Journal of Pharmaceutical and Biomedical Analysis 48 (2008) 708–715

Contents lists available at ScienceDirect

Journal of Pharmaceutical and Biomedical Analysis

journal homepage: www.elsevier.com/locate/jpba

Identification, synthesis, isolation and spectral characterization of potential impurities of montelukast sodium

M. Saravanan, K. Siva kumari, P. Pratap Reddy, M.N. Naidu, J. Moses Babu, Alok Kumar Srivastava, T. Lakshmi Kumar, B.V.V.N. Chandra Sekhar, Bollikonda Satyanarayana*

Dr. Reddy's Laboratories Ltd., IPDO-IPM Communication No. 00056, Hyderabad, India

ARTICLE INFO

Article history:
Received 22 April 2008
Received in revised form 1 July 2008
Accepted 2 July 2008
Available online 16 July 2008

Keywords:
Montelukast sodium
Preparative HPLC
Leukotriene
Bronchial asthma
LC–MS

ABSTRACT

During the process development of montelukast sodium, three polar impurities and one non-polar impurity with respect to montelukast sodium were detected by simple reverse phase high-performance liquid chromatography (HPLC). Initially, all the four impurities were identified by the liquid chromatography–mass spectrometry (LC–MS) data and out of four impurities, three have been prepared by the synthetic method and remaining one is isolated by preparative HPLC. Based on the spectral data (IR, ^{1}H NMR, ^{13}C NMR and MS), the structure of these impurities 1–4 were characterised as 1-[[[(1*R*)-1-[3-[(1*E*)-2-(7-chloro-2-quinolinyl)ethenyl]phenyl-3-[2-(1-hydroxy-1-methylethyl)phenyl]propyl]thio]methyl]cyclopropane acetamide (impurity-1), {1-[1-{3-[2-(7-chloro-quinolin-2-yl)-vinyl]-phenyl}-3-(2-isopropenyl-phenyl)-propylsulfanylmethyl]-cyclopropyl}-acetic acid (impurity-2), 1-[[[(1*R*)-1-[3-[(1*E*)-2-(7-chloro-2-quinolinyl)ethyl]phenyl-3-[2-(1-hydroxy-1-methylethyl)phenyl]propyl]thio]methyl]cyclopropaneacetic acid (impurity-3) and 1-[[[(1*R*)-1-[3-[(1*E*)-2-(2-quinolinyl)ethenyl]phenyl-3-[2-(1-hydroxy-1-methylethyl)phenyl]propyl]thio]methyl]cyclopropaneacetic acid (impurity-4).

1. Introduction

Montelukast sodium [1], a specific cysteinyl leukotriene receptor antagonist [2], belongs to quinoline series with the chemical name of 1-[[[(1*R*)-1-[3-[(1*E*)-2-(7-chloro-2-quinolinyl) ethenyl]phenyl-3-[2-(1-hydroxy-1-methylethyl)phenyl]propyl] thio]methyl]cyclopropaneacetic acid monosodium salt. Merck & Co. developed it, as a successful therapeutic agent for the treatment of bronchial asthma [3].

The presence of impurities or its related compounds in a drug substance can have a significant impact on the quality and safety of the drug product. During the process development of montelukast sodium, four impurities were observed in the range of 0.05–0.15% level along with the main product peak in the HPLC analysis. As per the general guidelines recommended by ICH [4] to qualify the drug substance, the amount of acceptable level for a known and unknown related compound (impurity) should be less than 0.15 and 0.10%, respectively. In order to meet the stringent regulatory requirements, the impurities present in the drug substance must be identified and characterised. Hence, a comprehensive study was undertaken to identify, synthesize and characterise these four impurities of montelukast sodium. In this article, we report synthesis, isolation and spectral characterization of impurities obtained during our process development of montelukast sodium [5] (Fig. 1).

2. Experimental

2.1. Samples and chemicals

The investigated samples of montelukast and impurities 1–3 were synthesized and impurity-4 was isolated through preparative HPLC in the laboratory after identification by HPLC and determination of molecular weight by liquid chromatography–mass spectrometry (LC–MS). HPLC grade acetonitrile and acetic acid were obtained from Merck, Mumbai, India. AR grade sodium dihydrogen phosphate, phosphoric acid and triethylamine were obtained from SD Fine Chemicals Limited, Mumbai, India. Water used for the preparation of mobile phase was purified using Millipore Milli-Q plus (Milford, MA, USA) purification system. Chloroform-d and dimethylsulfoxide-d_6 were purchased from Aldrich Chemicals Co., USA.

2.2. High-performance liquid chromatography (HPLC)

An in-house LC gradient method was developed for the separation of all possible related substances (impurities) of montelukast

* Corresponding author.
E-mail address: satyanarayanab@drreddys.com (B. Satyanarayana).

0731-7085/$ – see front matter © 2008 Elsevier B.V. All rights reserved.
doi:10.1016/j.jpba.2008.07.007

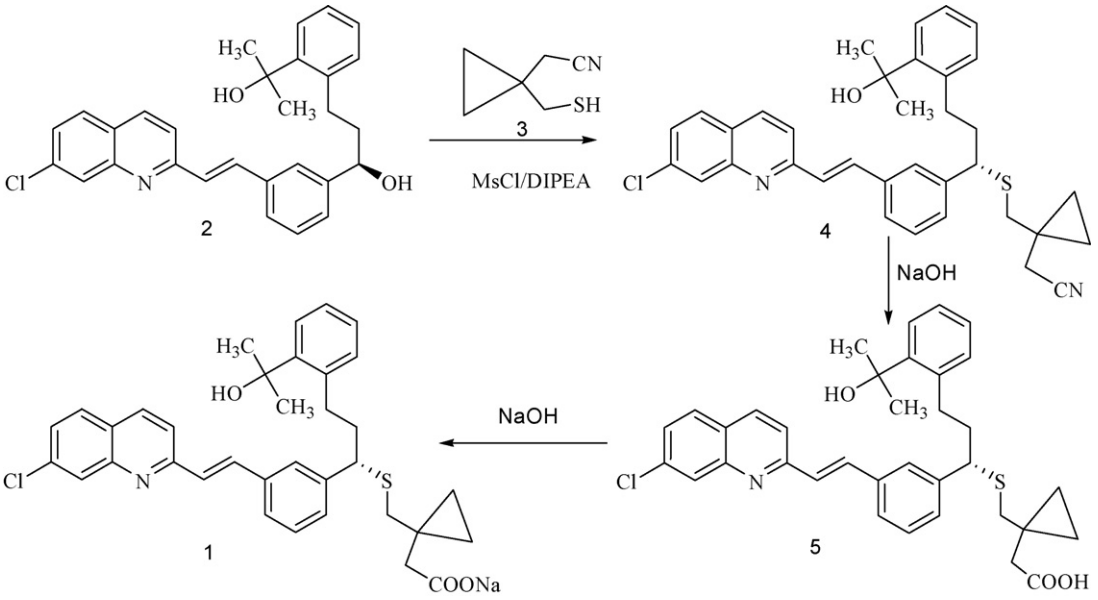


Fig. 1. Synthetic scheme for montelukast sodium.

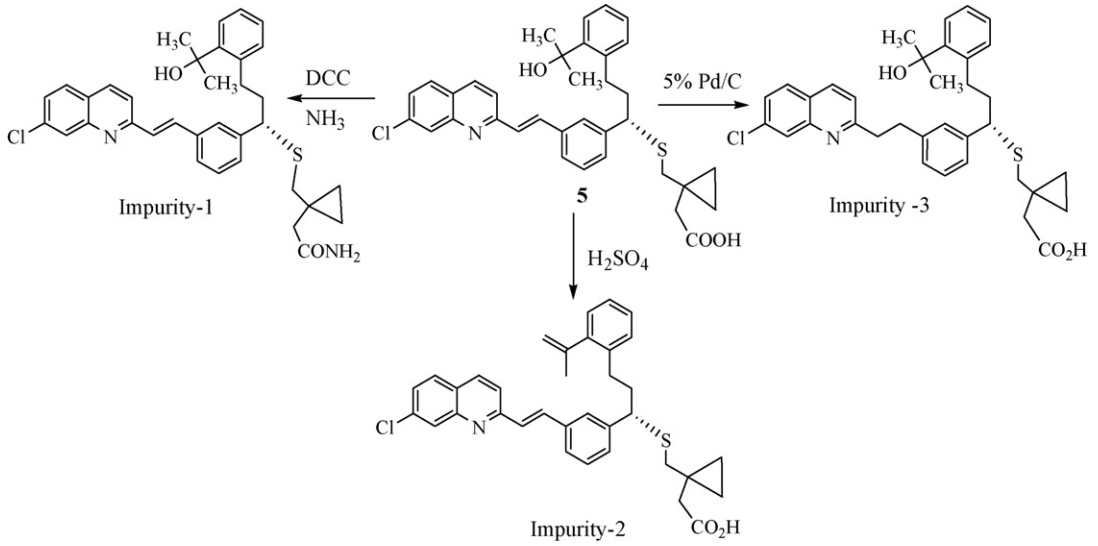


Fig. 2. Synthesis of montelukast sodium impurities.

sodium. Waters make HPLC system equipped with 515 pump and 2996-photodiode-array (PDA) detector was used for better separation and quantification of impurities. The buffer solution used for the preparation of mobile phases A and B consists of 0.05 M aqueous sodium dihydrogen phosphate monohydrate and its pH was adjusted to 3.7 with diluted phosphoric acid. Mobile phase A was prepared in the ratio of 800:200 (v/v) of buffer and acetonitrile; mobile phase B was prepared in the ratio of 200:800 (v/v) of buffer and acetonitrile. Hypersil BDS-C18, 100 mm × 4.6 mm, 3-μm particle size column was used with a time gradient program of *T* (min)/%B (v/v). Initial gradient starts with 47% of B and at 35 min it is 95%. The ratio being continued up to 58 min, at 62 min it is 47%, which is continued up to 70 min with a flow rate of 1.0 ml/min, column oven temperature was 30 °C and column eluent was monitored by PDA detector at 225 nm. This LC method was able to separate all the process-related substances with good resolution.

2.3. Liquid chromatography–mass spectrometry

The buffer solution used for the preparation of mobile phases A and B consists of 0.05 M aqueous ammonium acetate and its pH

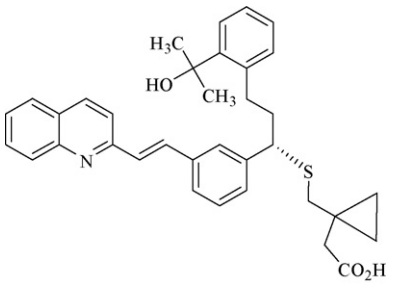


Fig. 3. Impurity-4 of montelukast sodium.

Table 1
LC Retention times, molecular weights of the impurities from LC–MS and atom numbering used for NMR assignment

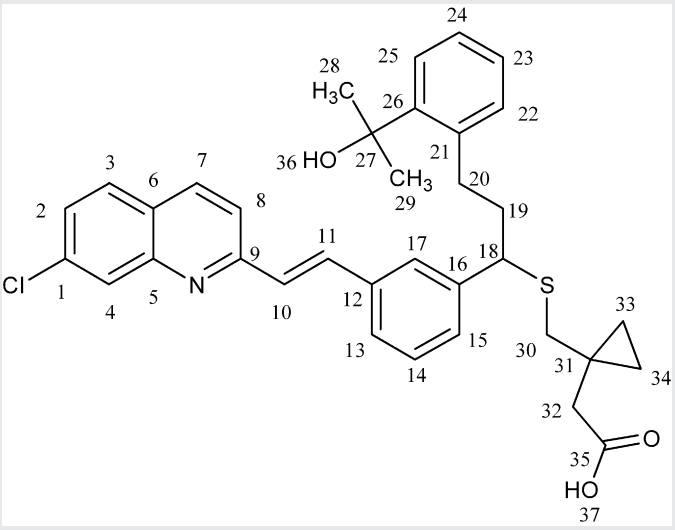
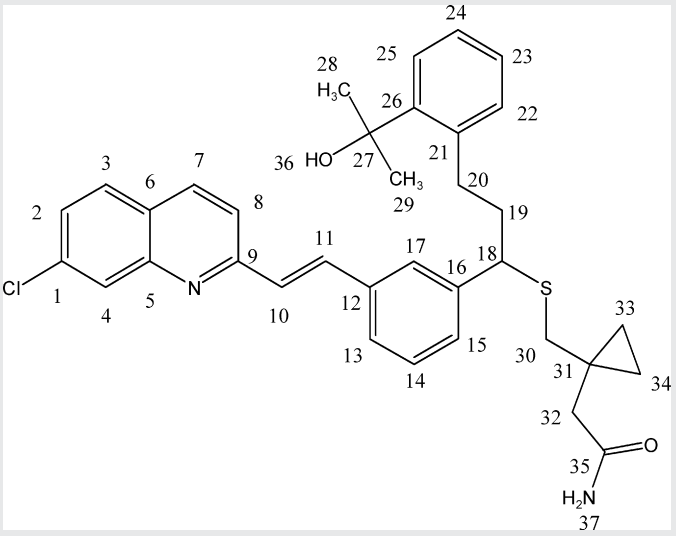
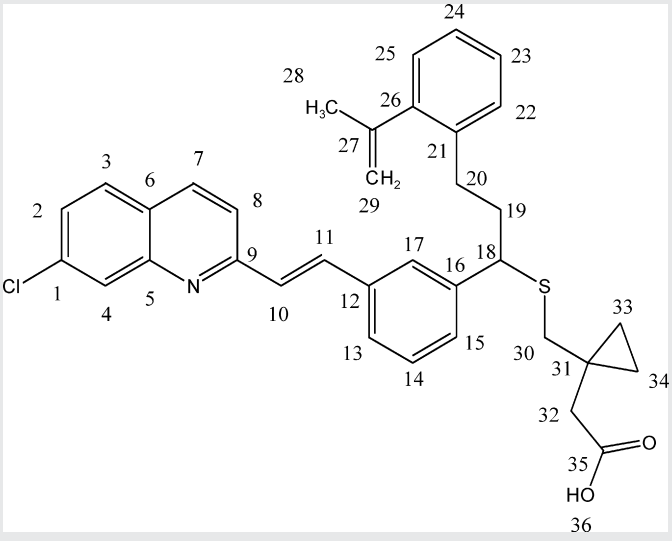
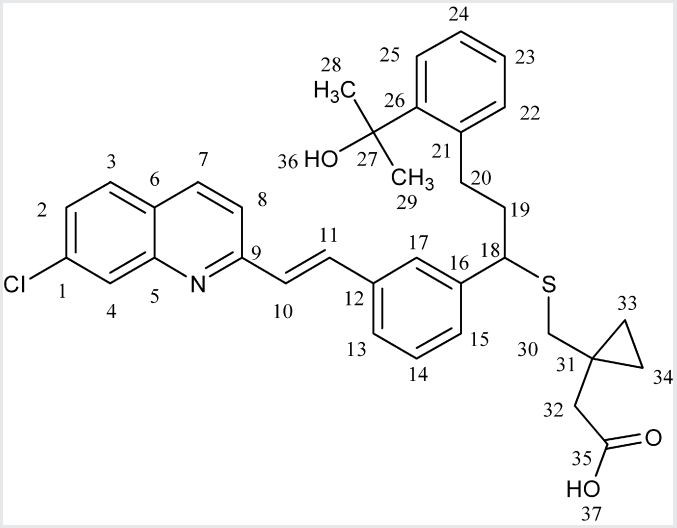
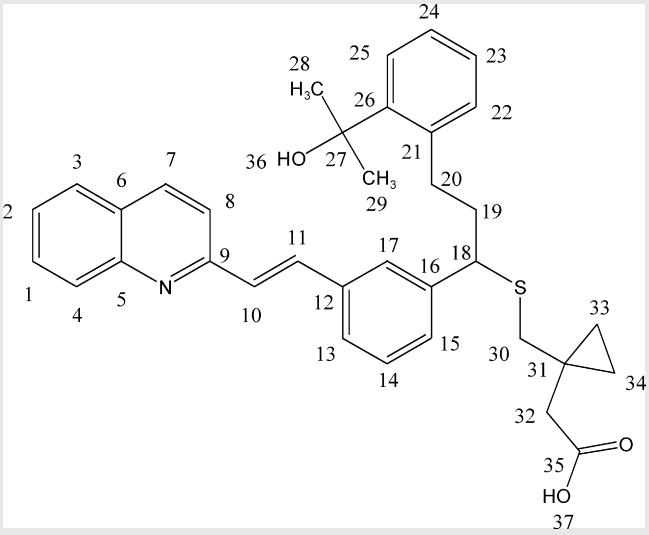
S.No.	Retention time (min)	Compound	Molecular weight	Structure
01	25.01	Montelukast	585	
02	19.56	Impurity-1	584	
03	37.03	Impurity-2	567	

Table 1 (*Continued*)

S.No.	Retention time (min)	Compound	Molecular weight	Structure
04	20.25	Impurity-3	587	
05	15.76	Impurity-4	551	

was adjusted to 3.7 with acetic acid. Mobile phase A was prepared in the ratio of 800:200 (v/v) of buffer and acetonitrile. Mobile phase B was prepared in the ratio of 200:800 (v/v) of buffer and acetonitrile. Hypersil BDS-C18, 100 mm × 4.6 mm, 3-μm particle size column was used with a time gradient program of *T* (min)/%B (v/v). Initial gradient of B starts with 47% and at 35 min it is 95%. The ratio being continued up to 70 min and at 75 min it is 47%, which is continued up to 80 min with a flow rate of 1.0 ml/min and column eluent was monitored by UV detector at 298 nm. This method was able to detect all the process-related substances with good resolution.

2.4. Mass spectrometry

The electrospray ionization and MS–MS studies were performed on a triple quadruple mass spectrometer PE Sciex model API 3000. The positive and negative electrospray MS data was obtained by switching the capillary voltage between +5000 and −4500 V, respectively. The MS–MS data was generated with the collision energy ramping from 30 to 60 V in nitrogen atmosphere.

2.5. NMR spectroscopy

The ^{1}H, ^{13}C, DEPT and 2D experiments for montelukast, impurity-1–3 were done on Varian Mercury plus 400 MHz FT NMR spectrometer and similar experiments for impurity-4 was done at Unity Inova 500 MHz FT NMR spectrometer. The solvents used for montelukast, impurity-2 and -3 were in DMSO-d_6 and for impurity-1 and -4 were in $CDCl_3$. The ^{1}H chemical shift values were reported on δ scale in ppm, relative to TMS (δ = 0.00 ppm) and in the ^{13}C NMR the chemical shift values were reported relative to $CDCl_3$ (δ = 77.00 ppm) and DMSO-d_6 (δ = 39.50 ppm) as internal standards, respectively. DEPT spectra revealed the presence of methyl and methine groups as positive peaks and methylenes as negative peaks.

2.6. FT-IR spectroscopy

The IR spectra were recorded in the solid state as KBr dispersion medium using PerkinElmer 1600 series FT-IR spectrophotometer.

2.7. Synthesis of impurities

Among the four impurities, impurity-1–3 were synthesized (Fig. 2) in the laboratory and impurity-4 (Fig. 3) was isolated through preparative HPLC method. Impurity-1 was synthesized by the reaction of montelukast acid 5 with ammonia gas in the presence of dicyclohexyl carbodiimide (DCC) and catalytic amount of 1-hydroxy benzotriazole. Dehydration of montelukast acid 5 with sulphuric acid yielded impurity-2. Impurity-3 was prepared by catalytic reduction of montelukast acid 5 using 5% palladium on carbon.

2.7.1. Preparative condition for impurity-4 of montelukast sodium

A Shimadzu LC-8A preparative liquid chromatograph equipped with SPD-10A VP, UV–vis detector (Shimadzu Corporation, Analytical Instruments Division, Kyoto, Japan) was used. 20 mm × 250 mm × 5 μm preparative column that contains L1 packing used for the isolation of impurity. The chromatograph was programmed as follows. Initial concentration of mobile phase starts at 100% with flow 10 ml/min and at 19 min the flow is 30 ml/min and continued up to 33 min and at 40 min the flow is 10 ml/min, which is continued up to 45 min. Buffer prepared by adjusting the Milli-Q water pH to 3.0 with acetic acid and mobile phase is a mixture of buffer and acetonitrile in the ratio of 200:800 (v/v). Sample concentration is 250 mg/ml in diluent (methanol).

2.7.2. Procedure

Injected 1.0 ml of methanol as blank and the test solution into chromatograph, recorded the chromatograms at 225 nm and collected the fractions and analyzed as per the conditions, the peak at RT 16.6 is matching with the mother sample peak (0.66 RRT). The collected millilitres (∼5 l) were distilled off under reduced pressure at below 50 °C to afford ∼100 mg of impurity-4. The *m*/*z* value for the distilled ml's is 551 and 2D NMR confirms the 0.66 RRT impurity as impurity-4 (deschloro montelukast).

3. Results and discussion

3.1. Detection of impurities 1–4

A typical analytical LC chromatogram (Fig. 4) of a laboratory batch of montelukast sodium recorded using the LC method is described in Section 2.2. The LC–MS compatible method which was used to detect the impurities is described in Section 2.3 (Fig. 4). Retention times in LC, structures and molecular weights of these impurities and montelukast (montelukast free acid) are shown in Table 1 . All the four impurities spectral data compared with montelukast for better visualization of five similar molecules.

3.2. Structure elucidation of montelukast and its impurities

3.2.1. Montelukast

ESI mass spectrum of montelukast exhibited protonated molecule peak at *m*/*z* 586.2 [$(MH)^+$] in positive ion mode, indicating the mass of this impurity to be 585. In ^{1}H NMR of this impurity, the signal at 11.9 ppm corresponding to acid OH and a broad signal at 5.6 ppm corresponding to tertiary alcohol OH were observed. In IR spectrum, a broad band at 3396 cm^{-1} corresponding to acid OH, a band at 1710 cm^{-1} corresponding to acid C=O, a broad band at 1132 cm^{-1} corresponding to ether C–O linkage of tertiary alcohol and a band at 1068 cm^{-1} corresponding to aromatic C–Cl were observed. The DEPT spectra displayed six negative signals due to six methylene groups and eighteen positive signals due to the presence of two methyl groups and the rest are due to the methine groups. Based on the above spectral data the molecular formula of montelukast acid was confirmed as $C_{35}H_{36}ClNO_3S$ and the corresponding structure was confirmed as 1-[[[(1*R*)-1-[3-[(1*E*)-2-(7-chloro-2-quinolinyl)ethenyl]phenyl-3-[2-(1-hydroxy-1-methylethyl)phenyl]propyl]thio]methyl]cyclopropaneacetic acid.

3.2.2. Impurity-1

ESI mass spectrum of impurity-1 displayed protonated molecule peak at *m*/*z* 585.4 [$(MH)^+$] in positive ion mode, indicating the mass of this impurity to be 584 which is 1 amu less than that of montelukast. In ^{1}H NMR of this impurity, the signal at 11.9 ppm corresponding to acid OH was disappeared and a broad signal at 5.6 ppm corresponding to amide NH was observed with two proton integration. In IR spectrum, acid OH band at 3396 cm^{-1} was absent and a strong NH signals observed at 3357 cm^{-1}, this was further confirms the band at 1671 cm^{-1} corresponding to C=O (amide I band) and 1630 corresponding N–H bending (amide II band). In ^{13}C NMR spectrum of the impurity, acid carbon signal at 174.84 ppm was absent and amide carbon signal at 172.9 ppm was observed. This observation confirms that acid OH group is replaced by amine group. From the above spectral data the molecular formula of impurity-1 was confirmed as $C_{35}H_{37}ClN_2O_2S$ and the corresponding structure was characterised as 1-[[[(1*R*)-1-[3-[(1*E*)-2-(7-chloro-2-quinolinyl)ethenyl]phenyl-3-[2-(1-hydroxy-1-methylethyl)phenyl]propyl]thio]methyl]cyclopropane acetamide.

3.2.3. Impurity-2

ESI mass spectrum of impurity-2 showed protonated molecule peak at *m*/*z* 568.3 [$(MH)^+$] in positive ion mode, indicating the mass of this impurity to be 567 which is 18 amu less than that of montelukast. In ^{1}H NMR of this impurity, the broad singlet signal at 5.24 ppm corresponding to tertiary alcohol was disappeared and singlet signal at 4.74 ppm and 5.1 ppm corresponding to alkene in position 29 (Table 1) were observed. In FT-IR spectrum C–O band at 1132 cm^{-1} was absent and a strong alkene C=C band was observed at 1610 cm^{-1}. In ^{13}C NMR spectrum, the signal corresponding to methyl group of montelukast was absent and an additional signal was observed at 104.7 ppm, from the DEPT spectrum it was confirmed as alkene CH_2 signal. This observation suggested the loss of water molecule from montelukast. Based on the above spectral data the molecular formula of impurity-2 was confirmed as $C_{35}H_{34}ClNO_2S$ and the corresponding structure was characterised as {1-[1-{3-[2-(7-chloro-quinolin-2-yl)-vinyl]-phenyl}-3-(2-isopropenyl-phenyl)-propylsulfanylmethyl]-cyclopropyl}-acetic acid (Table 2.).

3.2.4. Impurity-3

ESI mass spectrum of impurity-3 exhibited protonated molecule peak at *m*/*z* 588.2 [$(MH)^+$] in positive ion mode, indicating the mass of this impurity to be 587 which is 2 amu more than that of montelukast. In ^{1}H NMR of this impurity, doublet signal at 7.89 and 7.54 ppm were assigned to trans alkene group and which were absent in montelukast, triplet signal at 3.25 ppm and 3.09 ppm were observed corresponding to methylene proton. ^{13}C NMR spectrum showed the absence of signals at 134.95 and 128.1 ppm corresponding to alkene carbon. Additional signals at 39.5 and 34.2 ppm corresponding to alkane carbon were observed from the DEPT spectrum and it was confirmed as alkane CH_2 signal. All these observation confirms that olefin double bond is reduced to alkane group. Based on the above spectral data the molecular formula of impurity-3 was confirmed as $C_{35}H_{38}ClNO_3S$ and the corresponding structure was characterised as 1-[[[(1*R*)-1-[3-[(1*E*)-2-(7-chloro-2-quinolinyl)ethyl]phenyl-3-[2-(1-hydroxy-1-methylethyl)phenyl]propyl]thio]methyl]cyclopropaneacetic acid.

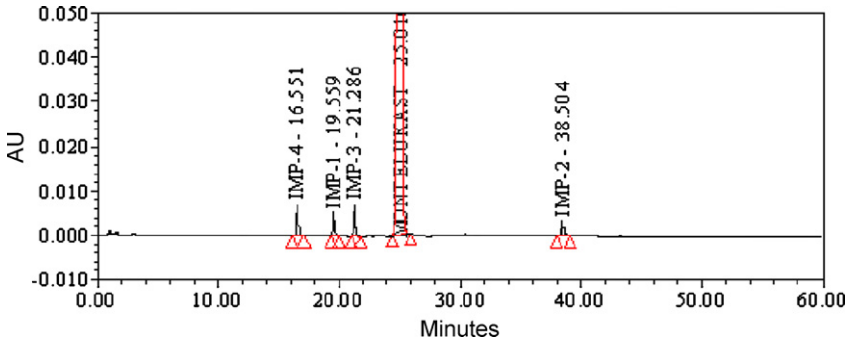


Fig. 4. LC chromatogram of montelukast sodium.

3.2.5. Impurity-4

ESI mass spectrum of impurity-4 exhibited protonated molecule peak at *m/z* 552.3 [$(MH)^+$] in positive ion mode, indicating the mass of this impurity to be 551 which is 35 amu less than that of montelukast. The ^{1}H NMR, this impurity showed the entire signal similar to that of montelukast, in addition to that a signal at 7.7 ppm corresponding to aromatic proton was observed with one proton integration. In FT-IR spectrum, aromatic C–Cl band at 1071 cm^{-1} was absent, except C–Cl band at 1071 cm^{-1} remaining FT-IR pattern was similar to that of montelukast. ^{13}C NMR spectrum of this

Table 2
^{1}H and ^{13}C NMR assignments for montelukast sodium and its impurities 1 and 2

Position	Montelukast sodium				Impurity-1				Impurity-2			
	^{1}H	ppm/J	^{13}C	DEPT	^{1}H	ppm/J	^{13}C	DEPT	^{1}H	ppm/J	^{13}C	DEPT
1	–	–	125.43	–	–	–	125.54	–	–	–	125.66	–
2	1H	7.59/dd 8.8, 2.4	126.54	CH	1H	7.44/dd 8.8, 2.0	127.00	CH	1H	7.59/dd 8.8, 2.0	126.64	CH
3	1H	8.00/d 8.8	129.56	CH	1H	7.71/d 8.8	128.59	CH	1H	8.01/d 8.8	129.69	CH
4	1H	8.03/d 2.4	127.03	CH	1H	8.07/d 2.0	127.97	CH	1H	8.02/d 2.0	127.18	CH
5	–	–	147.82	–	–	–	148.48	–	–	–	148.00	–
6	–	–	134.00	–	–	–	135.44	–	–	–	134.27	–
7	1H	8.37/d 8.4	136.32	CH	1H	8.10/d 8.4	136.08	CH	1H	8.41/d 8.8	136.50	CH
8	1H	7.95/d 8.4	120.16	CH	1H	7.65/d 8.4	119.46	CH	1H	7.94/d 8.8	120.26	CH
9	–	–	156.71	–	–	–	156.72	–	–	–	156.76	–
10	1H	7.89/d 16.4	134.95	CH	1H	7.70/d 16.4	134.92	CH	1H	7.88/d 16.0	134.94	CH
11	1H	7.50/d 16.4	128.10	CH	1H	7.39/d 16.4	128.93	CH	1H	7.49/d 16.0	128.35	CH
12	–	–	135.89	–	–	–	136.46	–	–	–	136.08	–
13	1H	7.61/d 7.2	125.44	CH	1H	7.51/d 7.2	126.10	CH	1H	7.63/d 7.6	125.89	CH
14	1H	7.39/m	125.13	CH	1H	7.37/m	125.38	CH	1H	7.41/t 7.6	128.87	CH
15	1H	7.35/m	128.13	CH	1H	7.34/m	128.34	CH	1H	7.32/d 7.6	128.23	CH
16			144.04		–	–	143.62	–	–	–	143.27	–
17	1H	7.73/s	126.63	CH	1H	7.66/s	126.62	CH	1H	7.68/s	126.61	CH
18	1H	4.02/t	49.08	CH	1H	3.93/t	50.27	CH	1H	3.93/t	48.81	CH
19	Ha Hb	2.22/m 2.11/m	39.10	CH_2	2H	2.23/m	39.56	CH_2	2H	2.10/m	38.10	CH_2
20	Ha Hb	3.08/m 2.74/m	31.58	CH_2	Ha Hb	3.20/m 2.90/m	32.25	CH_2	Ha Hb	2.65/m 2.54/m	30.58	CH_2
21	–	–	139.84	–	–	–	140.03	–	–	–	137.64	–
22	1H	7.11/m	130.83	CH	1H	7.17/m	131.37	CH	1H	7.18/m	126.92	CH
23	1H	7.11/m	126.13	CH	1H	7.18/m	126.95	CH	1H	7.19/m	129.06	CH
24	1H	7.06/m	124.94	CH	1H	7.13/m	125.56	CH	1H	7.15/m	125.72	CH
25	1H	7.38/m	128.39	CH	1H	7.36/m	128.61	CH	1H	7.05/d 7.2	127.76	CH
26	–	–	146.69	–	–	–	145.37	–	–	–	144.82	–
27	–	–	71.61	–	–	–	73.45	–	–	–	142.96	–
28	3H	1.45/s	31.29	CH_3	3H	1.62/s	31.79	CH_3	3H	1.90/s	24.71$^\beta$	CH_3
29	3H	1.44/s	31.29	CH_3	3H	1.60/s	31.75	CH_3	Ha Hb	5.10/s 4.74/s	24.71$^\beta$	CH_2
30	Ha Hb	2.16/d 14.0 1.97/d 14.0	43.74	CH_2	2H	2.27/s	41.67	CH_2	2H	2.50/s	38.48	CH_2
31	–	–	17.88	–	–	–	17.21	–	–	–	16.63	–
32	Ha Hb	2.69/d 12.8 2.55/d 12.8	39.45	CH_2	Ha Hb	2.48/d 13.0 2.44/d 13.0	39.14	CH_2	Ha Hb	2.32/d 16.3 2.27/d 16.3	39.36	CH_2
33	2H	0.20/m	11.84	CH_2	2H	0.42/m	12.71	CH_2	2H	0.39/m	11.83	CH_2
34	2H	0.41/m	11.84	CH_2	2H	0.48/m	12.20	CH_2	2H	0.40/m	12.05	CH_2
35	–	–	174.84	–	–	–	174.14	–	–	–	172.97	–
36	OH	5.24/br	–	–	OH	5.28/br	–	–	OH	12.01/br	–	–
37	–	–	–	–	NH	5.60/br	–	–	–	–	–	–

Table 3
^{1}H and ^{13}C NMR assignments for impurities 3 and 4

Position	Impurity-3				Impurity-4			
	^{1}H	ppm/J	^{13}C	DEPT	^{1}H	ppm/J	^{13}C	DEPT
1	–	–	125.21	–	1H	7.70t 7.00	130.23	CH
2	1H	7.56/dd 8.8, 2.0	126.18	CH	1H	7.50t 7.5	126.54	CH
3	1H	7.94/d 8.8	129.51	CH	1H	7.78d 7.5	127.74	CH
4	1H	7.99/d 2.0	126.82	CH	1H	8.06d 7.5	128.62	CH
5	–	–	147.59	–	–	–	147.99	–
6	–	–	136.36	–	–	–	135.35	–
7	1H	8.27/d 8.4	135.99	CH	1H	8.13d 8.5	137.09	CH
8	1H	7.47/d 8.4	122.00	CH	1H	7.72d 8.5	118.95	CH
9	–	–	162.91	–	–	–	156.30	–
10	2H	3.25/t 8.4	39.51	CH_2	1H	7.58d 16.0	135.06	CH
11	2H	3.09/t 8.4	34.20	CH_2	1H	7.54d 16.0	129.47	CH
12	–	–	140.99	–	–	–	137.03	–
13	1H	7.36/dd 7.6, 1.6	125.14	CH	1H	7.40d 8.0	126.74	CH
14	1H	7.09/ m	126.18	CH	1H	7.35/m	125.64	CH
15	1H	7.22/d 7.6	128.01	CH	1H	7.30t 7.0	128.55	CH
16	–	–	142.24	–	–	–	143.57	–
17	1H	7.24/s	127.55	CH	1H	7.79/m	126.92	CH
18	1H	3.86/t 7.6	49.05	CH	1H	4.01t 7.5	50.57	CH
19	2H	2.06/m	39.13	CH_2	2H	2.21/m	40.16	CH_2
20	Ha Hb	2.95/m 2.66/m	31.41	CH_2	Ha Hb	3.19/m 2.93/m	32.44	CH_2
21	–	–	139.66	–			140.57	
22	1H	7.03/m	130.73	CH	1H	7.15/m	131.73	CH
23	1H	7.36/m	125.14	CH	1H	7.15/m	127.34	CH
24	1H	7.07/m	124.98	CH	1H	7.10/m	125.86	CH
25	1H	7.15/m	125.14	CH	1H	7.33/m	129.24	CH
26	–	–	146.52	–	–	–	145.43	–
27	–	–	71.46	–	–	–	74.02	–
28	3H	1.41/s	31.24	CH_3	3H	1.62/s	32.07	CH_3
29	3H	1.40/s	31.24	CH_3	3H	1.60/s	32.07	CH_3
30	2H	2.28/s	39.80	CH_2	Ha Hb	2.59d 16.5 2.33d 16.5	40.88	CH_2
31	–	–	16.18	–	–	–	17.00	–
32	Ha Hb	2.42/d 13.2 2.37/d 13.2	38.08	CH_2	Ha Hb	2.70d 13.0 2.41d 13.0	38.95	CH_2
33	2H	0.29/m	11.63	CH_2	2H	0.51	12.60	CH_2
34	2H	0.41/m	11.55	CH_2	2H	0.51	12.60	CH_2
35	–	–	172.70	–	–	–	175.82	–
36	1H	4.84/br	–	–	1H	4.84/ br	–	–
37	1H	11.9/br	–	–	1H	11.9/ br	–	–

impurity showed all the signals similar to that of montelukast, in addition to that a signal at 130.23 ppm corresponding to aromatic carbon was observed, from the DEPT spectrum it was confirmed as aromatic CH signal. These observations suggested that; lose of chlorine atom from montelukast. Based on the above spectral data the molecular formula of impurity-4 was confirmed as $C_{35}H_{37}NO_3S$ and the corresponding structure was characterised as 1-[[[(1*R*)-1-[3-[(1*E*)-2-(2-quinolinyl)ethenyl]phenyl-3-[2-(1-hydroxy-1-methylethyl)phenyl]propyl]thio]methyl]cyclopropaneacetic acid (Tables 3 and 4).

3.3. Formation of impurities

3.3.1. Impurity-1

During the basic hydrolysis stage, the cyano group in four elaborated into carboxylic group and leads to acid 5 (Fig. 1) via the

Table 4
FT-IR and mass spectral data for montelukast and its impurities 1–4

S.No.	Compound	IR	Mass
1.	Montelukast	3396 (COOH stretching), 3057 (aromatic C–H stretching), 2925 (aliphatic C–H stretching), 1710 (C=O stretching), 1610 (C=C stretching), 1594 (C=N stretching), 1497 (aliphatic C–H bending), 1132 (C–O stretching), 1068 (aromatic C–Cl stretching), 837 (aromatic C–H bending), 697 (C–S stretching)	586.2
2.	Impurity-1	3357 (N–H stretching), 3196 (aromatic C–H stretching), 2927 (aliphatic C–H stretching), 1671 (C=O stretching), 1630 (N–H bending), 1442 (aliphatic C–H bending), 1132 (C–O stretching), 1070 (aromatic C–Cl stretching), 839 (aromatic C–H bending), 698 (C–S stretching)	585.4
3.	Impurity-2	3439 (COOH stretching), 3074 (aromatic C–H stretching), 2919(aliphatic C–H stretching), 1712 (C=O stretching), 1610 (C=C stretching), 1596 (C=N stretching), 1500 (aliphatic C–H bending), 1076 (aromatic C–Cl stretching), 770 (aromatic C–H bending), 697 (C–S stretching)	568.3
4.	Impurity-3	3437 (COOH stretching), 3090 (aromatic C–H stretching), 2910 (aliphatic C–H stretching), 1710 (C=O stretching), 1598 (C=N stretching), 1495 (aliphatic C–H bending), 1132 (C–O stretching), 1071 (aromatic C–Cl stretching), 790 (aromatic C–H bending), 690 (C–S stretching)	588.2
5.	Impurity-4	3430 (COOH stretching), 3083 (aromatic C–H stretching), 2911 (aliphatic C–H stretching), 1715 (C=O stretching), 1604 (C=N stretching), 1493 (aliphatic C–H bending), 1132 (C–O stretching), 796 (aromatic C–H bending), 695(C–S stretching)	552.3

transformation of amide intermediate, i.e. impurity-1. If amide intermediate could not be completely hydrolyzed into acid 5, impurity-1 will be resulted.

3.3.2. Impurity-2

To isolate the montelukast 5, acetic acid was used in the work up stage. Due to acidic nature, tertiary hydroxyl moiety gets protonated. Since protanated hydroxy being a good leaving group, it takes away the adjacent methyl group proton and leads to the formation of impurity-2.

3.3.3. Impurity-3 and -4

Starting material 2 contains saturated and deschloro analogue of 2, these compounds undergo sequential reactions (4, 5 and 1) and leads to formation of impurity-3 and impurity-4, respectively.

4. Conclusion

The process-related impurities in montelukast sodium bulk drug were identified, synthesized, isolated and characterised by HPLC (analytical and preparative), LC–MS, FT-IR and NMR (1H, ^{13}C and DEPT) techniques.

Acknowledgments

The authors are thankful to the management of Dr. Reddy's Laboratories Ltd. for supporting this work and cooperation from other colleagues also highly appreciated.

References

[1] M. Belley, S. Leger, P. Roy, Y.B. Xiang, M. Labelle, D. Guay, D. EP 0480717B1, April 15 (1998).
[2] M. Labelle, M. Belley, Y. Gareau, J.Y. Gauthier, D. Guay, R. Gordon, S.G. Grossman, T.R. Jones, Y. Leblanc, M. McAuliffe, C. McFarlane, P. Masson, K.M. Metters, N. Ouimet, D.H. Patrick, H. Piechuta, C. Rochette, N. Sawyer, Y.B. Xiang, C.B. Pickett, A.W. Ford-Hutchinson, R.J. Zamboni, R.N. Young, Bioorg. Med. Chem. Lett. 5 (1995) 283–288.
[3] D.F. Schoors, M.D. Smet, T. Reiss, D. Margolskee, H. Cheng, P. Larson, R. Amin, G. Somers, Br. J. Clin. Pharmacol. 40 (1995) 277–288.
[4] ICH Guideline Q3A (R), Impurities in New Drug Substances, February 7, 2002.
[5] P. Pratap Reddy, B. Satyanarayana, S. Alok kumar, K. Ravi kumar, J. Rajender Reddy, M.N. Naidu, WO2007/012075A2, January 25, 2007.