SYNTHESIS AND ANTIMICROBIAL ACTIVITY OF SOME BENZIMIDAZOLE DERIVATIVES

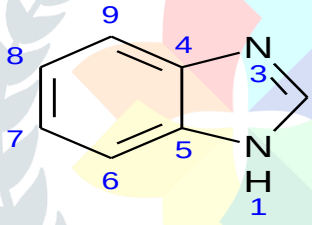
[1]Tajamul Khan, [2]Dr. Kishore Singh Chatrapati, [3]H.J.Kallur, Prabhudev S.M
[1,2,3]Department of Pharmaceutical Chemistry, RMES'S College of Pharmacy, Kalaburgi Karnataka, India

Abstract: In the present study, a series of substituted 3-(1H-benzimidazol-2-ylsulfanyl)-1-phenylpropane-1, 2-dione was prepared. The synthesis of titled compounds from starting material unsubstituted 2-mercapto benzimidazoles was prepared from o-phenylenediamine and carbon disulfide in presence of KOH in single step.2-mercapto Benzimidazole on reacting with substituted 3-chloro-1-phenylpropane-1, 2-dione yield different derivatives of Benzimidazole. The structure of new compounds prepared during present investigation has been authentically established by their IR, 1H NMR and Mass spectral studies. The antibacterial and antifungal activities of benzimidazole derivatives were reported.

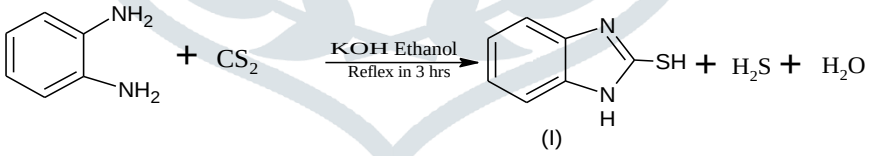
Keywords: - 2-mercapto benzimidazoles, phenylpropane-1, 2-dione, antibacterial, antifungal.

1. INTRODUCTION

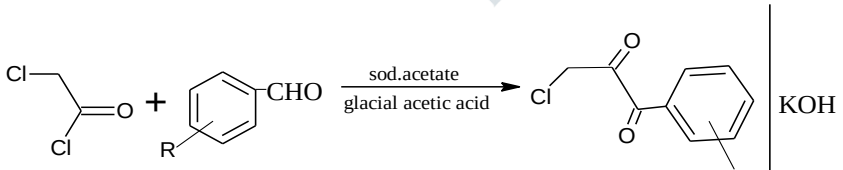
Benzimidazole is a fused aromatic imidazole ring system where a benzene ring is fused to the 4 and 5 positions of an imidazole ring resulting in benzimidazole and is numbered as follows. Benzimidazoles are also known as benziminazoles and 1, 3-benzodiazoles.1, 2 they possess both acidic and basic characteristics. The NH group present in benzimidazoles is relatively strongly acidic and also weakly basic. Another characteristic of benzimidazoles is that they have the capacity to form salts. benzimidazoles with unsubstituted–NH- groups exhibit fast prototropic tautomerism, which leads to equilibrium mixtures of asymmetrically substituted compounds.1 in 1953, KLAUS HOFMANN covered the entire chemistry of monocyclic imidazoles and benzimidazoles. It was discovered that benzimidazole is an integral part of the structure of vitamin b_{12}



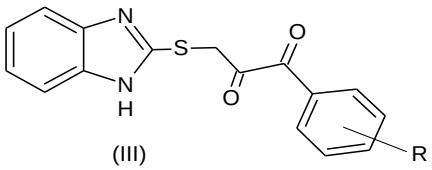
2. EXPERIMENTAL SCHEME



(I)



(II)



(III)

Where R=-H, 2-Cl, 3-Cl, -OCH_3, 4-CH_3.

3. MATERIALS AND METHODS

The chemicals and reagents used in the present project were of AR grade and LR grade, purchased from Lancaster, Sigma, Qualigens, NR Chem., Rolex, S.D. Fine Chem. Ltd., Merck, Loba, Himedia, Sahiyadri and, gravity suplyer. O-phenylenediamine, potassium hydroxide, carbon disulfide chloroacetylchloride, sodium acetate, Ethanol, Benzaldehyde, Anisaldehyde,2-chlorobenzaldehyde, 3-chlorobenzaldehyde, 4-methyl Benzaldehyde, etc were used.

The completion of reactions was monitored by TLC technique using Silica gel-G (for TLC) using suitable solvent. Determination of melting point was done by open capillary tube method using paraffin bath and are uncorrected. Recrystallization was done by suitable solvent. The 1H NMR of synthesized compounds were recorded in Bruker FT-NMR (400MHz & 200MHz) as $CDCL_3$ as internal standard and IR-spectra were recorded in 4000-650 cm^{-1}Bruker alpha FT-IR using KBr pellets. The Mass spectra were recorded on Shimadzu LC-MS with ESI sour

EXPERIMENTAL PROCEDURE

Step-1: Preparation of 2-mercapto benzimidazole:

A mixture of 10.8gm (0.1mole) of o-phenylenediamine, 5.65gm (0.1mole) of potassium hydroxide and 7.67 gm (0.1mole, 6.19ml) of carbon disulfide, 100ml of 95% ethanol and 15 ml of water in a 500ml round bottom flask heated under reflux for three hours. Then added 1-1.5 gm of charcoal cautiously and the mixture was further heated at the reflux for 10 minutes, the charcoal is removed by filtration. The filtrate was heated to 60-70 ^{0}C, 100ml of warm water is added, and acidified with dilute acetic acid with good stirring. The product separated as glistening white crystals, and the mixture is placed in a refrigerator for three hours to complete the crystallization. The product was collected on a Buckner funnel and dried over night at 40^{0}C. The dried product was recrystallized by ethanol the yield is 8.5gm (73%) melting point is 300-305^{0}C.

Step-2: General procedure for preparation of substituted 3-chloro-1-phenylpropane-1, 2-Dione:

The aromatic aldehyde (0.05mole) were dissolved in a mixture of glacial acetic acid (25ml) and saturated solution of sodium acetate (25ml) and cooled to 5 ^{0}C. To this, chloroacetylchloride (6.2ml. 0.075 mole) was added drop wise at 0-5 ^{0}C under constant to continuous stirring on magnetic stirrer. Then it was left at room temperature for 8-10 hrs and evaporates the mixtures to dryness. The crude product was collected filtered, washed with cold water. It was recrystallized from suitable solvent.

Step-3: General procedure for the preparation of substituted 3-(1H-benzimidazol-2-ylsulfanyl)-1-phenylpropane-1, 2-dione:

2-mercaptobenzimidazole (I) (1.50gm. 0.01 mole) was dissolved in aqueous potassium hydroxide solution (0.61gm in 10ml water) with stirring till a clear yellow solution was obtained. It was filtered to remove any suspended impurities. Then various aromatic aldehyde substituted chloroacetylchloride (II) (0.011mole) were added in small proportions with continuous stirring till the immediate product was obtained. The precipitate was filtered and washed with cold water to remove KCl and dry. Dried product (III) was recrystallized from aqueous ethanol.

Table No. 3.1
Physicochemical Data of Chloroacetylchloride Substituted Aromatic Aldehyde

Sl. NO	Compound Code	R	Molecular Formula	Molecular weight	M.P (^{0}C)	Yield (%)
1	B1	-H	$C_9H_7ClO_2$	182.60	228-230 ^{0}C	77%
2	B2	2-Cl	$C_9H_6Cl_2O_2$	217.05	270-272 ^{0}C	74%
3	B3	3-Cl	$C_9H_6Cl_2O_2$	217.05	241-243 ^{0}C	69%
4	B4	-OCH_3	$C_{10}H_9ClO_3$	247.07	196-198 ^{0}C	79%
5	B5	4-CH_3-	$C_{10}H_9ClO_2$	231.07	280-282 ^{0}C	75%

Table No. 3.2
Physicochemical data of substituted 3-(1H-benzimidazol-2- ylsulfanyl)-1-phenyl propane-1,2-dione

Sl. No.	Compound Code	R	Molecular Formula	Molecular weight	M.P (^{0}C)	Rf Value	Yield (%)
1	**C-1**	-H	$C_{16}H_{12}N_2O_2S$	296.34	248-250 ^{0}C	0.58	78%
2	**C-2**	2-Cl	$C_{16}H_{11}ClN_2O_2S$	330.78	220-222 ^{0}C	0.60	72%
3	**C-3**	3-Cl	$C_{16}H_{11}ClN_2O_2S$	330.78	240-242 ^{0}C	0.59	73%
4	**C-4**	-OCH_3	$C_{17}H_{14}N_2O_3S$	326.36	230-232 ^{0}C	0.62	77%
5	**C-5**	4-CH_3-	$C_{17}H_{14}N_2O_2S$	310.37	222-224 ^{0}C	0.65	73%

4. RESULTS

4.1 RESULTS OF IR SPECTRA

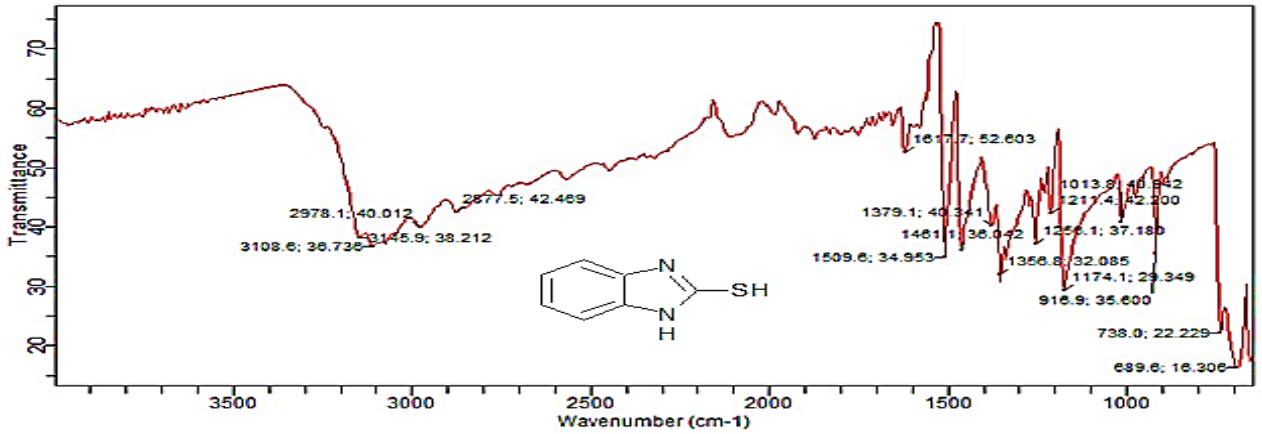


Figure No. 4.1
IR Spectra of Sample A

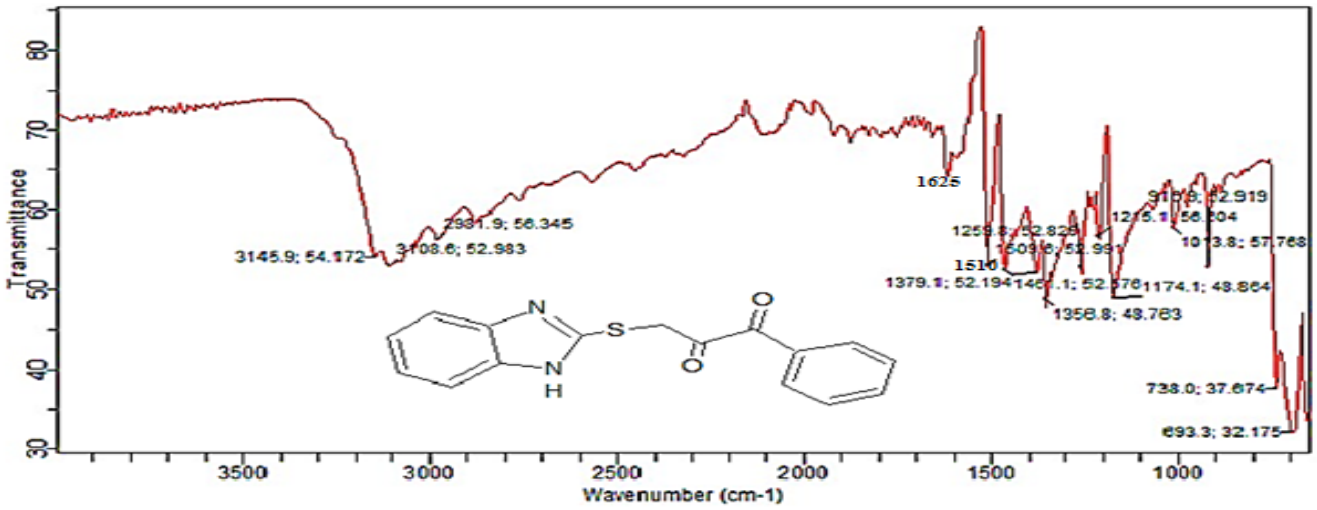


Figure No. 4.2
IR Spectra of Sample C 1

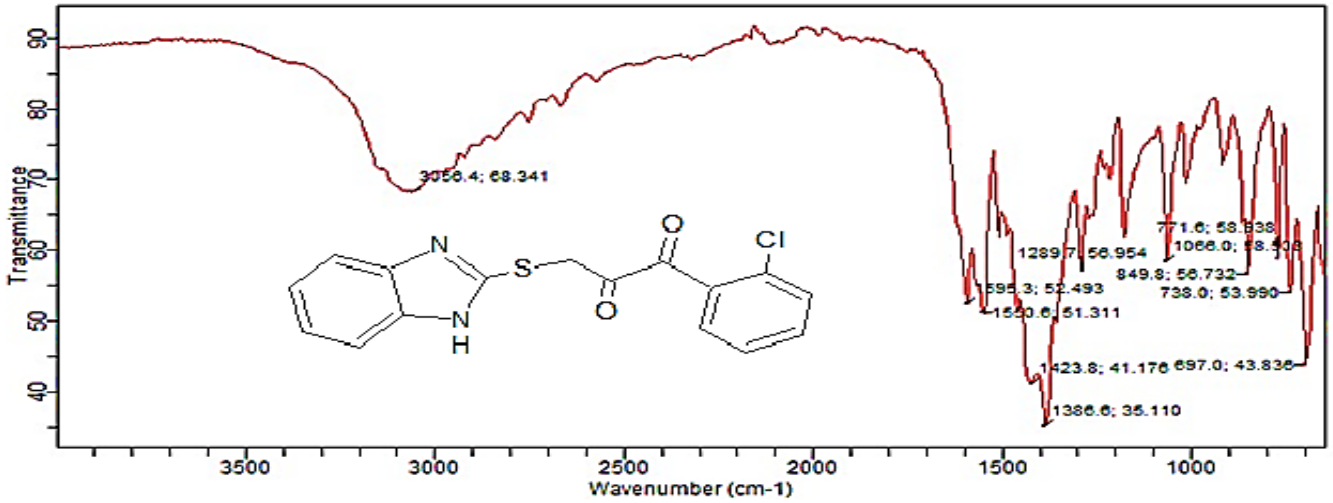


Figure No. 4.3
IR Spectra of Sample C 2

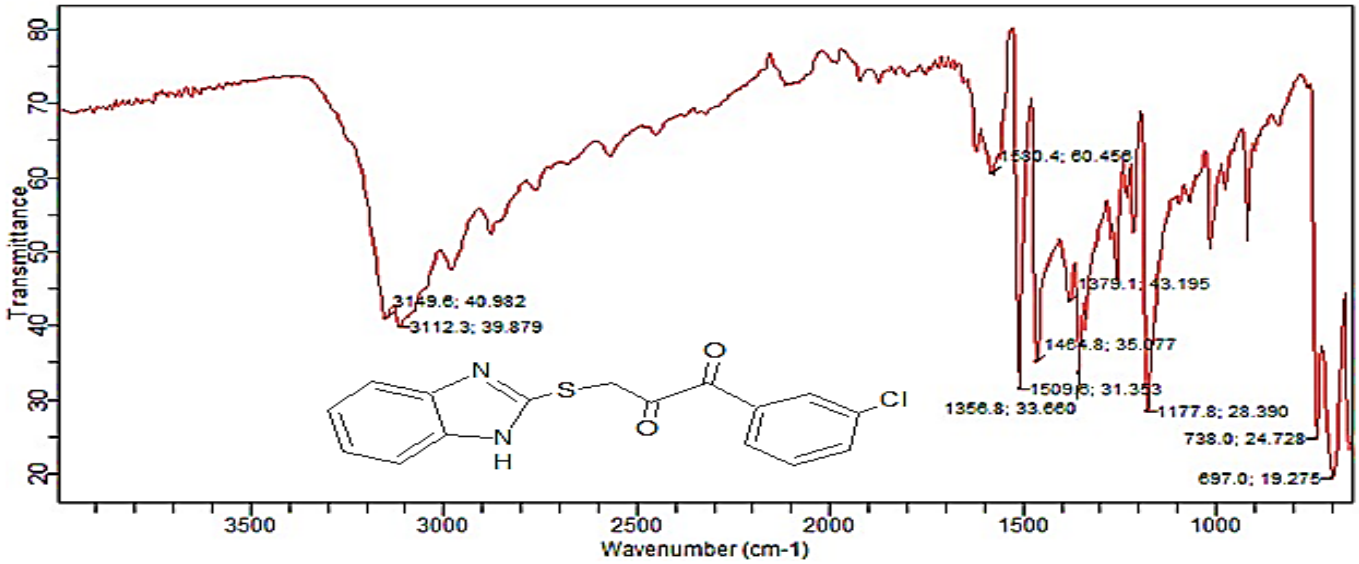
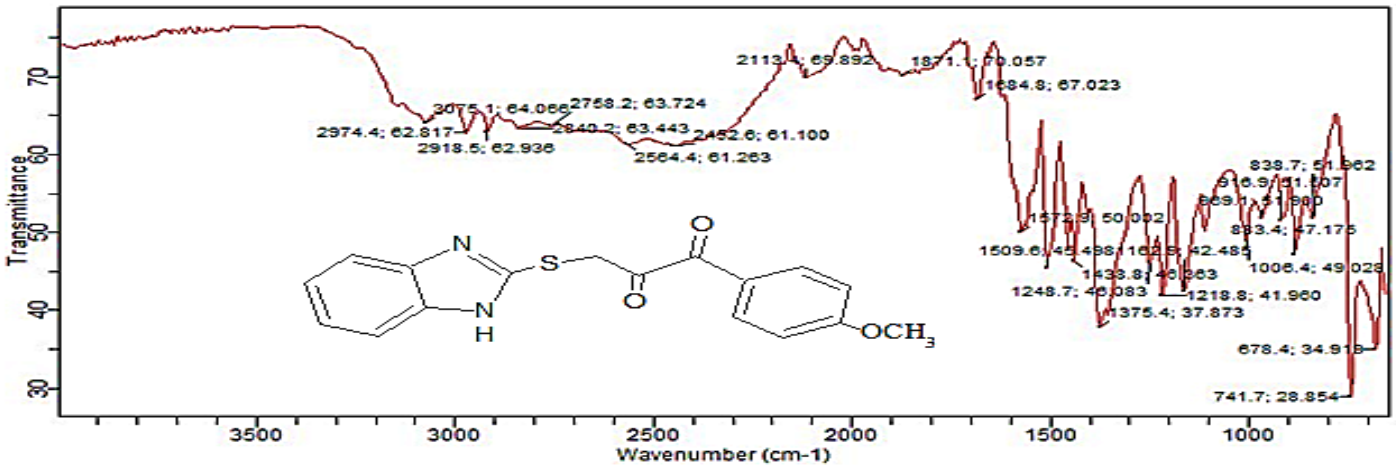


Figure No. 4.4



IR Spectra of Sample C 3
Figure No. 4.5

IR Spectra of Sample C 4

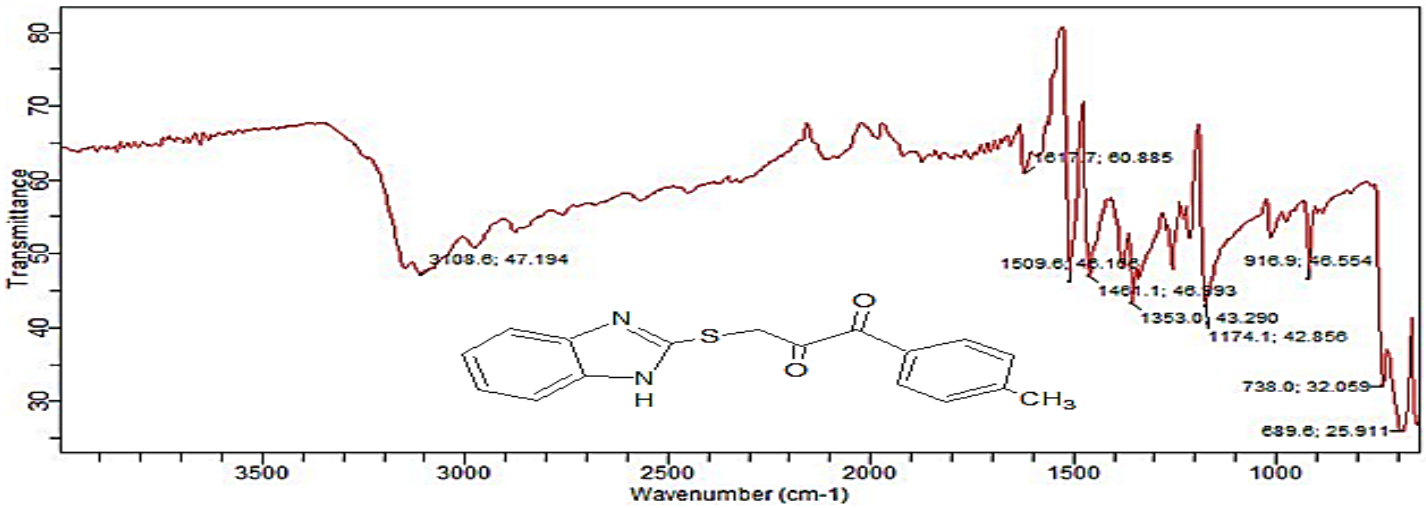


Figure No. 4.6
IR Spectra of Sample C 5

4.2 RESULTS OF ^{1}HNMR

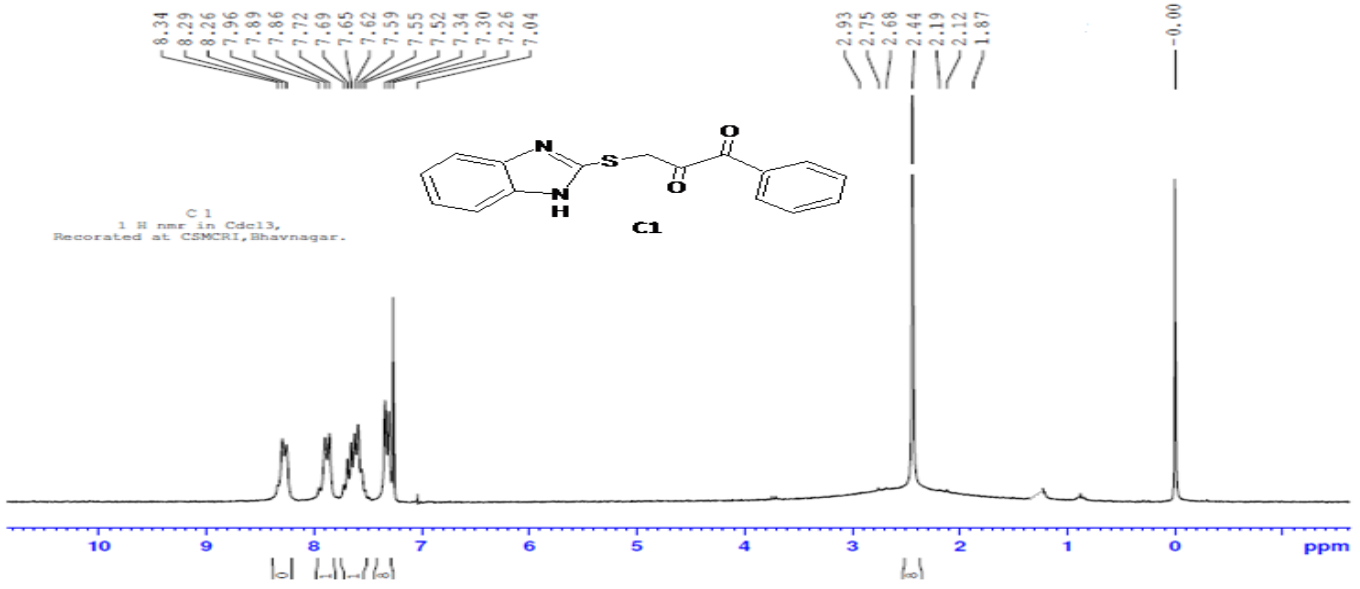


Figure 4.7
^{1}H-NMR Spectra for Sample C 1

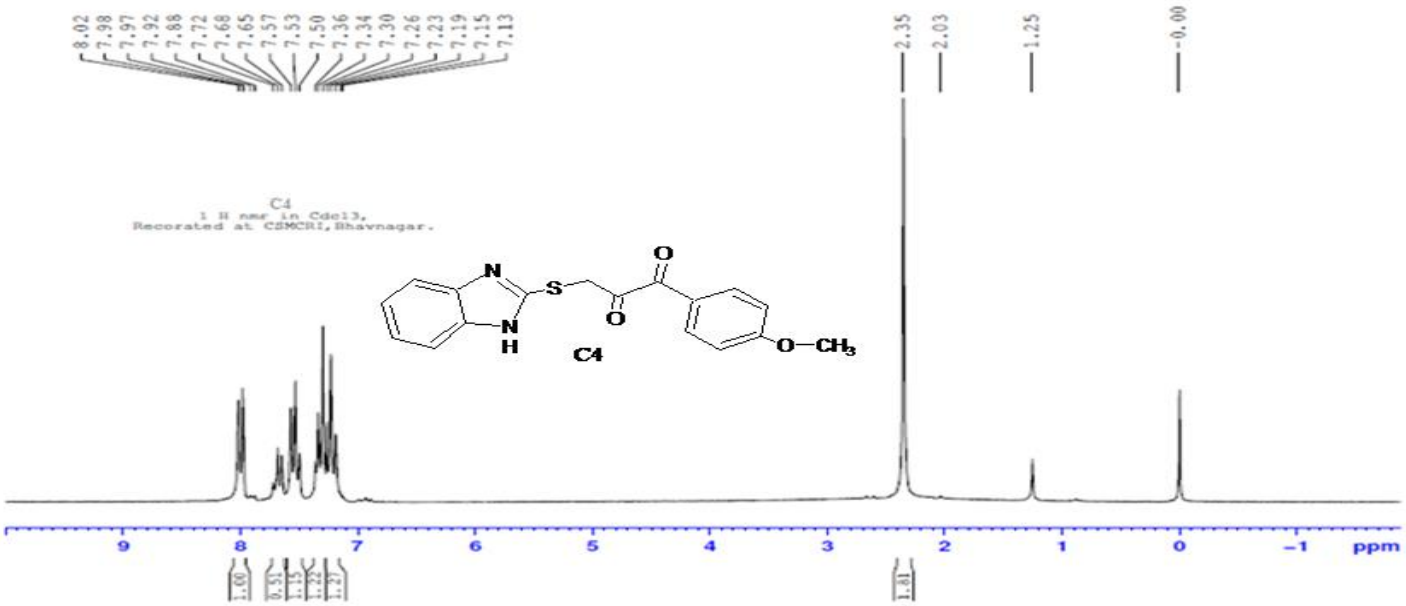


Figure 4.8
^{1}H-NMR Spectra for Sample C 4

4.3 RESULTS OF MASS SPECTROSCOPY

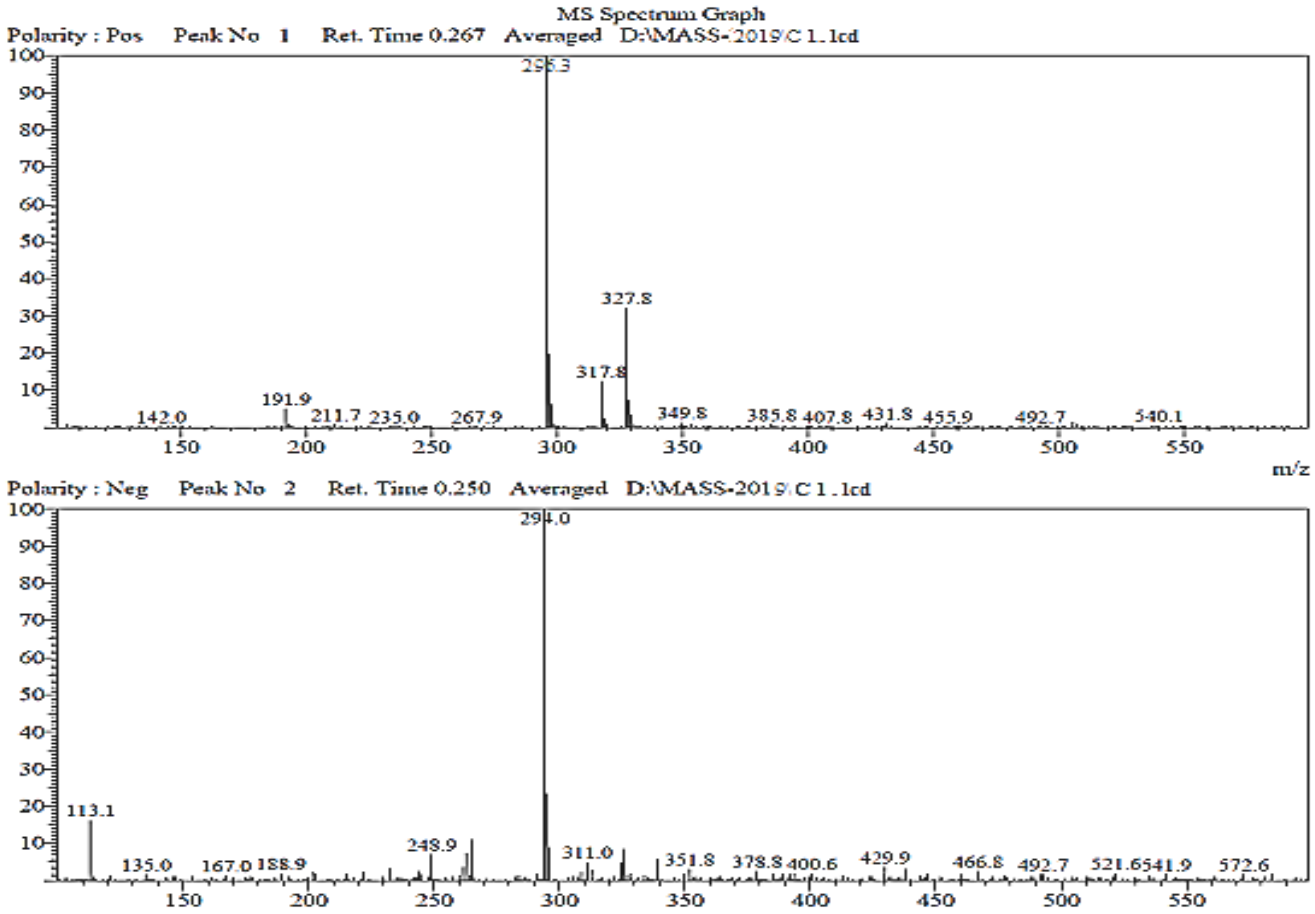


Figure 4.9
Mass Spectra for Sample C1

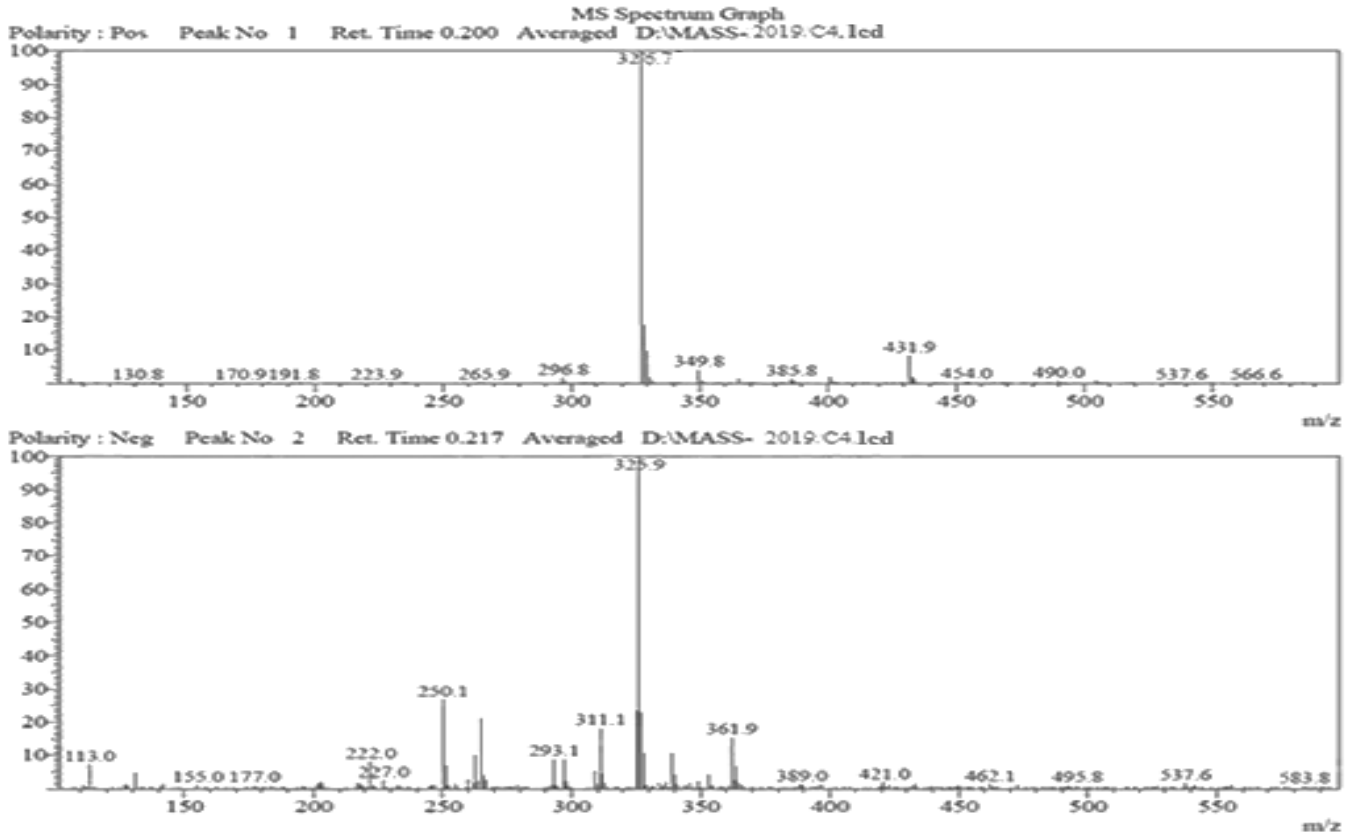


Figure 4.10
Mass Spectra for Sample C4

5. DISCUSSION

A series of substituted 3-chloro-1-phenylpropane-1,2-dione. / [**B1 to B5**] and substituted 3-(1H-benzimidazol-2-ylsulfanyl)-1-phenylpropane-1,2-dione. / [**C1 to C5**] were synthesized in good yield using the synthetic route outlined in Scheme 2 and 3.

The starting material of 2-mercapto benzimidazole was prepared according to the literature procedure. Structure of the synthesized compounds was stabilized on the basis of IR, ^{1}H-NMR, and Mass spectral data.

It was noteworthy that such a procedure for rapid preparation of various benzimidazoles derivatives affords advantages of short reaction time, moderate yields and simple workup. The IR spectrum of **C1** compound exhibited N-H absorption peak at 3145 cm^{-1} which was the normal place of absorption of N-H of benzimidazole moiety. The Ar-H of aryl resonated at 3108 cm^{-1}. The aromatic C=N peak was noticed 1509 cm^{-1} but aliphatic C-H at 2981 cm^{-1}. The C=O of the appeared at 1625 cm^{-1}. The aromatic C=C peak noticed 1510cm^{-1} and C-S peak was showed at 738 cm^{-1}.These data in confirmative with the structural propose of the synthesized compound.

The IR spectrum of **C2** also gave Ar-H absorption peak at 3056cm^{-1}. In these case the C=N peak showed at 1595 cm^{-1}and C=C peak showed at 1423 cm^{-1}, C=O of absorbed at 1550 cm^{-1} , C-Cl and C-S peaks showed at 697 cm^{-1}and 738 cm^{-1}respectively. These IR data expected concurrent for proposed molecule.

The next analogue of the compound **C3**.The IR spectrum of Ar-H of benzimidazole gave an absorption peak at 3112 cm^{-1} and C=C at 1356 cm^{-1}. The C=N peak found to be absorbed at 1509 cm^{-1}.The aliphatic C-H peak was found to be absorbed at 3149cm^{-1}. The C=O group gave absorption peak at 1580 cm^{-1}, C-Cl and C-S peaks showed at 697 cm^{-1}and 738 cm^{-1}respectively.These IR data expected for the molecule under investigation.

The **C4** compound the IR data obtained in resembling with the IR data found for the compounds already described. The H of N-H found at 2918 cm^{-1} and H of C-H aliphatic at 2840 cm^{-1}. The C=O group gave absorption peak at 1684 cm^{-1}.The groups of C=C and C=N gave the absorption peaks at 1438 cm^{-1} and 1509cm^{-1}.The Ar-H group gave the absorption peak at 2974 cm^{-1}.The groups of C-N, C-S and C-O-C (ether) gave the absorption peaks at 1248 cm^{-1},741 cm^{-1}and 1218 cm^{-1} respectively. These data obtained resembling with expected data. The next compound **C5** taken for IR measurement of groups C=C, C=N and C=O absorptions peaks at 1461 cm^{-1},1509 cm^{-1} and 1617 cm^{-1} This compound produce IR measurement identical with the previous spectrum discusses. In this case also Ar-H, C-N, C-S absorption peak appear at the places where they expected to appear.

^{1}H-NMR spectrum of compound **[C1]** showed the characteristic singlet around 2.44 δ ppm for 1H, -CH_2 proton at 2nd position of benzimidazole, a triplet at 7.04 - 7.34 δ ppm for 1H, aromatic proton at 1st position of benzimidazole a multiplet around 7.52 -7.72 δ ppm for 5H, aromatic proton at 2nd position and doublet around 7.86-8.34 δ ppm for 5H, aromatic proton at

4th and 5th fused benzimidazole. Similarly ^{1}H-NMR spectrum of compound **[C4]** showed the characteristic singlet around 1.25 δ ppm for 3H, -OCH_3 proton at 4-methoxyphenyl-propane-1,2 dione , a singlet at 2.35 δ ppm for 1H, -CH_2 proton at 2nd position of benzimidazole, triplet around 7.13-7.36 δ ppm for 1H, aromatic proton at 1st position of benzimidazole, multiplet around 7.50-7.72 δ ppm, 4H, aromatic proton at 2nd position and doublet around 7.88-8.02 δ ppm 5H, aromatic proton at 4th and 5th fused benzimidazole.

The mass spectra showed an accurate molecular ion peak data at 296 m/z [M+1] for compound [**C1]** and at 326 m/z [M+1] for compound [**C4]**. All the compounds give satisfactory chemical analysis.

6. ANTIMICROBIAL ACTIVITY

The test was performed by disc diffusion method. The nutrient agar plates containing an inoculums size of 106cfu/ml for bacteria and 2×105 spores for fungi on Saboraud glucose agar plates were used. Previously prepared compound impregnated disc (6mm in diameter) at the concentrations of 200 µg/ml for bacterial and 200 µg/ml for fungal strains were placed aseptically on sensitivity plates with appropriate controls Ofloxacin (200 µg/ml) and Griseofulvin (200 µg/ml) were used as standard antibacterial and antifungal antibiotics respectively. Plates were incubated at 37 ^{0}C for 24 hours for bacteria and 30 ^{0}C for 72 hours for fungal inoculums. Sensitivity was recorded by measuring the clean zone of growth inhibition on agar surface around the disc.

Table No. 6.1
Antibacterial Activity of Synthesized Compounds

Sl. No	Compound code (200 µg/ml)	S. aureus(mm)	E. coli (mm)
1	**C1**	11.00 ± 0.40	12.38 ± 0.01
2	**C2**	10.43 ±0.08	10.13 ± 0.07
3	**C3**	12.40 ± 0.02	11.25 ± 0.75
4	**C4**	13.90 ± 0.01	10.90 ± 0.14
5	**C5**	11.27 ± 0.99	10.02 ± 0.40
6	**Ofloxacin**	**16.13 ± 0.99**	**15.33 ± 0.09**

All values are expressed as mean ± S.E.M. of three replications.

Table No. 6.2

Antifungal Activity of Synthesized Compounds

Sl. No	Compound code (200 µg/ml)	A. niger(mm)	C. albicans(mm)
1	C_1	13.43 ± 0.88	12.16 ± 0.07
2	C_2	11.88 ± 0.00	10.87 ± 0. 76
3	C_3	12.08 ± 0.88	10.12 ± 0.88
4	C_4	13.08 ± 0.88	12.10 ± 0.76
5	C_5	12.78 ± 0.00	12.77 ± 0. 76
6	**Griseofulvin**	**16.13 ± 0.99**	**14.22 ± 0.14**

All values are expressed as mean ± S.E.M. of three replications

The synthesized all benzimidazole derivatives were screened for antibacterial activity using DMF as a solvent against the organisms, S.aureus and E.coli and Antifungal activity using Candida albicans .By disc diffusion method on nutrient agar media. The Ofloxacin was used as standard drug for antibacterial and Griseofulvin as standard for antifungal activity. **C1, C2, C3** and **C5**showed good activity at concentration 50µg/ml and at 100µg/ml they possess good activity against E.coli and S .Aureus. **C4** shows good activity at 50µg/ml but at 100µg/ml it shows very good activity against S.aureus when compared with Ofloxacin. The same Compounds also screened for the antifungal activity against Candida albicans the compounds **C1, C2, C3, C4** and **C5** shown no growth at 250µg/ml and 500µg/ml against C albicans when compared with the standard drug Griseofulvin. However the activities shown by all the compounds were less than that of the standard.

7. CONCLUSION

In this work, the reaction of 2-mercaptobenzimidazole and various substituted 3-chloro-1-phenylpropane-1,2-dione in presence of aq. KOH in the synthesis of benzimidazole derivatives is successfully carried out. The data obtained from IR, 1HNMR and Mass data resembled with expected data.

The data obtained in confirmative with the structural propose of the synthesized compound. Since some of the present new benzimidazole derivatives exhibit moderate antibacterial activity compared with the standard employed, it is desirable to determine their toxicity to decide on whether to go for further screening or not.

8. ACKNOWLEDGEMENT

Authors are thankful to Dr. Kishore Singh Chatrapati Professor & HOD, Dr. Ashok Kumar Malpani, Principal and Professor of RMES's College of Pharmacy, for providing necessary facilities. They are also thankful to Oxygen Healthcare Research Pvt. Ltd, Ahmedabad and CSMCRI, Bhavnagar for providing the Spectral Data.

9. REFERENCES

1. Chander Mohan, Rajesh Kumar, et al., "Evaluation and characterisation of antibacterial potential of Novel Schiff bases of benzimidazole": International Research Journal of Pharmacy; Jan .2019.10 (2).
2. Ansari Yaasir Ahmed, Mohd Farooque, Anwar Ahmad, Md. Azhruddin, M.S Sayyed 2019. Phytochemical Investigation Studies of Anti-Diabetic Phytochemicals of Plant Murraya Koenigii (Linn).Spreng belonging to Family Rutaceae. International Journal of Research in Advent Technology. 7(4S): 45-49.
3. Garima Shrivastava, Manjul Shrivastava. ''Microwave assisted synthesis and characterization of Schiff base of 2-amino benzimidazole": International Journal of Pharmaceutical Sciences and Drug Research 2018; 10(4): 293-296.
4. Ekta Khare and G. Mariappan. "Synthesis and biological evaluation of benzimidazole": International journal of research in pharmacy and chemistry; 2018, 8(1), 61-68.

5. G. ARORA, S. SHARMA, T. SAHNI AND P.SHARMA. "Antioxidant and Antimicrobial Activity of Some 2-Phenyl-1H-indoles and Benzimidazoles": Indian Journal of Pharmaceutical Sciences, 2018; 80(4):739-744.
6. Sangram U. Deshmukh, Dr. B. Rajeeva, et al., "Synthesis, chemical characterization and antimicrobial activity of some new benzimidazole derivatives": World journal of pharmacy and pharmaceutical sciences; 01 Feb. 2018, Volume 7, Issue 4, 1112-1129.
7. Hadole CD, Rajput JD and Bendre RS. "Concise on Some Biologically Important 2-Substituted Benzimidazole Derivatives": Organic Chemistry Current Research 2018, 7:3.
8. Jayakumar.A, Ramesh Dhani, Sindhu.P, et al., "Synthesis of some novel benzimidazole derivatives & evaluation of their antibacterial activity": International Journal of Chemistry and Pharmaceutical Sciences; 2018, 6(7): 194–200.
9. Navanath. V. Kalyane and Shivakumar. B, et. al., "Synthesis and Anti-Inflammatory Activity of Some New 2-Chloro-3-[3-(6-Nitro-1H-Benzimidazol-2-Yl)-1H-Pyrazol-5Yl] Quinoline": IOSR Journal of Pharmacy and Biological Sciences; Sep – Oct 2018, Volume 13, PP 83-88.
10. Girija K and Indhumathi S. "Synthesis, Characterization and Antimicrobial Study of 1, 2-Disubstituted Benzimidazoles": International Journal of Chemical Sciences; November 19, 2018; Vol-16.
11. Alaa Alkhafaji, Zuhair Muhi-Eldeen, et al. "Synthesis, Structural Elucidation, and Evaluation of Antimicrobial Activity of 5-Ethoxy-2-Mercaptobenzimidazole Derivatives": International Journal of Medical Research & Health Sciences, 2018, 7(5): 117-127.
12. Raja Sundararajan, Pullagura M Krishna Prasad. "Design, synthesis, antitubercular and antimicrobial activities of novel thiazole substituted benzimidazole derivatives": Scholars Research Library; Der Pharmacia Lettre, 2017, 9 [6]:270-284.
13. Thandra Yakaiah, Upender Rao Eslawath, et al. "Synthesis and antimicrobial evaluation of benzimidazole derivatives": IOSR Journal of Pharmacy and Biological Sciences; Volume 12, Issue 5 Ver. VII (Sep. – Oct. 2017), PP 23-28.
14. Snehlata Yadav, Balasubramanian Narasimhan, et al. "Synthesis, characterization, biological evaluation and molecular docking studies of 2-(1H-benzo[d]imidazol-2-ylthio)-N-(substituted4-oxothiazolidin-3-yl) acetamides": Chemistry Central Journal; (2017) 11:137.
15. Chandrashekhar G Devkate, Khandu D Warad, et al. "Green and efficient synthesis of benzimidazole derivatives and their antibacterial Screening": Journal of Chemical and Pharmaceutical Research, 2017, 9(8):209-213.
16. Mohammed Waseem, Mohammad Saqib, Kishore Singh Chatrapati, H. J. Kallur, et al. "Synthesis and antimicrobial activity of 2-(1h-benzimidazol -2-ylsulfanyl)-n phenyl- acetamide": American Journal of PharmTech Research. 2012; 2(2).