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## SYNTHESIS, CHARACTERIZATION AND BIOLOGICAL EVALUATION OF NOVEL IMIDAZO THIAZOLE CHALCONES AS ANTIOXIDANT AGENT

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### Keywords:

Antioxidant activity,  
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**ABSTRACT:** A series of condensed Imidazo [2, 1-*b*] [1,3] thiazole Chalcones have been synthesized by Claisen–Schmidt condensation reaction. Imidazo thiazole Chalcones are associated with immense biological activities. Novel Imidazo thiazole-based chalcones were synthesized using Imidazo thiazole-3-carboxaldehydes and various acetyl ketones in PEG-400. The structures of the compounds were characterized by IR, <sup>1</sup>H NMR and screened for their *in-vitro* antioxidant (DPPH and SOD) activity. DPPH free radical scavenging activity was given by the compounds 7a-j at with (0.5 mmol/L in methanol). The compounds having substituent hydroxyl, fluoro, methoxy, ethoxy, and fluoro as well as benzimidazole and a ferrocenyl moiety in chalcone structure (7a-j) shows enhancement in antioxidant activity. Particularly in compounds like 7e, 7f, 7g, and 7h show the moderate percentage and 7c show the strongest percentage of inhibition mainly at (C=1mM). Almost all the compounds indicate moderate to good antioxidant activity compared to that of standard ascorbic acid percentage antioxidant activity (1 mm) by DPPH and SOD as 44.18 and 74.07 respectively. The resultant absorbance was recorded at 517 nm after 30 min incubation at 37 °C. It was also found to be a potent scavenger of SOD, and this property may be responsible for the good anti-inflammatory activity of corresponding Imidazo-Thiazole Chalcones. Almost all the synthesized novel Imidazothiazole chalcones show potent antioxidant activity.

**INTRODUCTION:** Nitrogen and sulfur-containing heterocyclic are an important class of heterocyclic compounds useful in medicinal chemistry <sup>1-3</sup>.

Most of the five-membered nitrogen-containing heterocyclic are biologically active <sup>4-5</sup>. In the history of modern drug discovery, fused heterocyclic with Imidazothiazole moiety are interesting forthcoming objects.

Imidazothiazole derivatives have long been therapeutically used to cure various diseases. In recent years, the imidazothiazole and chalcone moieties play an important role as powerful pharmacophore units. Imidazo [2, 1-*b*] thiazoles are well-known heterocyclic compound, and a new

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series of imidazothiazoles chalcones system possess potent biological activities that could be further developed as a drug. Imidazothiazoles compounds have broad scope to synthesize a large number of new chemotherapeutic agents, and these are used in remedying new compounds which useful in scientific medicines. Intensively a great idea has been developed behind the synthesis, and biological activities of the condensed Imidazo [2, 1-b] [1, 3] thiazoles have been reported.

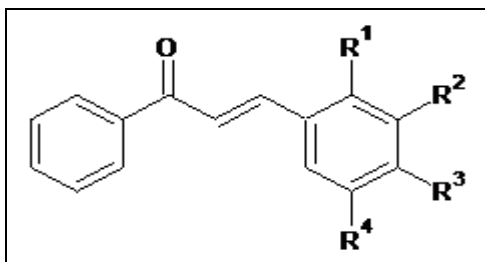


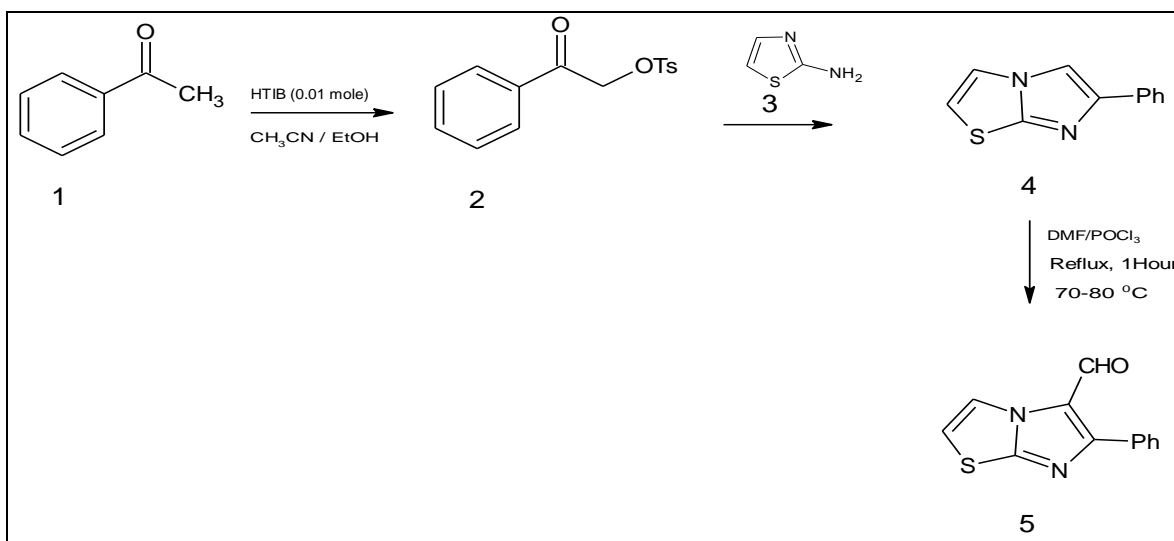
FIG. 1: STRUCTURE

This framework has been used as antifungal<sup>6</sup> anti-inflammatory, antimicrobial<sup>7-8</sup>, antitumor<sup>9-13</sup>, Imidazo [2, 1-b] [1, 3] thiazoles act as inhibitors of mitochondrial NADH dehydrogenase as specific inhibitors of the NADH-ubiquinone reductase

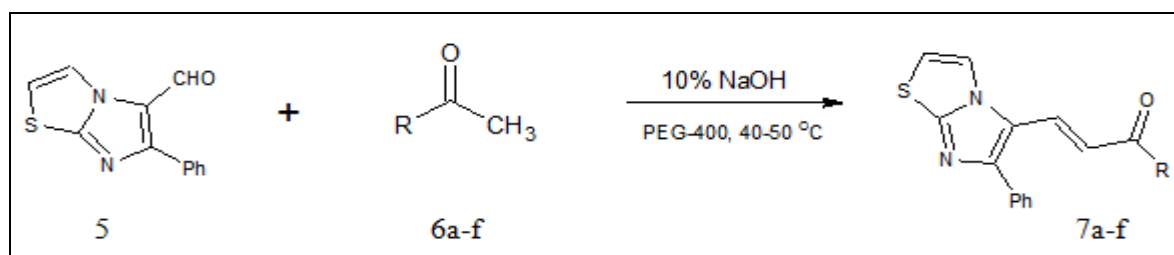
action in mitochondrial membranes, and antioxidant activities<sup>14-16</sup> among others. There are two aromatic rings are in unsaturation coupled together by a three-carbon  $\alpha$ ,  $\beta$ -unsaturated carbonyl system (Structure, Fig. 1).

Polyethylene glycol (PEG) has been well-known as a green solvent and is known to be reasonable, simply available, thermo logically secure, recyclable, non-hazardous and water-soluble compounds<sup>17-19</sup> reusable reaction medium and cost-effective to the catalytic reaction capable of recycling both solvent and catalyst easily. The structures of the compounds were supported by elemental and spectral data. Considering, these reports, we have synthesized and characterized new series of condensed Imidazo [2, 1-b] [1, 3] thiazoles Chalcones (7a-j) and evaluated pharmacology antioxidant (DPPH and SOD) activities by using PEG-400 as an alternative reaction medium. Among the synthesized compounds, most of the compounds were found to be potent inhibitors than with potential antioxidant activity.

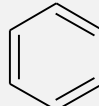
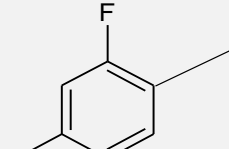
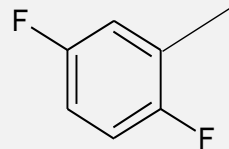
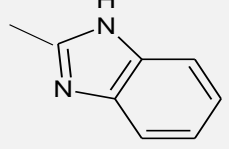
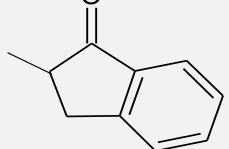
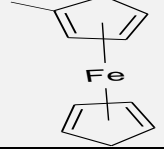
## MATERIALS AND METHODS:

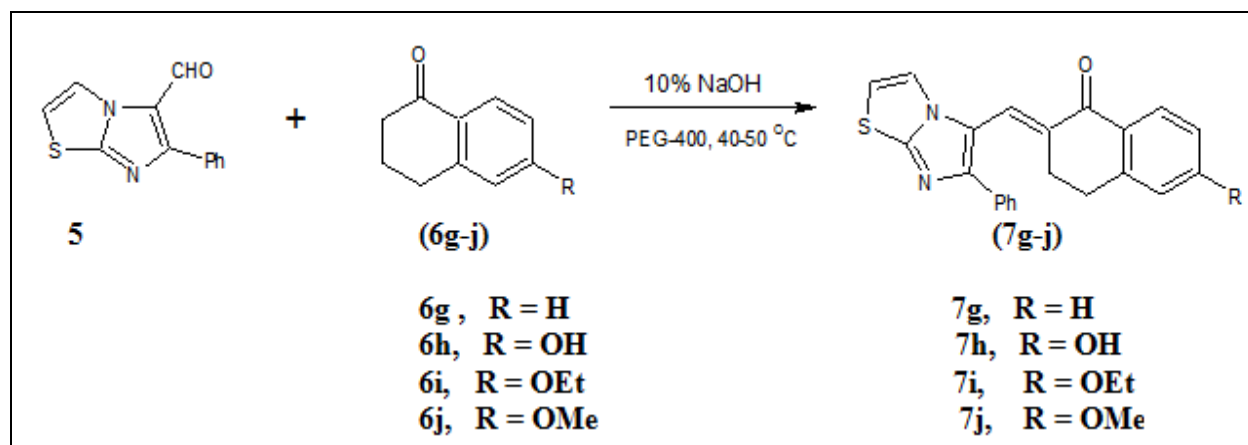


SCHEME 1: SYNTHESIS OF 6-Aryl Imidazo [2,1- b] thiazole aldehyde 5 from 6-Aryl Imidazo [2,1- b] thiazole 4



SCHEME 2: SYNTHESIS OF IMIDATHIAZOLE CHALCONE USING SUBSTITUTED ACETOPHENONE

Acetyl Ketone	R	Product
6a		7a
6b		7b
6c		7c
6d		7d
6e		7e
6f		7f



SCHEME 3: SYNTHESIS OF IMIDAZOTHIAZOLECHALCONE USING SUBSTITUTED TETRALONES

**General Instrumentation:** Melting points of the synthesized compounds were determined by using a digital thermometer and were found to be uncorrected. IR spectra were recorded on FT-IR spectrometer (Perkin Elmer, Maharashtra, India) using KBr disk method.  $^1\text{H}$ NMR spectra were recorded on  $^1\text{H}$ NMR (Varian-NMR-mercury 300 MHz) spectrometer in  $\text{CDCl}_3$  as a solvent. All chemical shifts ( $\delta$ ) are quoted in parts per million

downfield from TMS and coupling constants (J) are given in Hertz. Abbreviations used in the splitting pattern were as follows: s = singlet, d = doublet, t = triplet, q = quintet and m = multiplet. All the reagents and solvents were used of analytical grade and used as supplied unless otherwise stated. Thin layer chromatography was performed on silica gel coated plates for monitoring the reactions. The spots could be visualized easily under UV light.

**Scheme 1: General Method for the Synthesis of 6-Aryl Imidazo [2,1- b]thiazole 4 from acetophenone 1: One-Pot Synthesis:** Synthesis of Bridgehead Heterocycles and  $\alpha$ - tosyloxyketone (2) were synthesized by using Iodine according to the literature procedure<sup>21</sup>.

A mixture of acetophenone 1 (5mmol) and HTIB (5 mmol) in CH<sub>3</sub>CN (20 ml) was condensed for 2 h. The formation of  $\alpha$ - tosyloxyketone (2) was confirmed by TLC monitoring of the reaction mixture. Excess of acetonitrile was distilled off. Then a solution of 2 aminothiazole (3) (5 mmol) in ethanol (20 ml) was added dropwise. The resultant reaction mixture was refluxed until the completion of the reaction. The reaction was monitored by TLC. The solid obtained after cooling the reaction mixture was filtered, neutralized with saturated aq. Sodium bicarbonate solution, and washed with water to give 6-Aryl Imidazo [2,1- b] thiazole (4). MP 144 °C (lit. mp 144-145 °C); IR (cm<sup>-1</sup>): 3135 (aromatic C-H), 1619 (C=C and C=N), 1513 (C-N); 1H NMR (CDCl<sub>3</sub>): 6.82–6.83 (d, 1H), 7.40-7.42 (d, 1H), 7.80 (s, 1H).

**General Procedure for the Synthesis of aldehydes 5 from 4:** Synthesis of 6-Aryl Imidazo [2,1- b] thiazole aldehyde 5 from 6-Aryl Imidazo [2,1- b] thiazole by Vilsmeier- Haak reaction according to the literature method<sup>13</sup> which in turn were prepared from the appropriate 2- aminothiazole and  $\alpha$ - tosyloxyketone (2) as shown in Scheme 1. The Vilsmeier reagent was prepared at 0-5 °C by dropping POCl<sub>3</sub> (20 mmol) into a stirred solution of DMF (25 mmol) in CHCl<sub>3</sub> (5 mL). Compound 4 (10 mmol) in CHCl<sub>3</sub> (60 mL)

was added dropwise to the Vilsmeier reagent while maintaining stirring and cooling. The reaction mixture was kept at room temperature for 3 h and under reflux for 2 h. CHCl<sub>3</sub> was removed under reduced pressure, and the resulting oil was poured into ice. The crude aldehyde 5 thus obtained was collected by filtration and crystallized from ethanol with a yield of 70-80%.

**6 phenyl imidazo[2,1-b][1,3- thiazole-5- carbaldehyde] 5:** 80% yield, R<sub>f</sub> value is 0.28 (EtOAc/Hexane, 3:7), <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>):  $\delta$  9.61 (s, 1H),  $\delta$  5.1 (d, 1H),  $\delta$  6.4 (d, 1H),  $\delta$  7.48 (d, 2H),  $\delta$  7.32 (m, 2H),  $\delta$  7.22 (t, 1H),

**General Procedure for the Synthesis of Chalcones (7a-j):** The different imidazothiazole-chalcone derivatives (7a-j) were prepared by the Claisen-Schmidt condensation by using Imidazo [2,1-b]thiadazole aldehydes and appropriate different substituted acetyl ketones (6a-f) and (6g-j) according to literature method<sup>13,17-19</sup> in the presence of NaOH (10%) as shown in **Scheme 2**.

Imidazo [2,1-b]thiadazole aldehydes (0.01 mole) and appropriate acetyl ketones (0.01 mole) was dissolved in 15 mL PEG-400. To this mixture, NaOH (10%, 2 mL) was added, and the reaction mixture was stirred at 40-50 °C temperature for 2-4 h. After completion reaction (monitored by TLC) the reaction mixture was then poured into 100 mL ice-cold water and neutralized with dil. HCl solution to get solid. The product was purified from silica column using hexane: ethyl acetate as solvent out; it was filtered and processed out to afford pure compounds 7a-j.

S. no.	Structure	Product	Derivatives of Chalcones		M.P. °C
			M.F	M.W	
1		7a	C <sub>20</sub> H <sub>14</sub> ON <sub>2</sub> S	330	150
2		7b	C <sub>20</sub> H <sub>12</sub> ON <sub>2</sub> SF <sub>2</sub>	366	94
3		7c	C <sub>20</sub> H <sub>12</sub> ON <sub>2</sub> SF <sub>2</sub>	366	250

4		7d	C <sub>22</sub> H <sub>18</sub> ON <sub>2</sub> S	356	157
5		7e	C <sub>23</sub> H <sub>17</sub> O <sub>2</sub> N <sub>2</sub> S	360	140
6		7f	C <sub>23</sub> H <sub>20</sub> O <sub>2</sub> N <sub>2</sub> S	388	254
7		7g	C <sub>22</sub> H <sub>19</sub> O <sub>2</sub> N <sub>2</sub> S	374	--
8		7h	C <sub>21</sub> H <sub>15</sub> ON <sub>4</sub> S	371	254
9		7i	C <sub>21</sub> H <sub>14</sub> ON <sub>2</sub> S	330	178
10		7j	C <sub>24</sub> H <sub>20</sub> ON <sub>2</sub> SFe	384	118

### The Spectral Data of Synthesized Compounds 7(a-j):

**(2E)- 1- phenyl- 3- (6-phenylimidazo[2,1-b][1,3] thiazol-5-yl)prop-2-en-1-one (7a):** <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>): δ 7.81 (d, 2H), δ 7.45 (m, 1H), δ 7.54 (d, 1H), δ 7.90 (d, J= 15.5 Hz, 1H), δ 7.56 (d, J= 15.5 Hz, 1H), δ 7.48 (d, 2H), δ 7.32 (m, 2H), δ 7.22 (d, 1H), δ 7.41 (d, 1H), δ 7.98 (d, 1H).

**(2E)- 1- (2,4-difluorophenyl)-3-(6-phenylimidazo [2,1-b][1,3]thiazol-5-yl)prop-2-en-1-one (7b):** <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>): δ 6.87 (s, 1H), δ 6.93 (d, J= 8.0 Hz, 1H), δ 7.77 (d, J= 8.0 Hz, 1H), δ 7.92 (d, J= 15.8 Hz, 1H), δ 7.56 (d, J= 15.8 Hz, 1H), δ 7.48 (d, 2H), δ 7.32 (m, 2H), δ 7.22 (d, 1H), δ 7.41 (d, 1H), δ 7.98 (d, 1H).

**(2E)- 1- (2,5-difluorophenyl)-3-(6-phenylimidazo [2,1-b][1,3]thiazol-5-yl)prop-2-en-1-one (7c):** <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>): δ 7.14 (d, J=8.0 Hz, 1H), δ 7.23 (d, J=8.0 Hz, 1H), δ 7.50 (s, 1H), δ 7.90 (d, J= 15.5 Hz, 1H), δ 7.56 (d, J= 15.5 Hz, 1H), δ 7.48 (d, 2H), δ 7.32 (m, 2H), δ 7.22 (d, 1H), δ 7.41 (d, 1H), δ 7.98 (d, 1H).

**(2E)- 2- [(6-phenylimidazo [2,1-b][1,3] thiazol-5-yl)methylidene]-3, 4-dihydronaphthalen- 1(2H)-one (7d):** <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>): δ 7.49 (t, 1H), δ 7.27 (t, 1H), δ 7.76 (d, 1H), δ 7.31 (d, 1H), δ 2.29 (t, 2H), δ 2.59 (t, 2H), δ 7.62 (s, 1H), δ 7.48 (d, 2H), δ 7.32 (m, 2H), δ 7.22 (d, 1H), δ 7.41 (d, 1H), δ 7.98 (d, 1H).

**(2E)- 6- hydroxy-2-[(6-phenylimidazo[2,1-b][1,3] thiazol- 5- yl) methylidene]- 3, 4- dihydro naphthalen-1(2H)-one (7e):** <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>): δ 5.0 (s, 1H), δ 6.78 (s, 1H), δ 2.59 (t, 2H), δ 6.74 (d, J= 8.2 Hz, 1H), δ 7.59 (d, J= 8.2 Hz, 1H), δ 2.29 (t, 2H), δ 7.62 (s, 1H), δ 7.48 (d, 2H), δ 7.32 (m, 2H), δ 7.22 (d, 1H), δ 7.41 (d, 1H), δ 7.98 (d, 1H).

**(2E)- 6- ethoxy- 2- [(6-phenylimidazo [2,1-b][1,3] thiazol- 5- yl) methylidene]- 3, 4- dihydro naphthalen-1(2H)-one (7f):** <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>): δ 1.33 (t, 3H), δ 3.98 (q, 2H), δ 6.82 (s, 1H), δ 2.59 (t, 2H), δ 6.78 (d, J= 8.2 Hz, 1H), δ 7.65 (d, J= 8.2 Hz, 1H), δ 2.29 (t, 2H), δ 7.62 (s,

1H),  $\delta$  7.48 (d, 2H),  $\delta$  7.32 (m, 2H),  $\delta$  7.22 (d, 1H),  $\delta$  7.41 (d, 1H),  $\delta$  7.98 (d, 1H).

**(2E)-6-methoxy-2-[(6-phenylimidazo[2,1-b][1,3]thiazol-5-yl)methylidene]-3,4-dihydro-naphthalen-1(2H)-one (7g):**  $^1\text{H}$  NMR (300 MHz,  $\text{CDCl}_3$ ):  $\delta$  3.73 (s, 3H),  $\delta$  3.98 (q, 2H),  $\delta$  6.82 (s, 1H),  $\delta$  2.59 (t, 2H),  $\delta$  6.78 (d,  $J=8.2$  Hz, 1H),  $\delta$  7.65 (d,  $J=8.2$  Hz, 1H),  $\delta$  2.29 (t, 2H),  $\delta$  7.62 (s, 1H),  $\delta$  7.48 (d, 2H),  $\delta$  7.32 (m, 2H),  $\delta$  7.22 (d, 1H),  $\delta$  7.41 (d, 1H),  $\delta$  7.98 (d, 1H).

**(2E)-1-(1H-benzimidazol-2-yl)-3-(6-phenylimidazo[2,1-b][1,3]thiazol-5-yl)prop-2-en-1-one (7h):**  $^1\text{H}$  NMR (300 MHz,  $\text{CDCl}_3$ ):  $\delta$  5.0 (s, 1H),  $\delta$  6.67 (d,  $J=15.5$  Hz, 1H),  $\delta$  7. (d,  $J=15.5$  Hz, 1H), (d,  $J=8$  Hz, 2H),  $\delta$  7.26 (d,  $J=8$  Hz, 2H),  $\delta$  7.48 (d, 2H),  $\delta$  7.32 (m, 2H),  $\delta$  7.22 (d, 1H),  $\delta$  7.41 (d, 1H),  $\delta$  7.98 (d, 1H).

**(2E)-2-[(6-phenylimidazo[2,1-b][1,3]thiazol-5-yl)methylidene]-2,3-dihydro-1H-inden-1-one (7i):**  $^1\text{H}$  NMR (300 MHz,  $\text{CDCl}_3$ ):  $\delta$  7.69 (d, 1H),  $\delta$  7.26 (t, 1H),  $\delta$  7.42 (t, 1H),  $\delta$  7.25 (d, 1H),  $\delta$  3.22 (s, 2H),  $\delta$  7.58 (s, 1H),  $\delta$  7.48 (d, 2H),  $\delta$  7.32 (m, 2H),  $\delta$  7.22 (d, 1H),  $\delta$  7.41 (d, 1H),  $\delta$  7.98 (d, 1H).

### Biological Study:

#### Antioxidant Activity of Chalcones:

**DPPH Radical Scavenging Activity:** The molecule 1, 1-diphenyl-2-picrylhydrazyl (a, a-diphenyl-bipicrylhydrazyl; DPPH) is characterized as a stable free radical by virtue of the delocalization of the spare electron over the molecule as a whole, so that the molecule does not dimerize, as would be the case with most other free radicals. The delocalization of electron also gives rise to the deep violet color, characterized by an absorption band in ethanol solution centered at about 517 nm. When a solution of DPPH is mixed with that of a substrate (AH) that can donate a hydrogen atom, then this gives rise to the reduced form with the loss of this violet color. The ability of compounds to scavenge.

**Procedure:** DPPH radical was assessed using Ramanathan Sambath Kumar *et al.*, and Manzocco *et al.*, 1998 method with modification. Briefly, 1 ml of synthesized compounds as 1 mM was mixed with 3.0 mL DPPH (0.5 mmol/L in methanol), the resultant absorbance was recorded at 517 nm after 30 min. incubation at 37 °C. The percentage of

scavenging activity was derived using the following formula.

Percentage of inhibition (%) = [(A control - A sample) / A control]  $\times$  100

Where A control - absorbance of DPPH; A sample - absorbance reaction mixture (DPPH with Sample).

**Scavenging of Superoxide Radical by Alkaline DMSO Method (Kunchandy E, 1990):** To the reaction mixture containing 1 mL of alkaline DMSO, 0.3 mL of the drug samples and standard was added in DMSO at various concentrations followed by 0.1 mL of NBT (0.1 mg) to give a final volume of 1.4 mL. The absorbance was measured at 560 nm.

**RESULTS AND DISCUSSION:** In the present study, the synthesis of title compounds Imidazothiazole chalcone (7a-j) by Claisen Schmidt condensation has been carried out successfully by using selected acetyl ketones (6a-j) and Imida-thiazole aldehydes according to literature methods **Scheme 1**. TLC and melting point recognized the purity of the newly synthesized compounds. The characterization of all the listed synthesized Chalcones was made by melting point, IR, NMR spectral analysis. Chalcones showed the IR absorptions characteristics of carbonyl  $>\text{C}=\text{O}$  ( $1685\text{-}1600\text{ cm}^{-1}$ ) and aromatic  $\text{C}=\text{C}$  ( $1580\text{-}1400\text{ cm}^{-1}$ ) functionalities. The  $^1\text{H}$  NMR spectra of chalcones displayed multiplet due to aromatic protons at 6.92-8.00  $\delta$  (m, Ar-H). Particulars of the methods and conditions are described in the experimental section.

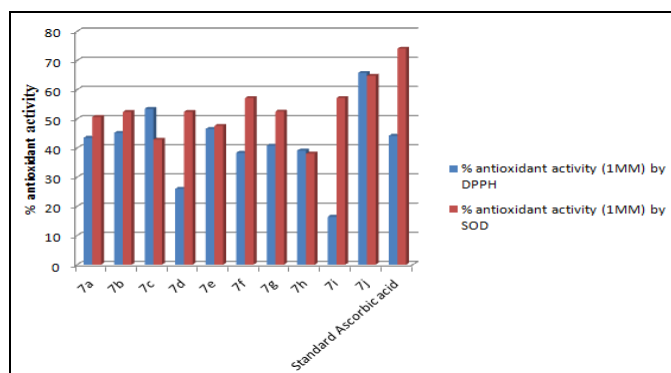
**Antioxidant Activity:** All the synthesized compounds were assessed to achieve the promising and selective inhibition is concerned, antioxidant reactivity by monitoring the ability of the compounds to reduce 2,2-diphenylpicrylhydrazyl (DPPH), a commonly used radical scavenger. A comparative assessment of these activities is shown in **Table 1**. The data are given in **Table 1** indicate that compounds were having electron withdrawing benzene ring with substituent hydroxyl, fluoro, methoxy and ethoxy on B ring as proposed for compounds (7a-j) gave very good antioxidant activity. Particularly in compounds like 7e, 7f, 7g, and 7h showed the moderate percentage, and 7c

shows the strongest percentage of inhibition mainly at (C=1mM). Almost all the compounds indicate moderate to good antioxidant activity compared to that of standard ascorbic acid percentage antioxidant activity (1MM) by DPPH and SOD as 44.18 and 74.07 respectively. The data indicate that the compounds with good activity in case of electron-withdrawing para fluoro substituted chalcone 7c instead of electron-withdrawing meta fluoro substituted in chalcone 7b. Importantly, the

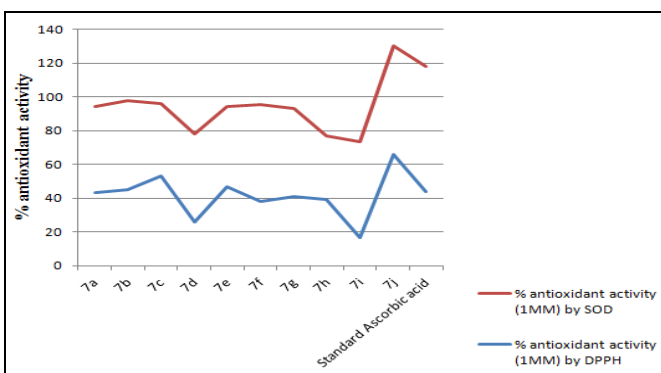
level of activity of these compounds is very close to the level of ascorbic acid, particularly at low concentrations. In contrast, compounds 7d, showing optimum while, 7i showing minimum and 7j though having pseudoaromatic character showing the strongest percentage of inhibition compared with ascorbic acid. This behavior, however, is consistent with that reported by Murti et al.<sup>23</sup>

**TABLE 1: ANTIOXIDANT ACTIVITY OF DIFFERENT SUBSTITUTED CHALCONE DERIVATIVES**

S. no.	Sample Number	% antioxidant activity (1MM) by DPPH	% antioxidant activity (1MM) by SOD
1	7a	43.51	50.65
2	7b	45.20	52.38
3	7c	53.42	42.85
4	7d	26.02	52.38
5	7e	46.57	47.61
6	7f	38.35	57.14
7	7g	40.79	52.51
8	7h	39.12	38.09
9	7i	16.43	57.14
10	7j	65.75	94.76
11	Standard Ascorbic acid	44.18	74.07



**FIG. 2: COMPARISON BETWEEN % ANTIOXIDANT ACTIVITY OF DPPH AND SOD**



**FIG. 3: COMPARISON BETWEEN % ANTIOXIDANT ACTIVITY OF DPPH AND SOD**

**CONCLUSION:** The main focus of this research work was to synthesize novel series of Imidazothiazole chalcones, purify, characterize and evaluate their antioxidant activity. The yield of the synthesized compound was found to be in the appropriate range. Structure of synthesized compounds was confirmed and characterized with the help of analytical data such as FTIR, <sup>1</sup>HNMR. Almost all the synthesized novel Imidazo thiazole chalcones show potent antioxidant activity. The activity is enhanced due to the presence of hydroxyl, methoxy, ethoxy and fluoro substitution as well as benzimidazole and ferrocenyl framework in chalcone structure. It was also found to be a potent scavenger of SOD, and this property may be responsible for the good anti-inflammatory activity

of corresponding imidazothiazole chalcones. From the results, it can be concluded that the modified chalcones show significant biological evaluation as antioxidant agents. However, further evaluation of Imidazothiazole chalcones will be undertaken, concerning the structural arrangements on B ring for antioxidant activity.

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