



- (51) **International Patent Classification:**  
*C07D 487/04* (2006.01)
- (21) **International Application Number:** PCT/IN2012/000299
- (22) **International Filing Date:** 24 April 2012 (24.04.2012)
- (25) **Filing Language:** English
- (26) **Publication Language:** English
- (30) **Priority Data:** 3079/MUM/2011 2 November 2011 (02.11.2011) IN
- (71) **Applicant (for all designated States except US):** **CADILA HEALTHCARE LIMITED** [IN/IN]; Zydus Tower, Satellite Cross Roads, Ahmedabad - 380 015, Gujarat (IN).
- (72) **Inventors; and**
- (75) **Inventors/Applicants (for US only):** **PANDEY, Bipin** [IN/IN]; Cadila Healthcare Limited, Zydus Tower, Satellite Cross Roads, Ahmedabad - 380 015, Gujarat (IN). **DAVE, Mayank, Ghanshyambhai** [IN/IN]; Cadila Healthcare Limited, Zydus Tower, Satellite Cross Roads, Ahmedabad - 380 015, Gujarat (IN). **KOTHARI, Himanshu, M.** [IN/IN]; Cadila Healthcare Limited, Zydus Tower, Satellite Cross Roads, Ahmedabad - 380 015, Gujarat (IN). **SHUKLA, Bhavin, Shriprasad** [IN/IN]; Cadila Healthcare Limited, Zydus Tower, Satellite Cross Roads, Ahmedabad - 380 015, Gujarat (IN). **MENDIRATTA, Sanjeev, Kumar** [IN/IN]; Cadila Healthcare Limited, Zydus Tower, Satellite Cross Roads, Ahmedabad - 380 015, Gujarat (IN). **JOSHI, Rupal** [IN/IN]; Cadila Healthcare Limited, Zydus Tower, Satellite Cross Roads, Ahmedabad - 380 015, Gujarat (IN). **TRIVEDI, Umang** [IN/IN]; Cadila Healthcare Limited, Zydus Tower, Satellite Cross Roads, Ahmedabad - 380 015, Gujarat (IN).
- (74) **Agents:** **SUBRAMANIAM, Hariharan** et al.; Subramaniam, Nataraj & Associates, E- 556, Greater Kailash-II, New Delhi 110 048 (IN).
- (81) **Designated States (unless otherwise indicated, for every kind of national protection available):** AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.
- (84) **Designated States (unless otherwise indicated, for every kind of regional protection available):** ARIPO (BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).
- Declarations under Rule 4.17:**
- *as to applicant's entitlement to apply for and be granted a patent (Rule 4.17(ii))*
- Published:**
- *with international search report (Art. 21(3))*



WO 2013/065066 A1

(54) **Title:** PROCESSES FOR PREPARING 4-OXO-4-[3-(TRIFLUOROMETHYL)-5,6-DIHYDRO [L,2,41-TRIAZOLO[4,3-A]PYRAZIN-7(8H)-YL]-L-(2,4,5-TRIFLUOROPHENYL)BUTAN-2-AMINE

(57) **Abstract:** The present invention relates to synthesis of 4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro [1,2,4]-triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine of Formula (I) either in its racemic (R/S) form or any of its optically active (S) or (R) forms or enantiomeric excess mixture of any of the forms by novel processes. The invention further relates to certain novel intermediates useful in the preparation of compound of Formula (I) and processes for their preparation.

**PROCESSES FOR PREPARING 4-OXO-4-[3-(TRIFLUOROMETHYL)-5,6-DIHYDRO [1,2,4]-TRIAZOLO[4,3-A]PYRAZIN-7(8H)-YL]-1-(2,4,5-TRIFLUOROPHENYL)BUTAN-2-AMINE**

5

**FIELD OF THE INVENTION**

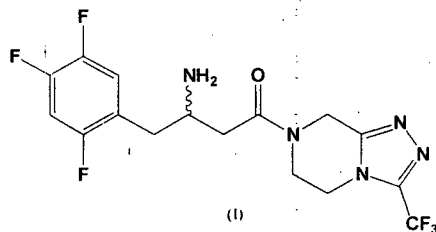
The present invention relates to synthesis of 4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro [1,2,4]-triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine of Formula (I) or its salts either in its racemic (R/S) form or any of its optically active (S) or (R) forms or enantiomeric excess mixture of any of the forms by novel processes. The invention further relates to certain novel intermediates useful in the preparation of compound of Formula (I) and processes for their preparation. The invention also relates to pharmaceutical compositions that include the compound of Formula (I), prepared according to the processes disclosed herein.

15

**BACKGROUND OF THE INVENTION**

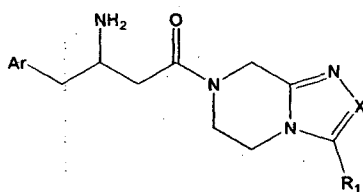
The compound of Formula (I) either in its racemic (R/S) form or any of its optically active (S) or (R) forms or enantiomeric excess mixture of any of the forms is an industrially useful compound having the chemical name 4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro [1,2,4]-triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine and the following structure.

20

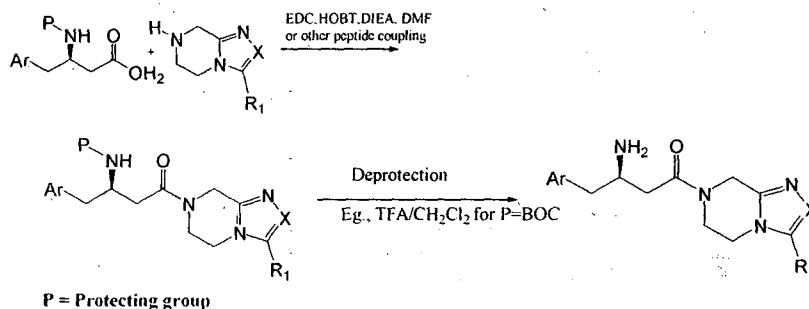


The preparation and use of beta-amino tetrahydrotriazolo[4,3-a]pyrazines, including (2R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro [1,2,4]-triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine (sitagliptin), are disclosed in U.S. patent No. 6,699,871, which are inhibitors of DPP-IV. Disclosed therein are compounds, whose general formula is,

25



(2R)-4-oxo-4-[3-(trifluoromethyl)-5, 6-dihydro [1,2,4]-triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine can be synthesized by method disclosed in the above specification, depicted by following scheme

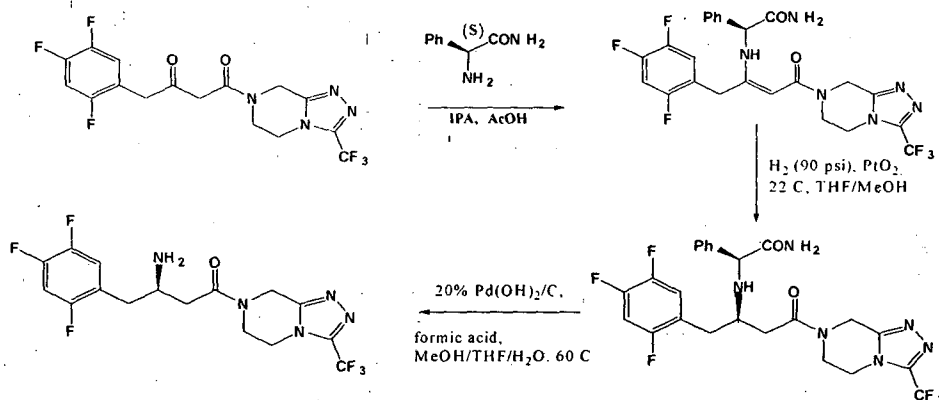


5

However here the overall chemical and optical yield reported is low and there is no mention of stereoisomeric yield.

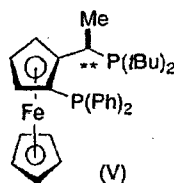
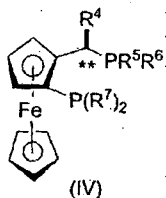
Another process for preparation of compound of Formula (I) in its (R) form is disclosed in WO 2004087650 which involves the formation of chiral benzyloxyazetidione intermediate, which later through subsequent steps is converted to the final product. In this process, chiral reagents have been used from the beginning.

WO 2004085661 describes another process for preparation of chiral beta amino acid derivatives including compound of Formula (I) in its (R) form. Here S-phenylglycine amide (S-PGA) as a chiral auxiliary is used to get pure Z-enamines from diketone. Here a metal catalyst like  $\text{PtO}_2$  is used, which is expensive and high pressure is required. Here > 90% e.e is reported for the reduced amine.

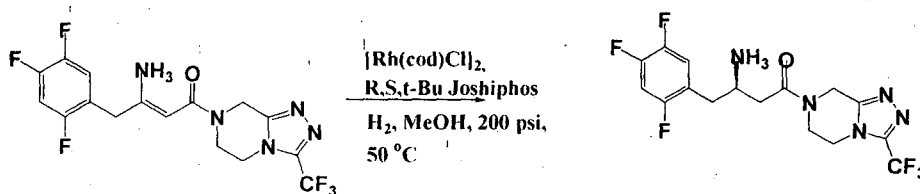


PCT Publication No. WO 2004085378 and WO 2006081151 describe a process of preparation of chiral beta amino acid derivatives which includes compound of

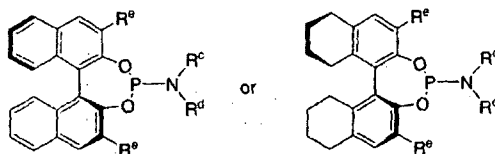
Formula (I) in its (R) form. The product is prepared by an enantioselective reduction via transition metal catalyzed asymmetric hydrogenation at high pressure, of a prochiral enamine. In WO 2004085378, hydrogenation of enamine is carried out to get final product using special catalysts such as R,S t-butyl Josiphos, as given below (IV), and another special catalyst is used in PCT Publication No. WO 2006081151 as given below (V),



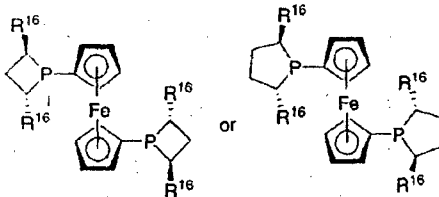
The chiral purity of the product after reduction is claimed to be > 70 % e.e. The scheme is depicted below:



PCT Publication No. WO 2005097733 describes a process of preparation of compound of Formula (I) in its (R) form by asymmetric hydrogenation using Rhodium metal precursor complexes with chiral mono or bisphosphine ligand. Such mono and bis phosphine ligand disclosed are depicted below:



Monophosphine ligand



Bisphosphine ligand

But in all the above processes, the chiral metal catalysts and the diastereoselective catalysts used are very difficult to prepare and are very expensive. The conditions in which the reactions are carried out are extreme i.e. at high pressure,

20

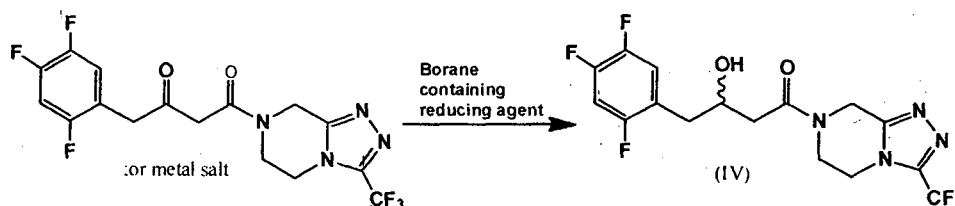
high temperature, etc. Hence there is a need in the art to develop a simple and efficient process for the bulk production of compound of Formula (I).

PCT Publication No. WO2009084024 discloses process for the preparation of compound of Formula (I) and its pharmaceutically acceptable salts by resolving the amine with a suitable resolving agent. Additionally, the same document also describes resolution of a racemic (50:50 mixture of R & S) amine with a chirally pure resolving agent which gives dibenzoyl-L-tartaric acid diastereomer of Formula (I) having ratio of (R) and (S) isomers 50: 50 (racemic mixture).

Of course, by subsequent solvent treatments the desired diastereomer is obtained by repeated recrystallization to get a chiral purity 85-90 %. (Example- 5a, 5b and 5c)

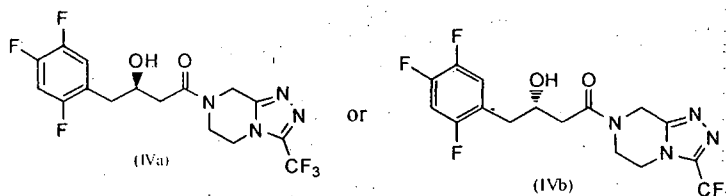
PCT Publication No. WO2009085990 discloses process for the preparation of compound of Formula (I) and its pharmaceutically acceptable salts, wherein the resolution was carried out with di-p-tolyl-L-tartaric acid. Process as disclosed in WO2009085990 involves the use of sodium cyanoborohydride with methanolic HCl (Example 4) during reduction step which may lead to generation of HCN gas. Naturally, generation of such toxic gases can be difficult to manage at large scale level.

PCT Publication No. WO2010032264 discloses the compound 3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl) butan-1-one (IV). The same application also discloses process for the preparation of the 3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl) butan-1-one (IV) by using chemical reduction method, the reduction is performed by a suitable borane containing reducing agent, in absence or presence of an acid in a suitable solvent to obtain 3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl) butan-1-one. The process is illustrated in scheme given below,



Moreover the WO2010032264 only provides the racemic form of 3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl) butan-1-one (Formula IV).

2805/MUM/2010 provides two enantiomers of 3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl) butan-1-one of opposite chirality of the following Formula,



WO2010122578 disclosed process for the preparation of formula (I) in optically active (R) form. The process involves the steps wherein all the intermediates are obtained in (R) configuration from (R)-isomer of starting material with retention throughout all the steps. Such a reaction proposition is difficult to follow. [See for refs. 1. Tetrahedron Assymetry, vol.3 (12), 1475-1504 (1992); 2. Russian Chem. Rev, 73(6), p-581-608 (2004). 3. Reagents for Organic Synthesis, Vol.5, p-728-729, John Wiley & sons by-Fieser & Fieser. 4. Advanced organic chemistry p-294-297 & 346-347, 4<sup>th</sup> Edition, John Wiley & Sons. By-Jerry March].

WO2011049344 and WO201102640 discloses processes for the preparation of compound of formula (I) by using chemical method.

EP2397141 discloses process for the preparation of 3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl) butan-1-one of formula (IV) and also compound of formula (I) comprising, (i) the (S)-epihalohydrin reacts with 1-metalo-2,4,5-trifluorobenzene to form halohydrin (ii) a conversion of halohydrin into cyanohydrin by reaction with cyanide anion (iii) hydrolysis of cyanohydrin leading to carboxylic derivatives (iv) which on amidation process with amines provides compound of formula (IV), which is further converted to formula (I).

The drawback associated with this process is a particular enantiomer of compound of formula (IV) is separated off by chiral chromatography which make the process less efficient to prepare the compound of formula (I) at low cost. Such a process using chiral chromatography, additionally are difficult to handle at large scale and moreover these are not environment friendly.

Several of the above processes either have a problem of low yield and purity or having improperly stated configuration of intermediates or, are difficult to carry out on a commercial scale and are expensive. Hence there is a high unmet need to develop a process which provides the compound of Formula (I) in either its racemic (R/S) form or any of its optically active (S) or (R) forms or enantiomeric excess mixture of any of the forms at low cost and which should be environment friendly, scalable, and industrially applicable. The present invention provides a process which is efficient, cost effective and does not result in impure product, thus making the process amenable for commercial scale use.

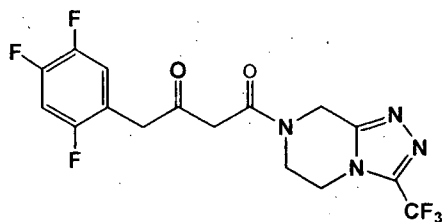
The enzymatic reduction processes in which the enzyme acts as a reduction catalyst are environmentally advantageous compared to the process as described in the EP2397141. The use of the enzymes is also typically lower in cost than the process as in EP2397141. Enzymes can have a unique stereo selective property of producing only one enantiomer of compound of formula (IV) with good chiral purity and therefore no need to do chiral chromatography. The use of compound of formula (IV) prepared by enzymatic route, as a starting material for the preparation of formula (I) is also advantageous because the purity of starting material plays an important role in the preparation of formula (I), in high yield and better purity. Thus the present process overcomes the problem of the prior art.

We herein disclose a process for the preparation of compound Formula (I) into its racemic (R/S) form or any of its optically active (S) or (R) forms or enantiomeric excess mixture of any of the forms by using compound of Formula (IV), which is obtained by enzymatic conversion, in racemic (R/S) form or any of its optically active (S) or (R) forms or as an enantiomeric excess mixture of any of the forms. We herein also disclose some of the novel intermediates generated during the process for the preparation of compound Formula (I).

#### **SUMMARY OF THE INVENTION**

In one general aspect there is provided a process for the preparation of compound Formula (I) either in its racemic (R/S) form or any of its optically active (S) or (R) forms or enantiomeric excess mixture of any of the forms. The process includes:

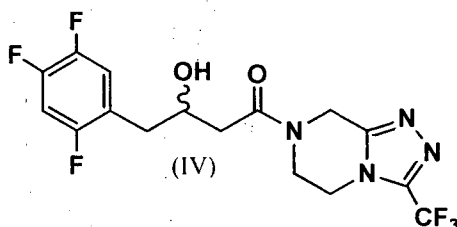
a) reacting 4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one of formula (V) or its metal ion salt



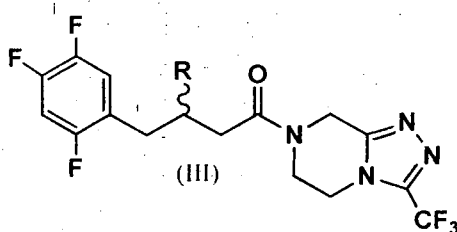
Formula (V)

with suitable enzymes or its variants that reduce a ketone to form an alcohol, by  
 5 maintaining under suitable conditions, to obtain 3-hydroxy-1-(3-(trifluoromethyl)-5,6-  
 dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl) butan-1-one  
 of formula (IV), in racemic (R/S) form or any of its optically active (S) or (R) forms or  
 their enantiomerically excess mixtures.

(b) reacting 3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-  
 10 a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl) butan-1-one of Formula (IV) either in its  
 racemic (R/S) form or any of its optically active (S) or (R) forms or enantiomeric  
 excess mixture of any of the forms



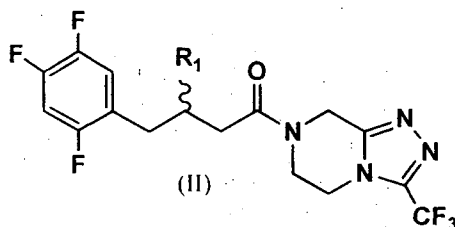
with suitable reagents that activates the hydroxyl group, wherein suitable conditions  
 15 are maintained depending on the corresponding reagents being used, to obtain  
 compound of Formula (III) wherein 'R' represents a suitable leaving group as defined  
 elsewhere in the specification, with retention of configuration;



(c) optionally, isolating the compound of Formula (III);

20 (d) converting compound of Formula (III) to compound of Formula (II) with inversion  
 of configuration, wherein R<sub>1</sub> is as defined elsewhere in the specification, by using  
 suitable amine equivalent, optionally with additives in suitable solvents depending on  
 the R<sub>1</sub> group to be introduced;





(e) suitably deprotecting or reducing the compound of Formula (II) or employing other suitable chemistry to obtain compound of Formula (I) having  $\text{NH}_2$  group with retention of configuration.

5 (f) optionally, the compound of Formula (IV) is directly converted to the compound of Formula (II) with inversion of configuration, by using suitable amine equivalent, optionally with suitable additives in suitable solvents.

(g) optionally, the compound of Formula (III) is directly converted to the compound of Formula (I) with inversion of configuration, by using suitable amine equivalent.

10 In another general aspect there is provided a compound of Formula (III) in racemic (R/S) form or any of its optically active (S) or (R) forms or their enantiomerically excess mixtures and compound of Formula (II) in racemic (R/S) form or any of its optically active (S) or (R) forms or their enantiomerically excess mixtures and their use for the preparation of compound of Formula (I).

15 In another general aspect there is provided a process for the preparation of pharmaceutically acceptable salts of compound of Formula (I)

In another general aspect there is provided a process for chiral enrichment of pharmaceutically acceptable salts of compound of Formula (I).

20 The product so obtained may be further or additionally purified by suitable salt formation, diastereomeric salt formation or by purification using suitable solvents or other techniques known to obtain desired purity levels.

The process may include further formulating the product so obtained into a finished dosage form.

25 The details of one or more embodiments of the invention are set forth in the description below. Other features, objects and advantages of the invention will be apparent from the description and claims.

#### DETAILED DESCRIPTION OF THE INVENTION

As used herein, the term "reflux temperature" refers to the boiling point of the solvent.

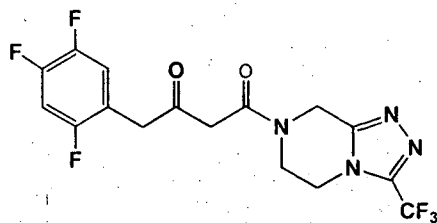
As used herein, the term "compound of Formula (I)" refers to 4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]-triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine in racemic (R/S) form or any of its optically active (S) or (R) forms or their enantiomerically excess mixture.

5 As used herein, the term "THF" refers to tetrahydrofuran, the term "DCM" refers to dichloro methane, the term "DMF" refers to dimethyl formamide, the term "DIPE" refers to di-isopropyl ether, the term "MTBE" refers to methyl t-butyl ether, the term "DMSO" refers to dimethyl sulfoxide, the term "DMA" refers to dimethylacetamide, the term "IPA" refers to isopropyl alcohol, the term TEA refers to  
10 Triethylamine, the term LDA refers to Lithium diisopropylamide, the term KHMDS refers to Potassium hexamethyldisilazane, the term DABCO refers to 1,4-diazabicyclo[2,2,2]octane, the term HMPA refers to Hexamethylphosphoramide, the term DIAD refers to diisopropyl azodicarboxylate, the term DBN refers to 1,5-Diazabicyclo[4.3.0]non-5-ene, the term DBU refers to 1,8-Diazabicyclo[5.4.0]undec-7-  
15 ene the term Aliquat refers to Trioctylmethylammonium chloride, Adogen refers to Methyltrialkyl(C<sub>8</sub>-C<sub>10</sub>)ammonium chloride, the term PEG refers to Polyethylene glycol, the term TDA refers to 1-Tris(3,6-dioxaheptyl)amine.

The term Lindlar catalyst refers to 5.0 % Palladium on calcium carbonate (poisoned with lead), The term DMAP refers to Dimethylamino pyridine, Co- factors include, for  
20 example, nicotinamide co-factors such as nicotinamide adenine dinucleotide ("NAD"), reduced nicotinamide adenine dinucleotide ("NADH"), nicotinamide adenine dinucleotide phosphate ("NADP<sup>+</sup>"), reduced nicotinamide adenine dinucleotide phosphate ("NADPH").

The inventors have developed a process for the preparation of compound of  
25 Formula (I) either in its racemic (R/S) form or any of its optically active (S) or (R) forms or enantiomeric excess mixture of any of the forms. The process includes:

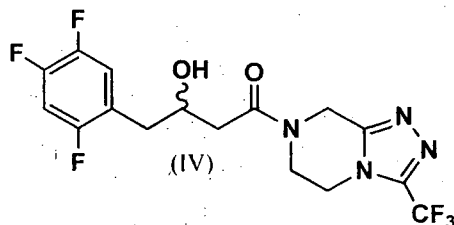
a) reacting 4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one of formula (V) or its metal ion salt



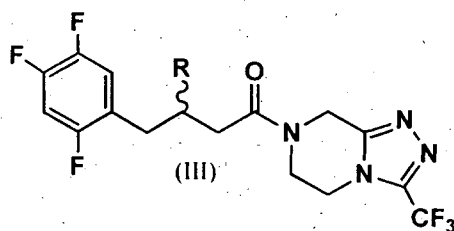
Formula (V)

with suitable enzymes or their variants that reduce a ketone to form an alcohol, by  
 5 maintaining under suitable conditions, to obtain 3-hydroxy-1-(3-(trifluoromethyl)-5,6-  
 dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl) butan-1-one  
 of compound of formula (IV), in racemic (R/S) form or any of its optically active (S)  
 or (R) forms or their enantiomerically excess mixtures;

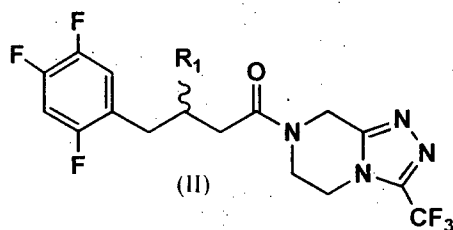
(b) reacting 3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-  
 10 a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl) butan-1-one of Formula (IV) either in its  
 racemic (R/S) form or any of its optically active (S) or (R) forms or enantiomeric  
 excess mixture of any of the forms of Formula (IV)



with suitable reagents that activate the hydroxyl group, by maintaining under suitable  
 15 conditions, to obtain compound of Formula (III) wherein 'R' represents a suitable  
 leaving group such as carboxylate, sulfonate, phosphate and oxyphosphonium ion etc.  
 with retention of configuration;



(c) optionally, isolating the compound of Formula (III);  
 20 (d) converting compound of Formula (III) to compound of Formula (II), wherein R<sub>1</sub> is  
 selected from an azido group, cyclic imido group, aralkylamino group and sulfonamido  
 group with inversion of configuration, by using suitable amine equivalent optionally  
 with additives in suitable solvents;



(e) suitably deprotecting or reducing the compound of Formula (II) or employing suitable chemistry to obtain compound of Formula (I) having  $\text{NH}_2$  group with retention of configuration;

5 (f) optionally, the compound of Formula (IV) is directly converted to the compound of Formula (II) with inversion of configuration, by using suitable amine equivalent, optionally with suitable additives in suitable solvents.

(g) optionally, the compound of Formula (III) may be directly converted to the compound of Formula (I) with inversion of configuration, by using suitable amine  
10 equivalent.

The process is further exemplified below:

**Step-(a)**

In general, the reaction of compound of Formula (V) or its suitable metal ion salts with suitable enzyme to give compound of Formula (IV) in racemic (R/S) form or  
15 any of its optically active (S) or (R) forms or their enantiomerically excess mixtures can be carried out using suitable polypeptides.

The metal ion salt of compound of formula (V) is selected from Na, K, Li, Mg, Ca, Zn, Cu and like.

In one embodiment of the present invention the polypeptide having desired  
20 enzymatic activity and variants thereof can be isolated from suitable bacteria, yeast or fungi. In one embodiment the suitable polypeptides having enzymatic activities are selected from oxidoreductases. In a preferred embodiment suitable enzymes are selected from aldo-keto reductases. In another embodiment, suitable enzymes are selected from dehydrogenases. In an embodiment the  $\text{NAD(P)}^+$  dependent reductase is  
25 selected from the *Saccharomyces species*. In an another preferred embodiment  $\text{NAD(P)}^+$  dependent reductase is selected (derived) from *Saccharomyces cerevisiae* and having Genebank id:- NP\_012630.1. In an another preferred embodiment  $\text{NAD(P)}^+$  dependent alcohol dehydrogenase is selected (derived) from *Saccharomyces cerevisiae* and having Genebank id:- NP\_013953.1, NP\_014763.1, NP\_011972.1, NP\_014068 and  
30 NP\_011330..

In a preferred embodiment, the 3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl) butan-1-one of compound of formula (IV), in racemic (R/S) form or any of its optically active (S) or (R) forms or their enantiomerically excess mixtures was prepared as per method similar to that disclosed in patent application No. 2805/MUM/2010.

#### **Step-(b)**

In general, the reaction of compound of Formula (IV) with suitable reagent to give compound of Formula (III) having a suitable leaving group 'R' as defined earlier, with retention of configuration, may be carried out using a suitable reagents, suitable base and suitable solvents.

Suitable reagents which can be used in step (b) may include one or more of suitable acid chlorides such as acetyl chloride, benzoyl chlorides and substituted benzoyl chloride such as p-nitrobenzoyl chloride and the like; anhydrides such as acetic anhydride, trifluoroacetic anhydride, triflic anhydride and the like; sulfonyl chlorides such as , 2,2,2-trifluoroethanesulfonyl chloride, benzenesulfonyl chloride, substituted benzenesulfonyl chlorides such as p-toluenesulfonyl chloride, p-nitro benzenesulfonyl chlorides and the like; acid chloro phosphates such as diethyl chloro phosphate and the like.

The compound of Formula (III) wherein 'R' represents suitable leaving group as defined earlier, was prepared with retention of configuration.

In one embodiment, the compound of Formula (III), wherein when 'R' represents a caboxylate leaving group such as  $-\text{OCOCH}_3$ ,  $-\text{OCOCF}_3$ ,  $-\text{OC(O)OEt}$ ,  $-\text{OCOPh}$ , substituted  $-\text{OCOPh}$  etc. was prepared with retention of configuration from the compound of formula (IV) by reacting with suitable acid chlorides such as acetyl chloride, benzoyl chlorides and substituted benzoyl chloride such as p-nitrobenzoyl chloride and the like; anhydrides such as acetic anhydride, trifluoroacetic anhydride, triflic anhydride and the like in suitable solvents in the presence of a base.

In another embodiment, the compound of Formula (III), wherein 'R' represents suitable sulfonate leaving group such as  $-\text{OSO}_2\text{CF}_3$ ,  $-\text{OSO}_2\text{CH}_2\text{CF}_3$ ,  $-\text{OSO}_2\text{Ph}$ , substituted  $-\text{OSO}_2\text{Ph}$ , etc. was prepared with retention of configuration from the compound of formula (IV) by reacting with suitable sulfonyl chlorides such as methanesulfonyl chloride, 2,2,2-trifluoroethanesulfonyl chloride, benzenesulfonyl chloride and substituted benzenesulfonyl chlorides such as p-toluenesulfonyl chloride,

p-nitro benzenesulfonyl chlorides and the like, in suitable solvents in presence of a base.

In another embodiment, the compound of Formula (III), wherein 'R' represents suitable phosphate leaving group such as  $-OP(O)(OEt)_2$  was prepared with retention of configuration from the compound of formula (IV) by reacting with suitable chloro phosphates such as diethyl chloro phosphate in suitable solvents in the presence of a base with or without suitable additives.

Suitable solvents which can be used at step-(b), when 'R' represents a caboxylate, sulfonate and phosphate may include one or more of ethers such as diethyl ether, 1,4-dioxane, dimethoxy ethane, DIPE, MTBE, THF; chlorinated solvents such as chloroform, dichloromethane, dichloroethane; nitriles such as acetonitrile; hydrocarbons such as hexane, heptane, cyclohexane, toluene, xylene, chlorobenzene; aprotic polar solvents such as DMF, DMA; N-methyl pyrrolidone, pyridine and the like or their suitable mixtures.

Suitable bases which can be used at step-(b), when 'R' represents a caboxylate, sulfonate and phosphate, may be selected from  $C_{(1-5)}$  alkyl amines,  $C_{(1-5)}$  substituted alkyl amines such as triethyl amine (TEA), diisopropyl amine, diisopropylethyl amine, heterocyclic saturated or unsaturated amines, preferably morpholine, piperidine, pyrrolidine, DBU, DBN, 1,4-diazabicyclo[2,2,2]octane, dimethylamino pyridine and pyridine; hydrides such as NaH; n-BuLi, LDA and KHMDS and the like or mixtures thereof.

The reaction mixture in step (b) is maintained at  $-10$  to  $70$  °C. Preferably, the temperature is at  $-10$  to  $50$  °C.

Optionally, the reaction of step-(b) may be carried out in an inert atmosphere such as  $N_2$ , He or Ar. The duration of the reaction may be from 1 to 24 hrs, more specifically 2 to 8 hrs.

In another embodiment, the compound of Formula (III) was prepared with retention of configuration, wherein 'R' represents the oxyphosphonium ion leaving group [e.g.  $-O^+PPh_3$ ], which is generated in situ using phosphines with azodicarboxylates or azoamides, from the compound of Formula (IV) in suitable solvents (Mitsunobu reaction).

An oxyphosphonium ion leaving group can be generated in situ by reacting suitable phosphines such as, trialkylphosphine such as tributylphosphine, tri-*t*-butylphosphine, trihexylphosphine, trioctylphosphine, cyanomethylenetri-*n*-

butylphosphorane; cycloalkylphosphine such as tricyclohexylphosphine, dicyclohexylphenylphosphine, or arylphosphine such as triphenylphosphine, dicyclohexylphenylphosphine, diethylphenylphosphine, 4-(dimethylamino)phenyldiphenylphosphine, diphenyl-2-pyridylphosphine, isopropyldiphenylphosphine with azodicarboxylates such as diethylazodicarboxylate, diisopropyl azodicarboxylate, dimethyl azodicarboxylate, dibenzyl azodicarboxylate, bis(2,2,2-trichloroethyl)azodicarboxylate or with azoamides such as 1,1'-(azodicarbonyl)dipiperidine, N,N,N',N'-tetramethylazodicarboxamide (Mitsunobu reaction) by techniques known in the art.

10 Suitable solvents which can be used in Mitsunobu reaction of step-(b) may include one or more of ethers such as diethyl ether, 1,4-dioxane, dimethoxy ethane, diethoxy ethane, DIPE, MTBE, THF, 2-methyl tetrahydrofuran or their suitable mixtures.

The reaction mixture in step (b) is maintained at -10 to 50 °C. Preferably, the temperature is at -10 to 30 °C.

15 Optionally, the reaction of step-(b) may be carried out in an inert atmosphere such as N<sub>2</sub>, He or Ar. The duration of the reaction may be from 1 to 24 hrs, more specifically 2 to 8 hrs.

#### Step-(c)

20 The compound of Formula (III), wherein 'R' represents a leaving group as defined earlier, can be isolated or it may be generated in situ.

#### Step-(d)

The conversion of Formula (III) to compound of Formula (II) where R<sub>1</sub> is as defined earlier with inversion of configuration, may be carried out using a suitable amine equivalent, in presence of suitable solvents and optionally in the presence of suitable additives.

Suitable amine equivalent which can be used in step-(d) may include -N<sub>3</sub> (azide), cyclic imides, araalkylamine, sulfonamide and the like.

Suitable azides which can be used for preparation of compounds of formula (II) may be selected from HN<sub>3</sub>, NaN<sub>3</sub>, LiN<sub>3</sub>, Zn(N<sub>3</sub>)<sub>2</sub>, Cu(N<sub>3</sub>)<sub>2</sub>, Tetrabutylammonium azide etc.; Suitable cyclic imides which can be used for preparation of compounds of formula (II) may be selected from phthalimide, succinimide etc.; Suitable araalkylamine which can be used for preparation of compounds of formula (II) may be selected from benzylamine; p-methoxybenzylamine, α-methylphenethylamine, tritylamine etc.;

Suitable sulfonamide which can be used for preparation of compounds of formula (II) may be selected from p-toluene sulfonamide, sulfamic acid, N-(t-butoxycarbonyl)-p-toluenesulfonamide and the like.

In an embodiment, the compound of Formula (II) where  $R_1$  is  $-N_3$  can be obtained by reacting the compound of formula (III) with suitable azidating agents like,  $HN_3$ ,  $NaN_3$ ,  $LiN_3$ ,  $Zn(N_3)_2$ ,  $Cu(N_3)_2$ , Tetrabutylammonium azide etc. in suitable solvents.

Suitable solvents which can be used in step-(d), where  $R_1$  is  $-N_3$  may include one or more of ethers such as 1,4-dioxane, dimethoxy ethane, THF; esters such as ethyl acetate, n-butyl acetate and isopropyl acetate; chlorinated solvents such as chloroform, dichloromethane, dichloroethane; hydrocarbons such as hexane, heptane, cyclohexane, toluene, xylene, chlorobenzene; aprotic polar solvents such as DMF, DMA, DMSO; N-methyl pyrrolidone, pyridine, HMPA and the like, water or their suitable mixtures.

In an embodiment, the compound of Formula (II) where  $R_1$  is suitable cyclic imide can be obtained by reacting the compound of Formula (III) with cyclic imides like phthalimide, succinimide etc. in suitable solvents.

In another embodiment, the compound of Formula (II) where  $R_1$  is an aralkylamine can be obtained by reacting the compound of Formula (III) with aralkylamine like benzylamine, p-methoxybenzylamine,  $\alpha$ -methylphenethylamine, tritylamine, etc. in suitable solvents.

The compound of Formula (II) where  $R_1$  is sulfonamide can be obtained by reacting the compound of Formula (III) with suitable sulfonamides like p-toluene sulfonamide, sulfamic acid, N-(t-butoxycarbonyl)-p-toluenesulfonamide etc. in suitable solvents.

The suitable solvents, where  $R_1$  is cyclic imide, aralkylamine, sulfonamide may be selected from ethers such as diethyl ether, 1,4-dioxane, dimethoxy ethane, THF; chlorinated solvents such as chloroform, dichloromethane, dichloroethane; nitriles such as acetonitrile; hydrocarbons such as hexane, heptane, cyclohexane, toluene, xylene, chlorobenzene; aprotic polar solvents such as DMF, DMA, DMSO; N-methyl pyrrolidone, HMPA, pyridine and the like, water or their suitable mixtures.

Optionally, suitable additives may be added in step (d) to enhance the conversion. Such additives may be selected from those known in the art, such as suitable phase transfer catalysts like, trialkyl ammonium halides, tetraalkyl ammonium halides, trialkyl ammonium acetates, tetraalkyl ammonium acetates, trialkyl ammonium hydroxides, tetraalkyl ammonium hydroxides, trialkyl ammonium sulfates, tetraalkyl



ammonium sulfates, trialkyl ammonium tetrafluoroborate, tetraalkyl ammonium tetrafluoroborate, trialkyl phosphonium halides, tetraalkyl phosphonium halides, tetrapenyl phosphonium halides, pyridinium halides, Aliquat, Adogen, crown ethers, silicon analogues of crown ethers, PEG, TDA-1 etc.

5 The reaction mixture in step (d) is maintained at -10 to 100 °C. Preferably, the temperature is at -10 to 30 °C.

Optionally, the reaction of step-(d) may be carried out in an inert atmosphere such as N<sub>2</sub>, He or Ar. The duration of the reaction may be from 1 to 48 hrs, more specifically 2 to 24 hrs.

#### 10 Step-(e)

The compound of formula (I) can be obtained from the compound of formula (II) either by reduction and/or deprotection or employing suitable chemistry as discussed herein under.

The compound of Formula (I) is prepared with retention of configuration by reduction  
15 of the compound of Formula (II) when R<sub>1</sub> is -N<sub>3</sub>. The reduction of compound of formula (II) when R<sub>1</sub> is -N<sub>3</sub> may be carried out using suitable reagents and suitable solvents.

Reduction of the compound of Formula (II) when R<sub>1</sub> is -N<sub>3</sub> by standard techniques reported in the literature provides the compound of Formula (I). The  
20 suitable reagent may be selected from precious metal catalyst like, Pd/H<sub>2</sub>, Pt/H<sub>2</sub>, Pd/Al<sub>2</sub>O<sub>3</sub>, Pd-CaCO<sub>3</sub>, Pd(OH)<sub>2</sub>, PdO, PtO<sub>2</sub>, Pd-C/HCOOH, Pd-C/HCOONH<sub>4</sub>, Pd(OH)<sub>2</sub>-C/N<sub>2</sub>H<sub>4</sub>, Pd-C/NaBH<sub>4</sub>, Lindlar catalyst; Metal catalyst like, Mg, Ca, Zn, Zn-HCl, Zn-CH<sub>3</sub>COOH, SnCl<sub>2</sub>, SnCl<sub>2</sub>/thiophenol, Zn/NH<sub>4</sub>Cl, Zn/AlCl<sub>3</sub>; Boron containing reducing agents like, BH<sub>3</sub>, NaBH<sub>4</sub>, LiAlH<sub>4</sub>, Zn(BH<sub>4</sub>)<sub>2</sub>, HBCl<sub>2</sub>-Me<sub>2</sub>S, LiBH<sub>3</sub>NMe<sub>2</sub>,  
25 NaBH<sub>4</sub>/Phase Transfer Catalyst, NaBH<sub>4</sub>-exchange resin, NaBH<sub>4</sub>/NiCl<sub>2</sub>·6H<sub>2</sub>O, NaBH<sub>4</sub>/Ni(OAc)<sub>2</sub>, NaBH<sub>4</sub>/1,3-propanedithiol; sulfur compounds like HS(CH<sub>2</sub>)<sub>3</sub>SH/Et<sub>3</sub>N, PhSH; Phosphorous compounds like PPh<sub>3</sub>/NH<sub>4</sub>OH; (See for refs. (I) Comprehensive organic transformations, 2nd Edition, 815-819, Publisher-WILEY-VCH, By-Richard C. Larock. (II) Comprehensive organic Synthesis Vol. 8-Reduction,  
30 383-386, Publisher- Pergamon Press, By-Barry M. Trost and Ian Fleming. (III) Handbook of reagents for organic synthesis-Oxidizing and Reducing agents, 6, John Wiley & Sons, By-S.D. Burke and R. L. Danheiser).

Suitable solvents which can be used at step-(e) may include one or more of alcohols such as methanol, ethanol, isopropanol, butanol, 1,2-dimethoxy ethanol, 2-

methoxy ethanol, 2-ethoxy ethanol and ethylene glycol; ethers such as diethyl ether, 1,4-dioxane, dimethoxy ethane, DIPE, MTBE, THF and the like, water or their suitable mixtures.

The reaction mixture in step (e) is maintained at 0 to 80 °C. Preferably, the  
5 temperature is at 20 to 40 °C.

Optionally, the reaction of step-(e) may be carried out in an inert atmosphere such as N<sub>2</sub>, He or Ar. The duration of the reaction may be from 1 to 48 hrs, more specifically 5 to 24 hrs.

Deprotection of the protected compound of Formula (II) when R<sub>1</sub> is cyclic  
10 imido group, araalkylamino group, carbamate group, hydroxyl amino group, alkenylamino group, sulfonamido group etc. as defined earlier, may be carried out by standard techniques reported in the literature provides the compound of formula (I). (See for e.g. Protection and Deprotection of amines in Text book -Title: Protective Groups in Organic Synthesis, 3rd Edition, John Wiley & Sons, By-T.W. Greene and  
15 Peter G. M. Wuts).

#### **Step-(f)**

The conversion of Formula (IV) to compound of Formula (II) having group R<sub>1</sub> as defined earlier with inversion of configuration may be carried out in one step using a suitable amine equivalent with suitable reagents, suitable solvents and optionally in the  
20 presence of suitable additives.

Suitable amine equivalent used may be selected from cyclic imides, araalkylamine, etc.

The compound of formula (II) where R<sub>1</sub> is cyclic imides can be obtained by reacting the compound of formula (IV) with cyclic imides via oxyphosphonium ion formation which is generated in situ using suitable phosphines with azodicarboxylates  
25 or azoamides (Mitsunobu reaction). The cyclic imides like phthalimide, succinimide can be used.

The compound of formula (II) where R<sub>1</sub> is aralkylamine can be obtained by reacting the compound of formula (IV) with suitable aralkylamines via oxyphosphonium ion formation which is generated in situ using suitable phosphines  
30 with azodicarboxylates or azoamides (Mitsunobu reaction). The araalkylamine like benzylamine, p-methoxybenzylamine, α-methylphenethylamine, tritylamine, etc can be used.

The compound of Formula (II) where R<sub>1</sub> is sulfonamide can be obtained by reacting the compound of Formula (III) with suitable sulfonamides like p-toluene

sulfonamide, sulfamic acid, N-(t-butoxycarbonyl)-p-toluenesulfonamide etc. in suitable solvents.

An oxyphosphonium ion leaving group can be generated in situ by reacting trialkylphosphine such as tributylphosphine, tri-t-butylphosphine, trihexylphosphine, 5 trioctylphosphine, cyanomethylenetri-n-butylphosphorane; cycloalkylphosphines such as tricyclohexylphosphine, dicyclohexylphenylphosphine, or arylphosphine such as triphenylphosphine, dicyclohexylphenylphosphine, diethylphenylphosphine, 4-(dimethylamino)phenyldiphenylphosphine, diphenyl-2-pyridylphosphine, isopropyldiphenylphosphine; with azodicarboxylates such as diethylazodicarboxylate, 10 diisopropyl azodicarboxylate, dimethyl azodicarboxylate, dibenzyl azodicarboxylate, bis(2,2,2-trichloroethyl)azodicarboxylate or with azoamides such as 1,1'-(azodicarbonyl)dipiperidine, N,N,N',N'-tetramethylazodicarboxamide (Mitsunobu reaction).

Suitable solvents which can be used in Mitsunobu reaction of step-(f) may include one 15 or more of ethers such as diethyl ether, 1,4-dioxane, dimethoxy ethane, diethoxy ethane, DIPE, MTBE, THF, 2-methyl tetrahydrofuran or their suitable mixtures.

The reaction mixture in step (f) is maintained at -10 to 50 °C. Preferably, the temperature is at -10 to 30 °C.

Optionally, the reaction of step-(f) may be carried out in an inert atmosphere 20 such as N<sub>2</sub>, He or Ar. The duration of the reaction may be from 1 to 24 hrs, more specifically 2 to 8 hrs.

Optionally, suitable additives can be selected from phase transfer catalysts like, trialkyl ammonium halides, tetraalkyl ammonium halides, trialkyl ammonium acetates, tetraalkyl ammonium acetates, trialkyl ammonium hydroxides, tetraalkyl ammonium 25 hydroxides, trialkyl ammonium sulfates, tetraalkyl ammonium sulfates, trialkyl ammonium tetrafluoroborate, tetraalkyl ammonium tetrafuloroborate, trialkyl phosphonium halides, tetraalkyl phosphonium halides, tetraphenyl phosphonium halides, pyridinium halides, aliquat, adogen, crown ethers, silicon analogues of crown ethers, PEG, TDA-1 etc.

### 30 Step-(g)

The compound of Formula (III) can be converted directly to compound of Formula (I) with inversion of configuration using ammonia or suitable ammonium salts in suitable solvents and optionally in the presence of suitable additives and under pressure.

Suitable ammonium salt which can be used at step-(f) may include one or more of ammonium acetate, ammonium carbonate, ammonium hydroxide, ammonium formate, ammonium lactate, ammonium citrate dibasic, ammonium carbamate, ammonium benzoate.

- 5 Suitable solvents which can be used at step-(g) may include one or more of alcohols such as methanol, ethanol, isopropanol, butanol, 1,2-dimethoxy ethanol, 2-methoxy ethanol, 2-ethoxy ethanol and ethylene glycol; ethers such as diethyl ether, 1,4-dioxane, dimethoxy ethane, DIPE, MTBE, THF and the like, water or their suitable mixtures.

The reaction mixture in step (g) is maintained at 0 to 80 °C. Preferably, the  
10 temperature is at 20 to 40 °C.

Optionally, the reaction of step-(g) may be carried out in an inert atmosphere such as N<sub>2</sub>, He or Ar. The duration of the reaction may be from 1 to 48 hrs, more specifically 5 to 24 hrs.

Optionally, suitable additives may be added to enhance the conversion. Such additives  
15 may be selected from those known in the art, such as suitable phase transfer catalysts like, trialkyl ammonium halides, tetraalkyl ammonium halides, trialkyl ammonium acetates, tetraalkyl ammonium acetates, trialkyl ammonium hydroxides, tetraalkyl ammonium hydroxides, trialkyl ammonium sulfates, tetraalkyl ammonium sulfates, trialkyl ammonium tetrafluoroborate, tetraalkyl ammonium tetrafuloroborate, trialkyl  
20 phosphonium halides, tetraalkyl phosphonium halides, tetrapenyl phosphonium halides , pyridinium halides, Aliquat , Adogen, crown ethers, silicon analogues of crown ethers, PEG, TDA-1 etc.

The product so obtained may be further or additionally purified by suitable salt formation, diastereomeric salt formation or by purification using suitable solvents to  
25 obtain desired purity levels.

The compound of Formula (I) thus obtained, is converted to its corresponding acid addition salts by reacting with corresponding acids in suitable solvents.

The suitable solvents used for salt preparation may be selected from alcohols such as methanol, ethanol, isopropanol, butanol, 1,2-dimethoxy ethanol, 2-methoxy ethanol, 2-  
30 ethoxy ethanol, ethylene glycol and the like; hydrocarbons such as hexane, cyclohexane, toluene, chloro benzene and like; esters such as ethyl acetate, isopropyl acetate & the like; nitriles such as acetonitrile & the like; ethers such as, DIPE, 1,4-dioxane, THF & the like; ketones such as acetone, MIBK and like; aprotic polar

solvents such as DMF, DMSO, DMA & the like; water and suitable mixtures of one or more of the solvents described above.

The suitable acid used may be selected from phosphoric acid, HCl, benzenesulfonic acid, p-toluenesulfonic acid, 10-camphorsulfonic acid, tartaric acid, 5 dodecylsulfuric acid, citric acid, maleic acid, fumaric acid and the like.

The compound of Formula (I) thus obtained, is converted to its corresponding diastereomeric salt salts by reacting with suitable chiral acids in suitable solvents.

Suitable chiral acids used for the diastereomeric salt formation may be selected from tartaric acid, di-p-toluoyl tartaric acid, dibenzoyltartaric acid, o-nitrobenzoyl 10 tartaric acid, lactic acid, 10-camphorsulfonic acid, 8-camphorsulfonic acid, malic acid, N-acetyl glutamic acid, mandelic acid, o-acetylmandelic acid, o-methylmandelic acid, thiazolidine-4-carboxylic acid,  $\alpha$ -methoxyphenylacetic acid,  $\alpha$ -methoxy- $\alpha$ -trifluoromethylphenylacetic acid, 2-pyrrolidone-5-carboxylic acid and the like. Preferably the chiral acid used for resolution may be selected from tartaric acid and 15 dibenzoyltartaric acid.

The suitable solvents used used for the diastereomeric salt formation may be selected from suitable alcohols like methanol, ethanol, isopropanol, butanol, 1,2-dimethoxy ethanol, 2-methoxy ethanol, 2-ethoxy ethanol, isoamyl alcohol and ethylene glycol; esters like ethyl acetate and isopropyl acetate; chlorinated solvents like 20 chloroform and dichloromethane; nitriles like acetonitrile; hydrocarbons like toluene, xylene and chlorobenzene; ketones like acetone, methyl ethyl ketone; ethers like diethyl ether, 1,4-dioxane, DIPE, MTBE, THF; aprotic polar solvents such as DMF, DMSO, DMA; water and their suitable mixtures.

Suitable diastereomeric salt obtain above may be further converted to its pure base by 25 employing standard techniques in the art.

The compound of Formula (I) and its salt thus obtained, is purified by using suitable solvents.

The suitable solvents used for the purification may be selected from alcohols like methanol, ethanol, isopropanol, butanol, 1,2-dimethoxy ethanol, 2-methoxy 30 ethanol, 2-ethoxy ethanol, ethylene glycol and the like; esters like ethyl acetate, isopropyl acetate & the like; chlorinated solvents like chloroform, dichloromethane & the like; nitriles like acetonitrile & the like; hydrocarbons like toluene, xylene, chlorobenzene & the like; ketones like acetone & the like; ethers like diethyl ether, 1,4-dioxane, DIPE, MTBE, THF & the like; aprotic polar solvents such as DMF, DMSO,

DMA & the like; water and suitable mixtures of one or more of the solvents described above.

The pure compound of Formula (I) thus obtained, is converted to its pharmaceutically acceptable salts such as acetate, benzenesulfonate, benzoate, camphorsulfonate, citrate, ethanesulfonate, fumarate, gluconate, glutamate, HBr, HCl, isethionate, lactate, maleate, malicate, mandelate, methanesulfonate, mucicate, nitrate, pamoate, pantothenate, phosphate, succinate, sulfate, tartarate, p-toluenesulfonate, HI, oxalate, galactarate, Hemi-L-malate, D-gluconate, succinate, thiocyanate, aspartate, ethane disulfonate, pyroglutamate, glutarate, glycolate, L-Malate.

All the compounds are analysing by standard analytical techniques.

**Analytical methods:**

i) The infrared (IR) spectrum has been recorded on a Shimadzu FTIR-8400 model spectrophotometer, between  $450\text{ cm}^{-1}$  and  $4000\text{ cm}^{-1}$ , with a resolution of  $4\text{ cm}^{-1}$  in a KBr pellet.

ii) (2R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]-triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine and its salts were analyzed for purity by analytical HPLC at  $\lambda_{\text{max}}$  210nm using column YMC-C8, 250 mm x 4.6mm x 4 mm or its equivalent on AGILENT 1100 series under the following conditions,

Detector: UV absorption photometer Wave length: 210 nm Column temp. : 25 °C

Flow rate: 1.0 mL/min. Injection Vol.: 10  $\mu\text{L}$

Mobile Phase: 10mM  $\text{KH}_2\text{PO}_4$  (pH-6.8): Acetonitrile (55:45)

(2R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]-triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine and its salts were analyzed for chiral purity by HPLC at  $\lambda_{\text{max}}$  268 nm using column Chiral-Cel OJ-H, 250mm x 4.6mm x 5 $\mu$  or its equivalent on Shimadzu LCVP model under the following conditions,

Detector: UV absorption photometer Wave length: 268 nm Column temp. : 35 °C

Flow rate: 0.8 mL/min. Injection Vol.: 10  $\mu\text{L}$

Mobile Phase: 0.1 % diethyl amine in [n-Hexane: Ethanol (90:10)]

iii) Melting points were taken on VEEGO make model VMP-D melting point apparatus and are uncorrected.

The inventors have developed a process for the preparation of compound of Formula (I) or its salts either in its racemic (R/S) form or any of its optically active (S) or (R) forms or enantiomeric excess mixture of any of the forms. The process includes:

- (i) reacting 3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl) butan-1-one of Formula (IV) either in its racemic (R/S) form or any of its optically active (S) or (R) forms or enantiomeric excess mixture of any of the forms with a p-toluenesulfonyl chloride or p-nitro benzenesulfonyl chloride to obtain compound of Formula (III) wherein 'R' represents p-toluenesulfonyl group or p-nitro benzenesulfonyl group, with retention of configuration;
- (ii) optionally, isolating the compound of Formula (III) wherein 'R' represents p-toluenesulfonyl group or p-nitro benzenesulfonyl group;
- (iii) converting compound of Formula (III) wherein 'R' represents p-toluenesulfonyl group or p-nitro benzenesulfonyl group to compound of Formula (II), wherein 'R<sub>1</sub>' represents -N<sub>3</sub> group with inversion of configuration by using suitable azides, optionally with suitable additives in suitable solvents;
- (iv) reducing the compound of Formula (II) wherein 'R<sub>1</sub>' represents -N<sub>3</sub> group to obtain compound of Formula (I) with retention of configuration.

Suitable solvents which can be used at step-(i), when 'R' represents p-toluenesulfonyl chloride or p-nitro benzenesulfonyl chloride may include one or more of chlorinated solvents such as chloroform, dichloromethane, dichloroethane; nitriles such as acetonitrile; hydrocarbons such as hexane, heptane, cyclohexane, toluene, xylene, chlorobenzene; aprotic polar solvents such as DMF, DMA; N-methyl pyrrolidone, pyridine and the like or their suitable mixtures.

Suitable bases which can be used at step-(i), when 'R' represents p-toluenesulfonyl chloride or p-nitro benzenesulfonyl chloride, may be selected from heterocyclic saturated or unsaturated amines, preferably morpholine, piperidine, pyrrolidone, DBU, DBN, 1,4-diazabicyclo[2,2,2]octane, dimethylamino pyridine and pyridine; hydrides such as KHMDS and the like or mixtures thereof.

Suitable azides which can be used at step-(iii), for preparation of compounds of formula (II) may be selected from HN<sub>3</sub>, NaN<sub>3</sub>, LiN<sub>3</sub>, Zn(N<sub>3</sub>)<sub>2</sub>, Cu(N<sub>3</sub>)<sub>2</sub>, Tetrabutylammonium azide etc.;

Suitable additives may be added in step (iii) to enhance the conversion. Such additives may be selected from those known in the art, such as suitable phase transfer catalysts like, trialkyl ammonium halides, tetraalkyl ammonium halides, trialkyl ammonium acetates, tetraalkyl ammonium acetates, trialkyl ammonium hydroxides, tetraalkyl ammonium hydroxides, trialkyl ammonium sulfates, tetraalkyl ammonium

sulfates, trialkyl ammonium tetrafluoroborate, tetraalkyl ammonium tetrafluoroborate, trialkyl phosphonium halides, tetraalkyl phosphonium halides, tetrapenyl phosphonium halides, pyridinium halides, Aliquat, Adogen, crown ethers, silicon analogues of crown ethers, PEG, TDA-I etc.

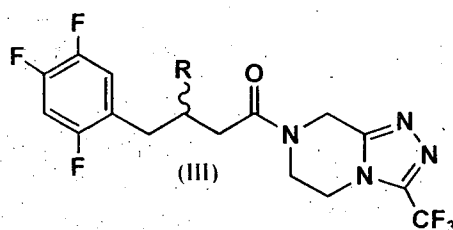
5            Suitable solvents which can be used in step-(iii), where  $R_1$  is  $-N_3$  may include one or more of ethers such as 1,4-dioxane, dimethoxy ethane, THF; esters such as ethyl acetate, n-butyl acetate and isopropyl acetate; chlorinated solvents such as chloroform, dichloromethane, dichloroethane; hydrocarbons such as hexane, heptane, cyclohexane, toluene, xylene, chlorobenzene; aprotic polar solvents such as DMF, DMA, DMSO; N-  
10 methyl pyrrolidone, pyridine, HMPA and the like, water or their suitable mixtures.

          The suitable reducing reagent can be used in step-(iv) may be selected from precious metal catalyst like, Pd/H<sub>2</sub>, Pt/H<sub>2</sub>, Pd/Al<sub>2</sub>O<sub>3</sub>, Pd-CaCO<sub>3</sub>, Pd(OH)<sub>2</sub>, PdO, PtO<sub>2</sub>, Pd-C/HCOOH, Pd-C/HCOONH<sub>4</sub>, Pd(OH)<sub>2</sub>-C/N<sub>2</sub>H<sub>4</sub>, Pd-C/NaBH<sub>4</sub>; Lindlar catalyst; Metal catalyst like, Mg, Ca, Zn, Zn-HCl, Zn-CH<sub>3</sub>COOH, SnCl<sub>2</sub>, SnCl<sub>2</sub>/thiophenol,  
15 Zn/NH<sub>4</sub>Cl, Zn/AlCl<sub>3</sub>; Boron containing reducing agents like, BH<sub>3</sub>, NaBH<sub>4</sub>, LiAlH<sub>4</sub>, Zn(BH<sub>4</sub>)<sub>2</sub>, HBCl<sub>2</sub>-Me<sub>2</sub>S, LiBH<sub>3</sub>NMe<sub>2</sub>, NaBH<sub>4</sub>/Phase Transfer Catalyst, NaBH<sub>4</sub>-exchange resin, NaBH<sub>4</sub>/NiCl<sub>2</sub>.6H<sub>2</sub>O, NaBH<sub>4</sub>/Ni(OAc)<sub>2</sub>, NaBH<sub>4</sub>/1,3-propanedithiol; sulfur compounds like HS(CH<sub>2</sub>)<sub>3</sub>SH/Et<sub>3</sub>N, PhSH; (See for refs. (I) Comprehensive organic transformations, 2nd Edition, 815-819, Publisher-WILEY-VCH, By-Richard C.  
20 Larock. (II) Comprehensive organic Synthesis Vol. 8-Reduction, 383-386, Publisher-Pergamon Press, By-Barry M. Trost and Ian Fleming. (III) Handbook of reagents for organic synthesis-Oxidizing and Reducing agents, 6, John Wiley & Sons, By-S.D. Burke and R. L. Danheiser).

          Suitable solvents which can be used at step-(iv) may include one or more of  
25 alcohols such as methanol, ethanol, isopropanol, butanol, 1,2-dimethoxy ethanol, 2-methoxy ethanol, 2-ethoxy ethanol and ethylene glycol; ethers such as diethyl ether, 1,4-dioxane, dimethoxy ethane, DIPE, MTBE, THF and the like, water or their suitable mixtures.

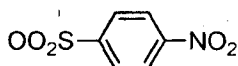
          In another aspect there is provided a novel intermediate of Formula (III) in  
30 racemic (R/S) form or any of its optically active (S) or (R) forms or their enantiomerically excess mixtures which can be used in the preparation of the compound of Formula (I).





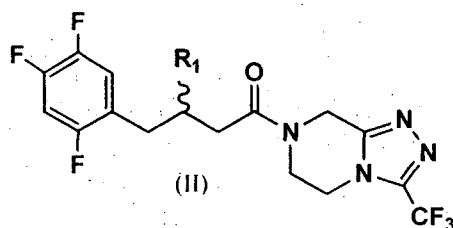
Wherein 'R' represents a suitable leaving group such as carboxylate, sulfonate, phosphate and oxyphosphonium ion etc.

In one of the preferred embodiments are provided the compound of formula (III) where 'R' represents suitable alkyl carboxylate group such as  $-\text{OCOCH}_3$ ; haloalkyl carboxylate group such as  $-\text{OCOCF}_3$ ; aryl carboxylate group such as  $-\text{OCOPh}$ , substituted  $-\text{OCOPh}$ ; alkoxy carboxylate group such as  $-\text{OC(O)OCH}_2\text{CH}_3$ ; alkyl sulfonates group such as  $-\text{OSO}_2\text{CH}_2\text{CH}_3$ ;  $-\text{haloalkyl sulfonate}$  group such as,  $-\text{OSO}_2\text{CF}_3$ ,  $-\text{OSO}_2\text{CH}_2\text{CF}_3$ ; aryl sulfonates group such as  $-\text{OSO}_2\text{Ph}$ , substituted  $-\text{OSO}_2\text{Ph}$  such as



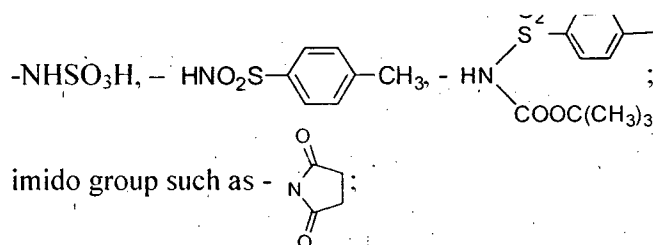
alkoxy phosphates group such as  $-\text{OP(O)(OEt)}_2$ ; an oxyphosphonium ion leaving group such as  $-\text{O}^+\text{P(X)}_3$  where 'X' may be alkyl groups, aryl groups, cycloalkyl groups.

In another aspect there is provided a novel intermediate of Formula (II) in racemic (R/S) form or any of its optically active (S) or (R) forms or their enantiomerically excess mixtures, which can be used in the preparation of the compound of Formula (I).



wherein 'R<sub>1</sub>' represent cyclic imido group, aralkylamino group, sulfonamido group.

In one of the preferred embodiments are provided the compounds of Formula (II) where 'R<sub>1</sub>' represents aralkylamino group such as  $-\text{NHCH}_2(4\text{-OCH}_3\text{Ph})$ ,  $-\text{NHCH}(\text{CH}_3)\text{Ph}$ ,  $-\text{NHCH}(\text{Ph})_3$ ; sulfonamido group such as,



5 The invention is further exemplified by the following non-limiting examples, which are illustrative representing the preferred modes of carrying out the invention. The invention's scope is not limited to these specific embodiments only but should be read in conjunction with what is disclosed anywhere else in the specification together with those information and knowledge which are within the general understanding of a person skilled in the art.

### Example 1

**Synthesis of (S)-3-hydroxy-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-1-one from sodium salt of 4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one using oxidoreductase in an enzyme coupled cofactor regeneration system**

Into a 250 ml round bottom flask equipped with a thermometer inlet a pH probe and an overhead stirrer, Glucose 6.28 gm (0.0349 moles) and  $\beta$ -Nicotinamide adenine dinucleotide phosphate disodium salt (10mg) was dissolved in 100 ml of water. Glucose Dehydrogenase lyophilized powder prepared as per example 4 of 2805/MUM/2010 (pET27bZBG13.1.1, 12.5 gm) was added to the reaction mixture to get a suspension. 50 gm cells prepared by the process as mentioned in the example no 6 of 2805/MUM/2010 (pET27BZBG2.0.9) suspended in 50 ml water was added to the reaction mixture and homogeneous preparation was incubated at 25-30 °C under stirring condition. 10 gm (0.02331 moles) of substrate, i.e., sodium salt of 4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one prepared as per process similar to WO2010/032264 was added in portions. Since this is a pH driven reaction (where pH is being maintained in the range of 7.0 to 8.0) and the substrate is basic in nature, substrate addition to the reaction mixture is carried out in a regulated, step-by-step manner in presence of NaOH, over a period of 3-4 hours, making the total volume of the reaction mixture to 200 ml. The progress of the reaction was observed on TLC. During 25 to 30 hrs, gradually the substrate almost disappeared and the product spot was seen. Reaction

mixture was extracted twice in equal volumes of ethyl acetate and upon evaporating the solvent the desired product was obtained in 60% yield.

The product was further analyzed by HPLC analysis showing an HPLC purity of >90% of the corresponding alcohol, followed by chiral HPLC analysis (as described in example no 5 of 2805/MUM/2010) showing an enantiomeric excess of >99% of single enantiomer.

The chiral configuration of this enzymatically synthesized alcohol, which appears as P1 in chiral HPLC analyses, is found to be (S). This chiral (S) alcohol was taken up for further synthesis.

### Example 2

**Synthesis of (S)-3-hydroxy-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-1-one from sodium salt of 4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one using crude lysate of whole-cell catalyst co-expressing oxidoreductase and glucose dehydrogenase**

Into a 1000 ml round bottomed flask equipped with a thermometer, an inlet, a pH probe and an overhead stirrer, Glucose (6.28 gm, 0.0349 moles) and  $\beta$ -Nicotinamide adenine dinucleotide phosphate disodium salt (10mg) was dissolved in 50 ml of water. 50 gm cells prepared as mentioned in the above example no 7 of 2805/MUM/2010 suspended in 500 ml water was subjected to cell lysis and clear cell free extract was added in the reaction mixture. The homogeneous reaction preparation was incubated at 25-30 °C under stirring condition. 10 gm (0.02331 moles) of Sodium salt of 4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one prepared as per process similar to WO2010/032264 was added in portions to the reaction mixture by maintaining pH of the reaction at 7.0 to 8.0 as has already been explained in example 9 above. The progress of the reaction was observed on TLC. During 25 to 30 hrs the starting material was almost disappeared and product spot was seen. Reaction mixture was extracted twice in equal volumes of Ethyl acetate and upon evaporating the solvent the desired product was obtained in 72% yield.

The product was future analyzed by HPLC analysis followed by chiral HPLC analysis (as described in example no 5 of 2805/MUM/2010). Which showed >90% HPLC purity of corresponding alcohol and >99% ee of single Enantiomer.

The chiral configuration of this enzymatically synthesized alcohol, which appears as P1 in chiral HPLC analyses, is found to be (S).

### Example 3

**Synthesis of (S)-3-hydroxy-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-1-one from sodium salt of 4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one using Whole-Cell Catalyst co-expressing oxidoreductase and glucose dehydrogenase at a large scale**

Into a 1000 ml round bottomed flask equipped with thermometer inlet, pH probe and overhead stirrer Glucose (15.66 gm, 0.087 moles) and  $\beta$ -Nicotinamide adenine dinucleotide phosphate disodium salt (12.5 mg) was dissolved in 100 ml of water. 250gm whole cells prepared as mentioned in example no 7 of 2805/MUM/2010 suspended in 250 ml water was added to the reaction mixture followed by 12.5 ml Toluene. The homogeneous reaction preparation was incubated at 25-30 °C under stirring condition. 25 gm (0.5827 moles) of Sodium salt of 4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one prepared as per process similar to WO2010/032264 was added in portions to the reaction mixture by maintaining pH of the reaction at 7.0 to 8.0. The progress of the reaction was observed on TLC. During 25 to 30 hrs the starting material was almost disappeared and product spot was seen. Reaction mixture was extracted twice in equal volumes of ethyl acetate and upon evaporating the solvent the desired product was obtained in 72% yield.

The enzymatically prepared alcohol product was analyzed by various classical tools i.e. Melting Point (m.p.), Specific Optical Rotation (SOR), Infra Red Spectroscopy (IR) and Nuclear Magnetic Resonance spectroscopy (NMR) and ESI-MS with the following results -

m.p. ; 116-120 °C

SOR  $[\alpha]_D^{25}$ : 23.2° (c = 1, CHCl<sub>3</sub>)

IR (cm<sup>-1</sup>): 3468, 1626, 1519

ESI-MS: 409 (M+H)<sup>+</sup>

<sup>1</sup>H NMR (400 MHz, DMSO-D<sub>6</sub>): δ 2.45-2.49 (m, 1H), 2.65-2.78 (m, 3H), 3.89-3.99 (m, 2H), 4.01-4.09 (m, 2H), 4.21-4.22 (m, 1H), 4.86-5.05 (overlapping m, 3H), 7.38-7.47 (m, 2H).

<sup>13</sup>C NMR (100 MHz, DMSO-D<sub>6</sub>): δ 35.4, 37.4, 38.3, 40.1, 41.4, 42.2, 43.0, 43.7, 67.3, 105.4, 114.5, 117.1, 119.5, 123.0, 142.3, 144.4, 146.5, 148.8, 151.0, 154.6, 156.9, 170.2.

The product was further analyzed by HPLC and chiral HPLC analysis (as described in example 5 of 2805/MUM/2010), which showed 96.1% HPLC purity of corresponding alcohol and 99.7 % chiral purity of single enantiomer .

The chiral configuration of this enzymatically synthesized alcohol, which appears as PI in chiral HPLC analyses, is found to be (S).

#### Example 4

#### **Preparation of (R/S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-yl p-toluenesulfonate**

In a dry, 100 mL round bottom flask, (R/S)-3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl)butan-1-one (5.0 g) and pyridine (25 mL) were charged at 25-30 °C and the reaction mixture was cooled to 0-5 °C. To the clear solution p-toluenesulfonyl chloride (7.0 g) was added portionwise at 0-5 °C and subsequently stirred for 24 h at 0-5 °C. The reaction mixture was added slowly at 0-5 °C into water and extracted with ethyl acetate. The organic layer was washed with dil. aq. HCl solution, saturated sodium bicarbonate solution and brine solution, it was dried over anhydrous sodium sulfate. It was distilled out at reduced pressure to obtain the title compound (Wt.- 6.0 g, % Purity by HPLC- 86.0 %).

The crude compound was stirred with diisopropyl ether at 25-30 °C for 30 min. After that it was filtered, washed with diisopropyl ether and dried to obtain the title compound. (Wt.-3.73 g, % Purity by HPLC-90.7 %).

#### **Purification:**

(R/S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-yl p-toluenesulfonate (200 mg) was stirred with methanol at 0-5 °C for 30 min.. After that it was filtered, washed with cold

methanol and dried to obtain the title compound. (Wt.-146 mg, % Purity by HPLC-95.4 %).

<sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>): δ 2.42 (s, 3H), 2.76-2.93, 2.96-3.06, 3.07-3.14(overlapping m, 4H), 3.96-4.12 (m, 2H), 4.19-4.26 (overlapping m, 2H), 4.95-5.08(overlapping m, 3H), 6.63-6.67 (m, 1H), 6.73-6.94 (m, 1H), 7.16-7.20 (m, 2H), 7.48-7.58 (m, 2H).

IR(cm<sup>-1</sup>): 3059, 1662, 1520 ESI-MS: 563 (M+H)<sup>+</sup>

#### Example 5

**Preparation of (S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-yl p-toluenesulfonate**

In a dry, 250 mL round bottom flask (S)-3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl)butan-1-one (10 g) and pyridine were charged at 25-30 °C and the reaction mixture was cooled at 0-5 °C. After that p-toluenesulfonyl chloride was added in 3 lots during 7h reaction time at 0-5 °C with stirring. Reaction mixture was added into diluted aqueous HCl solution at -5 to 0 °C and it was stirred for 2 h at -5 to 0 °C to obtain solid compound. It was filtered and washed with water and aqueous saturated sodium bicarbonate solution. It was dried at 45-50 °C. (Wt.-12.7 g, % Y-92.1, % Purity by HPLC-98.8 %, % Chiral purity-99.8 %).

m.p.: 161-164 °C, SOR:[α]<sub>D</sub><sup>25</sup> -42.3° (c 1 in CHCl<sub>3</sub>) (Range: -36 to -43°), % H<sub>2</sub>O - 0.16 % w/w.

#### Example 6

**Preparation of (S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-yl p-toluenesulfonate**

In a dry, 1.0 L round bottom flask (S)-3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl)butan-1-one (80 g) and pyridine (400 mL) were charged at 25-30 °C and the reaction mixture was cooled at 0-5 °C. After that p-toluenesulfonyl chloride (186.7 gm) was added in 3 lots during 7 h reaction time at 0-5 °C with stirring. Reaction mixture was added into diluted aqueous HCl solution at -5 to 0 °C and it was stirred for 2 h at -5 to 0 °C to obtain solid compound. It was filtered and washed with water and aqueous saturated sodium bicarbonate solution. It was dried at 45-50 °C. (Wt.-94.0 g, % Y-85.2, % Purity by HPLC-98.5%, % Chiral purity->99.5 %).

m.p.: 161-164 °C, SOR:[ $\alpha$ ]<sub>D</sub><sup>25</sup> -37.6 (c 1 in CHCl<sub>3</sub>) (Range: -36 to -43°), % H<sub>2</sub>O – 0.15 % W/W.

<sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>):  $\delta$  2.42 (s, 3H), 2.79-2.93, 2.95-3.04, 3.09-3.16 (overlapping m, 4H), 3.98-4.06 (m, 2H), 4.16-4.28 (overlapping m, 2H), 4.91-5.09 (overlapping m, 3H), 6.62-6.67 (m, 1H), 6.73-6.82 (m, 1H), 7.15-7.20 (m, 2H), 7.45-7.48 (m, 2H).

IR(cm<sup>-1</sup>): 3020, 2928, 1633, 1521 ESI-MS: 563 (M+H)<sup>+</sup>

#### Example 7

#### **Preparation of (S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-yl p-toluenesulfonate**

In a dry, 25 mL round bottom flask (S)-3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl)butan-1-one (0.2 g), DABCO (0.220 g) and toluene (1.5 mL) were charged at 25-30 °C. The reaction mixture was cooled at 0-5 °C. After that p-toluenesulfonyl chloride (0.230 g) was added and stirred for 3 h at 0-5 °C. Reaction mixture was diluted with toluene (5.0 mL). It was transferred into a separating funnel. It was washed four times with water. The organic layer was collected. It was dried over anhydrous sodium sulfate. It was distilled out at reduced pressure to obtain the title compound (Wt.- 0.260 g, % Purity by HPLC- 87 %).

20

#### Example 8

#### **Preparation of (S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-yl p-toluenesulfonate**

In a dry, 25 mL round bottom flask (S)-3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl)butan-1-one (0.2 g), DABCO (0.220 g) and dichloromethane (1.5 mL) were charged at 25-30 °C. The reaction mixture was cooled at 0-5 °C. After that p-toluenesulfonyl chloride (0.230 g) was added and stirred for 4 h at 0-5 °C. Reaction mixture was diluted with dichloromethane (5.0 mL). It was transferred into a separating funnel. It was washed with four times with water. The organic layer was collected. It was dried over anhydrous sodium sulfate. It was distilled out at reduced pressure to obtain the title compound (Wt.- 0.300 g, % Purity by HPLC- 87 %).

30

### Example 9

#### **Preparation of (S)-diethyl (4-oxo-4-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-1-(2,4,5-trifluorophenyl)butan-2-yl) phosphate**

5 In a dry, 100 mL round bottom flask, (S)-3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl)butan-1-one (0.5 g) and dichloromethane (10 mL) were charged at 25-30 °C and the reaction mixture was cooled to 0-5 °C. To the clear solution pyridine (0.118 mL) was added. After 30 min. diethylchlorophosphate (0.22 g) was added drop by drop at 0-5 °C. It was  
10 warmed to 25-30 °C and stirred for 21 h at 25-30 °C. The reaction mixture was diluted with dichloromethane (25 mL). It was transferred into a separating funnel and it was washed 3 times with 10 % aq. HCl solution, 3 times with saturated sodium bicarbonate solution and brine solution, it was dried over anhydrous sodium sulfate. It was distilled out at reduced pressure to obtain the title compound (Wt.- 0.633 g, % Purity by HPLC-  
15 84.0 %).

ESI-MS: 545 (M+H)<sup>+</sup>

### Example 10

#### **Preparation of (R/S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-yl p-nitrobenzenesulfonate**

20 In a dry, 25 mL round bottom flask, (R/S)-3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl)butan-1-one (0.5 g) and pyridine (2.5 mL) were charged at 25-30 °C and the reaction mixture was cooled to 0-5 °C. To the clear solution p-nitrobenzenesulfonyl chloride (0.32 g) was added portionwise at 0-5 °C and stirred for 5 h at 0-5 °C. After that another p-  
25 nitrobenzenesulfonyl chloride (0.54 g) was added and stirred for another 1 h at 0-5 °C. The reaction mixture was added slowly at 0-5 °C into aq. KHSO<sub>4</sub> solution and extracted with ethyl acetate. The organic layer was washed with dil. aq. HCl solution, saturated sodium bicarbonate solution and brine solution; it was dried over anhydrous sodium sulfate. It was distilled out at reduced pressure to obtain the title compound (Wt.- 0.630  
30 g, % Purity by HPLC- 80.8 %).



### Example 11

#### Preparation of (S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-yl p-nitrobenzenesulfonate

In a dry, 50 mL round bottom flask, (S)-3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl)butan-1-one (4.0 g) and pyridine (20 mL) were charged at 25-30 °C and the reaction mixture was cooled to 0-5 °C. To the clear solution p-nitrobenzenesulfonyl chloride (6.5 g) was added portionwise at 0-5 °C and stirred for 4 h at 0-5 °C. After that another p-nitrobenzenesulfonyl chloride (2.1 g) was added and stirred for another 1 h at 0-5 °C. The reaction mixture was added slowly at 0-5 °C into 10 % aq. HCl solution and stirred for 30 min. at 25-30 °C. Solid was filtered and washed with 5 % cooled aq. HCl, with 5 % cooled aq. NaHCO<sub>3</sub> solution and with cold water. It was dried under reduced pressure at 40 °C. (Wt.- 6.0 g, % Purity by HPLC- 87.5 %).

#### Purification:

(S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-yl p-nitrobenzenesulfonate (5.5 g) was stirred with methyl t-butyl ether at 25-30 °C for 30 min.. After that it was filtered, washed with cold methyl t-butyl ether and dried to obtain the title compound. (Wt.-4.3 g, % Purity by HPLC-91.4 %, % Chiral purity-99.2 %).

m.p. : 98-99 °C, SOR:[ $\alpha$ ]<sub>D</sub><sup>25</sup>-28.6° (c 1 in CHCl<sub>3</sub>), % H<sub>2</sub>O – 0.85 % W/W.

<sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>):  $\delta$  2.77-2.87 (m, 1H), 2.89-2.97 (m, 1H), 3.06-3.11 (m, 2H), 4.01-4.02 (m, 1H), 4.15-4.28 (m, 3H), 4.91-5.23 (overlapping m, 3H), 6.71-6.77 (m, 1H), 6.86-6.92 (m, 1H), 7.86-7.88 (d, 2H), 8.27-8.29 (d, 2H).

IR(cm<sup>-1</sup>): 3072, 1658, 1533, 1186, 914 ESI-MS: 594 (M+H)<sup>+</sup>

### Example 12

#### Preparation of (S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-yl acetate

In a dry, 50 mL round bottom flask, (S)-3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl)butan-1-one (3.0 g), dimethylaminopyridine (89 mg) and dichloromethane (30 mL) were charged at 25-30 °C. The reaction mixture was cooled to 0-5 °C. To the clear solution acetic anhydride (1.1 g) was added drop by drop over a period of 15-30 min. at 0-5 °C. It was stirred for 3 h at 0-5 °C. The reaction mixture was diluted with dichloromethane (15 mL). It was transferred into a separating funnel and it was washed 3 times with water

and brine solution. It was dried over anhydrous sodium sulfate. It was distilled out at reduced pressure to obtain the title compound (Wt.- 3.29 g, % Purity by HPLC- 92.5 %, % Chiral purity >99.5 %).

<sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>): δ 2.23 (s, 3H), 2.61-2.63 (m, 1H), 2.65-2.67 (m, 1H), 2.77-2.86 (overlapping m, 2H), 2.90-2.94 (overlapping m, 1H), 3.02-3.10 (m, 2H), 4.09-4.25 (overlapping m, 1H), 4.89- 5.04 (overlapping m, 2H), 5.31-5.44 (m, 1H), 6.87-6.91 (m, 1H), 7.04-7.11 (m, 1H), 7.86-7.88 (d, 2H), 8.27-8.29 (d, 2H).

IR(cm<sup>-1</sup>): 3020, 1739, 1662, 1521, 1215 ESI-MS: 451 (M+H)<sup>+</sup>

#### Example 13

#### 10 **Preparation of (R/S)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one**

In a 25 mL round bottom flask (R/S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-yl, p-toluenesulfonate (0.500 g) and dimethylsulfoxide were charged. Subsequently, sodium azide (22.8 mg) was added at 24-30 °C into the reaction mixture. The reaction mixture was stirred for 6h at 24-30 °C. After that second lot of sodium azide (22.8 mg) was added and stirred for 15 h, subsequently, third lot of sodium azide (22.8 mg) was added and further stirred for 9 h at 24-30 °C. The reaction mixture was added into cold water and extracted with ethyl acetate. The organic layer was washed with water and brine solution, it was dried over anhydrous sodium sulfate. It was distilled out at reduced pressure to obtain the title compound (Wt.-379 mg, % Purity by HPLC- 62.3 %).

#### Example 14

#### **Preparation of pure (R/S)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]-triazolo[4,3-a] pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one**

25 Crude (R/S)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]- triazolo[4,3-a] pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one (2 g) was purified by column chromatography over silica gel (100-200 mesh) using DIPE:EA (4:6) as an eluent (Wt. 1.4 g, % Purity by HPLC-94.3 %)

<sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>): δ 2.61-2.70 (m, 2H), 2.86-2.91(m, 2H), 4.20-4.28 (overlapping m, 4H), 4.95-5.02 (overlapping m, 2H), 6.93-6.95 (m, 1H), 7.10-7.16 (m, 1H),

IR(cm<sup>-1</sup>): 2123, 1654, 1521 ESI-MS: 434 (M+H)<sup>+</sup>

5

#### Example 15

#### **Preparation of (R/S)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one**

In a 25 mL round bottom flask (R/S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-yl, methanesulfonate (0.100 g) and dimethylformamide were charged. To the reaction mixture sodium azide (67 mg) was added at 24-30 °C. The reaction mixture was stirred for 24 h at 24-30 °C. After that second lot of sodium azide (26.0 mg) was added and further stirred for 4 h at 24-30 °C. The reaction mixture was added into cold water and extracted with ethyl acetate. The organic layer was washed with water and brine solution, it was dried over anhydrous sodium sulfate. It was distilled out at reduced pressure to obtain the title compound (Wt.-93 mg, % Purity by HPLC- 25.8 % and % of olefin impurity – 54.5 %).

10

15

#### Example 16

#### **Preparation of (R/S)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one**

In a 25 mL round bottom flask (R/S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-yl, methanesulfonate (0.200 g) and dimethylsulfoxide were charged. To the reaction mixture sodium azide (133mg) was added at 24-30 °C. The reaction mixture was stirred for 27 h at 24-30 °C. The reaction mixture was added into cold water and extracted with ethyl acetate. The organic layer was washed with water and brine solution, it was dried over anhydrous sodium sulfate. It was distilled out at reduced pressure to obtain the title compound (Wt.-160 mg, % Purity by HPLC- 46.0 % and % of olefin impurity – 49.8 %).

20

25

30

#### Example 17

#### **Preparation of (R)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one**

In a 25 mL round bottom flask (S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-yl, p-toluenesulfonate (0.700 g) and dimethylsulfoxide were charged. Subsequently, sodium azide was added at 25-30 °C into the reaction mixture. The reaction mixture was stirred for 28 h at 25-30 °C. The reaction mixture was added into cold water and extracted with ethyl acetate. The organic layer was washed with water and brine solution, it was dried over anhydrous sodium sulfate. It was distilled out at reduced pressure to obtain the title compound (Wt.-0.490 g, % Purity by HPLC- 70.9 %).

#### Example 18

#### 10 **Preparation of (R)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one**

In a 250 mL round bottom flask (S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-yl, p-toluenesulfonate (2.5 g) and dimethylsulfoxide were charged. Subsequently, sodium azide was added at 25-30 °C into the reaction mixture. The reaction mixture was stirred for 28 h at 25-30 °C. The reaction mixture was added into cold water and extracted with ethyl acetate. The organic layer was washed with water and brine solution, it was dried over anhydrous sodium sulfate. It was distilled out at reduced pressure to obtain the title compound (Wt.-1.9 g, % Purity by HPLC- 75.6 %).

20 ESI-MS: 434 (M+H)<sup>+</sup>

#### Example 19

#### **Preparation of (R)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one**

In a 2.0 L round bottom flask (S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-yl, p-toluenesulfonate (93 g) and dimethylsulfoxide were charged. Subsequently, sodium azide (53.7 g) was added at 25-30 °C into the reaction mixture. The reaction mixture was stirred for 22 h at 20-30 °C. The reaction mixture was added into cold water and extracted with ethyl acetate. The organic layer was washed with water and brine solution, it was dried over anhydrous sodium sulfate. It was distilled out at reduced pressure to obtain the title compound (Wt.-69.4 g, % Purity by HPLC- 77.9 %).

30 ESI-MS: 434 (M+H)<sup>+</sup>

### Example 20

#### **Preparation of pure (R)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]-triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one**

Crude (R)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]-triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one (2.9 g) was purified by column chromatography over silica gel (100-200 mesh) using DIPE:EA (7:3) as an eluent (Wt. 2.0 g, % Purity by HPLC-96.5 %)

SOR: $[\alpha]_D^{25}$  -2.37° (c 1 in CHCl<sub>3</sub>)

<sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>): δ 2.58-2.71 (m, 2H), 2.82-2.95(m, 2H), 3.98-4.26 (overlapping m, 4H), 4.96-5.01 (overlapping m, 2H), 6.93-6.97 (m, 1H), 7.09-7.16 (m, 1H),

IR(cm<sup>-1</sup>): 2123, 1666, 1521 ESI-MS: 434 (M+H)<sup>+</sup>

### Example 21

#### **Preparation of (R)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one**

In a 250 mL round bottom flask (S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-yl p-nitrobenzenesulfonate (0.200 g) and toluene were charged. Then tetrabutylammonium azide (97 mg) was added at 25-30 °C into the reaction mixture. The reaction mixture was stirred for 5 h at 25-30 °C. Then another tetrabutylammonium azide (24 mg) was added and further stirred for 6 h. The reaction mixture was added into cold water and extracted with ethyl acetate. The organic layer was washed with water and brine solution, it was dried over anhydrous sodium sulfate. It was distilled out at reduced pressure to obtain the title compound (Wt.-0.125 g, % Purity by HPLC- 72.4 %).

ESI-MS: 434 (M+H)<sup>+</sup>

### Example 22

#### **Preparation of (R/S)-3-(benzylamino)-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo [4,3-a]pyrazin-7(8H)-yl]-4-(2,4,5-trifluorophenyl)butan-1-one OR 3-[(benzyl)amino)-4-(2,4,5-trifluorophenyl)butanoyl]-3-trifluoromethyl-5,6,7,8-tetrahydro-1,2,4-triazolo[4,3-a]pyrazine**

In a 25 mL round bottom flask (R/S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-yl, p-toluenesulfonate (0.500 g), benzylamine (0.190 g) and toluene were charged. The

reaction mixture was heated to 80 °C and stirred for 5 h at 80-82 °C. Subsequently, the reaction mixture was cooled to 25-30 °C and water was added. The reaction mixture was extracted with ethyl acetate. The organic layer was washed with water and brine solution, it was dried over anhydrous sodium sulfate. It was distilled out at reduced pressure to obtain the title compound (Wt.- 0.446 g, % Purity by HPLC- 25.3 %).

#### Example 23

##### **Preparation of (R/S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine**

In a 50 mL round bottom flask crude (R/S)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one (1.4 g), methanol and 5 % Pd/C (280 mg) were taken. The reaction mixture was cooled to 0 to 5 °C and NaBH<sub>4</sub> (368 mg) was added. It was warmed to 25-30 °C and stirred for 20-24 h at 25 to 30 °C. After that water and hyflosupercell were added into the reaction mixture and stirred for 5-10 minutes. It was filtered and washed with methanol. Filtrate was taken in a 50 mL one neck round bottom flask. Solvent was distilled out at reduced pressure to obtain crude (R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine. After usual acid-base purification pure product was obtained. (Wt.-860 mg, % Purity by HPLC- 95.0 %).

#### Example 24

##### **Preparation of (R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine**

In a 25 mL round bottom flask crude (R)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one (1.35 g), methanol and 5 % Pd/C (270 mg) were taken. The reaction mixture was cooled to 0 to 5 °C and NaBH<sub>4</sub> (355 mg) was added. It was warmed to 25-30 °C and stirred for 42 h at 25 to 30 °C. After that water, methanol and hyflosupercell were added into the reaction mixture and stirred for 5-10 minutes. It was filtered and washed with methanol. Filtrate was taken in a 100 mL one neck round bottom flask. Solvent was distilled out at reduced pressure and to obtain crude (R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine. After usual acid-base purification pure product was obtained. (Wt.-0.852 g, % Purity by HPLC- 92.8 %, Chiral Purity by HPLC- >99.5 %).

<sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>): δ 2.58-2.80 (m, 2H), 2.82-2.95 (m, 2H), 3.64-3.69 (m, 1H),

3.70-3.98 (m, 1H), 4.07-4.22 (m, 3H), 4.88-5.06 (m, 2H), 6.88-6.94 (m, 1H), 7.10-7.16 (m, 1H).

5 IR(cm<sup>-1</sup>): 1649, 1518 ESI-MS: 408 (M+H)<sup>+</sup>

#### Example 25

#### **Preparation of (R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine**

In a 25 mL round bottom flask water (9.0 mL), Zinc dust (90 mg) and  
10 Aluminium chloride hexahydrate (184 mg) were taken. After that crude (R)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one (0.3 g) dissolved in ethanol (0.9 mL) was slowly added. It was stirred at 25-30 °C for 22 h. It was dumped into dil. aq. NaOH solution till 8-9 pH. Solid precipitated. It was filtered. Filterate was transferred into a separating funnel. It  
15 was extracted twice with ethyl acetate (20 mL). Both the extracts were combined and washed with water and brine solution. Organic layer was collected and dried over anhyd. Sodium sulfate. Solvent was distilled out at reduced pressure and to obtain crude (R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine. After usual acid-base purification pure  
20 product was obtained. (Wt.-0.180 mg, % Purity by HPLC- 59.4 %)

#### Example 26

#### **Preparation of (R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine**

In a 25 mL round bottom flask 35 % conc.HCl (4.6 mL), Zinc dust (225 mg)  
25 and crude (R)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one (0.3 g) were taken. It was heated to 80-85 °C and stirred for 4 h. It was dumped into dil. aq. NaOH solution till 8-9 pH. Solid precipitated. It was filtered. Filtrate was transferred into a separating funnel. It was extracted twice with ethyl acetate (20 mL). Both the extracts were combined and  
30 washed with water and brine solution. Organic layer was collected and dried over anhyd. sodium sulfate. Solvent was distilled out at reduced pressure and to obtain crude (R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine. (Wt.-0.105 mg, % Purity by HPLC- 53.6 %) After usual acid-base purification pure product was obtained.

**Example 27****Preparation of (R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine**

In a 25 mL round bottom flask methanol (3.0 mL) and crude (R)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one (0.3 g) were taken. It was cooled to 0-5 °C. To the reaction mixture NiCl<sub>2</sub>.6H<sub>2</sub>O (83 mg) and NaBH<sub>4</sub> (270 mg) were added. It was stirred for 5 h at 0-30 °C. After that again NiCl<sub>2</sub>.6H<sub>2</sub>O (83 mg) and NaBH<sub>4</sub> (270 mg) were added into the reaction mixture. It was further stirred for 4 h. It was dumped into water (12 mL). Solid precipitated. It was filtered. Filtrate was transferred into a separating funnel. It was extracted twice with ethyl acetate (12 mL). Both the extracts were combined and washed with water and brine solution. Organic layer was collected and dried over anhyd. Sodium sulfate. Solvent was distilled out at reduced pressure and to obtain crude (R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine. (Wt.-0.190 mg, % Purity by HPLC- 40.6 %). After usual acid-base purification pure product was obtained.

**Example 28****Preparation of (R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine**

In a 25 mL round bottom flask methanol (3.0 mL) and crude (R)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one (0.3 g) were taken. To the reaction mixture SnCl<sub>2</sub>.2H<sub>2</sub>O (265 mg) was added. It was stirred for 22 h at 25-30 °C. After that again SnCl<sub>2</sub>.2H<sub>2</sub>O (265 mg) was added into the reaction mixture. It was further stirred for 17 h at 25-30 °C and 7 h at 55-60 °C. Distilled out the solvent at reduced pressure. To the residue dil. aq. NaOH solution was added upto ~ 9 pH. The solution was transferred into a separating funnel. It was extracted twice with ethyl acetate (12 mL). Both the extracts were combined and washed with water and brine solution. Organic layer was collected and dried over anhyd. Sodium sulfate. Solvent was distilled out at reduced pressure and to obtain crude (R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine. (Wt.-0.170 mg, % Purity by HPLC- 45.9 %). After usual acid-base purification pure product was obtained.



**Example 29****Preparation of (R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine**

In a 25 mL round bottom flask pyridine (1.5 mL) and crude (R)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one (0.3 g) were taken. To the reaction mixture triphenylphosphine (363 mg) was added at 25-30 °C. After 1 h aq. 25 % NH<sub>3</sub> solution (0.55 mL) was added at 25-30 °C and stirred for 72 h. After that again triphenylphosphine (180 mg) and aq. 25 % NH<sub>3</sub> solution (0.27 mL) were added at 25-30 °C and stirred for 5 h. Distilled out the solvent at reduced pressure. The sticky mass was triturated with three times with hexane. Distilled out traces of solvent at reduced pressure to obtain crude solid. (Wt.-0.520 mg, % Purity by HPLC- 10.9 %).

**Example 30**

**Preparation of (R/S)-3-(1,3-dioxoisindolin-2-yl)-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo [4,3-a]pyrazin-7(8H)-yl]-4-(2,4,5-trifluorophenyl)butan-1-one**

**OR**

**(R/S)-2-(4-oxo-4(3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-1-(2,4,5-trifluorophenyl)butan-2-yl)isoindoline-1,3-dione**

In a dry, 50 mL round bottom flask, (R/S)-3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl)butan-1-one (1.0 g) and dry THF (20 mL) were charged at 25-30 °C under N<sub>2</sub> atmosphere. To the clear solution triphenylphosphine (0.960 g) and phthalimide (0.540 g) were added at 25-30 °C and stirred for 10 min. After that DIAD (0.740 g) was added drop by drop over a period of 10-15 min. It was stirred for 2 h at 25-30 °C. The reaction mixture was dumped into water and extracted with ethyl acetate. The organic layer was washed with water and brine solution. It was dried over anhydrous sodium sulfate. It was distilled out at reduced pressure to obtain the crude title compound (Wt.- 2.39 g)

ESI-MS shows 537 (M-H)<sup>-</sup> value of the product.

**Example 31**

**Preparation of (R/S)-3-(benzylamino)-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo [4,3-a]pyrazin-7(8H)-yl]-4-(2,4,5-trifluorophenyl)butan-1-one OR**

**3-[(benzyl)amino)-4-(2,4,5-trifluorophenyl)butanoyl]-3-trifluoromethyl-5,6,7,8-**  
5 **tetrahydro-1,2,4-triazolo[4,3-a]pyrazine**

In a dry, 50 mL round bottom flask, (R/S)-3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4.3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl)butan-1-one (1.0 g) and dry THF (20 mL) were charged at 25-30 °C under N<sub>2</sub> atmosphere. To the clear solution triphenylphosphine (0.835 g) was added at 25-30 °C. After 10 min.  
10 benzylamine (0.350 mL) and DIAD (0.630 g) was added drop by drop over a period of 10-15 min. It was stirred for 26 h at 25-30 °C. The reaction mixture was dumped into water and extracted with ethyl acetate. The organic layer was washed with water and brine solution. It was dried over anhydrous sodium sulfate. It was distilled out at reduced pressure to obtain the crude title compound (Wt.- 2.19 g, HPLC purity shows  
15 very small amount of product formation).

**Example 32**

**Preparation of (R)-N-(4-methylphenylsulfonyl)-N'-t-butoxycarbonyl)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine**

In a dry, 50 mL round bottom flask, (S)-3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4.3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl)butan-1-one (0.5 g), N-(t-butoxycarbonyl)-p-toluenesulfonamide (0.332 g) and dry THF (10 mL) were charged at 25-30 °C under N<sub>2</sub> atmosphere. To the clear solution triphenylphosphine (0.960 g) was added at 25-30 °C and stirred for 10 min. After that  
25 DIAD (0.618 g) was added drop by drop over a period of 10-15 min. It was stirred for 2 h at 25-30 °C. The reaction mixture was dumped into water and extracted with ethyl acetate. The organic layer was washed with water and brine solution. It was dried over anhydrous sodium sulfate. It was distilled out at reduced pressure to obtain the crude title compound (Wt.- 1.9 g)  
30 ESI-MS shows 560 (M-H)<sup>+</sup> value of the product.

**Example 33**

**Preparation of (R/S)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one**

In a 25 mL round bottom flask (R/S)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-yl, p-toluenesulfonate (0.200 g), and dimethylformamide (2.0 mL) and tributyl hexadecylphosphonium bromide (18 mg) were charged. Subsequently, sodium azide (57 mg) was added at 24-30 °C into the reaction mixture. The reaction mixture was stirred for 6h at 24-30 °C and 8 h at 40-45 °C. The reaction mixture was added into cold water and extracted with ethyl acetate. The organic layer was washed with water and brine solution, it was dried over anhydrous sodium sulfate. It was distilled out at reduced pressure to obtain the title compound (Wt.-186 mg, % Purity by HPLC- 44.4 %).

#### Example 34

##### **Preparation of (R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine**

In a 250 mL round bottom flask crude (R)-3-azido-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one (9.5 g), methanol(90 mL), ammonium chloride (2.1 g) and water (4.75 mL) were taken. The reaction mixture was stirred for 10-15 min. to dissolve the solid at 25 to 30 °C. After that Zinc metal powder (2.85 g) was added. It was stirred for 6 h at 25 to 30 °C. Again Zinc metal powder (2.85 g) and ammonium chloride (2.1 g) were added. It was further stirred for 17 h at 25 to 30 °C . It was filtered through hyflosupercell bed using Buchener flask and funnel. The hyflosupercell bed was washed with methanol. The filtrate was collected and transferred into a 500 mL one neck round bottom flask. Solvent was distilled out at reduced pressure to obtain crude (R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine (Wt.-9.2 g % Purity by HPLC- 75.6 %). After usual acid-base purification pure product was obtained. (Wt.-6.6 g, % Purity by HPLC- 96.9 %, Chiral Purity by HPLC- >99.5 %).

#### Example 35

##### **Preparation of Monohydrate phosphate salt of (R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine**

In a 250 mL round bottom flask (R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine (6.0 g) from example 34, isopropanol (25.5 mL), and water (6.6 mL) were taken. The

reaction mixture was stirred for 10-15 min. to dissolve the solid at 25 to 30 °C. After that 4.35 ml 45 % aq. phosphoric acid (1.14 mL 85 % H<sub>3</sub>PO<sub>4</sub> was diluted upto 4.35 mL volume by adding water) was added drop by drop over 20-30 min. at 25-35 °C. It was heated to 78 °C and stirred for 5-10 min. at 78 °C. Then it was cooled to 67-68 °C and stirred for 3 h at 67-68 °C. It was slowly cooled to 30 °C at a rate of 12 °C/h. To the clear reaction mixture seed of monohydrate phosphate salt of title compound (10 to 20 mg) was added. It was further stirred for 17 h at 25-30 °C. To the thick sultry isopropanol (18.6 mL) was added drop by drop and It was further stirred for 1 h at 25 to 30 °C. The solid salt was filtered using Buchener flask and funnel. The solid was washed with aq. isopropanol (18 mL, 14.4 mL isopropanol + 3.6 mL water). The solid was dried under reduced pressure to obtain Monohydrate phosphate salt of (R)-4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-amine (Wt.-6.7g, % Purity by HPLC- 99.5 %, Chiral Purity by HPLC- >99.5 %).

15 SOR  $[\alpha]_D^{25}$  : -20.9° (c 1 in H<sub>2</sub>O), % water -3.4 %.

20

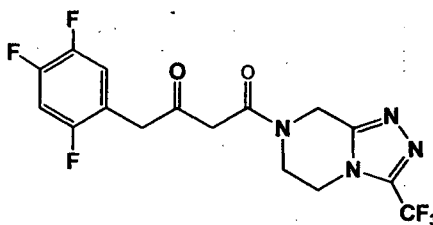
25

30

**We claim:**

1. A process for the preparation of compound Formula (I) or its salts either in its racemic (R/S) form or any of its optically active (S) or (R) forms or enantiomeric excess mixture of any of the forms comprising;

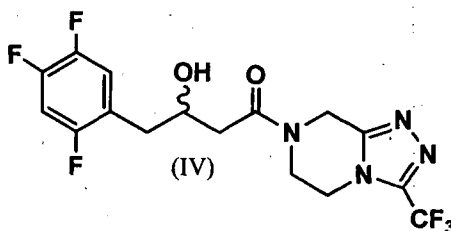
5 a) reacting 4-oxo-4-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl]-1-(2,4,5-trifluorophenyl)butan-2-one of formula (V) or its metal ion salt:



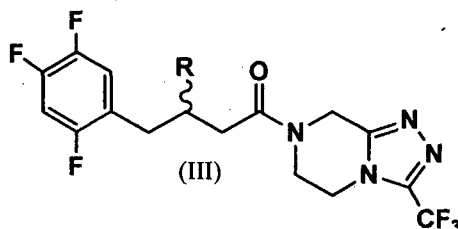
10 Formula (V)

with a suitable enzyme or its variants that reduce a ketone to form an alcohol, to obtain 3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl) butan-1-one of formula (IV), in racemic (R/S) form or any of its optically active (S) or (R) forms or their enantiomerically excess mixtures;

15 (b) reacting 3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl) butan-1-one of Formula (IV) either in its racemic (R/S) form or any of its optically active (S) or (R) forms or enantiomeric excess mixture of any of the forms

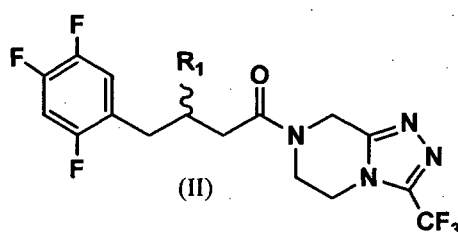


20 with a suitable reagents that activates the hydroxyl group to obtain compound of Formula (III) wherein 'R' represents a suitable leaving group such as suitable carboxylate, suitable sulfonate, suitable phosphate and suitable oxyphosphonium ion, with retention of configuration;



(c) optionally, isolating the compound of Formula (III);

(d) converting compound of Formula (III) to compound of Formula (II) with inversion of configuration, wherein  $R_1$  represent suitable azide group, suitable cyclic imido group, suitable aralkylamino group, suitable sulfonamido group, by using suitable amine equivalent, optionally with suitable additives in suitable solvents;



(e) suitably deprotecting or reducing the compound of Formula (II) or employing suitable chemistry to obtain compound of Formula (I) with retention of configuration.

- 10 2. The process as claimed in claim 1 in step (a), wherein the suitable enzyme is an oxidoreductase enzyme.
3. The process as claimed in claim 1 in step (b), wherein suitable reagents used is selected from suitable acid chlorides, suitable anhydrides, suitable sulfonyl chlorides, suitable chloro phosphates and suitable oxyphosphonium ions.
- 15 4. The process as claimed in claim 3, wherein suitable acid chlorides used is selected from acetyl chloride, benzoyl chlorides and substituted benzoyl chlorides;
5. The process as claimed in claim 3, wherein suitable anhydrides used is selected from acetic anhydride, trifluoroacetic anhydride, and triflic anhydride.
6. The process as claimed in claim 3, wherein suitable sulfonyl chlorides used is selected from methanesulfonyl chloride, 2,2,2-trifluoroethanesulfonyl chloride, benzenesulfonyl chloride, p-toluenesulfonyl chloride, p-nitro benzenesulfonyl chlorides.
- 20 7. The process as claimed in claim 3, wherein suitable chloro phosphates used is diethyl chloro phosphate

8. The process as claimed in claim 3, wherein suitable oxyphosphonium ion used is selected from trialkylphosphine such as tributylphosphine, tri-*t*-butylphosphine; cycloalkylphosphines, arylphosphines with azodicarboxylates.
9. The process as claimed in claim 3, wherein when suitable acid chlorides, suitable anhydrides, suitable sulfonyl chlorides, suitable chloro phosphates is used as reagent, suitable solvent used is selected from suitable ethers, suitable chlorinated solvents, suitable nitriles, suitable hydrocarbons, suitable aprotic polar solvents or their suitable mixtures.
10. The process as claimed in claim 1 in step (b), when suitable acid chlorides, suitable anhydrides, suitable sulfonyl chlorides, suitable chloro phosphates is used as reagent, suitable base used is selected from suitable C<sub>(1-5)</sub> alkyl amines, suitable C<sub>(1-5)</sub> substituted alkyl amines, heterocyclic saturated or unsaturated amines selected from as morpholine, piperidine, pyrrolidine, DBU, DBN, 1,4-diazabicyclo[2,2,2]octane, dimethylamino pyridine and pyridine; hydrides selected from NaH; *n*-BuLi, LDA and KHMDS or mixtures of suitable bases.
11. The process as claimed in claim 1 in step (b), wherein suitable acid chlorides, suitable anhydrides, suitable sulfonyl chlorides, suitable chloro phosphates is used as reagent, the reaction is carried out at temperature ranging from -10 °C to 70 °C, preferably the temperature is -10 °C to 50 °C.
12. The process as claimed in claim 1 in step (b), wherein when suitable oxyphosphonium ion is used as a reagent, the suitable solvent used is selected from suitable ethers selected from diethyl ether, 1,4-dioxane, dimethoxy ethane, diethoxy ethane, DIPE, MTBE, THF, 2-methyl tetrahydrofuran or their suitable mixtures.
13. The process as claimed in claim 12, wherein when suitable oxyphosphonium ion is used as a reagent, the reaction is carried out at temperature ranging from -10 to 50 °C, preferably the temperature is -10 to 30 °C.
14. A process as claimed in claim 1 in step (c), wherein the compound of formula (II), when 'R' represents a caboxylate, sulfonate and phosphate is isolated.
15. A process as claimed in claim 1 in step (d), wherein suitable amine equivalent used is selected from suitable -N<sub>3</sub> (azides), suitable cyclic imides, suitable aralkylamines and suitable sulfonamides.
16. A process as claimed in claim 15, wherein suitable azides is selected from HN<sub>3</sub>, NaN<sub>3</sub>, LiN<sub>3</sub>, Zn(N<sub>3</sub>)<sub>2</sub>, Cu(N<sub>3</sub>)<sub>2</sub>, tetrabutylammonium azide; suitable cyclic imides is selected from phthalimide, succinimide; suitable aralkylamines is selected from

benzylamine, p-methoxybenzylamine,  $\alpha$ -methylphenethylamine, tritylamine; suitable sulfonamides is selected from p-toluene sulfonamide, sulfamic acid, N-(t-butoxycarbonyl)-p-toluenesulfonamide.

17. A process as claimed in claim 1 in step (d), wherein suitable additives used is  
5 selected from suitable phase transfer catalysts selected from trialkyl ammonium halides, tetraalkyl ammonium halides, trialkyl ammonium acetates, tetraalkyl ammonium acetates, trialkyl ammonium hydroxides, tetraalkyl ammonium hydroxides, trialkyl ammonium sulfates, tetraalkyl ammonium sulfates, trialkyl ammonium tetrafluoroborate, tetraalkyl ammonium tetrafuloroborate, trialkyl phosphonium halides,  
10 tetraalkyl phosphonium halides, tetrapenyl phosphonium halides, pyridinium halides, Aliquat, Adogen, crown ethers, silicon analogues of crown ethers, PEG, TDA-1.

18. The process as claimed in claim 1 in step (d), wherein the suitable solvent used is selected from suitable ethers, suitable chlorinated solvents, suitable nitriles, suitable hydrocarbons, aprotic polar solvents selected from DMF, DMA, DMSO; N-methyl  
15 pyrrolidone, HMPA, pyridine and water or their suitable mixtures.

19. The process as claimed in claim 1 in step (d), wherein the reaction is carried out at temperature ranging from -10 to 100 °C, preferably the temperature is -10 to 30 °C

20. The process as claimed in claim 1 in step (e), wherein suitable reducing agent used is selected from metal catalyst selected from Pd/H<sub>2</sub>, Pt/H<sub>2</sub>, Pd/Al<sub>2</sub>O<sub>3</sub>, Pd-CaCO<sub>3</sub>,  
20 Pd(OH)<sub>2</sub>, PdO, PtO<sub>2</sub>, Pd-C/HCOOH, Pd-C/HCOONH<sub>4</sub>, Pd(OH)<sub>2</sub>-C/N<sub>2</sub>H<sub>4</sub>, Pd-C/NaBH<sub>4</sub>, Mg, Ca, Zn, Zn-HCl, Zn-CH<sub>3</sub>COOH, SnCl<sub>2</sub>, SnCl<sub>2</sub>/thiophenol, Zn/NH<sub>4</sub>Cl, Zn/AlCl<sub>3</sub>; Lindlar catalyst; Boron containing reducing agents selected from BH<sub>3</sub>, NaBH<sub>4</sub>, LiAlH<sub>4</sub>, Zn(BH<sub>4</sub>)<sub>2</sub>, HBCl<sub>2</sub>-Me<sub>2</sub>S, LiBH<sub>3</sub>NMe<sub>2</sub>, NaBH<sub>4</sub>/Phase Transfer Catalyst, NaBH<sub>4</sub>/NiCl<sub>2</sub>.6H<sub>2</sub>O, NaBH<sub>4</sub>/Ni(OAc)<sub>2</sub>, NaBH<sub>4</sub>/1,3-propanedithiol; sulfur compounds  
25 selected from HS(CH<sub>2</sub>)<sub>3</sub>SH/Et<sub>3</sub>N, PhSH; Phosphorous containing compounds .

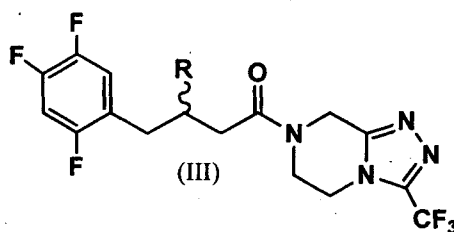
21. The process as claimed in claim 1 in step (e), wherein the suitable solvent used is selected from alcohols, ethers and water or their suitable mixtures.

22. The process as claimed in claim 1 in step (e), wherein the reaction is carried out at temperature ranging from 0 to 80 °C, preferably the temperature is 20 to 40 °C

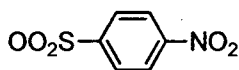
30 23. The process as claimed in claim 1, also including the step of, converting the compound of Formula (IV) directly to the compound of Formula (II) with inversion of configuration, by using suitable amine equivalent, optionally using suitable additives in suitable solvents.



24. The process as claimed in claim 23, wherein suitable amine equivalent used is selected from suitable cyclic imides, suitable aralkylamines, and suitable sulfonamides.
25. The process as claimed in claim 23, wherein suitable solvent used is selected from suitable ethers or their suitable mixtures.
- 5 26. The process as claimed in claim 23, wherein suitable additives used is selected from suitable phase transfer catalysts selected from trialkyl ammonium halides, tetraalkyl ammonium halides, trialkyl ammonium acetates, tetraalkyl ammonium acetates, trialkyl ammonium hydroxides, tetraalkyl ammonium hydroxides, trialkyl ammonium sulfates, tetraalkyl ammonium sulfates, trialkyl ammonium tetrafluoroborate, tetraalkyl ammonium tetrafuloroborate, trialkyl phosphonium halides, tetraalkyl phosphonium halides, tetrapenyl phosphonium halides, pyridinium halides, Aliquat, Adogen, crown ethers, silicon analogues of crown ethers, PEG, TDA-1.
- 10 27. The process as claimed in claim 1, also including the step of, converting the compound of Formula (III) directly to the compound of Formula (I) with inversion of configuration, by using suitable amine equivalent in suitable solvent.
- 15 28. The process as claimed in claim 27, wherein suitable amine equivalent used is selected from ammonium acetate, ammonium carbonate, ammonium hydroxide, ammonium formate, ammonium lactate, ammonium citrate dibasic, ammonium carbamate, ammonium benzoate.
- 20 29. The process as claimed in claim 27, wherein suitable solvent used is selected from alcohols, ethers and water or their suitable mixtures.
30. The compound of structural formula (III) in racemic (R/S) form or any of its optically active (S) or (R) forms or their enantiomerically excess mixtures

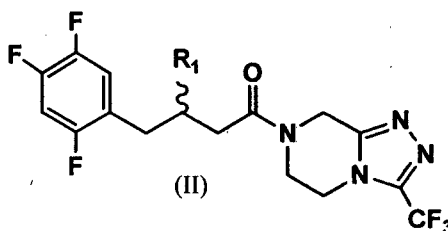


- 25 wherein 'R' represents suitable alkyl carboxylate group selected from  $-\text{OCOCH}_3$ ; haloalkyl carboxylate group selected from  $-\text{OCOCF}_3$ ; aryl carboxylate group selected from  $-\text{OCOPh}$ , substituted  $-\text{OCOPh}$ ; alkoxy carboxylate group selected from  $-\text{OC(O)OCH}_2\text{CH}_3$ ; alkyl sulfonates group selected from  $-\text{OSO}_2\text{CH}_2\text{CH}_3$ ; - haloalkyl sulfonate group selected from,  $-\text{OSO}_2\text{CF}_3$ ,  $-\text{OSO}_2\text{CH}_2\text{CF}_3$ ; aryl sulfonates group
- 30 selected from  $-\text{OSO}_2\text{Ph}$ , substituted  $-\text{OSO}_2\text{Ph}$  selected from



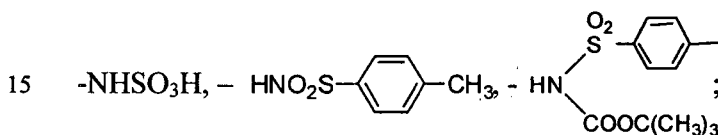
alkoxy phosphates group selected from  $-\text{OP}(\text{O})(\text{OEt})_2$ ; an oxyphosphonium ion leaving group selected from  $-\text{O}^+\text{P}(\text{X})_3$  where 'X' selected from alkyl groups, aryl groups, cycloalkyl groups.

31. A compound of structural formula (II) in racemic (R/S) form or any of its optically active (S) or (R) forms or their enantiomerically excess mixtures

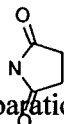


10

. Wherein 'R<sub>1</sub>' represents aralkylamino group selected from  $-\text{NHCH}_2(4\text{-OCH}_3\text{Ph})$ ,  $-\text{NHCH}(\text{CH}_3)\text{Ph}$ ,  $-\text{NHCH}(\text{Ph})_3$ ; sulfonamido group selected from,



imido group selected from -



32. The process for the preparation of compound Formula (I) or its salts either in its racemic (R/S) form or any of its optically active (S) or (R) forms or enantiomeric excess mixture of any of the forms comprising;

20 (i) reacting 3-hydroxy-1-(3-(trifluoromethyl)-5,6-dihydro-[1,2,4]triazolo[4,3-a]pyrazin-7(8H)-yl)-4-(2,4,5-trifluorophenyl) butan-1-one of Formula (IV) either in its racemic (R/S) form or any of its optically active (S) or (R) forms or enantiomeric excess mixture of any of the forms with a p-toluenesulfonyl chloride or p-nitro benzenesulfonyl chloride to obtain compound of Formula (III) wherein 'R' represents p-toluenesulfonyl group or p-nitro benzenesulfonyl group, with retention of configuration;

25 (ii) optionally, isolating the compound of Formula (III) wherein 'R' represents p-toluenesulfonyl group or p-nitro benzenesulfonyl group;

(iii) converting compound of Formula (III) wherein 'R' represents p-toluenesulfonyl group or p-nitro benzenesulfonyl group to compound of Formula (II), wherein 'R<sub>1</sub>' represents -N<sub>3</sub> group with inversion of configuration by using suitable azide, optionally with suitable additives in suitable solvents;

5 (iv) reducing the compound of Formula (II) wherein 'R<sub>1</sub>' represents -N<sub>3</sub> group to obtain compound of Formula (I) with retention of configuration.

33. The process as claimed in claim 32 in step (i), wherein suitable solvent used is selected from suitable chlorinated solvents, suitable nitriles, suitable hydrocarbons, suitable aprotic polar solvents, pyridine or their suitable mixtures.

10 34. The process as claimed in claim 32 in step (i), wherein suitable base used is selected from heterocyclic saturated or unsaturated amines selected from morpholine, piperidine, pyrrolidine, DBU, DBN, 1,4-diazabicyclo[2,2,2]octane, dimethylamino pyridine and pyridine; hydrides selected from KHMDS or mixtures of suitable bases.

35. A process as claimed in claim 32 in step (iii), wherein suitable azides is selected  
15 from HN<sub>3</sub>, NaN<sub>3</sub>, LiN<sub>3</sub>, Zn(N<sub>3</sub>)<sub>2</sub>, Cu(N<sub>3</sub>)<sub>2</sub>, tetrabutylammonium azide.

36. A process as claimed in claim 32 in step (iii), wherein suitable additives used is selected from suitable phase transfer catalysts selected from trialkyl ammonium halides, tetraalkyl ammonium halides, trialkyl ammonium acetates, tetraalkyl ammonium acetates, trialkyl ammonium hydroxides, tetraalkyl ammonium hydroxides,  
20 trialkyl ammonium sulfates, tetraalkyl ammonium sulfates, trialkyl ammonium tetrafluoroborate, tetraalkyl ammonium tetrafuloroborate, trialkyl phosphonium halides, tetraalkyl phosphonium halides, tetrapenyl phosphonium halides, pyridinium halides, Aliquat, Adogen, crown ethers, silicon analogues of crown ethers, PEG, TDA-1.

37. The process as claimed in claim 32 in step (iii), wherein the suitable solvent used is  
25 selected from suitable ethers, suitable chlorinated solvents, suitable nitriles, suitable hydrocarbons, aprotic polar solvents selected from DMF, DMA, DMSO; N-methyl pyrrolidone, HMPA, pyridine and water or their suitable mixtures.

38. A process as claimed in claim 32 in step (iv), wherein suitable reducing agent used is selected from metal catalyst selected from Pd/H<sub>2</sub>, Pt/H<sub>2</sub>, Pd/Al<sub>2</sub>O<sub>3</sub>, Pd-CaCO<sub>3</sub>,  
30 Pd(OH)<sub>2</sub>, PdO, PtO<sub>2</sub>, Pd-C/HCOOH, Pd-C/HCOONH<sub>4</sub>, Pd(OH)<sub>2</sub>-C/N<sub>2</sub>H<sub>4</sub>, Pd-C/NaBH<sub>4</sub>, Mg, Ca, Zn, Zn-HCl, Zn-CH<sub>3</sub>COOH, SnCl<sub>2</sub>, SnCl<sub>2</sub>/thiophenol, Zn/NH<sub>4</sub>Cl, Zn/AlCl<sub>3</sub>; Lindlar catalyst; Boron containing reducing agents selected from BH<sub>3</sub>, NaBH<sub>4</sub>, LiAlH<sub>4</sub>, Zn(BH<sub>4</sub>)<sub>2</sub>, HBCl<sub>2</sub>-Me<sub>2</sub>S, LiBH<sub>3</sub>NMe<sub>2</sub>, NaBH<sub>4</sub>/Phase Transfer Catalyst,

NaBH<sub>4</sub>/NiCl<sub>2</sub>·6H<sub>2</sub>O, NaBH<sub>4</sub>/Ni(OAc)<sub>2</sub>, NaBH<sub>4</sub>/1,3-propanedithiol; sulfur compounds selected from HS(CH<sub>2</sub>)<sub>3</sub>SH/Et<sub>3</sub>N, PhSH.

39. The process as claimed in claim 32 in step (iv), wherein the suitable solvent used is selected from alcohols, ethers and water or their suitable mixtures.

5

10

15

20

# INTERNATIONAL SEARCH REPORT

International application No <b>PCT/IN2012/000299</b>
--

<b>A. CLASSIFICATION OF SUBJECT MATTER</b> INV. C07D487/04 ADD.				
According to International Patent Classification (IPC) or to both national classification and IPC				
<b>B. FIELDS SEARCHED</b> Minimum documentation searched (classification system followed by classification symbols) C07D				
Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched				
Electronic data base consulted during the international search (name of data base and, where practicable, search terms used) EPO-Internal, WPI Data, CHEM ABS Data				
<b>C. DOCUMENTS CONSIDERED TO BE RELEVANT</b>				
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.		
Y	WO 2010/122578 A2 (MSN LAB LTD [IN]) 28 October 2010 (2010-10-28) cited in the application	1-31		
A	page 15 - page 16 page 35 - page 36 schemes 2, 8 examples 16,18,19,45,47,48,55-58,61 page 16, line 1 - line 2 claims 3, 31	32-39		
Y	----- WO 2006/045598 A1 (IEP GMBH [DE]) 4 May 2006 (2006-05-04) examples 1-4 claim 6 -----	1-31		
----- -/--				
<table style="width: 100%; border: none;"> <tr> <td style="width: 50%; border: none;"><input checked="" type="checkbox"/> Further documents are listed in the continuation of Box C.</td> <td style="width: 50%; border: none;"><input checked="" type="checkbox"/> See patent family annex.</td> </tr> </table>			<input checked="" type="checkbox"/> Further documents are listed in the continuation of Box C.	<input checked="" type="checkbox"/> See patent family annex.
<input checked="" type="checkbox"/> Further documents are listed in the continuation of Box C.	<input checked="" type="checkbox"/> See patent family annex.			
* Special categories of cited documents :				
"A" document defining the general state of the art which is not considered to be of particular relevance "E" earlier application or patent but published on or after the international filing date "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified) "O" document referring to an oral disclosure, use, exhibition or other means "P" document published prior to the international filing date but later than the priority date claimed	"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone "Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art "&" document member of the same patent family			
Date of the actual completion of the international search	Date of mailing of the international search report			
30 January 2013	05/02/2013			
Name and mailing address of the ISA/ European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Authorized officer  Cortés, José			

## INTERNATIONAL SEARCH REPORT

International application No

PCT/IN2012/000299

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 2011/025932 A2 (REDDYS LAB LTD DR [IN]; REDDYS LAB INC DR [US]) 3 March 2011 (2011-03-03) examples 4-7 -----	31
X	BECONI, MARIA G. ET AL: "Disposition of the dipeptidyl peptidase 4 inhibitor sitagliptin in rats and dogs", DRUG METABOLISM AND DISPOSITION, vol. 35, no. 4, 2007, pages 525-532, XP002686516, ISSN: 0090-9556, DOI: 10.1124/dmd.106.013110 page 526; compound M1 -----	31
X,P	WO 2012/046254 A2 (CADILA HEALTHCARE LTD [IN]) 12 April 2012 (2012-04-12) examples 11-18 claims 1, 31, 32 -----	1-39
X,P	EP 2 397 141 A1 (LEK PHARMACEUTICALS [SI]) 21 December 2011 (2011-12-21) cited in the application scheme 6 examples 6-8 claim 12 -----	1-39
E	WO 2012/072036 A1 (ZHEJIANG HISOAR PHARMACEUTICAL CO LTD [CN]) 7 June 2012 (2012-06-07) page 45; example 51 -----	31

# INTERNATIONAL SEARCH REPORT

International application No.  
PCT/IN2012/000299

## Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1.  Claims Nos.:  
because they relate to subject matter not required to be searched by this Authority, namely:
  
2.  Claims Nos.:  
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:
  
3.  Claims Nos.:  
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

## Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

see additional sheet

1.  As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.
2.  As all searchable claims could be searched without effort justifying an additional fees, this Authority did not invite payment of additional fees.
3.  As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:
4.  No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

### Remark on Protest

- The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.
- The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.
- No protest accompanied the payment of additional search fees.

**FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210**

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. claims: 1-31

the process of claim 1 and intermediates thereof  
---

2. claims: 32-39

the process of claim 32  
---



# INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No PCT/IN2012/000299
---

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 2010122578	A2	28-10-2010	NONE
WO 2006045598	A1	04-05-2006	AT 501928 A1 15-12-2006
			CA 2585411 A1 04-05-2006
			CN 101120094 A 06-02-2008
			EP 1805313 A1 11-07-2007
			JP 2008517612 A 29-05-2008
			KR 20070085458 A 27-08-2007
			US 2009148917 A1 11-06-2009
			WO 2006045598 A1 04-05-2006
WO 2011025932	A2	03-03-2011	EP 2470542 A2 04-07-2012
			WO 2011025932 A2 03-03-2011
WO 2012046254	A2	12-04-2012	NONE
EP 2397141	A1	21-12-2011	NONE
WO 2012072036	A1	07-06-2012	CN 102485718 A 06-06-2012
			WO 2012072036 A1 07-06-2012