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Identification and characterization of potential impurities of donepezil

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Abstract

Five unknown impurities ranging from 0.05 to 0.2% in donepezil were detected by a simple isocratic reversed-phase high performance liquid chromatography (HPLC). These impurities were isolated from crude sample of donepezil using isocratic reversed-phase preparative high performance liquid chromatography. Based on the spectral data (IR, NMR and MS), the structures of these impurities were characterised as 5,6-dimethoxy-2-(4-pyridylmethyl)-1-indanone (impurity I), 4-(5,6-dimethoxy-2,3-dihydro-1H-2-indenylmethyl) piperidine (impurity II), 2-(1-benzyl-4-piperdylmethyl)-5,6-dimethoxy-1-indanol (impurity III) 1-benzyl-4(5,6-dimethoxy-2,3-dihydro-1H-2-indenylmethyl) piperidine (impurity IV) and 1,1-dibenzyl-4(5,6-dimethoxy-1-oxo-2,3-dihydro-2H-2-indenylmethyl)hexahydropyridinium bromide (impurity V). The synthesis of these impurities and their formation was discussed.

Keywords: Donepezil; Impurities; Spectroscopy; Identification; Characterization and synthesis

1. Introduction

Alzheimer's disease is one of the most common causes of mental deterioration of elderly people. It is characterised by degeneration of various structures in the brain. Acetylcholinesterase inhibitors are the most frequently prescribed drugs for the treatment of Alzheimer's disease [1,2]. Donepezil, (−)2,3-dihydro-5,6-dimethoxy-2-[[1-(phenylmethyl)-4-piperidinyl] methyl]-1H-inden-1-one, a specific, selective and potent inhibitor of acetylcholinesterase [3–5] has been approved for the treatment of Alzheimer's disease.

Liquid chromatographic–spectroscopic [6] and capillary electrophoresis [7,8] methods have been described in the literature for the resolution and quantitative determination of enantiomers of donepezil.

During the analysis of different laboratory batches of donepezil, five unknown impurities were detected consistently in almost all the batches, whose area percentage ranged from 0.05 to 0.2%, by a simple isocratic reverse phase LC method. A comprehensive

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study has been undertaken to isolate and characterise these impurities by spectroscopic and spectrometric techniques. The impurity profile study has to be carried out for any final product to identify and characterise all the unknown impurities that are present at a level of even below 0.05%. The requirement of identifying and characterising the impurities in the final product is extremely necessary in the wake of stringent purity requirements from the regulatory authorities. This paper not only describes the isolation and characterization of five impurities that are present in the range of 0.05–0.2% in the bulk drug of donepezil but also explains the formation of these impurities.

2. Experimental

2.1. Samples

The investigated samples of donepezil bulk material (B. no. DON/B415/IIID/43) and crude sample (B. no. DON/B415/IIID/43C) were obtained from Dr. Reddy's Laboratories Ltd. Bulk Actives-III, Hyderabad, India.

2.2. High performance liquid chromatography (analytical)

A Waters Model Alliance 2690 Separation module equipped with a Waters 996 photo diode array UV detector was used. An in-house LC method was developed for the analysis of donepezil and its intermediates, where a C18 column (Hichrom-RPB, 250 mm × 4.6 mm i.d., Hichrom Ltd., UK) with a mobile phase consisting of a mixture of 0.01 M KH_2PO_4 and acetonitrile in the ratio of 65:35 (v/v) (pH:3.5) was used with UV detection at 240 nm at a flow rate of 1.0 ml/min for the resolution of all impurities. The data was recorded using Waters Millennium 32 software. This LC method was able to detect these impurities, which ranged from 0.05 to 0.1% in the presence of parent compound.

2.3. High performance liquid chromatography (preparative)

A Shimadzu preparative high performance liquid chromatography (HPLC) equipped with LC-8A pump, SCL-8A System controller, SPD-6AV UV–Vis detector, FCV-100B Fraction collector and Rheodyne Injector Model 7725i with 2.0 ml loop. The data was collected and processed using Shimadzu CR7A chromatopak. A 250 mm × 10 mm i.d. column packed with 5μ Hichrom-C18 (Hichrom Ltd., UK) was employed for separation. The mobile phase consisted of 0.01 M KH_2PO_4 and acetonitrile in the ratio of 50:50 (v/v) (pH 3.5). The flow rate was set at 3.0 ml/min. Detection was carried out at 240 nm.

2.4. Mass spectrometry

Mass spectra were obtained using HP5989A with an electron energy set to 70 eV. The samples were introduced via the particle beam inlet using a LC pump (HP 1050 series) and a manual injector (Rheodyne model 7725i). The source manifold and quadrupole temperatures were maintained at 250 and 100 °C, respectively.

Molecular ions of all the impurities were further confirmed by obtaining the mass spectra on a Perkin Elmer Sciex API 3000 ES/MS. The sample was introduced into the source through a turbo ion spray interface in positive ionisation mode. The nebuliser and curtain gases used were zero air and nitrogen, respectively. Ion source voltage was maintained at 4200 V. Focusing potential and declustering potential were kept at 300 and 80 V, respectively.

2.5. NMR spectroscopy

The ^{1}H and 2D experiments (COSY, gHSQC and gHMBC) were performed on Mercury plus 400 MHz and the ^{13}C and DEPT NMR experiments were performed on a 200 MHz instrument Varian Gemini model 2000 at 25 °C in $CDCl_3$. The ^{1}H chemical shift values were reported on the δ scale in ppm, relative to TMS ($\delta = 0.00$ ppm) and the ^{13}C chemical shift values were reported relative to $CDCl_3$ ($\delta = 77.0$ ppm) as internal standards, respectively.

2.6. FT-IR spectroscopy

The IR spectra for donepezil, impurities I–V were recorded in the solid state as KBr dispersion and neat for impurity II using Perkin Elmer 1650 FT-IR spectrophotometer.

2.7. Thermal analysis

Thermogravimetric analysis for impurity V was carried out on a Shimadzu TG-50 under nitrogen atmosphere at a heating rate of 5 °C/min. in the temperature range of 30–180 °C.

2.8. Moisture content determination

Moisture content for impurity V was determined using Metrohm Autotitrator Model 716 DMS Titrino using disodium tartarate as standard.

2.9. Synthesis of impurities

The impurity I was synthesised by hydrogenation of the intermediate VI under the hydrogen pressure of 2.0 kg/cm^2 for 2.0 h in methanol as a solvent. Impurity II was synthesised by reduction of intermediate VII with lithium aluminium hydride (LAH) [9]. Impurities III and IV were synthesised by reducing donepezil (VIII) with lithium hydride (LiH) and borane–THF, respectively [10]. The dibenzyl impurity V was synthesised by benzylation of donepezil VIII. Scheme for

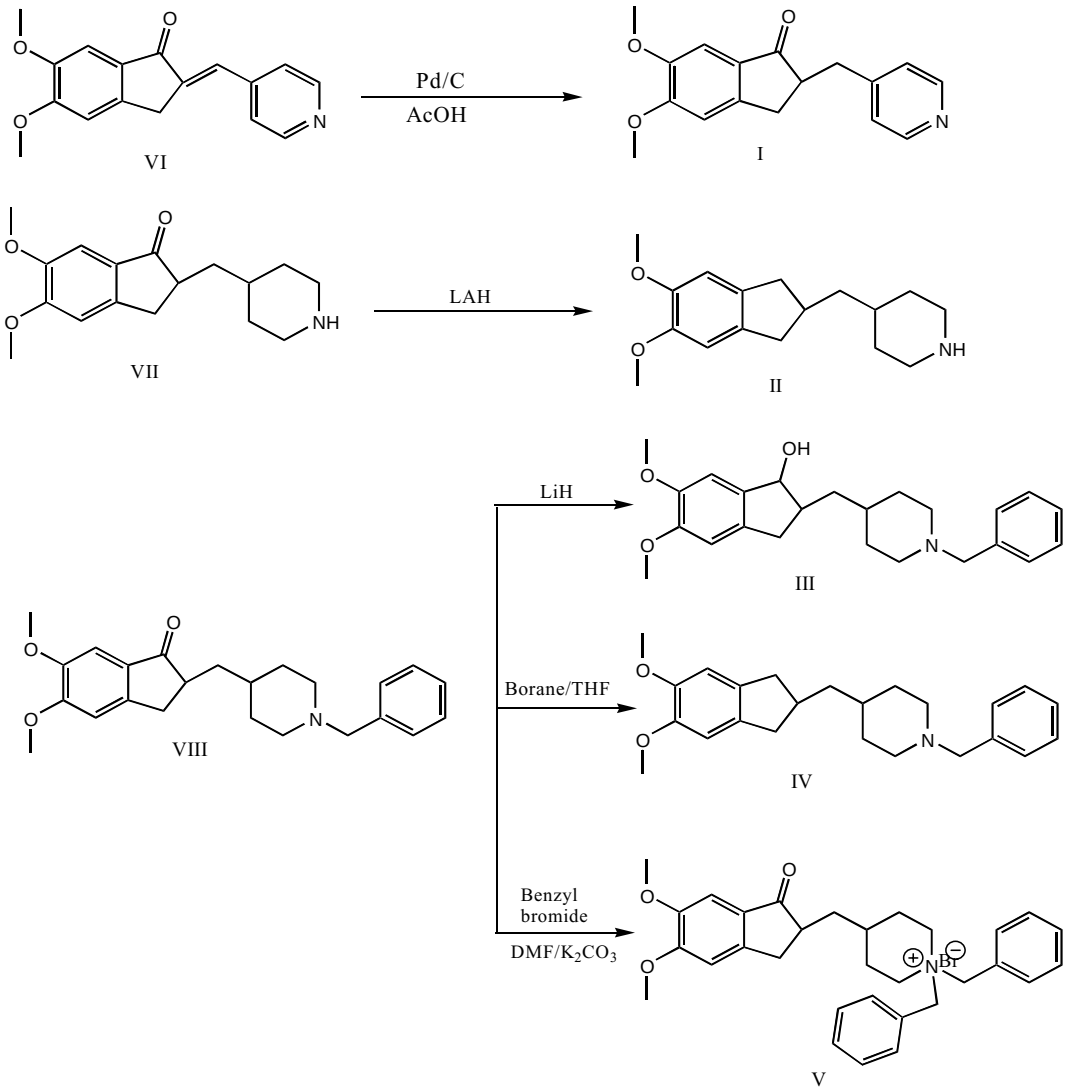


Fig. 1. Scheme for the synthesis of impurities I–V.

Table 1
Retention times and structures of donepezil impurities I–V

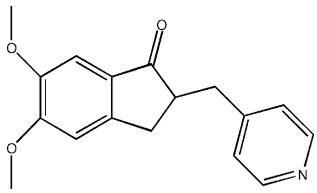
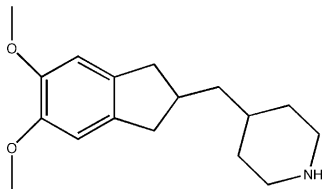
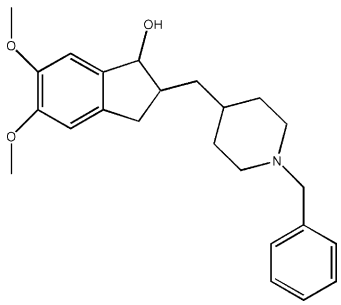
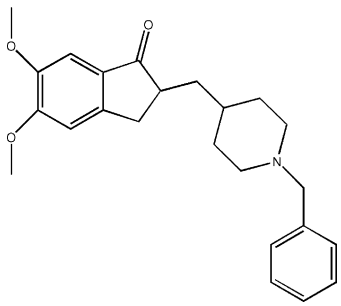
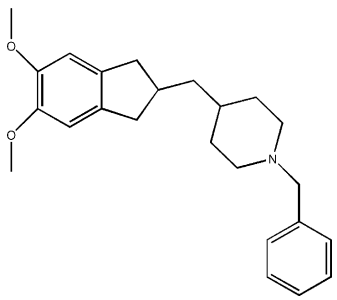
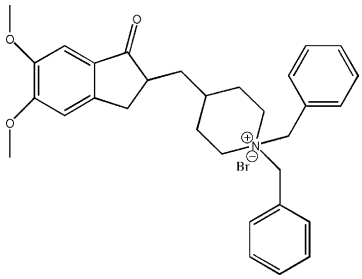
S. no.	Approximate retention time (min)		Compound	Structure	Nature
	Anal[a]	Prep[b]			
01	3.5	6–8	Impurity I		Process-related
02	4.5	10–12	Impurity II		Process-related
03	5.6	13–15	Impurity III		Process-related
03	6.0	17–19	Donepezil		–
04	13.0	25–28	Impurity IV		Process-related

Table 1 (*Continued*)

S. no.	Approximate retention time (min)		Compound	Structure	Nature
	Anal[a]	Prep[b]			
05	19.0	33–36	Impurity V		Process-related

[a] Analytical LC.
[b] Preparative LC.

the synthesis of impurities (I–V) is shown in Fig. 1.

3. Results and discussions

3.1. Detection of impurities I, II, III, IV and V

A typical analytical LC chromatogram of a laboratory batch of donepezil bulk drug recorded using the LC method as described in Section 2.2 is shown in Fig. 2a. The target impurities under study are marked as Imp-I, Imp-II, Imp-III, Imp-IV and Imp-V. Retention times and structures of these impurities and donepezil are shown in Table 1. These impurities were isolated from the crude sample of donepezil on preparative LC (Section 2.3) for spectroscopic studies. Impurities I–III are polar and impurities IV and V are non-polar, respectively, with respect to donepezil.

3.2. Isolation of the impurities by preparative HPLC

A simple reverse phase solvent system discussed under Section 2.3 was used for isolating these impurities. The retention times of donepezil and impurities are shown in Table 1. Collected fractions of these impurities were pooled together and kept in the refrigerator. All the fractions of impurities isolated were concentrated under high vacuum on a Buchi Rotavapor Model R124 to strip off the organic solvent. The remaining aqueous layer was subjected to solvent–solvent extraction with chloroform to extract the compounds into organic layer. The chloroform fractions ware again concentrated on rotavapor under vacuum. Purity of these impurities I–IV was tested separately before and after concentration in analytical mode (Section 2.2) and found to be 93.5, 97.2, 98.5, 95.1 and 96.3%, respectively. The isolated solids obtained from concentrated fractions of impurities were used to generate spectral data. The details of the elucidation of structures and formation of these impurities is presented in the following sections.

3.3. Structural elucidation of impurities

3.3.1. Structural elucidation of impurity I

The EI mass spectrum of impurity I exhibited molecular ion at *m*/*z* 283 atomic mass units (amu). The odd molecular ion in the mass spectrum indicates the presence of odd number of nitrogen atoms. The ^{13}C NMR spectrum displayed signals due to the presence of seventeen carbons. The DEPT spectrum displayed two negative signals due to two methylene groups and nine positive signals due to the presence of two methyl and the rest are due to methine groups (two in the aliphatic and the rest in aromatic region). The FT-IR spectrum displayed a characteristic absorption band at 1685 cm^{-1} indicating the presence of carbonyl functional group, which was supported

Table 2
FT-IR and mass spectral data of donepezil, impurities I–V

S. no.	Compound	IR[a]	MS data
1	Impurity I	2923 (aliphatic C–H stretching), 1685 (C=O stretching), 1602 (C=C stretching), 1442 (aliphatic C–H bending), 1312 (C–N stretching), 1269 and 1118 (C–O stretching)	*m/z* (EI; rel. int., %) 93(46), 163(17), 191(65), 237(28), 268(21), M^+ 283(100)
2	Impurity II	3341 (N–H stretching), 2923 (aliphatic C–H stretching), 2838 (aliphatic C–H stretching), 1583 (C=C stretching), 1463 (aliphatic C–H bending), 1220, 1100 (C–O stretching), 754 (aromatic C–H bending)	*m/z* (EI; rel. int., %) 98(100), 176(17), 189(14), M^+ 275(22)
3	Impurity III	3352 (O–H stretching), 2954 (aliphatic C–H stretching), 1610 (C=C stretching), 1462 (C–N stretching), 1228, 1175 (C–O stretching), 735 (aromatic C–H bending)	*m/z* (EI; rel. int., %) 91(100), 172(42), 188(23), 272 (37), 364 (12), M^+ 381(25)
4	Impurity IV	2923 (aromatic C–H stretching), 2840 (aliphatic C–H stretching), 1607 (C=C aromatic stretching), 1458 (aliphatic C–H bending), 1224, 1185 (C–O stretching), 754 (aromatic C–H bending)	*m/z* (EI; rel. int., %)91(100), 188(29), 274(13), M^+ 365(27)
5	Impurity V	3405 (O–H stretching, hydrate) 2934 (aromatic C–H stretching), 1693 (C=O stretching), 1591 (aromatic C=C stretching), 1498 and 1454 (aliphatic C–H bending), 1313 (C–N stretching), 1282, 1041 (C–O stretching), 760, 708 (aromatic C–H bending)	*m/z* (EI; rel. int., %) 91(100), 175(29), 238(31), 379(13), M^+ 470(2)
6	Donepezil	2926 (aromatic C–H stretching), 1694 (C=O stretching), 1591(aromatic C=C stretching), 1500, 1457 (aliphatic C–H bending), 1314 (C–N stretching), 1265, 1034 (C–O stretching), 748, 700 (aromatic C–H bending)	*m/z* (EI; rel. int., %) 91(100), 175(15), 205(3), 243(4), 288(12), M^+ 379(11)

[a] KBr (I, III, IV and V) and neat (II).

Table 3
Comparative ^{1}H NMR assignments for impurities I and II

Position[a]	Impurity I				Impurity II			
	^{1}H	ppm	^{13}C	DEPT	^{1}H	ppm	^{13}C	DEPT
1	–	–	205.18	–	2H	1.25, m	29.28	CH_2
2	1H	3.3, dd (7.8,2.0)	47.63	CH	1H	1.25, m	37.22	CH
3	Ha	2.7, m	36.17	CH_2	Ha	2.95, m	42.10	CH_2
	Hb	3.1			Hb			
4	–	–	178.53	–	–	–	134.43	–
5	–	–	128.68	–	–	–	134.43	–
6	1H	7.22, s	104.09	CH	1H	6.75	107.78	CH
7	–	–	149.34	–	–	–	147.84	–
8	–	–	155.53	–	–	–	147.84	–
9	1H	6.85, s	107.13	CH	1H	6.75	107.78	CH
10	Ha	2.65, m	31.53	CH_2	Ha	1.83	29.59	CH_2
	Hb	3.01, m			Hb	1.45		
11	–	–	148.32	–	1H	1.83	33.05	–
12 and 16	2H	7.22	124.01	CH	4H	1.45	39.23	CH_2
13 and 15	2H	8.42	149.37	CH	4H	2.51	43.71	CH_2
			149.58			3.25		
17	3H	3.95, s	55.81	CH_3	3H	3.92	55.97	CH_3
18	3H	3.97, s	55.96	CH_3	3H	3.92	55.97	CH_3

s: singlet; dd: doublet of a doublet; m: multiplet.
[a] Refer structural formula for numbering (Fig. 3 I and II)

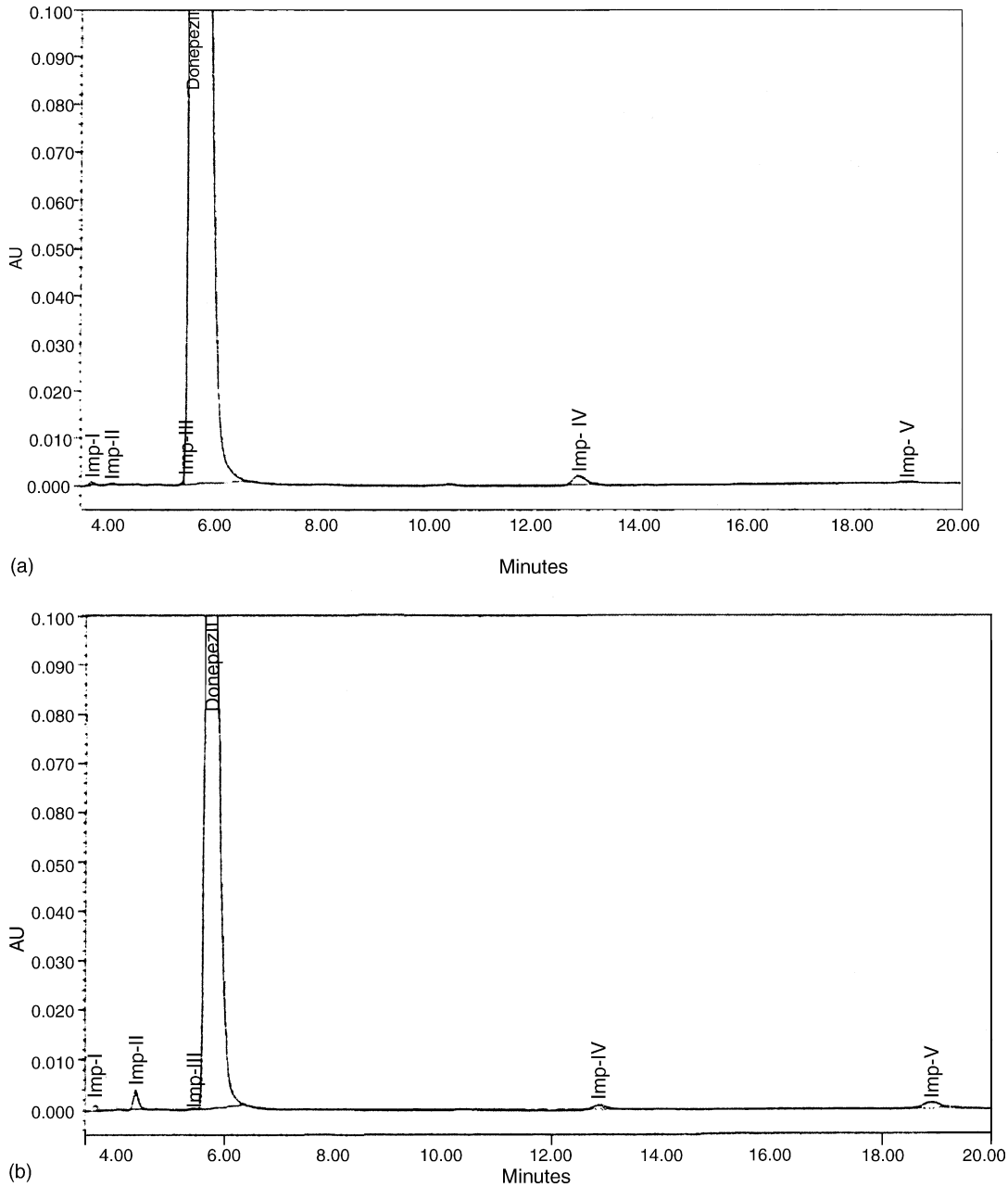


Fig. 2. (a) A typical analytical LC chromatogram of a laboratory batch of donepezil bulk drug. (b) The LC chromatogram of co-injection of the synthetic standards of impurities I–V with donepezil.

by the appearance of quaternary carbon signal due to carbonyl functional group in ^{13}C NMR spectrum. The peaks at 1269 and 1118 cm^{-1} in the IR spectrum are indicative of an aryl-alkyl ether functionality. Based on the above spectral data the molecular formula of impurity I could be $C_{17}H_{17}NO_3$. This molecular formula matched well with the molecular ion observed at 283 amu in the EI mass spectrum.

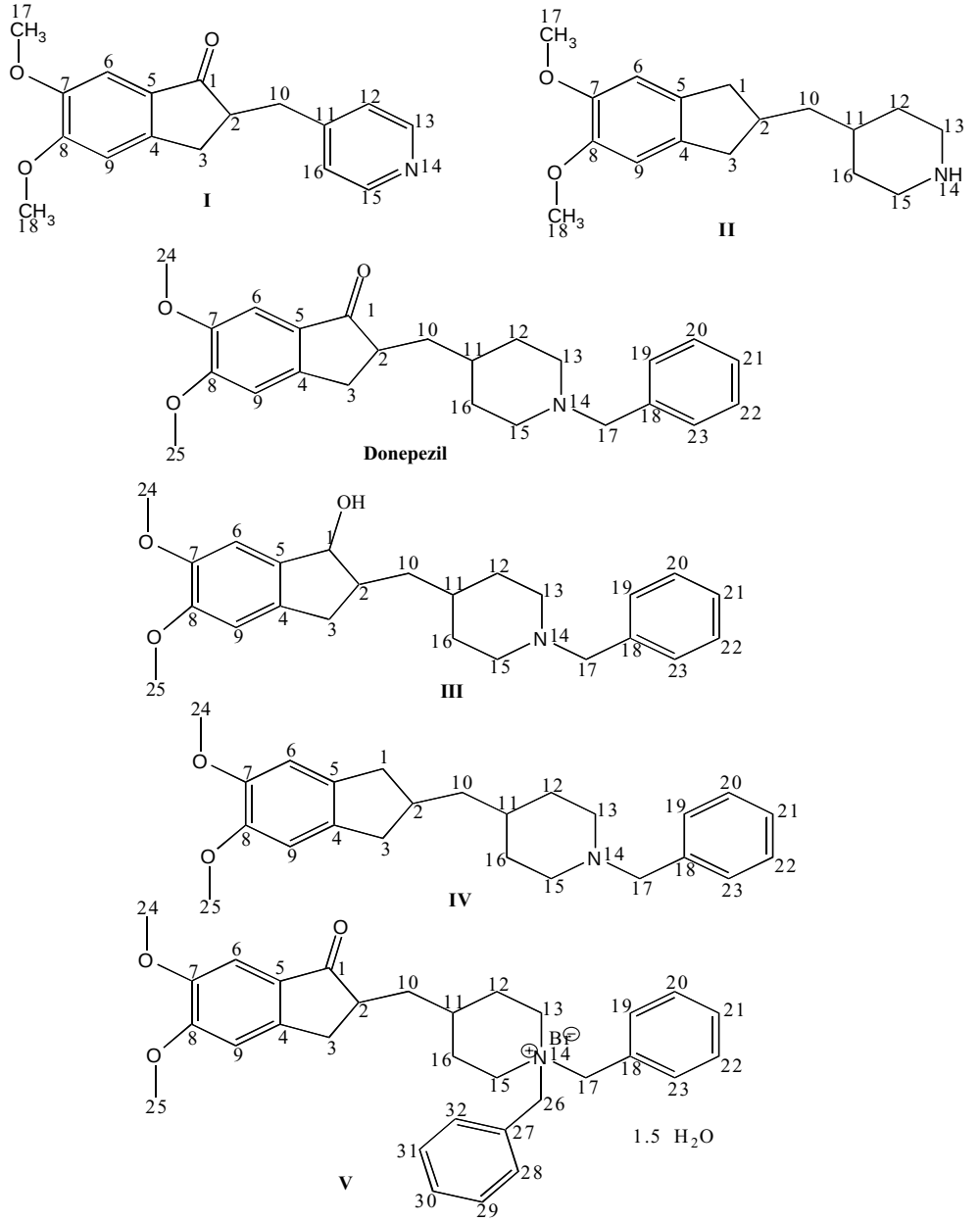


Fig. 3. The chemical structures of donepezil, impurities I–V and numberingscheme for NMR.

The data obtained form the spectral studies can be rationalised in terms of impurity I having the molecular formula $C_{17}H_{17}NO_3$ and the corresponding structure was characterised as 5,6-dimethoxy-2-(4-pyridylmethyl)-1-indanone.

3.3.2. Structural elucidation of impurity II

The spectral data of impurity II is quite similar to that of impurity I. The EI mass spectrum of impurity II exhibited molecular ion at *m/z* 275 atomic mass units (amu) which was eight amu less than that of impurity

I. The quaternary carbon signal at 206 ppm in the ^{13}C NMR spectrum and the characteristic signal due to carbonyl group in the FT-IR spectrum of impurity I were absent in the spectra of impurity II. The DEPT spectrum of impurity II exhibited five negative signals due the presence of seven methylene groups, whereas impurity I showed two negative signals due to two methylene groups. The DEPT spectrum of impurity II also exhibited an additional methine signal when compared to that of impurity I. The above data can be rationalised in terms of reduced carbonyl functionality and saturation of pyridine ring in impurity II.

Based on the above spectral data, the molecular formula of impurity II could be $C_{17}H_{25}NO_2$, which matched well with the observed molecular ion at 275 amu in the MS spectrum. The corresponding structure of impurity II was characterised as 4-(5,6-dimethoxy-2,3-dihydro-1H-2-indenylmethyl) piperdine.

3.3.3. Structural elucidation of impurity III

The spectral data of impurity III is compared with that of donepezil.

The EI mass spectrum of impurity III exhibited molecular ion at *m*/*z* 381 atomic mass units (amu), which was two atomic mass units more than that of donepezil. The characteristic carbonyl absorption band at $\sim$1690 cm^{-1} in the FT-IR spectrum and the quaternary carbon signal at 206 ppm in the ^{13}C NMR spectrum of donepezil were absent in the spectra of impurity III. In addition to this observation, a broad absorption band at $\sim$3330 cm^{-1} was observed which indicated the possible presence of hydroxy group in impurity III. An additional methine signal was observed in the DEPT spectrum of impurity III at 81.83 ppm when compared to that of donepezil. The observations can be rationalised in terms of conversion of carbonyl group in donepezil to hydroxy group in impurity III.

Based on the above spectral data the molecular formula of impurity III was confirmed as $C_{24}H_{31}NO_2$ and the corresponding structure was characterised as 2-(1-benzyl-4-piperdylmethyl)-5,6-dimethoxy-1-indanol.

3.3.4. Structural elucidation of impurity IV

The spectral data of impurity IV was compared with that of donepezil.

The EI mass spectrum of impurity IV exhibited molecular ion at *m*/*z* 365 atomic mass units (amu), which is fourteen atomic mass units less than that of donepezil. The characteristic carbonyl absorption band at $\sim$1690 cm^{-1} in the FT-IR spectrum and the quaternary carbon signal at 206 ppm in the ^{13}C NMR spectrum of donepezil were absent in the spectra of impurity IV. In addition to this observation, an extra methylene group signal was observed in the DEPT spectrum of impurity IV which was absent in that of donepezil. This observation indicates the addition of two hydrogens and removal of one oxygen atoms. These observations can be rationalised in terms of reduction of carbonyl group in donepezil to methylene group in impurity VI.

Based on the above spectral data the molecular formula of impurity IV was confirmed as $C_{24}H_{31}NO_2$ and the corresponding structure was characterised as 1-benzyl-4(5,6-dimethoxy-2,3-dihydro-1H-2-indenylmethyl) piperidine.

3.3.5. Structural elucidation of impurity V

The spectral data of impurity V are compared with that of donepezil.

The EI mass spectrum exhibited molecular ion at 470 atomic mass units (amu), which is 91 amu more than that of donepezil. The extra signals in the ^{1}H, ^{13}C and DEPT spectra in the aromatic region of impurity V indicate the presence of an additional aromatic ring. An additional signal at $\sim$5 ppm in the ^{1}H NMR spectrum appeared as two doublets integrating for two protons indicate the presence of an additional methylene group in impurity V. The splitting of this particular signal into two doublets (though there were no adjacent protons) could be explained in terms of pro-chiral nature of this methylene group. In addition, signals integrating for five additional protons are also observed in the aromatic region in the ^{1}H NMR spectrum of impurity V. These signals are absent in the ^{1}H NMR spectrum of donepezil. These observations are supported by the appearance of additional methylene signal and methine signals in the ^{13}C and DEPT spectra of impurity V. The FT-IR spectrum of impurity V showed a strong and broad absorption band at 3433 cm^{-1}, which is characteristic of hydroxy group. But based on the MS and NMR data this hydroxy group was not present in impurity V. The thermogravimetric (TG) analysis of impurity V showed a weight loss of 5.4% (w/w) in the temperature range of 30–200 °C. This weight loss was further confirmed by determining the moisture

Table 4
Comparative ^{1}H and ^{13}C NMR assignments for donepezil and impurities III–V

Position[a]	Donepezil				Impurity III				Impurity IV				Impurity V			
	^{1}H	*J* (ppm)	^{13}C	DEPT	^{1}H	*J* (ppm)	^{13}C	DEPT	^{1}H	*J* (ppm)	^{13}C	DEPT	^{1}H	*J* (ppm)	^{13}C	DEPT
1	–	–	207.61	–	1H	4.92, d (7.5)	81.83	CH	2H	2.98, m	42.17	CH_2	–	–	206.87	–
2	1H	3.28, m	45.21	CH	1H	3.06, m	42.38	CH	1H	2.12, m	39.32	CH	1H	3.28, m	44.26	CH
3	Ha	2.65, m	38.55	CH_2	Ha	2.62, m	36.12	CH_2	Ha	2.54, m	27.64	CH_2	Ha	2.67, m	37.56	CH_2
	Hb	2.99, m			Hb	2.88, m			Hb	2.98, m			Hb	3.14, m		
4	–	–	129.13	–	–	–	135.39	–	–	–	134.47	–	–		148.72	–
5	–	–	148.59	–	–	–	136.65	–	–	–	128.14	–	–		128.66	–
6	1H	6.82, s	104.19	CH	1H	6.72, s	106.76	CH	1H	6.63, s	107.77	CH	1H	6.82, s	104.15	CH
7	–	–	155.27	–	–	–	149.47	–	–	–	147.76	–	–		149.51	–
8	–	–	149.25	–	–	–	148.18	–	–	–	147.87	–	–		155.73	–
9	1H	7.09, s	107.21	CH	1H	6.95, s	107.52	CH	1H	6.72, s	107.77	CH	1H	7.09, s	107.38	CH
10	Ha	1.84, m	33.16	CH_2	Ha	1.97, m	32.79	CH_2	Ha	1.79, m	33.36	CH_2	Ha	1.83, m	33.39	CH_2
	Hb	1.53, m			Hb	1.42, m			Hb	1.42, m			Hb	1.67, m		
11	1H	1.84, m	34.31	CH	1H	1.97, m	34.06	CH	1H	1.42, m	37.43	CH	1H	1.90, m	31.15	CH
12 and 16	4H	2.06, m	32.89	CH_2	4H	1.75, m	32.69	CH_2	4H	2.15, m	39.32	CH_2	4H	2.06, m	25.66	CH_2
			31.66				31.72								26.15	
13 and 15	4H	3.47, m	53.63	CH_2	4H	1.97, m	53.56	CH_2	4H	2.54, m	56.41	CH_2	4H	3.87, m	54.89	CH_2
		2.05, m				2.88, m					56.66			3.16, m		
14	–	–	–	–	–	–	–		–	–	–	–	–		–	–
17	2H	4.17, s	63.29	CH_2	2H	3.51, s	63.28	CH_2	2H	4.09, s	70.10	CH_2	2H	4.88, s	59.57	CH_2
18	–	–	138.34	–	–	–	137.92	–	–	7.32–7.41	130.49	–	–		129.08	–
19 and 23	2H	7.61	129.03	CH	2H	7.31	129.17	CH	2H	7.32–7.41	128.68	CH	2H	7.74	130.66	CH
20 and 22	2H	7.43	127.96	CH	2H	7.31	127.93	CH	2H	7.51	132.76	CH	2H	7.37–7.54	129.35	CH
21	1H	7.43	126.73	CH	1H	7.31	126.76	CH	1H	7.34, s	127.74	CH	1H	7.37–7.54	130.55	CH
24	3H	3.82, s	55.91	CH_3	3H	3.84, s	55.79	CH_3	3H	3.89, s	56.00	CH_3	3H	3.87, s	55.99	CH_3
25	3H	3.95, s	56.03	CH_3	3H	3.89, s	55.79	CH_3	3H	–	56.00	CH_3	3H	3.94, s	56.23	CH_3
26	–	–	–	–	–	–	–	–	–	–	–	–	Ha	5.25, d	65.24	CH_2
27	–	–	–	–	–	–	–	–	–	–	–	–	–	–	126.61	–
28 and 32	–	–	–	–	–	–	–	–	–	–	–	–	–	7.37–7.54	133.20	CH
29 and 31	–	–	–	–	–	–	–	–	–	–	–	–	–	7.37–7.54	133.91	CH
30	–	–	–	–	–	–	–	–	–	–	–	–	–	7.37–7.54	127.28	CH

[a] Refer structural formula for numbering (Fig. 3 VIII, III, IV and V). s: singlet; d: doublet; m: multiple.

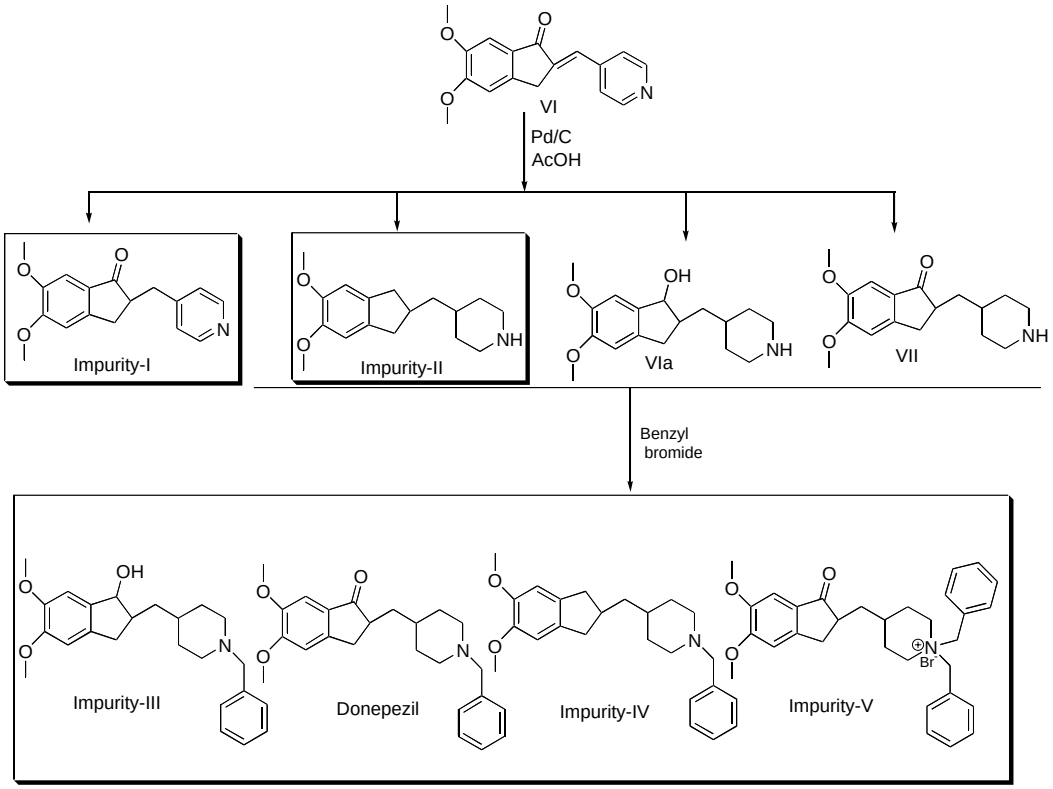


Fig. 4. The scheme for the formation of impurities.

content by Karl Fischer (KF) method. These observations from TG and KF experiments indicated impurity V was formed as a sesqui hydrate ($1.5H_2O$). The electrospray MS (ES-MS) spectrum in the negative mode showed the presence of an adduct of $[M^+ - Br^{2-}]$ in impurity V. The presence of bromide was further confirmed qualitatively by carrying out silver nitrate test, which yielded a light yellow precipitate. The above spectral data can be rationalised in terms of incorporation of benzyl group in impurity V. The position of attachment of this benzyl group was confirmed by carrying out 2D NMR experiments (COSY, HETCOR, gHMBC etc.).

Based on the above spectral data the molecular formula of impurity V was confirmed as $C_{24}H_{31}NO_2Br$ and the corresponding structure was characterised as 1,1-dibenzyl-4(5,6-dimethoxy-1-oxo-2,3-dihydro-2H-2-indenylmethyl) hexahydropyridinium bromide.

The spectral data of isolated and synthesised impurities were found to be identical. The synthetic standards of impurities I–V were co-injected on LC with donepezil and the area percentage at retention times 3.5, 4.0, 5.5, 13 and 19 min were enhanced and the LC chromatogram is shown in Fig. 2b.

The FT-IR and Mass Spectral data for donepezil, impurities I–V are shown in Table 2. The ^1H, ^{13}C and DEPT chemical shift values for impurities I and II are given in Table 3. The ^1H, ^{13}C NMR and DEPT assignments for donepezil, impurities III–V are given in Table 4. The chemical structures of donepezil, impurities I–V and numbering scheme for NMR are shown in Fig. 3.

3.4. Formation of impurities

One of the intermediates used in the synthesis of donepezil is VI. This on hydrogenation yields VII, which leads to another process intermediate. During this reaction, the double bond in VI gets reduced leading to the formation of impurity I. During the same

reaction, the reduction of carbonyl group as well as the saturation of pyridine ring leads to the formation of impurity II. During the reaction, the conversion of carbonyl group to hydroxy group may also take place leading to the formation of VIa.

The impurity II and VIa thus formed reacts with benzyl bromide during the final step of synthesis of donepezil leading to impurities IV and III, respectively. The excess of benzyl bromide reacts with donepezil (VIII) leading to the formation of impurity V. The scheme for the formation of impurities is shown in Fig. 4. The rationalisation given above clearly indicates that these impurities are process-related.

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