INVESTIGATIONS ON ANTIOXIDANT PROPERTIES OF SEMICARBAZONE BASED SCHIFF BASES OF 2-ANILINO-3 FORMYLCHROMONECHROMONE DERIVATIVES

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INTRODUCTION

Chromones are a class of substances with benzopyran ring **(Figure 1)** Many of the flavonoids, including flavones, isoflavones, and flavanols, have the chromone, or 1-benzopyran-4-one, ring system as a key component.

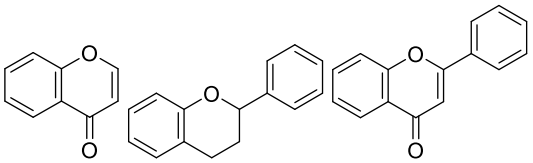
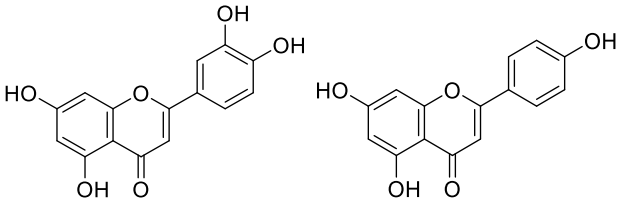


Figure 1Flavonoid (2) Flavone (3)

The biological activities shown by chromones[1] include antioxidants[2-4], anti-allergic[5], anti-hypertensive[6], anti-HIV[7,8], antimicrobial[9], antifungal[10], anticancer[11,12], antiviral[13], anti-inflammatory[14], antimalarial[15], antigiardial[16], cytotoxic[17], protein tyrosine kinase inihibitor[18], phosphoinositide-3-kinase inhibitor[19], anti-erectile-dysfunction[20], mono-amine-oxidase inhibitor[21], anti-inflammatory[22], adenosine receptor antagonists[23], antiplatelet[24], selective estrogen receptor modulator[25], anti-tumor/anti-proliferative[26], anti-psychotic[27], neuroprotective[28] and P-glycoprotein modulator[29]. These properties can also be produced using chromone derivatives, which motivates pharmaceutical and organic chemists to create chromone and its derivatives.[30]

Flavones with different replacement designs have been separated from verdant vegetables and herbs.[31] Flavones can cooperate with film bilayers since they contain both lipophilic (non-polar) and hydrophilic (polar) components.[32] This communication involves (a) the parcelling of the non-polar fragment of the particle between the water repelling inside of the lipid bilayers and (b) Production of hydrogen connections between the polar end gatherings of the lipids and the water attracting segments of flavones at the layer junction. The pace of film lipid and protein oxidation can be affected by the enlistment of changes in the actual attributes of layers. Moreover, flavones in the hydrophobic center may likewise prompt chain-breaking hostile to oxidant property, while cooperation between polyphenols at the bilayer surface through hydrogen bonds might restrict the entrance of possibly destructive particles, protecting the design and usefulness of films. It has been reported that there is a converse connection between the capacity of flavones to cause layer porousness by delivering calcein bound in liposomes and their maintenance in a phosphati-dylcholine-covered section.[33,34]



Luteolin (4)Apigenin(5)

Receptive oxygen species, or ROS, and responsive nitrogen species, or RNS, are two unique classifications of free radical that are tracked down in living frameworks. At the point when a free radical connects with a particle to kill itself, a second free radical is delivered. This sets off a chain response that delivers an enormous number of free radical, which thus causes infections related to maturing, for example, malignant growth, arteriosclerosis, coronary illness, maturing, safe framework decline, and mind dysfunction[35,36]. Majorly of the damage is caused by ROS species including lipid peroxidation and DNA damage in pneumoconiosis and carcinogenesis[37].Increased ROS affect various cardiovascular disorders as well as thrombus formation in arteries[38,39]. acute respiratory distress syndrome (ARDS) is characterised by pulmonary vascular lesions and inflammation, both of which are brought on by oxidative damage[40].

Previous studies on structure-activity relationship of flavonoids have shown the importance of the number and position of phenolic OH groups onto the chromone ring and ring stabilisation after a donation of Hydrogen atom to radicals. Compounds containing azomethine as functional group containing carbon, nitrogen double bond of which nnitrogen is attached to group other than hydrogen forms the non allylicbase class of compounds[41,42]. The substitution of electron withdrawing group at C-6 of chromone enhances the radical scavenging activity[43]. Antioxidant property of *flavonoids* is determined by the nature of substitutions on ring B and C. Position of OH group and degree of hydroxylation (3', 4', 5'-positions enhances the antioxidant) in the ring B enhances the antioxidant property of flavonoid. In ring C, the radical scavenging is enhanced by conjugation between the double bond between C_2-C_3 and 4-oxo group[44,45]. The structure activity relationship of flavonoids have highlighted the significance of the numerous electron donor and receptor sites that may directly affect the molecules' capacity to scavenge free radicals.The presence of various substituents in conjugation with C_2-$C_3\pi$-electrons, which may contribute to electron delocalization.

Substitution on the A-ring of chromone nucleus may cause a sudden change in its interaction with the free radicals accumulated in the reaction. Flavonoid derivatives act as radical scavengers by providing free radicals to the Hydrogen atom to stablise it.

In order to study radical scavenging behaviour, radical quenching mechanism was studied using DPPH-method where the entrapanent of radical species generated plays a role in the determination of antioxidant properties.

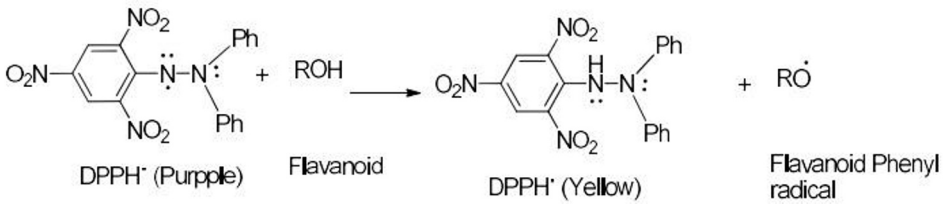


Fig. 1. Free radical scavenging of DPPH• by a radical scavenger (flavonoid)

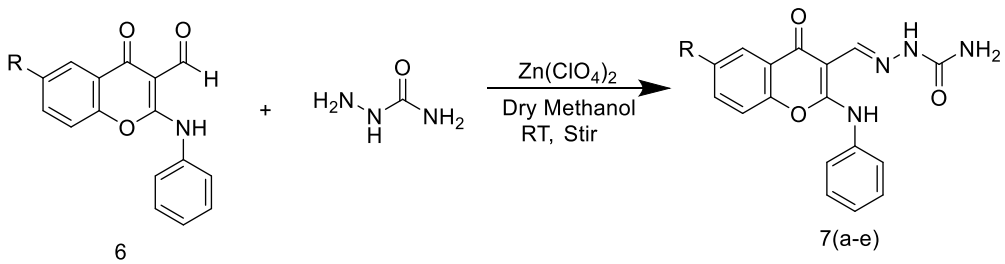
The effect of chromenederivatives on a purple 2,2′-diphenyl-1-picrylhydrazyl radical (DPPH) solution and observation of bleaching activity on the DPPH can be used to gauge their ability to donate electrons[47]. Flavonoids shows anti-oxidant (radical scavenging) property once added to the DPPH solution by decolourizing it. Considerable free radical scavenging activity of the sample is indicated by a considerable decrease in the absorbance of the reaction mixture[48].

MATERIALS AND METHODS

The aim of the research is to synthesize chromone derivatives and investigate their anti-oxidant properties. In order the test the radical scavenging capacity of compounds, semicarbazone was used in synthesis by using variously substituted chromones.[49]

Synthesis is done by mixing 2-anilino-3-formylchromone (1.0 eq) and semicarbazone (1.2 eq) in the presence of anhydrous methanol. The solution was stirred with continuous heating at 110°C till the completion of reaction (checked by TLC). The product so obtained was further tested for its free radical scavenging ability.[50]

The chromone derivatives so synthesised were then used to prepare the stock solutions of 100ppm using DMSO as the solvent and then further dilutions were done to change the concentration of the solutions.[51] Similar solutions of ascorbic acid were also prepared(standard). The general concentrations used are 10ppm, 20ppm, 40ppm, 60ppm and 80 ppm.



6

7(a-e)

7a R = H
7b R = CH_3
7c R = Cl
7d R = F
7e R = Br

0.1mM DPPH(2,2-diphenyl-1-picrylhydrazyl) was prepared of which 2 ml was added to each 10ml volumetric flask with different concentrations (10ppm, 20ppm, 40ppm, 60ppm, 80ppm). Then the reaction mixture was incubated for 30 minutes. Ascorbic acid was also prepared in a similar manner.[52] A change in the absorption behaviour of DPPH is determined in UV spectrum (UV-spectrum recorded on Schimadzu UV-1800S double beam spectrophotometer using glass cuvette). Radical scavenging capacity of the compounds is then calculated by the following formula :

DPPH Scavenging Activity(%) = $(A_{blank} – A_{sample})/A_{blank} * 100$

Where,

A_{blank}– Absorbance of the controlled reaction

A_{sample}– Absorbance in the presence of the compound under investigation

The change in absorption behaviour is observed with the change in concentration. As the concentration of the solutions increases, results get better.[53]

RESULTS AND DISCUSSION

The chromone derivatives synthesised show prominent radical scavenging ability and it is observed that as the concentration of the sample solution increases, Its radical scavenging % also increases.6- Flurochromene derivative have shown the best results, followed by 6-Chloro derivatives.Absorption studies were carried over as radical scavenging activities of DPPH at wave length of 470 - 575nm , where , Lembda max lies between 510-525nm. The decrease in the absorption behaviour of sample solutions on adding DPPH witnesses the radical scavenging activity. This study also shows the significant role of the substituents attached to the benzopyran ring. In case of the attachment of highly electronegative atoms, radical scavenging capacity is higher.

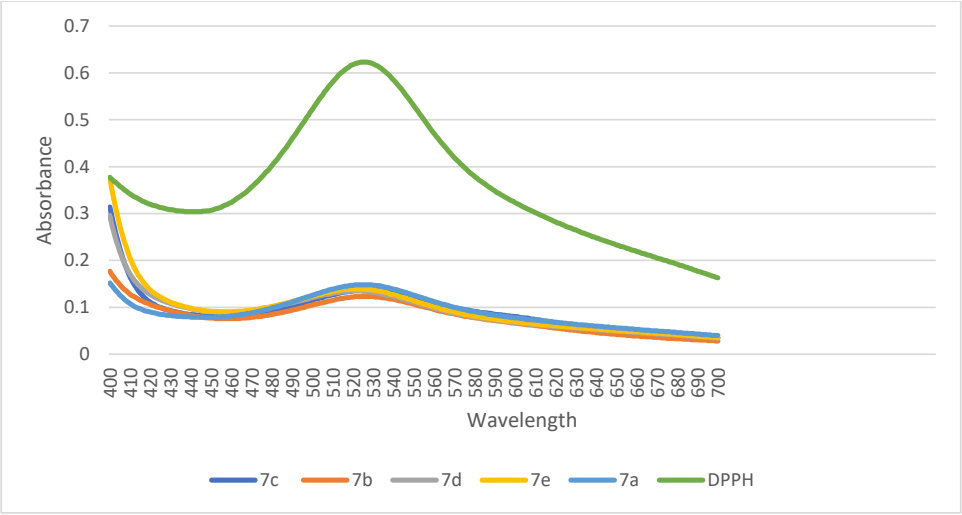


Fig 2. Absorbance variation on adding DPPH in Chromone derivatives (7a-e)

The absorption patterns of all the compounds were observed and compared to the standard ascorbic acid solution to determine the relative radical scavenging capacity. This revealed a very significant and comparable absorption pattern, indicating that chromone derivatives play the same type and function as ascorbic acid.

Entry	10ppm	20ppm	40ppm	60ppm	80ppm
7a	48.13	58.16	70.82	79.78	87.92
7b	51.25	57.83	74.06	82.34	91.38
7c	73.98	82.53	85.29	89.04	92.83
7d	81.27	89.78	88.53	92.46	95.76
7e	59.38	65.38	68.21	70.34	83.39
Ascorbic acid	69.30	75.25	81.12	87.34	88.79

Table 1. Radical scavenging percentage of chromone based semi-carbazones at 10, 20, 40, 60, 80 ppm concentrations

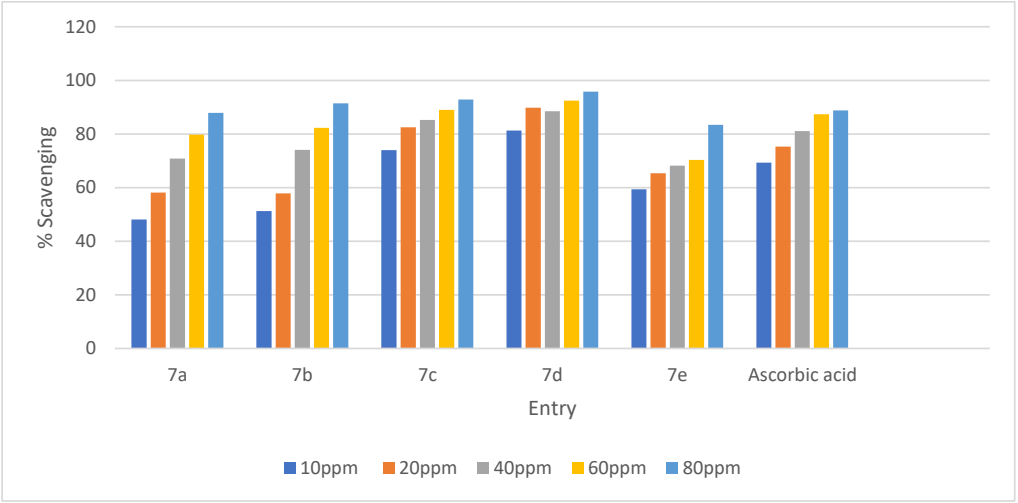


Fig 3. Relative absorption of chromone derivatives at different concentrations w.r.t. ascorbic acid

Here, the substituents with more electro negative atoms, such as fluorine and chlorine, have demonstrated similar radical scavenging activity, however the activity is reduced in the absence of electronegative group.

CONCLUSION

The dchiff base so designed have lead to an idea of radical scavenging activity possessed by chromone derivatives. From the result drawn, it has been observed that presence of group attached at C_6 position of chromone nuclei plays an important and significant role towards anti-oxidant activities. Derivatives with more electronegative atoms usually possess higher order of radical scavenging activity which can be attributed to electron delocalisation capacity of atom/group attached. Additionally, an alteration in the pie bonder conjugated system contributes to availability of free electrons to participate in radical scavenging activity. The present investigation may further be extended to determine the role /effect of groups attached silico methods and enzyme assay.

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