ChemPubSoc Europe

DOI: 10.1002/ejoc.201600705

EurJOC European Journal of Organic Chemistry

Full Paper

Radiopharmaceuticals

Preparation of No-Carrier-Added 6-[^{18}F]Fluoro-L-tryptophan via Cu-Mediated Radiofluorination

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Abstract: ^{18}F-Labeled aromatic amino acids exhibit great potential for diagnostic applications using positron emission tomography. However, the introduction of ^{18}F into aromatic compounds remains challenging, and novel fluorination methods facilitating easy access to ^{18}F-labeled arenes are highly sought after. In recent years, novel metal-mediated fluorination methods have been reported and transferred into radiochemistry. Based on Cu-mediated radiofluorination, a two-step synthesis of no-carrier-added (n.c.a.) 6-[^{18}F]fluoro-L-tryptophan was developed. 6-[^{18}F]Fluoro-L-tryptophan was synthesized with an overall radochemical yield of 16 ± 4% within 110 min and a specific activity of 280 GBq μmol^{-1}. The radiochemical purity was more than 99 %. The developed method allowed access to radiofluorinated tryptophan derivatives in high radiochemical yields and opens new ways to provide radiofluorinated amino acids. Furthermore, the reaction conditions were optimized to facilitate automation.

Introduction

Positron emission tomography (PET) is one of the most important molecular imaging techniques in clinical practice. PET imaging exploits the decay of positron emitters. The emitted positron is annihilated when in contact with an electron and produces a pair of photons that can be detected by a dedicated PET scanner, revealing a 3D-image of the radionuclide distribution. For these purposes, different PET nuclides, such as ^{11}C, ^{13}N, ^{15}O, and ^{18}F are available.[1] Among them, ^{18}F plays an outstanding role because of its favorable decay properties with low positron emission but highly intense energy and convenient half-life of 109.7 min, enabling multistep radiosyntheses to be performed.

The indole motif occurs in numerous natural products,[2] as well as in the essential α-amino acid tryptophan, which is involved in a range of physiological processes. In vivo, tryptophan is either converted enzymatically over two steps into serotonin, which is involved in various neurological functions and diseases,[3] or it is metabolized through the kynurenine pathway.[4] Kynurenine seems to be involved in many neurodegenerative disorders such as Alzheimer's disease,[5] Huntington's disease,[6] and multiple sclerosis.[7] Recently published results describe kynurenine as an important factor in "progressive" tumor growth and immune system suppression.[8] As described by Optiz et al.,[9] some tumors overexpress the liver enzyme tryptophan dioxygenase (TDO), resulting in an increase of tryptophan uptake into the tumor cells. Furthermore, tryptophan participates in the serotonin pathway and therefore a radiolabeled variant of tryptophan could enable to detect small alterations of tryptophan uptake in regions of serotonergic neurons to be traced.

First attempts to radiolabel tryptophan with positron emitters by using nucleophilic aromatic substitution (S_NAr) were reported by Atkins et al. in 1972.[10] In their work, the Balz–Schiemann reaction with subsequent hydrolysis was applied, providing carrier-added (c.a.) 5- or 6-[^{18}F]fluoro-D/L-tryptophan as a racemic mixture in radiochemical yields (RCY) of 7–10 %. The biodistribution of 5- and 6-[^{18}F]fluoro-D/L-tryptophan was studied in mice and rats. However, the results were of limited value because 6-fluorotryptophan is itself an inhibitor of tryptophan hydroxylase and lower molar concentrations are mandatory.[3e,11] Moreover, the lack of enantioselectivity and low specific activities precluded further investigations. The uptake of the tryptophan analogue 6-fluorotryptophan through the blood brain barrier (BBB) has been confirmed[12] and, furthermore, the interaction of 6-fluorotryptophan with tryptophan hydroxylase was examined.[3e]

Alternative approaches to ^{18}F-labeled tryptophan derivatives include electrophilic ^{18}F-fluorination of 5-hydroxytryptophan (5-HT) leading to a mixture of 4- and 6-[^{18}F]fluoro-5-hydroxytryptophan.[13] Moreover, ^{18}F-fluoroalkylation reactions at different positions of the indole motif were carried out.[14] Recently, a three-step synthesis using isotopic exchange through S_NAr was reported, giving c.a. 4-[^{18}F]fluoro-L-tryptophan in 13 % RCY and a specific activity of 70 MBq mmol^{-1}.[15]

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Supporting information and ORCID(s) from the author(s) for this article are available on the WWW under http://dx.doi.org/10.1002/ejoc.201600705.

In the last few years, several innovative ^{18}F-fluorination methods via S_NAr have been developed that allow direct introduction of ^{18}F into electron-rich or electron-neutral arenes starting from n.c.a. nucleophilic [^{18}F]fluoride.[16] Besides reactions using iodonium salts[17] as labeling precursors, transition-metal-mediated reactions under exceptionally mild reaction conditions were published. However, Pd-[18] or Ni-mediated[18c,19] ^{18}F-fluorination reactions turned out to be very air- and moisture-sensitive and therefore difficult to be applied in clinical routine.[20] Further developments were recently reported by Ichiishi et al.,[21] Tredwell et al.,[22] and Mossine et al.[23] using copper-mediated ^{18}F-fluorination of (mesityl)(aryl) iodonium salts, pinacol boronate esters or boronic acids, respectively. By using pinacol-derived aryl boronate esters as precursors, Tredwell et al. obtained protected 5-[^{18}F]fluoroindole in 19 % RCY.[22]

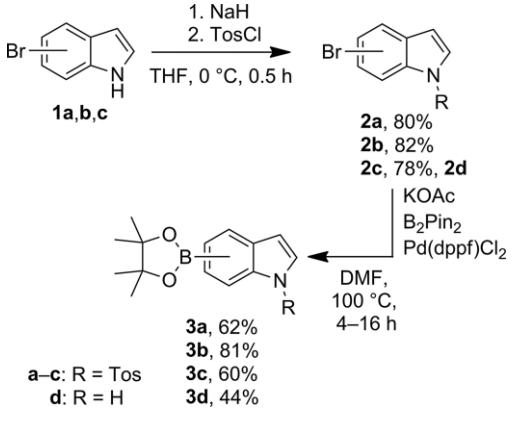
The aim of this work was firstly to determine the most reactive position of the indole scaffold towards substitution using copper-mediated ^{18}F-fluorination. Then, a method to prepare [^{18}F]fluorotryptophan derivatives in high specific activity and enantiomeric excess was developed.

Results and Discussion

Radiofluorination of Indolyl Pinacolyl Boronates

Synthesis of Indole Precursors

In preliminary studies, the most reactive substitution position of indole using the copper-mediated ^{18}F-fluorination had to be determined.[22] Therefore, an appropriate precursor synthesis was developed. As depicted in Scheme 1, the precursor synthesis started from the corresponding bromo-indole derivatives **1a**–**d**. The nitrogen of the bromoindole derivatives **1a**–**c** was protected with *p*-toluenesulfonyl chloride (TosCl) yielding **2a**–**c**. In the case of 7-bromo-1*H*-indole, the indole nitrogen could not be protected using tosyl- or *tert*-butyloxycarbonyl- (Boc-) as protecting groups, probably because of steric hindrance. Subsequently, bromine derivatives in the 4-, 5-, 6- or 7-position of the indole motif (**2a**–**d**) were substituted by a pinacol boronic ester group by using the Suzuki–Miyaura coupling reaction.[24]

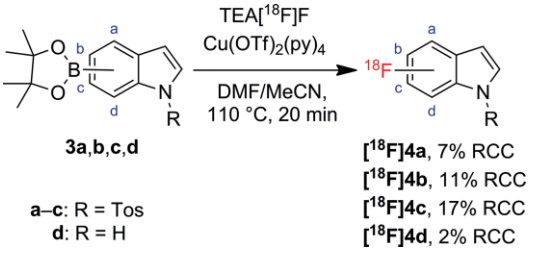


Scheme 1. Synthesis of indole precursors for radiolabeling and respective yields.

Protection of the indole nitrogen by using tosyl chloride was achieved in yields of 80 ± 2 %. The Suzuki–Miyaura coupling generated the boron pinacol ester substituted indole derivatives[22] in yields of 54–72 %. However, ^{18}F-labeling experiments of the unprotected compounds revealed that protective groups were not mandatory.

Radiolabeling of Indolyl Pinacolyl Boronates

[^{18}F]Fluoride obtained as an aqueous solution was fixed on a QMA®-cartridge and eluted with Et_4NHCO_3 ($TEAHCO_3$) in methanol (cf. optimization of radiosynthesis). After removal of methanol in vacuo, tetrakis(pyridine)copper(II)triflate [$Cu(OTf)_2(py)_4$] and the corresponding indole precursor dissolved in *N*,*N*-dimethylformamide (DMF)/MeCN (10:1) were added. After stirring at 110 °C for 20 min, the reaction was quenched with water to determine RCCs (cf. Scheme 2).



Scheme 2. Copper-mediated ^{18}F-fluorination of indole precursors **3a**–**d**; precursor (25 µmol), tetrakis(pyridine)copper(II) triflate (5.6 µmol), DMF/MeCN; n.c.a. tetraethylammonium [^{18}F]fluoride, 20 min, 110 °C.

The reaction mixture was analyzed by radio thin-layer chromatography (radio-TLC). The RCCs of the indole precursors refer to the whole radioactivity in the solution. Furthermore, RCCs were corrected for adsorption of [^{18}F]fluoride on the ion-exchange cartridge, syringes and the reaction vial and are summarized in Table 1. The highest RCC (17 %) was obtained for **[^{18}F]4c** with ^{18}F-substitution at the C-6 position of the indole ring. The RCC was also fair for **[^{18}F]4b**, with ^{18}F-substitution at the C-5 position (RCC = 11 %). However, the C-5 position in tryptophan is hydroxylated during the serotonin pathway,[25] hence, ^{18}F-substitution in this position seems impractical.

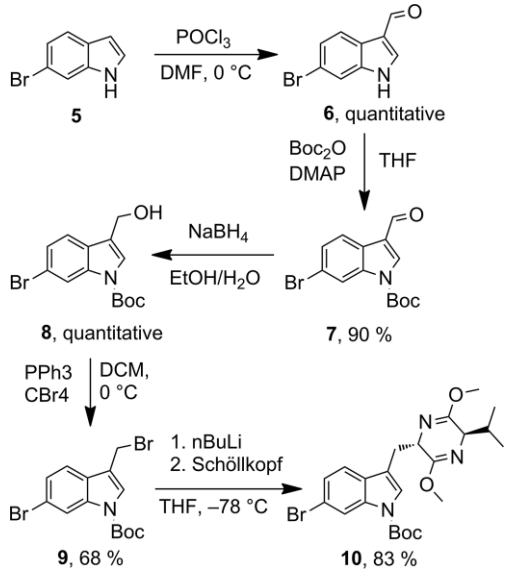
Table 1. Dependence of RCC on the substitution positions of the indole ring. Reaction conditions: [^{18}F]fluoride (ca. 30 MBq), precursor (25 µmol), $Cu(OTf)_2(py)_4$ (5.3 µmol), DMF (300 µL), MeCN (30 µL), 110 °C for 20 min. All reactions were carried out at least in triplicate.

Indication	Indole derivative	RCC [%]
[^{18}F]4a	4-Bpin-1-tosyl indole	7.3 ± 0.7 (n = 3)
[^{18}F]4b	5-Bpin-1-tosyl indole	11.4 ± 0.8 (n = 4)
[^{18}F]4c	6-Bpin-1-tosyl indole	17.0 ± 0.5 (n = 4)
[^{18}F]4d	7-Bpin-1*H*-indole	2.1 ± 1.0 (n = 3)

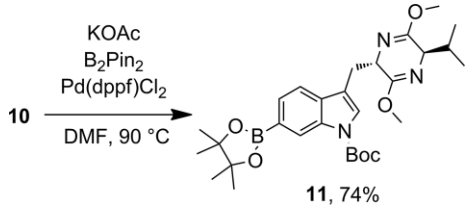
The presence of a pinacolyl ester at the C-4 position on the indole motif delivered only a low RCC of 7 %. This is in contrast to a previous study in which the authors observed the highest ^{18}F-labeling RCCs in the C-4 position of the indole motif using isotope exchange of carbonyl activated compounds.[15] Given that copper-mediated ^{18}F-labeling showed the best results at the C-6 position of the indole motif, only the precursor synthesis of 6-pinalcolester-substituted tryptophan was further investigated.

Synthesis of *tert*-Butyl 3-{[(2*R*,5*S*)-3,6-Dimethoxy-5-(propan-2-yl)-2,5-dihydropyrazin-2-yl]methyl}-6-tetramethyl-1,3,2-dioxaborolan-2-yl-1*H*-indole-1-carboxylate

An appropriate precursor for 6-[^{18}F]fluoro-L-tryptophan was synthesized within a linear six-step synthesis starting from 6-bromoindole according to Konas et al.[26] (cf. Scheme 3). Finally the Suzuki–Miyaura coupling[24] was used as a last step for the introduction of the boronic ester group (cf. Scheme 4).



Scheme 3. Synthesis of **10** according to Konas et al.[26]



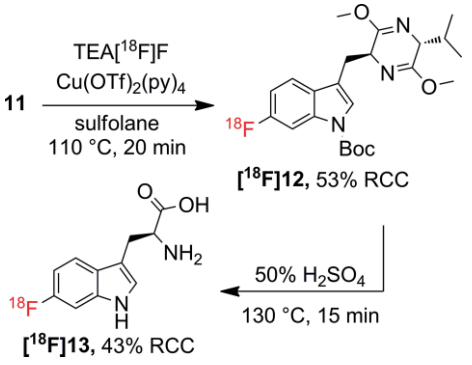
Scheme 4. Synthesis of the precursor *tert*-butyl 3-{[(2*R*,5*S*)-3,6-dimethoxy-5-(propan-2-yl)-2,5-dihydropyrazin-2-yl]methyl}-6-tetramethyl-1,3,2-dioxaborolan-2-yl-1*H*-indole-1-carboxylate (**11**).

In the first step, 6-bromoindole was formylated by a Vilsmeier–Haack reaction as described by Lauchli et al.,[27] providing **3** in quantitative yield. After protection of the amine with *tert*-butoxycarbonyl group (Boc) in 90 % yield of **6**, the aldehyde function was quantitatively reduced with $NaBH_4$ to the corresponding alcohol **7** followed by an Appel-bromination[28] using triphenylphosphine and tetrabromomethane in 68 % yield. The high reactivity of benzyl bromides meant that **9** was immediately coupled with the Schöllkopf chiral amino acid auxiliary,[29] which was previously activated by *n*-butyllithium (*n*BuLi) in 83 % yield.

In the final step, the boronic pinacol ester group was introduced by using a palladium-catalyzed Suzuki–Miyaura coupling[24] yielding the desired precursor **11** in 74 % yield. Precursor **11** was obtained within six steps in an overall yield of 37 %.

Radiosynthesis of 6-[^{18}F]Fluoro-L-tryptophan

The two-step radiosynthesis of the desired radiolabeled tryptophan **[^{18}F]13** is shown in Scheme 5. In the first step, the corresponding precursor **11** was radiolabeled in the presence of a Cu source in sulfolane and acetonitrile, based on similar conditions reported by Tredwell et al.[22] Highest hydrolysis RCCs were achieved by using 50 % H_2SO_4.



Scheme 5. Radiosynthesis of 6-[^{18}F]fluoro-L-tryptophan **[^{18}F]13** from the corresponding precursor **11** under optimized conditions.

Optimization of the Radiosynthesis

Different nucleophilicity enhancers of $^{18}F^-$ for the copper-mediated ^{18}F-fluorination were examined. In the original publication,[22] [^{18}F]fluoride was dried azeotropically in a separate vessel, redissolved in acetonitrile (1 mL), and only small aliquots (30 µL) were used for subsequent ^{18}F-labeling reactions. Since small aliquots are inappropriate for large-scale productions, a one-pot approach using the whole [^{18}F]fluoride target solution is indispensable. During our synthesis development (cf. Table 2), Zlatopolskiy et al. published optimized conditions for copper-mediated ^{18}F-fluorination.[30] In this report, the authors discovered the base sensitivity of the Cu complex and developed a "low-base" protocol in which only small amounts of K_2CO_3 (3 mg vs. 60 µg) and Kryptofix® 2.2.2 were used.

Table 2. RCC dependency of different bases for ^{18}F-labeling of **11** in the presence of [$Cu(OTf)_2(py)_4$]. Reaction conditions: base and [^{18}F]fluoride were azeotropically dried,[a] then DMF (300 µL), MeCN (30 µL), precursor **11** (25 µmol), and $Cu(OTf)_2(py)_4$ were added and heated up to 110 °C for 20 min. The reaction was quenched by addition of water (200 µL) and the radiochemical conversions were determined by radio-TLC. Each experiment was performed at least in triplicate.

	K_{222} + K_2CO_3	Bu_4NHCO_3	Et_4NHCO_3
Amount	0.5 µmol + 0.2 µmol	0.7 µmol	1.0 µmol
RCC [%]	20 ± 4 (n = 3)	7.3 ± 3 (n = 3)	32 ± 8 (n = 5)

[a] Aqueous [^{18}F]fluoride was added to the reaction vial with the respective base and dried azeotropically tree times with MeCN (1 mL) at 80 °C and 500 mbar.

The utilization of tetraethylammonium hydrogen carbonate in an amount of 1 µmol resulted in the highest RCCs for **[^{18}F]12** (32 ± 8 %) from precursor **11** (cf. Table 1). Recently published procedures described a significant shortening of the drying process. These methods consist of the fixation of the aqueous

[^{18}F]fluoride on a cartridge, followed by flushing with anhydrous solvents and elution of [^{18}F]fluoride with an appropriate base dissolved in anhydrous organic solvents. Brichard et al.[31] used *n*-hexane for flushing and $TEAHCO_3$ in acetonitrile for elution. In our study, according to Richarz et al.,[32] the best results were obtained by using anhydrous methanol as flushing reagent and also methanolic $TEAHCO_3$ (0.4–1.0 mg, low boiling point of methanol: 64.7 °C) for eluting the [^{18}F]fluoride from the cartridge. Compared with the conventional drying approach, the time required for the drying step was reduced from 20 to less than 5 minutes.

After drying the reaction vial containing TEA[^{18}F]F, the vial was flushed with air, which seems to improve reaction yields.[22] Then, precursor **11** and $Cu(OTf)_2(py)_4$ dissolved in 30 µL of acetonitrile and 300 µL of various polar aprotic solvents with a high boiling point were added (cf. Table 3). First experiments with less, or without MeCN in the radiolabeling reaction mixture afforded lower RCCs. Therefore, all labeling reactions were carried out in the presence of 10 % MeCN. The mixture was heated to 110 °C for 20 min and the reaction was quenched with water.

Table 3. Effect of solvents on the RCC of **[^{18}F]12**. Reaction conditions: methanolic $TEAHCO_3$ (0.8 mg) for elution of [^{18}F]fluoride from the QMA cartridge, precursor **11** (19 µmol), $Cu(OTf)_2(py)_4$ (5.6 µmol) in solvent (300 µL) and MeCN (30 µL); 110 °C, 20 min, subsequent addition of water (4 mL). The RCC of **[^{18}F]12** was determined based on radio-TLC. Losses of [^{18}F]fluoride on the reaction vial and syringes were taken into account. Each experiment was performed at least in triplicate.

Solvent	RCC [%]
Dimethylformamide	32.3 ± 8.1 (*n* = 6)
Dimethyl sulfoxide	16.5 ± 5.3 (*n* = 3)
Sulfolane	52.7 ± 10.4 (*n* = 5)
Propylene carbonate	38.4 ± 1.7 (*n* = 3)
N-Methyl-2-pyrrolidone	43.1 ± 6.1 (*n* = 3)
tert-Butanol	15.3 ± 5.4 (*n* = 3)

The use of aprotic solvents with a very high dipole moment substantially improved radiochemical yields. Accordingly, the highest labeling yields were obtained in sulfolane followed by *N*-methyl-2-pyrrolidone. Sulfolane has a similar toxicity compared to DMF,[33] and is therefore the solvent of choice for this kind of radiolabeling.

Hydrolysis of [^{18}F]12

In general, the Schöllkopf chiral auxiliary can be easily cleaved with 2 N hydrochloric acid (HCl) followed by 2 N sodium hydroxide (NaOH) within 2 h.[29] However, radiolabeling with short-lived radionuclides requires rapid reactions and harsher reaction conditions with regard to temperature and acidity.[34] A one-step deprotection with HCl at 150 °C for 30 min led to only incomplete hydrolysis of **[^{18}F]13** (20.1 ± 6 % RCC, *n* = 5). In contrast, significant decomposition of the radiofluorinated amino acid was observed by using the more acidic hydroiodic acid (130 °C, 15 min). As a compromise, less acidic hydrobromic acid at 165 °C for 25 min was used to obtain **[^{18}F]13** in 36.1 ± 5 % RCC (*n* = 5). Using shorter times for acidic hydrolysis with HCl followed by saponification in the presence of NaOH did not improve the overall hydrolysis yield (up to 21 % hydrolysis). Use of trifluoroacetic acid, followed by lithium hydroxide, also did not provide the desired compound. Only unidentified byproducts were observed in the reaction mixture. Finally, using 50 % sulfuric acid at 130 °C for 15 min yielded **[^{18}F]13** in the highest RCCs (43.3 ± 6 %; *n* = 4). Table 4 shows all hydrolysis reagents and their corresponding RCCs.

Table 4. Hydrolysis of **[^{18}F]12** using different hydrolysis reagents under the optimized reaction conditions. After applying the respective deprotection conditions, the mixture was diluted with water (1 mL) and analyzed by radioHPLC. Each experiment was carried out at least in triplicate.

Solvent	RCC [%]
HCl	20.0 ± 6.1 (*n* = 5)
HBr	36.1 ± 5.0 (*n* = 6)
HI	– (*n* = 3)
HCl/NaOH	17.2 ± 4.5 (*n* = 3)
TFA/LiOH	– (*n* = 3)
50 % H_2SO_4	43.3 ± 5.7 (*n* = 4)

The final purification was performed by using semipreparative high-performance liquid chromatography (HPLC) with 10 % ethanol in water as mobile phase. The isolated product could be used directly for further biological evaluations. The total radiosynthesis time of 6-[^{18}F]fluoro-L-tryptophan **[^{18}F]13** amounted to 110 min including HPLC purification with a total radiochemical yield of 15.8 ± 4 % (*n* = 4). 6-[^{18}F]Fluoro-L-tryptophan was obtained with a radiochemical purity of more than 99 % and an enantiomeric excess of 89 % (94.4 % L-enantiomer) and a specific activity of 280 GBq µmol^{-1}.

The relatively high amount of the D-enantiomer is inherent in the Schöllkopf auxiliary system.[29] Moreover, taking into account the fair hydrolysis yield, the synthesis of a precursor that can be deprotected under milder conditions is underway. However, the presented radiosynthesis provides a practical route to otherwise difficult-to-access ^{18}F-labeled tryptophan.

Conclusions

First, the influence of the substitution position for ^{18}F-fluorination in the indole structure was studied. Nucleophilic aromatic substitution was highest at the 6-position with 17 ± 1 % RCC (*n* = 4). The appropriately elaborated precursor synthesis for 6-[^{18}F]fluoro-L-tryptophan **[^{18}F]10** consisted of a linear six-step synthesis with an overall yield of 37 %. The copper-mediated ^{18}F-labeling step (53 ± 10 % RCC, *n* = 5) and the following hydrolysis (43 ± 6 % RCC, *n* = 6) were optimized. 6-[^{18}F]Fluoro-L-tryptophan was obtained in an overall RCY of 16 ± 4 % (*n* = 4). The enantiomeric excess amounted to 89 % and the specific activity was 280 GBq µmol^{-1} within a total synthesis time of 110 min.

The reported labeling results of n.c.a. 6-[^{18}F]fluoro-L-tryptophan open up new opportunities for PET diagnostics. Furthermore, the simplified procedure is amenable to remote-controlled synthesis.[35] Moreover, it should enable new insights into the serotonergic and kynurenine pathways. Further biological evaluations of 6-[^{18}F]fluorotryptophan will demonstrate the scope and limitations of the presented probe.

Experimental Section

General: Chemicals were purchased from Chempur (Karlsruhe, Germany), Merck (Darmstadt, Germany) or Sigma–Aldrich (Taufkirchen, Germany).

Thin-layer chromatography (TLC) was performed on precoated silica gel 60 F254 M (Merck, Darmstadt, Germany) and detection was carried out at 254 nm. Detection of radioactive products on the TLC was performed with a Raytest minigita device (Raytest, Straubenhardt, Germany).

Separations via high-performance liquid chromatography (HPLC) were performed with a Knauer pump, a Knauer K-2500 UV/Vis detector (Knauer, Berlin, Germany), and a Rheodyne manual injector (20 µL or 5 mL loop), and for radioactivity detection a NaI(Tl) well-type scintillation detector model 276 Photomultiplier Base with a ACE mate Amplifier and BIAS supply (EG&G Ortec Ametek, Meerbusch, Germany). The measured data were analyzed by using Gina software (Version 2.18, Raytest).

^{1}H and ^{13}C NMR spectra were recorded with either a Varian Inova 400 MHz spectrometer or a Bruker Avance III HD 600 MHz spectrometer using $CDCl_3$ or [D$_6$]DMSO as solvent. In the spectra, chemical shifts are given in δ ppm. HRMS spectra were measured with a FTICR "LTQ FT Ultra" (Thermo Fisher Scientific, Germany). Optical rotations were obtained with a Perkin–Elmer Polarimeter Model 341 (Rodgau, Germany) at a wavelength of 589 nm. Elemental analyses were recorded with a Vario EL cube (ZEA-3, Forschungszentrum Jülich).

HPLC Systems

System A: Analytical HPLC of 6-[^{18}F]fluorotryptophan was carried out with a Synergi 4 µm Hydro-RP column 80Å (250 × 4 mm, CS Chromatographie Langerwehe, Germany) using 10 % ethanol in water as mobile phase with a constant flow rate of 1 mL min^{-1} (k = 3.51).

System B: Enantiomeric and radiochemical purity of 6-[^{18}F]fluoro-D/L-tryptophan were determined via HPLC using a Supelco chirobiotic™ T column (250 × 4.6 mm, Sigma–Aldrich, Deidenhofen, Germany) and as mobile phase 40:60 methanol/water with 0.1 % triethylamine-acetic acid pH 4.1 at a constant flow rate of 0.8 mL min^{-1} [k(L-enantiomer) = 3.13; k (D-enantiomer) = 3.6; α = 1.15].

System C: Semipreparative HPLC purification of 6-[^{18}F]fluorotryptophan was carried out with a Synergi 4 µm Hydro-RP 80Å column (250 × 10 mm, Phenomenex, Aschaffenburg, Germany). As a mobile phase 10 % ethanol in water was used at a constant flow rate of 4 mL min^{-1} (k = 2.89) (Table 5).

Table 5. The k-values of labeled compounds analysed by radio-HPLC.

	HPLC System	k
[^{18}F]13	System A	3.51
[^{18}F]**13**	System B	L-enantiomer 3.13 D-enantiomer 3.60
[^{18}F]13	System C	2.89

4-Bromo-1-(4-methylphenylsulfonyl)-1*H*-indole (2a): NaH (60 % in mineral oil, 200 mg, 5.0 mmol) was added to a solution of 4-bromo-1*H*-indole (650 mg, 3.31 mmol) in THF (10 mL) at 0 °C. The resulting solution was stirred at 0 °C for 30 min before 4-toluenesulfonyl chloride (978 mg, 5.0 mmol) was added. The resulting mixture was allowed to reach room temperature (room temp.) and stirred overnight. After quenching the reaction with water, the mixture was repeatedly extracted with Et_2O, and the combined organic layers were dried with Na_2SO_4. After evaporation of the solvent, purification was performed by flash chromatography to give the desired compound (980 mg, 2.68 mmol, 80 %) as a white solid. R_f = 0.25 (PE/EA, 95:5); m.p. 117 °C. ^{1}H NMR (400 MHz, [D$_6$]DMSO): δ = 7.99–7.93 (m, 2 H), 7.90 (d, J = 8.4 Hz, 2 H), 7.49 (d, J = 7.8 Hz, 1 H), 7.40 (d, J = 8.1 Hz, 2 H), 7.29 (t, J = 8.1 Hz, 1 H), 6.78 (dd, J = 3.7, 0.5 Hz, 1 H), 2.32 (s, 3 H) ppm. ^{13}C NMR (101 MHz, [D$_6$]DMSO): δ = 145.87, 134.30, 133.83, 130.57, 130.34 (2 C), 128.07, 126.81 (2 C), 126.21, 126.08, 114.12, 112.55, 108.35, 21.01 ppm. HRMS: m/z calcd. for $C_{15}H_{12}BrNO_2S$ [M]$^+$ 348.9767; found 348.9766. $C_{15}H_{12}BrNO_2S$: calcd. C 51.44, H 3.45, N 4.00; found C 51.53, H 3.52, N 4.02.

5-Bromo-1-(4-methylphenylsulfonyl)-1*H*-indole (2b): Prepared as described for 4-bromo-1-tosyl-1*H*-indole but starting from 5-bromo-1*H*-indole giving the desired compound as colorless solid in 82 % yield. R_f = 0.17 (PE/EA, 95:5); m.p. 134 °C. ^{1}H NMR (400 MHz, [D$_6$]DMSO): δ = 7.93–7.79 (m, 5 H), 7.48 (dd, J = 8.8, 1.9 Hz, 1 H), 7.38 (d, J = 8.3 Hz, 2 H), 6.81 (d, J = 3.7 Hz, 1 H), 2.31 (s, 3 H) ppm. ^{13}C NMR (101 MHz, [D$_6$]DMSO): δ = 145.72, 133.89, 132.92, 132.42, 130.29 (2 C), 128.42, 127.20, 126.70 (2 C), 124.04, 116.09, 114.97, 108.77, 21.00 ppm. HRMS: m/z calcd. for $C_{15}H_{12}BrNO_2S$ [M]$^+$ 348.9767; found 348.9767. $C_{15}H_{12}BrNO_2S$: calcd. C 51.44, H 3.45, N 4.00; found C 51.52, H 3.45, N 4.02.

6-Bromo-1-(4-methylphenylsulfonyl)-1*H*-indole (2c): Prepared as described for 4-bromo-1-tosyl-1*H*-indole but starting from 6-bromo-1*H*-indole giving the desired compound as colorless solid in 78 % yield. R_f = 0.17 (PE/EA, 95:5); m.p. 132 °C. ^{1}H NMR (600 MHz, $CDCl_3$): δ = 8.17 (s, 1 H), 7.76 (d, J = 8.4 Hz, 2 H), 7.53 (d, J = 3.7 Hz, 1 H), 7.38 (d, J = 8.3 Hz, 1 H), 7.33 (dd, J = 8.3, 1.7 Hz, 1 H), 7.24 (d, J = 8.1 Hz, 2 H), 6.61 (dd, J = 3.7, 0.6 Hz, 1 H), 2.35 (s, 3 H) ppm. ^{13}C NMR (151 MHz, $CDCl_3$): δ = 145.43, 135.59, 135.17, 130.17 (2 C), 129.67, 126.94 (2 C), 126.78, 122.59, 118.35, 116.71, 108.89, 21.72 ppm. HRMS: m/z calcd. for $C_{15}H_{13}O_2NBrS$ [M + H]$^+$ 349.9848; found 349.9845. $C_{15}H_{12}BrNO_2S$: calcd. C 51.44, H 3.45, N 4.00; found C 51.44, H 3.44, N 4.18.

1-(4-Methylphenylsulfonyl)-4-(tetramethyl-1,3,2-dioxaborolan-2-yl)-1*H*-indole (3a): The borylation was carried out as described by Ishiyama et al.[24] In a dry round-bottomed flask, 4-bromo-1-(4-methylphenylsulfonyl)-1*H*-indole (732 mg, 2.0 mmol), potassium acetate (567 mg, 6.0 mmol), bis(pinacolato)diboron (1.0 mg, 4.0 mmol) and $PdCl_2$[*R*-bis(diphenylphosphanyl)ferrocene] (120 mg, 0.2 mmol) were dried in fine vacuum and flushed repeatedly with dry argon. Dry DMF (18 mL) was added and the stirring reaction mixture was heated to 100 °C for 3.5 h under argon. After cooling to room temp., the reaction was quenched with EA/Et_2O (1:1), filtered through silica gel, washed with water, and extracted repeatedly with the quenching solvent. The combined organic phases were washed thrice with brine and dried with Na_2SO_4. The solvent was removed in vacuo and the crude product was purified via flash chromatography (PE/EA, 95:5) and reversed-phase flash chromatography (gradient 50 % MeCN in water up to 70 % MeCN in water), to give the desired compound (520 mg, 1.25 mmol, 62 %) as a white solid. R_f = 0.35 (PE/EA, 95:5); 0.20 (RP-TLC; MeCN/H_2O, 70:30); m.p. 208 °C (decomp.). ^{1}H NMR (400 MHz, [D$_6$]DMSO): δ = 8.07 (d, J = 8.3 Hz, 1 H), 7.86–7.83 (m, 3 H), 7.59 (dd, J = 7.2, 0.8 Hz, 1 H), 7.41–7.33 (m, 3 H), 7.08 (d, J = 3.7 Hz, 1 H), 2.32 (s, 3 H), 1.32 (s, 12 H) ppm. ^{13}C NMR (101 MHz, [D$_6$]DMSO): δ = 145.46, 135.13, 134.08, 133.61, 130.63, 130.20 (2 C), 127.55, 126.63 (2 C), 124.07, 121.52 (broad), 116.07, 110.68, 83.63 (2 C), 24.68 (4 C), 20.98 ppm. HRMS: m/z calcd. for $C_{21}H_{25}O_4NBS$ [M + H]$^+$ 398.1592; found 398.1591. $C_{21}H_{25}BNO_4S$: calcd. C 63.49, H 6.09, N 3.53; found C 62.61, H 6.03, N 3.49.

1-(4-Methylphenylsulfonyl)-5-(tetramethyl-1,3,2-dioxaborolan-2-yl)-1*H*-indole (3b): The reaction conditions were similar to those reported for the preparation of 1-(4-methylphenylsulfonyl)-4-(tetramethyl-1,3,2-dioxaborolan-2-yl)-1*H*-indole, but using a reaction time of 16 h. For purification, the reaction was quenched with EA/Et_2O (1:1), filtered through silica, washed repeatedly with brine and the combined organic layers were dried with Na_2SO_4. After evaporation of the solvent, the crude product was dissolved in MeCN (5 mL) and 2 N NaOH (5 mL) were added. The mixture was then stirred for 30 min at room temperature. After quenching with satd. aq NH_4Cl solution, the mixture was extracted repeatedly with EA/Et_2O. The organic layers were combined, washed with brine, dried with Na_2SO_4, and the solvent was evaporated. The crude product was dissolved in a small amount of dichloromethane (CH_2Cl_2)/PE and given onto a flash chromatography column (gradient PE/CH_2Cl_2, 80:20 to 50:50) to give the desired product (360 mg, 0.9 mmol, 81 %) as a fine white foam. R_f = 0.21 (PE/EA, 95:5); 0.21 (PE/CH_2Cl_2, 3:2); m.p. 151 °C. ^{1}H NMR (400 MHz, [D$_6$]DMSO): δ = 7.98–7.94 (m, 2 H), 7.85 (d, J = 8.4 Hz, 2 H), 7.80 (d, J = 3.7 Hz, 1 H), 7.63 (dd, J = 9.3, 0.5 Hz, 1 H), 7.38 (d, J = 8.1 Hz, 2 H), 6.87 (dd, J = 3.7, 0.4 Hz, 1 H), 2.35–2.26 (m, 3 H), 1.29 (s, 12 H) ppm. ^{13}C NMR (101 MHz, [D$_6$]DMSO): δ = 148.17, 138.75, 136.70, 133.02, 132.85 (2 C), 132.83, 131.16, 129.77, 129.27 (2 C), 125.85 (br), 115.30, 112.32, 86.27 (2 C), 27.29 (4 C), 23.62 ppm. HRMS: *m/z* calcd. for $C_{21}H_{24}O_4NBS$ [M$^+$] 397.1513; found 397.1514. $C_{21}H_{24}BNO_4S$ (397.29): calcd. C 63.49, H 6.09, N 3.53; found C 63.64, H 6.19, N 3.53.

1-(4-Methylphenylsulfonyl)-6-(tetramethyl-1,3,2-dioxaborolan-2-yl)-1*H*-indole (3c): The compound was prepared as described for **3a**. Flash chromatography was carried out with PE/EA (95:5) to give the desired compound (81 %) as a white solid. R_f = 0.29 (PE/EA, 9:1); m.p. 146 °C. ^{1}H NMR (400 MHz, [D$_6$]DMSO): δ = 8.25 (d, J = 0.6 Hz, 1 H), 7.88 (d, J = 3.7 Hz, 1 H), 7.77 (d, J = 8.4 Hz, 2 H), 7.58 (ddd, J = 28.2, 7.9, 0.6 Hz, 2 H), 7.38 (d, J = 8.1 Hz, 2 H), 6.88 (dd, J = 3.7, 0.6 Hz, 1 H), 2.30 (s, 3 H), 1.33 (s, 12 H), 1.17 (s, 2 H) ppm. ^{13}C NMR (101 MHz, [D$_6$]DMSO): δ = 145.51, 134.36, 134.02, 133.05, 130.28 (2 C), 129.07, 128.46, 126.32 (2 C), 124.52 (br), 121.16, 118.85, 109.59, 83.77 (2 C), 24.67 (4 C), 20.97 ppm. HRMS: *m/z* calcd. for $C_{21}H_{25}O_4NBS$ [M + H]$^+$ 397.1513; found 397.1514. $C_{21}H_{24}BNO_4S$ (397.29): calcd. C 63.49, H 6.09, N 3.53; found C 63.27, H 6.24, N 3.39.

7-(Tetramethyl-1,3,2-dioxaborolan-2-yl)-1*H*-indole (3d): In a dry round-bottomed flask, 7-bromo-1*H*-indole (480 mg, 2.5 mmol), potassium acetate (710 mg, 7.5 mmol), bis(pinacolato)diboron (950 mg, 3.75 mmol) and $PdCl_2$[*R*-bis(diphenylphosphanyl)ferrocene] (160 mg, 0.25 mmol) were dried by repeatedly application of fine vacuum and flushed with dry argon. Anhydrous DMF (20 mL) was added and the stirring reaction mixture was heated to 100 °C for 4 h under argon. After cooling to room temp., the reaction was quenched by adding Et_2O and water. The mixture was filtered through silica gel, extracted repeatedly with the Et_2O, and the organic phases were combined. After washing the organic layer three times with brine and drying over Na_2SO_4, the solvent was removed in vacuo. The crude product was purified by flash chromatography (PE/EA, 98:2 to 90:10) to give the desired compound (270 mg, 1.11 mmol, 44 %) as a white solid. R_f = 0.57 (PE/EA, 9:1); m.p. 88 °C. ^{1}H NMR (600 MHz, [D$_6$]DMSO): δ = 10.19 (s, 1 H), 7.67 (d, J = 7.8 Hz, 1 H), 7.43 (dd, J = 7.0, 0.9 Hz, 1 H), 7.32–7.31 (m, 1 H), 7.06–6.95 (m, 1 H), 6.43 (dd, J = 3.0, 2.0 Hz, 1 H), 1.33 (s, 12 H) ppm. ^{13}C NMR (151 MHz, [D$_6$]DMSO): δ = 139.90, 128.39, 127.04, 125.87, 123.78, 118.50, 110.24 (br), 100.92, 83.49 (2 C), 24.67 (4 C) ppm. HRMS: *m/z* calcd. for $C_{14}H_{19}BNO_2$ [M + H]$^+$ 244.1503; found 244.1503. $C_{14}H_{18}BNO_2$ (243.11): calcd. C 69.17, H 7.46, N 5.76; found C 68.89, H 7.46, N 5.77.

6-Bromo-1*H*-indole-3-carbaldehyde (6): The formylation and purification was performed according to Lauchli et al.[27] Under an inert atmosphere, 6-bromo-1*H*-indole (2.5 g,12.8 mmol) dissolved in DMF (12 mL) was added dropwise to a stirred solution of $POCl_3$ (2.67 mL, 16.4 mmol) in DMF (17 mL) at 0 °C. The reaction was warmed to room temp. and stirred for ca. 1 h until a yellowish precipitate appeared. The reaction was slowly quenched by adding KOH (6.65 g) in water (20 mL) and kept just below boiling. After cooling to room temp., saturated $NaHCO_3$ was added and the aqueous phase was extracted repeatedly with ethyl acetate and washed four times with brine. The combined organic phases were dried with Na_2SO_4 and removal of solvent in vacuo gave the desired compound (2.84 g, 12.7 mmol, 99 %) as an off-white solid. R_f = 0.08 (PE/EA, 4:1); m.p. 195 °C. ^{1}H NMR (600 MHz, [D$_6$]DMSO): δ = 12.56 (s, 1 H), 10.68 (s, 1 H), 8.30 (s, 1 H), 7.57 (dd, J = 8.1, 0.7 Hz, 1 H), 7.46 (dd, J = 7.6, 0.6 Hz, 1 H), 7.16 (t, J = 7.9 Hz, 1 H) ppm. ^{13}C NMR (151 MHz, [D$_6$]DMSO): δ = 185.04, 138.71, 134.28, 126.44, 125.17, 124.25, 118.30, 112.91, 112.76 ppm. HRMS: *m/z* calcd. for C_9H_6BrNO [M + H]$^+$ 223.9705; found 223.9705, 225.9685 [^{81}BrM]$^+$. C_9H_6BrNO: calcd. C 48.25, H 2.7, N 6.25; found C 48.44, H 2.81, N 6.29.

***tert*-Butyl 6-Bromo-3-formylindole-1*H*-carboxylate (7):** The following reactions were performed as described by Konas et al.[26] Boc_2O (3.03 g,13.9 mmol) and 4-(dimethylamino)pyridine (16.05 mg, 130 µmol) were added to a stirred solution of 6-bromo-1*H*-indole-3-carbaldehyde (2.8 g, 12.6 mmol) in THF (35 mL). After 30 min, the reaction showed complete conversion on TLC and was stopped by adding H_2O. The aqueous phase was extracted repeatedly with ethyl acetate. The organic phases were combined, washed with brine and dried with Na_2SO_4. After removal of the organic solvent in vacuo, the compound was purified by flash chromatography (PE/EA, 8:1) to give the desired product (3.69 g, 11.3 mmol, 90 %) as a colorless solid. R_f = 0.33 (PE/EA, 9:1); m.p. 142 °C. ^{1}H NMR (600 MHz, [D$_6$]DMSO): δ = 10.07 (s, 1 H), 8.68 (s, 1 H), 8.27 (d, J = 1.7 Hz, 1 H), 8.08 (d, J = 8.4 Hz, 1 H), 7.57 (dd, J = 8.4, 1.8 Hz, 1 H), 1.67 (s, 9 H) ppm. ^{13}C NMR (151 MHz, [D$_6$]DMSO): δ = 185.52, 147.80, 136.61, 133.26, 128.37, 126.50, 125.92, 120.26, 114.59, 112.83, 86.25, 27.45 (3 C) ppm. HRMS: *m/z* calcd. for $C_{14}H_{14}BrNO_3$ [^{81}BrM + H]$^+$ 326.0293; found 326.02095. $C_{14}H_{14}BrNO_3$ (324.17): calcd. C 51.87, H 4.35, N 4.32; found C 52.11, H 4.37, N 4.40.

***tert*-Butyl 6-Bromo-3-(hydroxymethyl)-1*H*-indole-1-carboxylate (8):** The reduction was performed by adding $NaBH_4$ (626 mg, 16.6 mmol) to a stirred solution of **7** (3.6 g, 11.1 mmol) in THF/EtOH (2:1) at 0 °C. The mixture was warmed to room temp. and stirred for 30 min. NH_4Cl was added and the mixture was extracted repeatedly with Et_2O. After drying the combined organic phases over Na_2SO_4 and removal of the solvent in vacuo the desired compound (3.6 g, 11.1 mmol, quantitative) was obtained as a white solid. R_f = 0.11 (PE/EA, 9:1); m.p. 90 °C. ^{1}H NMR (600 MHz, [D$_6$]DMSO): δ = 8.20 (s, 1 H), 7.60 (d, J = 8.4 Hz, 1 H), 7.54 (s, 1 H), 7.39 (dd, J = 8.4, 1.8 Hz, 1 H), 5.12 (t, J = 5.5 Hz, 1 H), 4.61 (dd, J = 5.5, 0.9 Hz, 2 H), 1.61 (s, 9 H) ppm. ^{13}C NMR (151 MHz, DMSO): δ = 148.75, 135.76, 128.25, 125.33, 123.56, 121.87, 121.59, 117.37, 117.12, 84.15, 54.92, 27.58 (3 C) ppm. HRMS: *m/z* calcd. for $C_{14}H_{16}BrNO_3$ [^{81}BrM + Na]$^+$ 350.0187; found 350.0183. $C_{14}H_{16}BrNO_3$: calcd. C 51.55, H 4.94, N 4.29; found C 51.87, H 4.92, N 4.32.

***tert*-Butyl 6-Bromo-3-(bromomethyl)-1*H*-indole-1-carboxylate (9):** For the Appel reaction,[28] at 0 °C, PPh_3 (3.58 g, 15.0 mmol) and CBr_4 (5.09 g, 15.0 mmol) were added subsequently to a stirred solution of *tert*-butyl 6-bromo-3-(hydroxymethyl)-1*H*-indole-1-carboxylate (4.4 g, 13.6 mmol) in CH_2Cl_2 (40 mL). By monitoring the reaction with TLC, no starting material was found after 30 min. After removal of solvent at 0 °C partially in vacuo, the oily residue was applied on

a silica column and eluted with a mixture of PE/EE (6:1). Removal of solvents gave the desired product (68 %, 3.65 g, 9.4 mmol) as an off-white solid. Because of the fast decomposition, the next step was performed immediately after drying. R_f = 0.68 (PE/EE, 4:1); m.p. 83 °C (decomp.). ^{1}H NMR (600 MHz, [D$_6$]DMSO): δ = 8.20 (s, 1 H), 7.88 (s, 1 H), 7.64 (d, *J* = 8.4 Hz, 1 H), 7.47 (dd, *J* = 8.4, 1.3 Hz, 1 H), 4.90 (s, 2 H), 1.60 (s, 9 H) ppm. ^{13}C NMR (151 MHz, [D$_6$]DMSO): δ = 148.37, 135.67, 127.55, 126.34, 125.70, 121.42, 117.70, 117.57, 117.34, 84.75, 27.53, 25.40 ppm.

***tert*-Butyl 6-Bromo-3-{[(2*R*,5*S*)-3,6-dimethoxy-5-(propan-2-yl)-2,5-dihydropyrazin-2-yl]methyl}-1*H*-indole-1-carboxylate (10):** *n*BuLi (3.4 mL, 8.5 mmol in *n*-hexane) was added slowly to a stirred solution of (3*S*)3-*tert*-butyl-2,5-dimethoxy-3,6-dihydropyrazine (1.54 g, 8.5 mmol) in THF (10 mL) at –78 °C. After stirring for further 30 min at –78 °C, *tert*-butyl 6-bromo-3-(bromomethyl)-1*H*-indole-1-carboxylate (3.0 g, 7.71 mmol) in THF (12 mL) was added slowly and the resulting solution was stirred for 1 h at –78 °C. After quenching with saturated NH_4Cl, the aqueous phase was extracted repeatedly with Et_2O. The combined organic phases were dried with Na_2SO_4 and the solvent was removed in vacuo. Purification was carried out by flash chromatography to give the desired compound (3.16 g, 6.4 mmol, 83 %) as a colorless oil. R_f = 0.25 (PE/EA, 6:1). ^{1}H NMR (600 MHz, [D$_6$]DMSO): δ = 8.16 (s, 1 H), 7.53 (d, *J* = 8.4 Hz, 1 H), 7.39 (dd, *J* = 8.4, 1.8 Hz, 1 H), 7.34 (s, 1 H), 4.35 (dd, *J* = 8.7, 4.9 Hz, 1 H), 3.60 (s, 3 H), 3.58 (s, 3 H), 3.52 (t, *J* = 3.5 Hz, 1 H), 3.10 (d, *J* = 4.9 Hz, 2 H), 2.08 (dtd, *J* = 13.6, 6.8, 3.4 Hz, 1 H), 1.60 (s, 9 H), 0.89 (d, *J* = 6.9 Hz, 3 H), 0.57 (d, *J* = 6.8 Hz, 3 H) ppm. ^{13}C NMR (151 MHz, [D$_6$]DMSO): δ = 163.61, 162.72, 149.04, 130.45, 125.59, 125.05, 121.94, 117.62, 117.33, 116.68, 84.60, 60.22, 55.65, 52.57, 52.40, 49.06, 31.42, 28.86, 28.02 (3 C), 19.39, 16.81 (2 C) ppm. HRMS: *m/z* calcd. for $C_{23}H_{30}BrN_3O_4$ [^{81}BrM]$^+$ 494.14720; found 494.14676. $C_{23}H_{30}BrN_3O_4$ (492.41): calcd. C 56.10, H 6.14, N 8.53; found C 51.92, H 6.57, N 7.89.

***tert*-Butyl 3-{[(2*R*,5*S*)-3,6-Dimethoxy-5-(propan-2-yl)-2,5-dihydropyrazin-2-yl]methyl}-6-tetramethyl-1,3,2-dioxaborolan-2-yl-1*H*-indole-1-carboxylate (11):** The borylation was performed according to Ishiyama et al.[24] In a dry round-bottomed flask, 6-bromo-3-{[(2*S*,5*R*)-3,6-dimethoxy-5-(propan-2-yl)-2,5-dihydropyrazin-2-yl]methyl}-1-methyl-1*H*-indole (2.0 g, 4.06 mmol), potassium acetate (1.15 g, 12.2 mmol), bis(pinacolato)diboron (2.04 g, 8.12 mmol) and $PdCl_2$[*R*-bis(diphenylphosphanyl)ferrocene] (253 mg, 0.41 mmol) were dried by repeated application of fine vacuum and flushing with dry argon. Anhydrous DMF (30 mL) was added and the stirring reaction mixture was heated to 100 °C for 3 h under argon. After cooling to room temp., the reaction mixture was quenched with water, filtered through silica gel, extracted repeatedly with Et_2O and the combined organic phases were washed three times with brine. After drying over Na_2SO_4, the solvent was removed in vacuo and the mixture was purified by flash chromatography (PE 100 % to PE/EA, 9:1) to give the desired compound (1.83 g, 3.4 mmol, 83 %). R_f = 0.35 (PE/EA, 95:5); m.p. 62 °C. $[\alpha]_D^{20}$ = 0.26(*c* = 10 mg mL^{-1} $CHCl_3$). ^{1}H NMR (600 MHz, [D$_6$]DMSO): δ = 8.45 (s, 1 H), 7.58 (d, *J* = 7.9 Hz, 1 H), 7.52 (d, *J* = 7.8 Hz, 1 H), 7.39 (s, 1 H), 4.36 (dd, *J* = 8.6, 4.7 Hz, 1 H), 3.61 (s, 3 H), 3.60 (s, 3 H), 3.47 (t, *J* = 3.5 Hz, 1 H), 3.12 (d, *J* = 4.9 Hz, 2 H), 2.08 (dhept, *J* = 6.8, 3.4 Hz, 1 H), 1.61 (s, 9 H), 1.31 (s, 12 H), 1.17 (s, 1 H), 0.89 (d, *J* = 6.9 Hz, 3 H), 0.57 (d, *J* = 6.8 Hz, 3 H) ppm. ^{13}C NMR (151 MHz, [D$_6$]DMSO): δ = 163.04, 162.24, 148.70, 134.18, 133.30, 127.82, 125.36, 123.75 (broad), 120.80, 119.05, 116.16, 83.53 (2 C), 83.49, 59.68, 55.18, 52.10, 51.88, 30.87, 28.49, 27.58 (4 C), 24.71 (3 C), 18.92, 16.30 ppm. HRMS: *m/z* calcd. for $C_{29}H_{42}BN_3O_6$ [M + H]$^+$ 540.3239; found 540.32441. $C_{29}H_{42}BN_3O_6$ (539.32): calcd. C 64.57, H 7.85, N 7.79; found C 64.68; H 8.06; N 7.52.

Radiochemistry

Preparation of Tetraethylammonium [^{18}F]Fluoride: No-carrier-added [^{18}F]fluoride was obtained through an ^{18}O(p,n)^{18}F nuclear reaction by the bombardment of isotopically enriched [^{18}O]water as target with 17 MeV protons at the JSW cyclotron BC 1710 (INM-5, Forschungszentrum Jülich).[1c] An aliquot of aqueous [^{18}F]fluoride was diluted with methanol (500 µL) and loaded onto a 46 mg Sep-Pak® QMA light cartridge (Waters, Eschborn, Germany). Then, anhydrous methanol (1 mL), followed by air (20 mL) were flushed through the cartridge. Elution of the [^{18}F]fluoride was conducted by washing tetraethylammonium hydrogen carbonate (0.8 mg) in anhydrous methanol (800 µL) slowly through the cartridge into the reaction vial. Afterwards, the cartridge was washed with anhydrous methanol (500 µL). The solvent was evaporated at 80 °C and 500 mbar followed by an evacuation for 3 min at <10 mbar.

General Procedure for Radiosynthesis of [^{18}F]Fluoroindoles: The radiosyntheses were carried out as described by Tredwell et al.[22] **4a**–**c** (10 mg) or **4d** (15 mg), the corresponding pinacol boronate indole derivative (25 µmol) and $Cu(OTf)_2(py)_4$ (3.6 mg, 5.6 µmol) were dissolved in DMF/MeCN (10:1, 330 µL) and added on the dried residue of TEA[^{18}F]F. The reaction mixture was heated to 110 °C for 20 min and quenched with water (3 mL). The RCCs were determined by radioTLC (Table 6).

Table 6. RadioTLC was carried out using a 4:1 hexane/ethyl acetate mix. The R_f values were compared with fluorinated standards.

Indication	Indole derivative	R_f
[^{18}F]4a	4-F-1-tosyl-1*H*-indole	0.80
[^{18}F]**4b**	5-F-1-tosyl-1*H*-indole	0.54
[^{18}F]4c	6-F-1-tosyl-1*H*-indole	0.63
[^{18}F]4c	7-F-1*H*-indole	0.76

General Procedure for Radiosynthesis of 6-[^{18}F]Fluoro-L-tryptophan: A solution of the precursor (10 mg, 19 µmol) and $Cu(OTf)_2(py)_4$ (3.8 mg, 5.6 µmol) in sulfolane (300 µL) with acetonitrile (30 µL) was added to the dried residue of TEA[^{18}F]F. The reaction mixture was heated to 115 °C for 20 min. Then, after a short cooling time, the mixture was diluted with CH_2Cl_2 (2 mL) and passed through a 2 g silica cartridge into a dry reaction vial. The cartridge was washed with CH_2Cl_2 (3 mL). After removal of solvent at 500 mbar and 80 °C, 1 mL of 50 % sulfuric acid was added and the reaction mixture was heated to 130 °C for 15 min. The resulting mixture was diluted with water (1 mL), filtered through a 0.2 µm Millex® PTFE membrane filter (Merck, Darmstadt, Germany) and purified by semipreparative HPLC (System C).

Acknowledgments

The authors wish to thank Dirk Bier and Marcus Holschbach (all Forschungszentrum Jülich GmbH, INM-5), Patrick Bongen (Institut für Bio- und Geowissenschaften, Biotechnologie, IBG-1) and Sabine Willbold (Zentralinstitut für Engineering, Elektronik und Analytik, ZEA-3), all Forschungszentrum Jülich, for recording the spectroscopic data.

Keywords: Radiopharmaceuticals · Radiochemistry · Isotopic labeling · Positron emission tomography · Imaging agents · Amino acids

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Received: June 9, 2016
Published Online: ■

Radiopharmaceuticals

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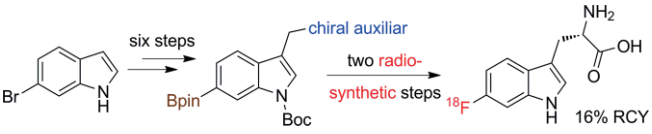
Preparation of No-Carrier-Added 6-[^{18}F]Fluoro-L-tryptophan via Cu-Mediated Radiofluorination



Tryptophan takes part in a range of physiological processes and is discussed as a potential tumor marker in positron emission tomography. An appropriate precursor for the introduction of [^{18}F]fluoride into the aromatic -system of tryptophan was developed. By using a new copper-mediated radiofluorination, the radiosynthesis was optimized with regard to amenability to automation.

DOI: 10.1002/ejoc.201600705