained by the elution of $^{18}F^-$ from a QMA cartridge with a solution of Et_4NHCO_3 (2.7 mg, 18 µmol) in *n*-BuOH (400 µL). A solution of indole-4-boronic acid (9.7 mg, 60 µmol) and $Cu(OTf)_2(py)_4$ (18 mg, 26.5 µmol) in DMA (800 µL) was added. The reaction mixture was heated at 110 °C for 20 min under air atmosphere. The mixture was cooled to room temperature, diluted with water (2 mL) and vigorously shaken for 30 s. Formation of 4-fluoroindole (12% based on the applied amount of Me_4NF) was confirmed by radio-HPLC.



Radiochemistry:

All radiosyntheses were carried out using anhydrous DMA and DMF stored over molecular sieves (available from "Acros" or "Aldrich") and under ambient air. All commercially available substrates for radiolabeling were used as received. $Cu(OTf)_2(py)_4$ was prepared according to the literature[7] and stored under ambient conditions without any precautions. StrataX cartridges were obtained from Phenomenex (Aschaffenburg, Germany) and Sep-Pak Accell Plus QMA carbonate plus light cartridges, 46 mg sorbent per cartridge from Waters GmbH (Eschborn, Germany).

HPLC analyses were carried out on a Dionex Ultimate® 3000 HPLC systems and a DAD UV detector coupled in series with a Berthold NaI detector. A Chromolith® SpeedROD RP-18e column (Merck, Darmstadt Germany), 50×4.6 mm was used.

UV and radioactivity detectors were connected in series, giving a time delay of 0.1–0.9 min depending on the flow rate. ^{18}F-Labeled compounds were identified by co-injection of the unlabelled reference compounds using HPLC.

[^{18}F]Fluoride was produced via the ^{18}O(p,n)^{18}F reaction by bombardment of enriched [^{18}O]water with 16.5 MeV protons using a MC16 cyclotron (Scanditronix, Uppsala, Sweden) at the Max Planck Institute for Metabolism Research.

Each radiochemical experiment was carried out at least in triplicates. In the majority of cases the standard deviation of RCC did not exceed 10% of its mean value. If the standard deviation of RCC exceeded 10% of its mean value the range of RCC is given.

Before radiosynthesis [^{18}F]fluoride was processed as follows. Aqueous [^{18}F]fluoride was loaded onto an anion-exchange resin (QMA cartridge). It should be noted, that aqueous [^{18}F]fluoride was loaded onto the cartridge form the male side, whereas flushing, washing and $^{18}F^-$ elution were carried out from the female side. If the QMA cartridge had been loaded, flushed and eluted from the female side only, sometimes a significant amount of [^{18}F]fluoride remained on the resin (this is probably because QMA-light (46 mg) cartridges have a single frit on the male side but four frits on the female side). If MeOH was used for elution, the resin was flushed with MeOH (2 mL) before use. If any other alcohol was used for the $^{18}F^-$ elution, the resin was washed with acetone (2 mL) and flushed with air (10 mL) before use.

If absolutely anhydrous *n*-BuOH was used for $^{18}F^-$ elution, radioactivity recovery from the anion exchange resin was amounted to 72–76%. In this case, addition of higher amounts of Et_4NOTf (10–15 mg) to the elution solution improved $^{18}F^-$ recovery to > 80% without decrease in RCC.

In order to account for surface-adsorbed $^{18}F^-$, reaction vials were completely emptied after the addition of water, and radioactivity in the aqueous solution and remaining radioactivity in the reaction vessel was separately determined and quantified. In all cases, ≥ 95% of radioactivity was observed in the aqueous solution.

UV and radioactivity detectors were connected in series, giving a time delay of 0.2–0.9 min depending on the flow rate. ^{18}F-Labeled compounds were identified by spiking of the reaction mixture with unlabeled standards using HPLC.

If not otherwise stated radiolabeled products were analyzed using as follows:

column: Chromolith SpeedROD®, 50×4.6 mm (Merck Millipore); gradient: 0–2 min: 5% MeCN, 2–2.5 min: 5→20% MeCN, 2.5–6 min: 20% MeCN, 6–7 min: 20→70% MeCN, 7–9 min: 70% MeCN, flow rate: 3.0 mL/min.

TLC analyses were carried out on TLC Al foils precoated with Silica gel 50×100 mm with fluorescent indicator (Sigma-Aldrich; 91835-50EA) as follows. Reaction mixtures (see GP-5) were cooled to ambient temperature, diluted with water (2 mL) and vigorously stirred for 30 s. Thereafter, aliquots of mixtures (1 µL) were applied onto TLC plates. The TLC plates were briefly dried at ambient temperature and developed using the appropriate solvent (EtOAc/hexane=1:4, for [^{18}F]**5**, [^{18}F]**9** and [^{18}F]**12**, and MeOH/CH_2Cl_2=1:9 for [^{18}F]**15**, respectively). After the development the TLC plates were placed into sealable PE bags. The bags were closed and the distribution of radioactivity on the TLC plates was quantified using a γ-miniGita TLC-radioactivity scanner (Elysia-raytest GmbH, Germany). Radiochemical conversions (RCCs) were

calculated by dividing the radioactivity of the product spots by the total radioactivity deposited on the TLC plate. It was impossible to carry out TLC analysis for relatively volatile compounds like [^{18}F]**5** (b.p. 154-155 °C) and [^{18}F]**8** (b.p 185 °C)

Preparation of K_2CO_3/K2.2.2./KOTf stock solution: K_2CO_3 (0.6 mg, 4.3 µmol), K2.2.2 (2.7 mg, 7.2 µmol) and Et_4NOTf (50 mg, 180 µmol) were dissolved in *n*-BuOH (4 mL). The solution was stored under ambient conditions and consumed within 5 days.

Synthesis of [^{18}F]fluorobenzene (1) from pinacolyl phenylboronate (PhBPin) using different alcohols for washing of the resin (GP-1):

$^{18}F^-$ (100–200 MBq) was loaded onto a QMA cartridge. The resin was washed with the corresponding alcohol (2 mL) and flushed with air (5 mL). Afterwards $^{18}F^-$ was eluted with a solution of pyridine (30 µL), K_2CO_3 (50 µg, 0.36 µmol), phenylboronic acid pinacol ester (12.2 mg, 60 µmol) and $Cu(OTf)_2(py)_4$ (18 mg, 26.5 µmol) in DMF (500 µL). The eluate was heated at 110 °C for 20 min under air, cooled to room temperature, diluted with water (2 mL) and the resulting mixture was shaken vigorously for 30 s. Thereafter, RCC was determined by radio-HPLC.

Table S1: Influence of different alcohols as washing solution on recovery and RCC.

	Elution yield [%]	RCC [%]
MeOH	48	55
EtOH	55	60
***n*-PrOH**	39	72
***iso*-PrOH**	35	73
***n*-BuOH**	39	82
***tert*-BuOH**	41	77

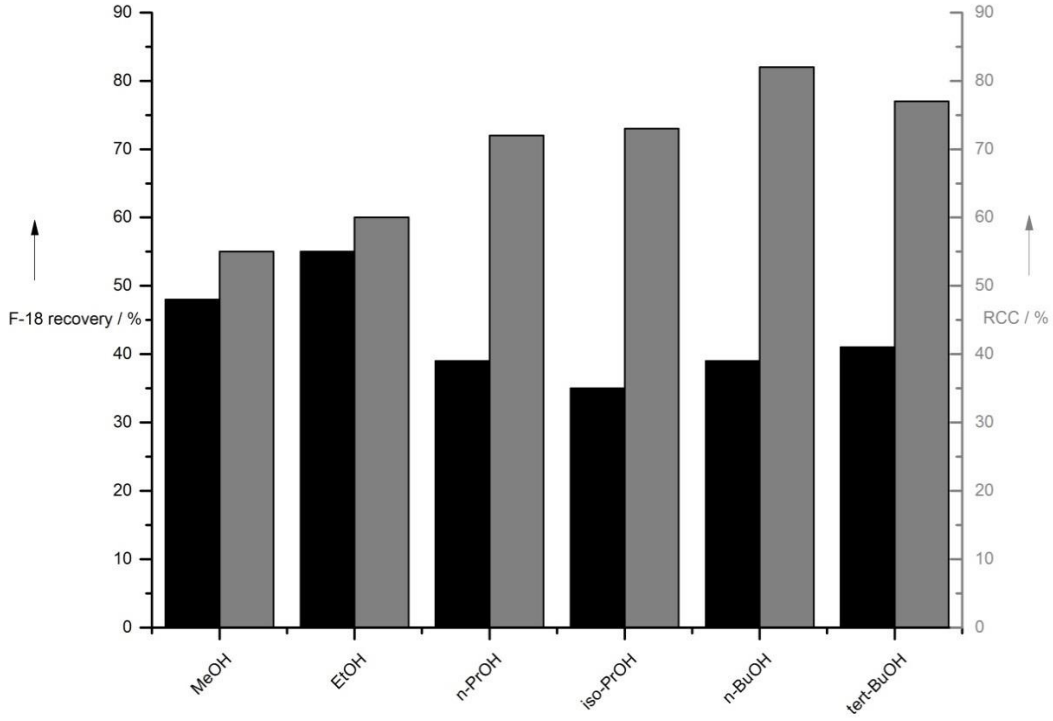


Figure S2: Influence of traces of alcohol on radiochemical conversion

Synthesis of [^{18}F]fluorobenzene (1) from PhBPin or phenylboronic acid [PhB(OH)$_2$] using alcohols as co-solvents (GP-2): $^{18}F^-$ (50–200 MBq) was eluted from a QMA cartridge with a solution of Et_4NHCO_3 (2.7 mg, 0.7 µmol) in the respective alcohol or water for comparison (400 µL). After that a solution of the PhBPin or PhB(OH)$_2$ precursor (60 µmol) and $Cu(OTf)_2(py)_4$ (18 mg, 26.5 µmol) in DMA (800 µL) was added. The reaction mixture was heated at 110 °C for 20 min under air. The mixture was cooled to room temperature, diluted with water (2 mL) and shaken vigorously for 30 s. Thereafter, RCC was determined by radio-HPLC.

Table S2: Alcohol screen.

	F-18 recovery [%]	**RCC from PhBPin [%]**	**RCC from PhB(OH)$_2$ [%]**
H$_2$O	99	0	0
MeOH	97	40	67
EtOH	91	59	90
n-PrOH	70	82	96

***iso*-PrOH**	71	62	86
***n*-BuOH**	80–95*	94	98
***iso*-BuOH**	84	89	93
***tert*-BuOH**	35	29	46
2-BuOH	43	82	85
1-OctOH	46	50	90
DMA w/o alcohol[a]		82	13

* If absolutely anhydrous *n*-BuOH was used for $^{18}F^-$ elution, radioactivity recovery from the anion exchange resin amounted to 72–76%. In this case, the addition of more Et_4NOTf (10–15 mg) to the elution solution increased $^{18}F^-$ recovery to > 80% without decrease in RCC. [a] Elution and synthesis was performed according to GP–3.

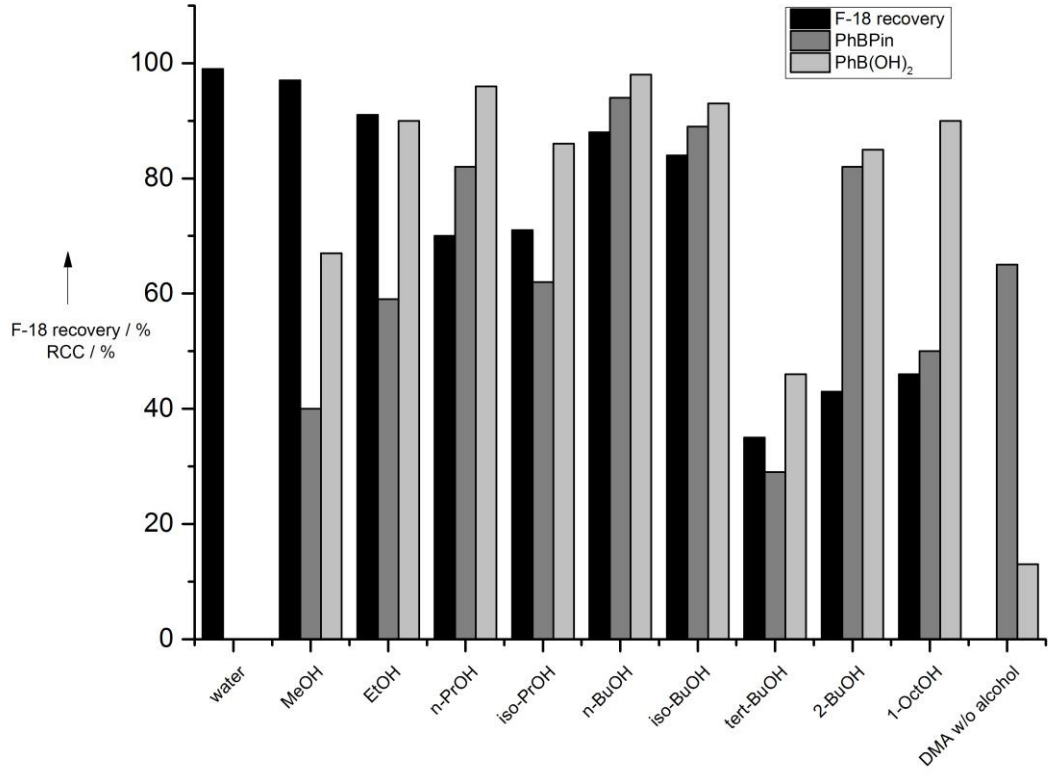


Figure S3: Influence of different alcohols and leaving groups on $^{18}F^-$ and RCC. For comparison RCCs in pure water and DMA are also shown.

Synthesis of [^{18}F]1 without alcohol (GP-3): Radiosynthesis starting from phenyl pinacolyl boronate was carried out according to Preshlock et al.[8]

For radiolabeling of $PhB(OH)_2$ [^{18}F]fluoride was eluted from the QMA cartridge with a solution of Et_4NHCO_3 (2.7 mg, 18 µmol) in MeOH (500 µL). The solvent was evaporated under Ar flow and reduced pressure at 70 °C. The residue was dissolved in a solution of $PhB(OH)_2$ (7.3 mg, 60 µmol) and $Cu(OTf)_2(py)_4$ (18 mg, 26.5 µmol) in DMA (1.2 mL). The mixture was stirred at

110 °C for 20 min, cooled to room temperature, diluted with water (2 mL) and shaken vigorously for 30 s. Thereafter, RCC was determined by radio-HPLC.

Effect of the alcohol content on RCC of [^{18}F]1 (GP-4): $^{18}F^-$ (50–150 MBq) was eluted from the QMA cartridge with a solution of Et_4NHCO_3 (2.7 mg, 18 µmol) in *n*-BuOH (for PhBPin)or EtOH [for $PhB(OH)_2$]. The necessary amount of the appropriate solution was added to a solution of the corresponding precursor (60 µmol) and $Cu(OTf)_2(py)_4$ (18 mg, 26.5 µmol) in DMA (1.2 mL total reaction volume). The reaction mixture was heated at 110 °C for 10 min under air atmosphere. The mixture was cooled to room temperature, diluted with water (2 mL) and vigorously shaken for 30 s. After that, RCC was determined by radio-HPLC.

Table S3: Dependency of alcohol proportion on RCC.

ROH [%]	PhBPin in *n*-BuOH/DMA	$Ph(BOH)_2$ in EtOH/DMA
0	65	13
5	97	98
10	97	96
20	> 99	96
30	> 99	97
40	> 99	91
50	98	92
60	97	76
70	90	70
80	82	70
90	40	66
100	8	45

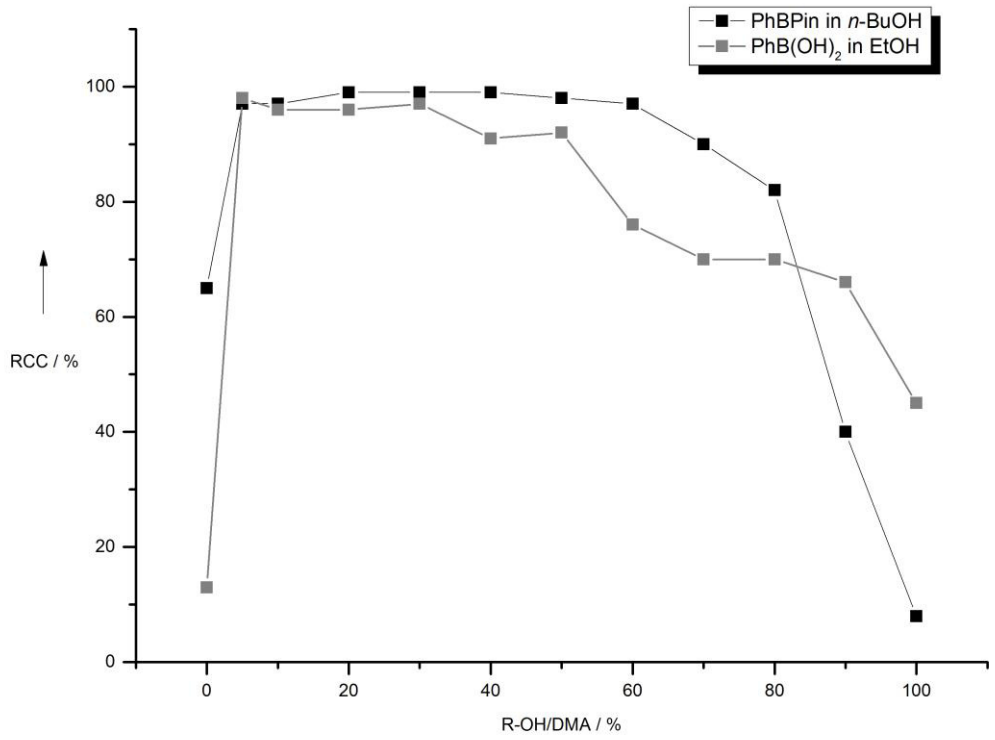


Figure S3: Dependency of alcohol proportion on RCC.

Base screen (GP-5): $^{18}F^-$ (100–200 MBq) was loaded onto a QMA cartridge. The resin was washed with *n*-BuOH (2 mL) and flushed with air (5 mL). Thereafter, $^{18}F^-$ was eluted from a QMA cartridge with a solution of the corresponding salt or salts (vide infra) in *n*-BuOH (400 µL). After that, a solution of $PhB(OH)_2$ (7.3 mg, 60 µmol) and $Cu(OTf)_2(py)_4$ (18 mg, 26.5 µmol) in DMA (800 µL) was added. The reaction mixture was heated at 110 °C for 20 min under air. The mixture was cooled to room temperature, diluted with water (2 mL) and shaken vigorously for 30 s. Thereafter, RCC was determined by radio-HPLC.

The following salts were tested:

K_2CO_3/K2.2.2 [(0.06 mg, 0.43 µmol) and (0.27 mg, 0.72 µmol), respectively];

Et_4NHCO_3 (2.7 mg, 18 µmol);

Et_4NOTf (5 mg, 18 µmol);

Et_4NHCO_3/Et_4NOTf [(2.7 mg, 18 µmol) and (10 mg, 35.8 µmol), respectively];

Bu_4POMes* (2 mg, 5.7 µmol).

*– Tetrabutylphosphonium methanesulfonate.

Table S4: Base screen.

Eluting agent	RCC [%]
K_2CO_3/K2.2.2	87

Et_4NHCO_3	99
Et_4NOTf	96
Et_4NHCO_3/Et_4NOTf	96
Bu_4POMes*	91

*– tetrabutylphosphonium methanesulfonate.

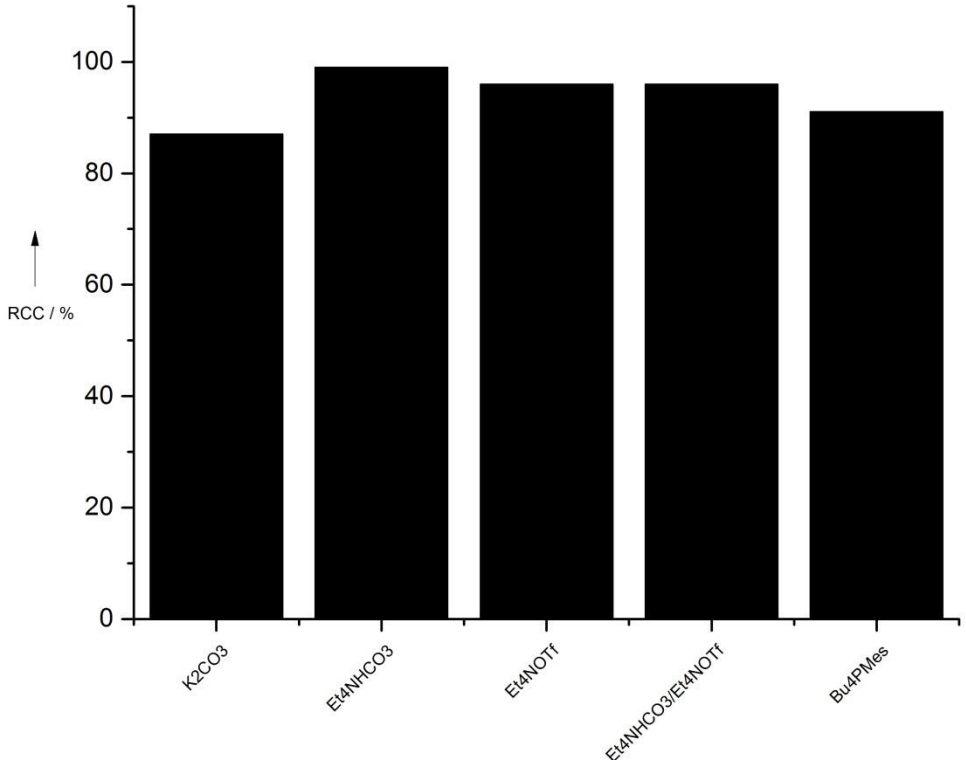


Figure S4: Dependency of different bases on the RCC.

Preparation of ^{18}F-labeled compounds from arylboronic acids, pinacolyl arylboronates and trialkylstannanes using *n*-BuOH as co-solvent (GP-6): $^{18}F^-$ (50–150 MBq) was eluted from a QMA cartridge with a solution of Et_4NHCO_3 (2.7 mg, 0.7 µmol), in *n*-BuOH (400 µL). A solution of the corresponding precursor (60 µmol) and $Cu(OTf)_2(py)_4$ (18 mg, 26.5 µmol) in DMA (800 µL) was added. The reaction mixture was heated at 110 °C for 20 min under air. The mixture was cooled to room temperature, diluted with water (2 mL) and vigorously shaken for 30 s. Thereafter, RCC was determined by radio-HPLC.

Effect of the aprotic solvent on the RCC of 4-[^{18}F]fluoroanisole ([^{18}F]3) (GP-7): $^{18}F^-$ (50–150 MBq) was eluted from a QMA cartridge with a solution of Et_4NOTf (5 mg, 18 µmol), K_2CO_3 (0.06 mg, 0.43 µmol) and K2.2.2. (0.27 mg, 0.72 µmol) in *n*-BuOH (400 µL). A solution of pinacolyl 4-methoxyphenyl boronate (14 mg, 60 µmol) and $Cu(OTf)_2(py)_4$ (18 mg, 26.5 µmol) in the corresponding aprotic solvent (800 µL) was added. The reaction mixture was heated at 110 °C

for 20 min under air. The mixture was cooled to room temperature, diluted with water (2 mL) and shaken vigorously for 30 s. Thereafter, RCC was determined by radio-HPLC.

Table S5: Dependency of RCC on aprotic solvent.

Solvent	RCC [%]
MeCN	2.5
DMSO	11
Sulfolane	50
DMF	50
NMP	77
DMA	87

NMP – *N*-Methylpyrrolidone

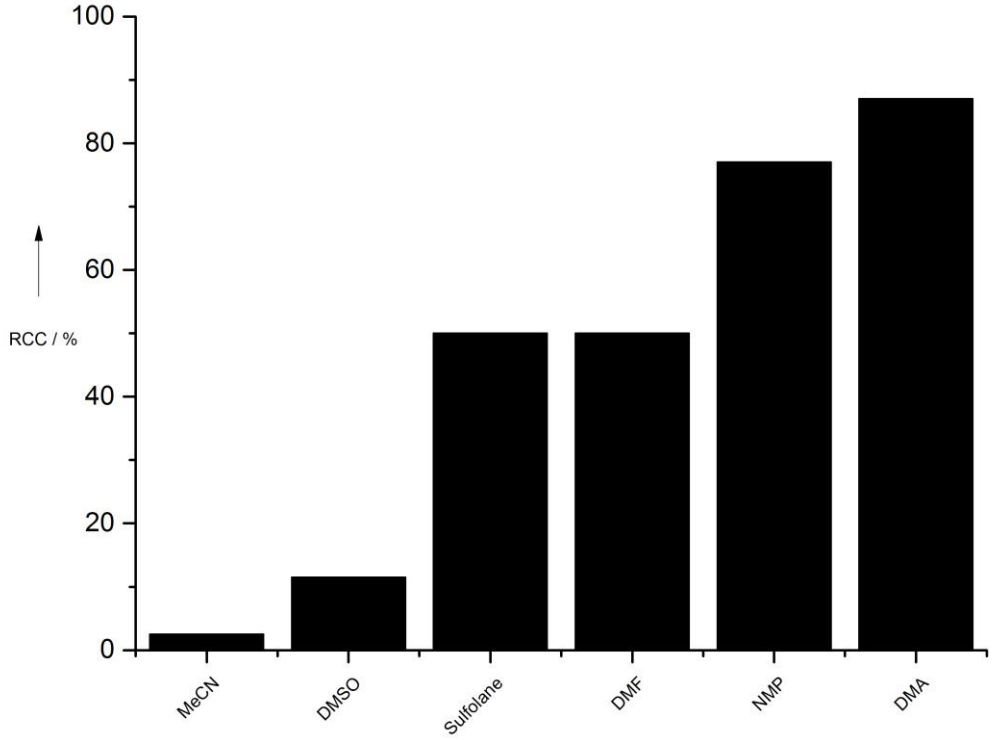


Figure S5: Dependency of RCC on aprotic solvent.

Dependence of the RCC of [^{18}F]1 on the reaction time (GP-8): $^{18}F^-$ (50–150 MBq) was eluted from a QMA cartridge with a solution of Et_4NHCO_3 (2.7 mg, 0.7 µmol), in *n*-BuOH (400 µL). A solution of PhBPin (12.2 mg, 60 µmol) and $Cu(OTf)_2(py)_4$ (18 mg, 26.5 µmol) in DMA (800 µL) was added. The reaction mixture was heated at 110 °C under air at a given time.. The mixture was cooled to room temperature, diluted with water (2 mL) and vigorously shaken for 30 s. Thereafter, RCC was determined by radio-HPLC.

Table S6: Kinetics of radiolabeling

t [min]	RCC [%]
5	85
10	91
20	89

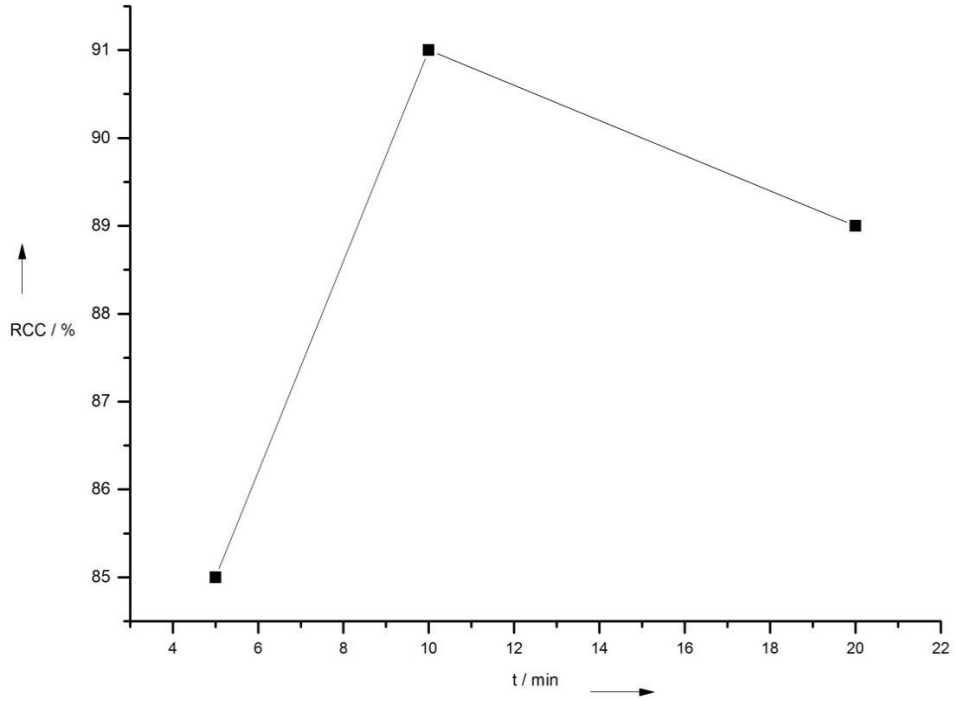


Figure S6: Kinetics of radiolabeling.

Effect of different Cu-catalysts on a RCC of [^{18}F]1 (GP-9):

$^{18}F^-$ (50–150 MBq) was eluted from a QMA cartridge with a solution of of Et_4NOTf (5 mg, 18 µmol), K_2CO_3 (0.06 mg, 0.43 µmol) and K2.2.2. (0.27 mg, 0.72 µmol) in *n*-BuOH (400 µL). A solution of PhBPin (12.2 mg, 60 µmol) and the corresponding Cu-catalyst (26.5 µmol) in DMA (800 µL) was added. The reaction mixture was heated at 110 °C for 20 min under air. The mixture was cooled to room temperature, diluted with water (2 mL) and shaken vigorously for 30 s. Thereafter, RCC was determined by radio-HPLC.

Table S7: Dependency of different copper sources on RCC of [^{18}F]1

Cu-catalyst	RCC [%]
$Cu(OTf)_2(Py)_4$	76
$Cu(OTs)_2(Py)_4$	25
$Cu(OMs)_2(Py)_4$	4

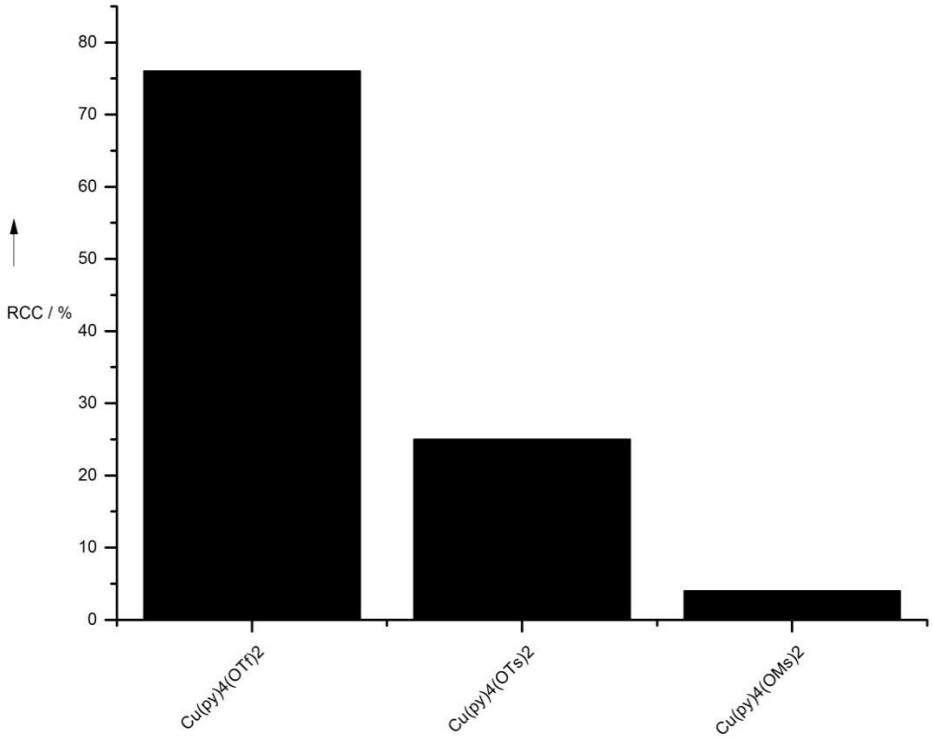
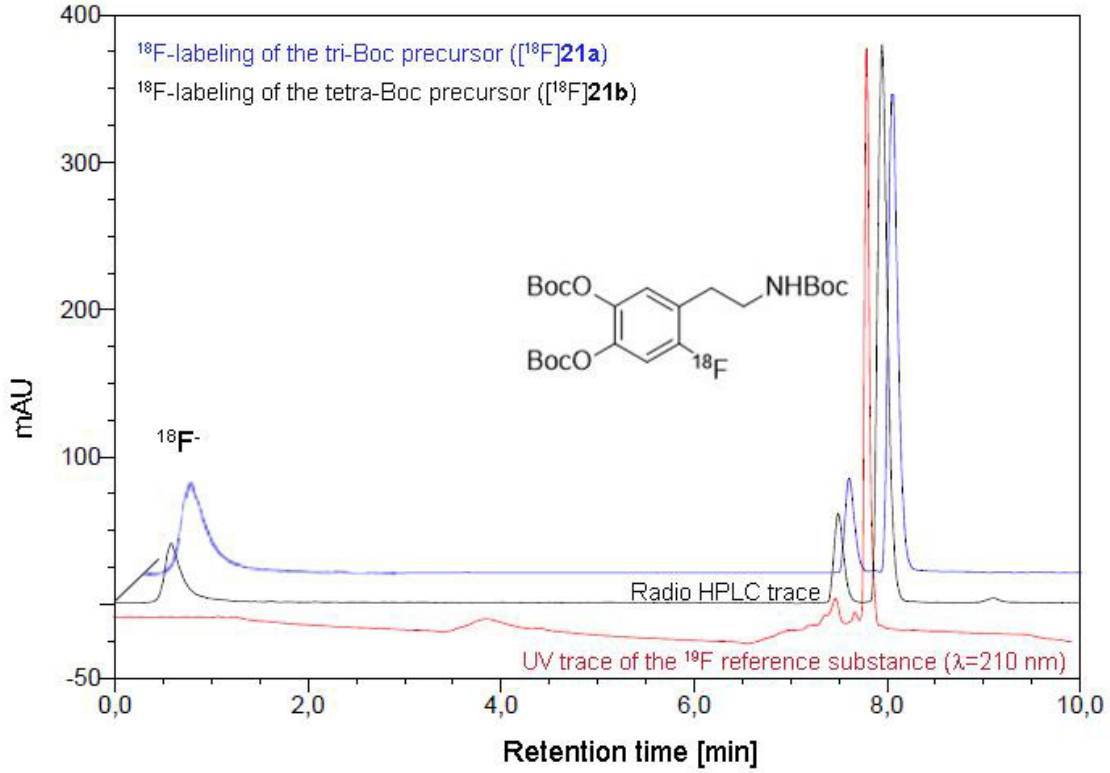
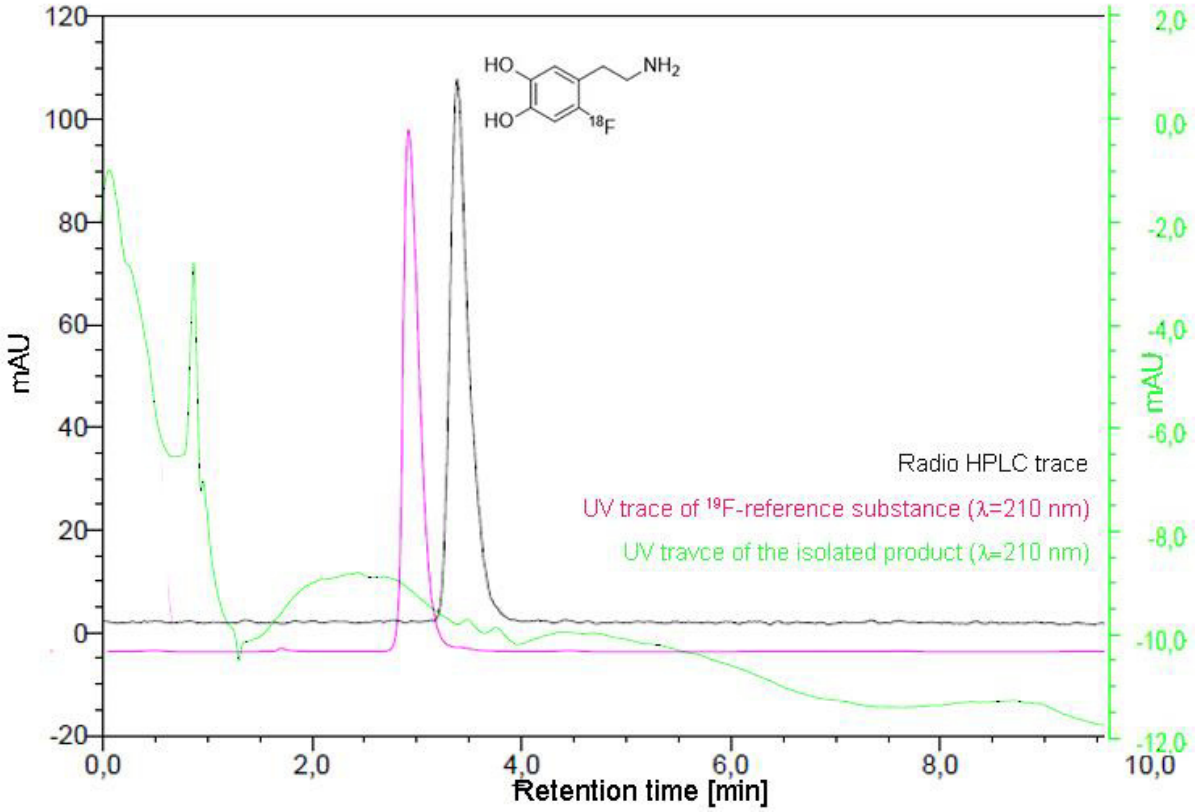


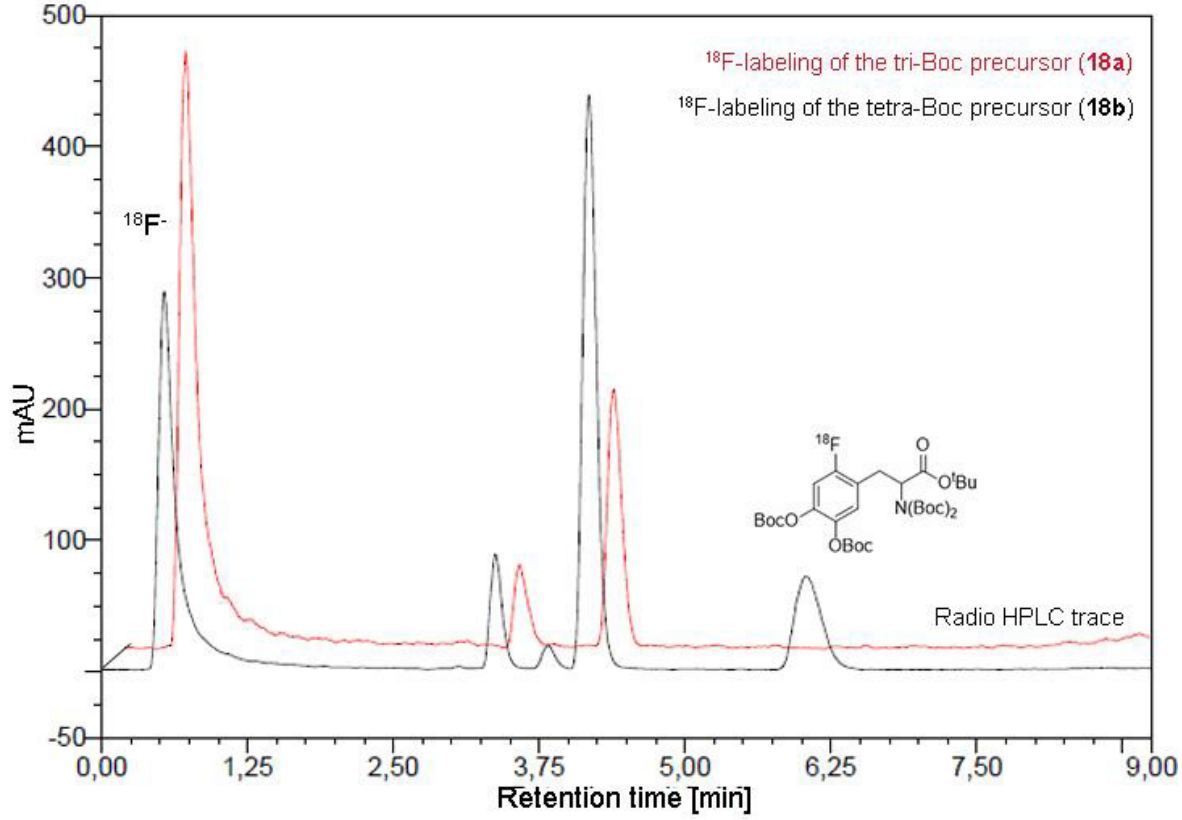
Figure S7: Dependency of different copper sources on RCC of [^{18}F]1.

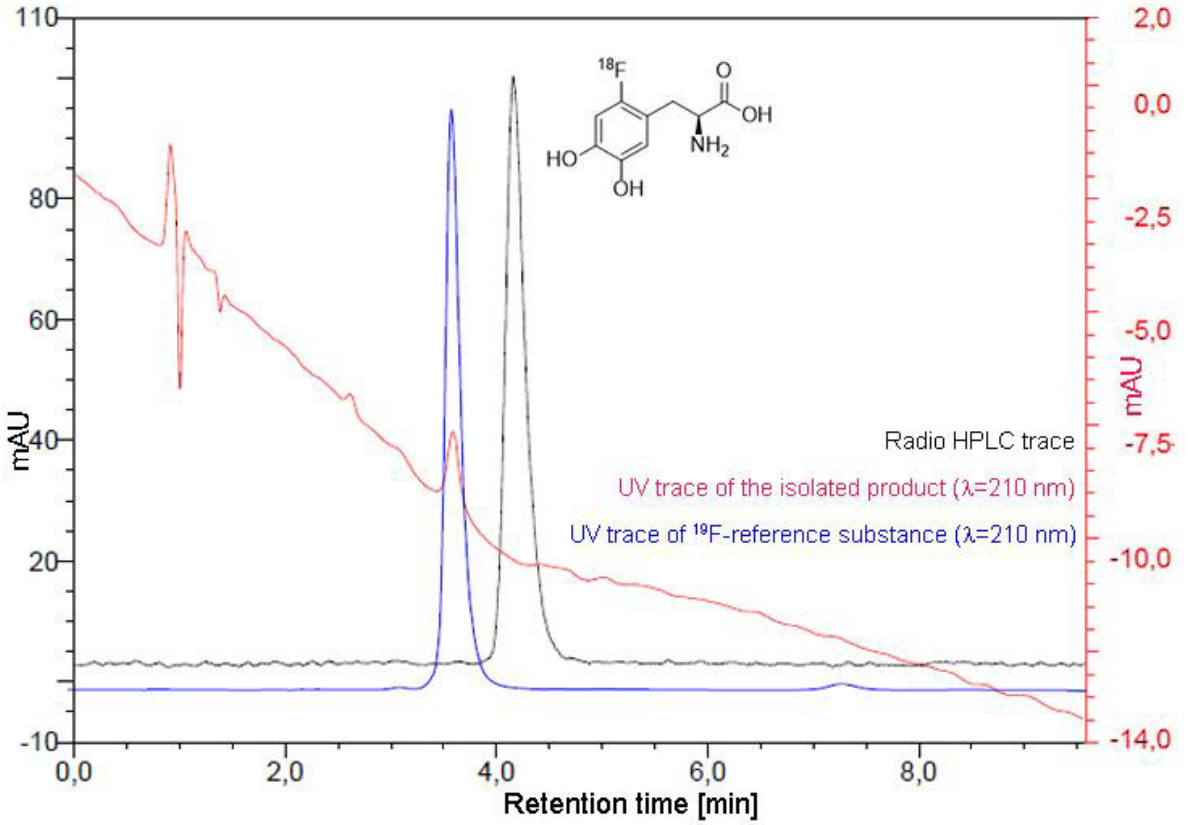
Preparation of 6-[^{18}F]FDA ([^{18}F]20) and 6-[^{18}F]FDOPA ([^{18}F]23): $^{18}F^-$ (1000–2000 MBq) was eluted from a QMA cartridge with a solution of Et_4NHCO_3 (2.7 mg, 0.7 µmol), in *n*-BuOH (400 µL). A solution of the respective precursor (**18a,b** for [^{18}F]FDA, **21a,b** for [^{18}F]FDOPA) (60 µmol) and $Cu(OTf)_2(py)_4$ (36 mg, 53 µmol) in DMA (800 µL) was added. The reaction mixture was heated at 110 °C for 10 min under air. Afterwards, the reaction mixture was quenched with water (4 mL) and passed through a C_{18} cartridge (500 mg), preconditioned with EtOH (1 mL) and water (10 mL). The cartridge was washed with water (10 mL) and dried with air. The protected product was eluated with MeOH (2 mL). KHF_2 (4.7 mg, 60 µmol) in water (50 µL) was added to the methanolic solution. The reaction mixture was stirred for 1 min at room temperature. H_2O (8 mL) was added and the resulting solution was immediately passed through a C_{18} cartridge (500 mg). The cartridge was washed with water (10 mL), dried with air and the protected ^{18}F-labeled intermediate was eluted with acetone (2 mL). Acetone was evaporated, 12 M HCl (300 µL) was added to the residue and the reaction mixture was stirred at 130 °C for 10 min. Thereafter, acetone (2 mL) was added and the resulting solution was concentrated to dryness at 80 °C under reduced pressure. The crude radiolabeled product was taken up into 4% EtOH in 0.02 M NaH_2PO_4 (500 µL, pH 2.5) and purified by HPLC to afford the desired tracer in a solution ready for injection. HPLC conditions for purification: Synergi 4u Hydro-RP 150×21.2 mm; eluent: 0.02 M NaH_2PO_4 (pH 2.5); flow rate: 9 mL/min; t_R=21.7 min (6-[^{18}F]FDA); t_R=16.0 min (6-[^{18}F]FDOPA).

Analytical HPLC conditions: column: Synergy 4µm Hydro-RP 150×4.6 mm; eluent: 4% EtOH in 0.02 M NaH_2PO_4 (pH 2.5); flow rate: 1.5 mL/min; t_R=3.6 min (6-[^{18}F]FDA), t_R=3.2 min (6-[^{18}F]FDOPA).



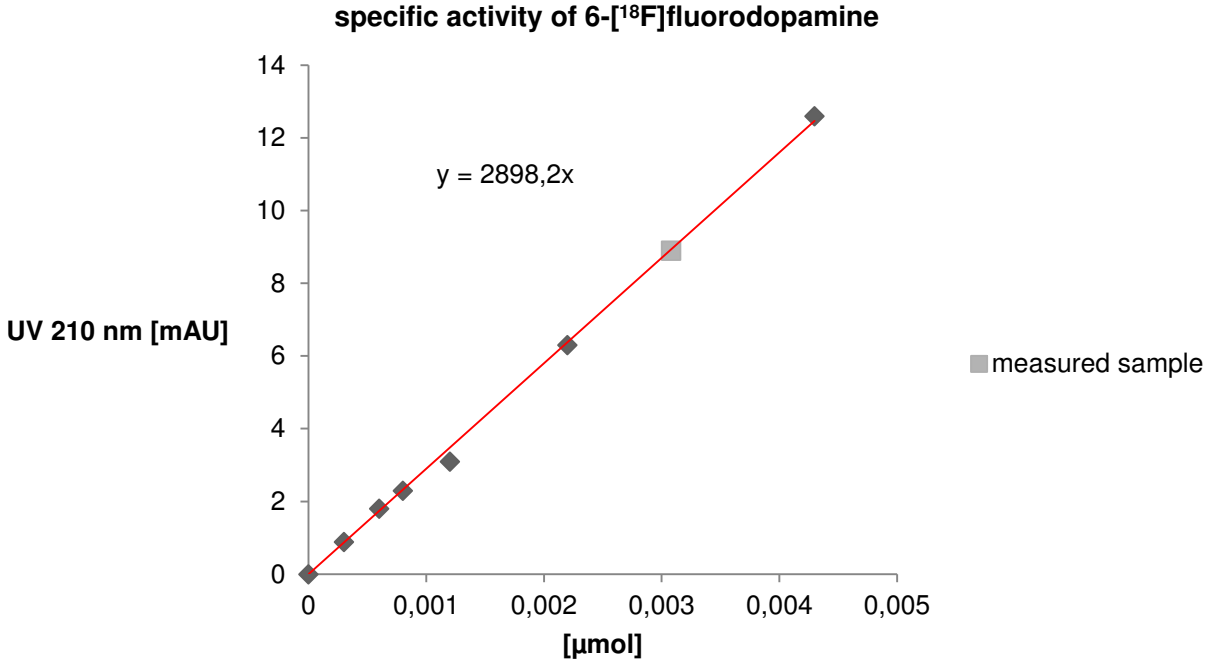


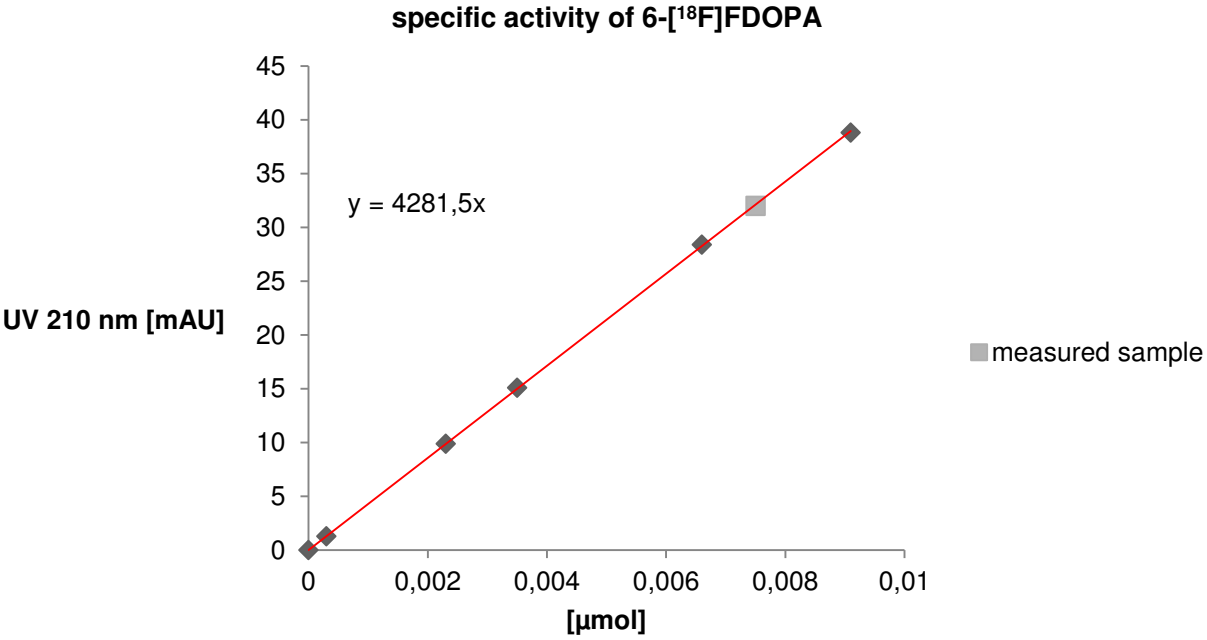




Specific activity calculation: The specific activities (GBq/μmol) were calculated by dividing the radioactivity of the $^{18}F^-$labeled product by the amount of the unlabeled tracer determined from the peak area in a UV-HPLC chromatograms (λ=210 nm). The amounts of unlabeled compounds were determined from the UVabsorbance/concentration calibration curve. The solutions of 6-

[^{18}F]FDOPA and 6-[^{18}F]FDA obtained after HPLC purification were concentrated under reduced pressure, the residues were redissolved in small amounts of 4% EtOH in 0.02 M sodium phosphate buffer (300 µL, pH 2.5). The resulting solutions were completely injected into the HPLC system. The peak area was determined and the amount of cold label was calculated according to the calibration curve. The specific activity of 6-[^{18}F]FDA (260 MBq) was determined to 39 GBq/µmol. The specific activity of 6-[^{18}F]FDOPA (278 MBq) a 37 GBq/µmol.

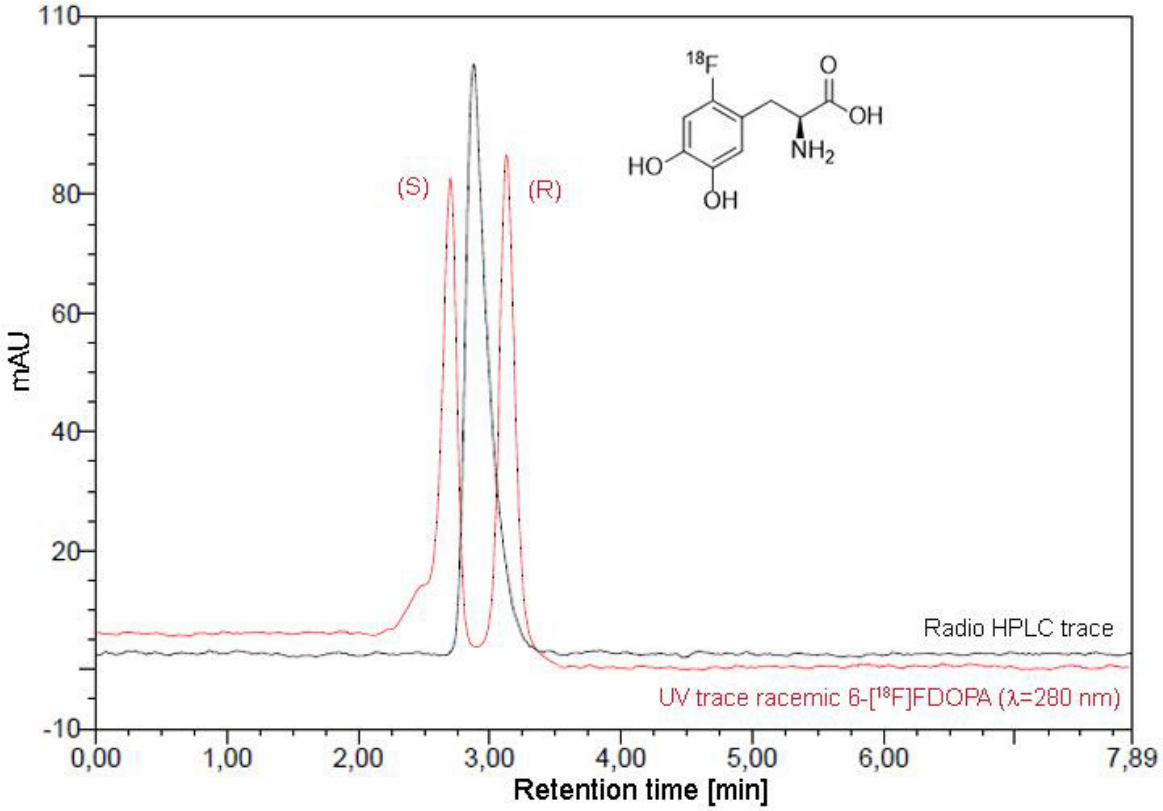




Determination of the enantiomeric purity of [^{18}F]FDOPA

An aliquot (20 µL) of the isolated 6-[^{18}F]FDOPA was analysed by HPLC.

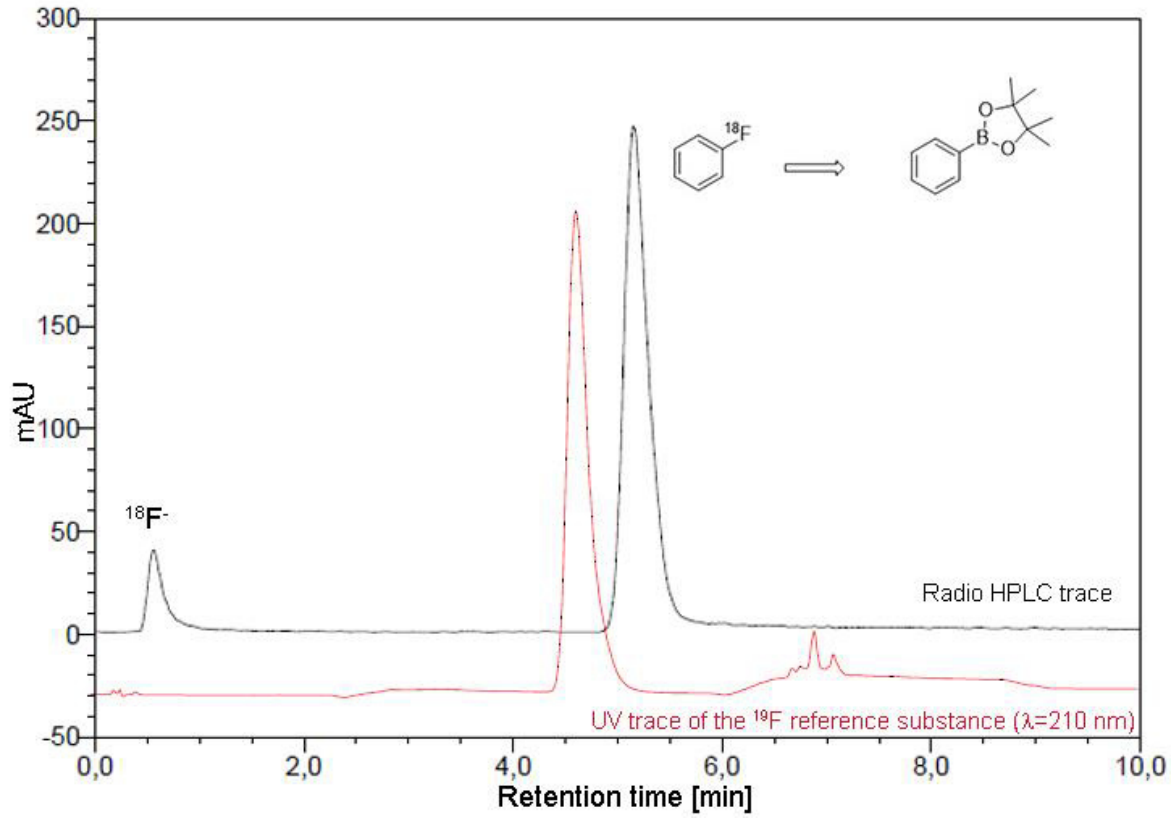
Conditions: column: Astec® CHIROBIOTIC® T 250×4.6 mm 5 µm (Supelco); eluent EtOH in 0.02 M NaH_2PO_4 (pH 2.5) (v/v 20:80); flow rate: 1.5 mL/min; t_R: 2.8 min [(*S*)-isomer t_R=2.67 min; (*R*)-isomer t_R=2.99 min].

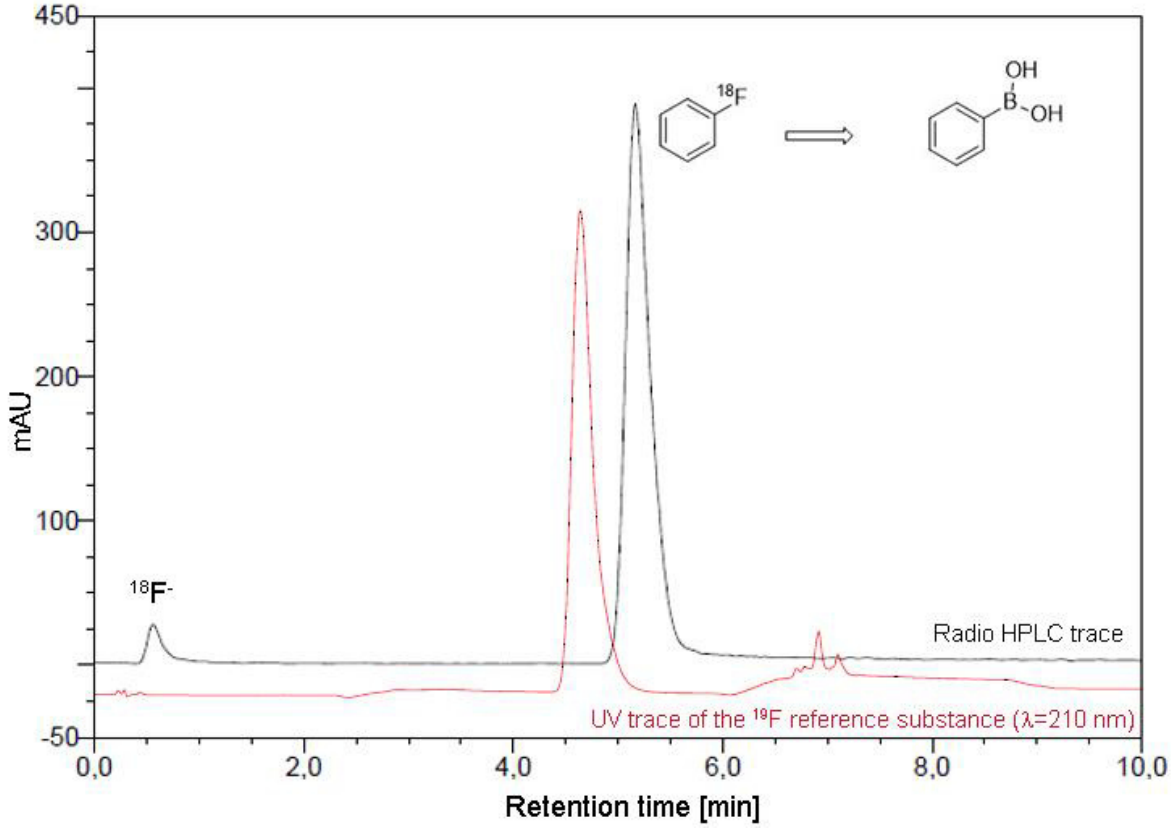


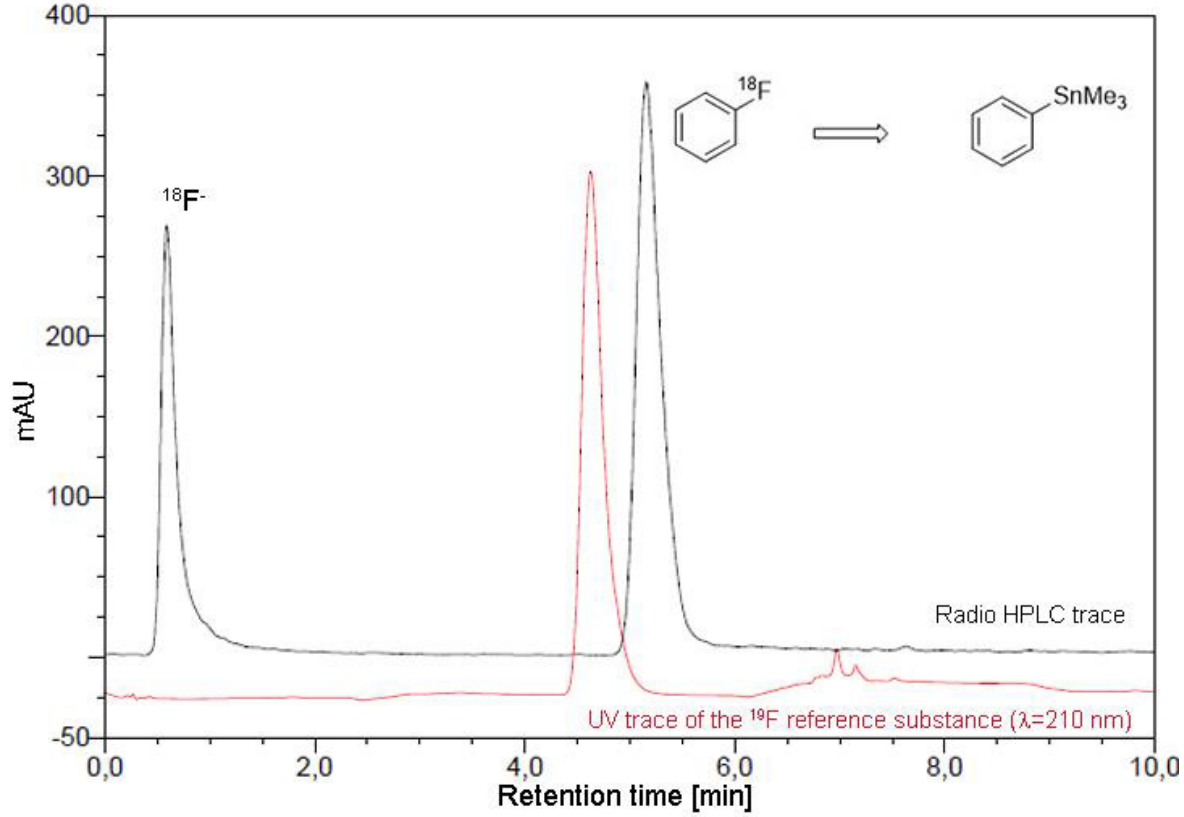
HPLC Chromatrogramms

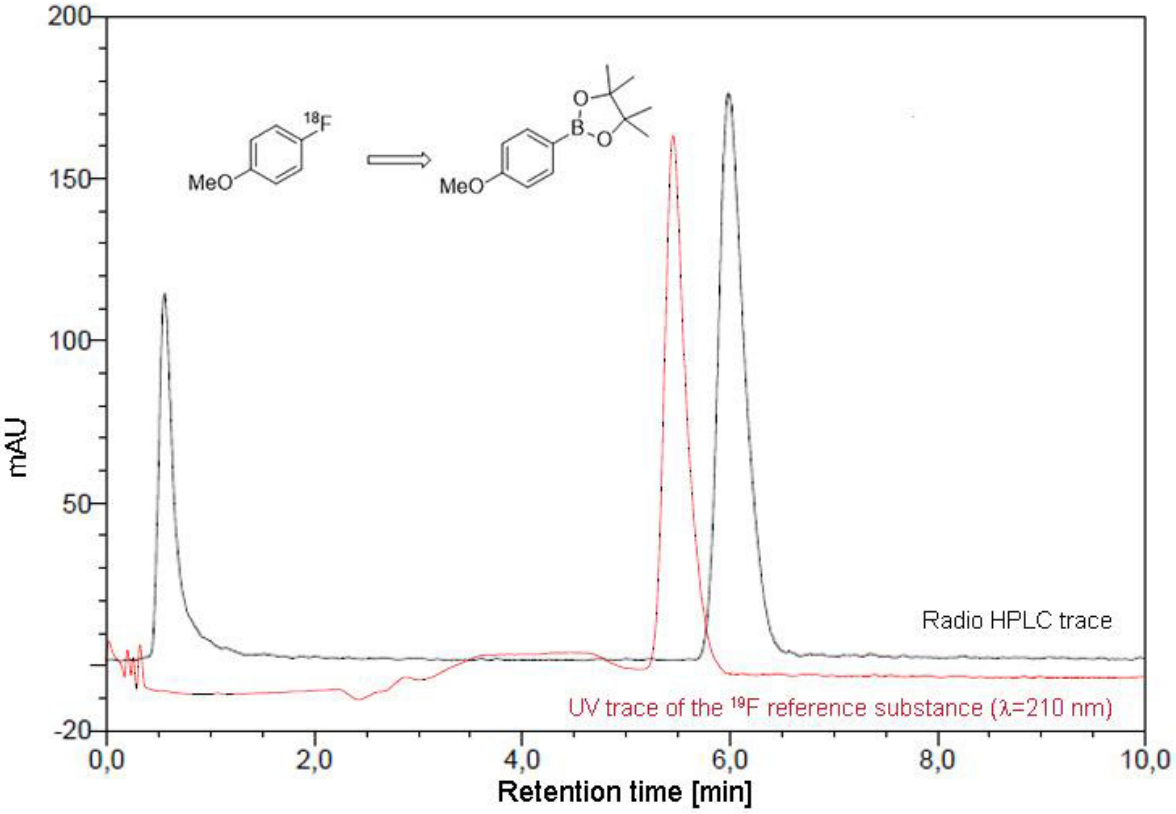
		t_R [min]
[^{18}F]1	[^{18}F]Fluorobenzene	5.2
[^{18}F]2	4-[^{18}F]Fluoroanisole	6.1
[^{18}F]3	2-[^{18}F]Fluoroanisole	5.2
[^{18}F]4	[^{18}F]Fluoronaphtalene	7.6
[^{18}F]5	3,4-Dimethoxy-[^{18}F]fluorobenzene	4.7
[^{18}F]6	4-[^{18}F]Fluorobenzaldehyde	3.5
[^{18}F]7	3-[^{18}F]Fluorobenzaldehyde	3.6
[^{18}F]8	4-[^{18}F]Fluorophenol	2.9
[^{18}F]9	4-[^{18}F]Fluoro-2-methylphenol	4.1
[^{18}F]10	2-[^{18}F]Fluoroaniline	2.4
[^{18}F]11	3-[^{18}F]Fluoroaniline	2.5
[^{18}F]12	4-[^{18}F]Fluoroindole	6.8
[^{18}F]13	5-[^{18}F]Fluoroindole	3.1[a]
[^{18}F]14	6-[^{18}F]Fluoroindole	3.1[a]
[^{18}F]15	*N*-Acetyl 4-[^{18}F]fluorotryptophan methyl ester	3.0[a]
[^{18}F]16	*N*-Boc-6-[^{18}F]fluorotryptophan ethyl ester	7.5
[^{18}F]17	[^{18}F]F-DPA	3.2[a]
[^{18}F]26	[^{18}F]DAA1106	3.3[a]

[a] HPLC-conditions: column: Chromolith SpeedROD®, 50×4.6 mm (Merck Millipore); gradient: 0–2 min: 5% MeCN, 2–2.5 min: 5→70% MeCN, 2.5–7 min: 70% MeCN, flow rate: 3.0 mL/min.






400
300
200
100
-50
mAU
$^{18}F^-$
^{18}F
$SnMe_3$
Radio HPLC trace
UV trace of the ^{19}F reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]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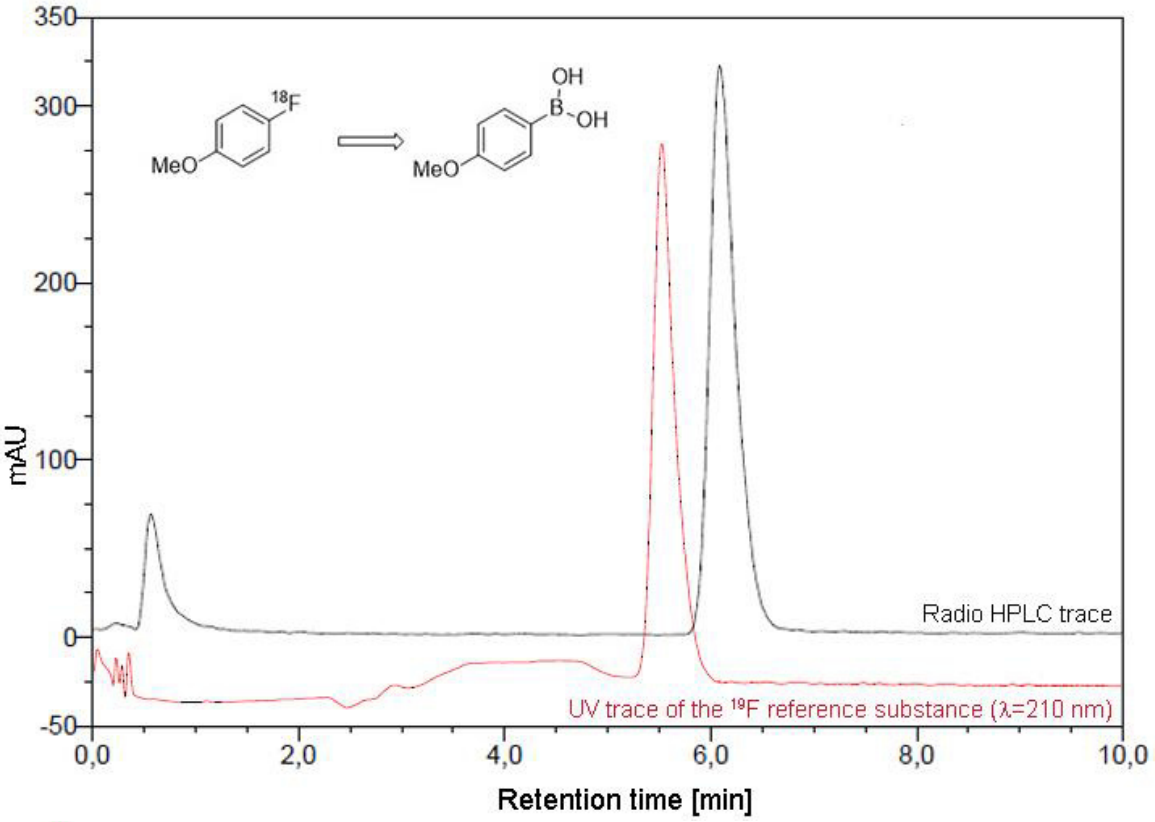
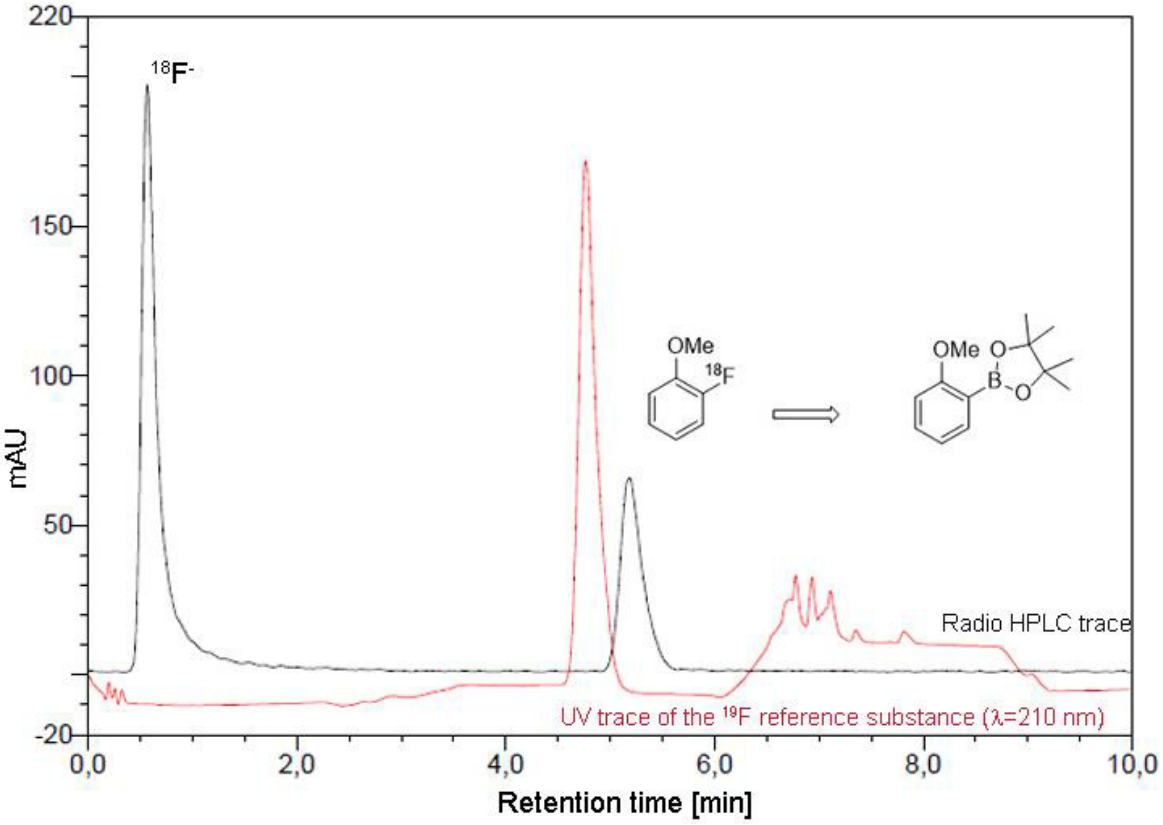
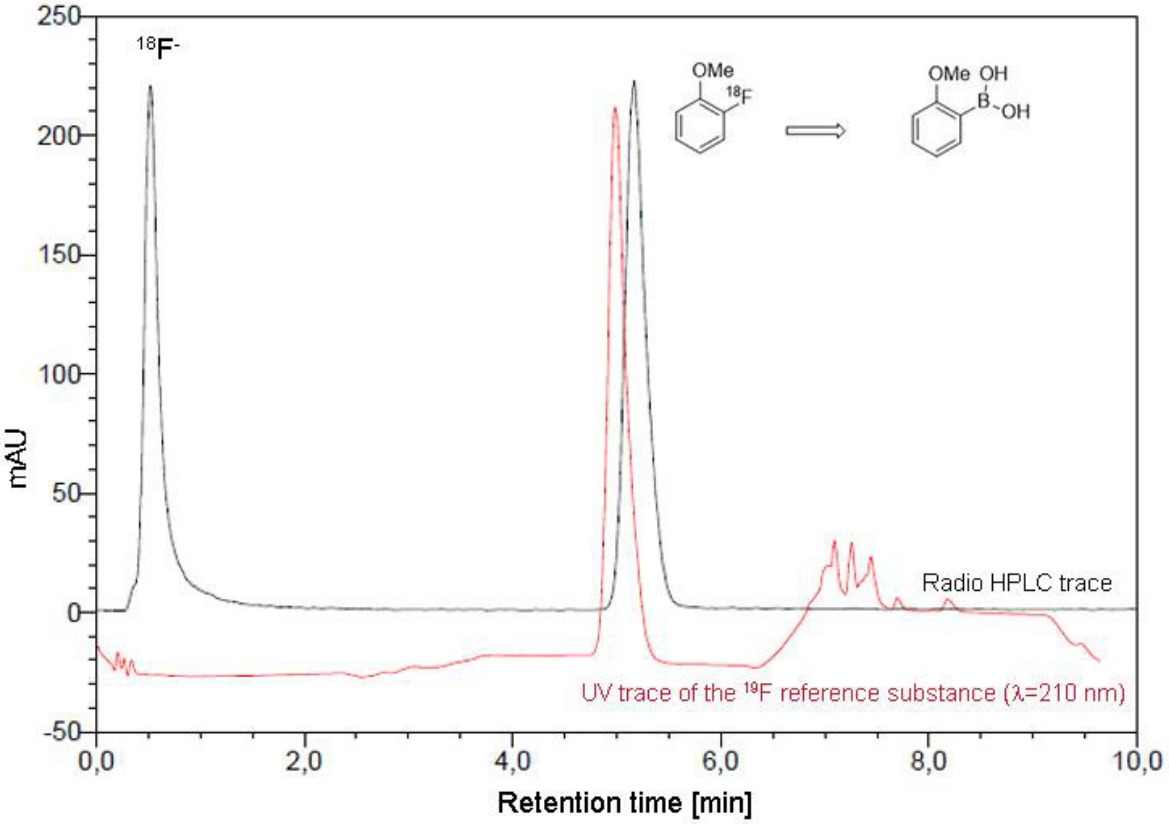
150
100
50
-20
mAU
^{18}F
MeO
MeO
O
B
O
Radio HPLC trace
UV trace of the ^{19}F reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]


350
300
200
100
0
-50
mAU
MeO
18F
OH
B
OH
Radio HPLC trace
UV trace of the 19F reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]


220
150
100
50
-20
mAU
18F-
OMe
18F
OMe
O
B
O
Radio HPLC trace
UV trace of the 19F reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]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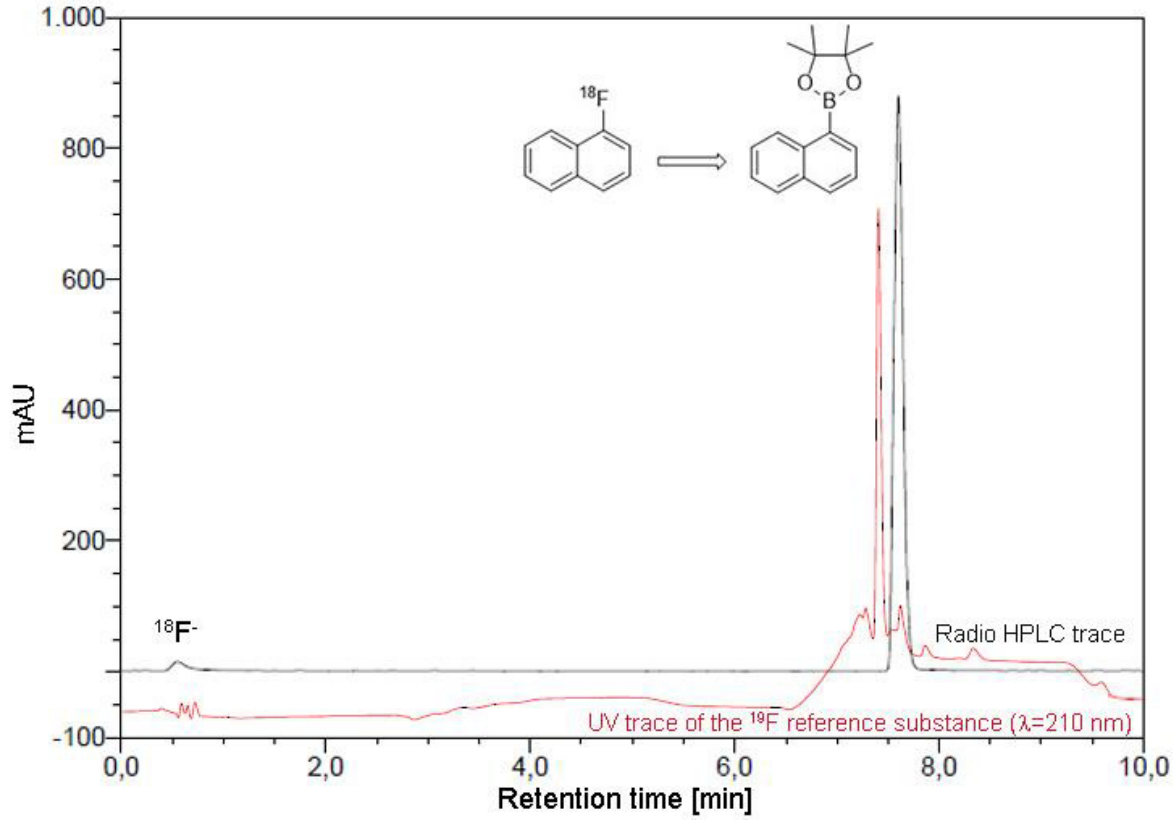
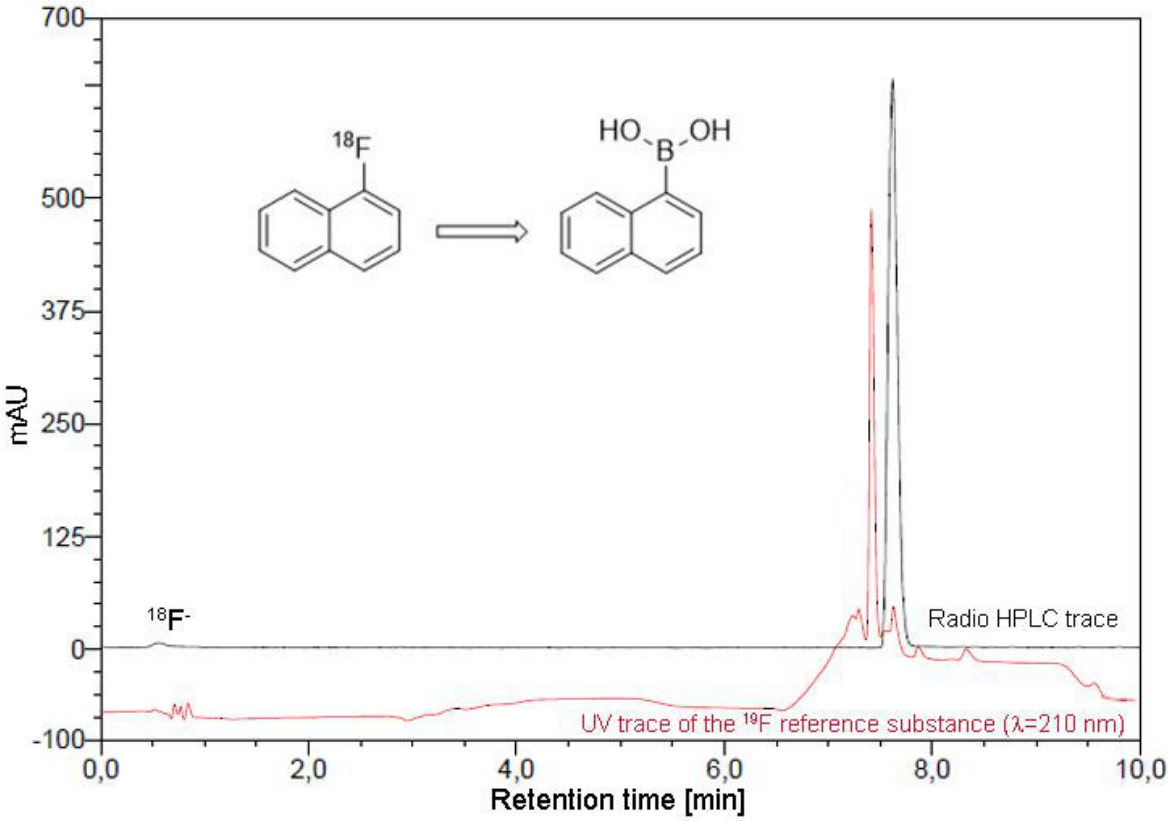
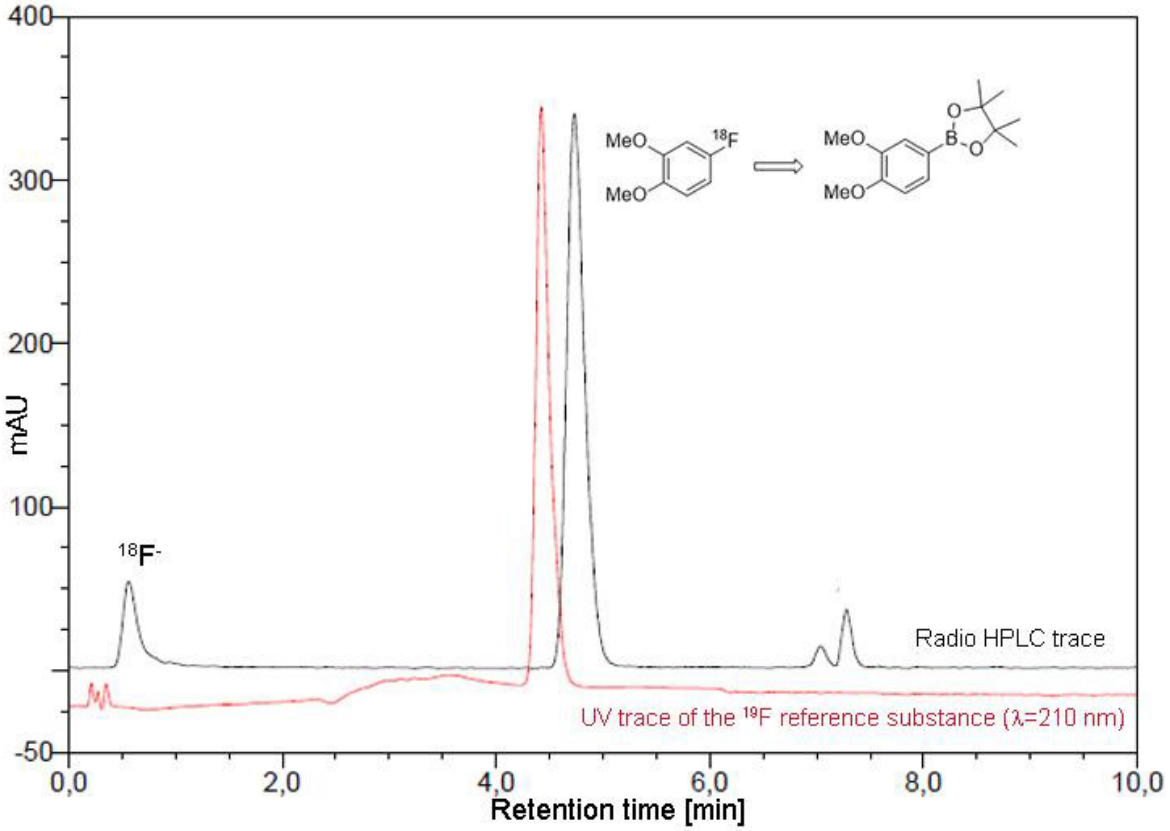
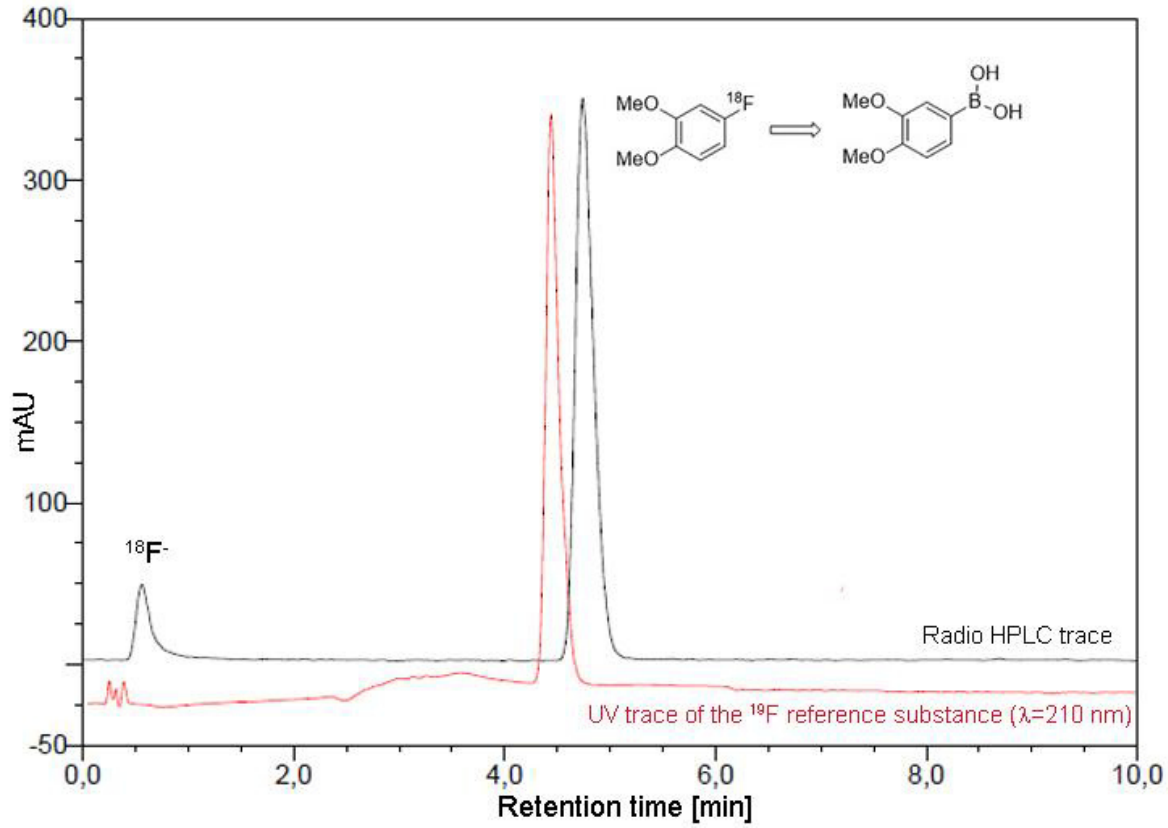
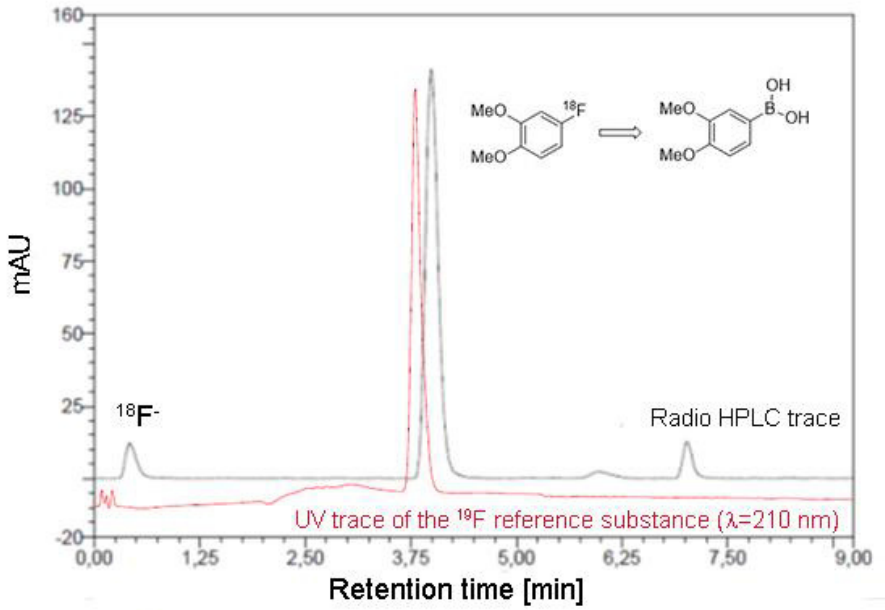
200
150
100
50
0
-50
mAU
$^{18}F^-$
OMe
^{18}F
OMe OH
B
OH
Radio HPLC trace
UV trace of the ^{19}F reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]


1.000
800
600
400
200
-100
mAU
^{18}F
O
O
B
$^{18}F^-$
Radio HPLC trace
UV trace of the ^{19}F reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]


700
500
375
250
125
0
-100
mAU
^{18}F
HO
B
OH
^{18}F-
Radio HPLC trace
UV trace of the ^{19}F reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]


400
300
200
100
-50
mAU
MeO
MeO
^{18}F
MeO
MeO
B
O
O
^{18}F-
Radio HPLC trace
UV trace of the ^{19}F reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]





Nr.	Retention time min	Intensity mAU	Integral %
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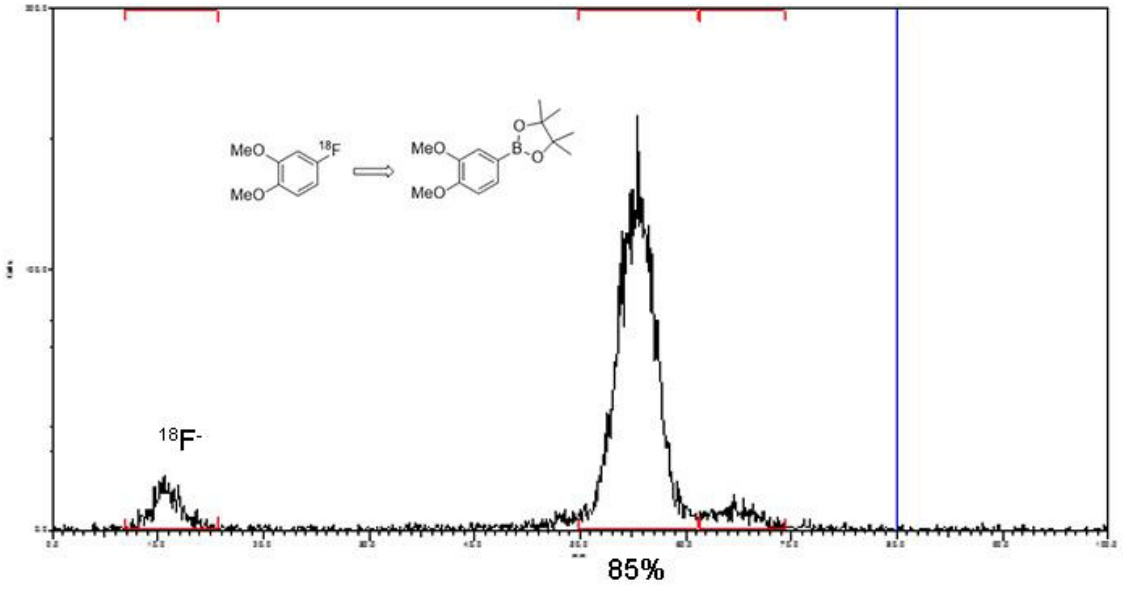
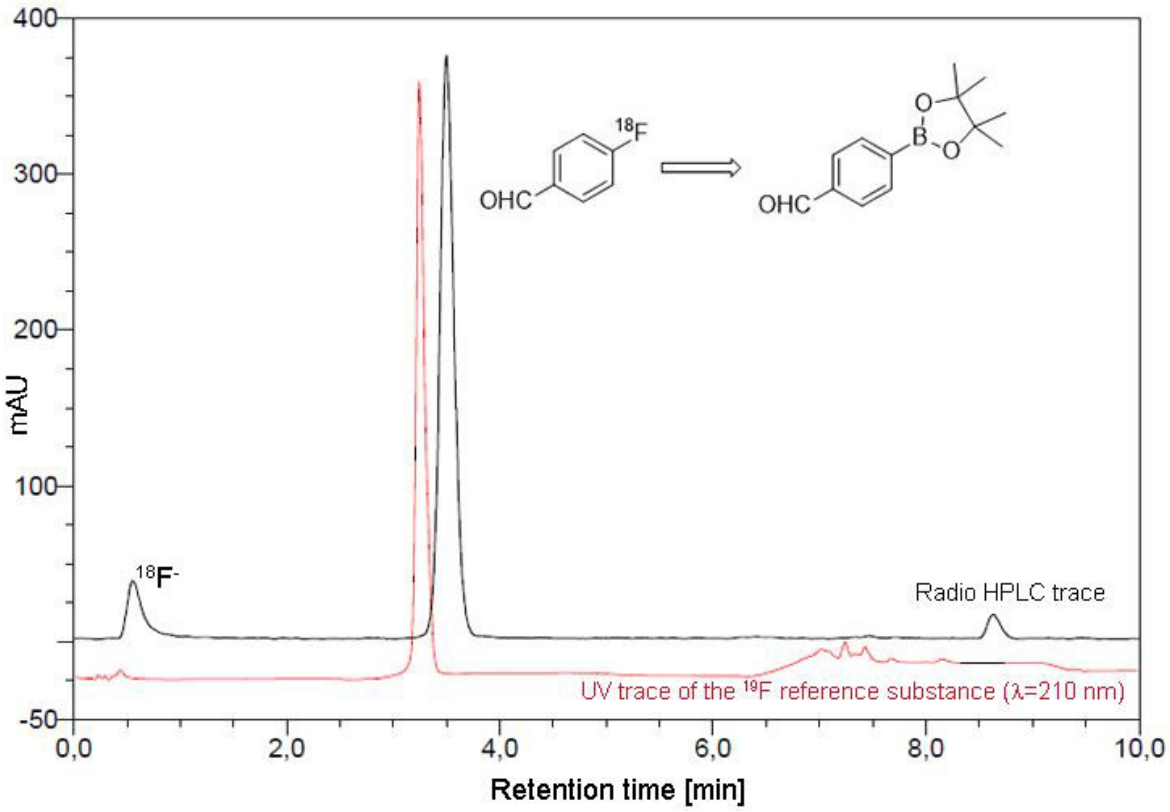
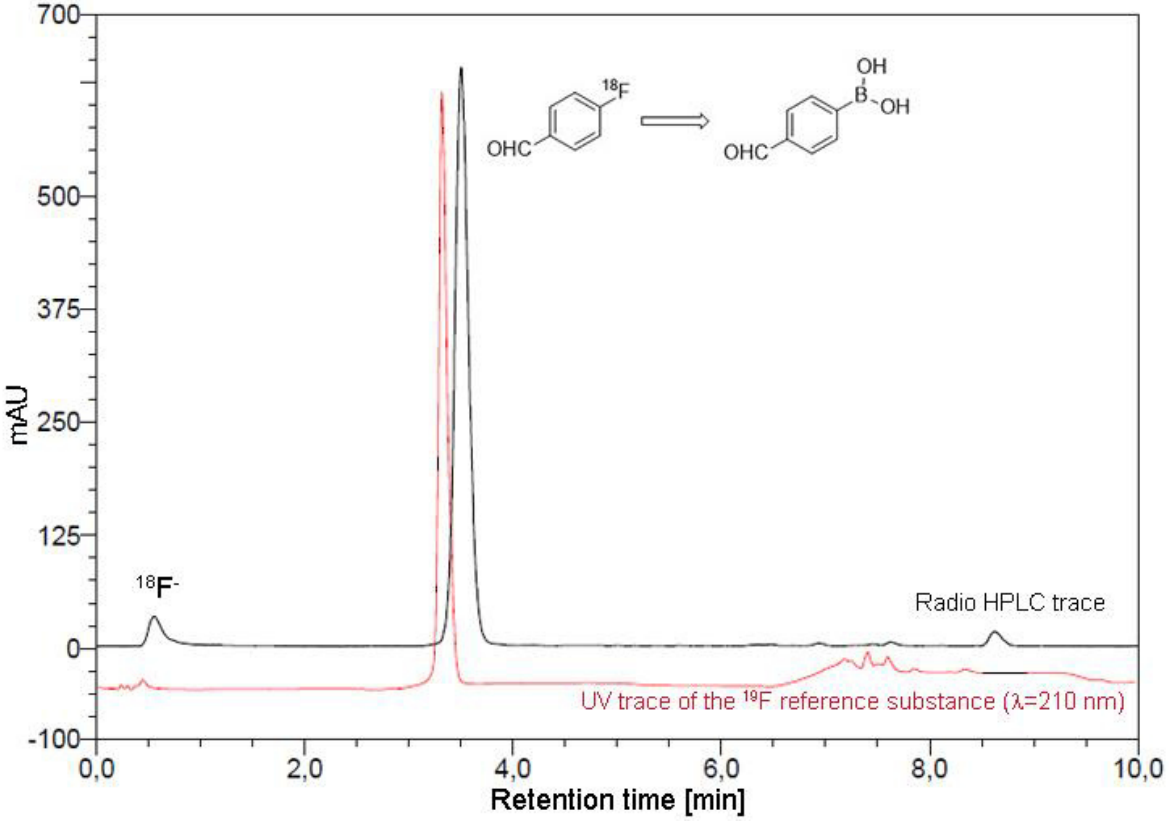
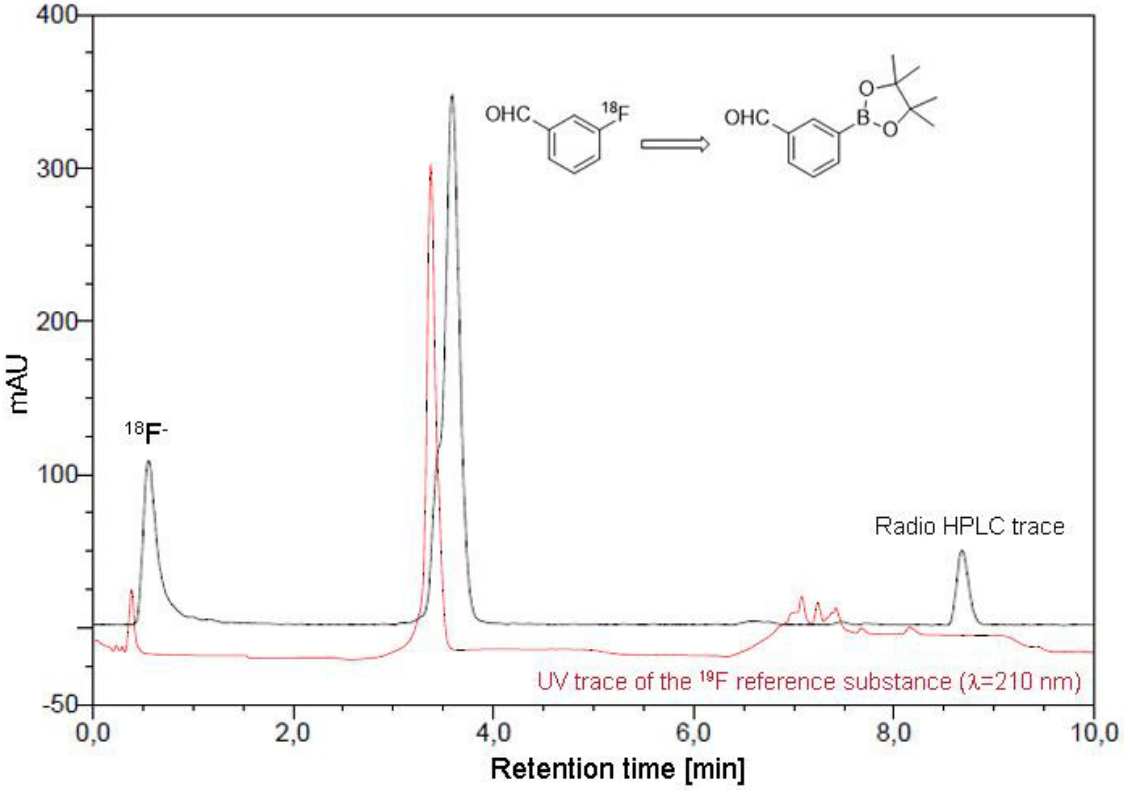
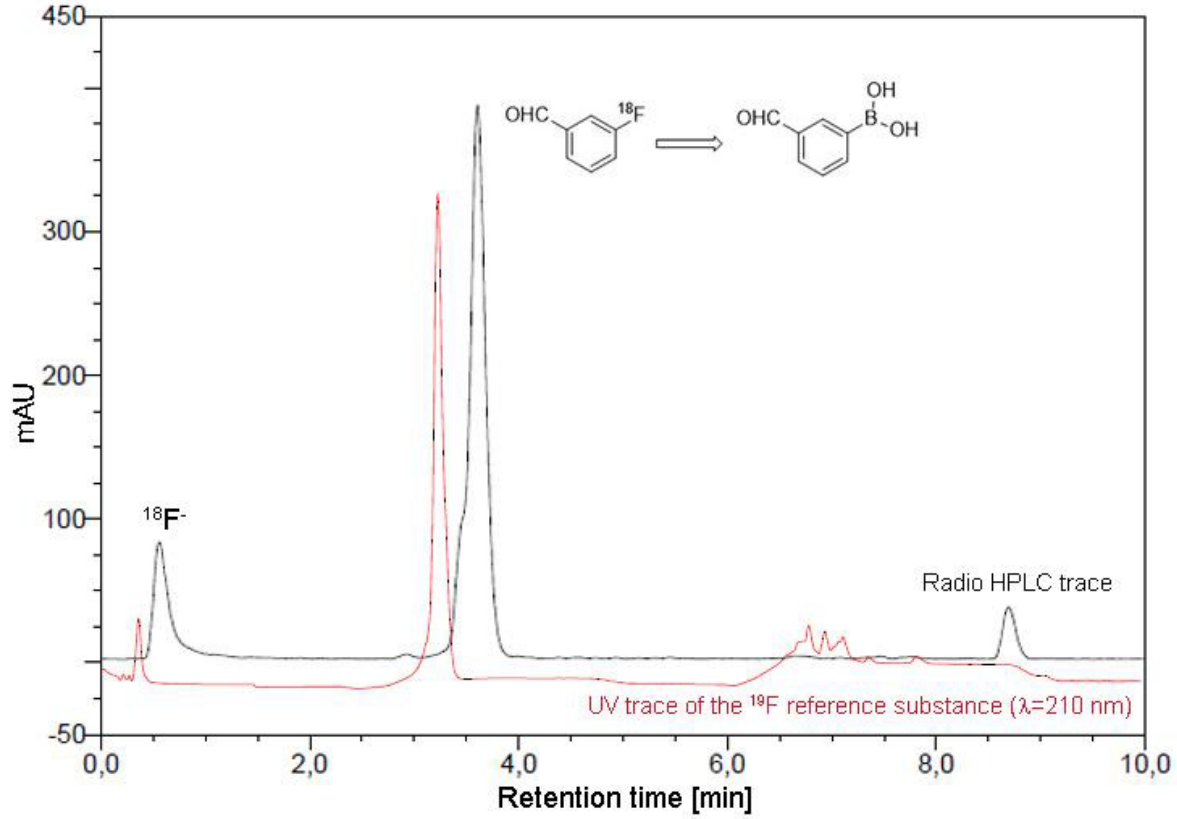
2	3,99	141,081	86,61
3	5,97	2,143	1,80
4	7,02	12,490	5,55
Total:		167,772	100,00

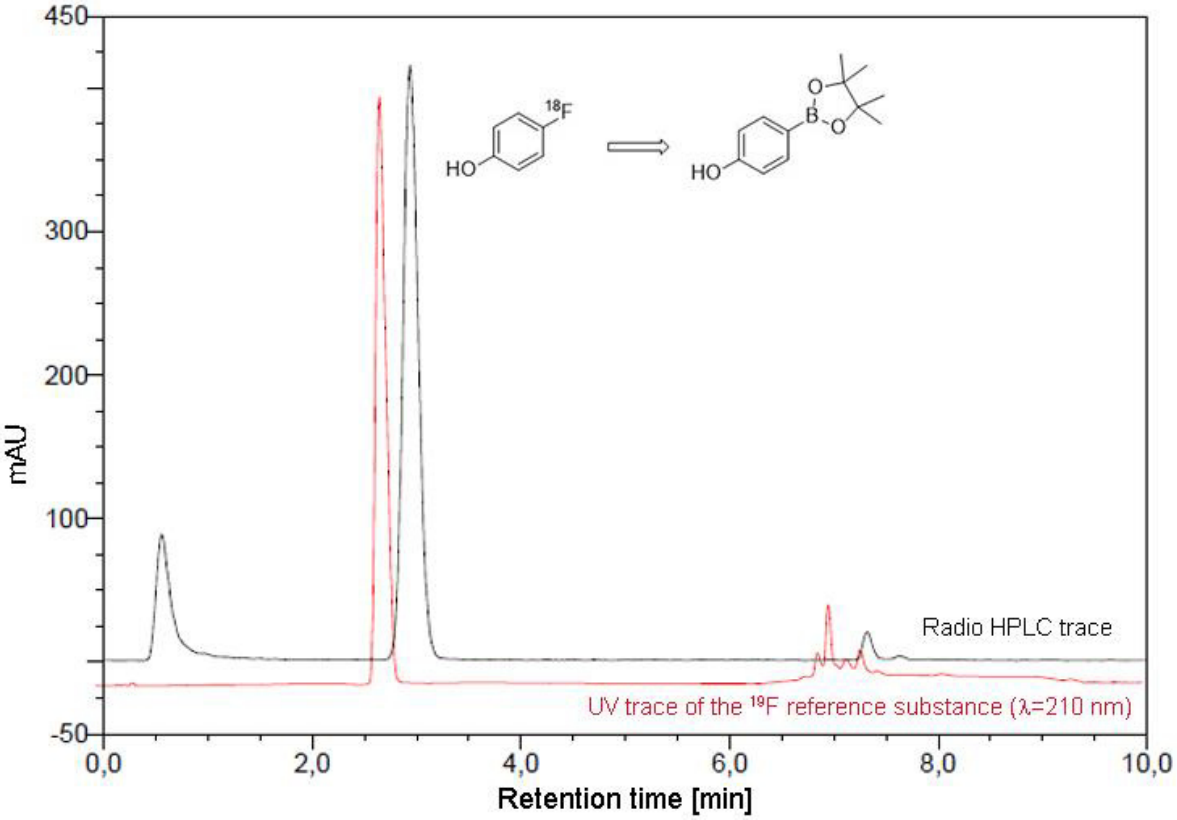

MeO
MeO
18F
MeO
MeO
O
B
O
18F-
85%

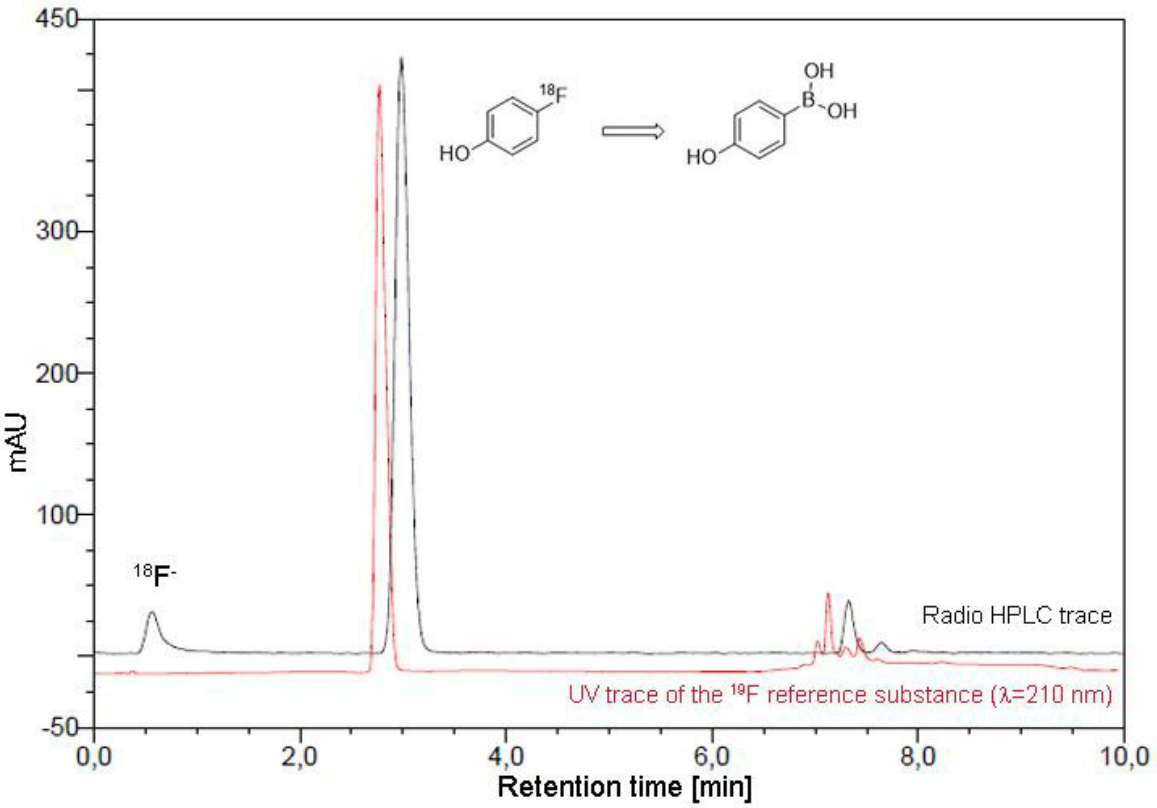

400
300
200
100
-50
mAU
OHC
18F
OHC
O
B
O
18F-
Radio HPLC trace
UV trace of the 19F reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]

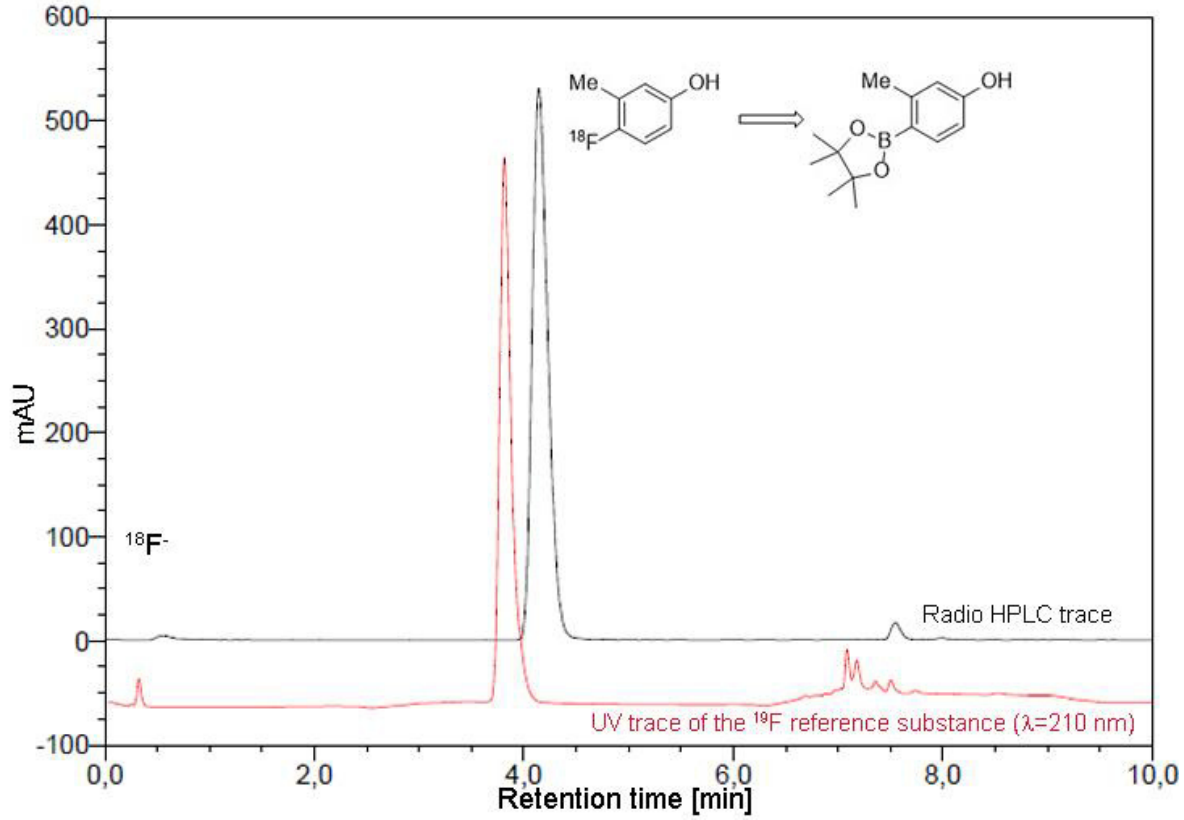

700
500
375
250
125
0
-100
mAU
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]
$^{18}F^-$
Radio HPLC trace
UV trace of the ^{19}F reference substance (λ=210 nm)
OHC
^{18}F
OH
B
OH

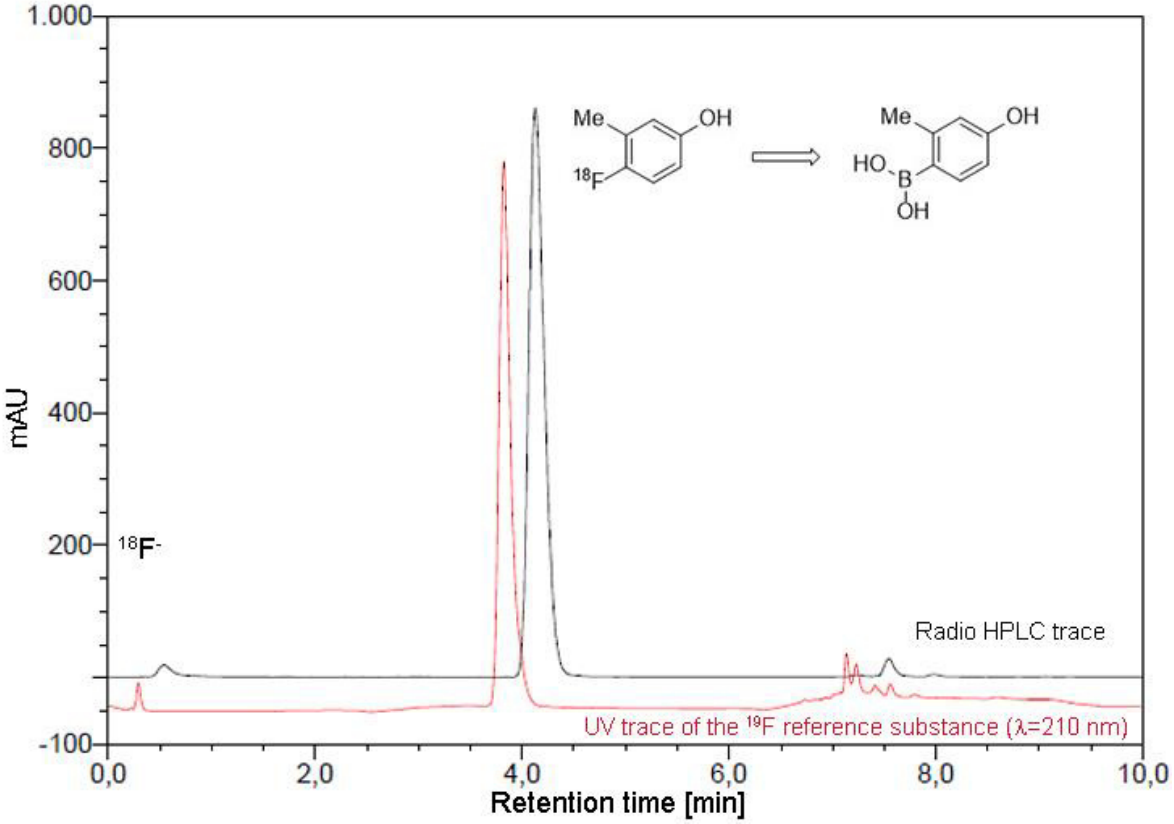

400
300
200
100
-50
mAU
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]
$^{18}F^-$
Radio HPLC trace
UV trace of the ^{19}F reference substance (λ=210 nm)
OHC
^{18}F
O
B
O

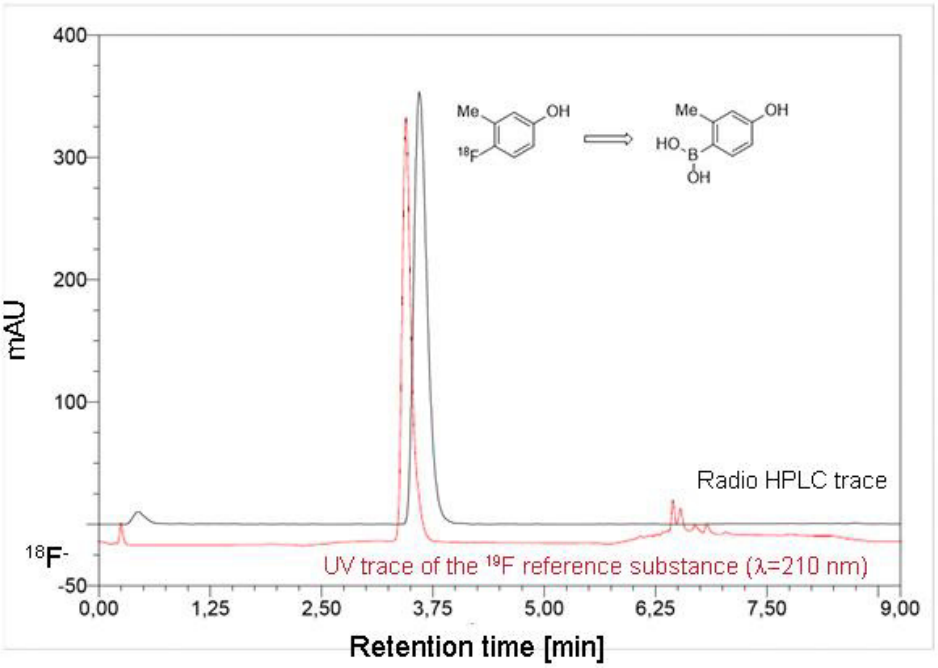

450
300
200
100
-50
mAU
OHC
18F
OHC
OH
B
OH
18F-
Radio HPLC trace
UV trace of the 19F reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]


450
300
200
100
-50
mAU
18F
HO
HO
O
B
O
Radio HPLC trace
UV trace of the 19F reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]

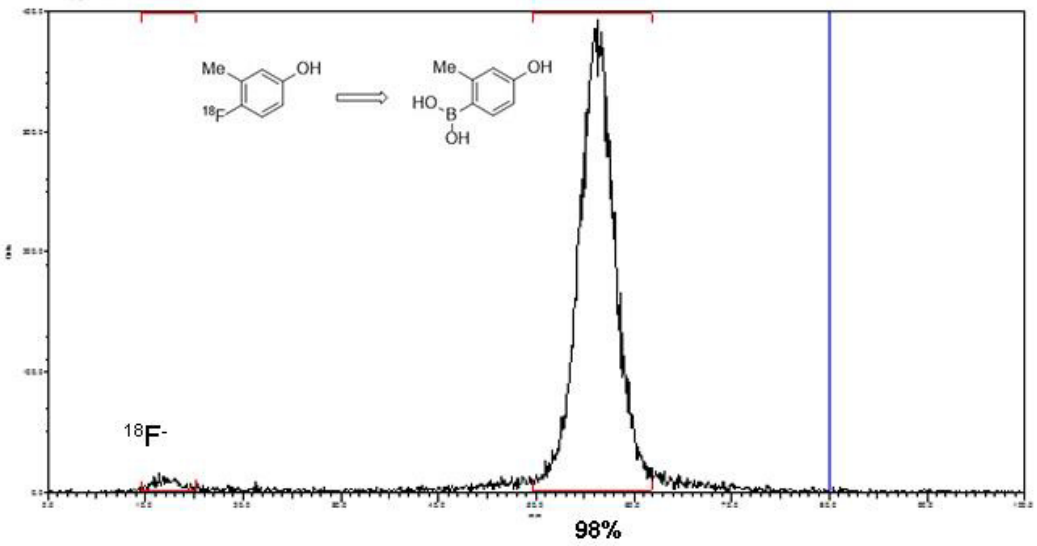

450
300
200
100
-50
mAU
^{18}F-
Radio HPLC trace
UV trace of the ^{19}F reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]

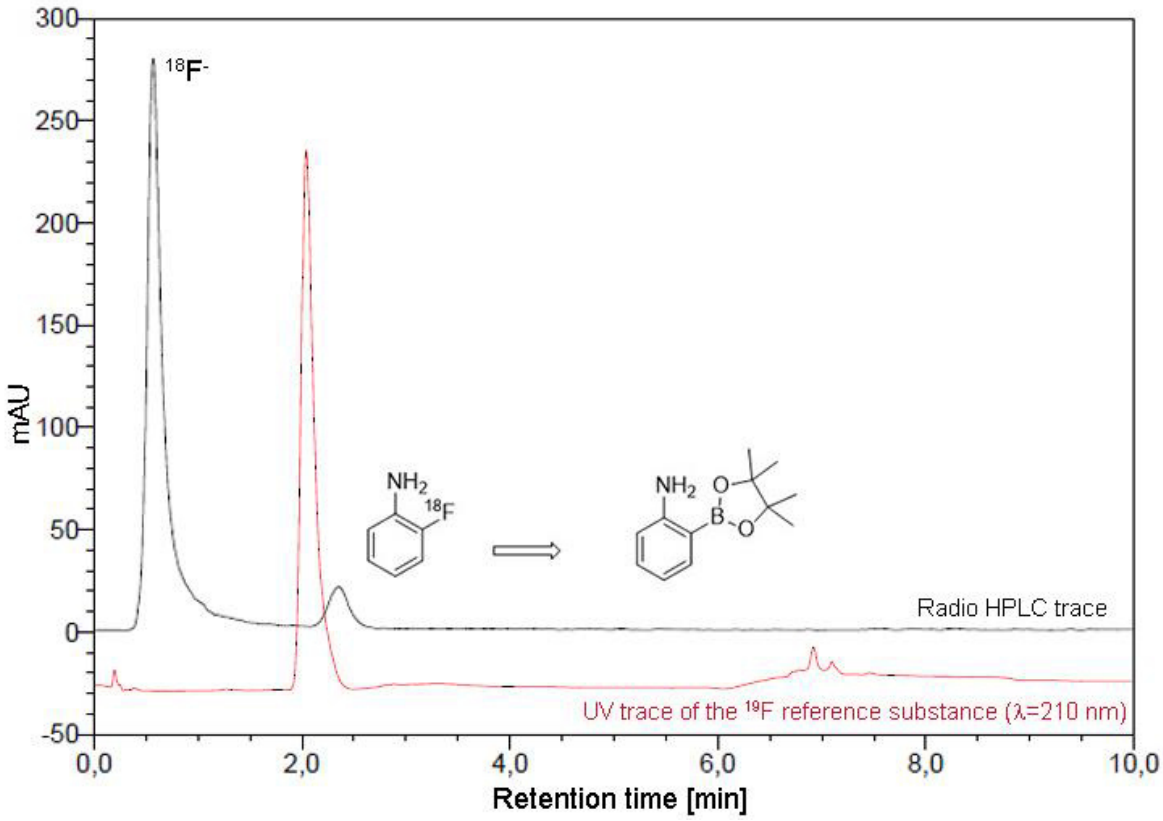

600
500
400
300
200
100
0
-100
mAU
^{18}F-
Radio HPLC trace
UV trace of the ^{19}F reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]

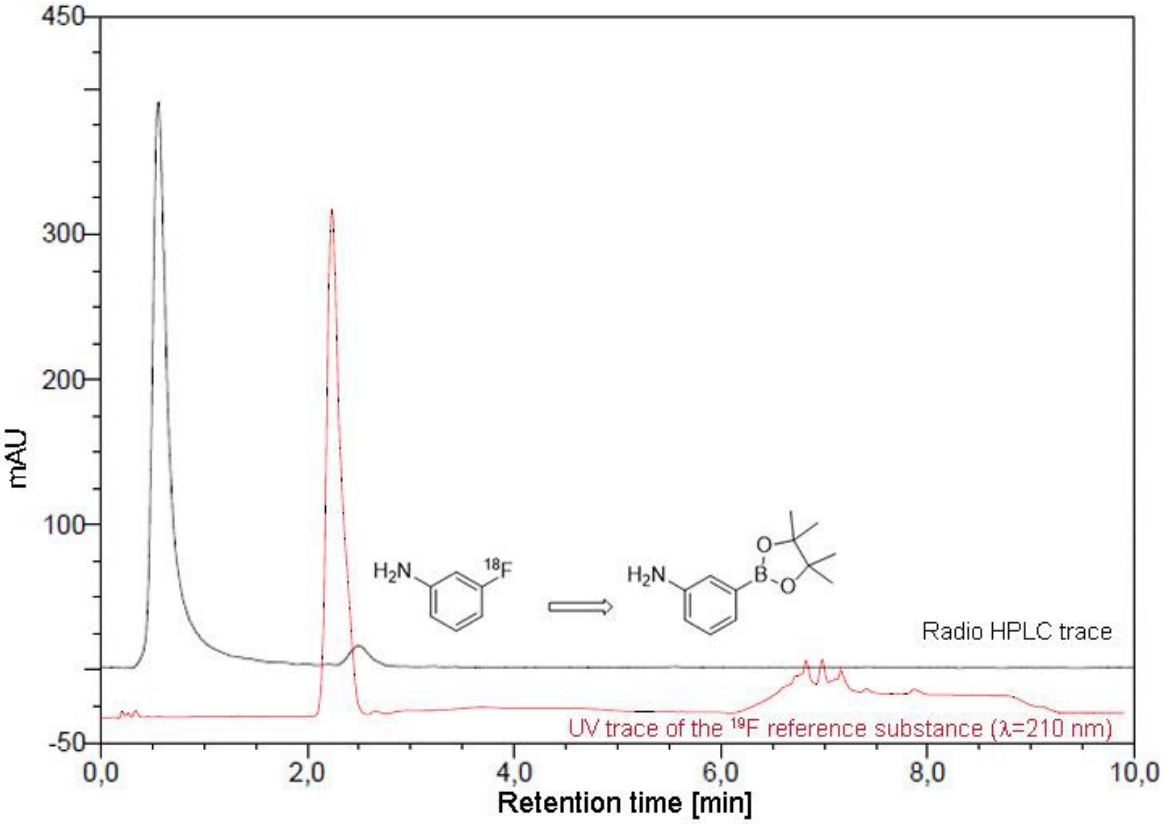


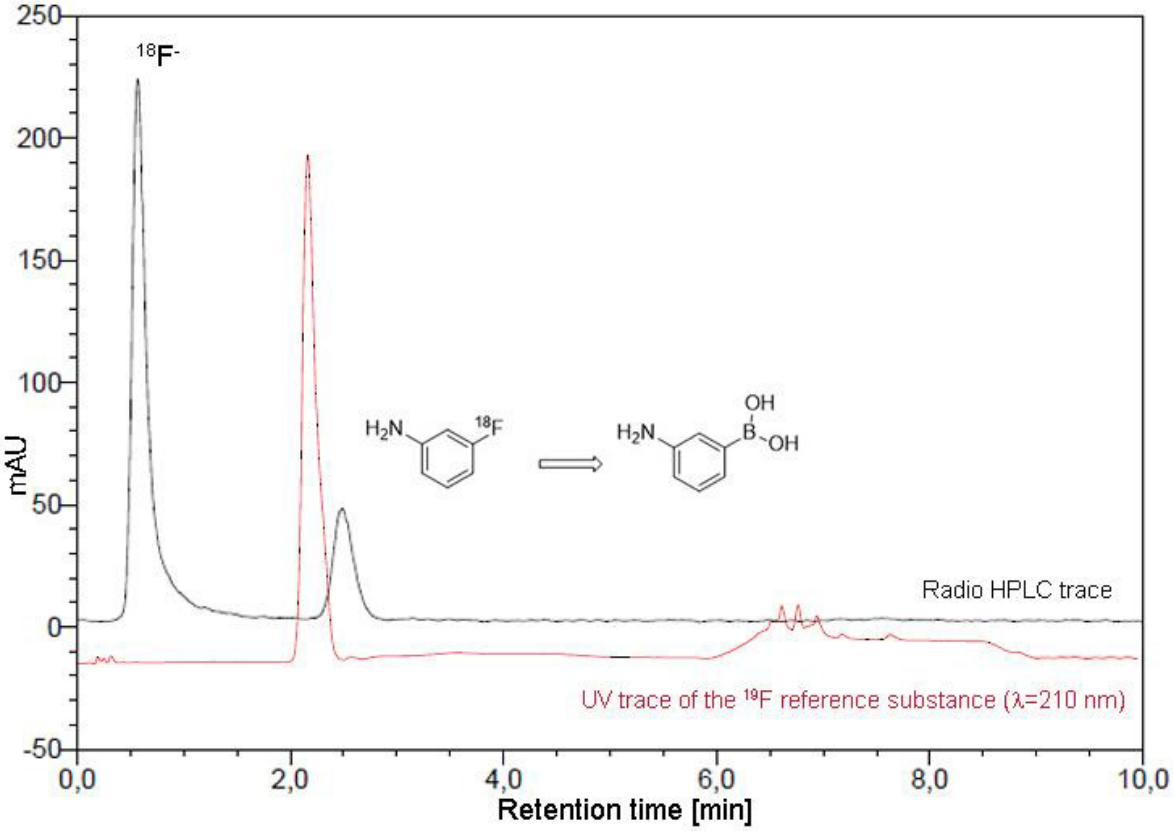


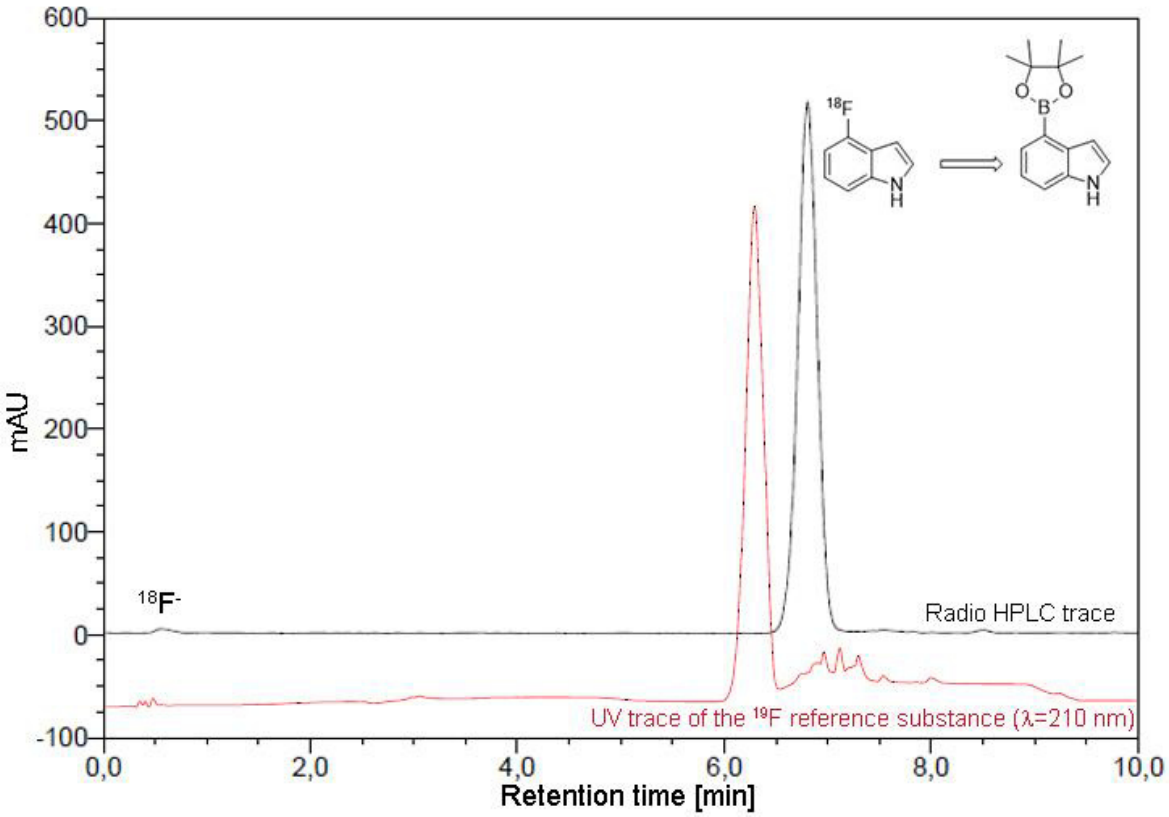
Nr.	Retention time	Intensity	Integral
	min	mAU	%
1	0,43	9,441	2,27
2	3,60	353,256	97,73
Total:		362,697	100,00

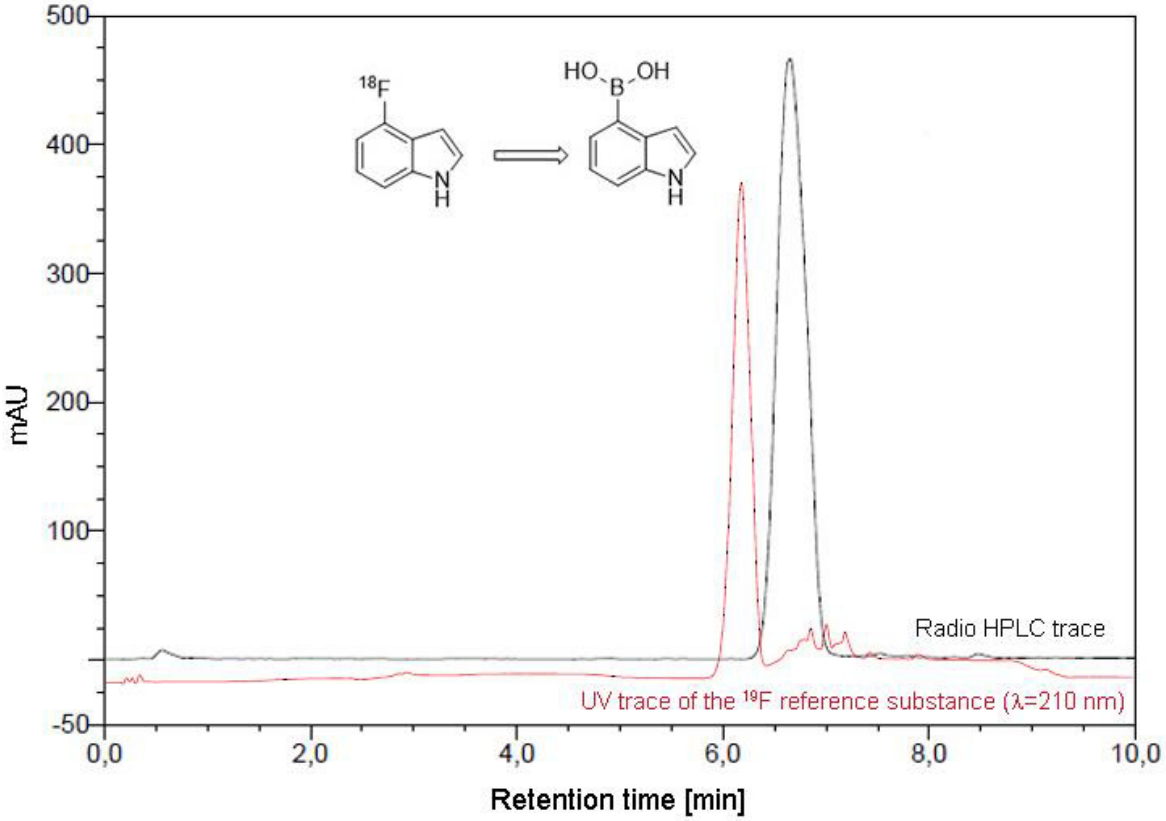

Me
OH
18F
Me
OH
HO
B
OH
18F-
98%

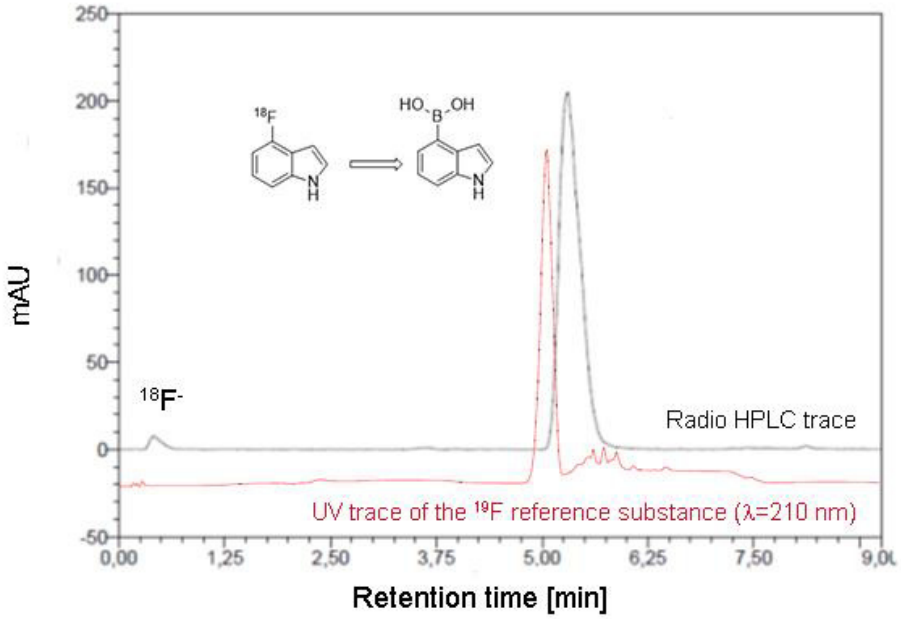

300
250
200
150
100
50
0
-50
mAU
18F-
NH2
18F
NH2
O
B
O
Radio HPLC trace
UV trace of the 19F reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]


450
300
200
100
-50
mAU
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]
Radio HPLC trace
UV trace of the 19F reference substance (λ=210 nm)

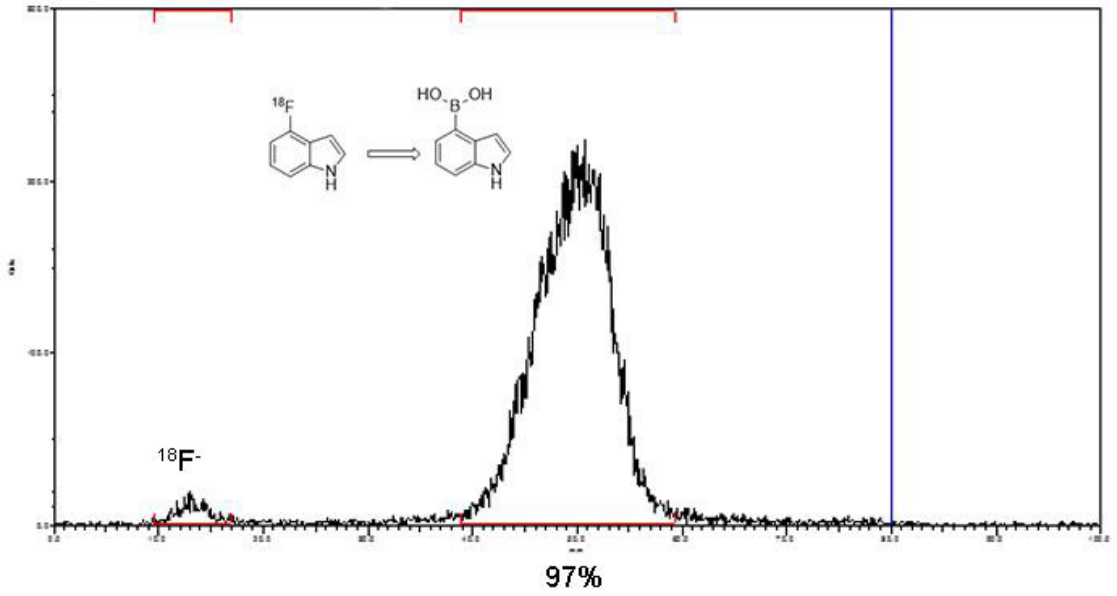

250
200
150
100
50
0
-50
mAU
18F-
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]
Radio HPLC trace
UV trace of the 19F reference substance (λ=210 nm)

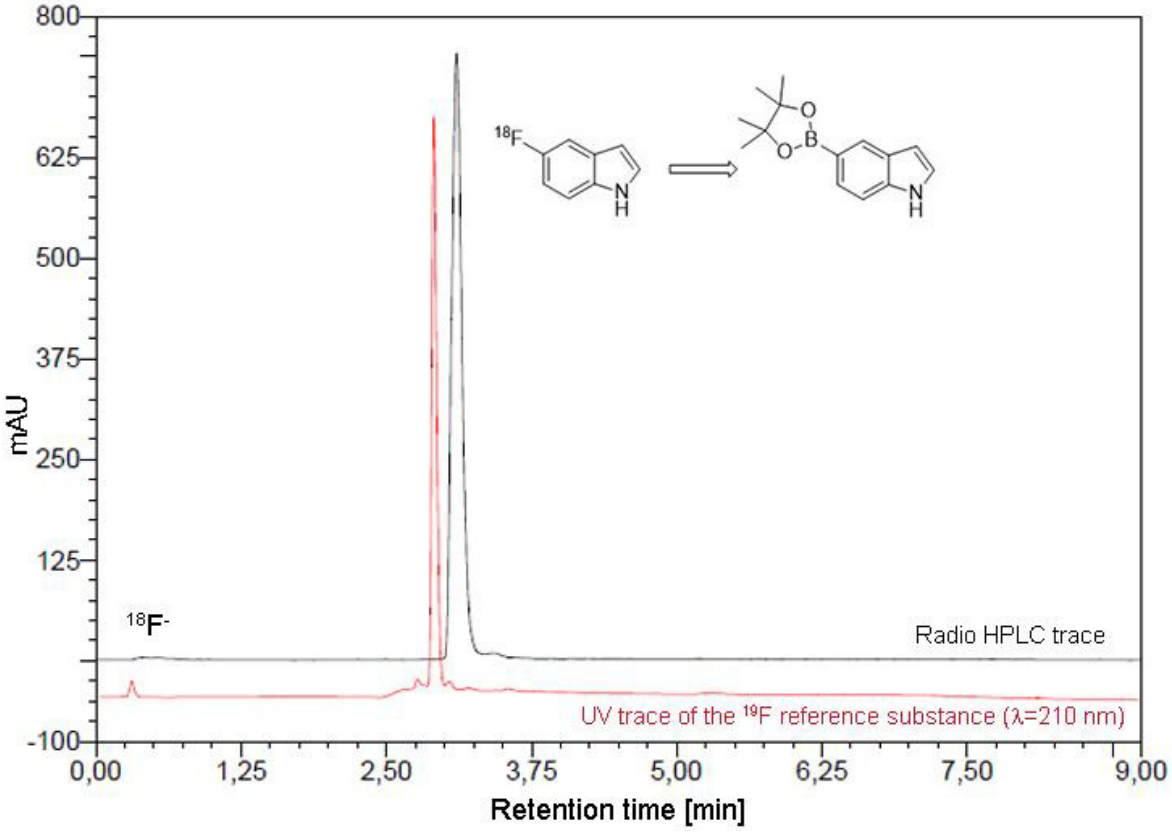

600
500
400
300
200
100
0
-100
mAU
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]
18F-
Radio HPLC trace
UV trace of the 19F reference substance (λ=210 nm)
18F
N
H
O
B
O
N
H

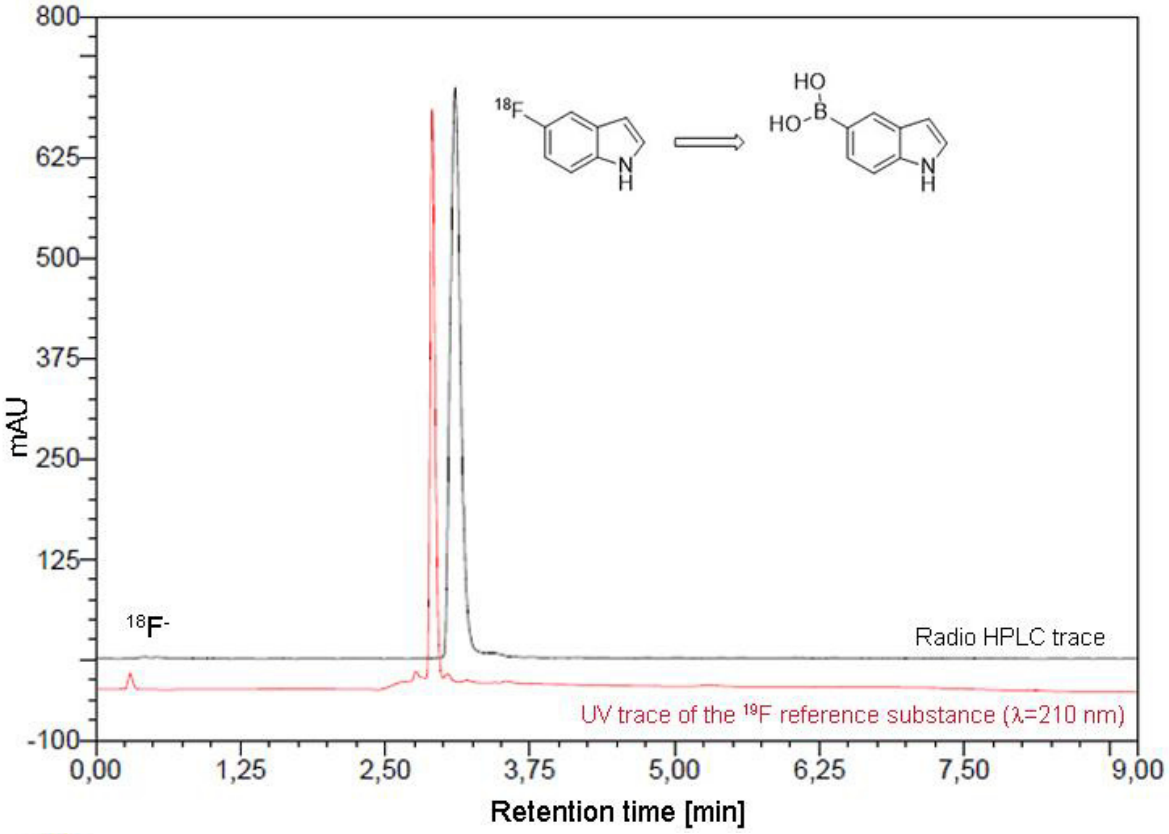


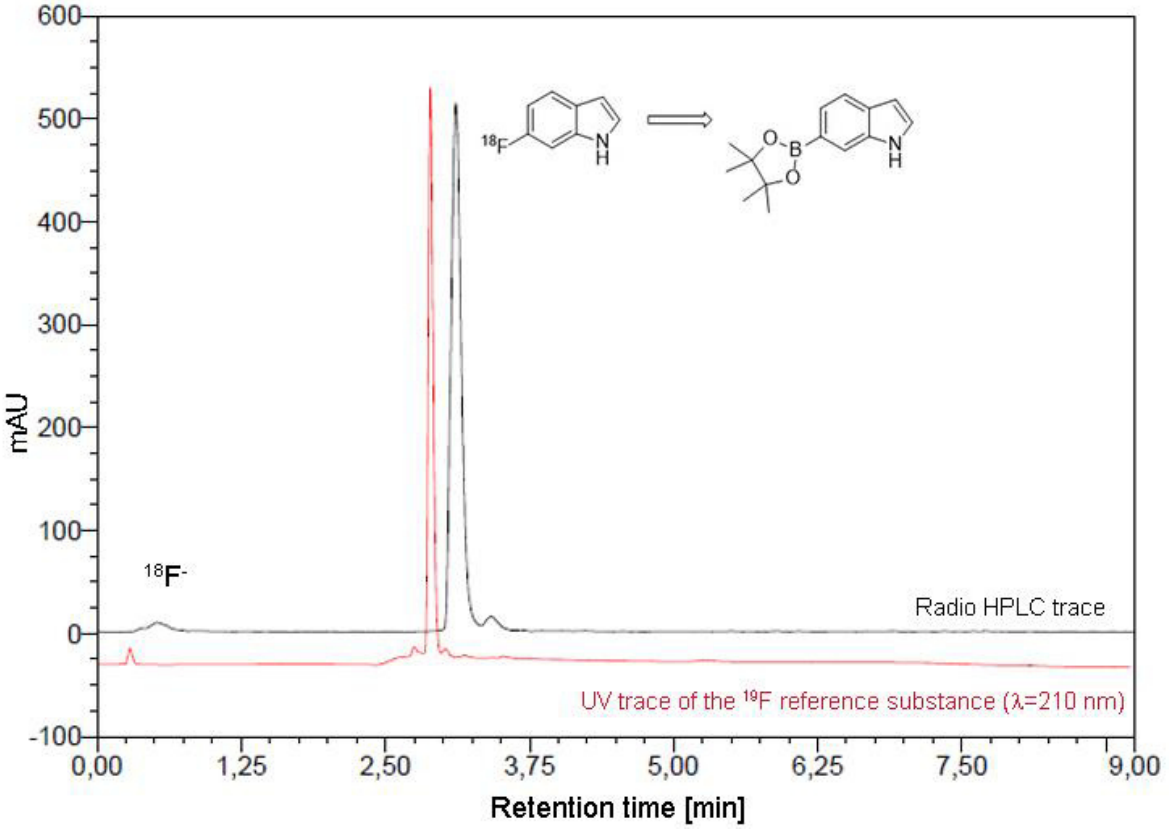


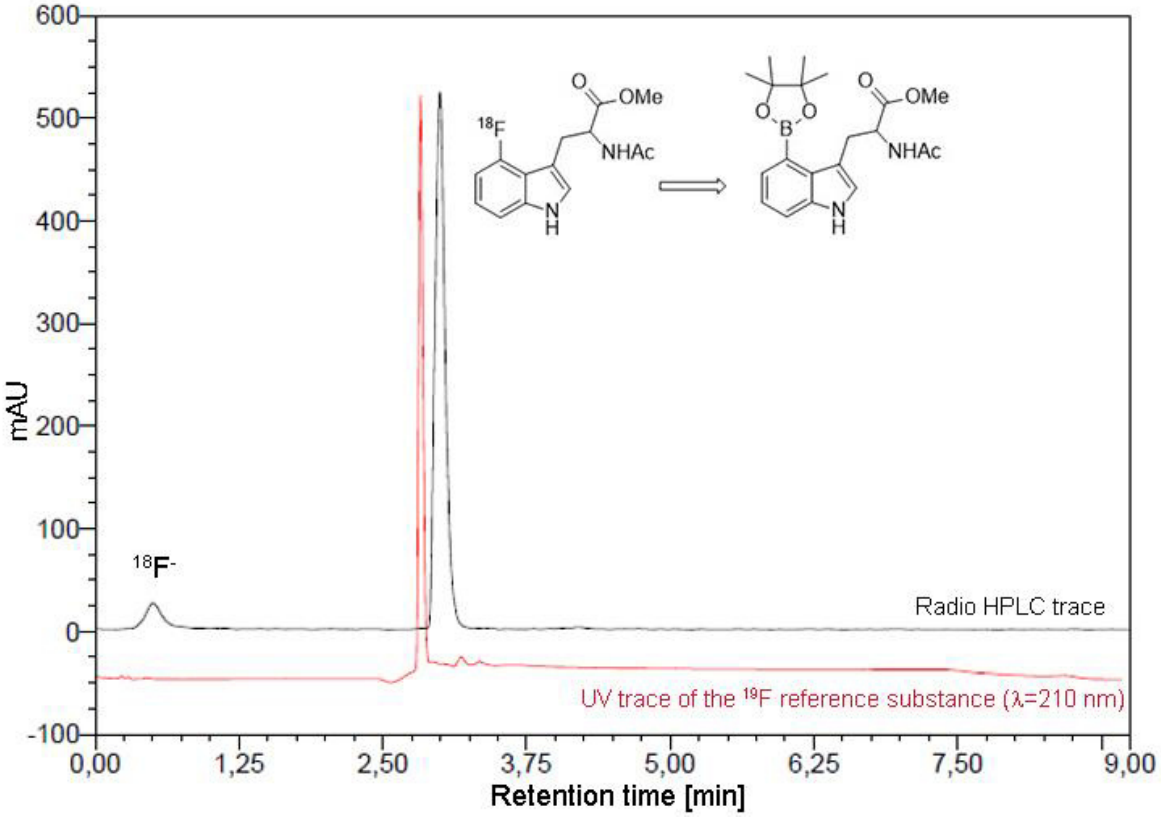
Nr.	Retention time min	Intensity mAU	Integral %
1	0,41	7,502	1,87
2	5,29	204,534	97,76
3	8,12	1,601	0,37
Total:		213,637	100,00

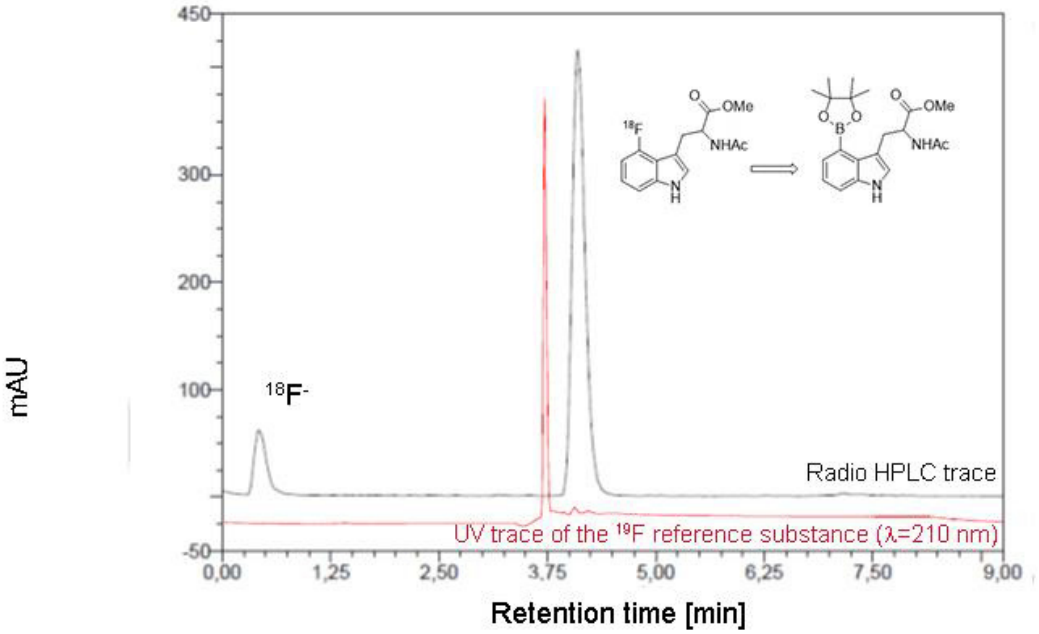

18F
HO
B
OH
N
H
18F-
97%


800
625
500
375
250
125
-100
mAU
18F
N
H
18F-
Radio HPLC trace
UV trace of the 19F reference substance (λ=210 nm)
0,00
1,25
2,50
3,75
5,00
6,25
7,50
9,00
Retention time [min]

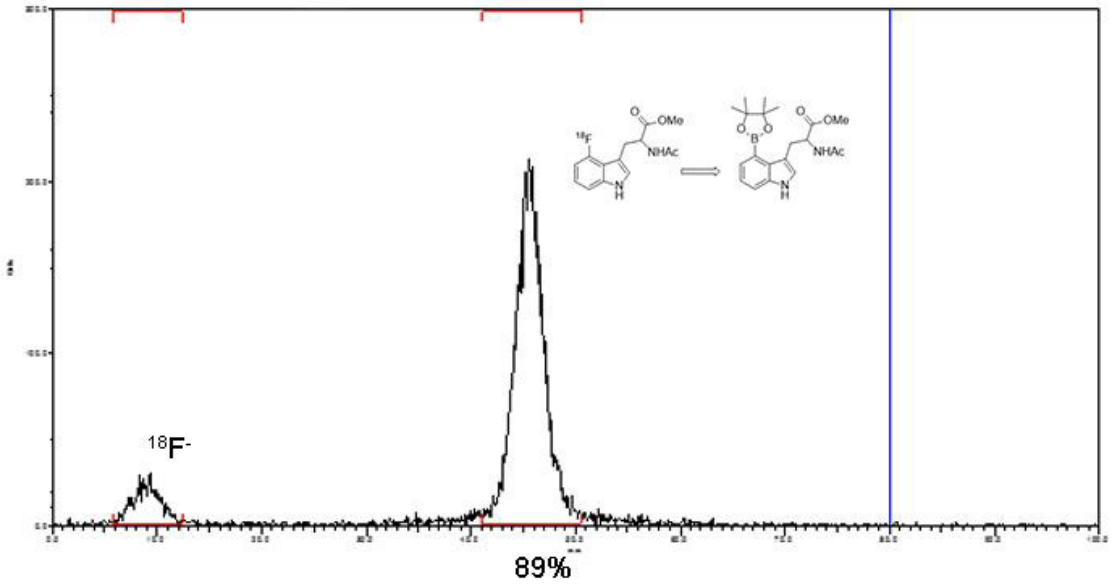

800
625
500
375
250
125
-100
mAU
^{18}F-
Radio HPLC trace
UV trace of the ^{19}F reference substance (λ=210 nm)
0,00
1,25
2,50
3,75
5,00
6,25
7,50
9,00
Retention time [min]

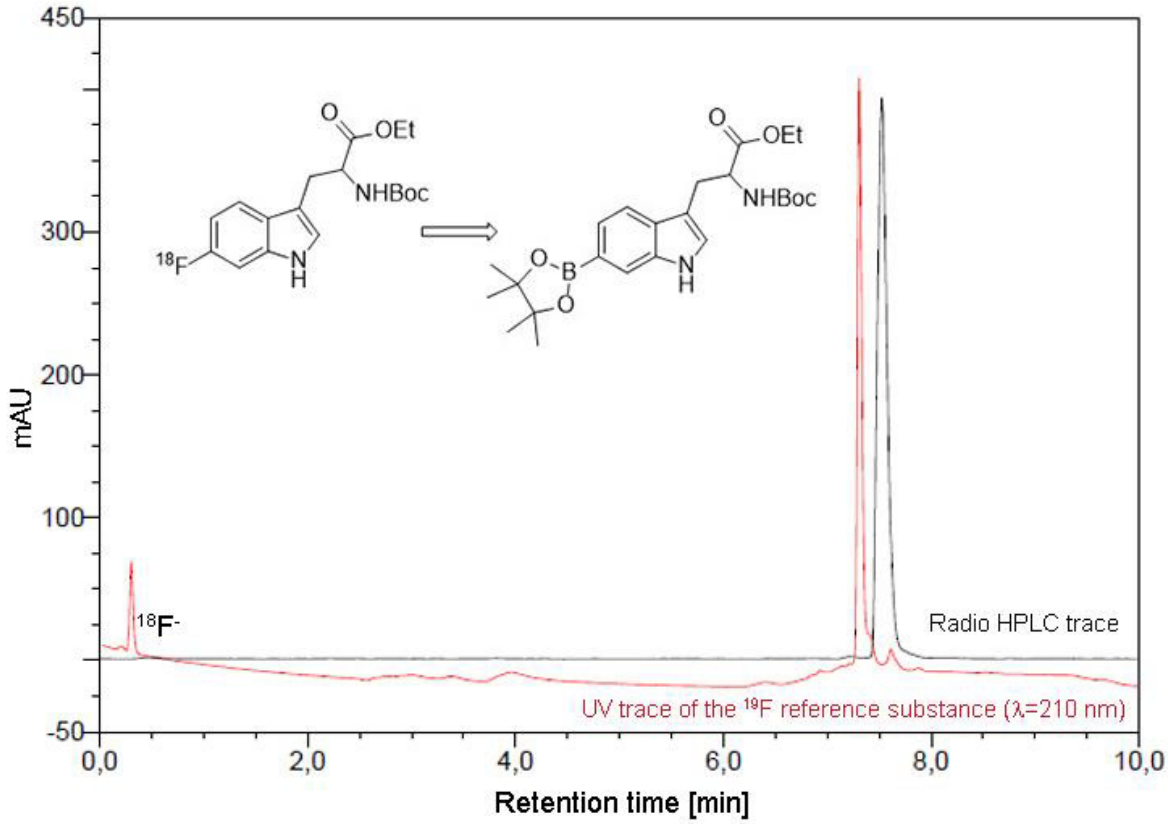

600
500
400
300
200
100
0
-100
mAU
^{18}F-
Radio HPLC trace
UV trace of the ^{19}F reference substance (λ=210 nm)
0,00
1,25
2,50
3,75
5,00
6,25
7,50
9,00
Retention time [min]

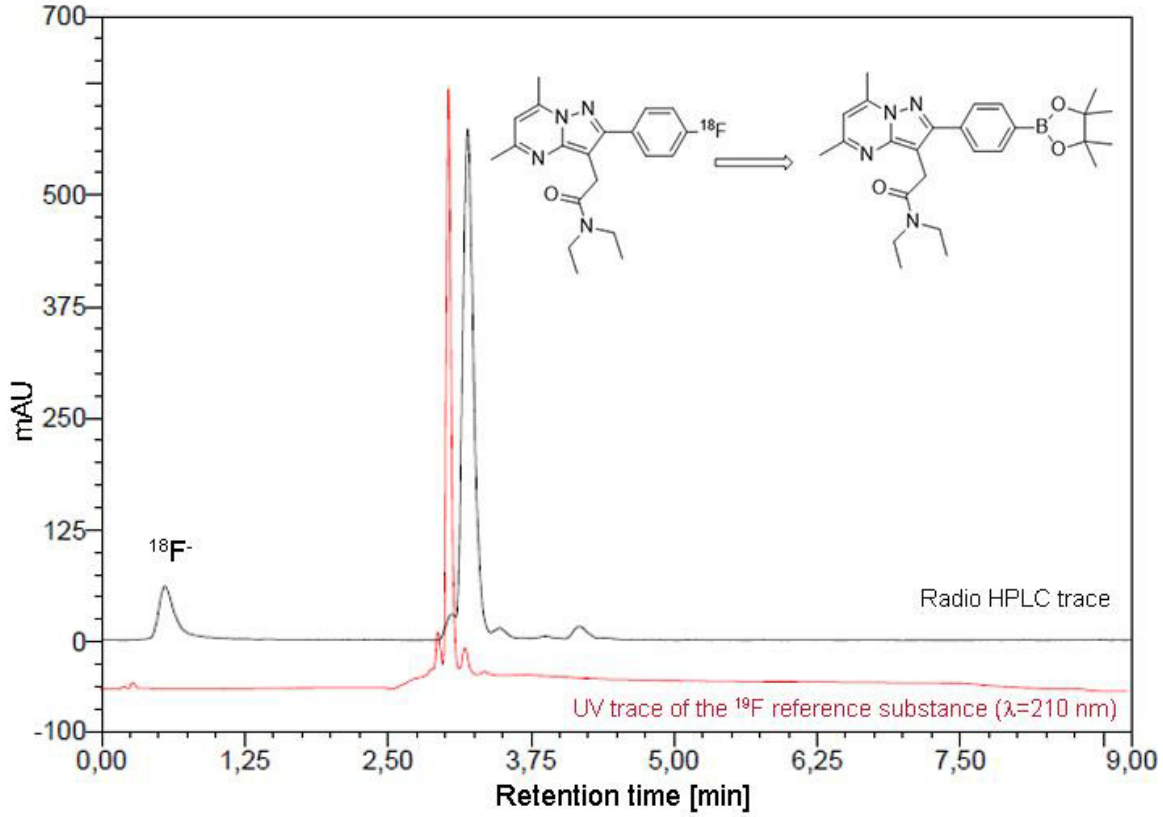


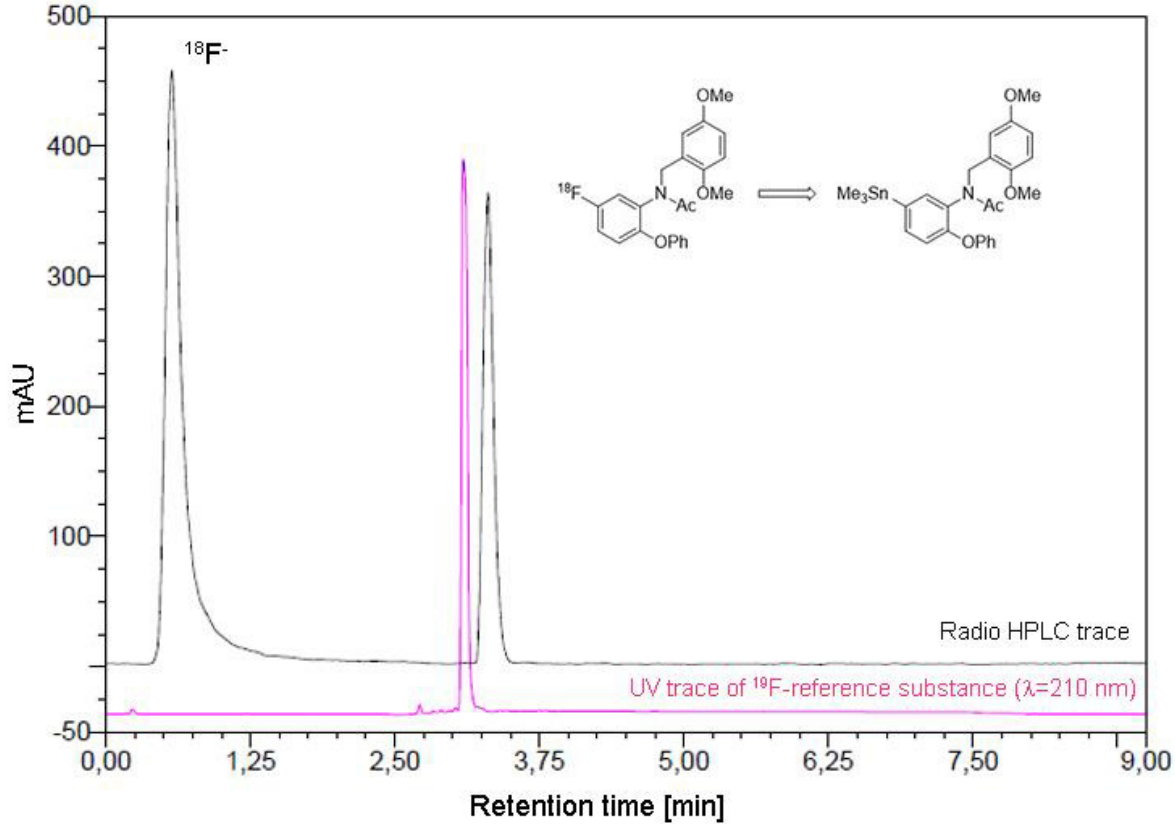


Nr.	Retention time	Intensity	Integral
	min	mAU	%
1	0,42	59,951	11,73
2	4,10	414,593	88,27
Total:		474,544	100,00


OMe
NHAc
^{18}F
^{18}F-
89%


450
300
200
100
-50
mAU
OEt
NHBoc
^{18}F
^{18}F-
Radio HPLC trace
UV trace of the ^{19}F reference substance (λ=210 nm)
0,0
2,0
4,0
6,0
8,0
10,0
Retention time [min]





References

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3 Zusammenfassende Diskussion

3.1 Entwicklung von neuen Radiofluorierungsverfahren für die Synthese von ^{18}F-markierten Aminen und Aminosäuren

Mit ^{18}F markierte aromatische Amine und Aminosäuren spielen in der klinischen Diagnostik mittels PET eine wichtige Rolle. Vor allem 6-[^{18}F]Fluor-L-3,4-dihydroxyphenylalanin (6-[^{18}F]FDOPA), ein wichtiger Tracer für die Visualisierung der Synthese, Einlagerung und dem Metabolismus von Catecholaminen, wird oft klinisch zur Diagnostik von neuroendokrinen Tumoren sowie Morbus Parkinson eingesetzt.[44] 6-[^{18}F]Fluor-L-*m*-tyrosin (6[^{18}F]FMT) wird für identische Zwecke verwendet, weist allerdings gegenüber 6-[^{18}F]FDOPA eine deutlich günstigere *in vivo* Verteilung auf.[45] Auch 6-[^{18}F]Fluordopamin (6-[^{18}F]FDA) hat in der Bildgebung von neuroendokrinen Tumoren eine gewisse Bedeutung erlangt.[46] Gemessen an der Gesamtzahl durchgeführter PET-Messungen, werden diese Verbindungen aktuell jedoch noch relativ selten verwendet. Die Einführung von ^{18}F in elektronenreiche aromatische Systeme, wie bei den oben genannten Verbindungen, ist nach wie vor eine der großen Herausforderungen der Radiochemie. Die meisten bisher beschriebenen Methoden verwirklichen dies auf elektrophilem Weg, wobei c.a. [^{18}F]F_2 verwendet wird. Die dadurch bedingten geringen Ausbeuten und molaren Aktivitäten (350–500 MBq/µmol oder niedriger)[47] sind dabei die wichtigsten Gründe dafür, dass die Tracer nur selten klinische Verwendung finden. Vor diesem Hintergrund war das vorrangige Ziel dieser Arbeit, effiziente Methoden zur Synthese von, mit ^{18}F markierten, aromatischen Aminen und Aminosäuren zu etablieren. Ein besonderer Fokus lag hierbei auf Übergangsmetall-vermittelten Radiofluorierungsverfahren.

3.2 Untersuchung der Ni-vermittelten Radiofluorierung elektronenreicher Aromaten

Die von Lee *et al.* beschriebene Ni-vermittelte Radiofluorierung ermöglichte den Zugang zu, mit ^{18}F markierten elektronenreichen Aromaten. Ein Ziel dieser Arbeit war es, die Eignung dieser Methode für die Herstellung von klinisch relevanten Tracern für die PET zu evaluieren. In ersten Experimenten zeigte sich, dass die Reaktionsbedingungen, die von Lee *et al.*[48] beschrieben wurden, nicht zur Produktbildung führten. Auch über das klassische Radiofluorierungsprotokoll mit K2.2.2 und K_2CO_3 (2,5–3 mg) konnte das gewünschte Produkt nicht detektiert werden. Daher wurde eine vollständige Optimierung aller Reaktionsparameter vorgenommen. Durch Elution mit sehr kleinen Mengen an K_2CO_3 (0,16 mg) in MeOH und

anschließender Trocknung und Umsetzung mit dem Ni-Vorläufer in Anwesenheit der hypervalenten Iodverbindung (Abb. 3.1), konnten schließlich erste RCUs von 1–4% für geschütztes 6-[^{18}F]Fluordopamin erreicht werden. Die Erhöhung der Vorläufermenge, sowohl absolut als auch im Verhältnis zum Oxidationsmittel, ermöglichte schließlich radiochemische Umsätze von bis zu 18±10%.

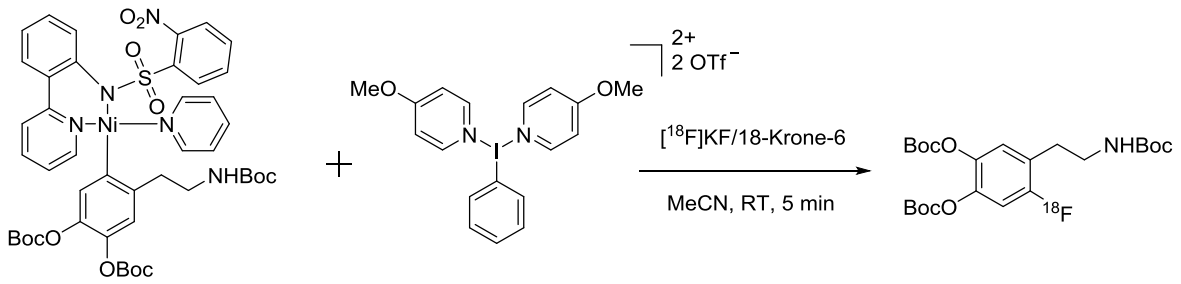


Abbildung 3.1 Synthese von tri-Boc geschütztem [^{18}F]FDA.

Eine darauf folgende Versuchsreihe mit verschiedenen Lösungsmitteln zeigte, dass, abweichend von den ursprünglich beschriebenen Reaktionsbedingungen, wasserfreies MeCN die höchsten RCUs ergibt. Die Verlängerung der Reaktionszeit von 1 auf 5 min führte schließlich zu optimierten Inkorporationsraten für $^{18}F^-$.

Tabelle 3.1 Vergleich der optimierten Reaktionsbedingungen von Lee *et al.* mit den in dieser Arbeit beschriebenen.

	Lee *et al.*[48]	**Zlatopolskiy, Zischler *et al.*[49]**
Base	Keine	0,16 mg K_2CO_3
Vorläufermenge	1 mg	5–10 mg
Verhältnis Oxidationsmittel/Vorläufer	1:1	1:1,3
Lösungsmittel	MeCN mit 1% H_2O	MeCN (wasserfrei)
Reaktionszeit	1 min	5 min

Unter optimierten Reaktionsbedingungen konnten die Boc-geschützten Vorstufen von 6-[^{18}F]FDOPA, 6-[^{18}F]FMT und 6-[^{18}F]FDA mit RCUs von 13, 9 bzw. 18% erhalten werden. Die anschließende saure Hydrolyse und Isolierung der radiofluorierten Produkte über HPLC ergab die PET-Tracer in radiochemischen Ausbeuten von 7±1, 5±1 und 12±2%. Ausgehend von 6,3 GBq $^{18}F^-$ wurden 220 MBq 6-[^{18}F]FDOPA erhalten. Für 6-[^{18}F]FDA wurde aus einer Startaktivität von 4 GBq 250 MBq der Zielverbindung isoliert. Für 6-[^{18}F]FDOPA und 6-[^{18}F]FDA wurden molare Aktivitäten von 175 bzw. 60 GBq/µmol bestimmt. Verglichen mit elektrophilen Synthesen lassen sich über die Ni-vermittelte Radiofluorierung elektronenreiche

Aromaten mit deutlich höheren molaren Aktivitäten erhalten. Allerdings liegen die sehr geringen RCAs von 5–12% teils noch deutlich unter den Ausbeuten moderner elektrophiler Radiosynthesen.[50] Vor diesem Hintergrund besteht weiterhin die Notwendigkeit, effizientere Radiofluorierungsmethoden für elektronenreiche aromatische Verbindungen zu entwickeln.

3.3 *In vivo* Vergleich von c.a. und n.c.a. 6-[^{18}F]FDOPA

Nach der erfolgreichen Synthese von n.c.a. 6-[^{18}F]FDOPA über die Ni-vermittelte Radiofluorierung ergab sich die Fragestellung, inwieweit die höhere molare Aktivität des PET-Tracers Einfluss auf die Qualität der rekonstruierten PET-Bilder hat. Als Beispiel diente hier ein unilaterales Modell des idiopathischen Parkinson-Syndroms in Ratten.[51] Zu diesem Zweck wurden einseitig die dopaminergen Mittelhirnneuronen, mittels stereotaktischer Injektion des Neurotoxins 6-Hydroxydopamin (6-OHDA) ins mediale Vorderhirnbündel, läsioniert. [52] 6–8 Wochen nach der Läsionierung wurden PET-Untersuchungen mit elektrophil und nucleophil hergestelltem 6-[^{18}F]FDOPA durchgeführt. 56–93 MBq des Tracers wurden in die laterale Schwanzvene injiziert. Die Messung wurden unmittelbar nach der Applikation gestartet und die Daten über eine Dauer von 60 min erfasst. Nach Auswertung der PET-Bilder konnte weder anhand des SUV (standardized uptake value), noch durch einen Vergleich des SUVR (standardized uptake value ratio), also dem Verhältnis von Aktivitätsaufnahme im Striatum zur Aktivitätsaufnahme im Cerebellum, welches als Referenzareal diente, signifikante Unterschiede detektiert werden. Auch rein visuell sind beide Aufnahmen nahezu identisch (Abb. 3.2).

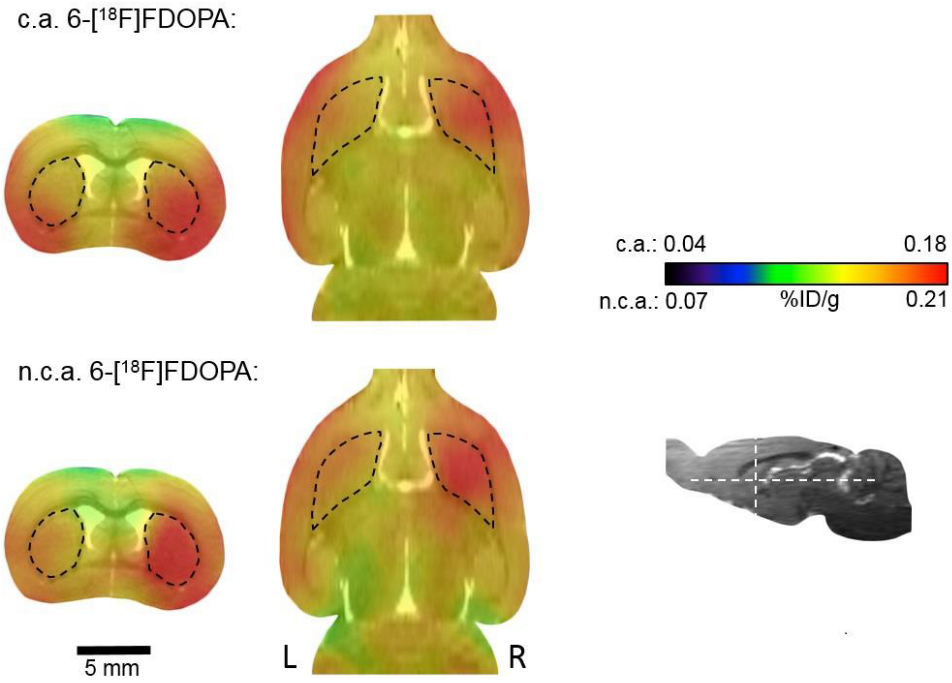


Abbildung 3.2 c.a. *versus* n.c.a. 6-[^{18}F]FDOPA: PET/MR-Bild in einem Rattenmodell der Parkinsonschen Krankheit. Gezeigt sind jeweils zwei transversale und zwei horizontale Schnittbilder desselben Tiers.

Dieses etwas unerwartete Ergebnis lässt sich durch die hohe Kapazität und/oder die starke Konkurrenz der biochemischen Prozesse erklären, die für die Akkumulation von 6-[^{18}F]FDOPA und dessen Metaboliten in dopaminergen Neuronen verantwortlich sind. Im Vergleich von 6-[^{18}F]FDOPA und anderen Substraten, wie Phenylalanin, Tyrosin oder Valin,[50] die um die gleichen Stoffwechselprozesse konkurrieren, kann der Beitrag des Trägers (6-FDOPA) zu diesen Prozessen offensichtlich vernachlässigt werden. Dementsprechend ist der Einfluss der molaren Aktivität von 6-[^{18}F]FDOPA in diesem geringen Konzentrationsbereich augenscheinlich von unwesentlicher Bedeutung für die Bildgebung des dopaminergen Systems. Gestützt werden diese Ergebnisse durch Kuik *et al*. An einem Modell für neuroendokrine Tumore in Mäusen konnten ebenfalls keine signifikanten Unterschiede in der Aufnahme von c.a. und n.c.a. 6-[^{18}F]FDOPA festgestellt werden.[53]

3.4 Cu-vermittelte Radiofluorierung von Mesitylaryliodoniumsalzen

Wie in Abschnitt 3.2 beschrieben, wies die Ni-vermittelte Radiofluorierung sowohl hinsichtlich der RCU als auch ihrer Praktikabilität deutliche Limitierungen auf. Vor diesem Hintergrund rückten Cu-vermittelte Fluorierungsstrategien, wie sie jüngst für die organisch-präparative Chemie beschrieben wurden, in den Fokus.

In ersten Experimenten wurde die Cu-vermittelte Fluorierung von Mesitylaryliodoniumsalzen nach Ichiishi *et al.*[32] aus der organischen in die Radiochemie

übertragen. Nach anfänglichen Anpassungen konnte ein erstes Protokoll für die Modellverbindungen [^{18}F]Fluorbenzol ([^{18}F]FB), 3-[^{18}F]Fluorbenzaldehyd (3-[^{18}F]FBA) und 4-[^{18}F]Fluoranisol ([^{8}F]FA) als Beispiele für elektronenneutrale, -ziehenden und –schiebende Substituenten, entwickelt werden.$^{18}F^-$ wurde mit einer methanolischen Lösung von K_2CO_3 von einer Anionenaustauschkartusche eluiert. Wie schon bei der Ni-vermittelten Radiofluorierung war bereits ein geringe Menge K_2CO_3 (0,5 mg) für eine vollständige [^{18}F]Fluorid-Wiedergewinnung ausreichend. Da MeOH innerhalb weniger Minuten entfernt werden konnte, konnte auf eine azeotrope Trocknung gänzlich verzichtet werden. Anschließend wurde der trockene Rückstand mit einer Lösung des jeweiligen Iodoniumvorläufers und $Cu(OTf)_2$ in Dimethylformamid (DMF) aufgenommen. Nach einer Reaktionszeit von 20 min bei 85 °C konnten die radiofluorierten Produkte in RCUs von 29–50% erhalten werden (siehe Veröffentlichung 2, Tabelle 1).

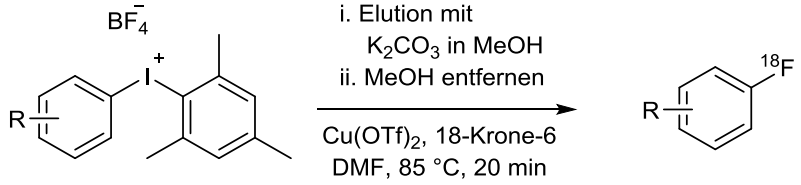


Abbildung 3.3 $Cu(OTf)_2$-vermittelte Radiofluorierung von Mesitylaryliodoniumsalzen.

Zunächst wurde der Einfluss des Onium-Gegenions auf die RCUs untersucht. Neben Tetrafluorborat wurden so auch Perchlorat und Triflat untersucht, die ähnliche Umsätze (35%) lieferten, während bei den Tosylat- und Bromid-Vorläufern nur eine sehr geringe Produktbildung beobachtet wurde. Die Verlängerung der Reaktionszeit auf 30 min erhöhte den RCU von [^{18}F]Fluorbenzol von 41 auf 61%.

Durch die hier beschriebenen Weiterentwicklungen konnte die Cu-vermittelte Radiofluorierung von Mesitylaryliodoniumsalzen erfolgreich in die Radiochemie übertragen werden. Jedoch setzt die Verwendung von hygroskopischem $Cu(OTf)_2$ die Einhaltung von streng inerten Bedingungen voraus, was einen routinemäßigen Herstellungsprozess deutlich erschwert.

3.5 Cu-vermittelte Radiofluorierung von Mesitylaryliodoniumsalzen unter „low-base“ Bedingungen

Während die Übertragung der im vorherigen Abschnitt beschriebenen Cu-vermittelten Radiofluorierung andauerte, veröffentlichten Ichiishi *et al.* eine Cu-vermittelte Radiofluorierungsmethode von Mesitylaryliodonium-Vorläufern.[33] Da der von den Autoren verwendete $(MeCN)_4CuOTf$-Komplex im Gegensatz zu, in den hier beschriebenen ersten

Experimenten eingesetzten, $Cu(OTf)_2$ weniger empfindlich gegenüber Hydrolyse ist, schien die Methode nach Ichiishi *et al.* zunächst dem bisher in dieser Arbeit entwickelten Protokoll überlegen zu sein. Es zeigte sich ferner, dass durch die Verwendung des $(MeCN)_4CuOTf$-Komplexes nicht nur auf die Schutzgasatmosphäre verzichtet werden konnte, sondern die Durchführung unter Luft sogar zu höheren Umsätzen führte. Demzufolge, und um weiterhin wasserfreie Bedingungen gewährleisten zu können, wurden sämtliche Synthesen unter synthetischer Luftatmosphäre durchgeführt. $^{18}F^-$ wurde zunächst azeotrop getrocknet und, wie bei Ichiishi beschrieben, in MeCN aufgenommen. Daraus wurde für jede Synthese ein Aliquot entnommen. Eventuelle Verluste während der Trocknung und durch Adsorption an den Gefäßwänden sind in den angegebenen Umsätzen nicht berücksichtigt. In unseren Bestimmungen zeigte sich, dass bis zu 80% der eingesetzten Aktivität nicht wieder gelöst werden konnte, sondern an den Gefäßwänden haften blieb. In einer Ein-Topf-Synthese zeigte sich demzufolge auch eine drastische Reduktion der RCUs auf 0,5–5%. Zudem wies das Auftreten von [^{18}F]Mesitylfluorid als Nebenprodukt auf einen Verlust an Selektivität hin. Dies führte zur Vermutung, dass $(MeCN)_4CuOTf$ unter basischen Bedingungen nicht stabil ist. Die Prüfung der Synthesebedingungen von Ichiishi, zeigte, dass in den Aliquoten der $^{18}F^-$-Acetonitril-Lösung lediglich 0,11 mg K_2CO_3 enthalten waren.

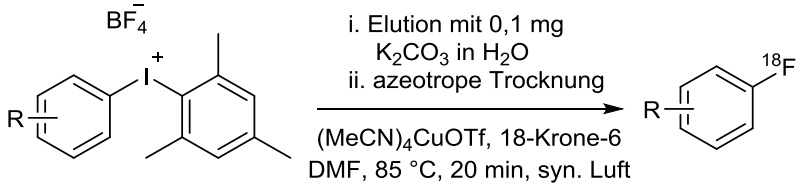


Abbildung 3.4 Cu-vermittelte Radiofluorierung von Mesitylaryliodoniumsalzen unter „low-base“ Bedingungen.

Aus diesem Grund wurde ein „low-base“ Protokoll entwickelt. Anstelle der sonst üblichen 3,5 mg K_2CO_3 wurden nur 0,11 mg für die Elution eingesetzt. Diese geringe Menge war für die vollständige ^{18}F-Elution ausreichend. Nach azeotroper Trocknung mit MeCN wurde der jeweilige Vorläufer und $(MeCN)_4CuOTf$ in DMF zugegeben und die Lösung bei 85 °C für 20 min unter synthetischer Luft gerührt. Durch Anwendung dieses Syntheseprotokolls konnten die RCUs deutlich gesteigert werden. Die Modellverbindungen [^{18}F]FB, 3-[^{18}F]FBA und [^{18}F]FA konnten in Umsätzen von 28, 56 bzw. 26% erhalten werden (Tab. 3.2). Dennoch konnten die hohen RCUs, wie sie mit den ursprünglich beschriebenen Reaktionsbedingungen unter Verwendung von Aliquoten der $^{18}F^-$-Lösung erreicht werden konnten, nicht reproduziert werden.

3.6 Cu-vermittelte Radiofluorierung unter „minimalistischen“ Bedingungen

Durch die Einführung des „low-base“ Protokolls konnten die RCUs der Cu-vermittelten Radiofluorierung von Mesitylaryliodoniumsalzen deutlich gesteigert werden. Daraus ergab sich die Fragestellung, ob es möglich ist, durch Übertragung der „minimalistischen“ Methode, vollständig auf den Einsatz einer Base zu verzichten.

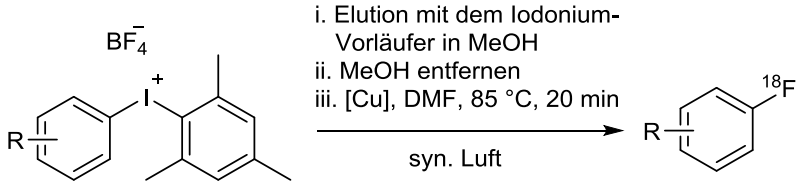


Abbildung 3.5 Cu-vermittelte Radiofluorierung von Mesitylaryliodoniumsalzen unter „minimalistischen“ Bedingungen.

Wie in der Literatur beschrieben wurde $^{18}F^-$ mit einer Lösung des entsprechenden Iodoniumvorläufers in MeOH von der Anionenaustauscherkartusche nahezu quantitativ eluiert (>97%). MeOH wurde entfernt, der $(MeCN)_4CuOTf$-Komplex in DMF zugegeben und die resultierende Lösung bei 85 °C für 20 min gerührt. Mit 90, 86 und 78% überstiegen die RCUs der Modellverbindungen [^{18}F]FB, 3-[^{18}F]FBA bzw. [^{18}F]FA deutlich die, die zuvor unter „low-base“ Bedingungen erhalten wurden. Auch die Umsätze, die unter Verwendung von Aliquots nach Ichiishi *et al*. erhalten wurden, konnten durch dieses Radiofluorierungsprotokoll zum Teil deutlich übertroffen werden (vgl. Tab. 3.2).

Tabelle 3.2 Vergleich der Radiofluorierungsmethoden von Mesitylaryliodoniumsalzen.

	RCU [%]		
	Aliquot	„low-base“	„minimalistisch“
[^{18}F]FB	56	28	90
3-[^{18}F]FBA	84	56	86
[^{18}F]FA	36	26	78

Durch die Etablierung eines „minimalistischen“ Protokolls für die Cu-vermittelte Radiofluorierung von Mesitylaryliodoniumsalzen gelang es, vollständig auf den Einsatz von Base zu verzichten. Ferner konnte auch auf die azeotrope Trocknung, die mit teils großen Aktivitätsverlusten einhergeht, verzichtet und so die RCUs erheblich gesteigert werden. Diese Weiterentwicklung der Cu-vermittelten ^{18}F-Markierung ermöglicht potentiell die Synthese von PET-Tracern in hohen Aktivitätsdosen.

3.7 „Minimalistische“ Cu-vermittelte Radiosynthese von 4-[^{18}F]Fluor-L-phenylalanin, [^{18}F]FDA und [^{18}F]DAA1106

Da die Ergebnisse der Ni-vermittelten Radiofluorierung bezüglich der Ausbeuten und Praktikabilität wenig überzeugend waren, sollte geklärt werden, ob sich die „minimalistische“ Cu-vermittelte Radiofluorierung zur Herstellung von PET-Tracern eignet. Um die Vergleichbarkeit der Ergebnisse zu gewährleisten, wurde erneut 6-[^{18}F]FDA als Modellverbindung ausgewählt. Das geschützte Zwischenprodukt konnte erfolgreich mit RCUs von 71–97% ^{18}F-markiert werden. Nach Hydrolyse wurde das Produkt mittels HPLC in einer RCA von 46% isoliert (Abb. 3.6). Die, im Verhältnis zum RCU geringe RCA ist auf die sehr aufwendige Entschützung des markierten Intermediats zurückzuführen. Da die *N*-Phthalimid-Gruppe selbst mit HI bei 130 °C nach 20 min nicht vollständig abgespalten werden konnte, wurde diese zunächst mit Hydrazinhydrat in EtOH enfernt. Im Anschluss wurden die *O*-4-Methylbenzyl-Gruppen mit HI abgespalten. Nach Entfernung der HI wurde das radiofluorierte Produkt mittels HPLC isoliert.

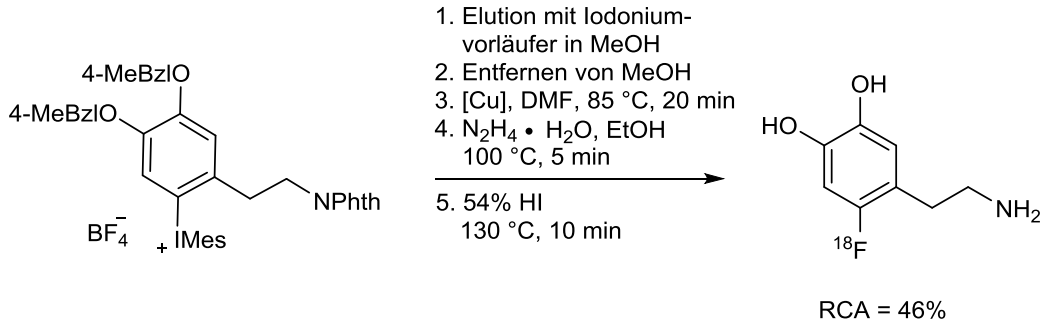


Abbildung 3.6 Synthese von 6-[^{18}F]Fluordopamin (6-[^{18}F]FDA)

Um das Anwendungsspektrum der „minimalistischen“ Cu-vermittelten Radiofluorierung von Mesitylaryliodoniumsalzen zu erweitern, wurden zwei weitere biologisch aktive Verbindungen mit ^{18}F markiert. 4-[^{18}F]Fluor-L-phenylalanin (4-[^{18}F]FPhe) wird in der Literatur als möglicher Tracer für die Visualisierung einer erhöhten Proteinsyntheserate, besonders bei Hirntumoren, beschrieben.[54] Klinische Anwendungsbeispiele sind bisher allerdings selten, was durch die schlechte Verfügbarkeit des Tracers bedingt ist. In der Praxis wird zur Diagnose von Hirntumoren fast ausschließlich 4-(2-[^{18}F]Fluorethyl)-L-tyrosin ([^{18}F]FET) als Surrogat verwendet.[44a, 55] [^{18}F]FET wird allerdings im Gegensatz zu 4-[^{18}F]FPhe nicht in Proteine eingebaut. Daher sollte die Aufnahme und Retention von 4-[^{18}F]FPhe im Tumorgewebe potentiell höher sein, was diese Verbindung zu einem sehr gut geeigneten Tracer für die Visualisierung von Hirntumoren machen könnte.

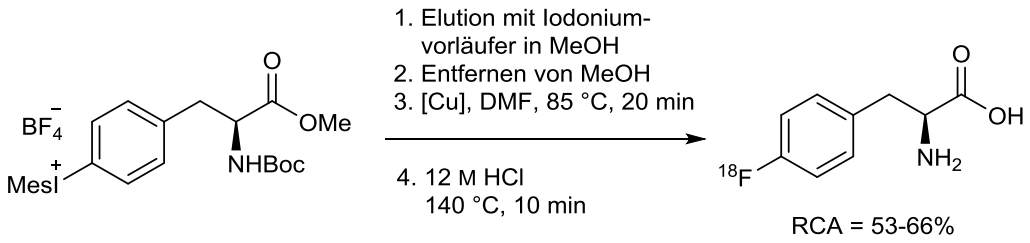


Abbildung 3.7 Synthese von 4-[^{18}F]Fluor-L-phenylalanin (4-[^{18}F]FPhe) über die Cu-vermittelte Radiofluorierung unter „minimalistischen" Reaktionsbedingungen.

Unter den, in dieser Arbeit beschriebenen, „minimalistischen" Reaktionsbedingungen konnte der *N*-Boc geschützte Methylester in RCUs von 81–92% erhalten werden. Saure Hydrolyse mit 12 N HCl ergab nach Isolation mittels HPLC 4-[^{18}F]FPhe in einer RCA von 66% (>98% *ee*). Es wurde eine Dosis von 14 GBq mit einer molaren Aktivität von 109 GBq/µmol aus einer Startaktivität von 49,5 GBq erhalten. Diese Ergebnisse liegen im gleichen Bereich, wie sie auch für [^{18}F]FET in der Literatur beschrieben sind.[19] Somit könnte [^{18}F]FPhe als eine potentielle Alternative zu [^{18}F]FET für die Bildgebung von Hirntumoren mittels PET dienen.

DAA1106 ist ein hoch selektiver Agonist für das mitochondriale 18 kDa Translokatorprotein (TSPO).[56] TSPO ist ein Biomarker für inflammatorische Prozesse im Gehirn, die bei einer Vielzahl von neurodegenerativen Erkrankungen, aber auch nach einem Schlaganfall oder in Folge einer Tumorerkrankung auftreten.[57] Bisher fand DAA1106 nur als, mit ^{11}C markierter, Tracer Anwendung.[58] In neueren Arbeiten konnte [^{18}F]DAA1106 bereits erfolgreich in guten RCUs synthetisiert werden.[36, 59] Die Abtrennung und biologische Evaluierung von [^{18}F]DAA1106 wurde bisher allerdings noch nicht beschrieben.

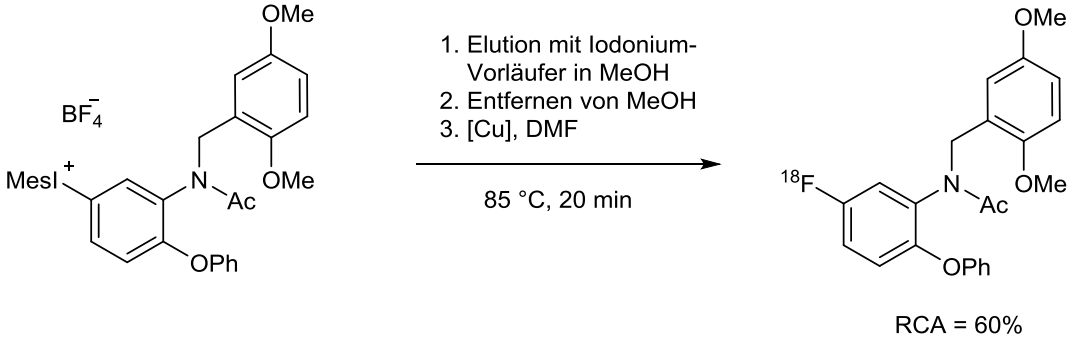


Abbildung 3.8 Synthese von [^{18}F]DAA1106.

Über die Cu-vermittelte Radiofluorierung unter „minimalistischen" Bedingungen konnte [^{18}F]DAA1106 in RCUs von 89–96% erhalten werden. Die anschließende Isolierung über HPLC und Reformulierung in ethanolischer Lösung ergab das Produkt in einer RCA von 60% und einer molaren Aktivität von 66 GBq/µmol.

Für die fertigen Radiopharmaka 6-[^{18}F]FDA, 4-[^{18}F]FPhe und [^{18}F]DAA1106 wurde aus der Formulierungslösung mittels Massenspektrometrie mit induktiv gekoppeltem Plasma (ICP-MS) der Kupfergehalt bestimmt. Mit 0,6–1,4 ppm liegt dieser sowohl deutlich unter der empfohlenen Tagesdosis von 1 mg/d für Erwachsene als auch unter der empfohlenen täglichen Maximaldosis bei parenteraler Gabe (130 µg/d).[60]

Abschließend wurde [^{18}F]DAA1106 *in vivo* auf seine Eignung zur Visualisierung von Neuroinflammation mit Hilfe eines Schlaganfallmodells in Ratten untersucht. Zu diesem Zweck wurde bei einer erwachsenen Long Evans Ratte operativ ein Verschluss der vorderen Gehirnschlagader erzeugt. 6 Tage nach dem Eingriff wurde die Messung mit [^{18}F]DAA1106 durchgeführt. Unmittelbar nach der intravenösen Injektion des Tracers erfolgte eine PET-Messung über eine Dauer von 70 min. Die aus den erhaltenen Daten rekonstruierten PET-Bilder sind in Abb. 3.9 dargestellt.

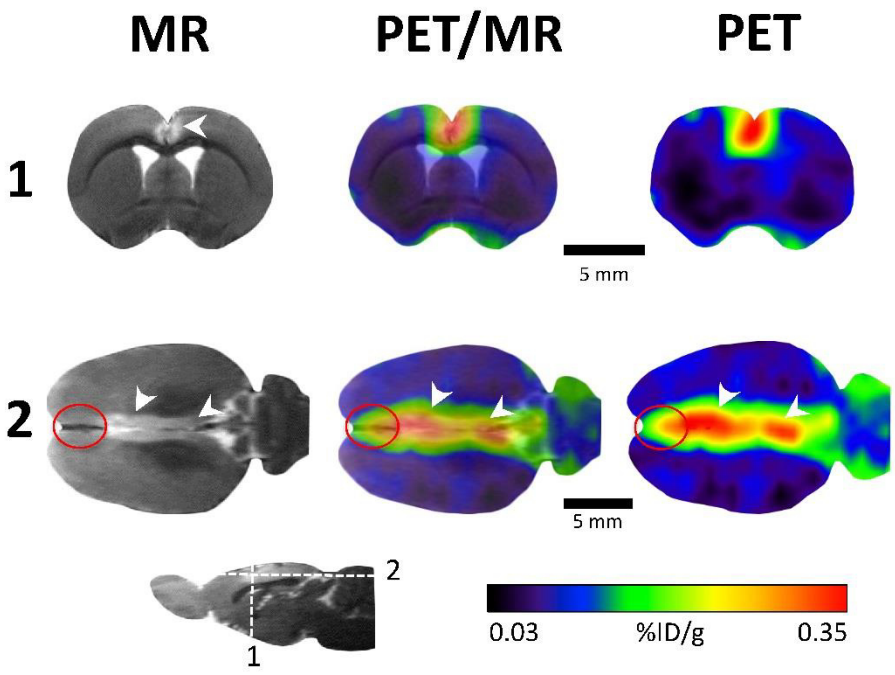


Abbildung 3.9 [^{18}F]DAA1106 PET/MR, sechs Tage nach dem induzierten Arterienverschluss. Gezeigt sind transversale (1) und horizontale (2) Schnittebenen.

Der Bereich erhöhter [^{18}F]DAA1106-Aufnahme überlappt genau mit der ischämische Läsion, die als heller Bereich im MRT-Bild zu sehen ist (weiße Pfeile in Abb. 3.9). Dabei zeigt das Radiopharmakon ein exzellentes Signal zu Hintergrund Verhältnis. Außerdem ist in der PET-Rekonstruktion eine Mehraufnahme an [^{18}F]DAA1106 im rostralen Bereich (roter Kreis in Abb. 3.9) und damit außerhalb des direkt vom Infarkt betroffenen Areals zu beobachten. Bedingt ist dies durch eine Ausdehnung der Neuroinflammation in die Periinfarktzone nach dem Schlaganfall.[61] Im Vergleich dazu ist dieser Bereich im MR-Bild unauffällig.

3.8 Automatisierte Cu-vermittelte Radiofluorierung unter „minimalistischen“ Bedingungen

Durch die stetig wachsende Bedeutung der PET erhöht sich auch der Bedarf an cGMP-konformen Synthesen zur Herstellung von PET-Tracern. Aus diesen Grund und aus Gründen des Strahlenschutzes ist es notwendig, die, im vorherigen Abschnitt beschriebenen Synthesen zu automatisieren, um die Produktion im multi-GBq-Maßstab zu ermöglichen. Nach den erfolgreichen manuellen Synthesen war es das Ziel, die Cu-vermittelte Radiofluorierung unter „minimalistischen“ Bedingungen auf ein automatisiertes Synthesemodul zu übertragen. Für die Entwicklung und den Aufbau einer geeigneten Konfiguration für das Modul (hotbox^{three}, Scintomics) wurde die Etablierung der Synthese der Modellverbindung [^{18}F]FA ausgewählt. Der erste wichtige Schritt für eine erfolgreiche Implementierung war die Beladung und Elution der Ionenaustauschkartusche. Kommerziell erhältliche Module sind standardmäßig so aufgebaut, dass $^{18}F^-$ von einer Seite auf die Kartusche geladen und von derselben Seite eluiert wird. Für eine vollständige Eultion muss diese aber über den weiblichen Luer-Anschluss erfolgen. Bei Beladung und Elution von derselben Seite verbleibt ein erheblicher Teil der Aktivität auf der Kartusche. Für Rückgewinnung von $^{18}F^-$ nach dem „minimalistischen“ Protokoll ist es jedoch essenziell, dass die Targetlösung über den männlichen Luer-Anschluss der Kartusche gesaugt wird. Realisiert wurde dies durch kleinere Modifikationen der Standardkonfiguration an der Syntheseapparatur. In Abbildung 3.10 sind beide Systeme schematisch gezeigt. Durch den Austausch der Gefäße zur Aufnahme der wässrigen ^{18}F-Lösung vom Zyklotron und der des abgetrennten [^{18}O]H_2O war es nun möglich, das „minimalistische“ Elutionsprotokoll auf das Synthesemodul zu übertragen. Wie in Abbildung 3.10 gezeigt, werden bei der klassischen Elutionsmethode (A) sowohl die vom Zyklotron kommende Aktivität als auch die Elutionslösung über 1 auf die Kartusche gegeben. Das restliche Wasser wird über 2 in ein Sammelgefäß geleitet. Für die „minimalistische“ Elutionsmethode wird die Aktivität von „unten“ über 2 in die Kartusche gesaugt. Spülung und Elution erfolgen entgegengesetzt von 1 in Richtung 2. Des Weiteren war es notwendig, das sonst übliche Betriebsgas Helium durch synthetische Luft zu ersetzten. Erste Synthesen von [^{18}F]FA ergaben signifikant niedrigere RCUs als in manuellen Synthesen. Unter Umständen waren Spuren von MeOH dafür verantwortlich. Durch die Verlängerung der Trocknungszeit von 1–2 min auf 5 min konnte die RCA für [^{18}F]FA auf 61% gesteigert werden.

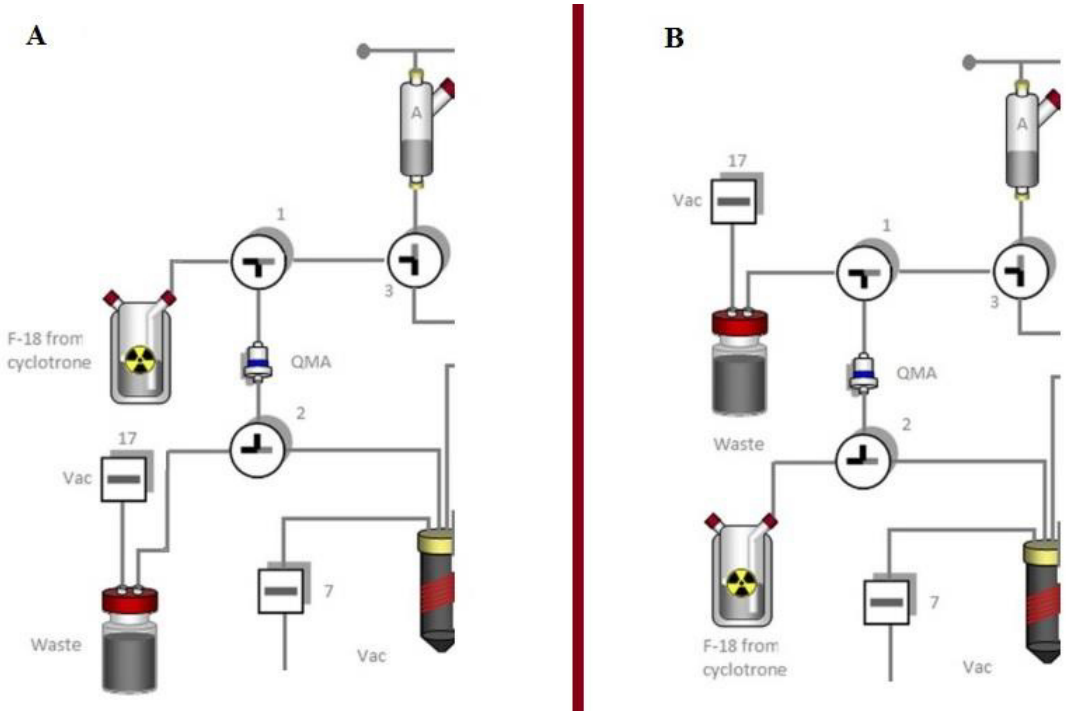


Abbildung 3.10 Flussschemata für verschiedene Synthesemodul-Konfigurationen. Gezeigt sind Schemata für die Standard-Elution (A)[62] sowie für die Rückspülelution (B).

Nachdem ein voll automatisiertes Syntheseprotokoll etabliert worden war, wurde im Folgenden die Synthese von [^{18}F]DAA1106 implementiert. Die Synthese konnte mit der gleichen Modulkonfiguration wie für [^{18}F]FA durchgeführt werden. Innerhalb von 30 min konnte [^{18}F]DAA1106 nach HPLC-Reinigung in einer RCA von 41% und einer molaren Aktivität von 66 GBq/µmol erhalten werden. Alternativ wurde eine Reinigung über SPE etabliert. Wie bei der Isolierung mittels HPLC wurde das Produkt in einer radiochemischen Reinheit (RCR) von >95% erhalten, allerdings beinhaltete die Produktlösung nicht-radioaktive Nebenprodukte.

Für eine Übertragung der Synthese von 4-[^{18}F]FPhe auf das automatische Synthesemodul musste der Aufbau um einen zweiten Reaktor für die Hydrolyse erweitert werden. Nach semi-präparativer HPLC wurde das radiomarkierte Produkt in einer RCA von 56% und einer RCR >95% in injektionsfertiger Lösung isoliert. Eine enantiomere Reinheit von >98% und eine molare Aktivität von 109 GBq/µmol wurden für das erhaltene Produkt ermittelt. Um die Synthese von 60 min zu verkürzen und die Reinigung zu vereinfachen wurde des Weiteren eine Reinigung mittels SPE entwickelt (Abb. 3.11). Hierzu wurde das Reaktionsgemisch mit Wasser

verdünnt und über eine HR-P-Kartusche (Macherey-Nagel) geleitet, auf der, neben dem ^{18}F-markierten Produkt, auch ein Großteil der Verunreinigungen fixiert wurde. Das Rohprodukt wurde mit einer 20%igen ethanolischen in 0,02 M NaH_2PO_4-Phosphatpuffer-Lösung eluiert. Das Eluat wurde über eine C18ec, sowie eine Oasis WAX Kartusche geleitet. Hier wurden die Verunreinigungen, die unpolarer als 4-[^{18}F]FPhe sind, fixiert, während das Produkt diese Kartuschen passieren konnte. Diese Reinigungsmethode ergab das Produkt in einer RCA von 42% und einer molaren Aktivität von 86 GBq/µmol. Trotz der etwas geringeren Ausbeute und molaren Aktivität, die mittels dieses Protokolls erreicht werden konnte, ist diese Methode mit Blick auf die Automatisierung von großem Vorteil.

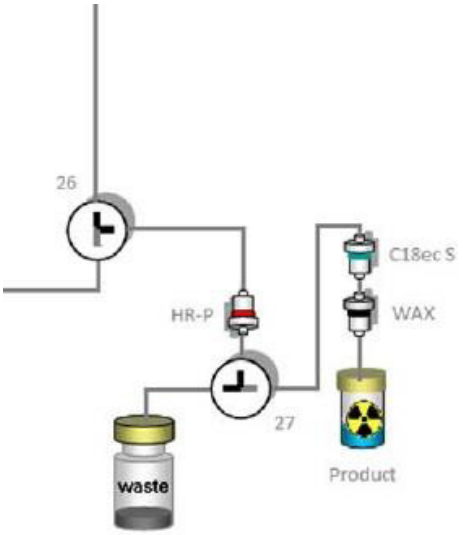


Abbildung 3.11 Schematische Darstellung der SPE-Reinigung von 4-[^{18}F]FPhe.

3.9 Cu-vermittlete Radiofluorierung von Arylboronsäurepinacolestern unter „low-base“ Bedingungen

Tredwell *et al.* beschrieben eine Cu-vermittelte Radiofluorierungsmethode, nach der Arylboronsäurepinacolester mit Hilfe von [^{18}F]KF/K2.2.2 und [$Cu(OTf)_2(Py)_4$] mit ^{18}F markiert werden.[36] Wie schon zuvor für die Fluorierung von Iodoniumsalzen nach Ichiishi *et al.* erwähnt, wurden auch bei dieser Methode zunächst nur Aliquots einer zuvor hergestellten [^{18}F]Fluorid-Lösung verwendet. Somit wurden auch hier etwaige Verluste während der azeotropen Trocknung in den Ausbeuten nicht berücksichtigt. Die Ein-Topf-Synthesen unter konventionellen Bedingungen (2,8 mg K_2CO_3) führten zu extrem niedrigen RCUs von nur 5–7%. Die Aliquotierung der $^{18}F^-$-Lösung ergab hingegen einen Umsatz von 66–76%, was zu der Vermutung führte, dass auch der hier verwendete Cu-Komplex unter basischen Bedingungen nicht ausreichend stabil ist. Dementsprechend wurde das für die Radiofluorierung von Mesitylaryliodonium-Salzen etablierte „low-base“ Protokoll auch auf die ^{18}F-Markierung von Arylboronsäurepinacolestern übertragen. 0,06 mg K_2CO_3 reichten hier für eine nahezu quantitative ^{18}F-Elution und wurde für Ein-Topf-Synthesen unter „low-base“ Bedingungen verwendet.

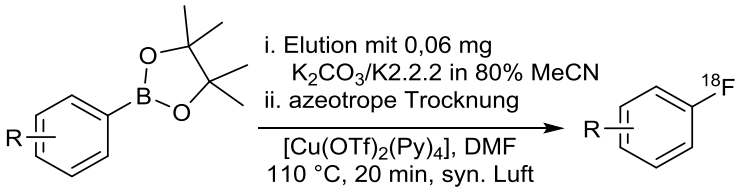


Abbildung 3.12 Cu-vermittelte Radiofluorierung von Arylboronsäurepinacolestern unter „low-base" Bedingungen.

Unter diesen Reaktionsbedingungen konnten, ähnlich wie bei der Radiofluorierung von Mesitylaryliodonium-Salzen, die RCUs auf 41–64% gesteigert werden. Bedingt durch die Beschaffenheit der Vorläufer, die keine Onium-Gruppen und somit kein Anion für die Elution enthalten, ist hier eine Übertragung der „minimalistischen" Methode nicht möglich und somit die Verwendung einer Base nicht vollständig zu vermeiden.

3.10 Unterstützende Wirkung von Alkoholen auf die Cu-vermittelte Radiofluorierung von Arylboronsäuren und Arylboronsäure-pinacolestern

Auch nach der erfolgreichen Übertragung der „low-base" Methode auf die Cu-vermittelte Radiofluorierung von Arylboronsäurepinacolestern bleiben die Ausbeuten noch deutlich unter denen, die für die Cu-vermittelte Radiofluorierung von (Aryl)(mesityl)iodonium-Salz-Vorläufern unter „minimalistischen" Bedingungen erreicht wurden. Da die „minimalistische" Methode hier nicht anwendbar ist, richtete sich das Augenmerk auf die Umgehung der azeotropen Trocknung und die Vermeidung der mit ihr einhergehenden Verluste. Erste Versuche zielten darauf ab, $^{18}F^-$ direkt mit dem aprotischen Reaktionslösungsmittel vom Anionentauscher zu eluieren. Zu diesem Zweck wurden die Kartuschen nach der ^{18}F-Fixierung mit verschiedenen Lösungsmitteln gespült. Im Anschluss wurde $^{18}F^-$ mit einer Lösung aus K_2CO_3, KOTf, K2.2.2 und Pyridin in DMF in ein Vial mit Phenylpinakolboronat (PhBpin) und $Cu(OTf)_2(Py)_4$ eluiert und das resultierende Reaktionsgemisch bei 110 °C für 20 min gerührt. Dabei wurden unerwartet hohe RCUs von 70–80% beobachtet, wenn zuvor Alkohole zur Spülung der Kartusche verwendet wurden. Dies führte zu der Vermutung, dass geringe Mengen Alkohol, die auf der Kartusche verblieben und mit in das Reaktionsgemisch eluiert wurden, die Radiomarkierung unterstützten. Um diese Hypothese zu überprüfen, wurde der Einfluss von Alkoholen auf die Cu-vermittelte Radiofluorierung von Arylboronsäurepinacolestern und Arylboronsäuren systematisch untersucht. Diese Experimente wurden wie folgt durchgeführt: $^{18}F^-$ wurde von einer QMA-Kartusche mit einer Lösung aus Et_4NHCO_3 in dem jeweiligen

Alkohol direkt in ein Reaktionsgefäß, in dem PhBpin bzw. Phenylboronsäure [$PhB(OH)_2$] und $Cu(OTf)_2(Py)_4$ in DMA vorgelegt war, eluiert.

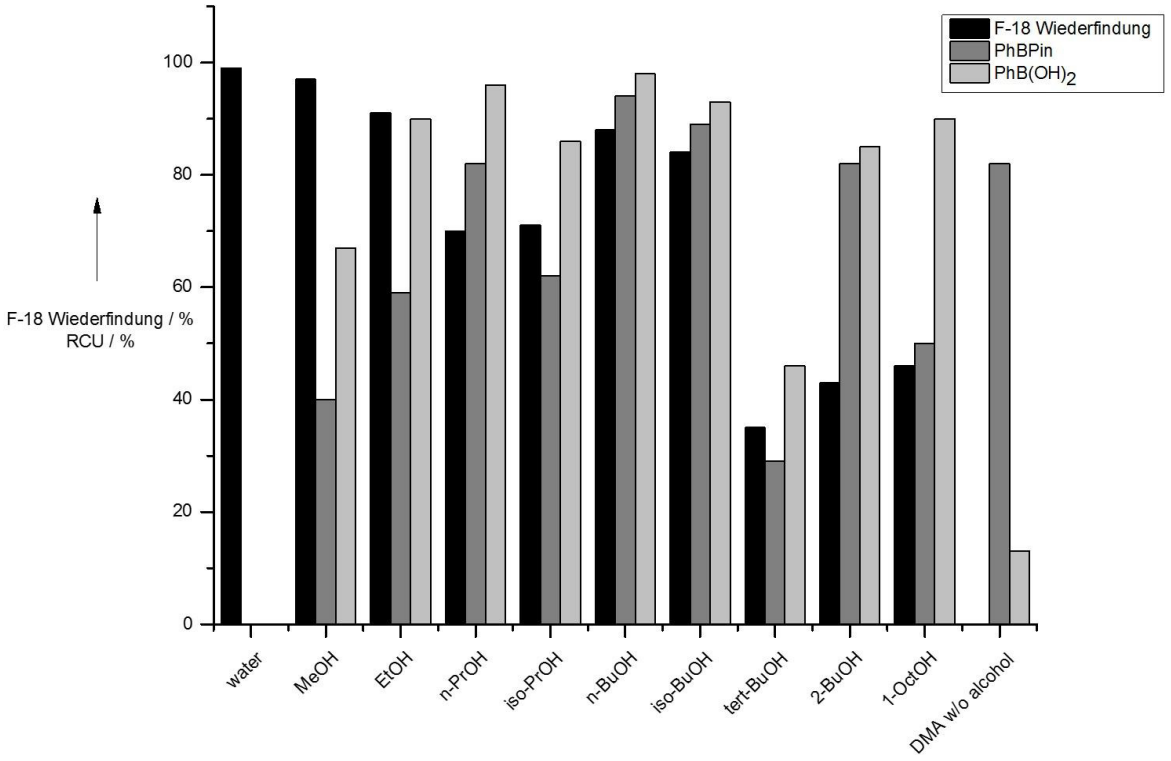


Abbildung 3.13 ^{18}F-Wiederfindungsraten und RCCs bei Verwendung verschiedener Alkohole als Co-Solvents.

Der Wasser- bzw. Alkoholanteil im Reaktionsmedium betrug jeweils 33%. Die erhaltene Reaktionslösung wurde für 20 min bei 110 °C gerührt und der RCU mittels analytischer HPLC bestimmt. Es zeigte sich, dass die Zugabe von Alkoholen zum Reaktionsmedium zu deutlich erhöhten Umsätzen, im Vergleich zu reinem DMA (in der Abb. 3.13 ganz rechts) führte, insbesondere bei Arylboronsäure-Vorläufern. Wie die Mitte von Abb. 3.13 zeigt, hatte *n*-BuOH einen stärksten unterstützenden Effekt auf die RCU. Auch wurde bei *n*-BuOH eine hohe ^{18}F-Wiederfindungsrate von 80–90% beobachtet.,

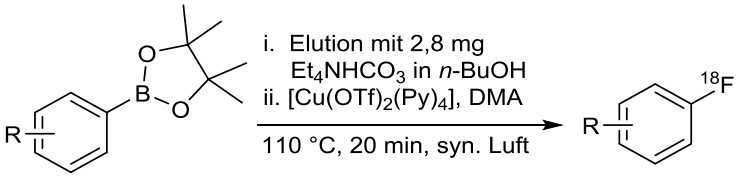


Abbildung 3.13 Alkohol-unterstütze Cu-vermittelte Radiofluorierung von Arylboronsäurepinacolestern.

In weiteren Experimenten wurde untersucht, wie die RCU durch den Alkoholgehalt beeinflusst wird. Es zeigte sich, dass bereits kleine Mengen *n*-BuOH (5%) zu deutlich höheren Umsätzen führten und der Alkoholanteil auf zu bis zu 60% gesteigert werden konnte, ohne

einen nachteiligen Effekt auf die Radiofluorierung von PhBPin zu zeigen. Auch durch eine weitere Erhöhung der *n*-BuOH Konzentration auf bis zu 80% fiel der RCU von [^{18}F]FB nicht unter 80%. Auffällig war überdies, dass $PhB(OH)_2$ selbst in 33% EtOH in sehr hohen RCUs (90%) mit ^{18}F markiert werden konnte. Auf Grund dieser Beobachtung wurde der Einfluss von EtOH auf den RCU untersucht. Bereits ein Alkoholanteil von 5% reichte aus, um eine deutliche Steigerung der RCUs zu erreichen. Bei höheren Mengen von bis zu 50% konnten weiterhin Umsätze von >90% beobachtet werden. Selbst in reinem EtOH wurde [^{8}F]FB mit einem Umsatz von 45% erhalten (vgl. Abb. 3.14).

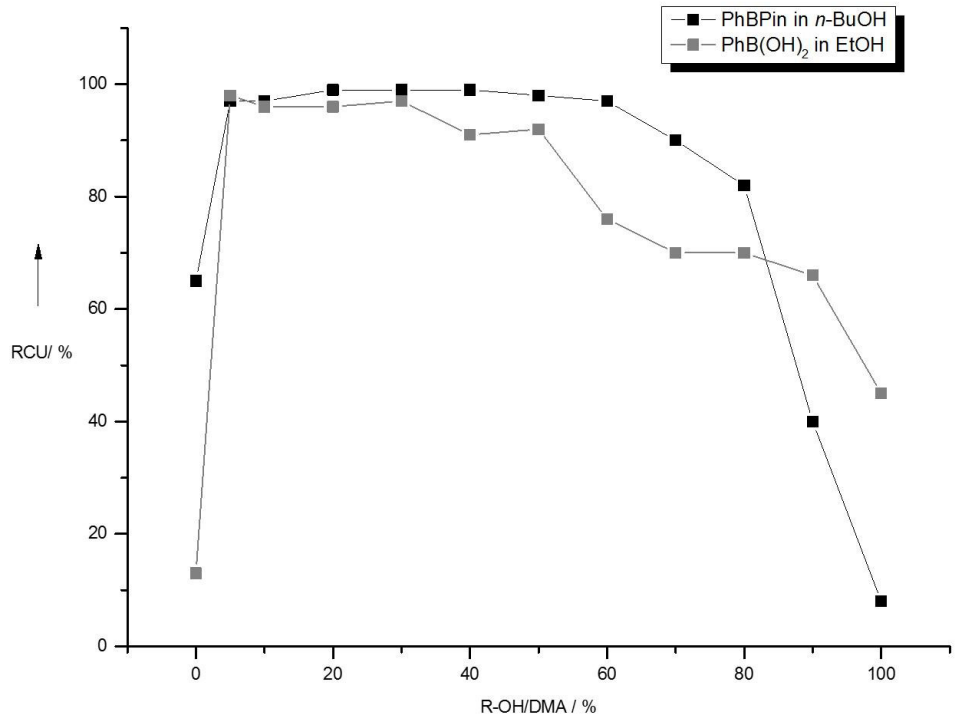


Abbildung 3.14 Abhängigkeit der RCU vom Alkoholanteil im Reaktionsmedium.

Diese Beobachtungen sind mit den Ergebnissen aus der bisherigen Literatur nicht vereinbar. So gelten protische Lösungsmittel, aufgrund ihrer Tendenz, Wasserstoffbrückenbindungen zu bilden, die die Nukleophilie von Fluorid sehr stark erniedrigen, als ungeeignet für S_NAr Fluorierungen.[63] Daher ist dies auch die erste Übergangsmetall-vermittelte aromatische Fluorierung in alkoholischem Medium, die bisher beschrieben wurde.

Für aliphatische Fluorierungsreaktionen ist dagegen der unterstützende Effekt allerdings von tertiären Alkoholen in der Literatur beschrieben.[64] Zurückgeführt wird dieser im Wesentlichen auf vier Einflussmöglichkeiten des Alkohols auf die Reaktion.[65]

- ❖ Schwächung der ionischen Bindung zwischen Fluorid und dem Gegenion durch die Bildung von Wasserstoffbrückenbindungen zwischen dem Alkohol und dem Fluoridion.
- ❖ Durch die geringe Solvatisierung mit dem Alkohol bleibt die Nucleophilie von Fluorid größtenteils erhalten.
- ❖ Wasserstoffbrückenbindungen zwischen dem Alkohol und der Abgangsgruppe erleichtern die Substitution.
- ❖ Wasserstoffbrückenbindungen zwischen dem Alkohol und reaktiven Heteroatomen im Substrat unterdrücken mögliche Nebenreaktionen.

Auf diese Arbeit übertragen, könnte die unterstützende Wirkung des Alkohols auf die Cu-vermittelte Radiofluorierung von Arylboronsäuren, -boronsäurepinacolestern und –stannanen wie folgt erklärt werden:

- ❖ Durch Solvatisierung des Fluoridions (vgl. Abb. 3.15) schwächt der Alkohol die ionische Bindung zum Tetraethylammonium-Kation.
- ❖ Die Anionenmetathese zum Cu(OTF)F-Komplex wird durch Wasserstoffbrückenbindungen zwischen dem Alkohol und der Triflatgruppe begünstigt.

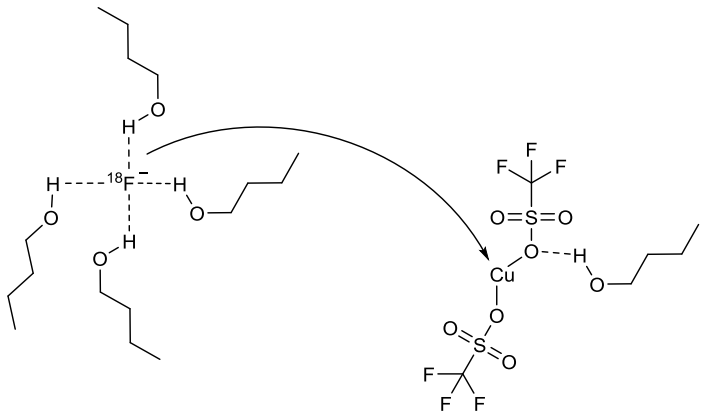
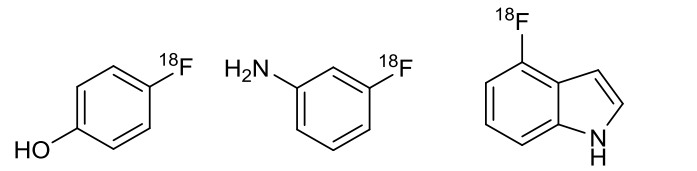


Abbildung 3.15 Mögliche Einflüsse des Alkohols auf die Cu-vermittelte Radiofluorierung von Arylboronsäuren.

Die Vielseitigkeit der Alkohol-unterstützen Cu-vermittelten Radiofluorierung wurde mit einer Auswahl an elektronenreichen, -armen und -neutralen Modellsubstraten, die mit teils sehr guten RCUs ^{18}F-markiert werden konnten, gezeigt. Einzig N- und O-Substituenten in *ortho*-Position führten zu geringeren Umsätzen, vermutlich aufgrund von Wechselwirkungen zwischen Substituent und Abgangsgruppe. Darüber hinaus gelang es, radiofluorierte Phenole

und Aniline direkt aus den jeweiligen ungeschützten Vorläufern zu synthetisieren. [^{18}F]Fluorindole wurden ebenfalls direkt, das heißt ohne vorherige Schützung des Stickstoffs, in nahezu quantitativen Umsätzen erhalten (Abb. 3.16).



4-[^{18}F]Fluorphenol	3-[^{18}F]Fluoranilin	4-[^{18}F]Fluor-1*H*-indol
81%[a]	3%[a]	99%[a]
88%[b]	21%[b]	99%[b]

Abbildung 3.16 RCUs für ^{18}F-markiertes Phenol, Anilin und Indol aus [a] Boronsäurepinacolester- und [b] Boronsäure-Vorläufern.

Als weitere potentielle Substrate wurden Trimethylstannyl-Verbindungen untersucht. Makaravage *et al.* beschrieben bereits eine Cu-vermittelte Radiofluorierung von Stannanen., die in der Radiochemie schon als Substrate für Radioiodierungen und elektrophile Radiofluorierungen angewendet werden.[42] Auch hier wird hydrolyseempfindliches $Cu(OTf)_2$ als Cu-Quelle verwendet und $^{18}F^-$ aufwendig mittels azeotroper Trocknung vorbereitet. Die Übertragung der Alkohol-unterstützen Cu-vermittelten Radiofluorierung auf die ^{18}F-Markierung von Trimethylstannyl-Vorläufern ergab [^{18}F]FB und [^{18}F]DAA1106 unter nicht weiter optimierten Reaktionsbedingungen in RCUs von 66 bzw. 31%.

3.11 Alkohol-unterstützte, Cu-vermittelte Radiosynthese von 6-[^{18}F]FDOPA und 6-[^{18}F]FDA

Um die Alkohol-unterstütze Cu-vermittelte Radiofluorierung mit den anderen hier beschriebenen Synthesestrategien vergleichen zu können, und ihre Eignung für die Radiopharmakaproduktion zu untersuchen, wurden als klinisch relevante Radiotracer 6-[^{18}F]FDA und 6-[^{18}F]FDOPA synthetisiert.

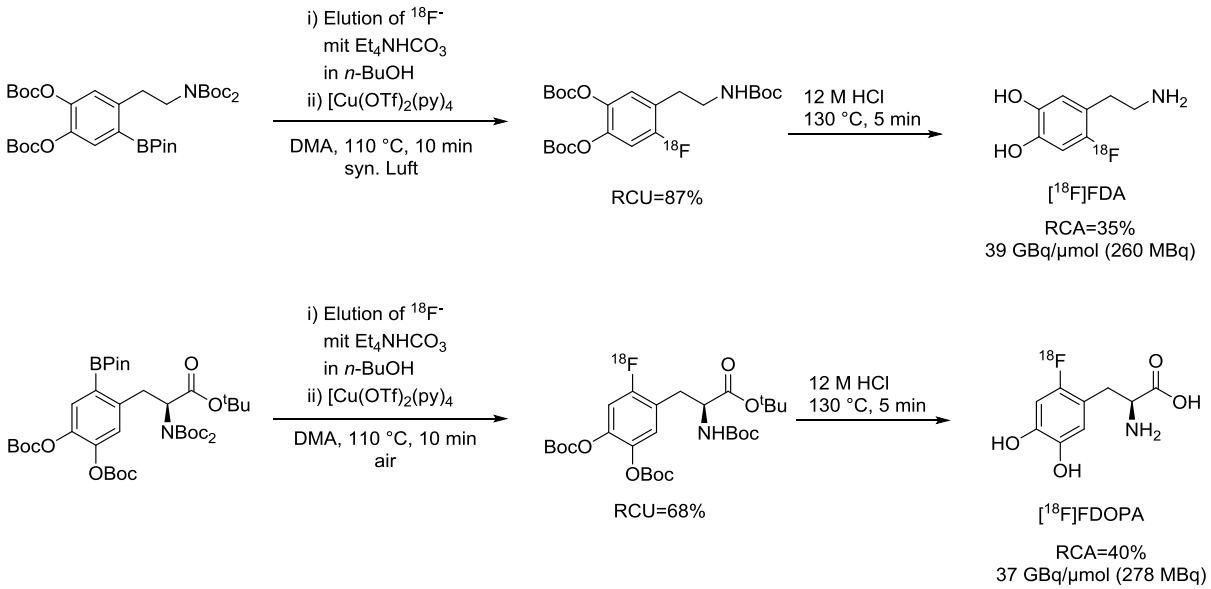


Abbildung 3.17 Synthese von 6-[^{18}F]FDA und 6-[^{18}F]FDOPA unter Alkohol-unterstützen Bedingungen.

Unter optimierten Reaktionsbedingungen konnten die geschützten, ^{18}F-markierten Intermediate aus *N,N,O,O*-geschützen Vorläufern mit RCUs von 68 bzw. 87% erhalten werden. Dabei war die vollständige Abspaltung einer *N*-Boc-Schutzgruppe sowie die partielle Abspaltung einer *O*-Boc-Gruppe während der Reaktion zu beobachten. Nach saurer Hydrolyse wurden 6-[^{18}F]FDA und 6-[^{18}F]FDOPA in RCAs von 35 und 40% und molaren Aktivitäten von 39 und 37 GBq/µmol isoliert. Der Cu-Gehalt der Produktlösungen wurde mittels ICP-MS bestimmt und belief sich auf 6,54 µg/Synthese für 6-[^{18}F]FDA und 5,92 µg/Synthese für 6-[^{18}F]FDOPA. Auch diese Werte liegen weit unterhalb der Richtwerte, bei denen toxische Wirkungen zu erwarten sind.[60]

3.12 Vergleich der Übergangsmetall-vermittelten Radiofluorierungs-strategien

Tabelle 3.3 Zusammenfassung der RCUs und RCAs für 6-[^{18}F]FDA

	Ni-vermittelt	Cu-vermittelt (Iodoniumvorläufer)	Cu-vermittelt (Alkohol-unterstützt)
RCU [%]	18	94	87
RCA [%]	12	46	35

Ziel der vorliegenden Arbeit war es, aromatische Verbindungen als Tracer für die PET in hohen Aktivitätsmengen herzustellen. Über die, in der Literatur beschriebenen, elektrophilen und nucleophilen Synthesestrategien sind derartige Verbindung bisher nur in geringen

Ausbeuten zugänglich, was ihre routinemäßige Anwendung für die PET stark limitiert. Durch die, in dieser Arbeit entwickelten innovativen Radiofluorierungsstrategien konnten effiziente Syntheserouten etabliert werden, die gleichzeitig zu hohen Ausbeuten führten. Dazu wurden verschiedene Synthesewege, die über Übergangsmetall-vermittelte Reaktionen abliefen etabliert, die die Herstellung ^{18}F-markierter Arene ermöglichte, die auf anderem Wege gar nicht oder nur schwer zugänglich waren. Wie die Übersicht in Tab 3.3 und die Ergebnisse dieser Arbeit zeigen, kommen nur Cu-basierte Methoden für eine praktische Anwendung in Frage. Während die Ni-vermittelte Radiofluorierung nur geringe RCUs und RCAs liefert, konnte mit den Cu-vermittelten ^{18}F-Markierungsverfahren deutlich höhere Ausbeuten erzielt werden. Die vorliegende Arbeit zeigt zudem, dass sich diese Methoden für eine routinemäßige Herstellung von aromatischen Radiopharmaka eignen. Vergleicht man die beiden, in dieser Arbeit entwickelten Cu-vermittelten Methoden untereinander, so erscheint zunächst die Methode, die von Iodoniumsalzen als Vorläufer ausgeht, aufgrund der etwas höheren RCAs als das überlegene Verfahren. Dieser Vorteil relativiert sich jedoch, wenn man die eingeschränkte Zugänglichkeit der Iodonium-Vorläufer berücksichtigt. Des Weiteren ist es notwendig, den $(MeCN)_4CuOTF$-Komplex bei längerer Lagerzeit unter inerten Bedingungen aufzubewahren, was die Handhabung ebenfalls erschwert. Demgegenüber nutzt die Alkohol-unterstützte Cu-vermittelte Radiofluorierung Vorläufer, wie Arylboronsäuren, -boronsäurepinacolestern und –stannanen, die im Gegensatz zu Iodoniumsalzen, vielfach kommerziell erhältlich und somit wesentlich leichter zugänglich sind. Da diese Vorläufer unter identischen Reaktionsbedingung radiofluoriert werden können, entfallen aufwendige Optimierungsarbeiten für jeden einzelnen Vorläufer. Darüber hinaus ermöglicht die Alkohol-unterstützte Cu-vermittelte Radiofluorierung den Zugang zu einem breiten Spektrum an, mit ^{18}F markierten aromatischen Verbindungen mit guten bis exzellenten RCUs. Insbesondere sind hierbei Indole und geschützte Tryptophane zu erwähnen, die nahezu quantitativ radiofluoriert werden konnten. In diesem Zusammenhang zeigte sich auch die große Toleranz der Alkohol-unterstützten Cu-vermittelten Radiofluorierung gegenüber funktionellen Gruppen. Neben Indolen konnten auch Phenole und Aniline direkt aus den ungeschützten Vorläufern in guten RCUs synthetisiert werden. Neben diesen Vorteilen erlaubt es die Alkohol-unterstützte Synthesestrategie, auf zeitintensive Trocknungsschritte, die bei bisher bekannten Methoden stets zu großen Aktivitätsverlusten führten, zu verzichten.

Die Alkohol-unterstützte Cu-vermittelte Radiofluorierung stellt eine äußerst robuste und effiziente Methode für die Produktion verschiedener aromatischer Radiopharmaka dar, die es ermöglichen wird, innovative PET-Tracer der nächsten Generation zu entwickeln. Dies wird

zukünftig völlig neue diagnostische Möglichkeiten eröffnen, die die Basis für effizientere Behandlungsstrategien und letztlich der Patientenversorgung bilden.

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Abkürzungsverzeichnis

6

6-OHDA 6-Hydroxydopamin

Å

Å Ångström

B

Boc .....tert-Butyloxycarbonyl

Bq .....Bequerel

BTMG 2-tert-Butyl-1,1,3,3-tetramethylguanidin

BuOH ..... Butanol

C

c.a .....Carrier-added

cGMP ..... Current Good Manufacturing Practice

CiCurie

COX-2 ..... Cyclooxygenase-2

CT.....Computertomographie

D

DAA1106 ..... (N-5-Fluor-2-phenoxyphenyl)-N-(2,5-dimethoxybenzyl)acetamid

DMA.....Dimethylacetamid

DMF ..... Dimethylformamid

E

eV ..... Elektronenvolt

F

FA.....Fluoranisol

FB .....Fluorbenzol

FBA ..... Fluorbenzaldehyd

FDA 6-Fluordopamin

FDOPA..... 6-Fluor-L-3,4dihydroxyphenylalanin

FET ..... 4-(2-Fluorethyl)-L-tyrosin
FMT 6-Fluor-L-m-tyrosin
FPEB ..... 3-Fluor-5-[(Pyridin-3-yl)-ethynyl]benzonitril
FPhe 4-Fluor-L-phenylalanin

H

HPLC ..... Hochleistungsflüssigkeitschromatographie

I

ICP-MS ..... Massenspektrometrie mit induktiv gekoppeltem Plasma

K

K2.2.2 ..... 1,10-diaza-4,7,13,16,21,24-hexaoxabicyclo[8.8.8]hexacosan

M

MeCN ..... Acetonitril
MeOH ..... Methanol
mg ..... Milligramm
min ..... Minute
mm ..... Millimeter
MPPF ..... 4-Fluoro-N-[2-[4-(2-methoxyphenyl)-1-piperazinyl]ethyl]-N-(2-pyridyl)benzamid
MRT ..... Magnetresonanztomographie

N

n.c.a ..... no-carrier-added

O

OTf ..... Trimethansulfonat

P

PET ..... Positronen-Emissions-Tomographie
ppm ..... parts per million, deutsch: Millionstel

R

RCA ..... radiochemische Ausbeute
RCR ..... radiochemische Reinheit
RCU ..... radiochemischer Umsatz

S

SUV .....standardized uptake value

T

TSPO .....mitochondriale 18 kDa Translokatorprotein

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Anhang

A. Eigener Anteil an den, dieser Arbeit zugrundeliegenden Publikationen

B. D. Zlatopolskiy, ‡ **J. Zischler**, ‡ E. A. Urusova, H. Endepols, E. Kordys, H. Frauendorf, F. M. Mottaghy, B. Neumaier#, *ChemistryOpen* **2015**, *4*, 457-462.

Das Konzept dieser Publikation wurde gemeinschaftlich von allen Autoren erstellt. Die radiochemische Umsetzung, einschließlich aller Optimierungen, erfolgte durch J. Zischler. Organische Vorläufersynthesen wurden durch J. Zischler und Dr. B. D. Zlatopolskiy durchgeführt. Das Manuskript wurde gemeinschaftlich von J. Zischler, Dr. B. D. Zlatopolskiy, PD Dr. H. Endepols und Prof. Dr. B. Neumaier erstellt.

B. D. Zlatopolskiy, ‡ **J. Zischler**, ‡ P. Krapf, F. Zarrad, E. A. Urusova, E. Kordys, H. Endepols, B. Neumaier#, *Chem. Eur. J.* **2015**, *21*, 5972-5979.

Das Konzept der Publikation wurde von J. Zischler und Dr. B. D. Zlatopolskiy, unterstützt von Prof. Dr. B. Neumaier, erarbeitet. Die experimentelle Umsetzung, Datenerhebung und Auswertung sowie die Entwicklung der präparativen Tracersynthesen erfolgte durch J. Zischler. Das Manuskript wurde gemeinsam von J. Zischler, Dr. B. D. Zlatopolskiy und Prof. Dr. B. Neumaier erstellt.

J. Zischler, ‡ P. Krapf, ‡ R. Richarz, B. D. Zlatopolskiy, B. Neumaier#, *Appl. Radiat. Isot.* **2016**, *115*, 133-137.

Das Konzept dieser Publikation wurde gemeinsam von allen Autoren erarbeitet. Entwicklung und Aufbau des Synthesemoduls erfolgte durch J. Zischler. Synthesen und Datenerhebung wurden durch J. Zischler, Dr. P. Krapf und Dr. R. Richarz durchgeführt. Die Entwicklung und Etablierung der Kartuschenreinigung erfolgte durch J. Zischler. Das Manuskript wurde gemeinschaftlich von J. Zischler, Dr. P. Krapf, Dr. B. D. Zlatopolskiy und Prof. Dr. B. Neumaier erstellt.

J. Zischler, N. Kolks, D. Modemann, B. Neumaier#, B. D. Zlatopolskiy, *Chem. Eur. J.* **2017**, *23*, 3251-3256

‡ Geteilte Erstautorenschaft
# Korrespondierender Autor

Die Konzeption dieser Publikation wurde von J. Zischler, Dr. B. D. Zlatopolskiy und Prof. Dr. B. Neumaier erarbeitet und erstellt. Die radiochemische Umsetzung, inklusive Datenerhebung und –auswertung, erfolgte durch J. Zischler. Das Manuskript wurde durch J. Zischler, Dr. B. D. Zlatopolskiy und Prof. Dr. B. Neumaier.

B. Erklärung gemäß § 4 Abs 1 Punkt 9

"Ich versichere, dass ich die von mir vorgelegte Dissertation selbständig angefertigt, die benutzten Quellen und Hilfsmittel vollständig angegeben und die Stellen der Arbeit − einschließlich Tabellen, Karten und Abbildungen −, die anderen Werken im Wortlaut oder dem Sinn nach entnommen sind, in jedem Einzelfall als Entlehnung kenntlich gemacht habe; dass diese Dissertation noch keiner anderen Fakultät oder Universität zur Prüfung vorgelegen hat; dass sie – abgesehen von unten angegebenen Teilpublikationen − noch nicht veröffentlicht worden ist sowie, dass ich eine solche Veröffentlichung vor Abschluss des Promotionsverfahrens nicht vornehmen werde.

Die Bestimmungen der Promotionsordnung sind mir bekannt. Die von mir vorgelegte Dissertation ist von Univ. Prof. Dr. Bernd Neumaier betreut worden."

Köln, 03.04.2017……

(Johannes Zischler)

C. Curriculum vitae

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Besondere Kenntnisse

Sprachkenntnisse:	Deutsch (Muttersprache), Englisch (sehr gut in Wort und Schrift)
EDV:	MS-Office, Windows, ChemBioOffice, MestRiNova, Origin, Photoshop, Endnote, Zotero, Joomla

Publikationen

J. Zischler, N. Kolks, D. Modemann, B. Neumaier*, B. D. Zlatopolskiy, *Chem. Eur. J.* **2017**, *23*, 3251-3256

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Tagungsbeiträge

2016 Resuscitation Science Symposium (ReSS):

D. de la Puente Bethencourt, D. C. Schroeder, E. Popp, **J. Zischler**, T. Annecke, T. Hucho, B. Neumaier, B. W. Böttiger, H. Endepols, *Circulation* **2016**, *134*, A19227. (Young Investigator-Award)

18. Hauptstadtkongress der DGAI für Anästhesiologie und Intensivtherapie (HAI):

Schroeder D.C., Popp E., De la Puente Bethencourt D., **Zischler J.**, Annecke T., Hucho T., Neumaier B., Böttiger BW., Endepols H., *HAI* **2016**, P042

29th Annual Congress of the European Association of Nuclear Medicine (EANM):

J. Zischler, B. D. Zlatopolskiy, P. Krapf, F. Zarrad, E. A. Urusova, E. Kordys, H. Endepols, B. Neumaier; *Eur. J. Nucl. Med. Mol. Imaging* **2015**, 42 (Suppl. 1): S 42

The 21th International Symposium on Radiopharmaceutical Sciences (ISRS):

J.Zischler, B. D. Zlatopolskiy, P. Krapf, F. Zarrad, E. A. Urusova, E. Kordys, H. Endepols, B. Neumaier; *J. Label. Compd. Radiopharm.* **2015**, 58 (S1): S158

J. Zischler, B. D. Zlatopolskiy, E. A. Urusova, B. Neumaier; *J. Label. Compd. Radiopharm.* **2015**, 58 (S1): S202

53. Jahrestagung der Deutschen Gesellschaft für Nuklearmedizin (DGN):

P. Krapf, **J. Zischler**, B. D. Zlatopolskiy, B. Neumaier, *Nuklearmedizin* 2015; 54(2): A65.

J. Zischler, B. D. Zlatopolskiy, P. Krapf, F. Zarrad, E. A. Urusova, E. Kordys, H. Endepols, B. Neumaier, *Nuklearmedizin* **2015**; 53 (V66).

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