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A practical large-scale synthesis of iohexol

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Abstract
A facile large scale synthesis developed for high pure iohexol 1 using an inexpensive synthetic route consist a total of 6 steps and easily isolatable intermediates. Key to the strategy is the preparation and isolation of 5-[*N*-(2,3-Dihydroxypropyl)acetamido]-2,4,6-triiodo-*N*,*N'*-bis(2,3-dihydroxypropyl)isophthalamide (crude iohexol, 9) in good yield using an efficient and practical protocol. The main advantages of the route include a straight forward reaction sequence and good overall yields at every stage. The process impurities have been well controlled without using any resins. Two best methods have proposed to eliminate *o*-acetyl and process impurities. The structures of all intermediates were confirmed by ^{1}H-NMR, ^{13}C-NMR, IR and MS and HPLC.

Keywords: Iohexol, x-ray contrast agent, synthesis, large scale, without resins, high pure

Introduction

Iodinated x-ray contrast media are among the most widely used pharmaceuticals for intravascular administration. More than 600 million x-ray examinations are conducted yearly and about 75 million of these procedures are performed with use of a contrast medium [1]. The available intravascular contrast media in the market are tri-iodinated benzene derivatives with iodine atoms in positions 2, 4 and 6. The other ring positions are occupied by side chains aimed at giving the contrast medium high water solubility which provides the enhanced contrast effect [2a]. All of these products depend on the presence of large quantities of iodine in the molecule to provide an *opaque* background, allowing their use to visualize internal organs. However, the quantities needed for this purpose are extremely high, being many times higher than those normally needed for compounds used as medicines [2b].

There are currently four types of products commercially available. These are classified into the ionic monomers, the ionic dimer, the non-ionic monomers and the non-ionic dimer [2c]. Most of newer generation products are of the non-ionic monomer in nature. These products have long side chains rich in hydroxy groups evenly distributed around the benzene core. Two of these side chains are usually identical, giving the compounds a divalent structure. Different products are very similar structure and may have one or two side chains in common. All types of compounds are of low molecular weight, are highly water-soluble, have low protein binding capacity. The search for best radiographic contrast media with more favorable properties than currently available ones led to the identification of iohexol.

Iohexol 1, [5-[*N*-(2,3-dihydroxypropyl)acetamido]-*N*,*N'*-bis(2,3-dihydroxypropyl)-2,4,6-tri iodoisophtalamide (Figure-1) is chemical drug substance of a non-ionic iodinated x-ray contrast agent which is one of the most used agents in diagnostic x-ray procedure [3a]. This tri-iodinated nonionic contrast agent, yields highly concentrated solutions with the best combination of low osmolality and low viscosity found to date. It has an attractive pharmacological profile with excellent clinical efficacy and extremely low toxicity and minimal interference with the normal functions of the organism [3b, c]. It is on the World Health Organization's list of essential Medicines, the most effective and safe medicines needed in a health system [3d-f]. Iohexol is administered at high doses and therefore it must have extremely high requirements of purity [4] and had been the pioneer molecule in this field and still is one of the most employed worldwide [5].

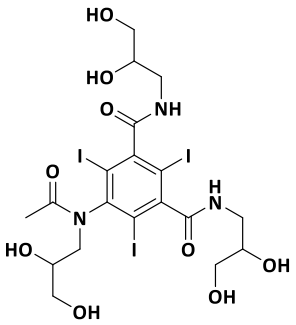
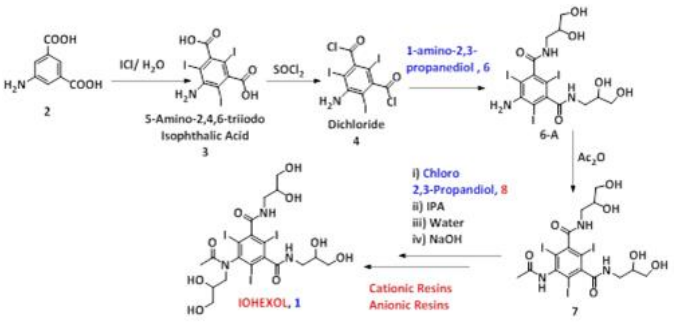


Fig 1: Iohexol

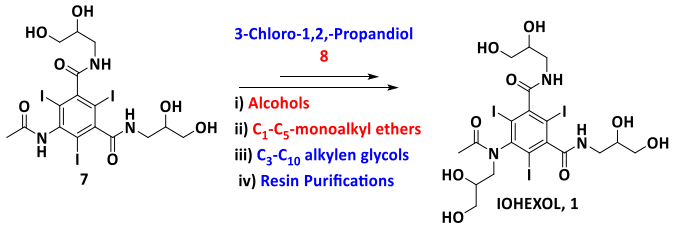
Several methods have been disclosed in the literature for the synthesis of Iohexol [6-9]. In general, iohexol synthesis involved seven stage linear syntheses and followed by *N*-alkylation of amide with 3-chloro-1,2-propanediol which determines the structure (Scheme 1).



Scheme 1: Commercial manufacturing of iohexol.

Most of the patents describes the synthesis of iohexol starting from 5-amino-2,4,6-triiodo isophthalic acid, 3. The preparation of 3 is a difficult task as observed practically in our large scale batches since the iodination never ends with desired product alone but always associated with mono and di iodo moieties as impurities in good amount. The generic patents revealed iodination techniques [10] always show discrepancy either in yield or in the quantity of mono and di iodo impurities in this stage. In order to obtain best purities in iohexol, its needs to be purified every stage in entire process (Scheme-1). Although, one pot synthesis of iohexol starting from compound 4 without isolating the intermediates has been described [11], this process apparently fails in explaining to achieve desired purity as required by pharmacopeia.

Another problem exists in the available manufacturing methods in the final reaction step is, the conversion of compound 7 to iohexol, 1 by means of 3-chloro-1,2-propanediol 8 in presence of alkali is cumbersome and large quantities of sodium chloride thus formed requires high volumes of ion-exchange resins for its removal which results inevitable increase of cost and loss of product. It was found that most of the readily available synthetic procedures describe the control over *N*-alkylation in the final step (Scheme-2) followed by appropriate high volume resin purifications to eliminate process impurities and get compound 1 in high purity. Some another procedures reveal the use of 2-methoxyethanol or mixtures of 2-methoxyethanol/ isopropanol and a solvent chosen from a C_1 to C_5 monoalkyl ethers of a C_3 to C_{10} alkaline glycols are respectively [12] as solvents in which the *N*-alkylation compound **7** to be carried out.



Scheme 2: Preparation and purification of iohexol.

Despite of difficulties mentioned above, there is a major problem observed is that the material is very hygroscopic and picks up the atmosphere moisture with in no time. To avoid, various solvent systems had considered adequate for the crystallization of 1. The first of these systems comprises the use of *n*-butanol [13a]. In this case; the so-obtained product required subsequent dissolution in water, followed by evaporation to dryness under vacuum, in order to remove the residual butanol from the crystallized product.

To address the purification problems, in general, chromatographic methods are best choice despite the chemical methods, such as crystallization or washing of a suspension. But they do not always allow the successful removal of impurities with molecular structures that are very similar to that of the primary product. In the very case of iohexol, the most difficult impurities are *o*-alkylated derivatives [13b,c]. In terms of the molecular configuration in the crystal, the properties of these substances are very similar to those of iohexol, being easily introduced in the crystalline structure, with the consequent difficulty in their removal. Another general consideration is the fact that the molecules used as x-ray contrast media are extremely soluble in aqueous media, and hence can be difficult to crystallize. It is due to the high degree of freedom associated with the hydrophilic side chains containing alcohol functions [14].

In order to make these procedures suitable for large-scale production of iohexol 1, several issues should be addressed. First, the iodination step (a) usage of ICl in triiodination of isophthalic acid is major problem mainly due to the corrosive properties of the iodinating agents and to their limited storage life. In addition, the presence of chlorine atoms within the iodinating agents themselves may lead to side-reactions and, thus, to the undesired formation of chlorine side-products as pharmacopeias listed impurities [15a]. Second, addition of 3-chloro-1,2-propanediol 8 for *N*-acetylation of 5-acetamido group where very possibility of generation *o*-alkyl impurities and these impurities substantially leads the material in end with low purity. Moreover, iohexol 1 is highly sensitive to atmospheric moisture and thus warrants high scale recrystallizations [15b]. Third, some solvents used in these procedures, such as C_1 to C_5-monoalkyl ethers, C_3 to C_{10} glycols, may participate in to the crystal lattice of final molecule which leads material to be substandard [15c]. Finally, usage of high volume resins and series of purifications are cumbersome and even may not promise the desired quality in bulk scale preparations for regulatory market.

After considering above all, a commercial route was hence developed that consists of a total five synthetic steps with all isolated intermediates starting with compound 3 [19] (Scheme-3). All intermediates prepared with good purity and characterized which otherwise very difficult to obtain from the commercial sources for low cost.

Results and Discussion

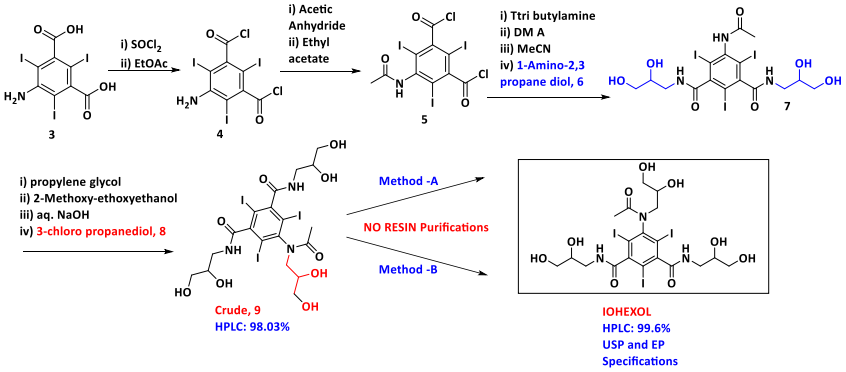
The key step in manufacture of iohexol in a linear synthesis is the penultimate stage where crude iohexol 9 is obtained from the reaction between compounds 7 and 8 at ambient temperature in higher alcohols. Thus obtained crude usually recrystallized twice from higher alcohols. To improve the *N*-alkylation and subsequent purification steps have been disclosed by patent literature by using 2-methoxyethanol[16a] and optionally isopropanol both in the alkylation step, or 1-methoxy-2-propanol [16b] as the solvent optionally in a mixture with other solvents. The *N*-alkylation step where 9 in solution is reacted with an alkylating agent such as 3-chloro-1,2-propandiol 8 to introduce the 2,3-dihydroxypropyl group at

the nitrogen of the 5-acetamido group is illustrated in Scheme-1.

Apparently, the *N*-alkylation step is challenging because the considerable amount of *o*-alkylated byproducts can also be formed when the alkylation occurs at the oxygen atoms of the hydroxyl groups [16c]. It is therefore a desire to limit the formation of impurities and thereby to limit their presence in the final purified iohexol. The upper limit for values for *o*-alkylated ones in the end product is fixed by the European Pharmacopeia to 0.6%[18] (HPLC by area). Further unidentified byproducts also referred as impurities which are also formed during the alkylation reaction and must be reduced to a tolerable level. In addition the solvents used should be easily available, be environmentally friendly and be of low toxicity.

There is, therefore a necessity to identify solvents which can be used in the *N*-alkylation reaction. It is further desired to improve the overall process including the *N*-alkylation step and the purification step in the manufacture of iohexol. If the crude product obtained by the *N*-alkylation step is to be recrystallized from a solvent that is different from the solvent used in the reaction, then it must first be completely removed e.g. by evaporation to dryness. It is known from crystallization theory and experience that even small quantities of residual solvents from previous steps may cause a crystallization process to get out of control due to changes in its supersaturating conditions, and thorough removal of the reaction solvent is an important step. Solvent removal is an energy consuming operation which also risks degradation of the product due to exposure to elevated temperature.

To address all observed difficulties, by introducing some facile modifications, a large scale preparation of iohexol described herein as covered in our recent patent[17] (Scheme **3**). The process is made simpler because the purification and hydrolysis are, in practice, carried out in a single step. The yields of the hydrolysis reaction are practically quantitative and the reaction itself does not give rise by-products which otherwise very difficult to remove in the final molecule. And overall yield of the process is good and, above all, the purity characteristics of the resultant product completely as per pharmacopeias requirements [18a].



Scheme 3: Preparation of High purity Iohexol.

The process of the present work yields 1 with purity 99.52% to 99.63% along with isolated yield in the range of 85% to 87%. Compound 3 [19] reacts with thionyl chloride in presence of EtOAc yields compound 4. Then compound 4 acylated with acetic anhydride in ethyl acetate to yield 5 which further reacts with two equivalents of 1-amino-2,3-propanediol 6 in DMA and MeCN mixture in presence of tri butylamaine to yield stable compound 7 in good yield. Compound 7, then reacts with 3-chloro-2,3-propanediol 8 in the presence of aq. NaOH and after workup gives off crude iohexol 9, in 70% yield and a minimum purity of 96.13%.

Although, various purification procedures readily available, [6-15] none of them works out to yield satisfactorily within the range of commercial viability. Another problem essentially address is that the complete elimination of resins which renders the process of the present invention economical over other the available processes.

Hence, we have chosen a different strategy to achieve ultrapurity and best yields by means of chemical purification methods by eliminating usage of any resins by protecting hydroxy groups in the crude molecule 9. This strategy found and proved to be very advantageous since the molecule turned out to be more of organic in nature so that hydrophilic structural analogues of iohexol will be completely washed away in water.

Two different strategic methods were tried to reach pharmaceutically accepted material. In Method-A where the crude iohexol is acylated with acetic anhydride with a catalytic amount of TFA and thus formed solid hexaacetyl iohexol 10 is filtered off and washed with chilled water then hydrolyzed with aq. ammonia to get iohexol 1 with 99.42% purity. Although this method allowed to get rid of maximum impurities from iohexol 1, but due to very possible incomplete hydrolysis, either in acid or base, always associated with some quantity of *o*-acetyl impurities which are detrimental to the purity of the material.

Hence acetal protection was chosen as an alternative by using acetone in Method-B where the crude compound 9 treated with acetone in 2,2-dimethoxy propane and *p*-TSA as catalyst. Thus formed triacetal iohexol 11 was isolated as a solid, as expected, after hydrolysis with aq. HCl, iohexol 1 yielded with an astonishing purity as per regulatory requirement [18] which warrants no further purifications. Thus obtained iohexol is ultrapure with kept *o*-acetyl impurities as minimum as <0.20%.

Conclusion

In summary, we have developed a highly inexpensive

approach for the preparation of iohexol and its intermediates for large scale synthesis with very high purity. This achieved with an easily accessible intermediate sequence, and newly introduced chemical purifications by limiting impurities [18]. The series of laborious purifications and usage of high volumes of resins have been completely eliminated. The proposed routes fully satisfied our aim of developing a safe, convenient, cost-effective, high-yielding, and environment-friendly process. We are confident that it should be valuable in the industrial manufacturing of iohexol for requirement of x-ray contrast agent.

Experimental Section

General. Compounds 5 prepared according to literature [19] and solvents were obtained from commercial suppliers and used without further purification. IR spectra were recorded Shimadzu FT-IR instrument; ^{1}H-NMR spectra were measured with reference to an internal standard of TMS at 0 ppm and ^{13}C-NMR spectra measured with DMSO signal at 39.5 ppm. The HPLC analysis was performed with an Agilent 1100 instrument with DAD detector.

Preparation of 5-amino-2,4,6,-triiodo isopthaloyl dichloride (4)

A stirred solution of 5-amino-2,4,6-triiodoisopthalic acid 3 [19] (201 kg, 0.359 Kmol), tertiary butyl ammonium bromide (200 g) in 2 L of ethyl acetate was slowly added thionyl chloride (222 kg, 1.865 Kmol) at room temperature. Then the temperature of mass increased to 75° C and stirred for 20 h. The reaction was cooled to 10°C and added 150 L of DM water. Then the ethyl acetate layer was washed with 50 L of (5%) sodium bicarbonate solution followed by 50 L of saturated ammonium chloride solution. The organic layer thus collected and evaporated to gives off compound 4 (182 kg, 85%). HPLC purity: 97.97%, (M+H): 596.9; IR (KBr): 3500, 3020, 1725 cm^{-1}; ^{1}H-NMR: (300 MHz, DMSO-d6), δ 6.11 (2H); ^{13}C-NMR (300 MHz, DMSO-*d6*), δ 169.55, 149.06, 148.92, 78.39, 66.17.

Preparation of 5-acetamido-2,4,6-triiodoisophthaloyl dichloride (5)

Compound 4 (18.2 kg, 0.0305 Kmol) dissolved in 108 L of ethyl acetate. Then the temperature of mass increased to 40 to 45°C and added acetic anhydride (9.3 kg, 0.091 Kmol) over 30 minutes. After stirring at ambient temperature for 1 h, reaction mass was cooled to 25°C and added 20 L of DM water. The organic layer was washed with 20 L of saturated ammonium chloride solution. The organic layer thus collected was evaporated to gives off **5** with a yield of 90.0% (17.5 Kg) with 99.34 % of purity. (M+H): 638.3; IR (KBr): 3100 cm$^{-1,}$ 3020, 1725, 1685; ^{1}H-NMR: (300 MHz, *DMSO-d$_6$*) δ : 2.33(3H), ^{13}C-NMR (300 MHz, *DMSO-d$_6$*) δ : 169.5, 169.4, 169.3, 168.1, 150.6, 149.1, 146.1, 143.7, 143.6, 22.6.

5-acetamido-N,N-bis(2,3-dihydroxypropyl)-2,4,6-triiodoisophthalamide (7)

A stirred solution of compound 5 (17.5 kg, 0.0234 Kmol) in 70L of MeCN and 18 L of DMF was slowly added tributylamine (9.3 kg, 0.0513 Kmol) at room temperature. Then the temperature of mass increased to 50 to 55° C and added 3-amino-1,2-propanediol **6** (5.3 kg, 0.0585 kmol) in 10L of DMF over about 3 h. Then the temperature of mass increased to 75° C and stirred for another 4 h. The organic solvents completely distilled off and reaction mass cooled to RT and charged 50 L of DM water with stirring. The pH of the mass adjusted to 4 to 4.5 with hydrochloric acid. The precipitated solid **7** filtered and washed with 10 L of cold water. Yield: 14.7 kg (72.0%); HPLC purity: 98.21%. (M+H) 748.2; IR (KBr): 3100, 3020, 1725, 1685, ^{1}H-NMR: (300 MHz, *DMSO-d6*) δ, 9.95 (1H); 8.52-8.49 (2H), 4.71 (2H), 4.51(2H); 3.69 (2H), 3.46(4H), 3.32-3.26 (2H), 3.17-3.13 (2H), 2.01 (2H); ^{13}C-NMR (300 MHz, *DMSO-d$_6$*) 169.6, 167.7, 149.9, 143.2, 99.4, 99.3, 99.1, 99.0, 90.1, 69.9, 63.9, 42.6, 42.5, 23.0.

Preparation of Crude Iohexol (9)

Compound **7** (14.7kg, 0.0196 Kmol) dissolved in the mixture of 22.5 L of 2-(2-methoxyethoxy) ethanol and 7 L of propylene glycol. The reaction mass cooled to 18°C to 25°C and sodium hydroxide (1.30 kg, 0.0323 kmol) in 650 mL of DM water was slowly added. Then temperature of reaction was increased to 40°C and stirred for 2 h. After that, 3-chloro-1,2-propanediol 8 (3.2 kg, 0.0294 Kmol) added slowly and stirred for 24 hours. Then reaction mass cooled 25°C and added 5 L of methanol. The *p*H of the mass adjusted to 4.5 and extracted with chloroform. The aqueous layer was collected and distilled out to get crude iohexol 9. Yield: 15.3 kg, (95%). HPLC purity: 96.13%.

Preparation of hexaacetyl iohexol (10)

A stirred solution of 70 L of DMA was added 10 g of DMAP, 10 g of TFA and compound **9** (15.3 kg, 0.0186 Kmol) and mixed well. The temperature of reaction cooled to 0° to 5°C. Then slowly added acetic anhydride (14.2 kg, 0.139 kmol) and stirred for about 8 h. Then *p*H of the reaction was adjusted to 7.5 with aq. NaOH and distilled off the solvent then slurred with hot water to get desired compound as a solid. Yield: 18.1 kg, (91%).

Synthesis of Iohexol (Method-A)

Compound 10 (18.1 kg, 0.0224 kmol) dissolved in 18 L of DM Water was added 56 L of 15% Ammonium hydroxide and mixed well at 35°C. The temperature of reaction mass was then raised to 60° to 65°C and stirred for about 9 h. After the temperature of the mass cooled to 35°C and added 3 L of DCM while stirring. Then pH of the mass adjusted to 7.2 and thus obtained aqueous layer was collected and completely distilled off. The resulted residue dissolved in 4 L of *iso*-butanol to yield 1 in 11.90kg, (86%) HPLC purity: 99.41%, (M+H) 822.4; IR (KBr): 3100 cm^{-1}, 3020 cm^{-1}, 1685 cm^{-1}: ^{1}H-NMR: (300 MHz, *DMSO-d$_6$*) δ: 8.58 - 8.50 (d, 2H), 4.76-4.75 (d, 2H), 4.61-4.55 (d, 4H), 3.92-3.83 (m, 3H), 3.70-3.34 (m, 6H), 3.32-3.10 (m, 6H), 1.75 (s, 3H); ^{13}C-NMR (300 MHz, *DMSO-d$_6$*) 170.9, 170.5, 170.5, 169.0, 151.4, 151.1, 147.8, 147, 100.1, 91.7, 70.2, 69.9, 69.7, 64.6, 64.4, 53.6, 53.1, 42.6, 42.4, 22.9.

Preparation of acetal protected iohexol (11)

A stirred solution of 70 L of acetone and 7.75 L of 2,2-dimethoxy propane, 100 g of *p*-TSA, was added compound 9

(15.3 kg, 0.0186 Kmol) and mixed well. The temperature of reaction mass stirred for 8 h at 50°C. Then reaction was cooled to 0 to 5°C and *p*H of the reaction mass adjusted to 6.5 to 7.5 with 50% NaOH. After removal of the solvent, slurred with 15 L of water at 40°C to get 11 as a solid. Yield: 15.7 kg, (90%).

Synthesis of Iohexol (Method-B)

Compound 11 (15.7 kg, 0.0167 kmol) dissolved in 15.7 L of DM Water contain 6.2 L of Con. HCl and mixed well at 35°C. Then temperature of reaction mass was increased to 60° to 65°C and stirred 1 h. After cooling the reaction mass to 35°C added 2 L of DCM. Then reaction mass pH was adjusted to 7.2 with aqueous ammonia. Thus obtained aqueous layer was collected and completely distilled off and slurried in 4 L of *iso*butanol to get 1 in pure. Yield: 11.60 kg, (85%); HPLC purity: 99.52%, (M+H) 822.4; IR (KBr): 3100 cm^{-1}, 3020, 1685: ^{1}H-NMR: (300 MHz, *DMSO-d*$_6$) δ: 8.58-8.50 (d, 2H), 4.76-4.75 (2H), 4.61-4.55 (4H), 3.92-3.83 (3H), 3.70-3.34 (6H), 3.32-3.10 (6H), 1.75 (3H), (D_2O): δ: 3.92-3.83 (3H), 3.70-3.34 (m, 6H), 3.32-3.10 (6H), 1.75 (s, 3H), ^{13}C-NMR (300 MHz, *DMSO-d*$_6$) 170.9, 170.5, 170.5, 169.0, 151.4, 151.1, 147.8, 147, 100.1, 91.7, 70.2, 69.9, 69.7, 64.6, 64.4, 53.6, 53.1, 42.6, 42.4, 22.9.

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References

1. (a) Amersham Health, Annual Report, 2002 (b) Lars-Göran W, Golman K, Radner F. *EP0828705A1*, 1996. (c) Lusic H, Grinstaff MW. Chem. Rev. 2013; 113:1641-1666, (c) Alvarez PCM, Du BVG, José Ganchas CA. *EP0919540A1*, 1999. (d) Krause W, Schneider PW. Topics in current chemistry. 2002, 107–150, Krause, W. (Ed.). Springer, Berlin. (e) Felder E, Grandi M, Pitrè D, Vittadini G. Analytical Profiles of Drug Substances, 17, K. Florey (Ed.), 115–154, 1988, Academic Press, San Diego.
2. (a) Bonnemain, B, Meyer D, Schaefer M, Dugast-Zrihen M, Legreneur S, Doucet D. *Invest Radiol*. 25, S104–S106. 1990 (b) Steger-Hartmann, T.; Länge, R.; Schweinfurth, H. Products in the Environment. 2005; 230-243, (c) Almen T. Invest. Radiol., 1994, 29, S37–S45. (d) Christiansen, C. *Toxicology*. 2005; 209:185-187.
3. (a) Katayama H, Heneine N, Van Gessel R, Taroni P, Spinazzi A. Investigative Radiology. 2001; 36:22-32, (b). Bonati F, Felder E, Tirone P. Invest Radiol. 1980; 15:S310-S316, (c) Drayer BP, Warner MA, Sudilovsky A, Luther J, Wilkins R, Allen S, *et al. Neuroradiology*. 1982; 24:77-84. (d) *WHO* List of Essential Medicines. 2015; (19). (e) Broe ME, Porter GA, Bennett WM, Verpooten GA. Drugs and Chemicals. 2013; 325, (f) Nordal, V; Holtermann, H. DE 2726196, 1983.
4. Pitrè D, Felder E. *Invest. Radiol*. 1980; 15:S301-336.
5. Felder E, Grandi M, Pitrè D, Vittadini G. Analytical Profiles of Drug Substances. 17, K. Florey (Ed.). 1988; 115-154. Academic Press, San Diego.
6. Sørenssen DM, Ingvoldstad OE, Myrbråten E. *WO1998008804 A1*, 1998.
7. Nordal V, Holtermann H. *US4250113 A*, 191.
8. Villax GDB, Jose A, De Carvalho G, Perez CMA. *US5204086*, 2002.
9. (a) Alvarez PCM, Du BVG, José GCA. EP0919540 B1, 2002. (b) José GSM, Bandarra J, Heggie H. US7541494 B2, 2009. (c) Holmaas, LT, Homestad OM. EP1641743B1, 2008.; (d) Ranga Reddy, K.; Vijayabhaskar Reddy, E. Indian Patent 2016/41042424, 2016.
10. (a) Felder E, Pitre DE. US4001323A, 1977. (b) Helen GF, US2820814, 1958. (c) Hu Z, Xing H, Li L. WO2013063737A1, 2013.
11. Turchetta S, Massardo P, Aromatario V. CA2443730, 2011.
12. (a) Bailey AR, *et al*. WO2011041275A1, 2011. (b) Galindro J, Marto S, Bandarra J, Heggie W. US7541494B2, 2009.
13. (a) Vegard Nordal, Hugo Holtermann, US4250113A, 1981. (b) USP Revision Bulletin. 2011; 1-2, (c) Turchetta S, Massardo P, Aromatario V. US6897339B2, 2005. (d) Turchetta S, Massardo P, Aromatario V. EP1395546 B1, 2008.
14. Villax GDB, Perez GA, De Carvalho CMG, Jose A. EP0 919 540A1, 1999.
15. (a) USP Monographs. 2017; 40:4649-4652.
16. (a) José Galindro, *et al.* US7541494 B2, 2009. (b) Turchetta S, Massardo P, WO2002083623A1, 2002. (c) Bailey AR, *et al.* US20120184772, 2012.
17. Ranga Reddy K, Vijayabhaskar Reddy. Indian *Patent* 2016/41012424, 2016.
18. EP monographs 9.0. 2017; 1114:2791-2795.
19. Ranga Reddy K, Vijayabhaskar Reddy E, Org. Proc. Res. Dev. 2017(submitted).