

Mono- or di-substituted indole derivatives as dengue viral replication inhibitors

The present invention relates to mono- or di-substituted indole derivatives, methods to prevent or treat dengue viral infections by using these compounds and also relates to these compounds for use as a medicine, more preferably for use as a medicine to treat or prevent dengue viral infections. The present invention furthermore relates to pharmaceutical compositions or combination preparations of the compounds, to the compositions or preparations for use as a medicine, more preferably for the prevention or treatment of dengue viral infections. The invention also relates to processes for preparation of these compounds.

BACKGROUND OF THE INVENTION

Flaviviruses, which are transmitted by mosquitoes or ticks, cause life-threatening infections in man, such as encephalitis and hemorrhagic fever. Four distinct, but closely related serotypes of the flavivirus dengue are known, so-called DENV1, -2, -3, and -4. Dengue is endemic in most tropical and sub-tropical regions around the world, predominantly in urban and semi-urban areas. According to the World Health Organization (WHO), 2.5 billion people of which 1 billion children are at risk of DENV infection (WHO, 2002). An estimated 50 to 100 million cases of dengue fever [DF], half a million cases of severe dengue disease (i.e. dengue hemorrhagic fever [DHF] and dengue shock syndrome [DSS]), and more than 20,000 deaths occur worldwide each year. DHF has become a leading cause of hospitalization and death amongst children in endemic regions. Altogether, dengue represents the most common cause of arboviral disease. Because of recent large outbreaks in countries situated in Latin America, South-East Asia and the Western Pacific (including Brazil, Puerto Rico, Venezuela, Cambodia, Indonesia, Vietnam, Thailand), numbers of dengue cases have risen dramatically over the past years. Not only is the number of dengue cases increasing as the disease is spreading to new areas, but the outbreaks tend to be more severe.

To prevent and/or control the disease associated with dengue viral infection, the only available methods at present are mosquito eradication strategies to control the vector. Although progress is being made in the development of vaccines against dengue, many difficulties are encountered. These include the

existence of a phenomenon referred to as antibody-dependent enhancement (ADE).

Recovery from an infection by one serotype provides lifelong immunity against that serotype but confers only partial and transient protection against a subsequent infection by one of the other three serotypes. Following infection with another serotype, pre-existing heterologous antibodies form complexes with the newly infecting dengue virus serotype but do not neutralize the pathogen. Instead, virus entry into cells is believed to be facilitated, resulting in uncontrolled virus replication and higher peak viral titers. In both primary and secondary infections, higher viral titers are associated with more severe dengue disease. Since maternal antibodies can easily pass on to infants by breast feeding, this might be one of the reasons that children are more affected by severe dengue disease than adults.

In locations with two or more serotypes circulating simultaneously, also referred to as hyper endemic regions, the risk of serious dengue disease is significantly higher due to an increased risk of experiencing a secondary, more severe infection. Moreover, in a situation of hyper-endemicity, the probability of the emergence of more virulent strains is increased, which in turn augments the probability of dengue hemorrhagic fever (DHF) or dengue shock syndrome.

The mosquitoes that carry dengue, including *Aedes aegypti* and *Aedes albopictus* (tiger mosquito), are moving north on the globe. According to the United States (US) Centers for Disease Control and Prevention (CDC), both mosquitoes are currently omnipresent in southern Texas. The spread north of dengue-carrying mosquitoes is not confined to the US, but has also been observed in Europe.

Recently (December 2015), the dengue vaccine produced by Sanofi Pasteur was first approved in Mexico. The vaccine has also been approved in Brazil, The Philippines and El Salvador. Regulatory review processes are continuing in other countries where dengue is a public health priority. Nevertheless, the vaccine leaves considerable room for improvement due to limited efficacy, especially against DENV-1 and -2, low efficacy in flavivirus-naïve subjects and the lengthy dosing schedule.

Despite these shortcomings, the vaccine is a game changer in endemic settings as it will offer protection to a large part of the population, but likely not

to very young infants, who bear the largest burden of dengue. In addition, the dosing schedule and very limited efficacy in flavivirus-naïve subjects make it unsuitable and likely not worthwhile/cost-effective for travelers from non-endemic areas to dengue-endemic areas. The above mentioned shortcomings
5 of the dengue vaccines are the reason why there is a need for a pre-exposure prophylactic dengue antiviral.

Furthermore, today, specific antiviral drugs for the treatment or prevention of dengue fever virus infection are not available. Clearly, there is still a great
10 unmet medical need for therapeutics for the prevention or treatment of viral infections in animals, more in particular in humans and especially for viral infections caused by Flaviviruses, more in particular Dengue virus. Compounds with good anti-viral potency, no or low levels of side-effects, a broad spectrum activity against multiple Dengue virus serotypes, a low toxicity and/or good
15 pharmacokinetic or -dynamic properties are highly needed.

The present invention now provides compounds, mono- or di-substituted indole derivatives, which show high potent activity against all four (4) serotypes of the Dengue virus. Also the compounds according to the invention possess a good
20 pharmacokinetic profile and surprisingly these specific compounds show an improved chiral stability.

SUMMARY

The present disclosure is based on the unexpected finding that at least one of
25 the above-mentioned problems can be solved by the current compounds of the disclosure.

The present disclosure provides compounds which have been shown to possess potent antiviral activity against all four (4) serotypes currently known. The present disclosure furthermore demonstrates that these compounds
30 efficiently inhibit proliferation of Dengue virus (DENV). Therefore, these compounds constitute a useful class of potent compounds that can be used in the treatment and/or prevention of viral infections in animals, mammals and humans, more specifically for the treatment and/or prevention of infections with Dengue viruses.

35 The present disclosure furthermore relates to the use of such compounds as medicines and to their use for the manufacture of medicaments for treating and/or preventing viral infections, in particular with viruses belonging to the

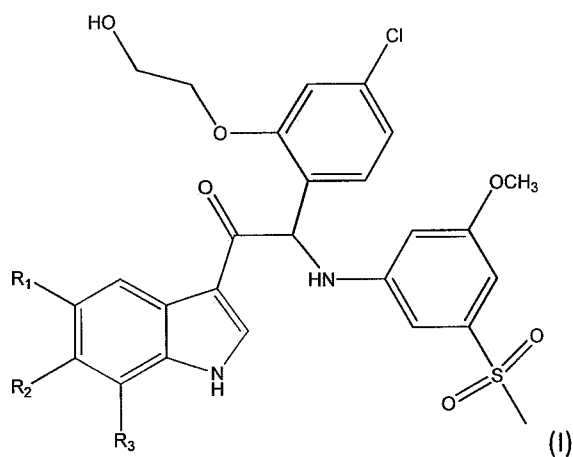
family of the Dengue viruses in animals or mammals, more in particular in humans. The disclosure also relates to methods for the preparation of all such compounds and to pharmaceutical compositions comprising them in an effective amount.

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The present disclosure also relates to a method of treatment or prevention of dengue viral infections in humans by the administration an effective amount of one or more such compounds, or a pharmaceutically acceptable salt thereof optionally in combination with one or more other medicines, like another antiviral agent, to a patient in need thereof.

10

One aspect of the disclosure is the provision of compounds of formula (I)



a stereo-isomeric form, a pharmaceutically acceptable salt, solvate or polymorph thereof comprising a mono- or di-substituted indole group; said

15 compound is selected from the group wherein:

R_1 is H, R_2 is F, Cl or OCH_3 and R_3 is H;

R_1 is H, R_2 is F or Cl and R_3 is CH_3 ;

R_1 is CH_3 , R_2 is OCH_3 and R_3 is H;

R_1 is F, R_2 is F and R_3 is H;

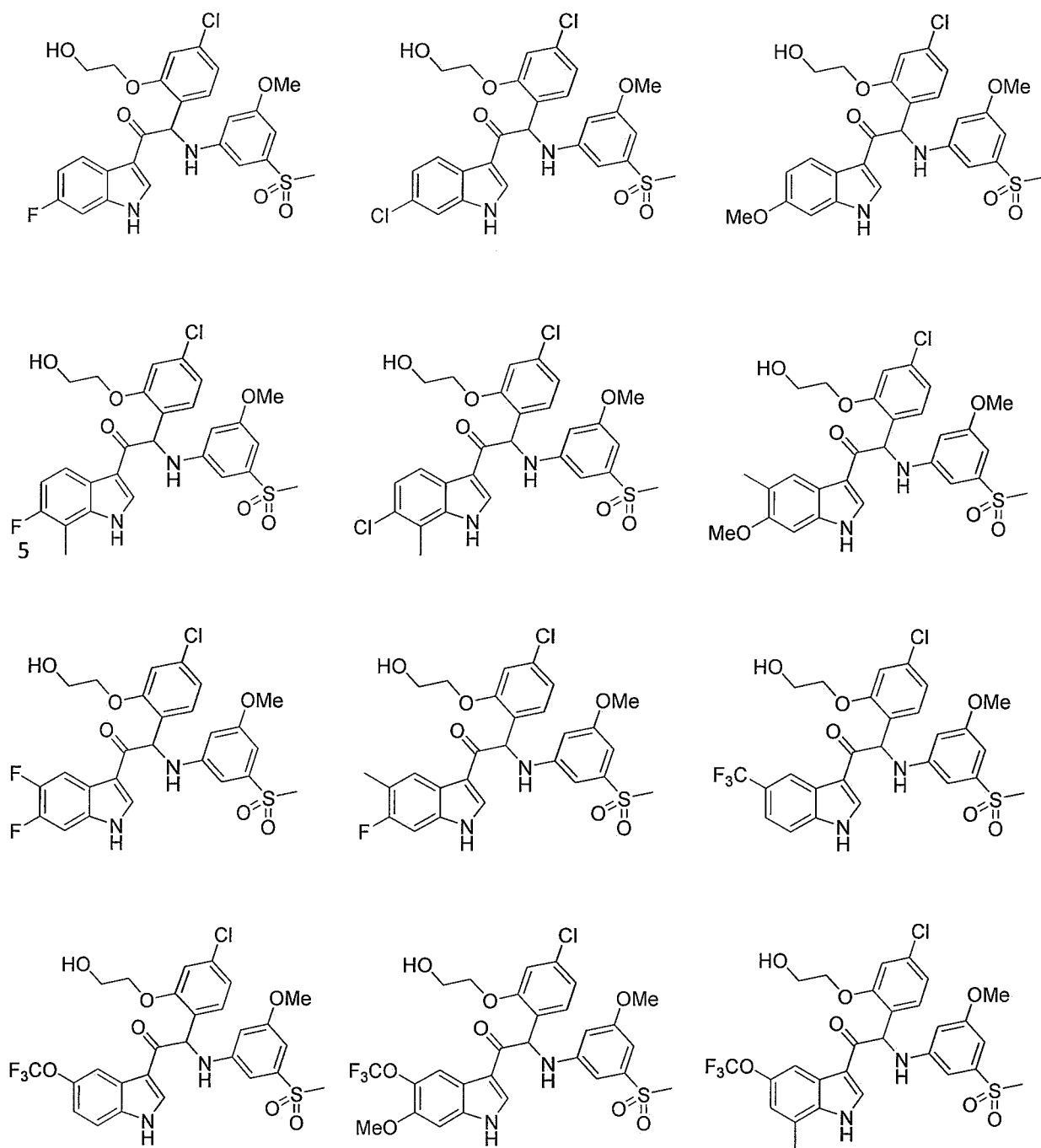
20 R_1 is CH_3 , R_2 is F and R_3 is H;

R_1 is CF_3 or OCF_3 and R_2 is H and R_3 is H;

R_1 is OCF_3 , R_2 is OCH_3 and R_3 is H or

R_1 is OCF_3 , R_2 is H and R_3 is CH_3 .

25 In particular the compounds of the disclosure or their stereo-isomeric form, a pharmaceutically acceptable salt, solvate or polymorph thereof are selected from the group:



10

Another aspect of the current disclosure is a pharmaceutical composition comprising a compound of formula (I) or a stereo- isomeric form , a pharmaceutically acceptable salt, solvate or polymorph as defined herein thereof together with one or more pharmaceutically acceptable excipients, diluents or carriers.

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Pharmaceutically acceptable salts of the compounds of formula (I) include the acid addition and base salts thereof. Suitable acid addition salts are formed from acids which form non-toxic salts. Suitable base salts are formed from bases which form non-toxic salts.

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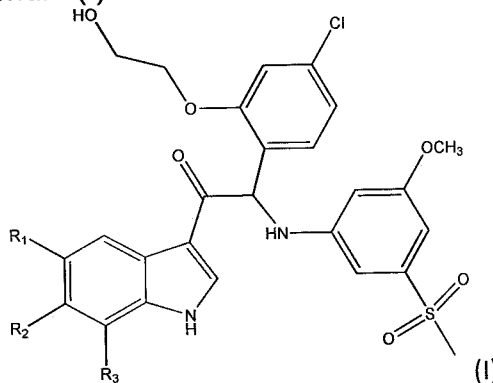
Another aspect of the current disclosure is a compound of formula (I) or a stereo- isomeric form, a pharmaceutically acceptable salt, solvate or polymorph thereof as defined herein or a pharmaceutical composition as defined herein for use as a medicament.

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Another aspect of the current disclosure is a compound of formula (I) or a stereo- isomeric form, a pharmaceutically acceptable salt, solvate or polymorph thereof as defined herein or a pharmaceutical composition as defined herein for use in the treatment of dengue.

15

Another aspect of the current disclosure is use of a compound represented by the following structural formula (I)



20

a stereo-isomeric form, a pharmaceutically acceptable salt, solvate or polymorph thereof comprising a mono- or di-substituted indole group; said compound is selected from the group wherein:

R₁ is H, R₂ is F, Cl or OCH₃ and R₃ is H;

R₁ is H, R₂ is F or Cl and R₃ is CH₃;

R₁ is CH₃, R₂ is OCH₃ and R₃ is H;

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R₁ is F, R₂ is F and R₃ is H;

R₁ is CH₃, R₂ is F and R₃ is H;

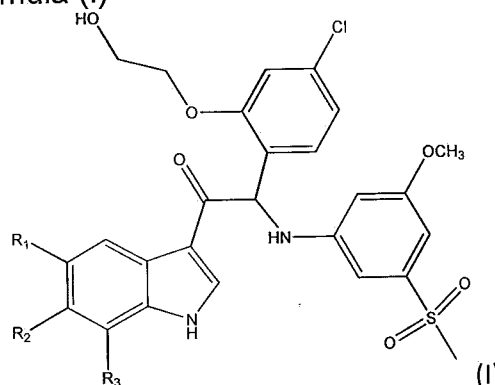
R₁ is CF₃ or OCF₃ and R₂ is H and R₃ is H;

R₁ is OCF₃, R₂ is OCH₃ and R₃ is H or

R₁ is OCF₃, R₂ is H and R₃ is CH₃

in the manufacture of a medicament for inhibiting the replication of dengue virus(es) in a patient.

Another aspect of the current disclosure is use of a compound represented by the following structural formula (I)



a stereo-isomeric form, a pharmaceutically acceptable salt, solvate or polymorph thereof comprising a mono- or di-substituted indole group; said compound is selected from the group wherein:

- 10 R_1 is H, R_2 is F, Cl or OCH_3 and R_3 is H;
 R_1 is H, R_2 is F or Cl and R_3 is CH_3 ;
 R_1 is CH_3 , R_2 is OCH_3 and R_3 is H;
 R_1 is F, R_2 is F and R_3 is H;
 R_1 is CH_3 , R_2 is F and R_3 is H;
15 R_1 is CF_3 or OCF_3 and R_2 is H and R_3 is H;
 R_1 is OCF_3 , R_2 is OCH_3 and R_3 is H or
 R_1 is OCF_3 , R_2 is H and R_3 is CH_3

for inhibiting the replication of dengue virus(es) in a biological sample.

20 The compounds of the invention may also exist in un-solvated and solvated forms. The term "solvate" is used herein to describe a molecular complex comprising the compound of the invention and one or more pharmaceutically acceptable solvent molecules, for example, ethanol.

 The term "polymorph" refers to the ability of the compound of the invention to
25 exist in more than one form or crystal structure.

The compounds of the present invention may be administered as crystalline or amorphous products. They may be obtained for example as solid plugs, powders, or films by methods such as precipitation, crystallization, freeze drying, spray drying, or evaporative drying. They may be administered alone or
5 in combination with one or more other compounds of the invention or in combination with one or more other drugs. Generally, they will be administered as a formulation in association with one or more pharmaceutically acceptable excipients. The term "excipient" is used herein to describe any ingredient other than the compound(s) of the invention. The choice of excipient depends largely
10 on factors such as the particular mode of administration, the effect of the excipient on solubility and stability, and the nature of the dosage form.

The compounds of the present invention or any subgroup thereof may be formulated into various pharmaceutical forms for administration purposes. As
15 appropriate compositions there may be cited all compositions usually employed for systemically administering drugs. To prepare the pharmaceutical compositions of this invention, an effective amount of the particular compound, optionally in addition salt form, as the active ingredient is combined in intimate admixture with a pharmaceutically acceptable carrier, which carrier may take a
20 wide variety of forms depending on the form of preparation desired for administration. These pharmaceutical compositions are desirably in unitary dosage form suitable, for example, for oral or rectal administration. For example, in preparing the compositions in oral dosage form, any of the usual pharmaceutical media may be employed such as, for example, water, glycols,
25 oils, alcohols and the like in the case of oral liquid preparations such as suspensions, syrups, elixirs, emulsions, and solutions; or solid carriers such as starches, sugars, kaolin, diluents, lubricants, binders, disintegrating agents and

the like in the case of powders, pills, capsules, and tablets. Because of their ease in administration, tablets and capsules represent the most advantageous oral dosage unit forms, in which case solid pharmaceutical carriers are obviously employed. Also included are solid form preparations that can be converted, shortly before use, to liquid forms.

It is especially advantageous to formulate the aforementioned pharmaceutical compositions in unit dosage form for ease of administration and uniformity of dosage. Unit dosage form as used herein refers to physically discrete units suitable as unitary dosages, each unit containing a predetermined quantity of active ingredient calculated to produce the desired therapeutic effect in association with the required pharmaceutical carrier. Examples of such unit dosage forms are tablets (including scored or coated tablets), capsules, pills, powder packets, wafers, suppositories, injectable solutions or suspensions and the like, and segregated multiples thereof.

Those of skill in the treatment of infectious diseases will be able to determine the effective amount from the test results presented hereinafter. In general it is contemplated that an effective daily amount would be from 0.01 mg/kg to 50 mg/kg body weight, more preferably from 0.1 mg/kg to 10 mg/kg body weight. It may be appropriate to administer the required dose as two, three, four or more sub-doses at appropriate intervals throughout the day. Said sub-doses may be formulated as unit dosage forms, for example, containing 1 to 1000 mg, and in particular 5 to 200 mg of active ingredient per unit dosage form.

The exact dosage and frequency of administration depends on the particular compound of formula (I) used, the particular condition being treated, the severity of the condition being treated, the age, weight and general physical condition of the particular patient as well as other medication the individual may be taking, as is well known to those skilled in the art. Furthermore, it is evident that the effective amount may be lowered or increased depending on the response of the treated subject and/or depending on the evaluation of the physician prescribing the compounds of the instant invention. The effective amount ranges mentioned above are therefore only guidelines and are not intended to limit the scope or use of the invention to any extent.

The present disclosure is also intended to include any isotopes of atoms present in the compounds of the invention. For example, isotopes of hydrogen include tritium and deuterium and isotopes of carbon include C-13 and C-14.

- 5 The present compounds used in the current invention may also exist in their stereo-chemically isomeric form, defining all possible compounds made up of the same atoms bonded by the same sequence of bonds but having different three-dimensional structures, which are not interchangeable. Unless otherwise mentioned or indicated, the chemical designation of compounds encompasses
- 10 the mixture of all possible stereo-chemically isomeric forms, which said compounds might possess.
- Said mixture may contain all dia-stereomers and/or enantiomers of the basic molecular structure of said compound. All stereo-chemically isomeric forms of the compounds used in the present invention either in pure form or in admixture
- 15 with each other are intended to be embraced within the scope of the present invention including any racemic mixtures or racemates.

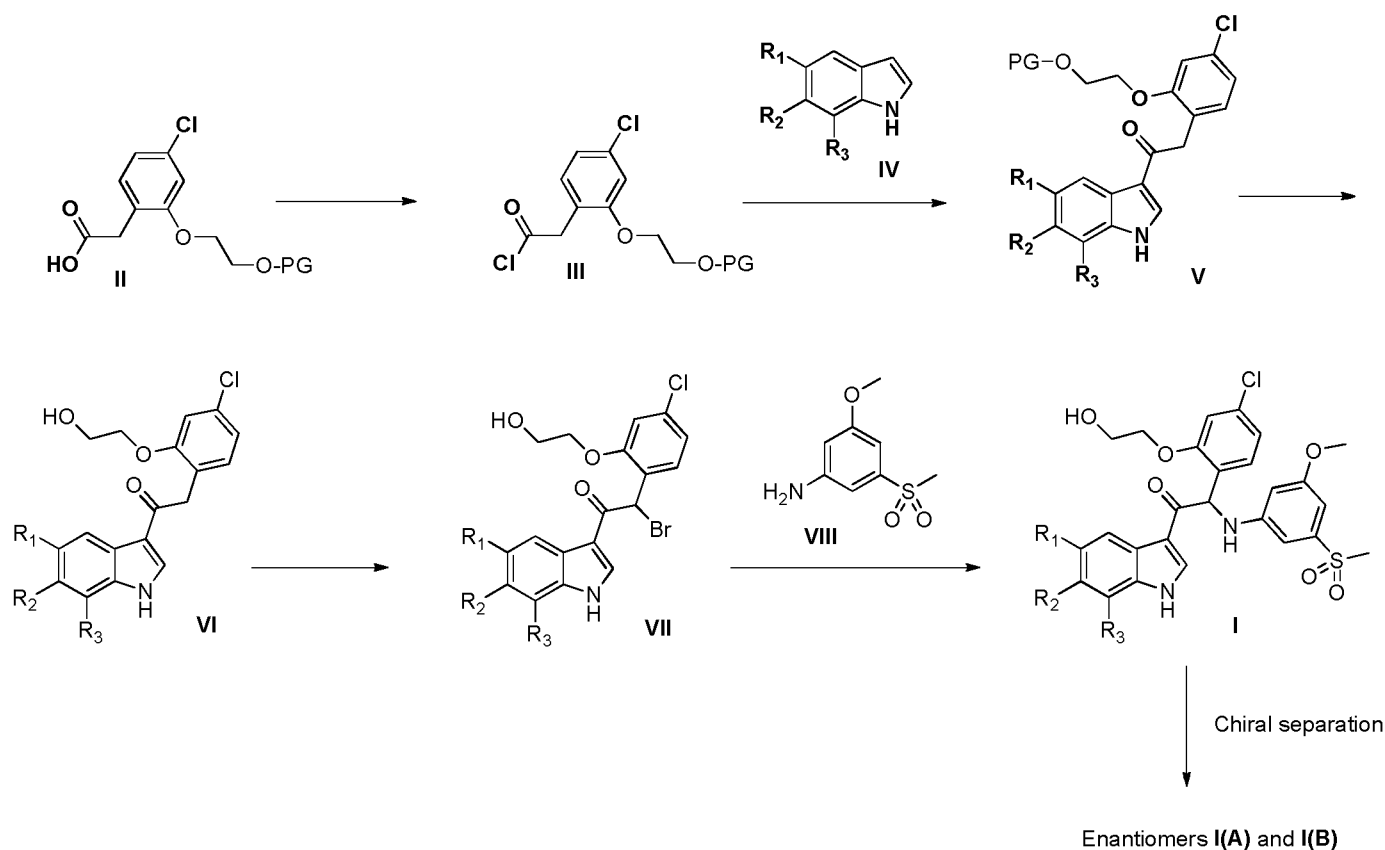
- Pure stereoisomeric forms of the compounds and intermediates as mentioned herein are defined as isomers substantially free of other enantiomeric or
- 20 diastereomeric forms of the same basic molecular structure of said compounds or intermediates. In particular, the term 'stereoisomerically pure' concerns compounds or intermediates having a stereoisomeric excess of at least 80% (i. e. minimum 90% of one isomer and maximum 10% of the other possible isomers) up to a stereoisomeric excess of 100% (i.e. 100% of one
- 25 isomer and none of the other), more in particular, compounds or intermediates having a stereoisomeric excess of 90% up to 100%, even more in particular having a stereoisomeric excess of 94% up to 100% and most in particular having a stereoisomeric excess of 97% up to 100%. The terms
- 'enantiomerically pure' and 'diastereomerically pure' should be understood in a
- 30 similar way, but then having regard to the enantiomeric excess, respectively the diastereomeric excess of the mixture in question.

- Pure stereoisomeric forms of compounds and intermediates used in this invention may be obtained by the application of art-known procedures. For
- 35 instance, enantiomers may be separated from each other by the selective crystallization of their diastereomeric salts with optically active acids or bases. Examples thereof are tartaric acid, dibenzoyltartaric acid, ditoluoyltartaric acid and camphosulfonic acid. Alternatively, enantiomers may be separated by

chromatographic techniques using chiral stationary phases. Said pure stereochemically isomeric forms may also be derived from the corresponding pure stereochemically isomeric forms of the appropriate starting materials, provided that the reaction occurs stereospecifically. Preferably, if a specific stereoisomer is desired, said compound will be synthesized by stereospecific methods of preparation. These methods will advantageously employ enantiomerically pure starting materials.

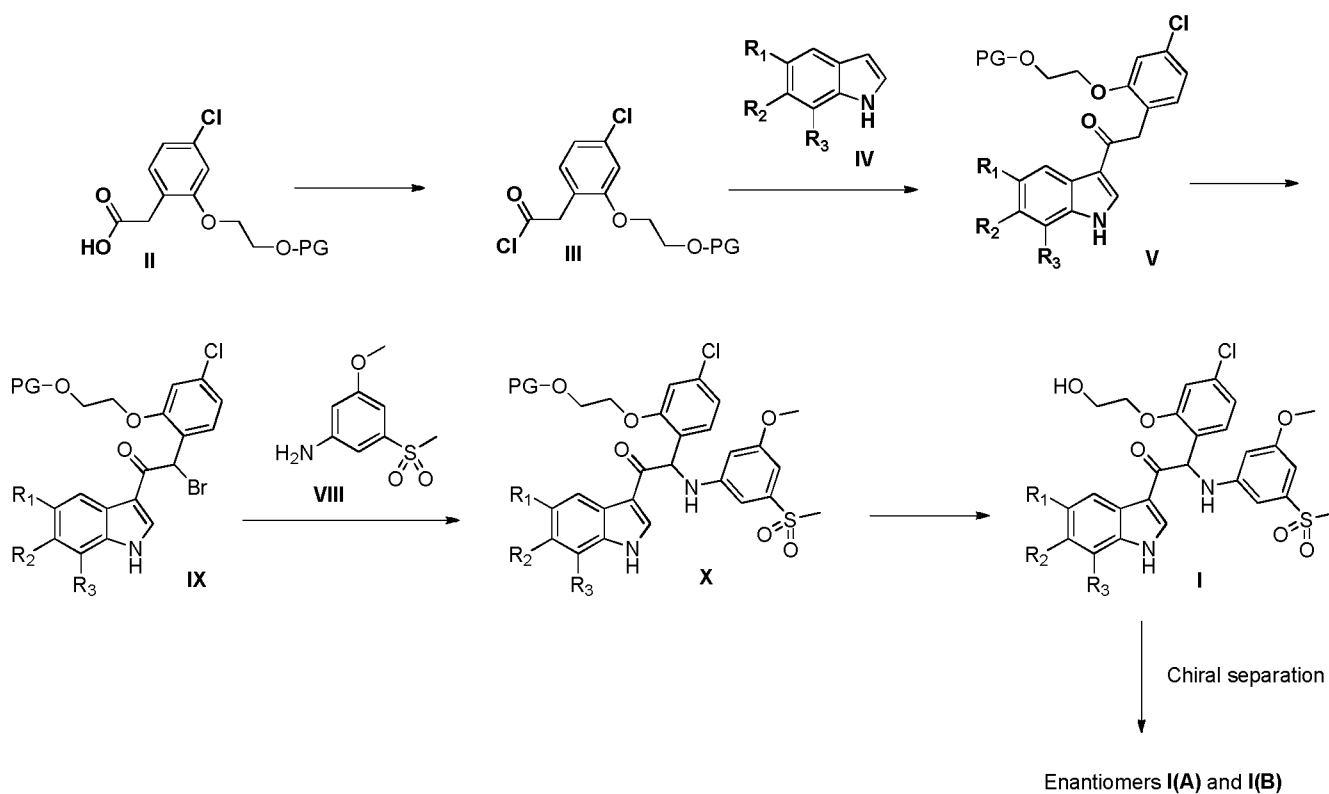
General synthetic approaches

The synthesis of compounds of general formula **I** can be performed as outlined in Scheme 1. A 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)acetic acid derivative of general formula **II**, containing an O-protecting group PG of the hydroxyl function (PG may be for example an O-benzyl protecting group), can be converted to the corresponding acid chloride derivative of general formula **III** with a chlorination reagent like for example oxalyl chloride or thionyl chloride. The Friedel-Crafts reaction of the acid chloride of general formula **III** with a substituted indole of general formula **IV** can be performed using a Lewis acid reagent like for example Et₂AlCl in a suitable solvent like for example CH₂Cl₂, and under suitable reaction conditions that typically involve cooling, to provide the 3-acylated indole of general formula **V**. Removal of the protecting group PG from the compounds of general formula **V** can be performed by for example reductive hydrogenolysis (PG = benzyl) in a suitable solvent like for example EtOAc, to provide the compounds of general formula **VI**. The introduction of an aniline moiety in alpha position to the carbonyl moiety of the compounds of general formula **VI** can be accomplished by a reaction sequence that involves for example bromination of **VI** with a reagent like for example phenyltrimethylammonium tribromide in a suitable solvent like for example THF, to provide the compounds of general formula **VII**, and subsequent reaction of the compounds of general formula **VII** with 3-methoxy-5-(methylsulfonyl)aniline (**VIII**) in a suitable solvent like for example CH₃CN, and optionally using a base like for example TEA or DIPEA, to provide the compounds of general formula **I** as racemic mixtures. The chiral separation of the compounds of general formula **I** can be performed by for example chiral chromatography to provide the Enantiomers **A** and **B** of general formula **I**.



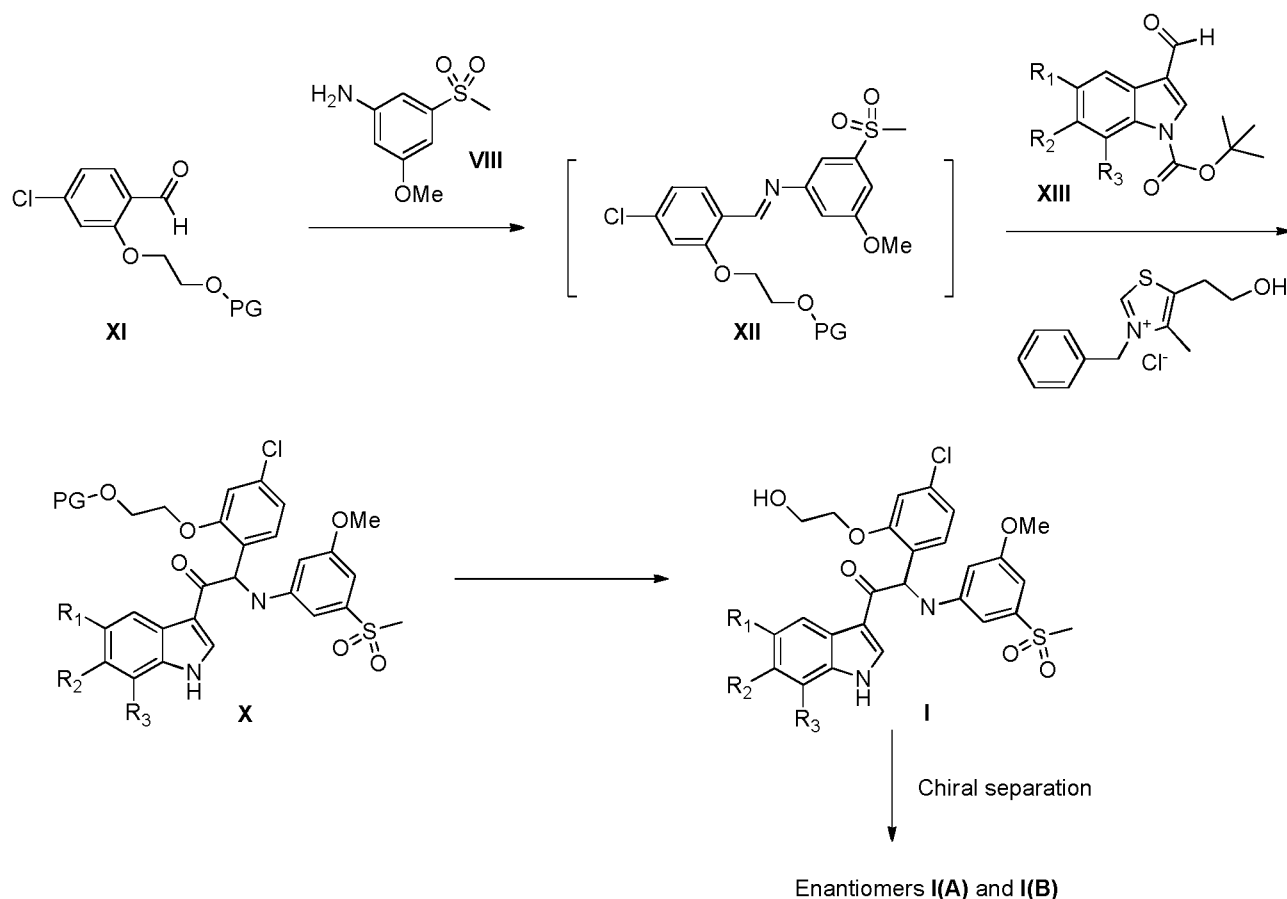
Scheme 1

- 5 Alternatively, the conversion of the intermediates of general formula **V** to the Compounds of general formula **I** can also be accomplished by the reaction sequence outlined in Scheme 2: bromination at the alpha position of the carbonyl function of the intermediates of general formula **V** with a suitable
- 10 bromination reagent such as for example phenyltrimethylammonium tribromide in a suitable solvent like for example THF, provides the compounds of general formula **IX**. Subsequent reaction of the compounds of general formula **IX** with 3-methoxy-5-(methylsulfonyl)aniline (**VIII**) in a suitable solvent like for example CH₃CN, and optionally using a base like for example TEA or DIPEA, provides the compounds of general formula **X**. After removal of the O-protecting group
- 15 (PG) from the compounds of general formula **X** by for example reductive hydrogenolysis (PG = benzyl) in suitable solvent like for example EtOAc or MeOH, the compounds of general formula **I** are generated as racemic mixtures. The chiral separation of the compounds of general formula **I** can be performed by for example chiral chromatography to provide the Enantiomers **A** and **B** of
- 20 general formula **I**.



Scheme 2

- 5 In a third approach, the synthesis of the Compounds of general formula I can be accomplished by the reaction sequence outlined in Scheme 3. This approach uses a one-pot three component reaction as the key reaction step to compose the central scaffold of the compounds of general formula I: condensation of 2-(2-(benzyloxy)ethoxy)-4-chlorobenzaldehyde (XI) with
- 10 3-methoxy-5-(methylsulfonyl)aniline (VIII) in a suitable solvent such as for example EtOH provides the intermediate imine XII, which upon addition of the substituted *N*-Boc-protected indole-carboxaldehyde of general formula XIII, and in the presence of an Umpolung catalyst such as for example 3-benzyl-5-(2-hydroxyethyl)-4-methylthiazol-3-ium, provides the compounds of general
- 15 formula X. After removal of the O-protecting group (PG) from the compounds of general formula X by for example reductive hydrogenolysis (PG = benzyl) in suitable solvent like for example EtOAc or MeOH, the compounds of general formula I are generated as racemic mixtures. The chiral separation of the compounds of general formula I can be performed by for example chiral
- 20 chromatography to provide the Enantiomers A and B of general formula I.



Scheme 3

Examples

5 LC/MS methods

The High Performance Liquid Chromatography (HPLC) measurement was performed using a LC pump, a diode-array (DAD) or a UV detector and a column as specified in the respective methods. If necessary, additional detectors were included (see table of methods below).

- 10 Flow from the column was brought to the Mass Spectrometer (MS) which was configured with an atmospheric pressure ion source. It is within the knowledge of the skilled person to set the tune parameters (e.g. scanning range, dwell time...) in order to obtain ions allowing the identification of the compound's nominal monoisotopic molecular weight (MW). Data acquisition was performed
- 15 with appropriate software.

- Compounds are described by their experimental retention times (R_t) and ions. If not specified differently in the table of data, the reported molecular ion corresponds to the $[M+H]^+$ (protonated molecule) and/or $[M-H]^-$ (deprotonated molecule). In case the compound was not directly ionizable the type of adduct
- 20 is specified (i.e. $[M+NH_4]^+$, $[M+HCOO]^-$, etc...). For molecules with multiple

isotopic patterns (Br, Cl), the reported value is the one obtained for the lowest isotope mass. All results were obtained with experimental uncertainties that are commonly associated with the method used.

- Hereinafter, "SQD" means Single Quadrupole Detector, "MSD" Mass Selective Detector, "RT" room temperature, "BEH" bridged ethylsiloxane/silica hybrid, "DAD" Diode Array Detector, "HSS" High Strength silica.

LC/MS Method codes (Flow expressed in mL/min; column temperature (T) in °C; Run time in minutes)

Method code	Instrument	Column	Mobile phase	Gradient	Flow ----- Col T	Run time (min)
LC-A	Waters: Acquity® UPLC® - DAD-SQD	Waters: BEH C18 (1.7µm, 2.1x50mm)	A: 10mM CH ₃ COONH ₄ in 95% H ₂ O + 5% CH ₃ CN B: CH ₃ CN	From 95% A to 5% A in 1.3 min, held for 0.7 min.	0.8 mL/min ----- 55°C	2
LC-B	Waters: Acquity® UPLC® - DAD-SQD	Waters: HSS T3 (1.8µm, 2.1x100mm)	A: 10mM CH ₃ COONH ₄ in 95% H ₂ O + 5% CH ₃ CN B: CH ₃ CN	From 100% A to 5% A in 2.10 min, to 0% A in 0.90 min, to 5% A in 0.5 min	0.7 mL/min ----- 55°C	3.5
LC-C	Waters: Acquity® UPLC® - DAD- Quattro Micro™	Waters: BEH C18 (1.7µm, 2.1x100mm)	A: 95% CH ₃ COONH ₄ 7mM / 5% CH ₃ CN, B: CH ₃ CN	84.2% A for 0.49 min, to 10.5% A in 2.18 min, held for 1.94 min, back to 84.2% A in 0.73 min, held for 0.73 min.	0.343 mL/min ----- 40°C	6.2
LC-D	Waters: Acquity® UPLC® - DAD- Acquity® TQ detector	Waters: BEH C18 (1.7µm, 2.1x50mm)	A: 10 mM CH ₃ COONH ₄ , pH 10 B: CH ₃ CN	80% A to 40% A in 3.4 min, to 10% A in 0.6 min, held for 1 min.	0.5 mL/min ----- 40°C	5
LC-E	Waters: Acquity® UPLC® - DAD- Acquity® TQ detector	Waters: BEH C18 (1.7µm, 2.1x50mm)	A: 10 mM CH ₃ COONH ₄ , pH 10 B: CH ₃ CN	50% A to 10% A in 3.5 min, held for 1.5 min.	0.5 mL/min ----- 40°C	5

SFC/MS methods

The SFC measurement was performed using an Analytical Supercritical fluid chromatography (SFC) system composed by a binary pump for delivering carbon dioxide (CO₂) and modifier, an autosampler, a column oven, a diode array detector equipped with a high-pressure flow cell standing up to 400 bars. If configured with a Mass Spectrometer (MS) the flow from the column was brought to the (MS). It is within the knowledge of the skilled person to set the tune parameters (e.g. scanning range, dwell time...) in order to obtain ions allowing the identification of the compound's nominal monoisotopic molecular weight (MW). Data acquisition was performed with appropriate software. Analytical SFC/MS Methods (Flow expressed in mL/min; column temperature (T) in °C; Run time in minutes, Backpressure (BPR) in bars.

Method code	column	mobile phase	gradient	Flow ----- Col T	Run time ----- BPR
SFC-A	Daicel Chiralpak® IC column (5 µm, 150 x 4.6 mm)	A: CO ₂ B: MeOH (+0.3% iPrNH ₂)	40% B hold 7 min,	3 ----- 35	7 ----- 100
SFC-B	Daicel Chiralcel® OD-3 column (3 µm, 100 x 4.6 mm)	A: CO ₂ B: EtOH (+0.3% iPrNH ₂)	30% B hold 7 min,	3.5 ----- 35	6 ----- 103
SFC-C	Daicel Chiralpak® AS3 column (3 µm, 150 x 4.6 mm)	A: CO ₂ B: EtOH +0.2% iPrNH ₂ +3% H ₂ O	10%-50% B in 6 min, hold 3.5 min	2.5 ----- 40	9.5 ----- 110
SFC-D	Daicel Chiralpak® AS-H column (5 µm, 250 x 4.6 mm)	A: CO ₂ B: EtOH +0.2% iPrNH ₂	35% B hold 4 min, to 50% in 1 min, hold 2 min	5 ----- 40	7 ----- 110
SFC-E	Daicel Chiralpak® IC column (5 µm, 150 x 4.6 mm)	A: CO ₂ B: MeOH	25% B hold 7 min,	3 ----- 35	7 ----- 100

Method code	column	mobile phase	gradient	Flow ----- Col T	Run time ----- BPR
SFC-F	Daicel Chiralpak® IA column (5 µm, 150 x 4.6 mm)	A:CO ₂ B: iPrOH +0.3% iPrNH ₂	30% B hold 7 min,	3 ----- 35	7 ----- 100

Melting Points

Values are either peak values or melt ranges, and are obtained with experimental uncertainties that are commonly associated with this analytical method.

DSC823e (indicated as DSC)

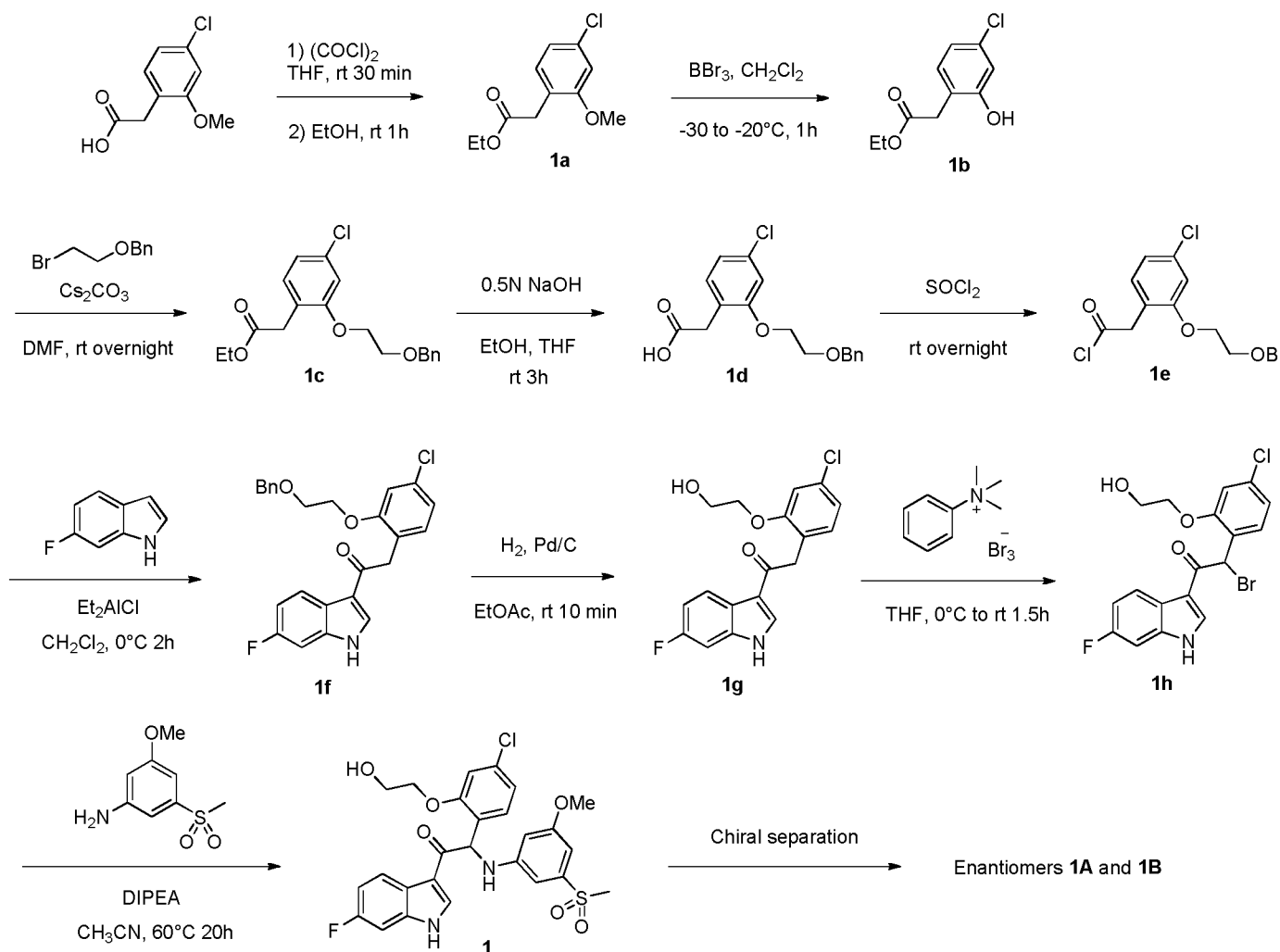
For a number of compounds, melting points were determined with a DSC823e (Mettler-Toledo). Melting points were measured with a temperature gradient of 10°C/minute. Maximum temperature was 300°C.

Optical Rotations:

Optical rotations were measured on a Perkin-Elmer 341 polarimeter with a sodium lamp and reported as follows: $[\alpha]^T_\lambda$ (λ , c g/100ml, solvent, T°C).

$[\alpha]^T_\lambda = (100\alpha) / (l \times c)$: where l is the path length in dm and c is the concentration in g/100 ml for a sample at a temperature T (°C) and a wavelength λ (in nm). If the wavelength of light used is 589 nm (the sodium D line), then the symbol D might be used instead. The sign of the rotation (+ or -) should always be given. When using this equation the concentration and solvent are always provided in parentheses after the rotation. The rotation is reported using degrees and no units of concentration are given (it is assumed to be g/100 ml).

Example 1: synthesis of 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-fluoro-1*H*-indol-3-yl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)ethanone (Compound **1**) and chiral separation into Enantiomers **1A** and **1B**.



Synthesis of intermediate 1a:

A solution of 2-(4-chloro-2-methoxyphenyl)acetic acid [CAS 170737-95-8] (20 g, 101 mmol) in dry THF (300 mL) was cooled at 0°C . Oxalyl chloride (18 mL, 202 mmol) and two drops of DMF were added. The reaction mixture was stirred at room temperature for 30 min. The solvent was evaporated under reduced pressure. The residue was dissolved in ethanol (300 mL) and the reaction mixture was stirred at room temperature for 1 h. The reaction mixture was concentrated under reduced pressure to give ethyl 2-(4-chloro-2-methoxyphenyl)acetate **1a** (23 g), which was used in the next step without further purification.

Synthesis of intermediate 1b:

To a solution of ethyl 2-(4-chloro-2-methoxyphenyl)acetate **1a** (10 g, 44 mmol) in CH₂Cl₂ (350 mL), cooled at -30 °C, was added dropwise a 1M BBr₃ solution in CH₂Cl₂ (87.5 mL, 87.5 mmol) while maintaining the temperature
5 below -20 °C. The reaction mixture was stirred at -30 °C for 1 h before quenching with methanol. The pH was adjusted to 8 by addition of an aqueous saturated solution of NaHCO₃. The phases were separated. The aqueous phase was extracted with CH₂Cl₂. The organic phases were combined, dried over MgSO₄, filtered and concentrated under reduced pressure to afford ethyl
10 2-(4-chloro-2-hydroxyphenyl)acetate **1b** (9.5 g), which was used in the next step without further purification.

Synthesis of intermediate 1c:

To a mixture of ethyl 2-(4-chloro-2-hydroxyphenyl)acetate **1b** [CAS 1261826-
15 30-5] (2.82 g, 13.1 mmol) and cesium carbonate (8.56 g, 26.3 mmol) in DMF (50 mL) was added benzyl 2-bromoethyl ether [CAS 1462-37-9] (2.29 g, 14.5 mmol). The reaction mixture was stirred at room temperature for 24 h. H₂O was added and the reaction mixture was extracted with EtOAc. The organic phase was dried over Na₂SO₄, filtered and concentrated under reduced
20 pressure. The residue was purified by flash chromatography on silica gel using a gradient of EtOAc (2% to 20%) in heptane to give ethyl 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)acetate **1c** (4.17 g).

Synthesis of intermediate 1d:

To a solution of ethyl 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)acetate **1c** (4.17 g, 12.0 mmol) in a mixture of EtOH (80 mL) and THF (40 mL) was added 0.5N NaOH (72 mL, 36.0 mmol). The reaction mixture was stirred at room temperature for 3 h. The reaction mixture was partially concentrated under reduced pressure to remove the organic solvents. The residue was acidified to
30 pH 2-3 with 1N HCl and the mixture was extracted with EtOAc. The organic phase was dried over MgSO₄, filtered and concentrated under reduced pressure to give 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)acetic acid **1d** (3.83 g).

Synthesis of intermediate 1e:

A solution of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)acetic acid **1d** (7.12 g, 22.2 mmol) in thionyl chloride (50 mL, 689 mmol) was stirred at room temperature for 18 h. The reaction mixture was concentrated under reduced

pressure and co-evaporated with toluene to give 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)acetyl chloride **1e** (7.53 g) which was used in the next step without further purification.

5 **Synthesis of intermediate 1f:**

Diethylaluminum chloride 1M in hexane (22.2 mL, 22.2 mmol) was added dropwise, at 0°C, to a solution of 6-fluoro-1*H*-indole [CAS 399-51-9] (2 g, 14.8 mmol) in CH₂Cl₂ (100 mL). After stirring for 15 min at 0°C, a solution of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)acetyl chloride **1e** (7.53 g, 22.2 mmol) in dichloromethane (75 mL) was added dropwise over 1 h, while
10 keeping the internal temperature of the reaction mixture below 4°C. The reaction mixture was stirred at 0°C for 2 h. A solution of potassium sodium tartrate tetrahydrate (Rochelle salt) [CAS 6100-16-9] (8.35 g, 29.6 mmol) in water (9 mL) was added dropwise. The reaction mixture was stirred at 0°C for
15 20 min. THF (200 mL) and Na₂SO₄ (35 g) were added and the mixture was stirred for 3 h at room temperature. The reaction mixture was filtered over dicalite® and the filter cake was washed several times with THF. The combined filtrates were evaporated under reduced pressure and co-evaporated with CH₃CN. The solid residue was stirred up in CH₃CN (20 mL) at 40°C. The
20 precipitate was filtered off, the solids were washed with CH₃CN (3x 3 mL) and dried under vacuum at 45°C to give crude compound **1f** (2.48 g). Concentration of the filtrate provided a second crop of **1f** (0.9 g). The combined fractions (3.4 g) were re-crystallized from EtOAc. The precipitate was filtered off, washed with EtOAc (3x) and dried under vacuum at 45°C to provide 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(6-fluoro-1*H*-indol-3-yl)ethanone **1f**
25 (2.21g).

Synthesis of intermediate 1g:

A mixture of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(6-fluoro-1*H*-indol-3-yl)ethanone **1f** (2.21 g, 5.05 mmol) and 10% palladium on carbon (1 g) in
30 EtOAc (75 mL) and THF (40 mL) was stirred at room temperature for 10 min under H₂ atmosphere. The reaction mixture was filtered over dicalite® and the filtrate was concentrated under reduced pressure. The residue was crystallized from a mixture of THF (10 mL) and diisopropyl ether (20 mL). The precipitate
35 was filtered off, washed with diisopropyl ether (3x) and dried under vacuum at 50°C to provide 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-fluoro-1*H*-indol-3-yl)ethanone **1g** (1.52 g).

Synthesis of intermediate 1h:

Phenyltrimethylammonium tribromide [CAS 4207-56-1] (1.73 g, 4.59 mmol) was added in portions to a cooled (0°C) solution of 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-fluoro-1*H*-indol-3-yl)ethanone **1g** (1.52 g, 4.37 mmol) in THF (60 mL). The mixture was stirred at 0°C for 45 min and at room temperature for 90 min. The precipitate was filtered off and washed with THF. The combined filtrates were concentrated under reduced pressure to give 2-bromo-2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-fluoro-1*H*-indol-3-yl)ethanone **1h** (1.87 g) which was used without further purification in the next step.

Synthesis of Compound 1 and chiral separation into Enantiomers 1A and 1B:

A mixture of 2-bromo-2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-fluoro-1*H*-indol-3-yl)ethanone **1h** (1.87 g, 4.37 mmol), 3-methoxy-5-(methylsulfonyl)aniline [CAS 62606-02-4] (1.76 g, 8.74 mmol) and diisopropylethylamine (1.51 mL, 8.74 mmol) in CH₃CN (125 mL) was stirred at 60°C for 20 h. The reaction mixture was diluted with water (500 mL) and extracted with Et₂O (2x). The combined organic fractions were washed with brine, dried over MgSO₄ and evaporated under reduced pressure. The residue was purified by flash chromatography (Stationary phase: Grace Reveleris® silica 80 g, Mobile phase: heptane/EtOAc/EtOH gradient 100/0/0 to 40/45/15). The desired fractions were combined, evaporated under reduced pressure, and co-evaporated with CH₃CN. The residue was purified via Reverse Phase HPLC (Stationary phase: Kromasil® C18 100A 5 µm (Eka Nobel), Mobile phase: Gradient from 50% ammoniumbicarbonate in water (0.25%), 50% acetonitrile to 0% ammoniumbicarbonate in water (0.25%), 100% acetonitrile). The product fractions were combined and evaporated under reduced pressure to provide Compound **1** (1400 mg) as a racemic mixture.

The chiral separation of the enantiomers of Compound **1** (1400 mg) was performed via Normal Phase Chiral separation (Stationary phase: (S,S) Whelk-O1, 5 µm with recycling peak shaving technique, Mobile phase: 100% ethanol). The product fractions were combined and evaporated under reduced pressure to provide Enantiomer **1A** (588 mg) as the first eluted product and Enantiomer **1B** (466 mg) as the second eluted product. Enantiomer **1A** was purified by flash chromatography (Stationary phase: Grace Reveleris® silica 12 g, Mobile phase: heptane/EtOAc/EtOH gradient 100/0/0 to 40/45/15). The desired fractions were combined and evaporated under reduced pressure. The residue

was stirred up in H₂O (7.5 mL) + MeOH (2.5 mL). The solids were filtered off, washed with a mixture of H₂O/MeOH 3/1 (3x), and dried at under vacuum at 45°C to provide Enantiomer **1A** (0.425 g). Enantiomer **1B** was purified by flash chromatography (Stationary phase: Grace Reveleris® silica 12 g, Mobile phase: heptane/EtOAc/EtOH gradient 100/0/0 to 40/45/15). The desired fractions were combined and evaporated under reduced pressure. The residue was stirred up in H₂O (7.5 mL) + MeOH (2.5 mL). The solids were filtered off, washed with a mixture of H₂O/MeOH 3/1 (4x), and dried under vacuum at 45°C to provide Enantiomer **1B** (0.375 g).

Compound 1:

¹H NMR (300 MHz, DMSO-*d*₆) δ ppm 3.09 (s, 3 H) 3.72 (s, 3 H) 3.85 - 4.09 (m, 2 H) 4.20 (t, *J*=4.3 Hz, 2 H) 5.30 (t, *J*=4.9 Hz, 1 H) 6.38 (d, *J*=7.5 Hz, 1 H) 6.58 (s, 1 H) 6.66 (s, 1 H) 6.91 - 7.00 (m, 2 H) 7.01 - 7.15 (m, 3 H) 7.25 (dd, *J*=9.4, 2.3 Hz, 1 H) 7.37 (d, *J*=8.3 Hz, 1 H) 8.16 (dd, *J*=8.7, 5.7 Hz, 1 H) 8.68 (s, 1 H) 12.17 (br. s., 1 H)

LC/MS (method LC-D): R_t 3.70 min, MH⁺ 547

Enantiomer 1A:

¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 3.08 (s, 3 H) 3.72 (s, 3 H) 3.88 - 4.05 (m, 2 H) 4.20 (t, *J*=4.6 Hz, 2 H) 5.26 (t, *J*=5.5 Hz, 1 H) 6.37 (d, *J*=7.8 Hz, 1 H) 6.58 (t, *J*=1.9 Hz, 1 H) 6.65 (t, *J*=2.1 Hz, 1 H) 6.92 - 6.97 (m, 2 H) 7.02 - 7.09 (m, 2 H) 7.11 (d, *J*=2.0 Hz, 1 H) 7.24 (dd, *J*=9.6, 2.4 Hz, 1 H) 7.37 (d, *J*=8.3 Hz, 1 H) 8.15 (dd, *J*=8.8, 5.6 Hz, 1 H) 8.67 (s, 1 H) 12.15 (br s, 1 H)

LC/MS (method LC-A): R_t 1.05 min, MH⁺ 547

[α]_D²⁰: +139.3° (c 0.435, DMF)

Chiral SFC (method SFC-C): R_t 3.52 min, MH⁺ 547, chiral purity 99.0%.

Enantiomer 1B:

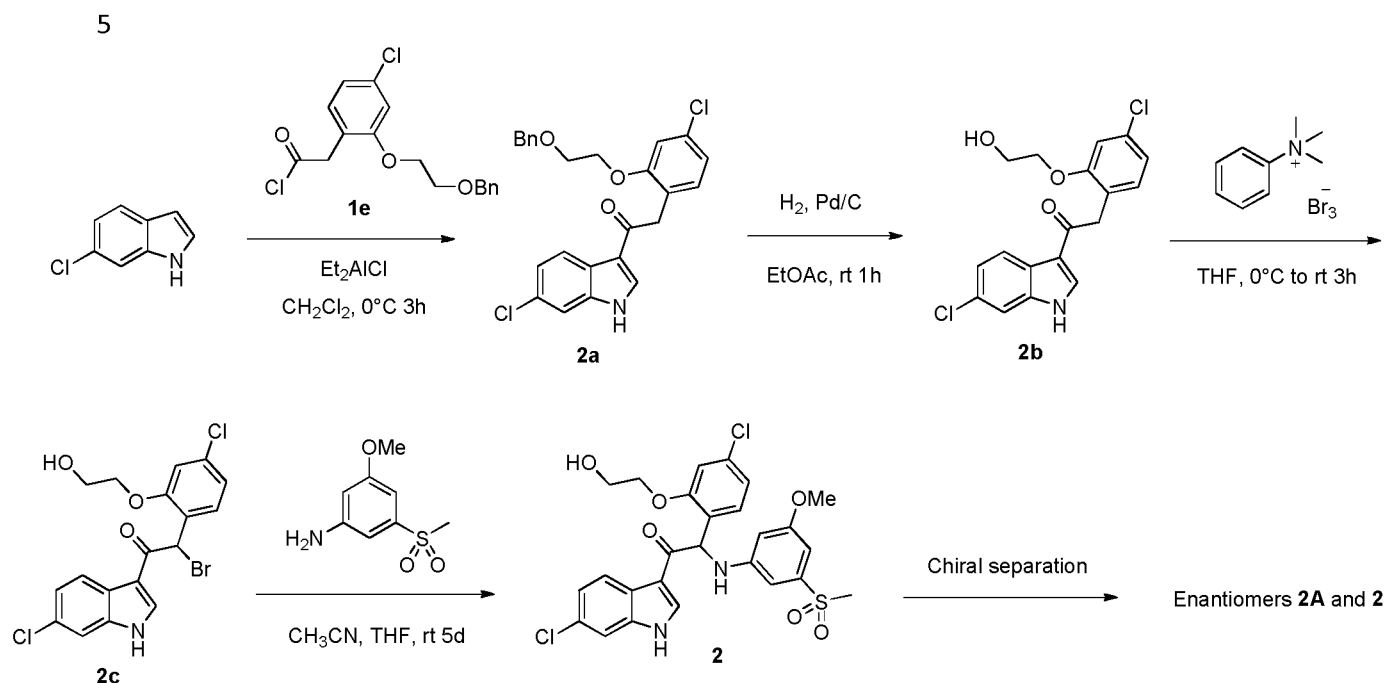
¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 3.08 (s, 3 H) 3.72 (s, 3 H) 3.88 - 4.05 (m, 2 H) 4.20 (t, *J*=4.6 Hz, 2 H) 5.27 (t, *J*=5.5 Hz, 1 H) 6.37 (d, *J*=7.7 Hz, 1 H) 6.58 (t, *J*=1.9 Hz, 1 H) 6.66 (t, *J*=2.1 Hz, 1 H) 6.92 - 6.97 (m, 2 H) 7.02 - 7.09 (m, 2 H) 7.11 (d, *J*=2.0 Hz, 1 H) 7.24 (dd, *J*=9.6, 2.4 Hz, 1 H) 7.37 (d, *J*=8.3 Hz, 1 H) 8.15 (dd, *J*=8.8, 5.6 Hz, 1 H) 8.67 (s, 1 H) 12.15 (br s, 1 H)

LC/MS (method LC-A): R_t 1.05 min, MH⁺ 547

[α]_D²⁰: -145.6° (c 0.605, DMF)

Chiral SFC (method SFC-C): R_t 4.02 min, MH⁺ 547, chiral purity 97.9%.

Example 2: synthesis of 1-(6-chloro-1*H*-indol-3-yl)-2-(4-chloro-2-(2-hydroxyethoxy) phenyl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)ethanone (Compound **2**) and chiral separation into Enantiomers **2A** and **2B**.



Synthesis of intermediate 2a:

- 10 Diethylaluminum chloride 1M in hexane (15.0 mL, 15.0 mmol) was added dropwise, at 0°C, to a solution of 6-chloro-1*H*-indole [CAS 17422-33-2] (1.52 g, 10.0 mmol) in CH₂Cl₂ (35 mL). After 30 min at 0°C, a solution of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)acetyl chloride **1e** (5.09 g, 15.0 mmol, synthesis: see Example 1) in CH₂Cl₂ (10 mL) was added slowly. The reaction
- 15 mixture was stirred at 0°C for 3 h. 1M Rochelle salt solution was added. The reaction mixture was stirred at room temperature for 1 h. The formed solids were filtered off and partitioned between EtOAc and 3N HCl. The phases were separated. The aqueous phase was extracted with EtOAc and the combined organic phases were washed with brine, dried over MgSO₄, filtered and
- 20 concentrated under reduced pressure. The residue was triturated with EtOAc. The precipitate was filtered off and dried under vacuum to yield a first batch of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(6-chloro-1*H*-indol-3-yl)ethanone **2a**. The filtrate was concentrated under reduced pressure. The residue was purified by flash chromatography on silica gel using a gradient of EtOAc (0% to
- 25 50%) in heptane. The fractions containing the desired product were combined

and concentrated under reduced pressure. The residue was triturated with EtOAc. The solids were filtered and dried under vacuum to yield a second crop of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(6-chloro-1*H*-indol-3-yl)ethanone **2a** (overall amount for the two batches: 2.10 g).

5

Synthesis of intermediate 2b:

A mixture of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(6-chloro-1*H*-indol-3-yl)ethanone **2a** (1.98 g, 4.36 mmol) and 10% palladium on carbon (0.2 g) in EtOAc (40 mL) was stirred at room temperature for 1 h under H₂ atmosphere.

10 The reaction mixture was diluted with THF and filtered through celite®. The filtrate was concentrated under reduced pressure. The residue was triturated with EtOAc. The solids were filtered off and dried under vacuum to give 1-(6-chloro-1*H*-indol-3-yl)-2-(4-chloro-2-(2-hydroxyethoxy)phenyl)ethanone **2b** (1.29 g).

15

Synthesis of intermediate 2c:

A solution of phenyltrimethylammonium tribromide [CAS 4207-56-1] (1.47 g, 3.91 mmol) in THF (10 mL) was added dropwise, at 0°C, to a solution of 1-(6-chloro-1*H*-indol-3-yl)-2-(4-chloro-2-(2-hydroxyethoxy)phenyl)ethanone **2b** (1.29 g, 3.55 mmol) in THF (25 mL). The mixture was stirred at room temperature for 3 h. The precipitate was filtered off and washed with THF. The filtrate was concentrated under reduced pressure to give 2-bromo-1-(6-chloro-1*H*-indol-3-yl)-2-(4-chloro-2-(2-hydroxyethoxy)phenyl)ethanone **2c** (1.57 g) which was used in the next step without further purification.

25

Synthesis of Compound 2 and chiral separation into Enantiomers 2A and 2B:

A mixture of 2-bromo-1-(6-chloro-1*H*-indol-3-yl)-2-(4-chloro-2-(2-hydroxyethoxy)phenyl)ethanone **2c** (1.57 g, 3.55 mmol) and 3-methoxy-5-(methylsulfonyl)aniline [CAS 62606-02-4] (2.14 g, 10.7 mmol) in CH₃CN (35 mL) and THF (10 mL) was stirred at room temperature for 5 days. The reaction mixture was concentrated under reduced pressure. The residue was dissolved in EtOAc and washed with 1N HCl. The organic phase was washed with 1N HCl and an aqueous saturated NaHCO₃ solution, dried over Na₂SO₄, filtered and concentrated under reduced pressure. The residue was triturated with EtOAc and THF. The solids were filtered off. The filtrate was concentrated under reduced pressure and the residue was purified by flash chromatography on silica gel using a gradient of EtOAc (10% to 100%) in CH₂Cl₂. The fractions

35

containing the desired product were combined with the solids previously obtained and concentrated under reduced pressure. The residue was purified by flash chromatography on silica gel using EtOAc (50%) in CH₂Cl₂ as eluent to afford 1-(6-chloro-1*H*-indol-3-yl)-2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)ethanone (Compound **2**, 0.99 g) as a racemic mixture.

The chiral separation of the enantiomers of Compound **2** (894 mg) was performed via Normal Phase Chiral separation (Stationary phase: AS 20 µm, Mobile phase: 100% methanol). The product fractions were combined and evaporated to provide Enantiomer **2A** (324 mg) as the first eluted product and Enantiomer **2B** (328 mg) as the second eluted product. Both enantiomers were solidified as follows: the solids were stirred up in a 1/1 mixture of water and MeOH (10 mL) for 1 h, filtered off and dried under vacuum at 50°C to give Enantiomer **2A** (253 mg) and Enantiomer **2B** (252 mg) as white powders.

Compound **2**:

¹H NMR (300 MHz, DMSO-*d*₆) δ ppm 3.08 (s, 3 H) 3.72 (s, 3 H) 3.83 – 4.10 (m, 2 H) 4.20 (m, 2 H) 5.31 (br. s., 1 H) 6.38 (d, *J*=7.9 Hz, 1 H) 6.58 (s, 1 H) 6.66 (s, 1 H) 6.90 - 6.99 (m, 2 H) 7.02 - 7.15 (m, 2 H) 7.23 (dd, *J*=8.6, 1.6 Hz, 1 H) 7.36 (d, *J*=8.3 Hz, 1 H) 7.50 (s, 1 H) 8.15 (d, *J*=8.5 Hz, 1 H) 8.70 (s, 1 H) 12.23 (br. s., 1 H)

LC/MS (method LC-E): R_t 1.37 min, MH⁺ 563

Enantiomer **2A**:

¹H NMR (360 MHz, DMSO-*d*₆) δ ppm 3.08 (s, 3 H) 3.72 (s, 3 H) 3.86 - 4.07 (m, 2 H) 4.20 (br t, *J*=4.5 Hz, 2 H) 5.30 (t, *J*=5.7 Hz, 1 H) 6.37 (br d, *J*=7.7 Hz, 1 H) 6.58 (s, 1 H) 6.65 (br s, 1 H) 6.90 - 6.99 (m, 2 H) 7.05 - 7.14 (m, 2 H) 7.22 (dd, *J*=8.5, 1.9 Hz, 1 H) 7.36 (d, *J*=8.3 Hz, 1 H) 7.50 (d, *J*=1.8 Hz, 1 H) 8.15 (d, *J*=8.5 Hz, 1 H) 8.70 (s, 1 H) 12.23 (s, 1 H)

LC/MS (method LC-A): R_t 1.11 min, MH⁺ 563

[α]_D²⁰: +149.5° (c 0.43, DMF)

Chiral SFC (method SFC-C): R_t 4.15 min, MH⁺ 563, chiral purity 100%.

Melting point: 122 °C

Enantiomer **2B**:

¹H NMR (360 MHz, DMSO-*d*₆) δ ppm 3.08 (s, 3 H) 3.72 (s, 3 H) 3.86 - 4.07 (m, 2 H) 4.19 (br t, *J*=4.5 Hz, 2 H) 5.30 (t, *J*=5.7 Hz, 1 H) 6.37 (br d, *J*=7.7 Hz,

1 H) 6.58 (s, 1 H) 6.66 (br s, 1 H) 6.90 - 6.99 (m, 2 H) 7.05 - 7.14 (m, 2 H) 7.23 (dd, $J=8.6, 1.8$ Hz, 1 H) 7.36 (d, $J=8.3$ Hz, 1 H) 7.50 (d, $J=1.9$ Hz, 1 H) 8.15 (d, $J=8.5$ Hz, 1 H) 8.70 (s, 1 H) 12.23 (s, 1 H)

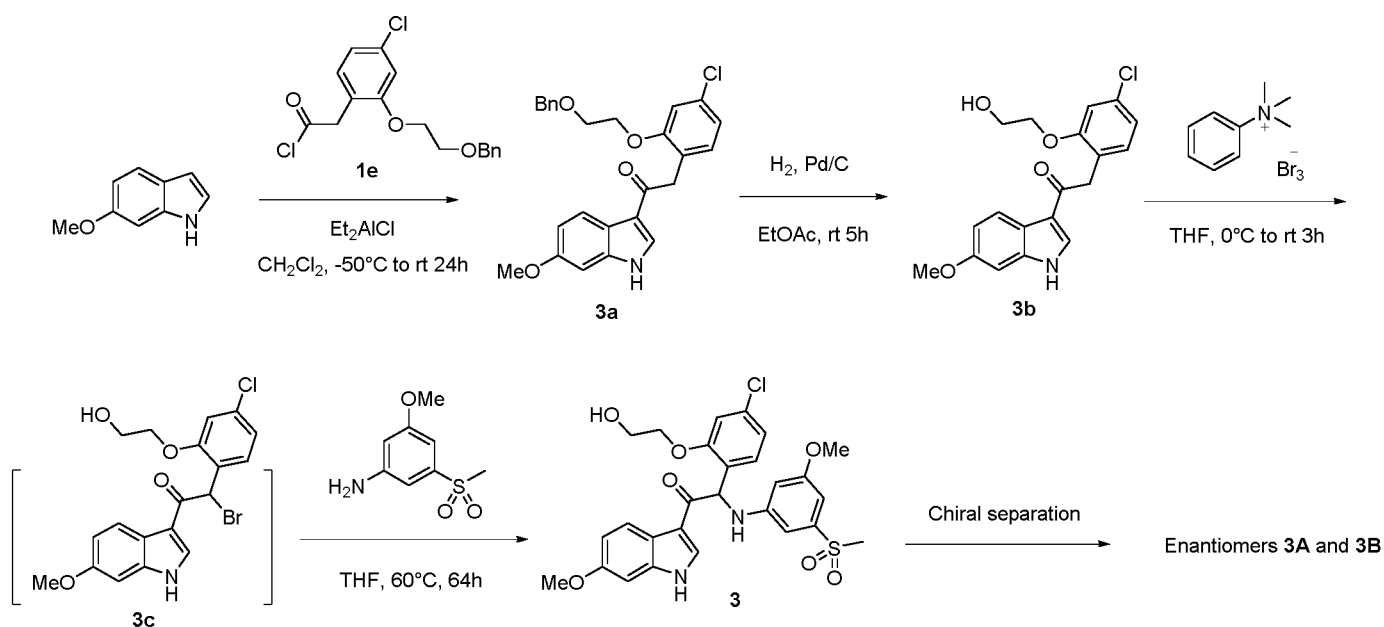
LC/MS (method LC-A): R_t 1.11 min, MH^+ 563

5 $[\alpha]_D^{20}$: -144.1° (c 0.401, DMF)

Chiral SFC (method SFC-C): R_t 4.68 min, MH^+ 563, chiral purity 100%.

Melting point: 121°C

Example 3: synthesis of 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-methoxy-1*H*-indol-3-yl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)ethanone (Compound **3**) and chiral separation into Enantiomers **3A** and **3B**.



Synthesis of intermediate **3a**:

- 15 Diethylaluminum chloride 1M in hexane (17.2 mL, 17.2 mmol) was added dropwise, at -50°C , to a suspension of 6-methoxy-1*H*-indole [CAS 3189-13-7] (1.69 g, 11.5 mmol) in CH_2Cl_2 (56 mL). After 30 min at -50°C , a solution of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)acetyl chloride **1e** (5.82 g, 17.2 mmol, synthesis: see Example 1) in CH_2Cl_2 (23 mL) was slowly added. The reaction
- 20 mixture was stirred at -50°C for 3 h, was allowed to slowly warm to room temperature and was stirred at room temperature overnight. The reaction mixture was diluted with CH_2Cl_2 and was poured out into a 1M Rochelle salt solution. The mixture was stirred at room temperature for 2 h. The phases were separated. The organic phase was washed with H_2O and brine, dried over
- 25 Na_2SO_4 , filtered and concentrated under reduced pressure. The residue was

purified by flash chromatography on silica gel using a gradient of EtOAc (20% to 60%) in heptane. The fractions containing desired product were combined and evaporated under reduced pressure. The residue was triturated with EtOAc. The solids were filtered off and dried under vacuum, giving 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(6-methoxy-1*H*-indol-3-yl)ethanone **3a** (1.39 g).

Synthesis of intermediate 3b:

A mixture of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(6-methoxy-1*H*-indol-3-yl)ethanone **3a** (2.13 g, 4.79 mmol) and 10% palladium on carbon (0.2 g) in EtOAc (125 mL) was stirred at room temperature for 5 h under H₂ atmosphere. The reaction mixture was filtered through celite®. The filter pad was washed with THF. The combined filtrates were concentrated under reduced pressure. The residue was triturated with CH₂Cl₂. The solids were filtered off and dried under vacuum to give 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-methoxy-1*H*-indol-3-yl)ethanone **3b** (1.38 g).

Synthesis of Compound 3 and chiral separation into Enantiomers 3A and 3B:

A solution of phenyltrimethylammonium tribromide [CAS 4207-56-1] (1.57 g, 4.18 mmol) in THF (27 mL) was added dropwise, at 0°C, to a solution of 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-methoxy-1*H*-indol-3-yl)ethanone **3b** (1.38 g, 3.83 mmol) in THF (37 mL). The mixture was stirred at 0°C for 1 h and at room temperature for 3 h. 3-Methoxy-5-(methylsulfonyl)aniline [CAS 62606-02-4] (3.85 g, 19.1 mmol) was added and the reaction mixture was heated at 60°C for 64 h. The reaction mixture was diluted with EtOAc and washed with 1N HCl. The phases were separated. The organic phase was washed with 1N HCl, an aqueous saturated NaHCO₃ solution, H₂O and brine, dried over Na₂SO₄, filtered and concentrated under reduced pressure. The residue was purified by flash chromatography on silica gel using a gradient of MeOH (0% to 10%) in CH₂Cl₂. Subsequent purification by flash chromatography on silica gel using a gradient of EtOAc (50% to 95%) in heptane followed by precipitation from EtOAc furnished 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-methoxy-1*H*-indol-3-yl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)ethanone (Compound **3**, 1.12 g), as a racemic mixture.

The chiral separation of the enantiomers of Compound **3** (1.12 g) was performed via Normal Phase Chiral separation (Stationary phase: AS 20 µm,

Mobile phase: 100% methanol). The product fractions were combined and evaporated under reduced pressure to provide Enantiomer **3A** as the first eluted product and Enantiomer **3B** as the second eluted product. Enantiomer **3A** was stirred up in a mixture of Et₂O (6 mL) and CH₃CN (0.3 mL). The solids were filtered off, washed with Et₂O (5x 1.5 mL) and dried under vacuum to provide Enantiomer **3A** (392 mg) as a powder. Enantiomer **3B** was stirred up in a mixture of Et₂O (3 mL) and CH₃CN (0.15 mL). The solids were filtered off, washed with Et₂O (5x 1.0 mL) and dried under vacuum to provide Enantiomer **3B** (172 mg) as a powder.

Compound 3:

¹H NMR (300 MHz, DMSO-*d*₆) δ ppm 3.08 (s, 3 H) 3.72 (s, 3 H) 3.76 (s, 3 H) 3.86 – 4.10 (m, 2 H) 4.19 (m, 2 H) 5.30 (t, *J*=5.3 Hz, 1 H) 6.34 (d, *J*=7.9 Hz, 1 H) 6.57 (s, 1 H) 6.65 (s, 1 H) 6.83 (dd, *J*=8.7, 2.0 Hz, 1 H) 6.94 (m, 3 H) 7.06 (d, *J*=7.7 Hz, 1 H) 7.10 (s, 1 H) 7.36 (d, *J*=8.6 Hz, 1 H) 8.02 (d, *J*=8.7 Hz, 1 H) 8.55 (s, 1 H) 11.92 (br. s., 1 H)

LC/MS (method LC-D): R_t 3.49 min, MH⁺ 559

Enantiomer 3A:

¹H NMR (360 MHz, DMSO-*d*₆) δ ppm 3.08 (s, 3 H) 3.72 (s, 3 H) 3.76 (s, 3 H) 3.89 - 4.07 (m, 2 H) 4.19 (t, *J*=4.7 Hz, 2 H) 5.26 - 5.33 (m, 1 H) 6.34 (d, *J*=7.8 Hz, 1 H) 6.55 - 6.58 (m, 1 H) 6.63 - 6.67 (m, 1 H) 6.83 (dd, *J*=8.7, 2.3 Hz, 1 H) 6.91 - 6.97 (m, 3 H) 7.05 (d, *J*=7.8 Hz, 1 H) 7.11 (d, *J*=2.0 Hz, 1 H) 7.36 (d, *J*=8.3 Hz, 1 H) 8.03 (d, *J*=8.7 Hz, 1 H) 8.55 (s, 1 H) 11.92 (br s, 1 H)

LC/MS (method LC-B): R_t 1.90 min, MH⁺ 559

[α]_D²⁰: +139.1° (c 0.445, DMF)

Chiral SFC (method SFC-C): R_t 4.04 min, MH⁺ 559, chiral purity 100%.

Melting point: 146 °C

Enantiomer 3B:

¹H NMR (360 MHz, DMSO-*d*₆) δ ppm 3.08 (s, 3 H) 3.72 (s, 3 H) 3.76 (s, 3 H) 3.89 - 4.07 (m, 2 H) 4.19 (t, *J*=4.7 Hz, 2 H) 5.26 - 5.33 (m, 1 H) 6.35 (d, *J*=7.7 Hz, 1 H) 6.55 - 6.58 (m, 1 H) 6.63 - 6.67 (m, 1 H) 6.83 (dd, *J*=8.7, 2.3 Hz, 1 H) 6.91 - 6.97 (m, 3 H) 7.05 (d, *J*=7.8 Hz, 1 H) 7.11 (d, *J*=2.0 Hz, 1 H) 7.36 (d, *J*=8.3 Hz, 1 H) 8.03 (d, *J*=8.7 Hz, 1 H) 8.55 (s, 1 H) 11.92 (br s, 1 H)

LC/MS (method LC-B): R_t 1.89 min, MH⁺ 559

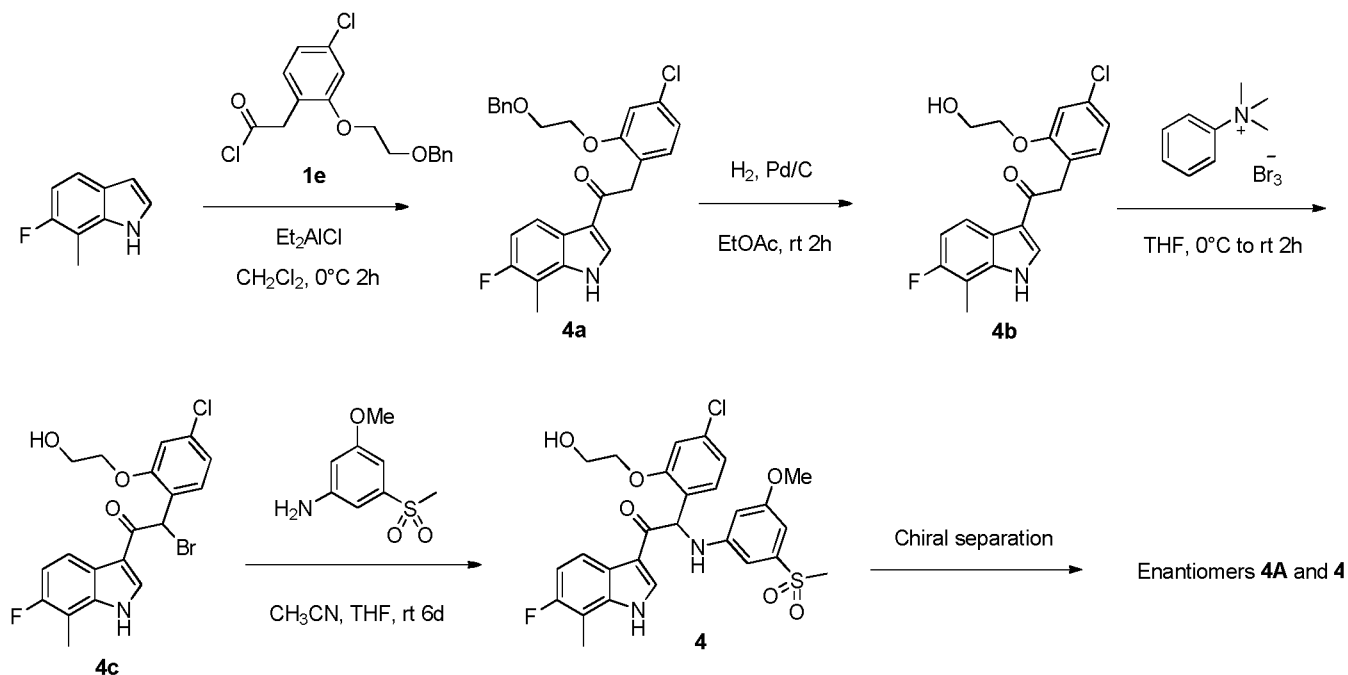
[α]_D²⁰: -136.0° (c 0.455, DMF)

Chiral SFC (method SFC-C): R_t 4.47 min, MH⁺ 559, chiral purity 100%.

Melting point: 149°C

Example 4: synthesis of 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-fluoro-7-methyl-1*H*-indol-3-yl)-2-((3-methoxy-5-

- 5 (methylsulfonyl)phenyl)amino)ethanone (Compound **4**) and chiral separation into Enantiomers **4A** and **4B**.



10 **Synthesis of intermediate 4a:**

Diethylaluminum chloride 1M in hexane (12.2 mL, 12.2 mmol) was added dropwise, at 0°C, to a suspension of 6-fluoro-7-methyl-1*H*-indole [CAS 57817-10-4] (1.20 g, 8.04 mmol) in CH₂Cl₂ (17 mL). After 30 min at 0°C, a solution of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)acetyl chloride **1e** (4.09 g,

- 15 12.1 mmol, synthesis: see Example 1) in CH₂Cl₂ (17 mL) was slowly added.

The reaction mixture was stirred at 0°C for 2 h. 1M Rochelle salt solution was added and the mixture was vigorously stirred for 2 h. EtOAc was added. The phases were separated. The aqueous phase was extracted with EtOAc. The organic phases were combined, dried over MgSO₄, filtered and concentrated

- 20 under reduced pressure. The residue was purified by flash chromatography on silica gel using a gradient of EtOAc (5% to 50%) in CH₂Cl₂. Further purification by flash chromatography on silica gel using a gradient of EtOAc (5% to 50%) in heptane furnished 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(6-fluoro-7-methyl-1*H*-indol-3-yl)ethanone **4a** (2.94 g).

Synthesis of intermediate 4b:

A mixture of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(6-fluoro-7-methyl-1*H*-indol-3-yl)ethanone **4a** (2.69 g, 5.95 mmol) and 10% palladium on carbon (0.3 g) in EtOAc (150 mL) was stirred at room temperature for 2 h under H₂ atmosphere. The reaction mixture was filtered through celite®. The filtrate was concentrated under reduced pressure. The residue was triturated with EtOAc. The solids were filtered off and dried under vacuum to give 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-fluoro-7-methyl-1*H*-indol-3-yl)ethanone **4b** (1.15 g).

Synthesis of intermediate 4c:

A solution of phenyltrimethylammonium tribromide [CAS 4207-56-1] (1.09 g, 2.89 mmol) in THF (18 mL) was added dropwise, at 0°C, to a solution of 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-fluoro-7-methyl-1*H*-indol-3-yl)ethanone **4b** (0.95 g, 2.63 mmol) in THF (25 mL). The mixture was stirred at 0°C for 15 min and at room temperature for 2 h. The precipitate was filtered off and washed with EtOAc. The filtrate was concentrated under reduced pressure to give 2-bromo-2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-fluoro-7-methyl-1*H*-indol-3-yl)ethanone **4c** (1.16 g) which was used in the next step without further purification.

Synthesis of Compound 4 and chiral separation into Enantiomers 4A and 4B:

A mixture of 2-bromo-2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-fluoro-7-methyl-1*H*-indol-3-yl)ethanone **4c** (1.16 g, 2.63 mmol) and 3-methoxy-5-(methylsulfonyl)aniline [CAS 62606-02-4] (1.59 g, 7.90 mmol) in CH₃CN (6 mL) and THF (6 mL) was stirred at room temperature for 6 days. The reaction mixture was diluted with EtOAc and washed with 1N HCl. The organic phase was washed with 1N HCl, an aqueous saturated NaHCO₃ solution, H₂O and brine, dried over MgSO₄, filtered and concentrated under reduced pressure.

The residue was purified by flash chromatography on silica gel using a gradient of EtOAc (15% to 70%) in CH₂Cl₂. The fractions containing the desired product were combined and concentrated under reduced pressure. The residue was triturated with CH₂Cl₂. The solids were filtered off and dried under vacuum to give 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-fluoro-7-methyl-1*H*-indol-3-yl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)ethanone (Compound **4**, 1.05 g) as a racemic mixture.

The chiral separation of the enantiomers of Compound **4** (1.37 g) was performed via chiral SFC (Stationary phase: Chiralpak® IA 5 µm 250 x 20 mm,

Mobile phase: 36.2% MeOH, 60% CO₂, 3.8% DCM). The product fractions were combined and evaporated under reduced pressure to provide Enantiomer **4A** (548 mg) as the first eluted product and Enantiomer **4B** (574 mg) as the second eluted product. Enantiomer **4A** was stirred up in a mixture of Et₂O (6 mL) and CH₃CN (0.25 mL). The solids were filtered off, washed with Et₂O (3x 1.5 mL) and dried under vacuum at 50°C to provide Enantiomer **4A** (369 mg) as a powder. Enantiomer **4B** was stirred up in a mixture of Et₂O (6 mL) and CH₃CN (0.25 mL). The solids were filtered off, washed with Et₂O (5x 1.0 mL) and dried under vacuum at 50°C to provide Enantiomer **4B** (352 mg) as a powder.

Compound 4:

¹H NMR (300 MHz, DMSO-*d*₆) δ ppm 2.39 (s, 3 H) 3.09 (s, 3 H) 3.72 (s, 3 H) 3.88 – 4.10 (m, 2 H) 4.19 (m, 2 H) 5.32 (br. s., 1 H) 6.40 (d, *J*=7.7 Hz, 1 H) 6.57 (s, 1 H) 6.67 (s, 1 H) 6.89 - 7.00 (m, 2 H) 7.01 - 7.09 (m, 2 H) 7.11 (d, *J*=1.8 Hz, 1 H) 7.36 (d, *J*=8.3 Hz, 1 H) 7.98 (dd, *J*=8.5, 5.2 Hz, 1 H) 8.64 (s, 1 H) 12.24 (br. s., 1 H)

LC/MS (method LC-E): R_t 1.31 min, MH⁺ 561

Enantiomer 4A:

¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 2.38 (s, 3 H) 3.08 (s, 3 H) 3.72 (s, 3 H) 3.90 - 4.07 (m, 2 H) 4.19 (t, *J*=4.7 Hz, 2 H) 5.28 (br t, *J*=5.3 Hz, 1 H) 6.40 (d, *J*=7.8 Hz, 1 H) 6.57 (t, *J*=1.9 Hz, 1 H) 6.65 - 6.68 (m, 1 H) 6.92 - 6.97 (m, 2 H) 6.98 - 7.06 (m, 2 H) 7.10 (d, *J*=2.0 Hz, 1 H) 7.36 (d, *J*=8.3 Hz, 1 H) 7.98 (dd, *J*=8.7, 5.2 Hz, 1 H) 8.62 (s, 1 H) 12.21 (br s, 1 H)

LC/MS (method LC-A): R_t 1.10 min, MH⁺ 561

[α]_D²⁰: +116.9° (c 0.575, DMF)

Chiral SFC (method SFC-C): R_t 3.54 min, MH⁺ 561, chiral purity 100%.

Enantiomer 4B:

¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 2.35 - 2.44 (m, 3 H) 3.09 (s, 3 H) 3.72 (s, 3 H) 3.90 - 4.08 (m, 2 H) 4.20 (t, *J*=4.7 Hz, 2 H) 5.29 (br s, 1 H) 6.40 (d, *J*=7.7 Hz, 1 H) 6.56 - 6.60 (m, 1 H) 6.64 - 6.70 (m, 1 H) 6.92 - 6.97 (m, 2 H) 6.98 - 7.07 (m, 2 H) 7.11 (d, *J*=2.0 Hz, 1 H) 7.36 (d, *J*=8.3 Hz, 1 H) 7.98 (dd, *J*=8.8, 5.2 Hz, 1 H) 8.63 (s, 1 H) 12.21 (br s, 1 H)

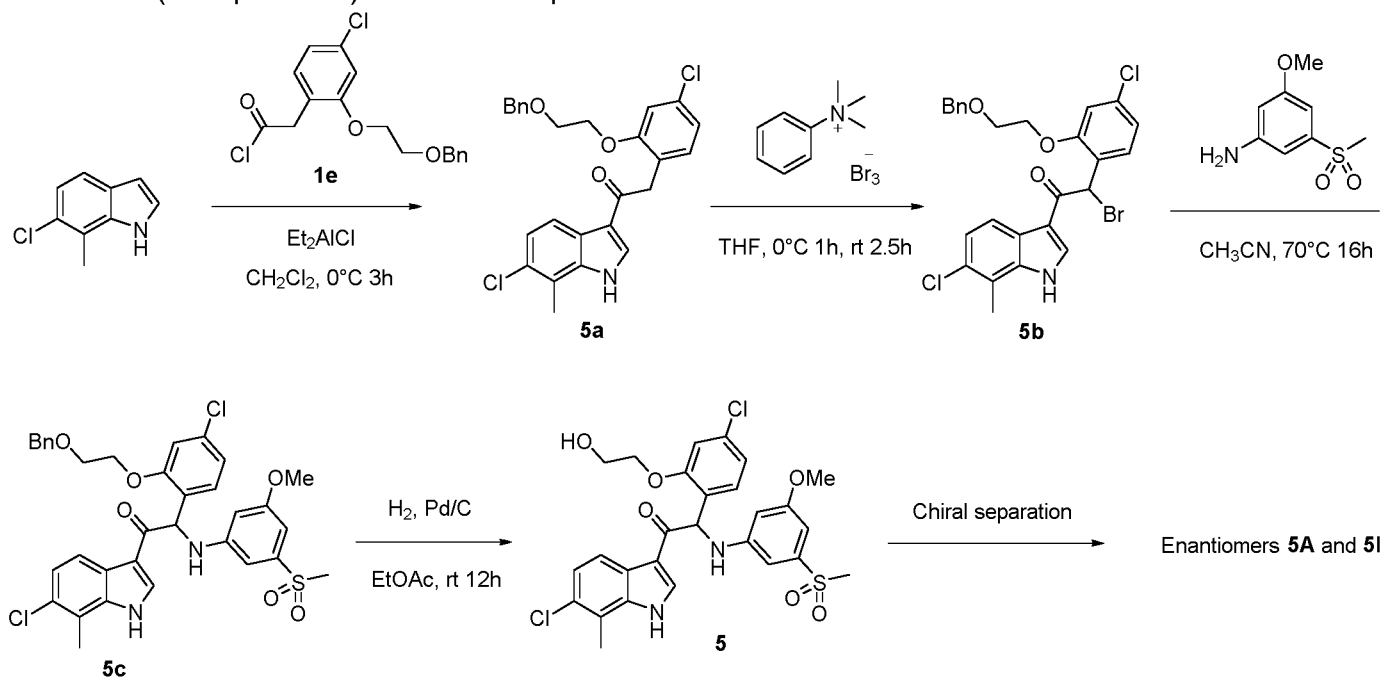
LC/MS (method LC-B): R_t 2.04 min, MH⁺ 561

[α]_D²⁰: -115.4° (c 0.455, DMF)

Chiral SFC (method SFC-C): R_t 4.09 min, MH^+ 561, chiral purity 100%.

Melting point: 173°C

- Example 5:** synthesis of 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-chloro-7-methyl-1*H*-indol-3-yl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)ethanone (Compound **5**) and chiral separation into Enantiomers **5A** and **5B**.



10 **Synthesis of intermediate 5a:**

- Diethylaluminum chloride 1M in hexane (8.91 mL, 8.91 mmol) was added dropwise, at 0°C, to a solution of 6-chloro-7-methyl-1*H*-indole [CAS 57817-09-1] (0.984 g, 5.94 mmol) in CH_2Cl_2 (40 mL). After 30 min at 0°C, 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)acetyl chloride **1e** (2.11 g, 6.22 mmol) in CH_2Cl_2 (40 mL) was added slowly at 0°C. The reaction was stirred at 0°C for 3 h. Ice-water was added and the reaction mixture was extracted with CH_2Cl_2 . The organic layer was dried over $MgSO_4$, filtered and the solvent was evaporated under reduced pressure. The crude product was purified by flash chromatography on silica gel (15-40 μm , 120 g, heptane/EtOAc 70/30). The pure fractions were combined and evaporated to dryness to give 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(6-chloro-7-methyl-1*H*-indol-3-yl)ethanone **5a** (1.08 g).

Synthesis of intermediate 5b:

Under a N₂ flow at 0°C, a solution of phenyltrimethylammonium tribromide [CAS 4207-56-1] (0.91 g, 2.42 mmol) in THF (40 mL) was added dropwise to a solution of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(6-chloro-7-methyl-1*H*-indol-3-yl)ethanone **5a** (1.08 g, 2.31 mmol) in THF (40 mL). The mixture was stirred at 0°C for 1 h, the cooling bath was removed and stirring was continued at room temperature for 2.5 h. The precipitate was filtered off and rinsed with EtOAc. The filtrate was concentrated under reduced pressure to give 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-2-bromo-1-(6-chloro-7-methyl-1*H*-indol-3-yl)ethanone **5b** (1.3 g).

Synthesis of intermediate 5c:

A mixture of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-2-bromo-1-(6-chloro-7-methyl-1*H*-indol-3-yl)ethanone **5b** (1.3 g, 2.38 mmol) and 3-methoxy-5-(methylsulfonyl)aniline [CAS 62606-02-4] (1.43 g, 7.13 mmol) in CH₃CN (80 mL) was stirred at 70°C for 16 h. The solvent was removed under reduced pressure. The crude product was purified by flash chromatography on silica gel (15-40 µm, 24 g, heptane/EtOAc 70/30). The pure fractions were combined and evaporated to dryness to give 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(6-chloro-7-methyl-1*H*-indol-3-yl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)ethanone **5c** (1.1 g).

Synthesis of Compound 5 and chiral separation of Enantiomers 5A and 5B:

A mixture of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(6-chloro-7-methyl-1*H*-indol-3-yl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)ethanone **5c** (0.8 g, 1.2 mmol) in EtOAc (40 mL) was hydrogenated under atmospheric pressure of H₂ for 12 h with Pd/C (10%) (54 mg, 0.05 mmol) as catalyst. The mixture was filtered through a pad of celite® and washed with EtOAc, and then with CH₂Cl₂/CH₃OH 90/10. The filtrate was concentrated under reduced pressure. CH₃CN was added and the solid formed was filtered off and dried to give 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-chloro-7-methyl-1*H*-indol-3-yl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)ethanone (Compound **5**) as a racemic mixture.

The enantiomers of Compound **5** (642 mg) were separated via chiral SFC (Stationary phase: Chiralpak® ICOD-H 5 µm 250 x 30 mm, Mobile phase: 60% CO₂, 40% MeOH (+ 0.3% *i*PrNH₂)) yielding 301 mg of the first eluted enantiomer and 320 mg of the second eluted enantiomer. The first eluted

enantiomer was crystallized from CH₃CN. The precipitate was filtered off and dried to give 198 mg of Enantiomer **5A**. The second eluted enantiomer was crystallized from CH₃CN. The precipitate was filtered off and dried to give 198 mg of Enantiomer **5B**.

5

Compound 5:

¹H NMR (500 MHz, DMSO-*d*₆) δ ppm 3.09 (s, 3 H) 3.72 (s, 3 H) 3.91 - 4.06 (m, 2 H) 4.19 (t, *J*=4.6 Hz, 2 H) 5.31 (br s, 1 H) 6.41 (d, *J*=7.6 Hz, 1 H) 6.58 (m, 1 H) 6.67 (br s, 1 H) 6.92 - 6.98 (m, 2 H) 7.06 (d, *J*=7.9 Hz, 1 H) 7.11 (d, *J*=1.9 Hz, 1 H) 7.22 (d, *J*=8.5 Hz, 1 H) 7.35 (d, *J*=8.2 Hz, 1 H) 7.99 (d, *J*=8.5 Hz, 1 H) 8.64 (s, 1 H) 12.29 (br s, 1 H)
LC/MS (method LC-C): *R*_t 3.20 min, MH⁺ 577

Enantiomer 5A:

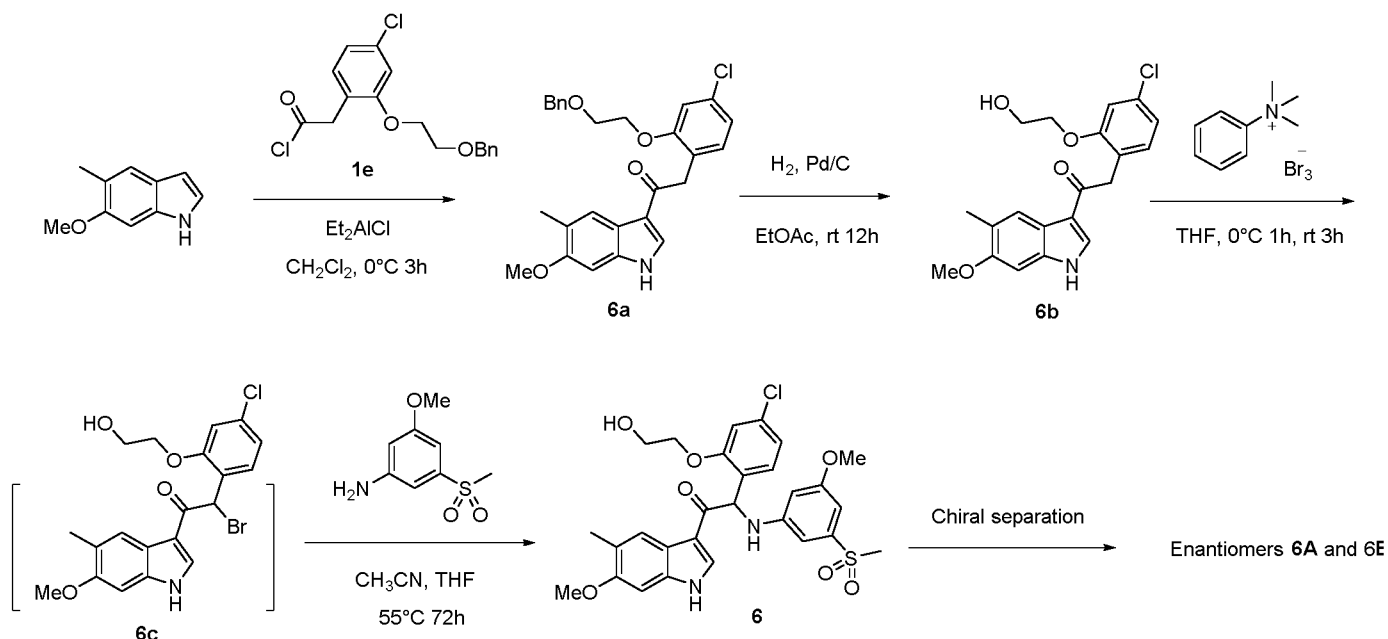
¹H NMR (500 MHz, DMSO-*d*₆) δ ppm 3.09 (s, 3 H) 3.72 (s, 3 H) 3.90 - 4.07 (m, 2 H) 4.19 (t, *J*=4.6 Hz, 2 H) 5.31 (br s, 1 H) 6.41 (d, *J*=7.9 Hz, 1 H) 6.58 (t, *J*=1.9 Hz, 1 H) 6.67 (s, 1 H) 6.93 - 6.96 (m, 2 H) 7.06 (d, *J*=7.6 Hz, 1 H) 7.11 (d, *J*=2.2 Hz, 1 H) 7.22 (d, *J*=8.5 Hz, 1 H) 7.35 (d, *J*=8.2 Hz, 1 H) 7.99 (d, *J*=8.5 Hz, 1 H) 8.64 (s, 1 H) 12.29 (br s, 1 H)
LC/MS (method LC-C): *R*_t 3.20 min, MH⁺ 577
[α]_D²⁰: -119.5° (c 0.37, DMF)
Chiral SFC (method SFC-A): *R*_t 2.30 min, MH⁺ 577, chiral purity 100%.
Melting point: 207°C

Enantiomer 5B:

¹H NMR (500 MHz, DMSO-*d*₆) δ ppm 3.09 (s, 3 H) 3.72 (s, 3 H) 3.90 - 4.07 (m, 2 H) 4.19 (t, *J*=4.7 Hz, 2 H) 5.31 (br s, 1 H) 6.41 (d, *J*=7.9 Hz, 1 H) 6.58 (t, *J*=1.7 Hz, 1 H) 6.67 (s, 1 H) 6.92 - 6.98 (m, 2 H) 7.07 (d, *J*=7.9 Hz, 1 H) 7.11 (d, *J*=1.9 Hz, 1 H) 7.23 (d, *J*=8.5 Hz, 1 H) 7.36 (d, *J*=8.5 Hz, 1 H) 7.99 (d, *J*=8.2 Hz, 1 H) 8.64 (s, 1 H) 12.29 (br s, 1 H)
LC/MS (method LC-C): *R*_t 3.20 min, MH⁺ 577
[α]_D²⁰: +126.1° (c 0.334, DMF)
Chiral SFC (method SFC-A): *R*_t 2.93 min, MH⁺ 577, chiral purity 99.2%.
Melting point: 206°C

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Example 6: synthesis of 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)-1-(6-methoxy-5-methyl-1*H*-indol-3-yl)ethanone (Compound **6**) and chiral separation into Enantiomers **6A** and **6B**.



Synthesis of intermediate **6a**:

- 5 Diethylaluminum chloride 1M in hexane (18.6 mL, 18.6 mmol) was added dropwise, at 0°C , to a solution of 6-methoxy-5-methyl-1H-indole [CAS 1071973-95-9] (2 g, 12.4 mmol) in CH_2Cl_2 (30 mL). After 30 min at 0°C , 2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)acetyl chloride **1e** (4.41 g, 13.0 mmol) in CH_2Cl_2 (30 mL) was added slowly at 0°C . The reaction was stirred at 0°C for
- 10 3 h. Ice-water was added and the reaction mixture was extracted with CH_2Cl_2 . The organic layer was dried over MgSO_4 , filtered and the solvent was evaporated under reduced pressure. The crude product was solidified by trituration with CH_3CN /diisopropyl ether. The solid was filtered off and dried to give 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(6-methoxy-5-methyl-1H-indol-3-yl)ethanone **6a** (2.65 g).
- 15

Synthesis of intermediate **6b**:

- A mixture of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(6-methoxy-5-methyl-1H-indol-3-yl)ethanone **6a** (1.7 g, 3.66 mmol) in EtOAc (70 mL) was
- 20 hydrogenated under an atmospheric pressure of H_2 for 12 h with Pd/C (10%) (164 mg, 0.154 mmol) as a catalyst. The mixture was filtered through a pad of celite® and washed with EtOAc. The filtrate was concentrated under reduced pressure to give 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-methoxy-5-methyl-1H-indol-3-yl)ethanone **6b** (910 mg).

Synthesis of Compound 6 and chiral separation of Enantiomers 6A and 6B:

Under a N₂ flow, at 0°C, a solution of phenyltrimethylammonium tribromide [CAS 4207-56-1] (1.09 g, 2.89 mmol) in THF (20 mL) was added dropwise to a solution of 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-methoxy-5-methyl-1*H*-indol-3-yl)ethanone **6b** (1.08 g, 2.89 mmol) in THF (30 mL). The mixture was stirred at 0°C for 1 h, the cooling bath was removed and stirring was continued at room temperature for 3 h. 3-Methoxy-5-(methylsulfonyl)aniline [CAS 62606-02-4] (1.74 g, 8.67 mmol) in CH₃CN (20 mL) was added dropwise and the resulting mixture was stirred at 55°C for 72 h. The mixture was diluted with CH₂Cl₂ and washed with HCl 1N (twice), dried over MgSO₄, filtered and the solvent was evaporated under reduced pressure. Purification was performed by flash chromatography on silica gel (15-40 µm, 80 g, CH₂Cl₂/CH₃OH 98.5/1.5). The pure fractions were combined and evaporated to dryness to give 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)-1-(6-methoxy-5-methyl-1*H*-indol-3-yl)ethanone (Compound **6**, 862 mg) as a racemic mixture.

The enantiomers of Compound **6** (1.3 g) were separated via chiral SFC (Stationary phase: Chiralcel® OD-H 5 µm 250 x 30 mm, Mobile phase: 55% CO₂, 45% EtOH (+ 0.3% *i*PrNH₂)). The first eluted enantiomer was solidified from petroleum ether/diisopropyl ether. The precipitate was filtered off and dried to give 441 mg of Enantiomer **6A**. The second eluted enantiomer was crystallized from petroleum ether /diisopropyl ether. The precipitate was filtered off and dried to give 461 mg of Enantiomer **6B**.

Compound 6:

¹H NMR (500 MHz, DMSO-*d*₆) 2.21 (s, 3 H) 3.08 (s, 3 H) 3.72 (s, 3 H) 3.79 (s, 3 H) 3.89 - 4.06 (m, 2 H) 4.19 (t, *J*=4.6 Hz, 2 H) 5.30 (br s, 1 H) 6.33 (d, *J*=7.9 Hz, 1 H) 6.57 (t, *J*=1.7 Hz, 1 H) 6.65 (br s, 1 H) 6.89 (s, 1 H) 6.92 - 6.96 (m, 2 H) 7.01 (d, *J*=7.9 Hz, 1 H) 7.10 (d, *J*=1.9 Hz, 1 H) 7.35 (d, *J*=8.2 Hz, 1 H) 7.90 (s, 1 H) 8.48 (s, 1 H) 11.84 (br s, 1 H)
LC/MS (method LC-C): R_t 2.99 min, MH⁺ 573

Enantiomer 6A:

¹H NMR (500 MHz, DMSO-*d*₆) δ ppm 2.22 (s, 3 H) 3.09 (s, 3 H) 3.72 (s, 3 H) 3.79 (s, 3 H) 3.90 - 4.07 (m, 2 H) 4.20 (t, *J*=4.4 Hz, 2 H) 5.30 (br s, 1 H) 6.34 (d, *J*=7.6 Hz, 1 H) 6.57 (s, 1 H) 6.65 (br s, 1 H) 6.90 (s, 1 H) 6.93 - 6.97 (m, 2 H)

7.01 (d, $J=7.9$ Hz, 1 H) 7.11 (d, $J=1.6$ Hz, 1 H) 7.36 (d, $J=8.5$ Hz, 1 H) 7.91 (s, 1 H) 8.49 (s, 1 H) 11.84 (br s, 1 H)

LC/MS (method LC-C): R_t 2.98 min, MH^+ 573

$[\alpha]_D^{20}$: +147.1° (c 0.2936, DMF)

- 5 Chiral SFC (method SFC-B): R_t 1.86 min, MH^+ 573, chiral purity 100%.

Enantiomer 6B:

1H NMR (500 MHz, DMSO- d_6) δ ppm 2.22 (s, 3 H) 3.09 (s, 3 H) 3.72 (s, 3 H)

3.79 (s, 3 H) 3.90 - 4.07 (m, 2 H) 4.20 (br t, $J=4.4$ Hz, 2 H) 5.30 (br s, 1 H) 6.34

- 10 (d, $J=7.6$ Hz, 1 H) 6.57 (s, 1 H) 6.65 (br s, 1 H) 6.90 (s, 1 H) 6.92 - 6.97 (m, 2 H)

7.02 (d, $J=7.6$ Hz, 1 H) 7.11 (d, $J=1.6$ Hz, 1 H) 7.36 (d, $J=8.5$ Hz, 1 H) 7.91

(s, 1 H) 8.49 (s, 1 H) 11.84 (br s, 1 H)

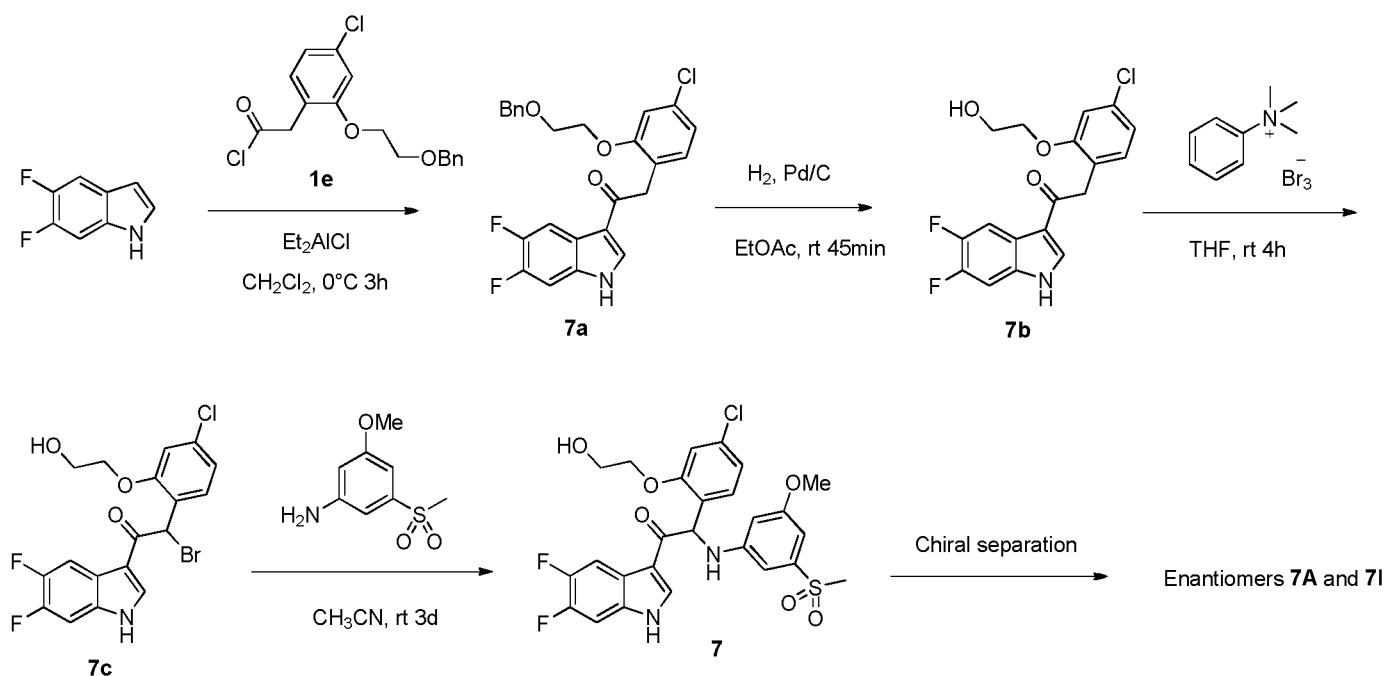
LC/MS (method LC-C): R_t 2.98 min, MH^+ 573

$[\alpha]_D^{20}$: -152.4° (c 0.2927, DMF)

- 15 Chiral SFC (method SFC-B): R_t 3.43 min, MH^+ 573, chiral purity 100%.

Example 7: synthesis of 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(5,6-difluoro-1*H*-indol-3-yl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)ethanone (Compound **7**) and chiral separation into Enantiomers **7A** and **7B**.

20



Synthesis of intermediate 7a:

Diethylaluminum chloride 1M in hexane (12.5 mL, 12.5 mmol) was added dropwise, at 0°C, to a solution of 5,6-difluoro-1*H*-indole [CAS 169674-01-5] (1.27 g, 8.30 mmol) in CH₂Cl₂ (50 mL). After 30 min at 0°C, a solution of 2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)acetyl chloride **1e** (4.23 g, 12.5 mmol, synthesis: see Example 1) in CH₂Cl₂ (20 mL) was slowly added. The reaction mixture was stirred at 0°C for 3 h. 1M Rochelle salt solution was added and the mixture was stirred vigorously for 30 min. H₂O was added and the phases were separated. The aqueous phase was extracted twice with EtOAc. The organic phases were combined, washed with brine, dried over MgSO₄, filtered and concentrated under reduced pressure. The residue was triturated with Et₂O. The solids were filtered off and dried under vacuum to give 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(5,6-difluoro-1*H*-indol-3-yl)ethanone **7a** (1.37 g).

Synthesis of intermediate 7b:

A mixture of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(5,6-difluoro-1*H*-indol-3-yl)ethanone **7a** (1.43 g, 3.14 mmol) and 10% palladium on carbon (0.07 g) in EtOAc (70 mL) was stirred at room temperature for 45 min under H₂ atmosphere. The reaction mixture was filtered through celite®. The filtrate was concentrated under reduced pressure. The residue was purified by flash chromatography on silica gel using a gradient of EtOAc (0% to 15%) in CH₂Cl₂ to provide 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(5,6-difluoro-1*H*-indol-3-yl)ethanone **7b** (0.88 g).

Synthesis of intermediate 7c:

A solution of phenyltrimethylammonium tribromide [CAS 4207-56-1] (1.68 g, 4.47 mmol) in THF (20 mL) was added dropwise to a solution of 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(5,6-difluoro-1*H*-indol-3-yl)ethanone **7b** (1.49 g, 4.07 mmol) in THF (45 mL). The mixture was stirred at room temperature for 4 h. The precipitate was filtered off and washed with EtOAc. The filtrate was concentrated under reduced pressure to give 2-bromo-2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(5,6-difluoro-1*H*-indol-3-yl)ethanone **7c** (1.81 g) which was used in the next step without further purification.

Synthesis of Compound 7 and chiral separation into Enantiomers 7A and 7B:

A mixture of 2-bromo-2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(5,6-difluoro-1*H*-indol-3-yl)ethanone **7c** (1.81 g, 4.07 mmol) and 3-methoxy-5-(methylsulfonyl)aniline [CAS 62606-02-4] (2.46 g, 12.2 mmol) in CH₃CN (40 mL) was stirred at room temperature for 3 d. The reaction mixture was concentrated under reduced pressure. The residue was partitioned between EtOAc and 1N HCl. The phases were separated. The aqueous phase was extracted with EtOAc. The organic phases were combined, washed with brine, dried over MgSO₄, filtered and concentrated under reduced pressure. The residue was purified by flash chromatography on silica gel using a gradient of EtOAc (50% to 100%) in heptane. The fractions containing the desired product were combined and concentrated under reduced pressure. The residue was triturated with CH₂Cl₂. The solids were filtered off and dried under vacuum to give 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(5,6-difluoro-1*H*-indol-3-yl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)ethanone (Compound **7**, 0.97 g) as a racemic mixture.

The chiral separation of the enantiomers of Compound **7** (914 mg) was performed via Normal Phase Chiral separation (Stationary phase: AS 20 µm, Mobile phase: 100% methanol). The product fractions were combined and evaporated to provide Enantiomer **7A** (351 mg) as the first eluted product and Enantiomer **7B** (337 mg) as the second eluted product. Enantiomer **7A** was stirred up in a 1/1 mixture of MeOH and water (10 mL). The solids were filtered off, washed with a small amount of MeOH/water 1/1 and dried under vacuum at 50°C to provide Enantiomer **7A** (323 mg) as a white powder. Enantiomer **7B** was further purified via preparative chiral SFC (Stationary phase: Chiralpak® Diacel AS 20 x 250 mm, Mobile phase: CO₂, EtOH (+ 0.4% *i*PrNH₂)). The fractions containing desired product were combined and evaporated under reduced pressure. The residue was stirred up in a 1/1 mixture of MeOH and water (10 mL) for 30 min. The solids were filtered off, washed with a small amount of MeOH/water 1/2 and dried under vacuum at 50°C to provide Enantiomer **7B** (209 mg) as a white powder.

Compound 7:

¹H NMR (300 MHz, DMSO-*d*₆) δ ppm 3.09 (s, 3 H) 3.72 (s, 3 H) 3.86 - 4.07 (m, 2 H) 4.19 (m, 2 H) 5.29 (t, *J*=5.3 Hz, 1 H) 6.37 (d, *J*=7.9 Hz, 1 H) 6.58 (s, 1 H) 6.65 (s, 1 H) 6.91 - 7.01 (m, 2 H) 7.05 - 7.16 (m, 2 H) 7.36 (d, *J*=8.3 Hz,

1 H) 7.50 (dd, $J=10.7$, 7.0 Hz, 1 H) 8.01 (dd, $J=11.3$, 8.3 Hz, 1 H) 8.72 (s, 1 H)
12.31 (br. s., 1 H)

LC/MS (method LC-E): R_t 1.21 min, MH^+ 565

5 **Enantiomer 7A:**

1H NMR (360 MHz, DMSO- d_6) δ ppm 3.09 (s, 3 H) 3.72 (s, 3 H) 3.87 - 4.05 (m, 2 H) 4.19 (br t, $J=4.6$ Hz, 2 H) 5.31 (br t, $J=5.4$ Hz, 1 H) 6.37 (d, $J=7.8$ Hz, 1 H) 6.58 (br s, 1 H) 6.65 (br s, 1 H) 6.92 - 7.01 (m, 2 H) 7.06 - 7.16 (m, 2 H) 7.36 (d, $J=8.3$ Hz, 1 H) 7.50 (dd, $J=10.7$, 6.9 Hz, 1 H) 8.01 (dd, $J=11.1$, 8.1 Hz, 1 H) 8.72 (s, 1 H) 12.31 (br s, 1 H)

LC/MS (method LC-A): R_t 1.15 min, MH^+ 565

$[\alpha]_D^{20}$: +120.2° (c 0.499, DMF)

Chiral SFC (method SFC-C): R_t 3.47 min, MH^+ 565, chiral purity 100%.

15 **Enantiomer 7B:**

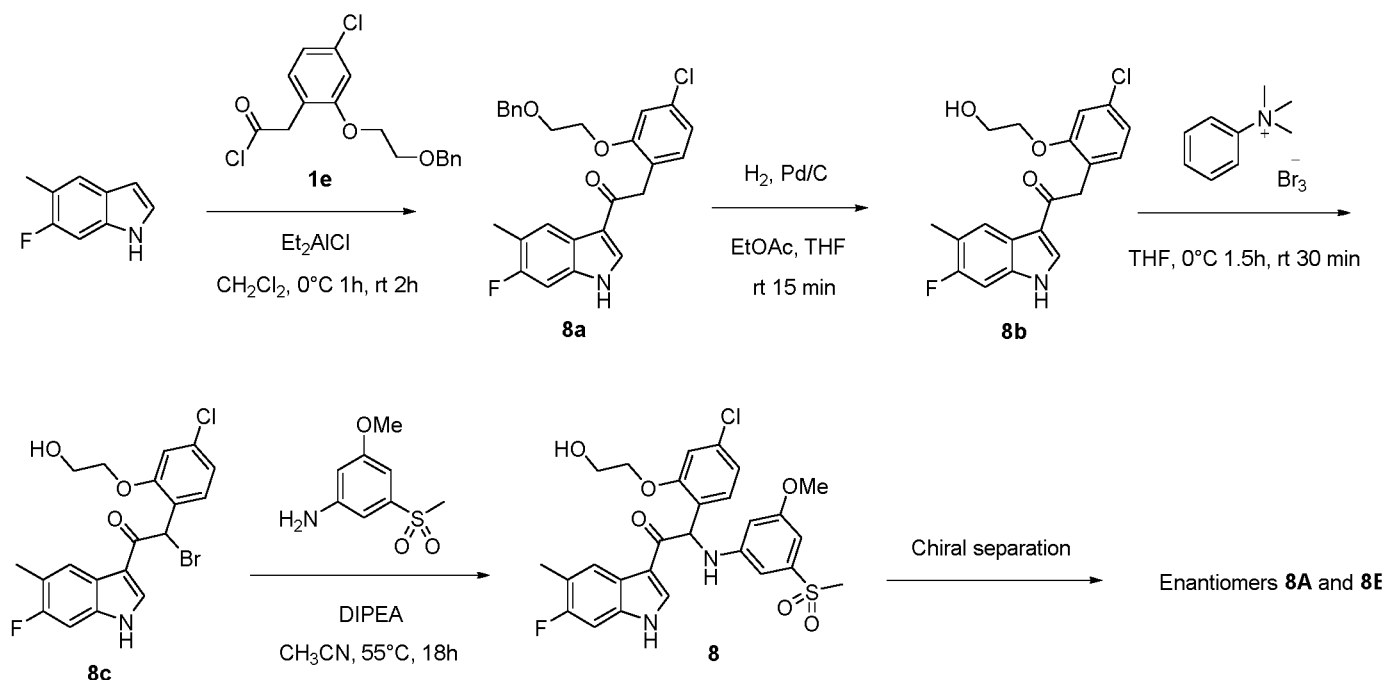
1H NMR (360 MHz, DMSO- d_6) δ ppm 3.09 (s, 3 H) 3.72 (s, 3 H) 3.86 - 4.06 (m, 2 H) 4.19 (br t, $J=4.5$ Hz, 2 H) 5.30 (br t, $J=5.4$ Hz, 1 H) 6.37 (d, $J=7.8$ Hz, 1 H) 6.58 (s, 1 H) 6.65 (s, 1 H) 6.92 - 7.00 (m, 2 H) 7.06 - 7.14 (m, 2 H) 7.36 (d, $J=8.3$ Hz, 1 H) 7.50 (dd, $J=10.7$, 6.8 Hz, 1 H) 8.01 (dd, $J=11.1$, 8.0 Hz, 1 H) 8.72 (s, 1 H) 12.30 (s, 1 H)

LC/MS (method LC-A): R_t 1.10 min, MH^+ 565

$[\alpha]_D^{20}$: -125.0° (c 0.414, DMF)

Chiral SFC (method SFC-D): R_t 1.60 min, MH^+ 565, chiral purity 100%.

- 25 Example 8: synthesis of 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-fluoro-5-methyl-1*H*-indol-3-yl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)ethanone (Compound **8**) and chiral separation into Enantiomers **8A** and **8B**.



Synthesis of intermediate **8a**:

- 5 Diethylaluminum chloride 1M in hexane (17.0 mL, 17.0 mmol) was added dropwise, at 0°C and under N_2 -atmosphere, to a solution of 6-fluoro-5-methyl-1H-indole [CAS 162100-95-0] (1.69 g, 11.3 mmol) in CH_2Cl_2 (150 mL). After 15 min at 0°C , a solution of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)acetyl chloride **1e** (5.37 g, 15.8 mmol, synthesis: see Example 1) in CH_2Cl_2 (100 mL)
- 10 was slowly added. The reaction mixture was stirred at 0°C for 1 h and at room temperature for 2 h. The reaction mixture was poured out into an ice/Rochelle salt solution and the mixture was vigorously stirred. The layers were separated. The organic layer was dried over MgSO_4 and filtered over a short pad of dicalite®. The filter cake was rinsed a few times with THF and the combined
- 15 filtrates were concentrated under reduced pressure. The solid residue was suspended in CH_3CN (20 mL), filtered off, washed with a small amount of CH_3CN , and dried under vacuum at 50°C to give 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(6-fluoro-5-methyl-1H-indol-3-yl)ethanone **8a** (2.39 g) as a white solid.

20

Synthesis of intermediate **8b**:

A mixture of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(6-fluoro-5-methyl-1H-indol-3-yl)ethanone **8a** (2.39 g, 4.71 mmol) and 10% palladium on carbon (0.5 g) in EtOAc (135 mL) and THF (15 mL) was stirred at room temperature for

15 min under H₂ atmosphere. The reaction mixture was filtered through dicalite® and the filter cake was washed with EtOAc and THF. The combined filtrates were concentrated under reduced pressure. The residue was stirred up in DIPE/THF (2/1), filtered off, washed with DIPE (3x) and dried under vacuum at 50°C to provide 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-fluoro-5-methyl-1*H*-indol-3-yl)ethanone **8b** (0.90 g).

Synthesis of intermediate 8c:

Phenyltrimethylammonium tribromide [CAS 4207-56-1] (982 mg, 2.61 mmol) was added to a cooled (0°C) solution of 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-fluoro-5-methyl-1*H*-indol-3-yl)ethanone **8b** (900 mg, 2.49 mmol) in THF (60 mL), under N₂-atmosphere. The reaction mixture was stirred at 0°C for 90 min and at room temperature for 30 min. The precipitate was filtered off and washed with THF (2x). The combined filtrates were concentrated under reduced pressure to give 2-bromo-2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-fluoro-5-methyl-1*H*-indol-3-yl)ethanone **8c** (1.1 g) which was used in the next step without further purification.

Synthesis of Compound 8 and chiral separation into Enantiomers 8A and 8B:

A mixture of 2-bromo-2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-fluoro-5-methyl-1*H*-indol-3-yl)ethanone **8c** (1.10 g, 2.49 mmol), 3-methoxy-5-(methylsulfonyl)aniline [CAS 62606-02-4] (1.00 g, 4.97 mmol) and diisopropylethylamine (857 µL, 4.97 mmol) in CH₃CN (60 mL) was stirred at 55°C for 18 h. The reaction mixture was cooled to room temperature and poured out into stirring water (250 mL). The product was extracted with a mixture of Et₂O/2-MeTHF 9/1, and with Et₂O. The combined organic layers were washed with brine, dried over MgSO₄, filtered, and evaporated under reduced pressure. The residue was purified by flash chromatography (Stationary phase: Grace Reveleris® silica 40 g, Mobile phase: heptane/EtOAc/EtOH gradient 100/0/0 to 40/45/15). The desired fractions were combined and evaporated under reduced pressure. The residue was purified via preparative HPLC (Stationary phase: RP XBridge® Prep C18 OBD – 10 µm, 50 x 150 mm, Mobile phase: 0.25% NH₄HCO₃ solution in water, CH₃CN). The desired fractions were combined and evaporated under reduced pressure and the solid residue was dried at 50°C under vacuum to give racemic 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(6-fluoro-5-methyl-1*H*-indol-3-yl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)ethanone (Compound **8**, 600 mg).

The chiral separation of the enantiomers of Compound **8** (570 mg) was performed via Normal Phase Chiral separation (Stationary phase: AS 20 μm , Mobile phase: 100% methanol). The product fractions were combined and evaporated to provide Enantiomer **8A** as the first eluted product and

- 5 Enantiomer **8B** as the second eluted product. Both enantiomers were stirred up in water/MeOH 4/1 (5 mL), filtered off, washed with water/MeOH 4/1 and dried under vacuum at 50°C to provide Enantiomer **8A** (178 mg) and Enantiomer **8B** (189 mg).

10 **Compound 8:**

- ^1H NMR (400 MHz, DMSO- d_6) δ ppm 2.30 (d, $J=1.3$ Hz, 3 H) 3.08 (s, 3 H) 3.72 (s, 3 H) 3.88 - 4.05 (m, 2 H) 4.19 (t, $J=4.5$ Hz, 2 H) 5.27 (br s, 1 H) 6.35 (d, $J=7.7$ Hz, 1 H) 6.57 (t, $J=1.9$ Hz, 1 H) 6.64 (t, $J=1.5$ Hz, 1 H) 6.92 - 6.97 (m, 2 H) 7.01 (d, $J=7.7$ Hz, 1 H) 7.10 (d, $J=2.0$ Hz, 1 H) 7.19 (d, $J=10.1$ Hz, 1 H) 7.36 (d, $J=8.4$ Hz, 1 H) 8.02 (d, $J=7.9$ Hz, 1 H) 8.61 (s, 1 H) 12.02 (br s, 1 H)
15 LC/MS (method LC-B): R_t 2.01 min, MH^+ 561

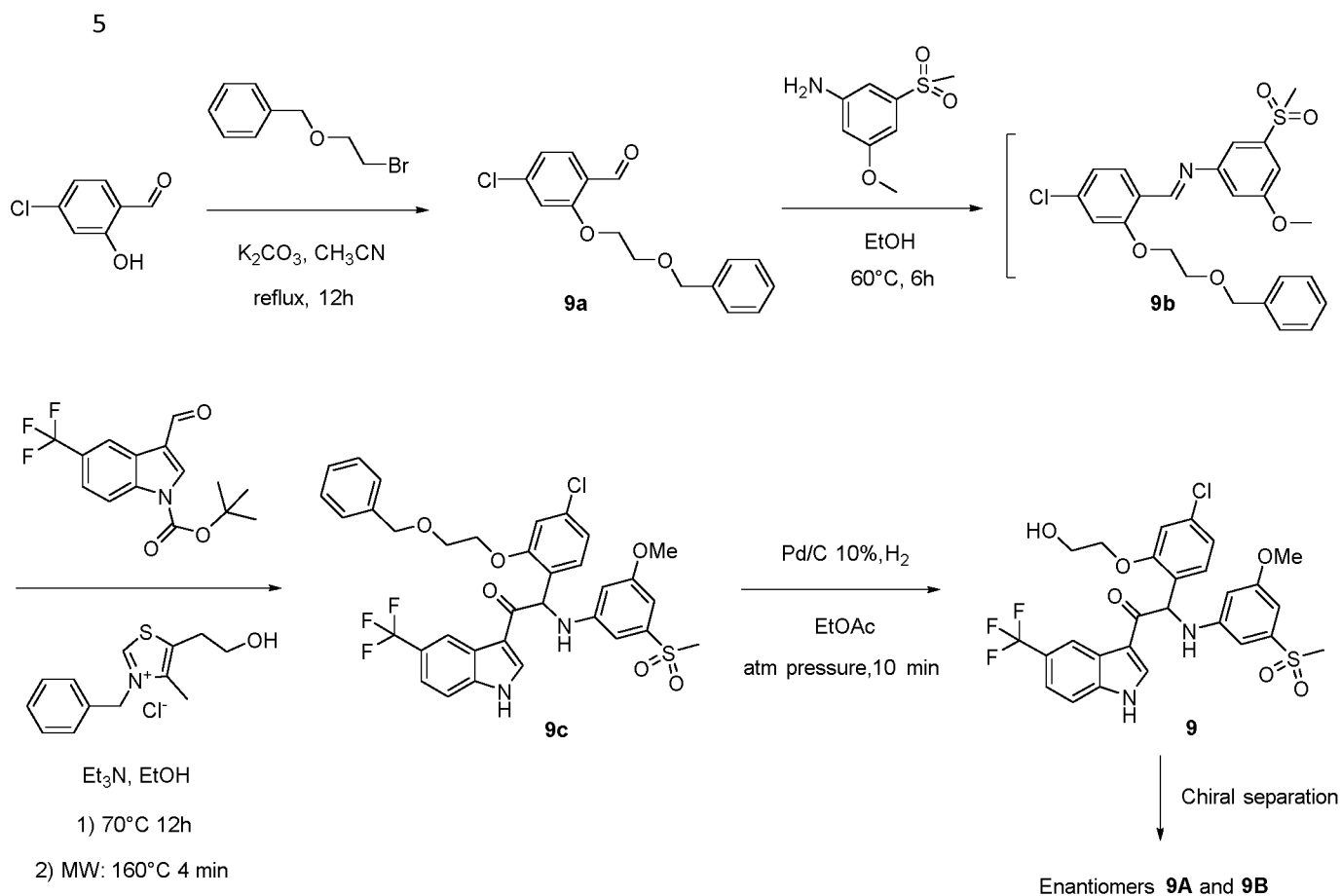
Enantiomer 8A:

- ^1H NMR (400 MHz, DMSO- d_6) δ ppm 2.30 (d, $J=1.3$ Hz, 3 H) 3.08 (s, 3 H) 3.72 (s, 3 H) 3.87 - 4.06 (m, 2 H) 4.19 (t, $J=4.5$ Hz, 2 H) 5.26 (br t, $J=5.1$ Hz, 1 H) 6.35 (d, $J=7.7$ Hz, 1 H) 6.57 (t, $J=1.8$ Hz, 1 H) 6.65 (t, $J=1.9$ Hz, 1 H) 6.92 - 6.97 (m, 2 H) 7.01 (d, $J=7.7$ Hz, 1 H) 7.10 (d, $J=1.8$ Hz, 1 H) 7.19 (d, $J=10.1$ Hz, 1 H) 7.36 (d, $J=8.1$ Hz, 1 H) 8.03 (d, $J=7.9$ Hz, 1 H) 8.61 (s, 1 H) 12.03 (br s, 1 H)
20 LC/MS (method LC-A): R_t 1.10 min, MH^+ 561
 $[\alpha]_D^{20}$: +172.4° (c 0.485, DMF)
Chiral SFC (method SFC-C): R_t 3.59 min, MH^+ 561, chiral purity 100%.

Enantiomer 8B:

- ^1H NMR (400 MHz, DMSO- d_6) δ ppm 2.30 (d, $J=1.3$ Hz, 3 H) 3.08 (s, 3 H) 3.72 (s, 3 H) 3.88 - 4.05 (m, 2 H) 4.19 (t, $J=4.6$ Hz, 2 H) 5.27 (br s, 1 H) 6.35 (d, $J=7.7$ Hz, 1 H) 6.57 (t, $J=1.8$ Hz, 1 H) 6.65 (t, $J=1.8$ Hz, 1 H) 6.91 - 6.97 (m, 2 H) 7.01 (d, $J=7.9$ Hz, 1 H) 7.10 (d, $J=2.0$ Hz, 1 H) 7.19 (d, $J=10.3$ Hz, 1 H) 7.36 (d, $J=8.4$ Hz, 1 H) 8.03 (d, $J=7.7$ Hz, 1 H) 8.61 (s, 1 H) 11.94 (br s, 1 H)
30 LC/MS (method LC-A): R_t 1.10 min, MH^+ 561
 $[\alpha]_D^{20}$: -170.6° (c 0.425, DMF)
Chiral SFC (method SFC-C): R_t 4.06 min, MH^+ 561, chiral purity 98.7%.

Example 9: synthesis of 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)-1-(5-(trifluoromethyl)-1*H*-indol-3-yl)ethanone (Compound **9**) and chiral separation into Enantiomers **9A** and **9B**.



Synthesis of the intermediate **9a**:

- 10 A mixture of 4-chloro-2-hydroxy-benzaldehyde [CAS 2420-26-0] (7.72 g, 49.31 mmol), benzyl 2-bromoethyl ether [CAS 1462-37-9] (7.8 mL, 49.31 mmol) and potassium carbonate (8.2 g, 59.17 mmol) in CH_3CN (200 mL) was heated under reflux for 12 h. The mixture was evaporated under reduced pressure. The residue was taken up with $EtOAc$, washed with water (twice), dried over
- 15 $MgSO_4$, filtered and the solvent was evaporated to afford 2-(2-(benzyloxy)ethoxy)-4-chlorobenzaldehyde **9a** (14.2 g).

Synthesis of the intermediate **9b**:

- A mixture of 2-(2-(benzyloxy)ethoxy)-4-chlorobenzaldehyde **9a** (2.1 g, 7.22 mmol), 3-methoxy-5-(methylsulfonyl)aniline [CAS 62606-02-4] (1.45 g,
- 20

7.22 mmol) in EtOH (18 mL) was stirred at 60°C for 6 h. The resulting solution containing imine **9b** was used as such in the next step.

Synthesis of the intermediate **9c**:

- 5 To a solution of 3-benzyl-5-(2-hydroxyethyl)-4-methylthiazol-3-ium chloride [CAS 4568-71-2] (1.95 g, 7.22 mmol) in EtOH (10 mL) was added triethylamine (1 mL, 7.22 mmol) and the resulting mixture was stirred at 70°C for 10 min. This solution was added to a stirred mixture of imine **9b** (3.42 g, 7.22 mmol, solution in EtOH, see above: synthesis of intermediate **9b**) and *tert*-butyl
- 10 3-formyl-5-(trifluoromethyl)-1*H*-indole-1-carboxylate [CAS 1493799-60-2] (2.7 g, 8.67 mmol) at room temperature. The mixture was stirred at 70°C for 12 h. The reaction mixture was cooled down to room temperature and transferred into a sealed tube that was subsequently heated at 160°C using one single mode microwave (*Biotage Initiator EXP 60*) with a power output ranging from 0 to
- 15 400 W for 4 min (fixed hold time). The mixture was evaporated under reduced pressure. Purification was performed by flash chromatography on silica gel (15-40 µm, 120 g, eluent: CH₂Cl₂/CH₃OH 99.5/0.5). The pure fractions were combined and evaporated under reduced pressure. The residue (3.48 g) was taken up with CH₃OH and stirred at room temperature for 1 h. The precipitate
- 20 was filtered off and dried to yield 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)-1-(5-(trifluoromethyl)-1*H*-indol-3-yl)ethanone **9c** (1.23 g).

Synthesis of Compound **9** and chiral separation into Enantiomers **9A** and **9B**:

- 25 A mixture of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)-1-(5-(trifluoromethyl)-1*H*-indol-3-yl)ethanone **9d** (1.10 g, 1.60 mmol) in EtOAc (20 mL) was hydrogenated for 10 min under atmospheric pressure of H₂ with 10% Pd/C (340 mg, 0.32 mmol) as the catalyst.
- 30 The reaction was diluted with EtOAc and filtered through a pad of celite®. The filtrate was evaporated under reduced pressure to afford 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)-1-(5-(trifluoromethyl)-1*H*-indol-3-yl)ethanone (compound **9**, 910 mg) as a racemic mixture.
- 35 The enantiomers of Compound **9** (1.15 g) were separated via chiral SFC (Stationary phase: Chiralpak® IC 5 µm 250 x 30 mm, Mobile phase: 75% CO₂, 25% EtOH + 0.3% iPrNH₂) yielding 544 mg of the first eluted enantiomer and 464 mg of the second eluted enantiomer. The first eluted enantiomer was

crystallized from CH₃OH and water. The precipitate was filtered off and dried to give 362 mg of Enantiomer **9A**. The second eluted enantiomer was crystallized from CH₃OH and water. The precipitate was filtered off and dried to give 348 mg of Enantiomer **9B**.

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Compound 9:

¹H NMR (500 MHz, DMSO-*d*₆) δ ppm 3.09 (s, 3 H) 3.72 (s, 3 H) 3.87 - 4.09 (m, 2 H) 4.20 (t, *J*=4.6 Hz, 2 H) 5.32 (t, *J*=5.5 Hz, 1 H) 6.42 (d, *J*=7.9 Hz, 1 H) 6.59 (t, *J*=1.9 Hz, 1 H) 6.66 (s, 1 H) 6.94 - 6.98 (m, 2 H) 7.10 (d, *J*=7.9 Hz, 1 H) 7.12 (d, *J*=1.9 Hz, 1 H) 7.38 (d, *J*=8.2 Hz, 1 H) 7.54 (dd, *J*=8.7, 1.7 Hz, 1 H) 7.67 (d, *J*=8.5 Hz, 1 H) 8.50 (s, 1 H) 8.83 (s, 1 H) 12.53 (br s, 1 H)
LC/MS (method LC-C): R_t 3.12 min, MH⁺ 597
Melting point: 228°C

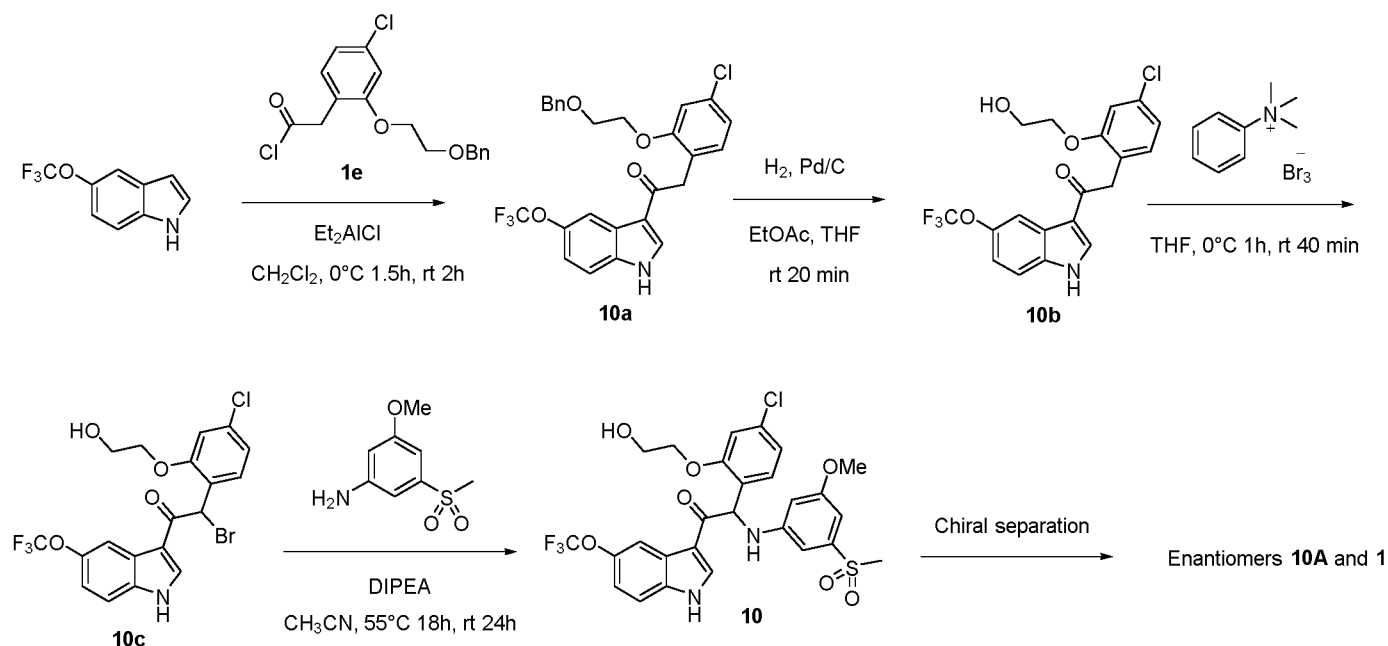
15 **Enantiomer 9A:**

¹H NMR (500 MHz, DMSO-*d*₆) δ ppm 3.09 (s, 3 H) 3.73 (s, 3 H) 3.87 - 4.07 (m, 2 H) 4.20 (t, *J*=4.4 Hz, 2 H) 5.32 (br t, *J*=4.4 Hz, 1 H) 6.42 (d, *J*=7.9 Hz, 1 H) 6.59 (s, 1 H) 6.66 (br s, 1 H) 6.94 - 6.99 (m, 2 H) 7.10 (d, *J*=7.9 Hz, 1 H) 7.12 (d, *J*=1.3 Hz, 1 H) 7.38 (d, *J*=8.2 Hz, 1 H) 7.54 (d, *J*=8.5 Hz, 1 H) 7.67 (d, *J*=8.5 Hz, 1 H) 8.50 (s, 1 H) 8.83 (s, 1 H) 12.52 (br s, 1 H)
LC/MS (method LC-C): R_t 3.12 min, MH⁺ 597
[α]_D²⁰: -154.3° (c 0.245, DMF)
Chiral SFC (method SFC-E): R_t 1.75 min, MH⁺ 597, chiral purity 100%.

25 **Enantiomer 9B:**

¹H NMR (500 MHz, DMSO-*d*₆) δ ppm 3.09 (s, 3 H) 3.72 (s, 3 H) 3.88 - 4.06 (m, 2 H) 4.20 (t, *J*=4.4 Hz, 2 H) 5.32 (br t, *J*=4.7 Hz, 1 H) 6.42 (d, *J*=7.9 Hz, 1 H) 6.59 (s, 1 H) 6.66 (br s, 1 H) 6.94 - 6.98 (m, 2 H) 7.10 (d, *J*=7.9 Hz, 1 H) 7.12 (d, *J*=1.6 Hz, 1 H) 7.38 (d, *J*=8.2 Hz, 1 H) 7.54 (d, *J*=8.5 Hz, 1 H) 7.67 (d, *J*=8.5 Hz, 1 H) 8.50 (s, 1 H) 8.83 (s, 1 H) 12.50 (br s, 1 H)
LC/MS (method LC-C): R_t 3.12 min, MH⁺ 597
[α]_D²⁰: +142.6° (c 0.284, DMF)
Chiral SFC (method SFC-E): R_t 2.15 min, MH⁺ 597, chiral purity 100%.

35 Example 10: synthesis of 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)-1-(5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone (Compound **10**) and chiral separation into Enantiomers **10A** and **10B**.



Synthesis of intermediate **10a**:

- 5 Diethylaluminum chloride 1M in hexane (18.2 mL, 18.2 mmol) was added dropwise, at 0°C and under N_2 -atmosphere, to a solution of 5-(trifluoromethoxy)-1H-indole [CAS 262593-63-5] (2.44 g, 12.1 mmol) in CH_2Cl_2 (150 mL). After stirring for 15 min at 0°C , a solution of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)acetyl chloride **1e** (6.17 g, 18.2 mmol,
- 10 synthesis: see Example 1) in CH_2Cl_2 (100 mL) was slowly added. The reaction mixture was stirred at 0°C for 90 min and at room temperature for 2 h. The reaction mixture was cooled to 0°C and a solution of Rochelle salt [CAS 6100-16-9] (6.85 g, 24.3 mmol) in water (7 mL) was added dropwise. The mixture was vigorously stirred for 30 min at 0°C . The ice-bath was removed and THF
- 15 (200 mL) was added. After stirring for 30 min at room temperature, Na_2SO_4 (25 g) was added. The mixture was stirred for 90 min and filtered over dicalite®. The filter cake was washed with THF (4x 150 mL) and the combined filtrates were evaporated under reduced pressure and co-evaporated to dryness with a mixture CH_3CN and toluene. The solid residue was stirred up in a mixture of
- 20 toluene (5 mL) and CH_3CN (2.5 mL), filtered off, washed with a small amount of toluene/ CH_3CN (2/1), and dried under vacuum at 50°C to give 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(5-(trifluoromethoxy)-1H-indol-3-yl)ethanone **10a** (1.89 g). The filtrate was evaporated under reduced pressure. The residue (6.8 g) was purified by flash chromatography (Biotage® SNAP
- 25 Ultra silica 100 g, eluent: heptane/ EtOAc / EtOH gradient 100/0/0 to 40/45/15).

The desired fractions were combined and evaporated under reduced pressure, and co-evaporated with EtOAc. The product was stirred up in a mixture of DIPE (15 mL) and EtOAc (1 mL), filtered off, washed with DIPE (2x), and dried at 50°C under vacuum to provide a second batch of **10a** (1.62 g).

5

Synthesis of intermediate 10b:

A mixture of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone **10a** (1.62 g, 3.22 mmol) and 10% palladium on carbon (0.5 g) in EtOAc (75 mL) and THF (10 mL) was stirred at room temperature for 20 min under H₂ atmosphere. The reaction mixture was filtered through dicalite® and the filter cake was washed with THF. The combined filtrates were concentrated under reduced pressure. The solid residue was combined with another fraction (total amount: 3 g), stirred up in CH₂Cl₂ (8 mL), filtered off, washed with CH₂Cl₂ (5x 1 mL) and dried under vacuum at 45°C to provide 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone **10b** (1.64 g).

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Synthesis of intermediate 10c:

Phenyltrimethylammonium tribromide [CAS 4207-56-1] (1.56 g, 4.16 mmol) was added portionwise to a cooled (0°C) solution of 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone **10b** (1.64 g, 3.96 mmol) in THF (75 mL), under N₂-atmosphere. The reaction mixture was stirred at 0°C for 1 h and at room temperature for 40 min. The precipitate was filtered off and washed with THF (2x). The combined filtrates were concentrated under reduced pressure to give 2-bromo-2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone **10c** (1.92 g) which was used in the next step without further purification.

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Synthesis of Compound 10 and chiral separation into Enantiomers 10A and 10B:

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A mixture of 2-bromo-2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone **10c** (1.95 g, 3.96 mmol), 3-methoxy-5-(methylsulfonyl)aniline [CAS 62606-02-4] (1.60 g, 7.92 mmol) and diisopropylethylamine (1.37 mL, 7.92 mmol) in CH₃CN (75 mL) was stirred at 55°C for 18 h and at room temperature for 24 h. The reaction mixture was cooled to room temperature and poured out into stirring water (350 mL). The product was extracted with Et₂O (2x). The combined organic layers were washed with brine, dried over MgSO₄, filtered, and evaporated under reduced

- pressure. The residue was purified by flash chromatography (Stationary phase: Grace Reveleris® silica 80 g, Mobile phase: heptane/EtOAc/EtOH gradient 100/0/0 to 40/45/15). The desired fractions were combined, evaporated under reduced pressure and co-evaporated with CH₃CN. The residue was purified via preparative HPLC (Stationary phase: RP XBridge® Prep C18 OBD – 10 µm, 50 x 150 mm, Mobile phase: 0.25% NH₄HCO₃ solution in water, CH₃CN). The desired fractions were combined, evaporated under reduced pressure, co-evaporated with MeOH and was dried at 50°C under vacuum to give racemic 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)-1-(5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone (Compound **10**, 700 mg). A small sample of Compound **10** (50 mg) was solidified by slow evaporation from a solution in MeOH/water using a Rotavapor. The solids were filtered off, washed with water (3x) and dried at 45°C under vacuum to provide an analytical sample of Compound **10** (46 mg).
- The chiral separation of the Enantiomers of Compound **10** (650 mg) was performed via Normal Phase Chiral separation (Stationary phase: Whelk-O1 (R,R), Mobile phase: 20% ethanol, 80% heptane). The product fractions were combined and evaporated to provide Enantiomer **10A** as the first eluted product and Enantiomer **10B** as the second eluted product. Both enantiomers were stirred up in a mixture of water (4 mL) and MeOH (1.25 mL), filtered off, washed with water/MeOH 4/1 (4x) and dried under vacuum at 45°C to provide Enantiomer **10A** (115 mg) and Enantiomer **10B** (140 mg).

Compound 10:

- ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 3.08 (s, 3 H) 3.72 (s, 3 H) 3.87 - 4.05 (m, 2 H) 4.20 (t, *J*=4.6 Hz, 2 H) 5.27 (t, *J*=5.6 Hz, 1 H) 6.38 (d, *J*=7.9 Hz, 1 H) 6.58 (t, *J*=1.8 Hz, 1 H) 6.65 (t, *J*=2.4 Hz, 1 H) 6.92 - 6.98 (m, 2 H) 7.07 (d, *J*=7.7 Hz, 1 H) 7.12 (d, *J*=2.0 Hz, 1 H) 7.21 (dd, *J*=8.8, 1.8 Hz, 1 H) 7.38 (d, *J*=8.1 Hz, 1 H) 7.56 (d, *J*=8.8 Hz, 1 H) 8.07 (d, *J*=1.1 Hz, 1 H) 8.77 (s, 1 H) 12.37 (s, 1 H)
- LC/MS (method LC-A): R_t 1.15 min, MH⁺ 613

Enantiomer 10A:

- ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 3.09 (s, 3 H) 3.72 (s, 3 H) 3.87 - 4.07 (m, 2 H) 4.20 (br t, *J*=4.4 Hz, 2 H) 5.29 (br t, *J*=5.4 Hz, 1 H) 6.39 (d, *J*=7.7 Hz, 1 H) 6.59 (t, *J*=1.9 Hz, 1 H) 6.65 (t, *J*=2.2 Hz, 1 H) 6.91 - 7.00 (m, 2 H) 7.09 (d, *J*=7.9 Hz, 1 H) 7.12 (d, *J*=1.8 Hz, 1 H) 7.21 (dd, *J*=8.7, 1.7 Hz, 1 H) 7.38

(d, $J=8.4$ Hz, 1 H) 7.57 (d, $J=8.8$ Hz, 1 H) 8.08 (br s, 1 H) 8.77 (s, 1 H) 12.38 (br s, 1 H)

LC/MS (method LC-A): R_t 1.15 min, MH^+ 613

$[\alpha]_D^{20}$: -139.3° (c 0.425, DMF)

- 5 Chiral SFC (method SFC-C): R_t 3.27 min, MH^+ 613, chiral purity 100%.

Enantiomer 10B:

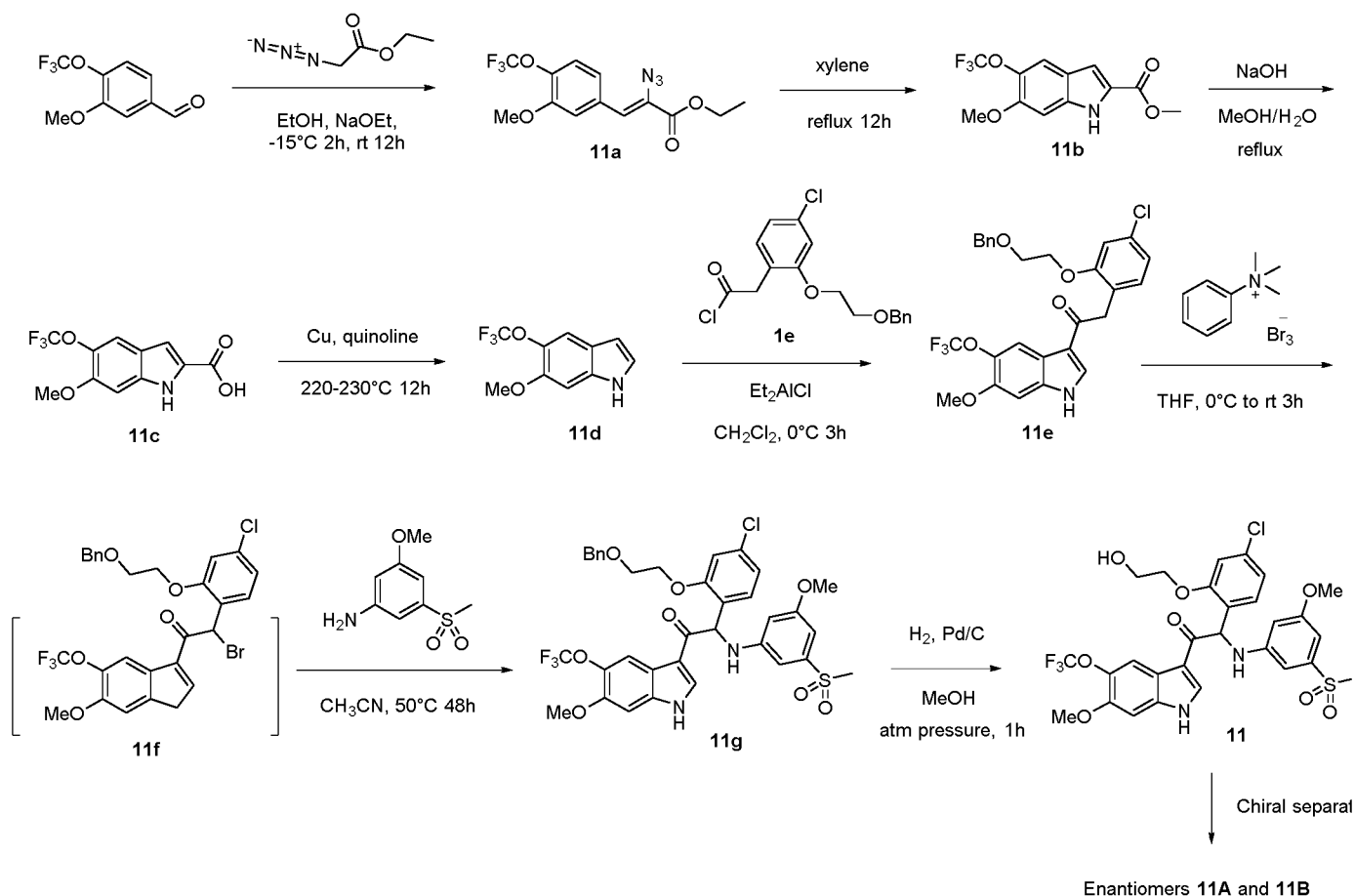
1H NMR (400 MHz, DMSO- d_6) δ ppm 3.09 (s, 3 H) 3.72 (s, 3 H) 3.88 - 4.05 (m, 2 H) 4.20 (t, $J=4.5$ Hz, 2 H) 5.29 (t, $J=5.5$ Hz, 1 H) 6.39 (d, $J=7.7$ Hz, 1 H)
10 6.58 (t, $J=1.8$ Hz, 1 H) 6.65 (t, $J=1.9$ Hz, 1 H) 6.92 - 6.99 (m, 2 H) 7.09 (d, $J=7.7$ Hz, 1 H) 7.12 (d, $J=2.0$ Hz, 1 H) 7.21 (dd, $J=8.7, 1.9$ Hz, 1 H) 7.38 (d, $J=8.4$ Hz, 1 H) 7.57 (d, $J=8.8$ Hz, 1 H) 8.07 (d, $J=0.9$ Hz, 1 H) 8.77 (s, 1 H) 12.38 (br s, 1 H)

LC/MS (method LC-A): R_t 1.15 min, MH^+ 613

- 15 $[\alpha]_D^{20}$: $+141.7^\circ$ (c 0.525, DMF)

Chiral SFC (method SFC-C): R_t 2.92 min, MH^+ 613, chiral purity 100%.

- Example 11: synthesis of 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)-1-(6-methoxy-5-(trifluoromethoxy)-
20 1*H*-indol-3-yl)ethanone (Compound **11**) and chiral separation to afford the Enantiomers **11A** and **11B**



Synthesis of intermediate 11a:

- To a cooled (-15°C) solution of 3-methoxy-4-(trifluoromethoxy)benzaldehyde [CAS 853771-90-1] (50 g, 230 mmol) and ethyl azidoacetate (89 g, 690 mmol) in EtOH (400 mL) was added dropwise, over a period of 2 h, a solution of NaOEt (0.69 mol, prepared from 15.9 g Na and 700 mL of EtOH). The reaction mixture was stirred at room temperature overnight. After cooling on an ice-bath, the reaction was quenched with a saturated NH₄Cl solution (1.2 L), and stirred for 10 min. The precipitate was filtered off, washed with water, and dried to give (Z)-ethyl 2-azido-3-(3-methoxy-4-(trifluoromethoxy)phenyl)acrylate **11a** (32 g) as a yellowish solid.

Synthesis of intermediate 11b:

- A solution of (Z)-ethyl 2-azido-3-(3-methoxy-4-(trifluoromethoxy)phenyl)acrylate **11a** (3 g, 10 mmol) in xylene (40 mL) was heated under reflux overnight. After cooling to room temperature, the solvent was evaporated to dryness. The residue was triturated with hexane (50 mL) and the precipitate was filtered off

to afford methyl 6-methoxy-5-(trifluoromethoxy)-1*H*-indole-2-carboxylate **11b** (yield: 1.4-1.6 g) as a yellow solid.

Synthesis of intermediate 11c:

- 5 To a mixture of methyl 6-methoxy-5-(trifluoromethoxy)-1*H*-indole-2-carboxylate **11b** (25 g, 87 mmol) in MeOH/H₂O (2/1, 300 mL) was added NaOH (7 g, 175 mmol) and the mixture was heated under reflux until a clear solution was obtained. After cooling to room temperature, most of the methanol was removed under reduced pressure and the remaining aqueous solution was
- 10 acidified with conc. HCl to pH 3-4. The product was extracted with EtOAc (2x 250 mL). The combined organic layers were washed with brine, dried, and evaporated under reduced pressure to give 6-methoxy-5-(trifluoromethoxy)-1*H*-indole-2-carboxylic acid **11c** (22.7 g) as a grey solid.

15 **Synthesis of intermediate 11d:**

- A suspension of 6-methoxy-5-(trifluoromethoxy)-1*H*-indole-2-carboxylic acid **11c** (7.5 g, 27 mmol) and Cu (1.22 g, 0.7 equiv.) in quinoline (150 mL) was heated to 220-230°C under inert atmosphere for 12 h. After cooling to room temperature, the mixture was diluted with methyl *tert*-butyl ether (MTBE,
- 20 400 mL) and washed with a saturated aqueous NaHSO₄ solution (2x 500 mL). The organic layer was dried over MgSO₄, filtered through short pad of silica gel, and evaporated under reduced pressure. The residue was purified by column chromatography to afford 6-methoxy-5-(trifluoromethoxy)-1*H*-indole **11d** (3.75 g) as a yellow solid.

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Synthesis of the intermediate 11e:

- Diethylaluminum chloride 1M in hexane (9.7 mL, 9.7 mmol) was added dropwise at 0°C to a solution of 6-methoxy-5-(trifluoromethoxy)-1*H*-indole **11d** (1.5 g, 6.5 mmol) in CH₂Cl₂ (25 mL). After 30 min at 0°C, 2-(2-(2-
- 30 (benzyloxy)ethoxy)-4-chlorophenyl)acetyl chloride **1e** (2.4 g, 7.13 mmol) in CH₂Cl₂ (25 mL) was added dropwise. The reaction was stirred at 0°C for 3 h. The reaction was carefully quenched at 0°C with ice. Water was added, the layers were separated and the aqueous layer was extracted with CH₂Cl₂. The combined organic layers were dried over MgSO₄, filtered, and solvent was
- 35 evaporated under reduced pressure. Purification was carried out by flash chromatography on silica gel (15-40 µm, 120 g, heptane/EtOAc 80/20). The pure fractions were combined and evaporated to dryness to afford 2-(2-(2-

(benzyloxy)ethoxy)-4-chlorophenyl)-1-(6-methoxy-5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone **11e** (2.1 g).

Synthesis of the intermediate 11g:

5 Under a N₂-flow, at 0°C, a solution of phenyltrimethylammonium tribromide [CAS 4207-56-1] (1.06 g, 2.81 mmol) in THF (37 mL) was added dropwise to a solution of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(6-methoxy-5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone **11e** (1.5 g, 2.81 mmol) in THF (38 mL). The mixture was stirred at 0°C for 1 h. The cooling bath was removed
10 and stirring was continued at room temperature for 3 h. A solution of 3-methoxy-5-(methylsulfonyl)aniline [CAS 62606-02-4] (1.69 g, 8.43 mmol) in CH₃CN (30 mL) was added and the resulting mixture was stirred at 50°C for 48 h. The mixture was concentrated under reduced pressure. The residue was taken up with EtOAc, washed with water, 1N HCl (3 times), and then with a
15 solution of 10% K₂CO₃ in water. The organic layer was dried over MgSO₄, filtered, and the solvent was concentrated under reduced pressure. Purification was performed by flash chromatography on silica gel (15-40 µm, 80 g, eluent: CH₂Cl₂/CH₃OH 99.5/0.5). The pure fractions were combined and evaporated to dryness. The residue was purified again by flash chromatography on silica gel
20 (15-40 µm, 40 g, eluent: heptane/EtOAc gradient 60/40 to 50/50). The pure fractions were combined and evaporated to dryness yielding 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)-1-(6-methoxy-5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone **11g** (1.075 g).

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Synthesis of Compound 11 and chiral separation into Enantiomers 11A and 11B:

A mixture of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)-1-(6-methoxy-5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone **11g** (934 mg, 1.27 mmol) in CH₃OH (18 mL) was hydrogenated for
30 1 h under atmospheric pressure of H₂ using 10% Pd/C (271 mg, 0.255 mmol) as the catalyst. The reaction was diluted with CH₂Cl₂ and filtered through a pad of celite®. The filtrate was evaporated under reduced pressure. Purification was carried out by flash chromatography on silica gel (15-40 µm, 24 g, eluent:
35 CH₂Cl₂/CH₃OH:99/1). The pure fractions were combined and evaporated to dryness to afford 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)-1-(6-methoxy-5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone (compound **11**, 540 mg) as a racemic mixture. The enantiomers of

Compound **11** (540 mg) were separated via chiral SFC (Stationary phase: Chiralpak® IA 5 μ m 250 x 20 mm, Mobile phase: 70% CO₂, 30% iPOH + 0.3% iPrNH₂) yielding 250 mg of the first eluted enantiomer and 260 mg of the second eluted enantiomer. The first eluted enantiomer was precipitated from diisopropyl ether/Et₂O/heptane. The precipitate was filtered off and dried to give 209 mg of Enantiomer **11A**. The second eluted enantiomer was precipitated from diisopropyl ether. The precipitate was filtered off and dried to give 172 mg of Enantiomer **11B**.

10 **Compound 11:**

¹H NMR (500 MHz, DMSO-*d*₆) δ ppm 3.08 (s, 3 H) 3.72 (s, 3 H) 3.84 - 4.04 (m, 5 H) 4.15 - 4.23 (m, 2 H) 5.28 (br s, 1 H) 6.36 (d, *J*=7.9 Hz, 1 H) 6.58 (t, *J*=1.9 Hz, 1 H) 6.64 (s, 1 H) 6.93 (s, 1 H) 6.96 (dd, *J*=8.2, 1.9 Hz, 1 H) 7.06 (d, *J*=7.9 Hz, 1 H) 7.12 (d, *J*=1.9 Hz, 1 H) 7.17 (s, 1 H) 7.37 (d, *J*=8.5 Hz, 1 H) 8.03 (d, *J*=1.3 Hz, 1 H) 8.63 (s, 1 H) 12.16 (br s, 1 H)

LC/MS (method LC-C): *R*_t 3.10 min, MH⁺ 643

Melting point: 212°C

Enantiomer 11A:

20 ¹H NMR (500 MHz, DMSO-*d*₆) δ ppm 3.08 (s, 3 H) 3.72 (s, 3 H) 3.87 (s, 3 H) 3.89 - 4.05 (m, 2 H) 4.19 (br t, *J*=4.4 Hz, 2 H) 5.30 (br s, 1 H) 6.36 (d, *J*=7.6 Hz, 1 H) 6.58 (t, *J*=1.6 Hz, 1 H) 6.64 (br s, 1 H) 6.93 (s, 1 H) 6.96 (dd, *J*=8.4, 1.7 Hz, 1 H) 7.07 (d, *J*=7.9 Hz, 1 H) 7.12 (d, *J*=1.9 Hz, 1 H) 7.17 (s, 1 H) 7.37 (d, *J*=8.5 Hz, 1 H) 8.04 (d, *J*=0.6 Hz, 1 H) 8.63 (s, 1 H) 12.10 (br s, 1 H)

25 LC/MS (method LC-C): *R*_t 3.09 min, MH⁺ 643

$[\alpha]_D^{20}$: -102.3° (c 0.208, DMF)

Chiral SFC (method SFC-F): *R*_t 3.61 min, M-F 625, chiral purity 100%.

Enantiomer 11B:

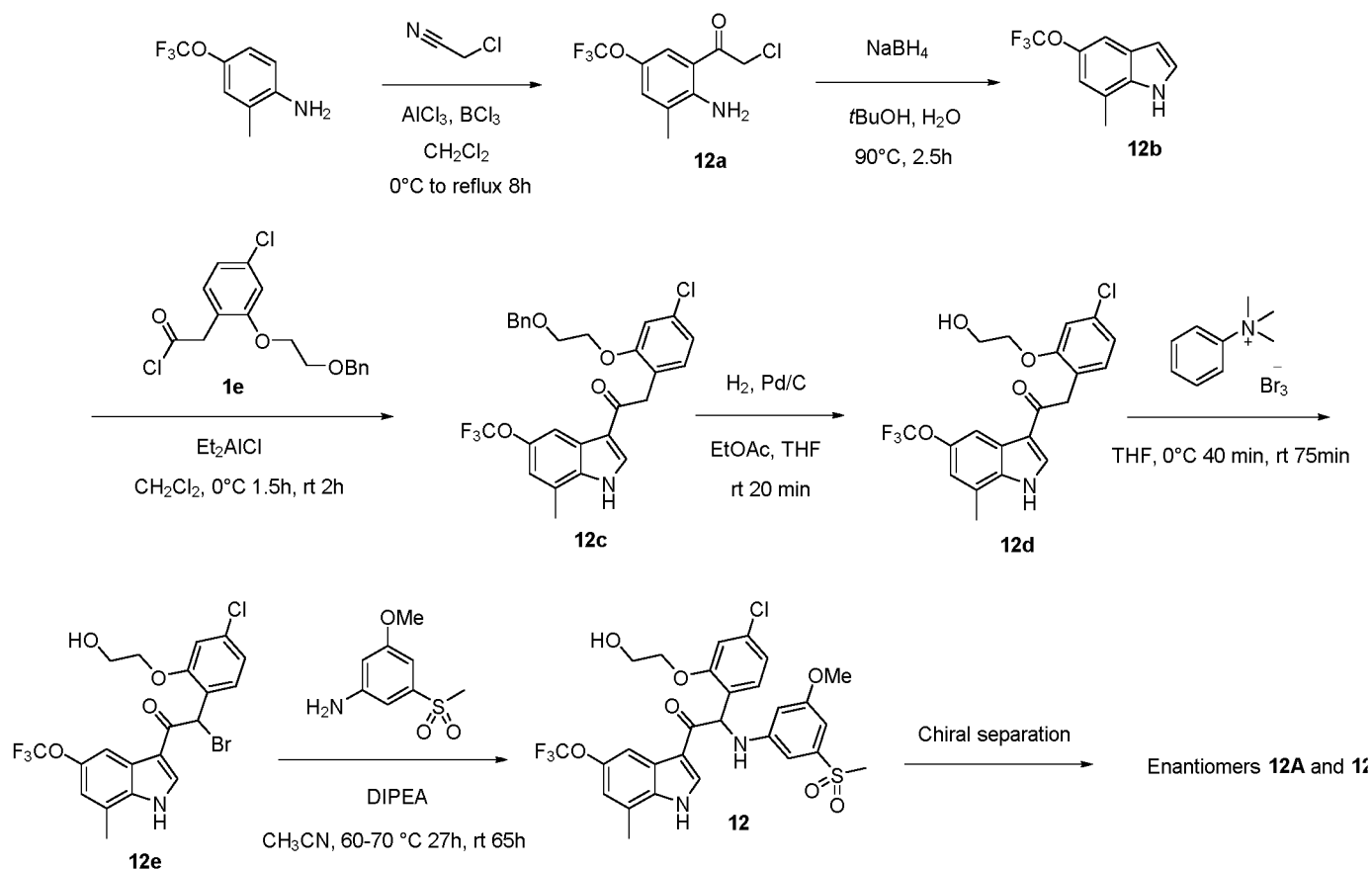
30 ¹H NMR (500 MHz, DMSO-*d*₆) δ ppm 3.09 (s, 3 H) 3.72 (s, 3 H) 3.87 (s, 3 H) 3.88 - 4.04 (m, 2 H) 4.19 (t, *J*=4.6 Hz, 2 H) 5.30 (t, *J*=5.5 Hz, 1 H) 6.36 (d, *J*=7.9 Hz, 1 H) 6.58 (t, *J*=1.7 Hz, 1 H) 6.64 (br s, 1 H) 6.93 (s, 1 H) 6.96 (dd, *J*=8.2, 1.9 Hz, 1 H) 7.07 (d, *J*=7.9 Hz, 1 H) 7.12 (d, *J*=1.9 Hz, 1 H) 7.18 (s, 1 H) 7.37 (d, *J*=8.2 Hz, 1 H) 8.04 (d, *J*=0.9 Hz, 1 H) 8.63 (s, 1 H) 12.10 (br s, 1 H)

35 LC/MS (method LC-C): *R*_t 3.09 min, MH⁺ 643

$[\alpha]_D^{20}$: +101.8° (c 0.208, DMF)

Chiral SFC (method SFC-F): R_t 4.38 min, MH^+ 643, chiral purity 98.7%.

Example 12: synthesis of 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)-1-(7-methyl-5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone (Compound **12**) and chiral separation into Enantiomers **12A** and **12B**.



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Synthesis of intermediate 12a:

A mixture of boron(III) chloride 1M in CH_2Cl_2 (25.5 mL, 25.5 mmol) and aluminum(III) chloride (3.40 g, 25.5 mmol) was diluted with CH_2Cl_2 (20 mL) and cooled on an ice-bath under N_2 -atmosphere. A solution of 2-methyl-4-(trifluoromethoxy)aniline [CAS 86256-59-9] (4.88 g, 25.5 mmol) and chloroacetonitrile (3.24 mL, 51.0 mmol) in CH_2Cl_2 (7.5 mL) was added dropwise. After addition, the ice-bath was removed and the mixture was heated under reflux for 8 h. The mixture was cooled again to $0^\circ C$ using an ice-bath. 2N HCl (75 mL) was added dropwise, causing heavy precipitation. The resulting suspension was heated under reflux for 90 min, and cooled to room

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temperature. The solids were removed by filtration. The filter cake was washed with CH₂Cl₂ (4x). The filtrates were combined and the phases were separated. The organic layer was isolated, washed with an aqueous NaHCO₃ solution, dried over MgSO₄, filtered and evaporated under reduced pressure. The residue was purified by flash chromatography (Stationary phase: Biotage® SNAP Ultra Silica 100 g, Mobile phase: heptane/CH₂Cl₂ gradient 100/0 to 0/100). The desired fractions were combined and concentrated to a residual volume of 30 mL. The precipitate was filtered off, washed with heptane and CH₂Cl₂, and dried under vacuum at 50°C to provide 1-(2-amino-3-methyl-5-(trifluoromethoxy)phenyl)-2-chloroethanone **12a** (1.37 g). The filtrate was concentrated under reduced pressure. The solid residue was stirred up in a mixture of heptane (20 mL) and diisopropyl ether (3 mL), filtered off, washed with heptane (3x) and dried under vacuum at 50°C to provide a second fraction of **12a** (0.24 g).

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Synthesis of intermediate 12b:

Sodium borohydride (326 mg, 8.61 mmol) was added to a stirred solution of 1-(2-amino-3-methyl-5-(trifluoromethoxy)phenyl)-2-chloroethanone **12a** (1.92 g, 7.17 mmol) in *tert*-butanol (50 mL) and water (5 mL). The reaction mixture was stirred at room temperature for 30 min and at 90°C for 2.5 h. Water (50 mL) was added and the product was extracted with diethyl ether (2x). The combined organic layers were washed with brine, dried over MgSO₄, filtered and evaporated under reduced pressure. The residue was purified by flash chromatography (Stationary phase: Biotage® SNAP Ultra silica 25 g, Mobile phase: heptane/EtOAc gradient 100/0 to 20/80). The desired fractions were combined, concentrated under reduced pressure, co-evaporated with heptane and dried under vacuum at 50°C to provide 7-methyl-5-(trifluoromethoxy)-1*H*-indole **12b** (1.2 g).

30 **Synthesis of intermediate 12c:**

Diethylaluminum chloride 1M in hexane (18.2 mL, 18.2 mmol) was added dropwise, at 0°C and under N₂-atmosphere, to a solution of 7-methyl-5-(trifluoromethoxy)-1*H*-indole **12b** (2.0 g, 9.3 mmol) in CH₂Cl₂ (150 mL). After stirring for 25 min at 0°C, a solution of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)acetyl chloride **1e** (4.72 g, 13.9 mmol, synthesis: see Example 1) in CH₂Cl₂ (75 mL) was slowly added, keeping the reaction temperature below 5°C. The reaction mixture was stirred at 0°C for 90 min and at room temperature for 2 h. The reaction mixture was cooled to 0°C and a solution of

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Rochelle salt [CAS 6100-16-9] (5.25 g, 18.6 mmol) in water (5.5 mL) was added dropwise. The mixture was vigorously stirred for 30 min at 0°C. The ice-bath was removed and THF (200 mL) was added. After stirring for 1 h at room temperature, Na₂SO₄ (25 g) was added. The mixture was stirred for 18 h and filtered over dicalite®. The filter cake was washed with THF (4x 150 mL) and the combined filtrates were evaporated under reduced pressure. The remaining oil was purified by flash chromatography (Stationary phase: Biotage® SNAP Ultra silica 100 g, Mobile phase: heptane/EtOAc/EtOH gradient 100/0/0 to 40/45/15). The desired fractions were combined and evaporated under reduced pressure. The solid residue was stirred up in a mixture of DIPE (25 mL) and EtOAc (2 mL), filtered off, washed with DIPE (3x), and dried at 50°C under vacuum to give 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(7-methyl-5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone **12c** (2.88 g).

Synthesis of intermediate 12d:

A mixture of 2-(2-(2-(benzyloxy)ethoxy)-4-chlorophenyl)-1-(7-methyl-5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone **12c** (2.88 g, 5.56 mmol) and 10% palladium on carbon (0.5 g) in EtOAc (75 mL) and THF (10 mL) was stirred at room temperature for 20 min under H₂ atmosphere. The reaction mixture was filtered through dicalite® and the filter cake was washed with THF. The combined filtrates were concentrated under reduced pressure. The residue was purified by flash chromatography (stationary phase: Biotage® SNAP Ultra silica 50 g, Mobile phase: heptane/EtOAc/EtOH gradient 100/0/0 to 40/45/15). The fractions containing product were combined and evaporated under reduced pressure. The solid residue was stirred in DIPE (7.5 mL), filtered off, washed with DIPE (2x), and dried under vacuum at 45°C to provide 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(7-methyl-5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone **12d** (780 mg).

Synthesis of intermediate 12e:

Phenyltrimethylammonium tribromide [CAS 4207-56-1] (327 mg, 0.869 mmol) was added to a cooled (0°C) solution of 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(7-methyl-5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone **12d** (354 mg, 0.827 mmol) in THF (15 mL), under N₂-atmosphere. The reaction mixture was stirred at 0°C for 45 min and at room temperature for 75 min. The precipitate was filtered off and washed with THF (2x). The combined filtrates were concentrated under reduced pressure to give 2-bromo-2-(4-chloro-2-(2-

hydroxyethoxy)phenyl)-1-(7-methyl-5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone **12e** (419 mg) which was used in the next step without further purification.

Synthesis of Compound 12 and chiral separation into Enantiomers 12A and 12B:

A mixture of 2-bromo-2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-1-(7-methyl-5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone **12e** (419 mg, 0.827 mmol), 3-methoxy-5-(methylsulfonyl)aniline [CAS 62606-02-4] (333 mg, 1.65 mmol) and diisopropylethylamine (285 μ L, 1.65 mmol) in CH₃CN (30 mL) was stirred at 60°C for 20 h. The reaction was continued at 70°C for 7 h, and at room temperature for 65 h. The volatiles were evaporated under reduced pressure. The solid residue was combined with another fraction (1.14 g) and purified by flash chromatography (stationary phase: Grace Reveleris® silica 40 g, Mobile phase: heptane/EtOAc/EtOH gradient 100/0/0 to 40/45/15). The desired fractions were combined and evaporated under reduced pressure. The residue was purified via preparative HPLC (Stationary phase: RP XBridge® Prep C18 OBD – 10 μ m, 50 x 150 mm, Mobile phase: 0.25% NH₄HCO₃ solution in water, CH₃CN). The desired fractions were combined and evaporated under reduced pressure. The product was crystallized at room temperature from EtOH (10 mL), filtered off, washed with EtOH (2x), and dried at 45°C under vacuum to provide racemic 2-(4-chloro-2-(2-hydroxyethoxy)phenyl)-2-((3-methoxy-5-(methylsulfonyl)phenyl)amino)-1-(7-methyl-5-(trifluoromethoxy)-1*H*-indol-3-yl)ethanone (Compound **12**, two crops: 485 mg and 169 mg). The chiral separation of the enantiomers of Compound **12** (602 mg) was performed via Normal Phase Chiral separation (Stationary phase: Whelk-O1 (R,R), Mobile phase: 30% ethanol, 70% heptane). The product fractions were combined and evaporated to provide Enantiomer **12A** as the first eluted product and Enantiomer **12B** as the second eluted product. Both enantiomers were stirred up in a mixture of water (3.5 mL) and MeOH (1.25 mL), filtered off, washed with water/MeOH 3/1 (4x) and dried under vacuum at 45°C to provide Enantiomer **12A** (202 mg) and Enantiomer **12B** (166 mg).

Compound 12:

¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 2.51 (s, 3 H) 3.08 (s, 3 H) 3.72 (s, 3 H) 3.90 - 4.06 (m, 2 H) 4.19 (t, *J*=4.6 Hz, 2 H) 5.28 (t, *J*=5.7 Hz, 1 H) 6.41 (d, *J*=7.7 Hz, 1 H) 6.58 (t, *J*=1.8 Hz, 1 H) 6.66 (t, *J*=2.1 Hz, 1 H) 6.92 - 6.98 (m, 2 H) 7.02 - 7.08 (m, 2 H) 7.11 (d, *J*=2.0 Hz, 1 H) 7.37 (d, *J*=8.4 Hz, 1 H) 7.92 (br s, 1 H) 8.70 (s, 1 H) 12.38 (s, 1 H)

LC/MS (method LC-A): R_t 1.18 min, MH^+ 627

Enantiomer 12A:

1H NMR (400 MHz, DMSO- d_6) δ ppm 2.51 (br s, 3 H) 3.09 (s, 3 H) 3.72 (s, 3 H)
5 3.88 - 4.09 (m, 2 H) 4.19 (br t, $J=4.5$ Hz, 2 H) 5.28 (br t, $J=5.4$ Hz, 1 H) 6.41 (br
d, $J=7.7$ Hz, 1 H) 6.58 (br s, 1 H) 6.66 (br s, 1 H) 6.91 - 6.99 (m, 2 H)
7.01 - 7.08 (m, 2 H) 7.11 (br s, 1 H) 7.37 (d, $J=8.4$ Hz, 1 H) 7.91 (br s, 1 H)
8.70 (s, 1 H) 12.38 (br s, 1 H)

LC/MS (method LC-A): R_t 1.21 min, MH^+ 627

10 $[\alpha]_D^{20}$: -111.0° (c 0.51, DMF)

Chiral SFC (method SFC-C): R_t 3.31 min, MH^+ 627, chiral purity 100%.

Enantiomer 12B:

1H NMR (400 MHz, DMSO- d_6) δ ppm 2.51 (br s, 3 H) 3.09 (s, 3 H) 3.72 (s, 3 H)
15 3.90 - 4.06 (m, 2 H) 4.19 (t, $J=4.6$ Hz, 2 H) 5.28 (br s, 1 H) 6.41 (d, $J=7.9$ Hz,
1 H) 6.58 (t, $J=1.8$ Hz, 1 H) 6.66 (br t, $J=2.4$ Hz, 1 H) 6.92 - 6.99 (m, 2 H)
7.01 - 7.08 (m, 2 H) 7.11 (d, $J=1.8$ Hz, 1 H) 7.37 (d, $J=8.4$ Hz, 1 H) 7.92 (br s,
1 H) 8.70 (s, 1 H) 12.38 (br s, 1 H)

LC/MS (method LC-A): R_t 1.21 min, MH^+ 627

20 $[\alpha]_D^{20}$: +105.2° (c 0.515, DMF)

Chiral SFC (method SFC-C): R_t 2.91 min, MH^+ 627, chiral purity 98.5%.

ANTIVIRAL ACTIVITY OF THE COMPOUNDS OF THE INVENTION

DENV-2 antiviral assay

25 The antiviral activity of all the compounds of the invention was tested against
the DENV-2 16681 strain which was labeled with enhanced green fluorescent
protein (eGFP). The culture medium consists of minimal essential medium
supplemented with 2% of heat-inactivated fetal calf serum, 0.04% gentamycin
(50mg/mL) and 2mM of L-glutamine. Vero cells, obtained from ECACC, were
30 suspended in culture medium and 25 μ L was added to 384-well plates
(2500 cells/well), which already contain the antiviral compounds. Typically,
these plates contain a 5-fold serial dilution of 9 dilution steps of the test
compound at 200 times the final concentration in 100% DMSO (200nL). In
addition, each compound concentration is tested in quadruplicate (final
35 concentration range: 25 μ M – 0.000064 μ M or 2.5 μ M – 0.000064 μ M for the
most active compounds). Finally, each plate contains wells which are assigned
as virus controls (containing cells and virus in the absence of compound), cell

controls (containing cells in the absence of virus and compound) and medium controls (containing medium in the absence of cells, virus and compounds). To the wells assigned as medium control, 25µL of culture medium was added instead of Vero cells. Once the cells were added to the plates, the plates were

5 incubated for 30 minutes at room temperature to allow the cells to distribute evenly within the wells. Next, the plates were incubated in a fully humidified incubator (37°C, 5%CO₂) until the next day. Then, DENV-2 strain 16681, labeled with eGFP, was added at a multiplicity of infection (MOI) of 0.5. Therefore, 15 µL of virus suspension was added to all the wells containing test

10 compound and to the wells assigned as virus control. In parallel, 15µL of culture medium was added to the medium and cell controls. Next, the plates were incubated for 3 days in a fully humidified incubator (37°C, 5% CO₂). At the day of the read out, the eGFP fluorescence was measured using an automated fluorescence microscope at 488 nm (blue laser). Using an in-house LIMS

15 system, inhibition dose response curves for each compound were calculated and the half maximal effective concentration (EC₅₀) was determined. Therefore, the percent inhibition (I) for every test concentration is calculated using the following formula: $I = 100 \cdot (S_T - S_{CC}) / (S_{VC} - S_{CC})$; S_T, S_{CC} and S_{VC} are the amount of eGFP signal in the test compound, cell control and virus control wells,

20 respectively. The EC₅₀ represents the concentration of a compound at which the virus replication is inhibited with 50%, as measured by a 50% reduction of the eGFP fluorescent intensity compared to the virus control. The EC₅₀ is calculated using linear interpolation (Table 1).

In parallel, the toxicity of the compounds was assessed on the same plates.

25 Once the read-out for the eGFP signal was done, 40µL of ATPlite, a cell viability stain, was added to all wells of the 384-well plates. ATP is present in all metabolically active cells and the concentration declines very rapidly when the cells undergo necrosis or apoptosis. The ATPlite assay system is based on the production of light caused by the reaction of ATP with added luciferase and

30 D-luciferin. The plates were incubated for 10 minutes at room temperature. Next, the plates were measured on a ViewLux. The half maximal cytotoxic concentration (CC₅₀) was also determined, defined as the concentration required to reduce the luminescent signal by 50% compared to that of the cell control wells. Finally, the selectivity index (SI) was determined for the

35 compounds, which was calculated as followed: $SI = CC_{50} / EC_{50}$.

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10

Table 1: EC_{50} , CC_{50} , and SI for the compounds of the invention in the DENV-2 antiviral assay

compound#	EC_{50} (μM)	N	CC_{50} (μM)	N	SI	N
1	0.00043	4	5.1	4	11800	4
1A	0.00024	7	3.7	7	>13500	7
1B	0.0035	4	8.4	4	2420	4
2	0.00027	4	4.2	4	>10300	4
2A	0.00011	10	3.4	10	>37500	9
2B	0.0049	4	11	4	2130	4
3	0.00054	5	12	5	>31100	5
3A	0.00032	6	5.2	6	16400	6
3B	0.056	4	13	4	226	4
4	0.00042	4	3.7	4	>7400	4
4A	0.00013	9	3.0	9	>23200	9
4B	0.011	5	5.7	6	509	5
5	0.00010	11	4.3	12	>35600	11
5A	0.0084	4	5.0	4	595	4
5B	0.000066	5	3.4	5	>43900	5
6	0.00037	4	4.7	4	12700	4
6A	0.00013	5	3.8	6	>38500	5
6B	0.036	5	5.9	4	164	4
7	0.00026	5	2.8	6	>10500	5
7A	0.00022	5	2.7	4	12573	3
7B	0.0050	3	9.2	3	1850	3
8	0.00027	3	2.8	3	>9820	3
8A	0.00011	4	>2.5	4	25900	4

compound#	EC ₅₀		CC ₅₀		SI	
	(μ M)	N	(μ M)	N		N
8B	0.0017	4	>2.4	5	>1450	4
9	0.00010	4	>2.5	4	>30100	4
9A	0.0019	4	11	4	5720	4
9B	0.000061	4	>2.4	4	>39700	4
10	0.00010	4	>2.4	4	>29600	4
10A	0.0029	3	2.3	3	783	3
10B	0.000067	5	2.4	5	>42700	5
11	0.00020	4	>2.5	4	17200	4
11A	0.0015	4	>2.3	4	>2220	4
11B	0.000089	3	>2.3	3	>26400	3
12	0.00010	6	2.5	6	>45600	6
12A	0.0050	6	2.5	7	1030	6
12B	0.000034	9	2.5	11	>390600	9

Tetravalent reverse transcriptase quantitative-PCR (RT-qPCR) assay: Protocol A.

The antiviral activity of the compounds of the invention was tested against

5 DENV-1 strain TC974#666 (NCPV), DENV-2 strain 16681, DENV-3 strain H87 (NCPV) and DENV-4 strains H241 (NCPV) and EDEN (SG/06K2270DK1/2005; GenBank accession number QG398256) in a RT-qPCR assay. Therefore, Vero cells were infected with either DENV-1, or -2, or -3, or -4 in the presence or absence of test compounds. At day 3 post-infection, the cells were lysed and

10 cell lysates were used to prepare cDNA of both a viral target (the 3'UTR of DENV; Table 2) and a cellular reference gene (β -actin, Table 2). Subsequently, a duplex real time PCR was performed on a Lightcycler480 instrument. The generated Cp value is inversely proportional to the amount of RNA expression of these targets. Inhibition of DENV replication by a test compound results in a

15 shift of Cp's for the 3'UTR gene. On the other hand, if a test compound is toxic to the cells, a similar effect on β -actin expression will be observed. The comparative $\Delta\Delta$ Cp method is used to calculate EC₅₀, which is based on the relative gene expression of the target gene (3'UTR) normalized with the cellular housekeeping gene (β -actin). In addition, CC₅₀ values are determined based on

20 the Cp values acquired for the housekeeping gene β -actin.

Table 2: Primers and probes used for the real-time, quantitative RT-PCR.

Primer/probe	Target	Sequence ^{a, b}
F3utr258	DENV 3'-UTR	5'-CGGTTAGAGGAGACCCCTC-3'
R3utr425	DENV 3'-UTR	5'-GAGACAGCAGGATCTCTGGTC-3'
P3utr343	DENV 3'-UTR	FAM -5'-AAGGACTAG- ZEN - AGGTTAGAGGAGACCCCCC-3'- IABkFQ
Factin743	β -actin	5'-GGCCAGGTCATCACCATT-3'
Ractin876	β -actin	5'-ATGTCCACGTCACACTTCATG-3'
Pactin773	β -actin	HEX -5'-TTCCGCTGC- ZEN -CCTGAGGCTCTC-3'- IABkFQ

^a Reporter dyes (FAM, HEX) and quenchers (ZEN and IABkFQ) elements are indicated in bold and italics.

^b The nucleotide sequence of the primers and probes were selected from the conserved region in the 3'UTR region of the dengue virus genome, based on the alignment of 300 nucleotide sequences of the four dengue serotypes deposited in Genbank (Gong et al., 2013, Methods Mol Biol, Chapter 16).

The culture medium consisted of minimal essential medium supplemented with 2% of heat-inactivated fetal calf serum, 0.04% gentamycin (50mg/mL) and 2mM of L-glutamine. Vero cells, obtained from ECACC, were suspended in culture medium and 75 μ L/well was added in 96-well plates (10000 cells/well), which already contain the antiviral compounds. Typically, these plates contain a 5-fold serial dilution of 9 dilution steps of the test compound at 200 times the final concentration in 100% DMSO (500nL; final concentration range: 25 μ M - 0.000064 μ M or 2.5 μ M – 0.0000064 μ M for the most active compounds). In addition, each plate contains wells which are assigned as virus controls (containing cells and virus in the absence of compound) and cell controls (containing cells in the absence of virus and compound). Once the cells were added in the plates, the plates were incubated in a fully humidified incubator (37°C, 5%CO₂) until the next day. Dengue viruses serotype-1, 2, 3 and 4 were diluted in order to obtain a Cp of ~22-24 in the assay. Therefore, 25 μ L of virus suspension, was added to all the wells containing test compound and to the wells assigned as virus control. In parallel, 25 μ L of culture medium was added to the cell controls. Next, the plates were incubated for 3 days in a fully humidified incubator (37°C, 5%CO₂). After 3 days, the supernatant was removed from the wells and the cells were washed twice with ice-cold PBS (~100 μ L). The cell pellets within the 96-well plates were stored at -80°C for at

least 1 day. Next, RNA was extracted using the Cells-to-CT™ lysis kit, according to the manufacturer's guideline (Life Technologies). The cell lysates can be stored at -80°C or immediately used in the reverse transcription step. In preparation of the reverse transcription step, mix A (table 3A) was prepared and 7.57 µL/well was dispensed in a 96-well plate. After addition of 5 µL of the cell lysates, a five minute denaturation step at 75°C was performed (table 3B). Afterwards, 7.43 µL of mix B was added (table 3C) and the reverse transcription step was initiated (table 3D) to generate cDNA.

Finally, a RT-qPCR mix was prepared, mix C (table 4A), and 22.02 µL/well was dispensed in 96-well LightCycler qPCR plates to which 3 µL of cDNA was added and the qPCR was performed according to the conditions in table 4B on a LightCycler 480.

Using the LightCycler software and an in-house LIMS system, dose response curves for each compound were calculated and the half maximal effective concentration (EC₅₀) and the half maximal cytotoxic concentration (CC₅₀) were determined (Tables 5-8).

Table 3: cDNA synthesis using Mix A, denaturation, Mix B and reverse transcription.

A	Mix A				
	Plates	8			
	Samples	828			
	Mix Item	Concentration			Reaction Vol. (µl)
		Unit	Stock	Final	20
					Volume for (µl)
					1 sample
					x samples
	Milli-Q H ₂ O				7.27
	R3utr425	µM	20	0.27	0.15
	Ractin876	µM	20	0.27	0.15
			Volume mix/well (µl)		7.57
			Cell lysates		5.00

Denaturation**B step:**

Step	Temp	Time
Denaturation	75°C	5'
Hold	4°C	hold

C Mix B

Samples	864				
Mix Item	Concentration			Volume for (μl)	
	Unit	Stock	Final	1 sample	x samples
Expand HIFI buffer 2	X	10.00	1.00	2.00	1728.0
MgCl ₂	mM	25.00	3.50	2.80	2419.2
dNTPs	mM	10.00	1.00	2.00	1728.0
Rnase inhibitor	U/μl	40.00	1.00	0.50	432.0
Expand RT	U/μl	50.00	0.33	0.13	112.3
			Total Volume Mix (μl)	7.43	

D Protocol cDNA synthesis

Step	Temp	Time
Rev transc	42°C	30'
Denaturation	99°C	5'
Hold	4°C	hold

Table 4: qPCR mix and protocol.

A Mix C

Samples	833			Reaction Vol. (μl)	25
Mix Item	Concentration			Volume for (μl)	
	Unit	Stock	Final	1 sample	x samples
H ₂ O PCR grade Roche				7.74	6447.42
Roche 2xMM mix	X	2	1	12.50	10412.50
F3utr258	μ M	20	0.3	0.38	316.54
R3utr425	μ M	20	0.3	0.38	316.54
P3utr343	μ M	20	0.1	0.13	108.29
Factin743	μ M	20	0.3	0.38	316.54
Ractin876	μ M	20	0.3	0.38	316.54
Pactin773	μ M	20	0.1	0.13	108.29
Volume Mix / Tube (μl)				22.02	
cDNA				3.00	

B Protocol qPCR3

Step	Temp	Time	Ramp rate	
preincub/denat	95°C	10 min	4.4	
Denaturation	95°C	10 sec	4.4	40 cycles
annealing	58°C	1 min	2.2	
Elongation	72°C	1 sec	4.4	
Cooling	40°C	10 sec	1.5	

Table 5: EC₅₀, CC₅₀, and SI for the compounds against serotype 1 in the RT-qPCR assays

compound#	Protocol A					
	RT-qPCR serotype 1 TC974#666					
	EC50 (μ M)	N	CC50 (μ M)	N	SI	N
1A	0.0013	3	>2.5	3	>2200	3
2A	0.0016	5	>2.5	5	>1980	5
3A	0.0042	4	5.8	4	1380	4
4A	0.0017	4	>2.5	4	>2160	4
5B	0.0016	3	>2.5	3	>967	3
6A	0.0016	6	4.9	5	2830	5
7A	0.00097	3	2.9	2	5380	2
8A	0.00039	3	6.5	3	17200	3
9B	0.000083	6	2.9	6	>38700	6
10B	0.000071	6	>2.4	7	>41300	6
11B	0.00012	3	>2.5	3	>26300	3
12B	0.000086	5	>2.3	5	>26700	5

N= the number of independent experiments in which the compounds were tested.

5

Table 6: EC₅₀, CC₅₀, and SI for the compounds against serotype 2 in the RT-qPCR assays

compound#	Protocol A					
	RT-qPCR serotype 2 16681					
	EC50 (μ M)	N	CC50 (μ M)	N	SI	N
1A	0.00024	4	4.1	4	29700	4
2A	0.00014	7	4.0	9	>29700	7
3A	0.00040	5	5.3	5	16800	5
4A	0.00016	7	3.7	8	40100	7
5B	0.000065	5	3.6	6	>52300	5
6A	0.00021	5	4.5	8	65400	5
7A	0.00021	3	3.8	4	>14800	3
8A	0.00027	3	6.1	4	89000	3
9B	0.000054	3	>2.5	4	>49900	3

compound#	Protocol A					
	RT-qPCR serotype 2 16681					
	EC50 (μ M)	N	CC50 (μ M)	N	SI	N
10B	0.000049	3	>2.5	3	>62800	3
11B	0.000062	5	2.9	4	>46200	4
12B	0.000053	3	>2.5	4	>56600	3

N= the number of independent experiments in which the compounds were tested.

Table 7: EC₅₀, CC₅₀, and SI for the compounds against serotype 3 in the RT-qPCR assays

5

compound#	Protocol A					
	RT-qPCR serotype 3 H87					
	EC50 (μ M)	N	CC50 (μ M)	N	SI	N
1A	0.0096	3	>2.5	3	>421	3
2A	0.018	5	>2.5	5	>186	5
3A	0.042	4	4.0	4	110	4
4A	0.018	4	>2.5	4	>173	4
5B	0.012	4	>2.5	3	>218	3
6A	0.017	6	3.9	4	198	4
7A	0.011	3	>2.5	3	299	3
8A	0.0042	3	4.0	3	1020	3
9B	0.0012	3	>2.2	3	1970	3
10B	0.0014	3	>2.5	3	>2220	3
11B	0.0013	3	>2.5	3	>2210	3
12B	0.0011	3	>2.2	3	2720	3

N= the number of independent experiments in which the compounds were tested.

Table 8: EC₅₀, CC₅₀, and SI for the compounds against serotype 4 in the RT-qPCR assays.

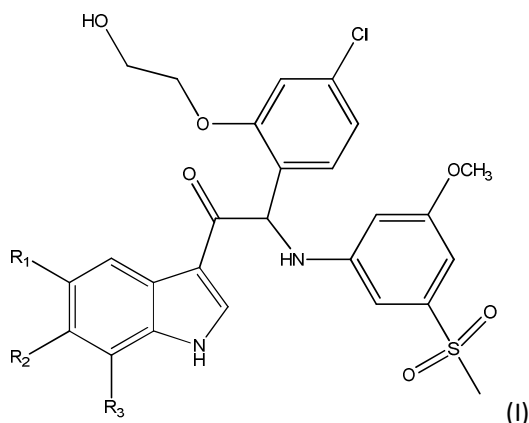
compound#	Protocol A					
	RT-qPCR serotype 4 H241					
	EC50 (μ M)	N	CC50 (μ M)	N	SI	N
1A	0.071	4	>2.4	3	>29	3
2A	0.074	7	2.5	6	39	6
3A	0.11	5	3.0	3	21	3
4A	0.078	8	>2.2	6	24	6
5B	0.074	6	>2.2	6	33	6
6A	0.053	7	>2.5	7	58	7
7A	0.064	4	>2.3	3	37	3
8A	0.036	4	2.6	4	93	4
9B	0.0081	3	>2.5	3	>409	3
10B	0.0069	4	>2.2	4	>331	4
11B	0.011	4	2.2	3	202	3
12B	0.0055	3	>2.1	3	514	3

compound#	Protocol A					
	RT-qPCR serotype 4 EDEN					
	EC50 (μ M)	N	CC50 (μ M)	N	SI	N
1A	ND		ND		ND	
2A	0.0013	5	> 2.5	5	> 2406	5
3A	0.0031	4	4.8	4	2088	4
4A	0.0014	4	> 2.5	4	> 2455	4
5B	0.0011	3	> 2.5	3	> 2989	3
6A	0.0010	5	> 2.5	4	> 2825	4
7A	0.00064	1	3.2	1	4953	1

N= the number of independent experiments in which the compounds were tested.

Claims

1. A compound of formula (I)



5 a stereo-isomeric form, a pharmaceutically acceptable salt, solvate or polymorph thereof comprising a mono- or di-substituted indole group; said compound is selected from the group wherein:

R₁ is H, R₂ is F, Cl or OCH₃ and R₃ is H;

R₁ is H, R₂ is F or Cl and R₃ is CH₃;

10 R₁ is CH₃, R₂ is OCH₃ and R₃ is H;

R₁ is F, R₂ is F and R₃ is H;

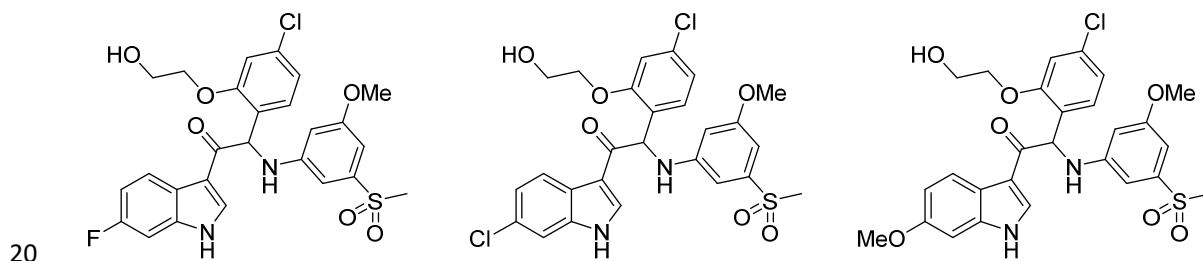
R₁ is CH₃, R₂ is F and R₃ is H;

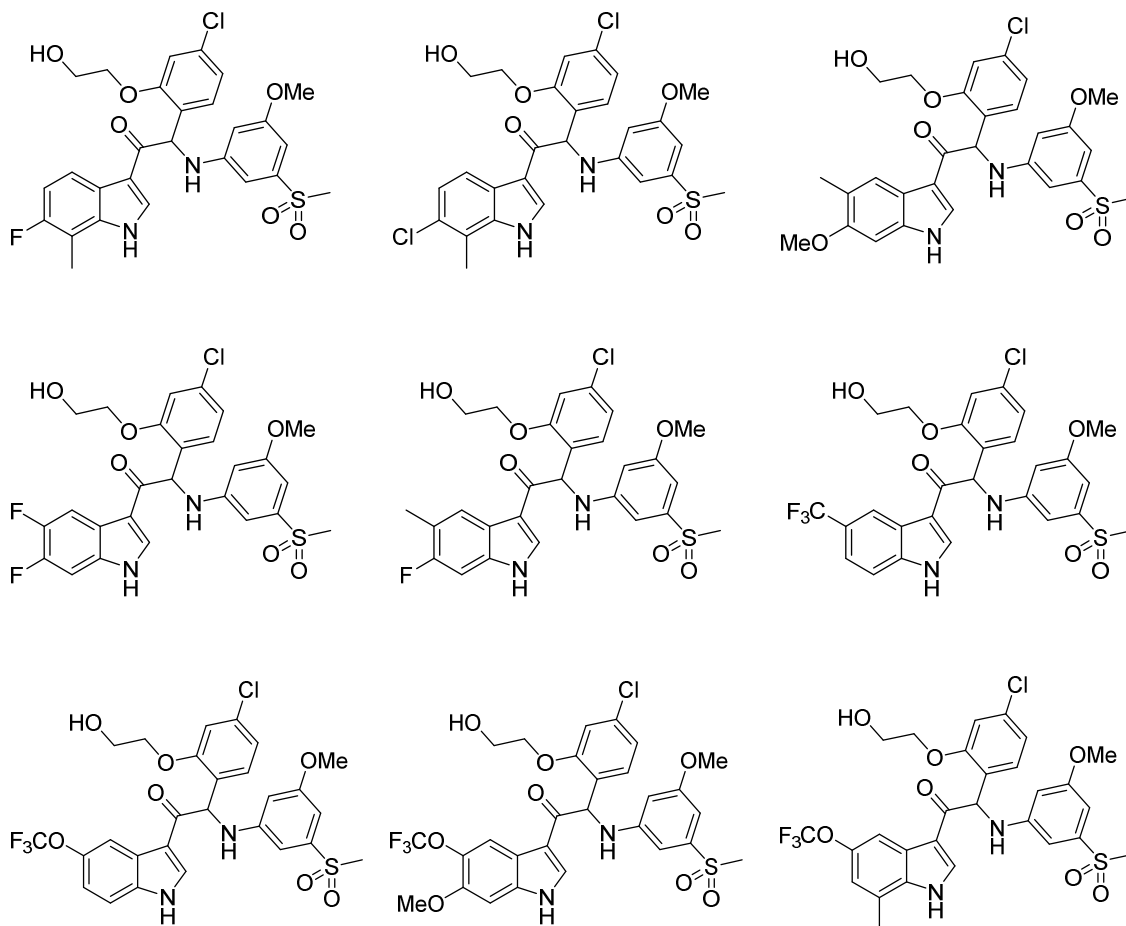
R₁ is CF₃ or OCF₃ and R₂ is H and R₃ is H;

R₁ is OCF₃, R₂ is OCH₃ and R₃ is H or

15 R₁ is OCF₃, R₂ is H and R₃ is CH₃.

2. A compound or its stereo-isomeric form, a pharmaceutically acceptable salt, solvate or polymorph thereof according to claim 1 wherein said compound is selected from the group:

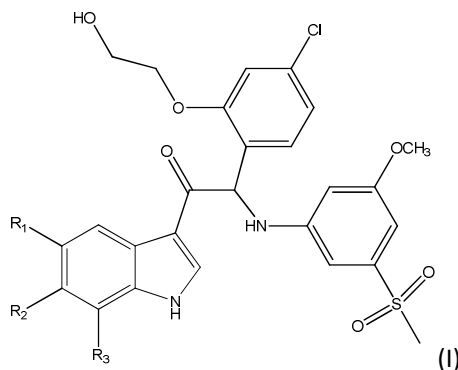




3. A pharmaceutical composition comprising a compound of formula (I) or a stereo- isomeric form , a pharmaceutically acceptable salt, solvate or polymorph thereof according to claim 1 or 2 together with one or more pharmaceutically acceptable excipients, diluents or carriers.
4. A compound of formula (I) or a stereo- isomeric form , a pharmaceutically acceptable salt, solvate or polymorph thereof according to claim 1 or a pharmaceutical composition according to claim 3 for use as a medicament.
5. A compound of formula (I) or a stereo- isomeric form, a pharmaceutically acceptable salt, solvate or polymorph thereof according to claim 1 or a pharmaceutical composition according to claim 3 for use in the treatment

of dengue.

6. Use of a compound represented by the following structural formula (I)



a stereo-isomeric form, a pharmaceutically acceptable salt, solvate or polymorph thereof comprising a mono- or di-substituted indole group; said compound is selected from the group wherein:

R₁ is H, R₂ is F, Cl or OCH₃ and R₃ is H;

R₁ is H, R₂ is F or Cl and R₃ is CH₃;

R₁ is CH₃, R₂ is OCH₃ and R₃ is H;

R₁ is F, R₂ is F and R₃ is H;

R₁ is CH₃, R₂ is F and R₃ is H;

R₁ is CF₃ or OCF₃ and R₂ is H and R₃ is H;

R₁ is OCF₃, R₂ is OCH₃ and R₃ is H or

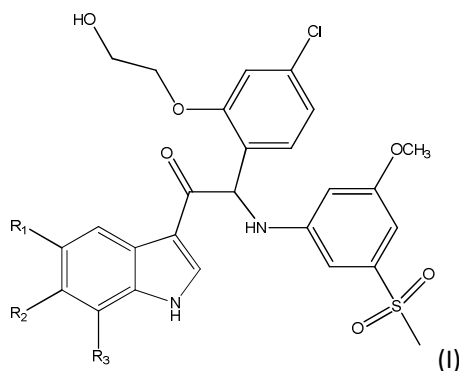
R₁ is OCF₃, R₂ is H and R₃ is CH₃

in the manufacture of a medicament for inhibiting the replication of dengue virus(es) in a patient.

7. The use according to claim 6 further comprising an additional therapeutic agent.

8. The use according to claim 7 wherein said additional therapeutic agent is another antiviral agent.

9. Use of a compound represented by the following structural formula (I)



a stereo-isomeric form, a pharmaceutically acceptable salt, solvate or polymorph thereof comprising a mono- or di-substituted indole group; said compound is selected from the group wherein:

R₁ is H, R₂ is F, Cl or OCH₃ and R₃ is H;

R₁ is H, R₂ is F or Cl and R₃ is CH₃;

R₁ is CH₃, R₂ is OCH₃ and R₃ is H;

R₁ is F, R₂ is F and R₃ is H;

R₁ is CH₃, R₂ is F and R₃ is H;

R₁ is CF₃ or OCF₃ and R₂ is H and R₃ is H;

R₁ is OCF₃, R₂ is OCH₃ and R₃ is H or

R₁ is OCF₃, R₂ is H and R₃ is CH₃

for inhibiting the replication of dengue virus(es) in a biological sample.