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Research Article

Wheat Rootlet Growth Inhibition Assay and Antimitotic studies of Some N^1, N^3 Bis (6-chlorobenzo (d) thiazol-2-yl) 2–substituted methyl malonamides

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Abstract: DNA intercalatiors are one of the most important and widely used classes of anti-cancer drugs. Based on the essentially required structural features hereby we have proposed to synthesize a novel series of dimeric compounds which would be capable of intercalating DNA in the minor groove and could serve as a model for developing a novel class of anti-cancer agents. It is evident from the literature review that benzothiazoles possess wide range of biological & pharmacological properties including anticancer properties. in present investigations we have attempted to design a novel series of compounds which contained N^1, N^3 Bis (6-chlorobenzo (d) thiazol-2-yl) 2 – substituted methyl malonamide molecular framework of possible Bis-DNA intercalators. Based on this hypothesis and the proposed model a new series of N^1, N^3 Bis (6-chlorobenzo (d) thiazol-2-yl) 2–substituted methyl malonamides were developed and screened for their anticancer activity by Wheat rootlet growth inhibition assay Antimitotic studies using onion root model. The compounds were characterized by FT-IR and ^{1}H-NMR spectral studies. Solubility studies, R_f value determination, Percentage yield determination were done. In wheat rootlet growth inhibition assay Compounds IVg (Fluoro) and Vie (Chloro) were most active with percentage inhibition of 50 and 54 respectively. Compounds IVg (Fluoro) and Vie (Chloro) were most active with percentage inhibition of 50 and 54 respectively. In antimitotic studies using onion root model the series compounds IVj (Nitro) was found to be more potent with 54 and 74% inhibition of mitosis at 10 and 20µg/ml respectively. It was followed by the compounds IVc (Aniline) & IVe (Chloro) which showed about 70% inhibition at 20µg/ml concentration.
Keywords: DNA intercalatiors, Mannich bases, Anticancer, Antimitotic.

INTRODUCTION

It is 50 years since Watson & Crick determined that genetic material exists structurally as a double helix with now well-established characteristics [1]. Its role in the control of cellular functions immediately suggested it as an excellent target for treating illnesses of genetic origin, such as cancer. The first compounds discovered to act on DNA were the sulfur mustards, but their high toxicity prompted to a search for less toxic and more efficient compounds [2]. With cytotoxic activities were the 1960s, some compounds with cytotoxic activity were discovered to act as anticancer agents, although their mechanism of action was unknown. Interestingly, after Lerman reported the occurrence of a noncovalent interaction between acridine and DNA, suggesting an intercalative process, it was established that some of these anticancer agents worked by interacting with DNA [3, 4].

DNA intercalation represent are of the most important and widely used classes of Anti-cancer drugs. The binding of foreign molecules to DNA causes substantial alterations in the normal transcription machinery of the genes, resulting in general modifications of some genes, including those involved in cell-cycle or differentiation. Intercalators are molecules that insert perpendicularly into DNA without forming covalent bonds. The only recognized forces that maintain the stability of the DNA-intercalators complex, even more the DNA alone, are van der Waals, hydrogen bonding, hydrophobic, and/or charge transfer forces [5- 8]. A frontier orbital interaction has also been suggested [8]. This means that such a process has the possibility of being reversed, and as a consequence it must have equilibrium constant. It has been argued that the 9-amino group is important in the DNA-recognizing region because of its ability to form hydrogen bonds [5, 9].

Drugs exerting their biological activity by non-covalent binding to the A-T rich region in the minor groove of DNA have emerged as a novel class of anti cancer drugs. These agents exhibit their activity by bis-intercalaion of the double stranded DNA. Any agent having two aromatic / hetero aromatic rings separated by a linker chain & containing are/more polar groups capable of forming H2 bonds can serve as a potential candidate for binding with minor groove in DNA.

A good number of reports from the literature clearly prove the utility of bis-DNA intercalator, where activity is associated with high DNA binding affinity several researchers have proved that dimeric molecules have a potential to intercalate both chains of a double stranded DNA which is more desirable.

Based on the essentially required structural features we have proposed to synthesize a novel series of dimeric compounds which would be capable of intercalating DNA in the minor groove and could serve as a model for developing a novel class of anti-cancer agents. With the main objective to develop novel anti-tumor agents are planned to synthesize a series of symmetrical dimeric compounds containing two hetero aryl groups possessing at least one polar group joined through a linker chain and screen for their anti-tumor activity by using standard protocols.

It is evident from the literature review benzothiazoles possess wide range of biological & pharmacological properties such as anti microbial [10, 11], anticonvulsant [12], antihelminthic [13], antidiabetic [14], antimalarial [15], anti-inflammatory [16], antitubercular [17], antiviral [18] etc. Many derivatives of Benzothiazoles were found to be very Potent anti tumor activities [19- 21] and anticancer activities [22].

Literature survey indicated that Benzothiazole derivatives, Bis-intercalators have potent anti-tumor activity. It could be noted from the literature that benzothiazole bis-intercalators are not reported so far, keeping in view of the biological importance of DNA-intercalators, it all our evidence for the first time, to synthesize new N^1, N^3 Bis (6-chlorobenzo (d) thiazol-2-yl) 2 – substituted methyl malonamides by appropriate synthetic routes this will stand not only as a source for new biologically active compounds but also a model for molecular conjugation in the design of new anti-cancer drug.

Review of the literature indicated therapeutic uses of the Bis- DNA intercalators, possess strong antitumor properties. Hence, in present investigations we have attempted to design a novel series of compounds which contained N^1, N^3 Bis (6-chlorobenzo (d) thiazol-2-yl) 2 – substituted methyl malonamide molecular framework of possible Bis-DNA intercalators. Based on this hypothesis and the proposed model a new series of N^1, N^3 Bis (6-chlorobenzo (d) thiazol-2-yl) 2–substituted methyl malonamides were developed and screened for their possible anticancer activity.

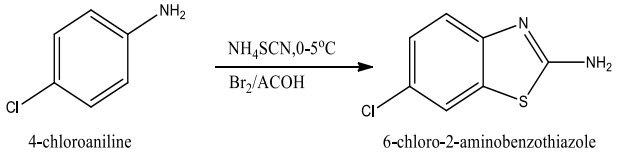
MATERIALS AND METHODS

Synthesis and analytical studies of the title compound were carried out using laboratory grade and analytical reagents as the case may be. Standard procedures or reported methods were followed with or without modification appropriately as and when required. Standard techniques like TLC were used to monitor reactions and to determine purity of the products. All the chemicals were obtained from SD fine chemicals Ltd. and the solvents were of laboratory grade. Each reaction of every step was monitored by using appropriate solvent system, which was selected by trail and error method. Precoated TLC plates (Silica gel GF 254) were obtained from E.Merck. All the melting points reported in this thesis were determined in open capillaries using SISCO melting point apparatus and KRION digital melting point apparatus expressed in °C and are uncorrected. The IR spectra of the compounds were recorded Perkin-Elmer 1720 FT-IR spectrometer (KBr pellets) expressed in cm^{-1}. The ^{1}H-NMR spectra were recorded on a Bruker AV III 400 spectrometer using DMS as the internal standard and were obtained from Central Facilities' for Research and Development (CFRD), Osmania University, Hyderabad.

Synthesis

Procedure for the synthesis of 6-chloro-2-aminobenzothiazole [24] (II)

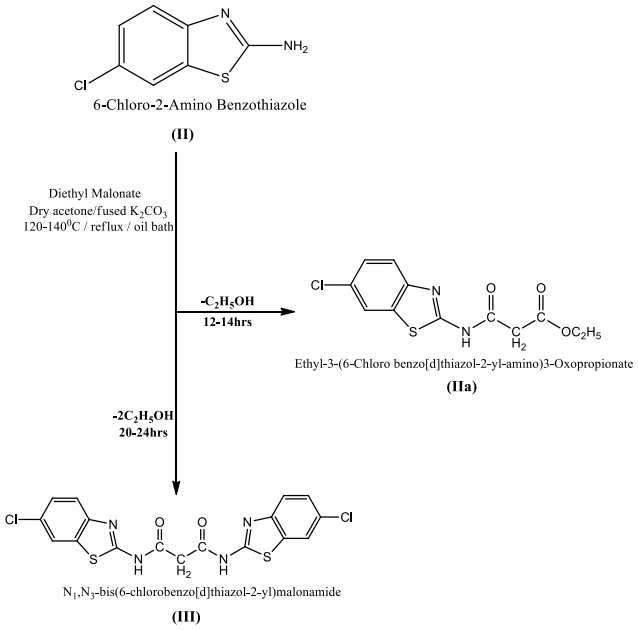
The p-substituted aniline (I, 0.1M) and ammonium thiocyanate (0.2M) in 150ml of glacial acetic acid were cooled in an ice bath and stirred mechanically. To the solution bromine (0.2M) in 25ml of glacial acetic acid was added drop wise at such a rate to keep the temperature below 10°C throughout the addition. The precipitate of the thiazole hydro bromide was collected dissolved in hot water & basified with saturated sodium solution. The free substituted benzothiazole was colleted washed with water &dried under vaccum recrystallization with 50% ethanol.



4-chloroaniline (I) 6-chloro-2-aminobenzothiazole (II)

Procedure for the preparation of ethyl 3-(6-chlorobenzo[d]thiazol-2-ylamino)-3-oxopropionate (IIa) and N^1, N^3 –bis(6-chlorobenzo[*d*]thiazol-2-yl)malonamide (III)

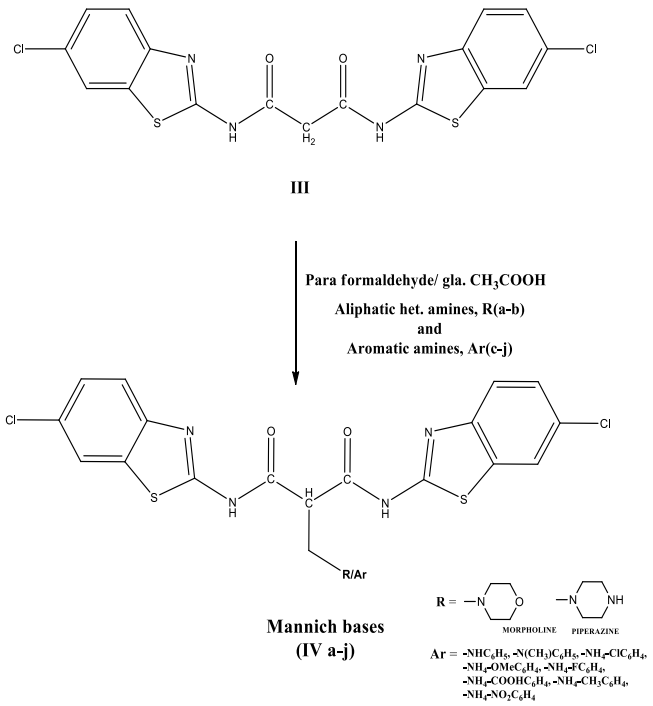
0.01 mole of II was dissolved in 30ml of dry acetone. To this diethyl malonate (0.01 mole) and 1.38 gm of freshly fused potassium carbonate (0.01 mole) were added and refluxed on an oil bath at 120-140°C for 12-14 hrs (reaction progress was monitored by TLC using (7:3 chloroform: ethylacetate as eluent). The reaction mixture was then poured onto the crushed ice and the precipitate was filtered and washed with cold water. The product was dried and purified by crystallization from aqueous alcohol to get monomer, IIa. Similarly III was prepared by the above method using 0.02 mole of II, 30ml dry acetone, 0.01 mole of diethyl malonate and 0.01 mole of fused potassium carbonate while refluxing on an oil bath at 120-140°C for 20-24 hrs to get the dimer, III.



Procedure for the preparation of mannich base IV (c)

A mixture of III (4.37gm, 0.1 mole), paraformaldehyde (0.36gm, 0.12 mole), aniline, (0.1 mole) in glacial acetic acid was refluxed on a mantle for a period of 5-8hrs and left overnight at room temperature. The solution was filtered to obtain crystalline compound and dried. The purification is effected by recrystallization by using methanol.

Similarly nine mannich bases (IVa, IVb, IVd, IVe, IVf, IVg, IVh, IVi and IVj) were also synthesized by employing above procedure and purified by using appropriate solvent.



Characterization

Compound II: 6-chloro-2-amino benzothiazole, $C_7H_5ClN_2S$, Molecular Weight: 184.5, Solubility: Methanol, R_f value : 0.71 (Chloroform: Ethylacetate) (7:3), Yield: 67.2 % , Melting point: 195-200^{0}C. IR (KBr, cm^{-1}): 3450(N-H str), 3076(Ar-H Str), 2926(C-H Str), 1445(Ar C=C Str), 1534(Cyclic C=N Str), 762(C-Cl Str), 645(C-S Str).

Compound II a: ethyl 3-(6-chlorobenzo[d]thiazol-2-ylamino)-3-oxopropanoate , Molecular formula: $C_{12}H_{11}N_2O_3SCl$, Molecular Weight: 298, Solubility: Methanol, R_f value : 0.60 (Chloroform: Ethyl acetate) (7:3), Yield: 62 %, Melting point: 170-174^o C. IR (KBr, cm^{-1}): 3457(N-H str), 3089(Ar-H Str), 2926(C-H Str), 1743(Ester C=O Str), 1633(Amide C=O Str), 1445(Ar C=C Str), 1534(Cyclic C=N Str), 762(C-Cl Str), 645(C-S Str).

Compound III: N^1,N^3 –bis(6-chlorobenzo[*d*]thiazol-2-yl) malanomide, Molecular formula: $C_{12}H_{11}Cl_2N_4O_2S_2$, Molecular Weight: 437.3, Solubility: Methanol, R_f value: 0.47 (Chloroform: Ethylacetate) (7:3) , Yield: 57.2 %, Melting point: 210-214^o C. IR (KBr, cm^{-1}): 3456(N-H str), 3093(Ar-H Str), 2926(C-H Str), 1632(Amide C=O Str), 1534(Cyclic C=N Str) 1445, (Ar C=C Str), 762(C-Cl Str),646(C-S Str). ^{1}H NMR Spectrum (DMSO,δ PPM): 3.1(2H,-CH_2),9.3(2H,-NH),7.2,7.6(6H,Ar-H).

Compound IVa: N^1,N^3 –bis (6-Chlorobenzo [*d*]thiazol-2-yl)-2-(morpholinomethyl) malonamide, Molecular formula: $C_{25}H_{19}Cl_2N_5O_3S_2$, Molecular Weight: 536.4, Solubility: Methanol, R_f value: 0.70 (Chloroform: Ethyl acetate) (7:3), Yield: 59.8 %, Melting point: 215-225^o C. IR (KBr, cm^{-1}): 3364(N-H str), 3176(Ar-H Str), 2969(methylene C- H Str), 1268(C-O str),2925(Amide C-H Str),1658(Amide C=O Str), 1549(Cyclic C=N Str),1598, 1445(Ar C=C Str) 1112(C-N Str), : 766(C-Cl Str),684(C-S Str). ^{1}H NMR Spectrum (DMSO,δ PPM): 7.56-7.69(6H,Ar-H), 3.65(8H,-CH2),9.3(2H,-CO-NH),3.25(1H,-CH),3.45(2H,-CH2).

Compound IVb: N^1,N^3 –bis(6-Chlorobenzo[*d*]thiazol-2-yl)-2-(piperazin -1-ylmethyl) malonamide, Molecular formula: $C_{22}H_{20}Cl_2N_6O_2S_2$, Molecular Weight: 535.4, Solubility: Methanol, R_f value: 0.57(Chloroform: Ethylacetate) (7:3), Yield: 68 %, Melting point: 247-256^o C. IR (KBr, cm^{-1}): 3416(N-H str), 3180(Ar-H Str),2970(methylene C-H Str), 2924(Amide C-H Str),1656(Amide C=O Str), 1541(Cyclic C=N Str),1599, 1445(Ar C=C Str),1098(C-N Str), 764(C-Cl Str), 688(C-S Str).^{1}H NMR Spectrum (DMSO,δ PPM) :7.35-7.89(6H,-Ar-H),2.37-2.65(8H,-CH2),1.90(1H,Cyclic –NH),2.95(2H,-CH2),9.15(2H,-NH),3.6(1H,-CH).

Compound IVc: N^1,N^3 –bis(6-Chlorobenzo[*d*]thiazol-2-yl)-2-((phenylamino)methyl)malonamide, Molecular

formula: $C_{24}H_{17}Cl_2N_5O_2S_2$, Molecular Weight: 542.4, Solubility: Methanol, R_f value: 0.53 (Chloroform: Ethylacetate) (7:3), Yield: 52 %, Melting point: 217-220^o C, IR (KBr, cm^{-1}): 3263(N-H str), 3024(Ar-H Str),2977(methylene C-H Str), 2928(Amide C-H Str),1697(Amide C=O Str), 1534(Cyclic C=N Str), 1597- 1445(Ar C=C Str, 776(C-Cl Str), 682(C-S Str). ^{1}H NMR Spectrum (DMSO,δ PPM): 3.4(2H,-CH_2),3.8(1H,-CH),8.1(2H,-CONH), 7.2- 7.7(11H,-Ar-H), 3..92(1H,-NH).

Compound IVd: N^1,N^3 –bis(6-Chlorobenzo[*d*]thiazol-2-yl)-2-((methyl(phenyl)amino)methyl)malonamide, Molecular formula: $C_{25}H_{19}Cl_2N_5O_2S_2$, Molecular Weight: 556.4, Solubility: Methanol, R_f value: 0.47 (Chloroform: Ethylacetate) (7:3), Yield: 45.3 %, Melting point: 228-232^o C. IR (KBr, cm^{-1}): 3268(N-H str), 3075(Ar-H Str),2977(methylene C-H Str),2930(Amide C-H Str),1696(Amide C=O Str), 1548(Cyclic C=N Str),1598- 1444(Ar C=C Str, 766(C-Cl Str), 682(C-S Str). ^{1}H NMR Spectrum (DMSO,δ PPM): 7.25-7.71(10H,-Ar-H),8.2(2H,CONH)2.75(3H,-CH3),3.67(2H,-CH2),3.9(1H,-CH).

Compound IVe: N^1,N^3 –bis(6-Chlorobenzo[*d*]thiazol-2-yl)-2-((4-chlorophenylamino)methyl)malonamide, Molecular formula: $C_{24}H_{16}Cl_3N_5O_2S_2$, Molecular Weight: 576, Solubility: Methanol, R_f value: 0.45 (Chloroform: Ethylacetate) (7:3), Yield: 54.2 %, Melting point: 238-242^o C. IR (KBr, cm^{-1}): 3219(N-H str), 3074(Ar-H Str),2979(methylene C-H Str) 2929(Amide C-H Str),1698(Amide C=O Str), 1548(Cyclic C=N Str), 1597, 1443(Ar C=C Str), 1421(Amide C-N Str),767(C-Cl Str),684(C-S Str). ^{1}H NMR Spectrum (DMSO,δ PPM): 3.6(2H,-CH_2),3.9(1H,-CH),7.2-7.69(10H,- Ar-H),8.62(2H,-CONH),3.9(1H,-NH).

Compound IVf: N^1,N^3 –bis(6-Chlorobenzo[*d*]thiazol-2-yl)-2-((4-methoxyphenylamino)methyl)malonamide, Molecular formula: $C_{25}H_{19}Cl_2N_5O_3S_2$, Molecular Weight: 572.8, Solubility: Methanol, R_f value: 0.53 (Chloroform: Ethylacetate) (7:3), Yield: 48 %, Melting point: 234-239^o C. IR (KBr, cm^{-1}): 3365(N-H str), 3124(Ar-H Str),3002(Ar C-H Str), 2963(Methyl C-H Str), 2935(Amide C-H Str), 1661(Amide C= Str), 1554(Cyclic C=N Str), 1598, 1445(Ar C=C Str), 767(C-Cl Str), 684(C-S Str). ^{1}H NMR Spectrum (DMSO,δ PPM):8.62(2H,-NH-C-O),6.51-7.51(10H,-Ar-H),3.92(1H,-NH),3.61(3H,-CH3),3.4(2H,-CH2), 3.6(1H,-CH).

Compound IVg: N^1,N^3 –bis(6-Chlorobenzo[*d*]thiazol-2-yl)-2-((4-fluorophenylamino)methyl)malonamide, Molecular formula: $C_{24}H_{16}Cl_2FN_5O_2S_2$, Molecular Weight: 560.4, Solubility: Methanol, R_f value: 0.49 (Chloroform: Ethylacetate) (7:3), Yield: 59 %, Melting point: 210-215^o C. IR (KBr, cm^{-1}): 3271(N-H str), 3074(Ar-H Str), 2977(methylene C-H Str) 2930(Amide C-H Str), 1612(Amide C=O Str), 1537(Cyclic C=N Str), 1598, 1447(Ar C=C Str), 1053(C-F Str), 766(C-Cl Str). ^{1}H NMR Spectrum (DMSO,δ PPM): 8.62(2H,-C0NH),6.51-7.5(10H,-Ar-H),3.9(1H,-NH),3.42(2H,-CH2),3.6(1H,-CH).

Compound IVh: 4–(3-(6-Chlorobenzo[*d*]thiazol-2-ylamino)-2-(6-chlorobenzo[*d*]thiazol-2-ylcarbamoyl)-3-oxopropylamino) benzoic acid, Molecular formula: $C_{25}H_{17}Cl_2N_5O_4S_2$, Molecular Weight: 586.4, Solubility: Methanol, R_f value:0.58 (Chloroform: Ethylacetate) (7:3), Yield: 47.3 %, Melting point: 225-235^o C. IR (KBr, cm^{-1}): 3304(N-H str), 3112(Ar-H Str),2970(methylene C-H Str) Str),2963(Methylene, C-H Str), 2935(Amide C-H Str), 1661(Amide C=O Str),1554(Cyclic C=N, 1598, 1445(Ar C=C Str),767(C-Cl Str), 684(C-S Str). ^{1}H NMR Spectrum (DMSO,δ PPM):8.6(2H,-NH-C-O),11.2(1H,-O-H),6.5-7.5(10H,-Ar-H),3.25(2H,-CH2),3.6(1H,-CH),3.9(1H,-NH).

Compound IVi: N^1,N^3 –bis(6-Chlorobenzo[*d*]thiazol-2-yl)-2-((*p*-tolylamino)methyl)malonamide, Molecular formula: $C_{25}H_{19}Cl_2N_5O_2S_2$, Molecular Weight: 556.4, Solubility: Methanol, R_f value: 0.47 (Chloroform: Ethylacetate) (7:3), Yield: 62 %, Melting point: 215-222^o C. IR (KBr, cm^{-1}): 3364(N-H str), 3119(Ar-H Str),2974(methylene C-H Str), 2945(Methyl C-H Str),2920(Amide C-H Str), 1663(Amide C=O Str), 1552(Cyclic C=N Str1599, 1445(Ar C=C Str), 766(C-Cl Str), 684(C-S Str). ^{1}H NMR Spectrum (DMSO,δ PPM):2.56(3H,-CH3),3.6(3H,-CH2&CH),3.9(1H,-NH),8.25(2H,-NH-C-O),7.2-7.82(10H,-Ar-H).

Compound IVj: N^1,N^3 –bis(6-Chlorobenzo[*d*]thiazol-2-yl)-2-((4-nitrophenylamino)methyl)malonamide, Molecular formula: $C_{24}H_{17}Cl_2N_5O_4S_2$, Molecular Weight: 586.4, Solubility: Methanol, R_f value: .52 (Chloroform: Ethylacetate) (7:3), Yield: 43.5% , Melting point: 228^o C. IR (KBr, cm^{-1}): 3361(N-H str), 3084(Ar-H Str), 2970(methylene C-H Str) 2935(Amide C-H Str), 1661(Amide C=O Str)1538(Cyclic C=N Str), 1598, 1446(Ar C=C Str), 1372(Ar-N=0 Str), 766(C-Cl Str), 690(C-S Str). ^{1}H NMR Spectrum (DMSO,δ PPM):8.15(2H,-NH-C-O),7.2-7.82(10H,-Ar-H),3.45(2H,-CH2),3.81(1H,NH),3.6(1H,-CH).

BIOLOGICAL EVALUATION

All the ten title compounds including monomer dimer analogues were subjected for screening by following various standard in vitro methods.

Wheat rootlet growth inhibition assay

[25] This model was used to determine the effect of the compounds on growth of wheat roots. It serves as a preliminary measure to identify the inhibitory effect of a drug on rapidly dividing normal cell. This assay can help in identifying the inhibitory effect on cell growth.

Antimitotic studies using onion root model

It was carriedout by standard procedures [26s]. Antimitotic study of all the test compounds was performed using standard onion root tip model to determine the effect of the drug on mitosis and cell division at to different concentration of 10 and 20µg/ml.

RESULTS

Wheat rootlet growth inhibition assay

Except three test compounds (below 20%), all others showed moderate inhibitory effect on growth of wheat roots.Compounds IVg (Fluoro) and IVe (Chloro) were most active with percentage inhibition of 50 and 54 respectively. Interestingly out of ten mannich compounds possessed better inhibitory effect as compared to the dimer. The monomeric compound IIa (Monomer) dimer compound IVh (PABA) possessed very low activity.

Table1: Wheat Rootlet Inhibition study of N^1,N^3-bis(6-chlorobenzo[d]thiazole-2-yl)-2-((phenylamino) methyl) malonamides.

Sl. No.	Wheat rootlet growth(mm)	Root growth Inhibition (%)*
Control	4.6	-
IIa (Monomer)	3.8	17.4
III (Dimer)	2.8	39.4
IVa	2.8	39.1
IVb	3.2	30.5
IVc	3.5	23.9
IVd	4.1	10.9
IVe	2.3	50
IVf	3.3	28.3
IVg	2.1	54.4
IVh	4.0	13.1
IVi	3.6	21.8
IVj	2.6	43.5

Average number of 30 seedlings

Table 2: Antimitotic Activity of N^1,N^3-bis(6-chlorobenzo[d]thiazole-2-yl)-2-((phenylamino)methyl)malonamides.

Group No.	Concentration (µg/ml)	Cells counted Non-dividing (a)	Dividing (b)	Percentage of dividing cells	Percentage Inhibition of mitosis
Control-I		66	74	100	
Control-II		48	62	100	
IIa (Monomer)	10	32	64	66.66	33.34
	20	56	38	50.42	49.58
III (Dimer)	10	38	66	63.46	36.53
	20	52	42	31.68	58.31
IVa	10	46	68	59.64	43.36
	20	52	32	38.09	61.91
IVb	10	40	72	60	40.00
	20	58	46	44.23	55.71
IVc	10	42	56	57.14	42.85
	20	76	32	29.62	70.37
IVd	10	54	56	50.9	49.09
	20	52	38	42.22	57.77
IVe	10	52	46	46.93	53.06
	20	28	24	23.26	70.73
IVf	10	52	54	50.94	49.05
	20	58	56	49.92	50.68
IVg	10	32	64	66.66	33.34
	20	62	27	30.33	69.66
IVh	10	58	48	45.28	54.71
	20	42	26	38.23	61.76
IVi	10	44	52	54.11	45.83
	20	62	72	34.04	65.95
IVj	10	56	48	46.15	53.85
	20	62	22	26.19	73.81

Antimitotic studies using onion root model

Results clearly indicate that all the compounds possess significant antimitotic activity at both the test concentrations. In the series compounds IVj (Nitro) was found to be more potent with 54 and 74% inhibition of mitosis at 10 and 20µg/ml respectively. It was followed by the compounds IVc (Aniline) & IVe (Chloro) which showed about 70% inhibition at 20µg/ml concentration.

From the results, it can be observed that the anti mitotic activity of the test compounds was dose dependent. In most cases the activity increased by more than 20% when the test concentrations was doubled. However such strong increase in activity was not observed with monomeric compounds to dimer and mannich bases. In general all thus test compounds possess significant antimitotc activity.

DISCUSSION:

In this study 10 new title compounds (IVa-j) were synthesized, purified and characterized by appropriate standard techniques. All the intermediates and title compounds were obtained in satisfactory yields and purity and were found to be stable.

In vitro biological screening of the final title compounds (mannich bases), a monomer (IIa) and benzothiazole dimer (III) was performed by employing three different preliminary models i.e. wheat rootlet growth inhibitory studies, antimitotic studies using onion root tip method. In wheat rootlet growth inhibition assay Compounds IVg (Fluoro) and IVe (Chloro) were most active with percentage inhibition of 50 and 54 respectively. Compounds IVg (Fluoro) and IVe (Chloro) were most active with percentage inhibition of 50 and 54 respectively. In antimitotic studies using onion root model the series compounds IVj (Nitro) was found to be more potent with 54 and 74% inhibition of mitosis at 10 and 20µg/ml respectively. It was followed by the compounds IVc (Aniline) & IVe (Chloro) which showed about 70% inhibition at 20µg/ml concentration.

In the present investigation, based on our hypothesis, we have developed a novel series of N^1, N^3 Bis (6-chlorobenzo (d) thiazol-2-yl) 2 –substituted methyl malonamides as a new class of possible antitumor agents, which act by binding in the minor grove of DNA. This conclusive study will serve as an excellent platform for development of new drugs to be useful for cancer chemotherapy in future.

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