ANTIOXIDANT ACTIVITY OF CHROMENO-PYRIMIDINEFUSED HETEROCYCLES OBTAINED IN GREEN REACTION

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Abstract - Chromeno [2,3-d] pyrimidin derivatives were obtained insimple, one-pot, and low-cost reactions of barbituric acid with a variety of substituted salicylaldehides in lemon juice, in high yields. All synthetized compounds were characterized using spectroscopic techniques NMR and IR. The products were subjected to evaluation of their in vitro antioxidative activity using stable 2,2-diphenyl-1-picryl-hydrazyl (DPPH) radical. It was shown that all investigated compounds exhibited moderate scavenging activity (about 80 % at concentration of 100 μM), but better than starting barbituric acid.

Keywords - Barbituric acid, Salicylaldehydes, Lemon juice, Chromeno[2,3-d]pyrimidines, Antioxidant activity.

I. INTRODUCTION

Barbituric acid is important partof the many biological active pyrimidinecompounds, commonly known as barbiturates [1-4]. They possess numerous biological activities such as hypnotic [5,6], sedative [7], anticonvulsant [8,9], antimicrobial [10], anesthetic [11], anticancer and antitumor properties [12,13].

Similarly, chromeno derivatives display remarkable pharmaceutical effects including antifungal [14], anti-microbial [15], anticoagulant, spasmolytic, diuretic, and anticancer [16], Fig. 1.

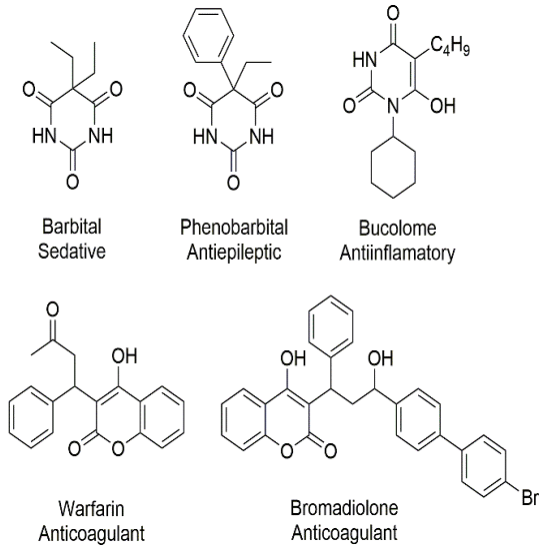


Fig. 1. Structures of some drugs which containpyrimidine or chromenomoiety.

The literature data revel that chrome no[2,3-d] pyramid in compound class exhibits a huge spectrum of pharmacological properties such as antitumor, cytotoxic [17], antioxidant [18], antithrombotic, anti-inflammatory [19], antimicrobial [20], and antidiabetic activities [21], Fig. 2.

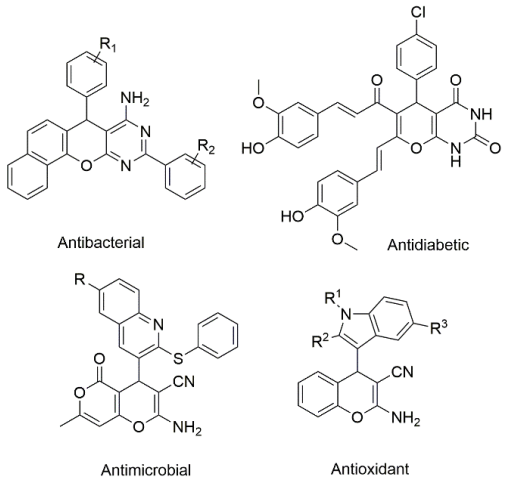


Fig. 2. Structures of some bioactive heterocycles compound which contain chromeno and pyrimidine moiety.

Therefore, synthesis of fused heterocyclic compounds which contain both moieties, chrome no and pyrimidine, is a big challenge by chemists. Few methods are known for the cascade synthesis of chrome no[2,3-d] pyrimidines from salicylaldehydes and barbituric acids.

In these reactions both, bases and acids, are usedas catalysts [22,23]. Synthesis of chrome no[2,3-d]pyramid in derivatives in boiling ethanol without catalyst also has been described [24]. These methods have limitations includinga low yield, a long reaction time, or using a more specific catalyst. An efficient formation of chromeno[2,3-d]pyramid in derivatives in water catalyzed by triethylbenzylammoium chloride is presented [25].

In this paper, we report a simple and eco-friendly protocol, where lemon juice was used as catalyst and solvent in one-pot synthesis of chrome no[2,3-d]pyramid in derivatives. In addition, the obtained products were studiedfor their in vitro antioxidative activity using DPPH radical.

II. DETAILS EXPERIMENTAL

2.1. Materials and Procedures

All the chemicals were procured from either Sigma-Aldrich Co. or Merck & Co. The IR spectra were recorded on a Perkin-Elmer Spectrum One FT-IR spectrometer using the KBr plates. The ^{1}H NMR and ^{13}C NMR spectra were run on a Varian Gemini spectrometer (200 MHz and 50 MHz for ^{1}H and ^{13}C respectively) using DMSO-d_6 as solvent. The UV-Vis measurements were performed on the UV/Vis Spectrophotometer, Agilent Technologies, Cary 300 Series. Lemon was purchased from local market.

Preparation of lemon juice Lemon was squeezed, and obtained juice was filtered through Büchner funnel with a sintered glass disc in order toremove pulp. The pH value of the filtrate was about 2. The obtained juice was then used in the reaction.

General procedure for the synthesis of chromeno[2,3-d]pyrimidin derivatives 3

A mixture of barbituric acid (2 mmol) and corresponding aldehyde (1 mmol) was stirred in lemon juice (3 ml) at 80 ℃, for one hour. Reaction progress was monitored using thin layer chromatography (TLC). The reaction product was precipitated during reaction. After completion of the reaction the mixture was filtered and washed with water. All obtained products were characterized without further purification.

The purity of isolated products was confirmed by^{1}HNMR, ^{13}CNMR and IR spectra. Herein, we present structural dates for one representative product.

5-(2,4-dioxo-1,3,4,5-tetrahydro-2H-chromeno[2,3-d]pyrimidin-5-yl)pyrimidine-2,4,6(1H,3H,5H)-trione (3a): Yellow powder ^{1}H NMR (200 MHz, DMSO) δ:11.98 (s, 1H), 11.30 (s, 1H), 11.18 (s, 1H), 10.99 (s, 1H), 7.40 – 7.28 (m, 1H), 7.27 – 7.04 (m, 3H), 4.71 (d, J = 2.4 Hz, 1H), 3.85 (d, J = 2.4 Hz, 1H);^{13}C NMR (50 MHz, DMSO) δ: 169.61, 168.96, 163.58, 155.49, 150.60, 149.62, 149.23, 129.32, 128.15, 125.68, 121.02, 116.58, 85.37, 53.50, 33.86; IR (cm^{-1}): 3485, 3278, 3222, 3038, 2860,1697, 1673, 1651, 1522, 1493, 1442, 1380, 1280, 1226, 1100, 895, 764, 617, 538;

DPPH free radical scavenging assay

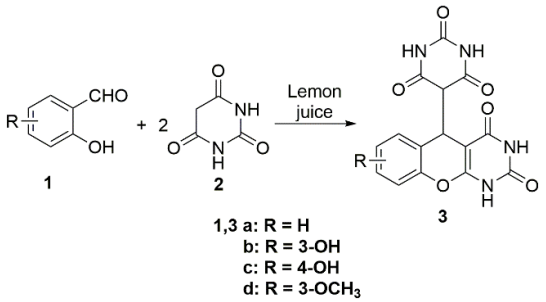
The free radical scavenging activity of the inspected compounds was achieved using the DPPH method, according to ref. [26]. DPPH solution (1 mL, 0.05 mM) in methanol was mixed with the tested compound (20 µL of compound solution in DMSO and 980 µL of methanol).

The reaction mixture was allowed to stand at room temperature for 20 and 60 min. After incubation the absorbance was determined spectro photometrically at 517 nm. As control solution, methanol was used. NDGA was used as positive control. All measurements were performed on three replicates.

III. RESULTS AND DISCUSSION

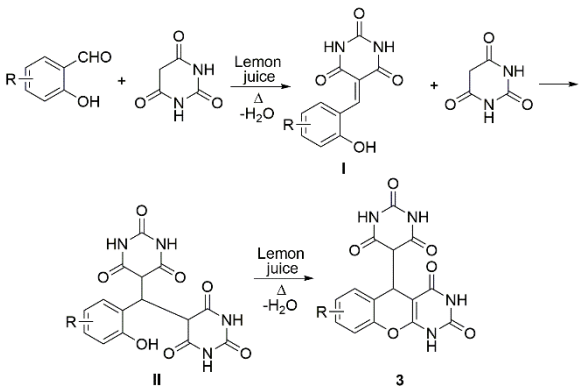
3.1. Chemistry

One-pot and pseudo three-component reactions of a salicylaldehyde or substituted salicylaldehydes (1, 1 equiv.)and barbituric acid(2, 2 equiv.)were performed in the presence of lemon juice. The reactions were performed by heating to 80 ℃ for one hour. In all reactions, chromeno[2,3-d]pyramid in derivatives3 were obtained in high yields (87-92 %),Scheme 1. It is worth pointing out that all products were pure, and no further purification was required.



Scheme 1.One pot synthesis of chromeno[2,3-d]pyrimidinderivatives

The proposed mechanism for the formation ofchrome no[2,3-d]pyramid in derivatives is presented in Scheme 2.The reaction starts with the Knoevenagel condensation of salicylaldehyde with barbituric acidyielding the intermediate I. Next step of the reaction is formation of the adduct II by Michael addition of the second molecule of barbituric acid to the arylidene intermediate I. Cyclization of the productII leads to the final chrome no[2,3-d]pyrimidine3.

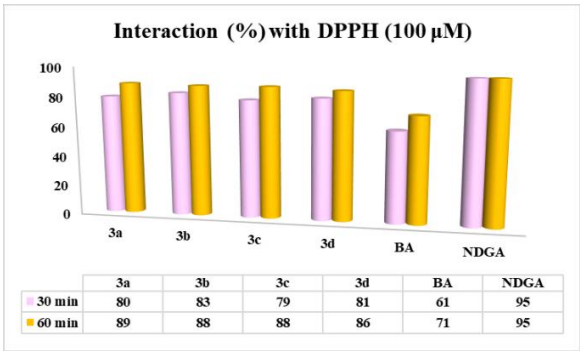


Scheme 2. The suggested mechanism for the considered reaction

3.2. DPPH radical scavenging activity

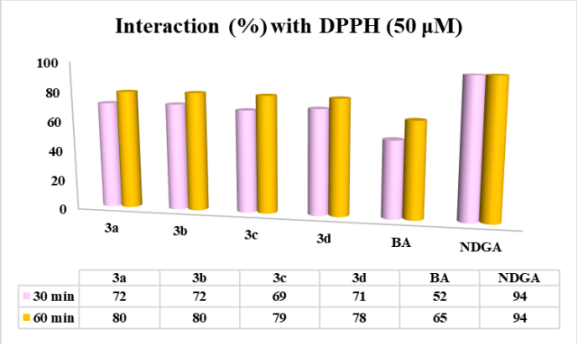
Synthetized chrome no[2,3-d]pyramid in derivatives3a-d, as well as barbituric acid, were tested for their in vitro antioxidative potential against the 1,1-diphenyl-2-picryl-hydrazyl (DPPH) stable free

radical. Antioxidant activity of these compounds was testedat concentrations (100 µM, 50 µM, and 25 µM) and for incubation period of 30 and 60 minutes. The obtained results reveal that compounds3a-d exhibited scavenging activities of about 80 % and 70 % at concentrations of100 µM and 50 µM. Compounds 3a-c evince activity of about 60 % at concentration of 25 µM, while compound 3d about 50 %. It is worth point out, that all compounds express better activity than starting compound barbituric acid, and that antioxidant capacity of these compounds is increasing(5-10 %) during prolongation of the incubation period to 60 minutes, Figs. 3-5.



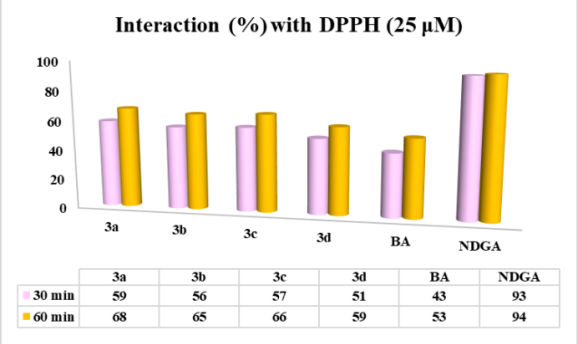
	3a	3b	3c	3d	BA	NDGA
30 min	80	83	79	81	61	95
60 min	89	88	88	86	71	95

Fig. 3. DPPH radical scavenging activity at compounds concentration of 100 µM



	3a	3b	3c	3d	BA	NDGA
30 min	72	72	69	71	52	94
60 min	80	80	79	78	65	94

Fig. 4. DPPH radical scavenging activity at compounds concentration of 50 µM.



	3a	3b	3c	3d	BA	NDGA
30 min	59	56	57	51	43	93
60 min	68	65	66	59	53	94

Fig. 5. DPPH radical scavenging activity at compounds concentration of 25µM.

CONCLUSIONS

A new efficient, lemon juice catalysed, synthesis of chromeno[2,3-d]pyrimidin derivatives3a-d has been performed. The products were obtained in excellent yields and characterized using NMR and IR spectroscopy. These compounds were screened for their DPPH radical scavenging potential. The obtained results reveal that compounds 3a-d exhibited moderate scavenging activity, but better than starting compound barbituric acid.

ACKNOWLEDGMENTS

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