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Pharmaceutical Sciences **Research Article……!!!**

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SYNTHESIS AND BIOLOGICAL EVALUATION OF SOME NOVEL N-SUBSTITUTED PHTHALAZINE AND PYRIDAZINE DERIVATIVES

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Keywords:

Synthesis, Anti-cancer activity, Anti convulsant activity, Anti microbial activity, Pthalazine and pyridazine Derivatives

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ABSTRACT

A new series of pthalazine and pyridazine derivatives were prepared .The structure of the synthesized compounds were confirmed by IR and 1HNMR spectral data.To investigate the possible Anti-cancer activity, Anti convulsant activity and anti microbial activity of Pthalazine and pyridazine Derivatives. Their antibacterial activity against *Pseudomonas aeruginosa* (MTCC96), *Escherchia coli* (MTCC722) were compared to standard on Amikacin. Anti fungal activity of compounds against *Candida albicans* (MTCC227) compared to standard Ketconazole (30µg/disc). In the DLA tumor control group, the average life span of animal was found to be 46% where as various synthetic drugs at the dose of 10 mg/kg body weight increase the life span to 70%,67%,71%,72% and 65% respectively. These values were significant. However the average life span of 5- FU treatment was found to be 90%, indicating its potent antitumor nature. The table values reveals that after treatment with various synthetic compounds, the compounds such as Ia, Ib, Ia, IIb. IIIa, IIIb. IVa, IVb, VIa, VIb, XIIIa & XIIIb possess significant Anti convulsant effects. All above said cmpounds were reduced the duration of extensor phase significantly. The anti microbial studies reveal that the compounds like Ia, IIIa, Vb and VIa shown to possess good activity against gram (-)ve micro organism. Similarly synthetic compounds like IIIa , VIa and VIb shown good activity against gram (+) microorganism. It also possess excellent activity against *Candida albicans* reveals that the synthesized compounds like XIVa and XI possess antifungal activity.

INTRODUCTION

The past studies on the parent moieties ,ie Phthalazines [1,2]and Pyridazines[3,4] shown compounds derived from it exhibits various activities like anticonvulsant , anti platelet aggregatory[5], anti-tumour[6], antitubercular[7], antimicrobial[8], antiviral, etc. The compounds phthalazines and pyridazines are particularly in interest of showing activity like anti-tumour , anticonvulsant and vasodilatory effect. Antibiotics are among the most prescribed drug in the world today, and since their development and commercialization, have saved countless millions of lives. The ideal antimicrobial agents are selective in only targeting the microorganism but not host cells. Resistance to antimicrobial agents is now recognized as a major global public health problem. In addition, because of the increased number of immunocompromised patients (AIDS. Cancer and transplants), primary and opportunistic fungal infections continue to increase rapidly, and as consequence, invasive fungal infections constitute a major cause of mortality for these patients. Although there are a new classes of compounds that are now frequently used to treat fungal infections, the frequency of deeply invasive microbial agents has increased 10 fold during the past decade. Moreover, many infections are actually refractory to antimicrobial therapy. With the emergence of new bacterial strain resistant to many currently available antibiotic treatment, there is increasing interest in the discovery of novel antibacterial agents. Certain small heterocyclic molecules are known as pharmacophores of a number of biologically active and medicinally useful molecules.

EXPERIMENTAL

The purity of the synthesized compounds were ascertained by thin layer chromatography on silica gel G in various solvent systems using iodine vapors a detecting agent. Melting points were determined by Toshniwal Melting Point Apparatus, Boiling point Determination in open capillary tubes and are uncorrected. Infra-red spectra were recorded on Shimadzu 8000FTIR Spectrometer in KBr phase. Proton NMR spectra were recorded in $CDCl_3$ on Brucker Avance DRX-300 FT-NMR spectrometer using tetra methyl silane as internal standard[9,10]. Table 1, 2

Scheme-I: (synthesis of some N-substituted Phthalazine and pyridazine derivatives.)

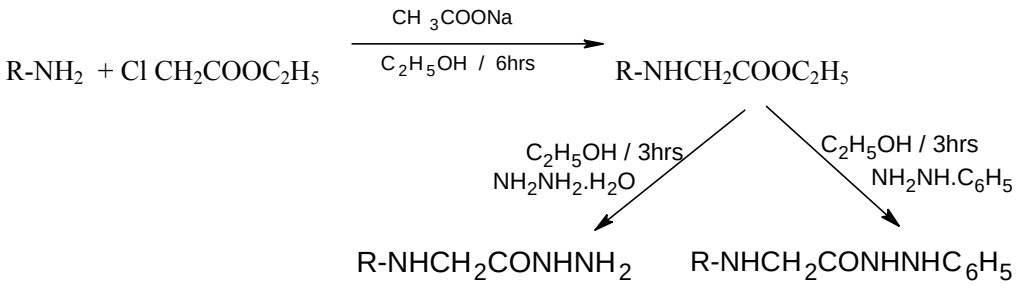
Synthesis of N-substituted hydrazides.

A mixture of substituted anline (0.1 mol), ethyl chloroacetate (0.12 mol), anhydrous sodium acetate (0.15 mol), in 50ml absolute ethanol was refluxed for 6 hrs. The mixture was cooled and left overnight at room temperature. It was poured into 150ml cold water. The solid **ethyl N-aryl glycinate** obtained was washed with cold water. Dried and divided into two portions.

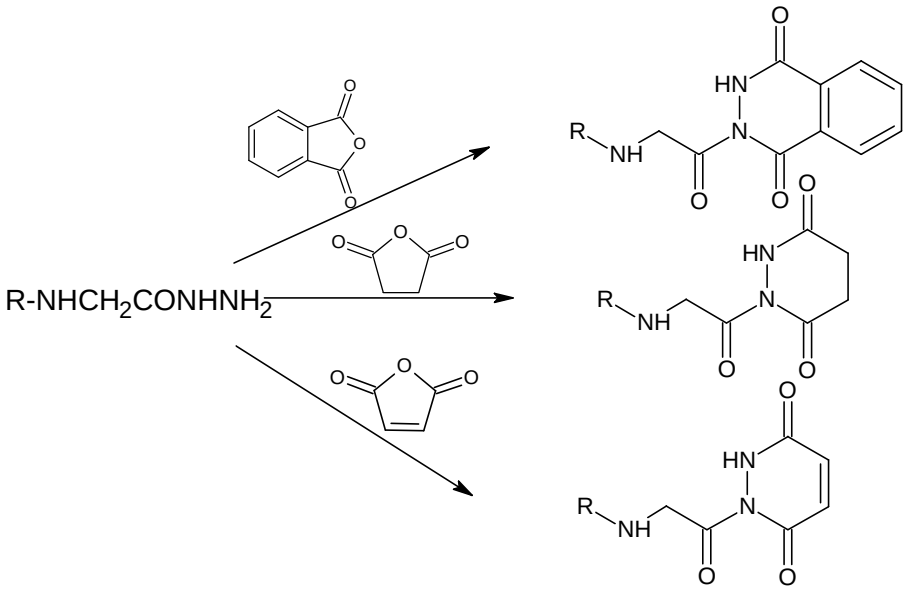
To a portion, hydrazine hydrate (0.1 mol) was added drop wise with constant stirring the mixture was gently refluxed for 15 minutes. Then about 25ml absolute ethanol was added to produce a clear solution. The reaction mixture was refluxed for 3 hrs. The ethanol was distilled of under reduced pressure, cooled. The resulting solid was recrystallised from ethanol

To other portion of ethyl N-aryl glycinate, added phenyl hydrazine (0.1 mol) drop wise with constant stirring the mixture was gently refluxed for 15 minutes. Then about 25ml absolute ethanol was added to produce a clear solution. The reaction mixture was refluxed for 3 hrs. The ethanol was distilled of under reduced pressure, cooled. The resulting solid was recrystallised from ethanol.

Step I- Synthesis of N-substituted Hydrazides.



Step II- Synthesis of phthalazine and pyridazine derivative.



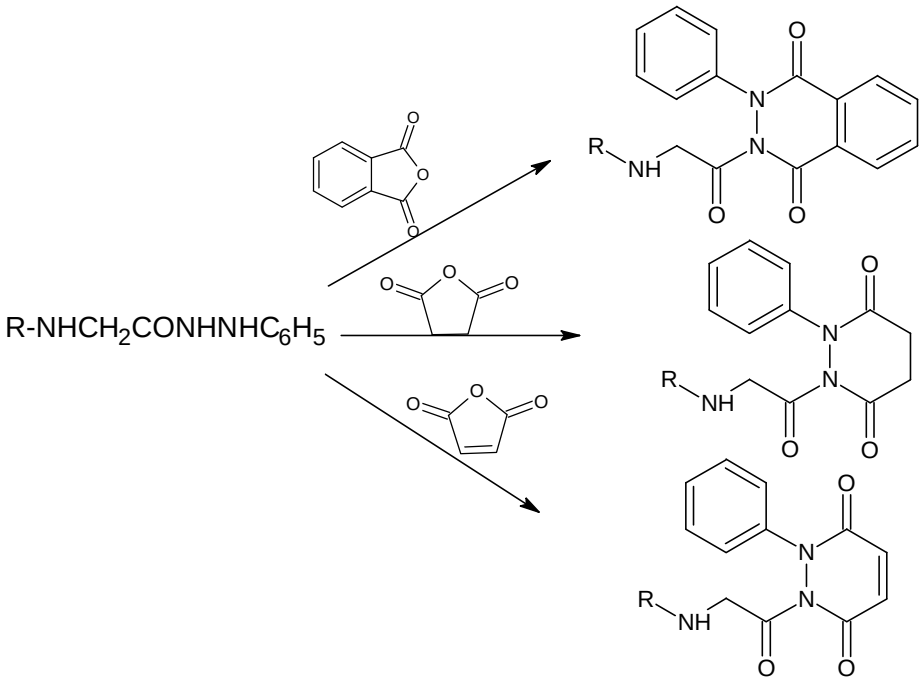
Synthesis of phthalazine and pyridazine derivatives

The above N-substituted hydrazide (0.01mol) was reacted with phthalic anhydride (0.01ml), succinic anhydride (0.01ml) and maleic anhydride (0.01ml) in presence of 5ml, absolute ethanol and glacial acetic acid (0.005ml) and was refluxed for 3hrs. Cooled and the reaction mixture was poured into crushed ice. The solid obtained was filtered , washed with diluted sodium bicarbonate solution and recrystallised with suitable solvent.

Synthesis of phthalazine and pyridazine derivatives

The above N-substituted phenyl hydrazide (0.01mol) was reacted with phthalic anhydride (0.01ml), succinic anhydride (0.01ml) and maleic anhydride (0.01ml) in presence of 5ml, absolute ethanol and glacial acetic acid (0.005ml) and was refluxed for 3hrs. Cooled and the reaction mixture was poured into crushed ice. The solid obtained was filtered, washed with diluted sodium bicarbonate solution and recrystallised with suitable solvent.

Step III- Synthesis of phthalazine and pyridazine derivative.

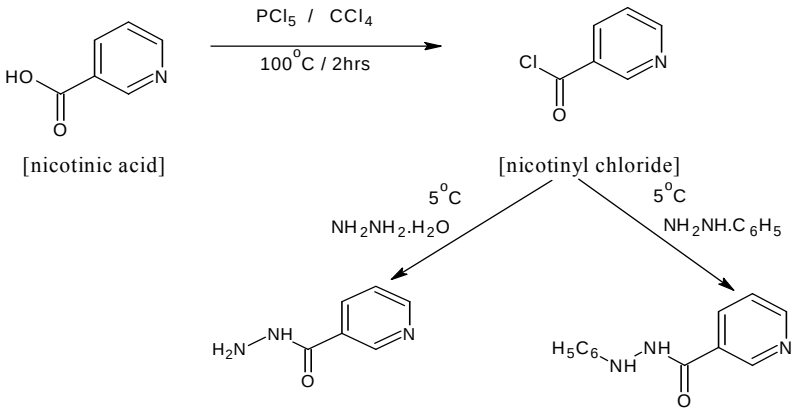


SchemeII-(synthesis of some N-substituted Phthalazine and pyridazine derivatives)

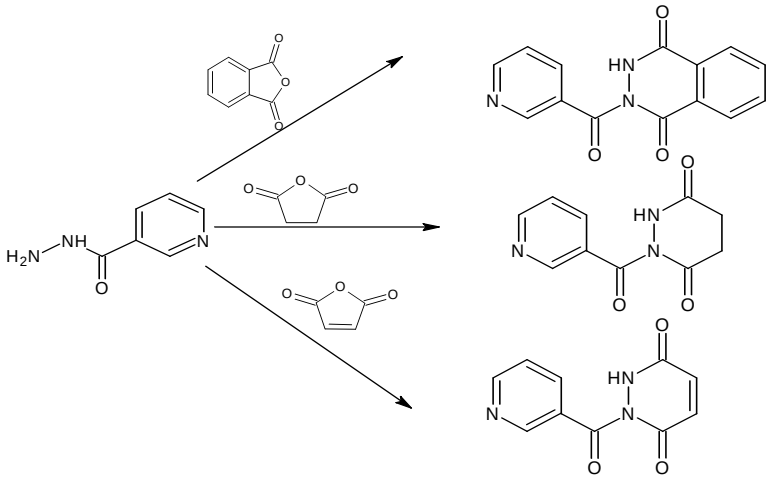
Synthesis of nicotinic acid hydrazides

A mixture of nicotinic acid [4.1gm], phosphorous pentachloride [10.3gm] in anhydrous carbontetrachloride(20ml) were refluxed for 2hrs at 100^{o}C .Solvents were distilled off and solid acid chloride was then divided into two equimolar ratio and was used for further reaction.

Step I- Synthesis of Nicotinic acid hydrazides .



Step II- Synthesis of phthalazine and pyridazine derivative.



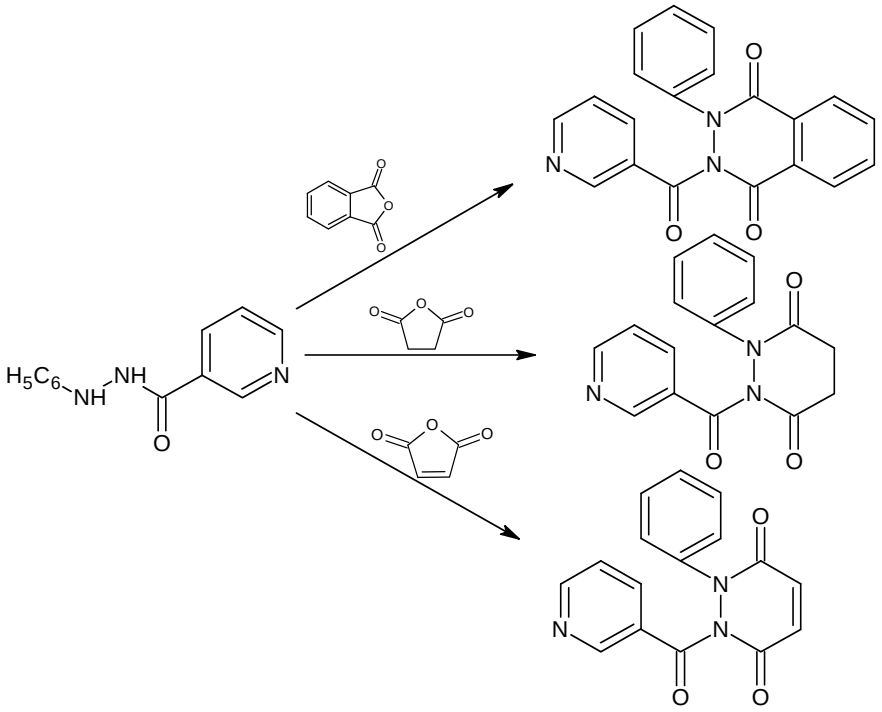
To the first portion of nicotinyl chloride [0.03mol] was added with hydrazine hydrate [0.1mol] and to other nicotinyl chloride[0.03mol] was added with phenyl hydrazine [0.1mol]. This was carried out cautiously below 5°C and are added slowly in drop wise with constant stirring. The reaction mixtures were then stirred for 5 hrs at room temperature. The solids were washed with dilute sodium bicarbonate solution. Dried vacuum and recrystallized from methanol.

To the second portion of nicotinyl chloride [0.03mol] was added with phenylhydrazine [0.1mol] and to other nicotinyl chloride[0.03mol] was added with phenyl hydrazine [0.1mol]. This was carried out cautiously below 5°C and are added slowly in drop wise with constant stirring. The reaction mixtures were then stirred for 5 hrs at room temperature. The solids were washed with dilute sodium bicarbonate solution. Dried vacuum and recrystallized from methanol.

Synthesis of phthalazine and pyridazine derivatives

The hydrazine hydrate and phenylhydrazine (each [0.01mol] was reacted with phthalic anhydride[0.01mol], succinic anhydride [0.01mol] , and maleic anhydride [0.01mol] in presence of 5ml, absolute ethanol and glacial acetic acid (0.005ml) and was refluxed for 3hrs. Cooled and the reaction mixture was poured into crushed ice. The solid obtained was filtered , washed with diluted sodium bicarbonate solution and recrystallised with suitable solvent.

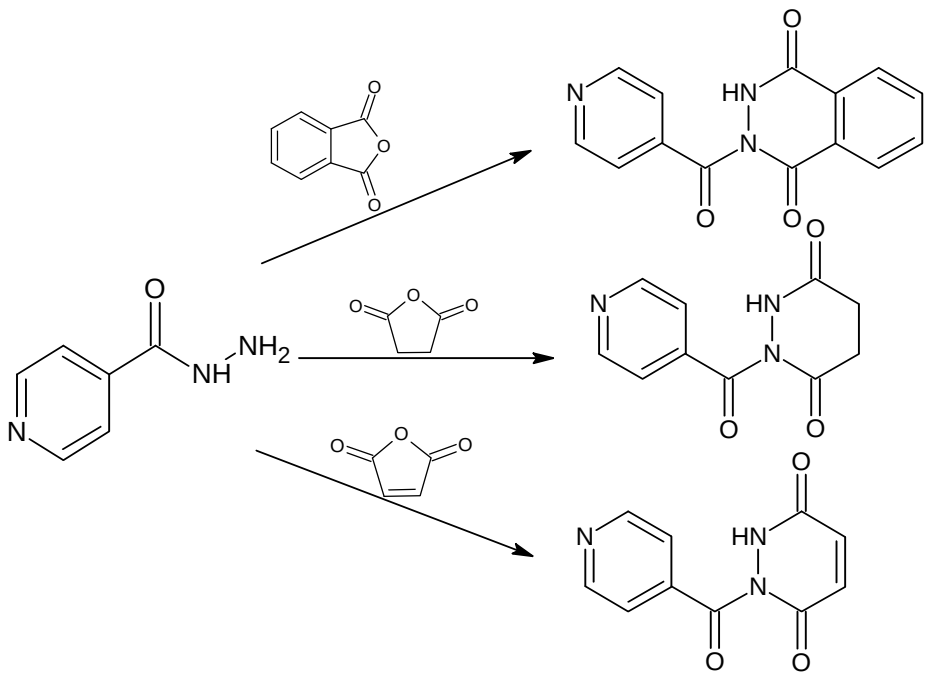
Step III- Synthesis of phthalazine and pyridazine derivative.



Scheme-III: (Synthesis of N- substituted Phthalazine and pyridazine derivatives)

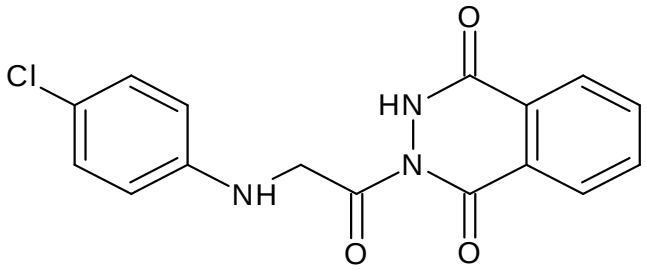
Isoniacid [65,81][0.01mol] was treated individually with phthalic anhydride[0.01mol], succinic anhydride [0.01mol] , and maleic anhydride [0.01mol] in separate round bottom flask. These are refluxed for 3 hrs with 5ml absolute ethanol and glacial acetic acid [0.005mol]. Cooled and the reaction mixture was poured into crushed ice. The solid obtained was filtered, washed with dilute sodium bicarbonate solution and recrystallized from suitable solvent.

Synthesis of phthalazine and pyridazine derivative.



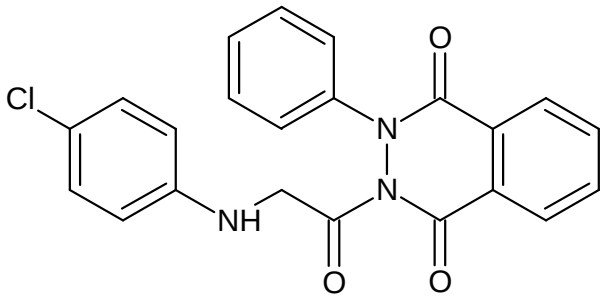
PHYTOCHEMICAL INVESTIGATION[11,12,13]

Ia. Compound name: 2-{[(4-chlorophenyl)amino]acetyl}-2,3-dihydrophthalazine-1,4- dione



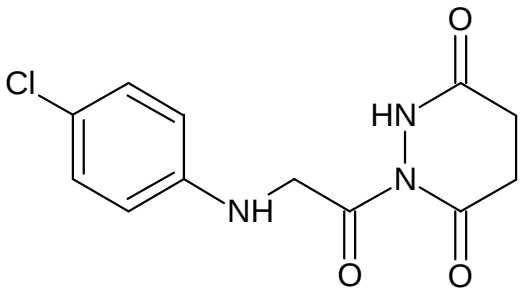
IR spectral data 3473cm-[1] O-H stretching, 3068-3184cm-[1] aromatic stretching,2854cm-[1] C-H stretching,1652,1697cm-[1] C=N stretching,1446-1591cm[1] C=C stretching,1168cm-[1] C-O stretching (aromatic),835cm-[1] aromatic bending. [1]HNMR 6.82-6.85 δ(ppm) aromatic proton,8.01-8.144 δ(ppm) N=CH, 3.52 δ(ppm) OH proton.

Ib. compound name : 2-{[(4-chlorophenyl)amino]acetyl}-3-phenyl-2,3-dihydrophthalazine-1,4-dione



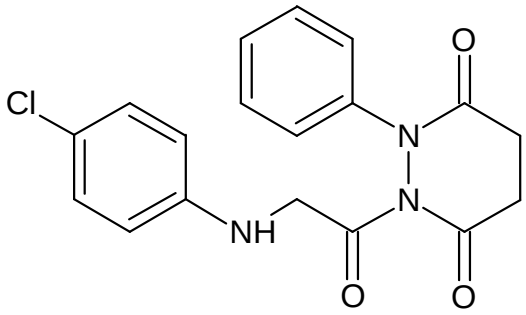
IR spectral data 3471cm-^{1} O-H stretching, 3069-3183cm^{-1} aromatic stretching, 2853cm-^{1} C-H stretching,1654,1698,1592cm-^{1}C=Nstretching,1446-1591cm^{1}C=C stretching, 1270cm-^{1} C-O stretching (aromatic),1025cm-1, O-C stretching (aliphatic), 864cm-^{1} aromatic bending.

IIa. compound name : 1-{[(4-chlorophenyl)amino]acetyl}tetrahydropyridazine-3,6-dione



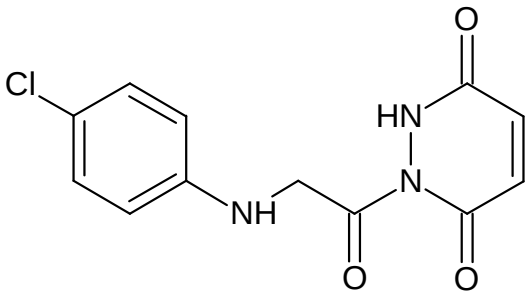
IR spectral data 3406cm-^{1} O-H stretching, 2919cm^{-1} aromatic stretching,2850cm-^{1} C-H stretching,1621cm-^{1} C=N stretching,1462cm-^{1} C=C stretching,1166cm-^{1} C-O stretching (aromatic),872cm-^{1} aromatic bending.

IIb.compoundname:1-{[(4-chlorophenyl)amino]acetyl}-2phenyltetrahydropyridazine-3,6-dione.



IR spectral data 3456cm-^{1} O-H stretching, 3055cm^{-1} aromatic stretching,2854cm-^{1} C-H stretching,1681,1608cm-^{1} C=N stretching,1571cm^{1} C=C stretching,1168cm-^{1} C-O stretching (aromatic),833cm-^{1} aromatic bending.

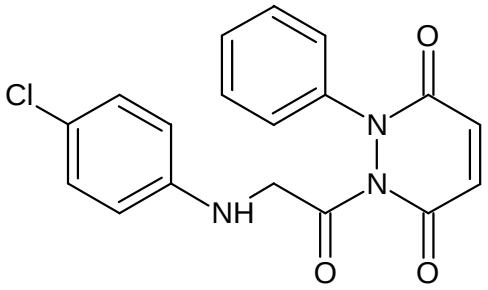
IIIa. compound name : 1-{[(4-chlorophenyl)amino]acetyl}-1,2-dihydropyridazine-3,6-dione



IR spectral data 3134-3323cm^{-1} aromatic stretching, 2848-2918cm-^{1}C-H stretching,1600,1604cm-^{1}C=N stretching,1458-1500cm^{1}C=C stretching, 1174cm-^{1} C-O stretching (aromatic), 1020-1078cm-1, O-C stretching (aliphatic), 831cm-^{1} aromatic bending.

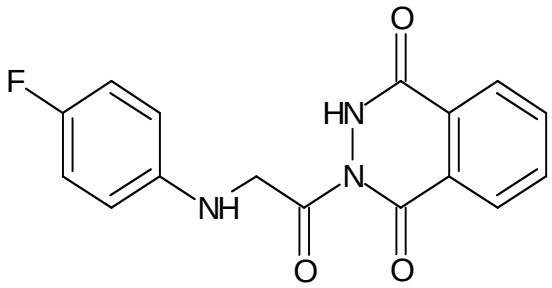
IIIb. compound name : 1-{[(4-chlorophenyl)amino]acetyl}-2-phenyl-1,2-

dihydropyridazine- 3,6-dione



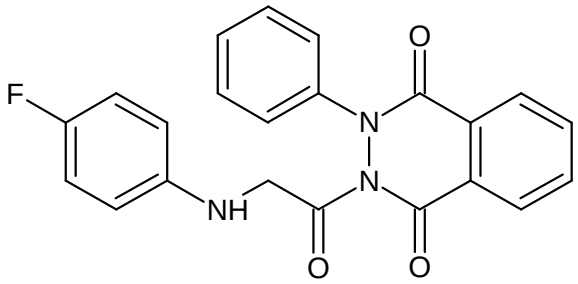
IR spectral data 3324cm-^{1} O-H stretching, 3134cm^{-1} aromatic stretching,2854cm-^{1} C-H stretching,1604,1668cm-^{1} C=N stretching,1500cm^{1} C=C stretching,1178cm-^{1} C-O stretching (aromatic)1020cm^{-1}O-Cstretching (aliphatic),831cm-^{1} aromatic bending.

IVa. compound name : 2-{[(4-fluorophenyl)amino]acetyl}-2,3-dihydrophthalazine-1,4-dione



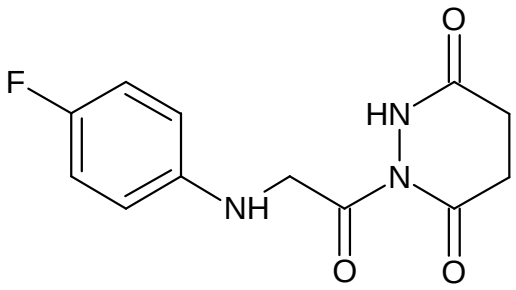
IR spectral data stretching, 3168cm^{-1} aromatic stretching,2995cm-^{1} C-H stretching, 1689cm-^{1} C=N stretching,1508cm^{1} C=C stretching,1249cm-^{1} C-O stretching (aromatic)1026cm^{-1}O-Cstretching (aliphatic),823cm-^{1} aromatic bending .

IVb. compound name : 2-{[(4-fluorophenyl)amino]acetyl}-3-phenyl-2,3-dihydrophthalazine1,4-dione



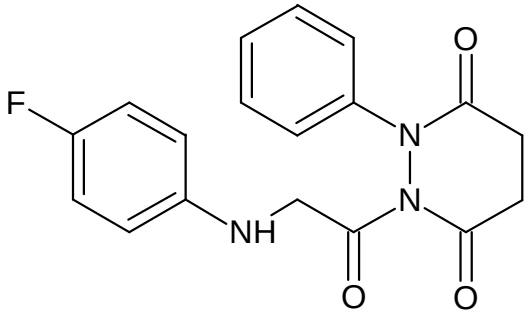
IR spectral data 3134cm^{-1} aromatic stretching,2854cm-^{1} C-H stretching,1606,1656cm-^{1} C=N stretching,1500cm^{1} C=C stretching,1178cm-^{1} C-O stretching (aromatic)1020cm^{-1}O-Cstretching (aliphatic),831cm-^{1} aromatic bending.

Va. compound name : 1-{[(4-fluorophenyl)amino]acetyl}tetrahydropyridazine-3,6-dione



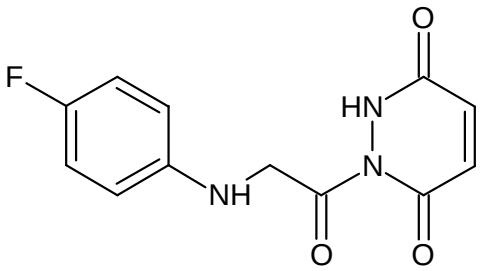
IR spectral data 3458cm-^{1} O-H stretching, 3055cm^{-1} aromatic stretching,2923cm-^{1} C-H stretching,1647,1689cm-^{1} C=N stretching,1512cm^{1} C=C stretching,1139cm-^{1} C-O stretching (aromatic)1027cm^{-1}O-Cstretching (aliphatic),854cm-^{1} aromatic bending.

Vb. compound name : 1-{[(4-fluorophenyl)amino]acetyl}-2-phenyltetrahydropyridazine-3,6-dione



IR spectral data 3463cm-^{1} O-H stretching, 3197-3072cm^{-1} aromatic stretching,2935cm-^{1} C-H stretching,1602,1647cm-^{1} C=N stretching,1514cm^{1} C=C stretching,1274cm-^{1} C-O stretching (aromatic)1029cm-^{1}O-C stretching(aliphatic),810cm-^{1} aromatic bending.

VIa. compound name : 1-{[(4-fluorophenyl)amino]acetyl}-1,2-dihydropyridazine-3,6-Dione

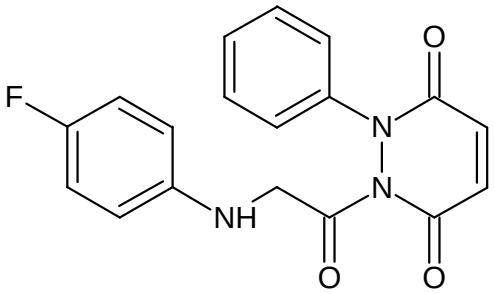


IR spectral data 3514cm-^{1} O-H stretching, 3070cm^{-1} aromatic stretching,2920cm-^{1} C-H stretching,1602,1647cm-^{1} C=N stretching,1514cm^{1} C=C stretching,1274cm-^{1} C-O stretching (aromatic)1029cm-^{1}O-C stretching(aliphatic),854cm-^{1} aromatic bending.

VIb.Compound name: 1-{[(4-fluorophenyl)amino]acetyl}-2-phenyl-1,2-dihydropyridazine-3,6-dione

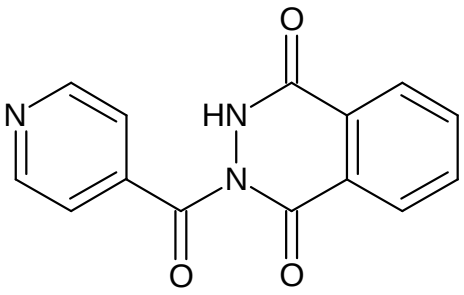
IR spectral data 3422.96 N-H stretching, 2921.88 C-H stretching, 1706.1 C=O stretching,

1615.82, N-H bending, 1392.36 C-N stretching(aromatic).

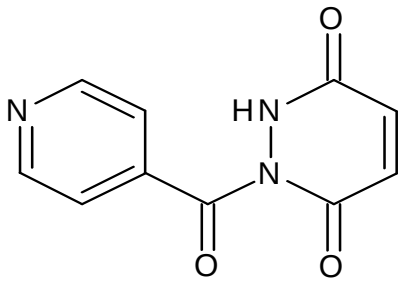


X. Compound name : 2-(pyridin-4-ylcarbonyl)-2,3-dihydrophthalazine-1,4-dione

IR spectral data 3414.36 N-H stretching, 2921.53 C-H stretching, 1695.53C=O stretching, 1615.1 N-H bending, 1393.03 C-N stretching(aromatic).



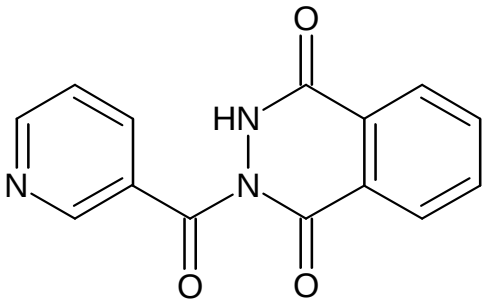
XII. compound name : 1-(pyridin-4-ylcarbonyl)-1,2-dihydropyridazine-3,6-dione



IR Spectral Data

3421.02 N-H stretching, 2920.02 C-H stretching, 1706.07C=O stretching, 1614.94 N-H bending, 1392 C-N stretching(aromatic).

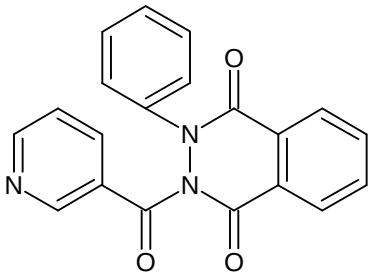
XIIIa. compound name : 2-(pyridin-3-ylcarbonyl)-2,3-dihydrophthalazine-1,4-dione



IR Spectral Data

3194.5 N-H stretching, 1700.2 C=O stretching, 1504.76 N-H bending, 1093.67 C-Cl stretching(aromatic).

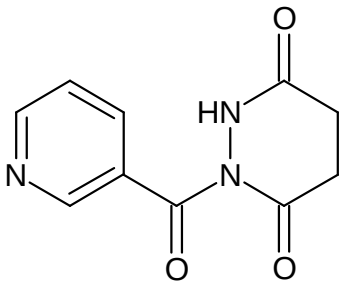
XIIIb. Compound name : 2-phenyl-3-(pyridin-3-ylcarbonyl)-2,3-dihydrophthalazine-1,4-dione



IR Spectral Data

3043.6 (C-H stretching aromatic), 2872.24 (C-H stretching aldehyde), 1688.65(C=O stretching), 1613.94, 1578.4, 1489.93(C=C &C=N stretching(aromatic), 1045.74(C-Cl stretching(aromatic).

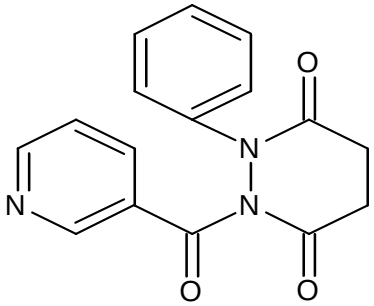
XIVa. compound name : 1-(pyridin-3-ylcarbonyl)tetrahydropyridazine-3,6-dione



IR Spectral Data

3423.47 N-H stretching, 1708.3 C=O stretching, 1653.75 N-H 0vertone, 1404.08 C-H bending, 1036.41 C-Cl stretching(aromatic).

XIVb. compound name : 1-phenyl-2-(pyridin-3-ylcarbonyl)tetrahydropyridazine-3,6-dione



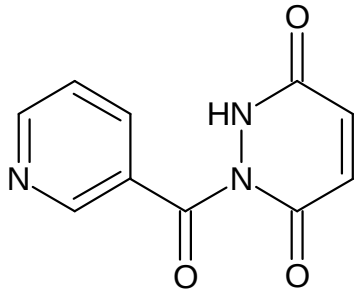
IR Spectral Data

3194.5 N-H stretching, 1700.2 C=O stretching, 1504.76 N-H bending, 1093.67 C-Cl stretching(aromatic).

NMR Spectral Data

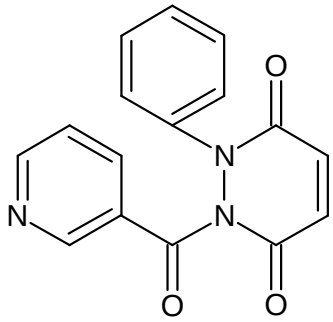
1.2 Methyl proton, 4.675-4.777 Methylene proton, 1.76 Amino proton, 7.26-8.13 Hetro aromatic hydrogen.

XVa. compound name : 1-(pyridin-3-ylcarbonyl)-1,2-dihydropyridazine-3,6-dione.



NMR spectral data 2.93-2.96 Methyl proton, 3.94-3.99 Methylene proton(quartet), 2.404 Amino proton, 6.914-7.525 Hetro aromatic hydrogen.

XVb. compound name : 1-phenyl-2-(pyridin-3-ylcarbonyl)-1,2-dihydropyridazine-3,6-dione



IR spectral data 2925cm-^{1} C-H stretching,1654cm^{1} C=C stretching,1651N-H bending,1499cm-^{1} C-H bending (aromatic),754cm-^{1}C-C bending, 689 cm-^{1} C-H bending.

PHARMACOLOGICAL ACTIVITY

EVALUATION OF ANTICANCER ACTIVITY

Selection Grouping and Acclimatization of Laboratory Animal[14,15]

Male Swiss albino mice (20-25 gm) were produced from animal experimental laboratory, and used throughout the study. They were housed in micro nylon boxes in a control environment (temp 25±2°C) and 12 hrs dark /light cycle with standard laboratory diet and water *ad libitum.* The study was conducted after obtaining institutional animal ethical committee clearance. As per the standard practice, the mice were segregated based on their gender and quarantined for 15 days before the commencement of the experiment. They were fed on healthy diet and maintained in hygienic environment in our animal house.

Induction of cancer using DLA cells

Dalton's Lymphoma ascites (DLA) and Ehrlich Ascites Carcinoma (EAC) cells were supplied by Amala cancer research center, Trissur, Kerala, India. Cells maintained in vivo in Swiss albino mice by intraperitoneally transplantation. While transforming the tumor cells to the grouped animal the DLA cells were aspirated from peritoneal cavity of the mice using saline. The cell counts were done and further dilution were made so that total cell should be 1×10^6 , this dilution was given intraperitonealy.Let the tumor grow in the mice for minimum seven days before starting treatments. Table 3,4

Treatment Protocol

Swiss Albino mice were divided in to eight group of six each. All the animals in seven groups were injected with DLA cells (1×10^6 cells per mouse) intraperitoneally, and the remaining one group is normal control group.

Group 1 served as the normal control.

Group 2 served as the tumor control. Group 1 and 2 receives normal diet and Water.

Group 3 served as the positive control, was treated with injection fluorouracil at 20

Mg/kg body weight, Intra peritoneally[1]

Group 4 served as the treatment control, treated with synthetic drugs such as I a at 10 mg/kg dissolved with 0.5ml of DMSO administered through orally.

Group 5 served as the treatment control and was treated with synthetic drugs such as IV- b at 10 mg/kg dissolved with 0.5ml of DMSO administered through orally.

Group 6 served as the treatment control, which was treated with synthetic drugs such as X at 10 mg/kg dissolved with 0.5ml of DMSO administered through orally.

Group 7 served as the treatment control and was treated with synthetic drugs such as XIII-a at 10 mg/kg dissolved in 0.5ml of DMSO administered through orally.

Group 8 served as the treatment control and was treated with synthetic drugs such as XIII-B at 10 mg/kg dissolved in 0.5ml of DMSO administered through orally.

Treatment

In this study, drug treatment was given after the 24 hrs of inoculation, once daily for 14 days.
On day 14, after the last dose, all mice from each group were sacrificed; the blood was withdrawn from each mouse by retro orbital plexus method and the following parameters were checked.

1. Hematological parameters[16,17]
    a. WBC count
    b. RBC count
    c. Hb content
    d. Platelet count
    e. Packed cell volume
2. Derived parameter
    f. Body weight
    g. Life span (%)

ANTICONVULSANT ACTIVITY [18,19]

Supra maximal electrical shock

Healthy albino wistar rats weighing from 200-250 gm were selected. They were kept in separate cages, fed with balanced diet, water and libitum. Then the animal were divided into 23 groups, each containing six animals.The first group of animals were served as control, which received 10ml/kg of normal saline. Second group served as standard which received phenytoin sodium[25mg/kg]. Group III to 23 treated with various synthetic compounds at, 10mg/kg dissolved in 0.5ml of DMSO. All the test compounds were dissolved in solvent like DMSO and administered through intra-peritoneal route.The evaluation was started 30 minutes after administration of the test compounds. Pinna electrodes with the intensity of 150 mA current were used to deliver the stimuli.

Inhibition of seizure relative to the control was calculated . Table5

MICROBIOLOGICAL STUDIES

ANTIBACTERIAL AND FUNGAL ACTIVITY[20,21,22,23,24,25]

Each compound of 10mg was weighed in sterile boiling tubes, dissolved using 0.5ml DMSO and mixed using remi cyclo mixer. Then it was made up to 10ml, 1ml of the test solution contains 1mg of test compound Assay was carried out by diffusion plate method. The method followed was spread plate technique. The agar plates free from contamination were spread with 50µl of 48h old culture of bacterial test organism using sterile buds. The std disc of Amikacin (sterile) of 5 mm diameter was in the

Petri plates. Then the filter paper discs (sterile) of 5 mm were soaked in 1ml (1µg/ml) of the test solution and in solvent control DMSO. After evaporating the solvent in a sterile atmosphere the drug impregnated discs were placed in Petri plates. The plates were refrigerated for 1h to arrest the growth and for easier diffusion of test compounds. Then the plates were removed from refrigerator and incubated at 37^0C over night is an inverted position. The clear zones of inhibition were measured using Hi media zone reader scale. The values are tabulated. The zones of test solutions were compared with standard Amikacin. Test with gram positive and gram negative bacteria were carried out in Muller Hilton agar. Anti fungal activity was also evaluated in Sabouraud Dextrose Agar. Table 6.

RESULTS AND DISCUSSION

Anticancer

Effect on Tumor Growth

In the DLA tumor control group, the average life span of animal was found to be 46% where as various synthetic drugs at the dose of 10 mg/kg body weight increase the life span to 70%,67%,71%,72% and 65% respectively. These values were significant. However the average life span of 5- FU treatment was found to be 90%, indicating its potent antitumor nature. The antitumor nature of various synthetic drugs were evidenced by the significant reduction in percent increase in body weight of animal treated with various synthetic drugs at the dose of 10 mg/kg body weight when compared to DLA tumor bearing mice. It was also supported by the significant reduction in packed cell volume In the extent of treatment when compared to the DLA tumor control. (Table No .4)

Effect on Hematological Parameters

As shown in (Table No.4) RBC, Hgb, Platelets were decreased and WBC count was significantly increased in the DLA control group compared to the normal control group. Treatment with various synthetic drugs at the dose (10 mg/kg) significantly increases the Hgb content, RBC, Platelets and significantly decreased the WBC count to about normal level. All these results suggest the anticancer nature of the various synthetic drugs However, the standard 5-FU at the dose of 20 mg/kg body weight produced better result in all these parameters.

In DLA tumor bearing, a regular rapid increase in ascitic tumor volume was observed. Ascitic fluid is the direct nutritional source for tumor cells and a rapid increase in ascitic fluid with tumor growth would be a means to meet the nutritional requirement of tumor cells. [86] Treatment with various synthetic drugs inhibited the tumor volume, viable tumor cell count and increased the life span of the tumor bearing mice. The reliable criteria for judging the value of any anticancer drug are the prolongation of the lifespan of animals.[4] It may be concluded that various synthetic drugs by

decreasing the nutritional fluid volume and arresting the tumor growth increases the life span of DLA bearing mice. Thus various synthetic drugs have antitumor activity against DLA bearing mice.

Usually, in cancer chemotherapy the major problems that are being encountered are of myelo suppression and anemia. [5,6] The anemia encountered in tumor bearing mice is mainly due to reduction in RBC or hemoglobin percentage and this may occur either due to iron deficiency or due to hemolytic or myelopathic conditions.[7] Treatment with various synthetic drugs brought back the hemoglobin (Hb) content, RBC and WBC count more or less to normal levels significantly. This clearly indicates that various synthetic drugs possess protective action on the haemopoietic system.

Anticonvulsant activity

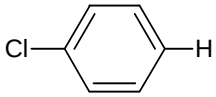
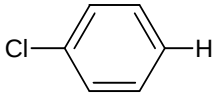
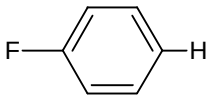
The table values reveals that after treatment with various synthetic compounds, the compounds such as Ia,Ib,IIa,IIb.IIIa,IIIb.IVa,IVb,VIa,VIb,XIIIa & XIIIb possess significant Anti convulsant effects. All above said cmpounds were reduced the duration of extensor phase significantly.

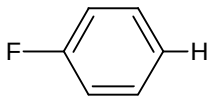
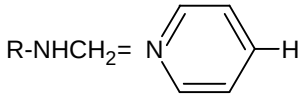
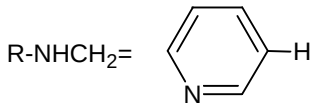
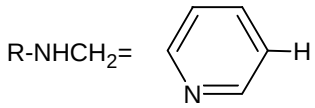
Anti microbial activity

The anti microbial studies reveal that the compounds like Ia,IIIa, Vb and VIa shown to possess good activity against gram (-)ve micro organism.Similarly synthetic compounds like IIIa VIa and VIb shown good activity against gram (+) microorganism. It also possess excellent activity against ***Candida albicans*** reveals that the synthesized compounds like XIVa and XI possess antifungal activity.

Structure of compounds

Table : 1

COMPOUND	R	R_1
I,II,III. (a)		H
I,II,III. (b)		C_6H_5
IV,V, VI. (a)		H

IV,V, VI. (b)		C_6H_5
X,XI,XII.		H
XIII,XIV,XV. (a)		H
XIII,XIV,XV. (b)		C_6H_5

Solubility of compounds

Table: 2

Sl. No	Solvents	Compounds																				
		I		II		III		IV		V		VI		X	XI	XII	XIII		XIV		XV	
		a	b	a	b	a	b	a	b	a	b	a	b	a	a	a	a	b	a	b	a	b
1.	H_2O	-	-	-	-	-	-	-	-	-	-	-	-	+	+	+	-	-	-	-	-	-
2.	CH_3OH	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
3.	CH_3COCH_3	+	+	+	+	+	+	+	+	+	+	+	+	-	-	-	-	-	-	-	-	-
4.	CH_3COOC_2H5	-	-	-	-	-	-	-	-	-	-	-	-	-	-	±	-	-	-	-	-	-
5.	$C_4H_{10}O$	±	±	±	±	±	±	±	±	±	±	±	±	-	-	-	±	±	±	±	±	±
6.	$CHCl_3$	+	+	+	+	+	+	+	+	+	+	+	+	-	-	-	+	+	+	+	+	+
7.	C_6H_6	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
8.	CCL_4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	±	±	±	±	±	±
9.	C_6H_{12}	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
10.	DMF	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
11.	DMSO	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
12.	Dil.HCl	-	±	-	±	-	±	-	±	±	±	-	-	-	-	-	-	-	±	-	-	-
13.	10%NaOH	-	-	-	-	-	-	-	-	-	-	-	-	±	±	±	-	-	-	-	-	-

H_2O : Water 10%NaOH :Dilute sodium hydroxide solution

CH_3OH : Methanol DMF : Dimethyl formamide

C_6H_6 : Benzene DMSO : Dimethyl sulfoxide

C_6H_{12} : Hexane Dil.HCl : Dilute hydrochloric acid

CH_3COCH_3 : Acetone CCL_4 : Carbon tetrachloride

$CHCl_3$: Chloroform $C_4H_{10}O$: Diethylether

CH_3COOC_2H5 : Ethyl acetate

[+] : Freely soluble [±] : Partially soluble

[-] : Insoluble

Table: 3

Effect of various synthetic drugs on Hematological parameters

TREATMENT	Total WBC Cells /mlx10^3	Rbc Count Mill/cumm	Hb Gm/dl	PCV %	Platelets Lakhs/cumm
G1	9.85 ±0.28	4.50±0.40	12.30 ±1.32	14.45±1.20	3.20±0.45
G2	14.35 ±1.45^{a**}	2.47±0.22^{a**}	7.06 ±0.65^{a**}	28.50±2.60^{a**}	1.70±0.25^{a**}
G3	11.30 ±1.03^{b**}	4.08±0.55^{b**}	9.4 ±0.55^{b**}	18.42±1.55^{b**}	2.65±0.10^{b**}
G4	12.42±0.95^{b**}	3.30±0.45^{b**}	11.30±0.40^{b**}	26.06±1.65^{b**}	2.22±0.18^{b**}
G5	12.03 ±1.10^{b**}	3.25±0.24^{b**}	11.70±0.45^{b**}	24.06±1.48^{b**}	2.40 ±0.10^{b**}
G6	12.86 ±0.92^{b**}	3.10±0.32^{b**}	11.55±0.40^{b**}	22.10±1.52^{b**}	2.20 ±0.16^{b**}
G7	11.95 ±0.85^{b**}	2.95±0.40^{b**}	11.44±0.52^{b**}	23.45±1.54^{b**}	2.16 ±0.24^{b**}
G8	12.10 ±0.78^{b**}	3.05±0.37^{b**}	11.25±1.04^{b**}	21.12±1.05^{b**}	2.05 ±0.28^{b**}

G_1 – Normal Control, G_2 – Cancer Control, G_3 – Positive control, G_4 to G_8Treatment controls All values are expressed as mean ± SEM for 6 animals in each group.

**a – Values are significantly different from control (G_1) at $P < 0.001$

**b – Values are significantly different from cancer control (G_2) at $P < 0.001$

Table No.4

Effect of various synthetic drugs on the life span and body weight of tumor induced mice.

Treatment	Number of animals	% ILS Life span	Increase in Body weight grams
G_1	6	>>30 days	1.90±0.32
G_2	6	46%	8.10±0.33^{a**}
G_3	6	90%	3.80±0.44^{b**}
G_4	6	70%	6.40±0.40^{b**}
G_5	6	67%	6.25±0.32^{b**}
G6	6	71%	6.12±0.28^{b**}
G7	6	72%	6.78±0.42^{b**}
G8	6	65%	6.45±0.35^{b**}

G_1 -Normal Control, G2 -Tumor Control, G_3- Positive control, G_4 to G_8Treatment controls

All values are expressed as mean ± SEM for 6 animals in each group.
**a – Values are significantly different from control (G_1) at $p<0.001$

**b – Value are significantly different from tumor control (G_2) at $p<0.001$

All values are found out by using one way ANOVA followed by Newman Keul's multiple range tests.

ANTI CONVULSANT ACTIVITY OF VARIOUS SYNTHESISED DRUGS BY SUPRAMAXIMAL ELECTRICAL SHOCK METHOD.

Table :5

Groups	Treatment	Drug & dose	Duration of extension phase in sec.	% inhibition of extension phase

Group I	Normal control	Normal saline 10ml/kg	13.6 ± 0.96	—
Group II	Positive control	Phenytoin sodium.25mg/kg	1.6 ± 0.12	87.8 %
Group III	Treatment control	Ia. - 10mg/kg	4.10 ± 0.36	68.8 %
Group IV	Treatment control	Ib- 10mg/kg	3.96 ± 0.28	68.8 %
Group V	Treatment control	IIa- 10mg/kg	4.28 ± 0.32	67.4 %
Group VI	Treatment control	IIb- 10mg/kg	3.60 ± 0.18	72.6 %
Group VII	Treatment control	IIIa- 10mg/kg	4.52 ± 0.42	65.6 %
Group VIII	Treatment control	IIIb- 10mg/kg	4.36 ± 0.39	66.8 %
Group IX	Treatment control	IVa- 10mg/kg	3.85 ± 0.20	70.7 %
Group X	Treatment control	IVb- 10mg/kg	3.60 ± 0.20	72.6 %
Group XI	Treatment control	Va- 10mg/kg	8.62 ± 1.06	34.4 %
Group XII	Treatment control	Vb- 10mg/kg	9.60 ± 1.20	27 %
Group XIII	Treatment control	VIa- 10mg/kg	4.30 ± 0.35	67.32 %
Group XIV	Treatment control	VIb - 10mg/kg	4.15 ± 0.42	68.4 %
Group XV	Treatment control	X- 10mg/kg	7.48 ± 0.67	43.16 %
Group XVI	Treatment control	XI- 10mg/kg	8.26 ± 0.60	37.2 %
Group XVII	Treatment control	XII- 10mg/kg	8.20 ± 0.52	37.2 %
Group XVIII	Treatment control	XIIIa- 10mg/kg	3.62 ± 0.30	72.4 %
Group XIX	Treatment control	XIIIb- 10mg/kg	3.80 ± 0.36	71.0 %
Group XX	Treatment control	XIVa- 10mg/kg	7.65 ± 0.55	41.8 %
Group XXI	Treatment control	XIVb- 10mg/kg	8.12 ± 0.65	38.2 %
Group XXII	Treatment control	XVa- 10mg/kg	9.0 ± 1.10	31.6 %
Group XXIII	Treatment control	XVb- 10mg/kg	8.65 ± 0.82	33.1 %

Table 6 Antimicrobial activity

Sl.no	Compound	Zone of inhibition against		
		E.coli #(%)	**Pseudomonas *(%)**	**Candida.Albicans**(%)
1.	Ia	**80**	40	23
2.	IIIa	70	60	45
3.	IIIb	40	**R**	**R**
4.	Vb	60	35	16
5.	VIa	60	75	46
6.	VIb	45	**90**	30
7.	XIVa	30	47	62
8.	XI	30	**R**	**77**

* *P. Aerugnosa* , # *Escherichia coli*, *R Resistant strain*

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