EVALUATION OF THE FUNGITOXIC PROPERTIES OF BENZOYL PROTECTED HETEROCYCLIC THIOMALTOSIDES AGAINST PLANT PATHOGENS

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Abstract:

Several 1-Aryl-2-*o*-tolyl-5-*S*-hepta-*O*-benzoyl maltosyl-1, 2-dihydro-1, 2, 4-triazol-3-ones II (a-f) and 4-Aryl-5-phenylimino-3-*S*-hepta-*O*-benzoyl maltosyl-1, 2, 4-thiadiazolines III (a-f) were synthesized by the interaction of *S*-Hepta-*O*-benzoyl maltosyl-1-arylisothiocarbamides I(a-f)with *o*-tolyl isocyanate followed by oxidative cyclization and interaction of *S*-Hepta-*O*-benzoyl maltosyl-1-arylisothiocarbamides I(a-f) with the *S*-Chloro-*N*-phenyl isothocarbamoyl chloride respectively. In the present investigation fungitoxic activities of these new S-maltosides against plant pathogenic fungi such as *Drechslera tetramera, Curvularia eragrostidis, Bipolaris sorokiniana*, *Xamthomonas campestris* and *Rhizopus species* were studied *In vitro* by poisoned food technique. Percent inhibition of these thiomaltosides was found to be ranging from 3.48 to 54.83%.

Key words: 1, 2, 4-triazol-3-ones, 1, 2, 4-thiadiazolines, isothioburets, isothocarbamoyl chloride, plant pathogen, fungitoxic properties.

Introduction:-

The resistance towards available drugs is rapidly becoming a major worldwide problem. The need to design new compounds to deal with this resistance has become one of the most important areas of research today. The nitrogen and sulfur containing 5/6 membered heterocyclic compounds have special[1, 2] biological importance. 1, 2, 4-triazoles are an important class of heterocycles, and have been the subject of great interest due to their pharmacological properties[3-5]. Very promising therapeutic applications have been obtained using the 1, 2, 4-triazole system. There are a number of drugs containing 1, 2, 4-triazole nucleus, such as itraconazole, flucanazole and voriconazole (antifungal), that have been used for the treatment of fungal infections[6-8].

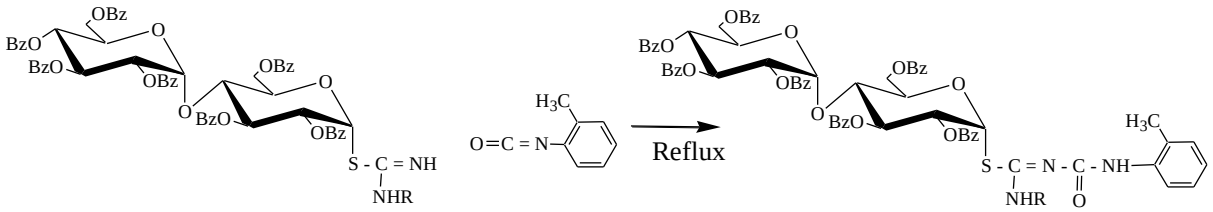
Chemistry of *S*-Chloro-*N*-phenyl isothocarbamoyl chloride with special utility in the synthesis of nitrogen and sulfur containing heterocyclic compounds has been exhaustively investigated by number of chemists[9-11]. However, there is an increasing resistance to these drugs. Moreover, some of azole derivatives used as common antibiotics posses a toxic effect on humans as well as their antimicrobial effects.

In view of applications of these compounds in medicine, agrochemical industry and detergents it appeared interesting to carry out synthesis of some novel thiomaltosides and study their fungitoxic activity against fungal phytopathogens.

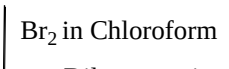
Fungi rank second only to insects as a cause of plant disease, which result in heavy loss of plant products. *Drechslera tetramera*, a pathogen causes seed born disease of Wheat and Sorghum, *Curvularia eragrostidis* is a pathogen of Crabgrass (Digiteria sanguinalis), *Bipolaris.sorokiniana*, a pathogen causes spot blotch and common root rot of wheat and barley crops, *Rhizopus sp*., a pathogen causes the most common disease on Wheat viz., were chosen for the investigation against the S-maltosides.

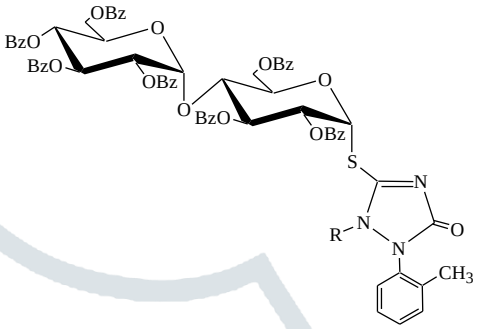
Schemes

Scheme I



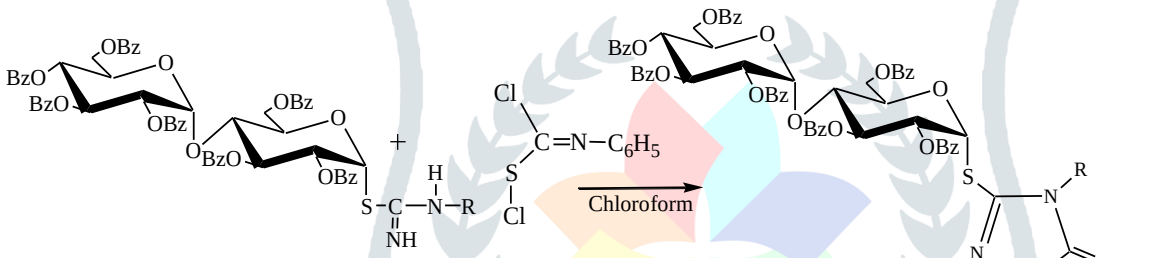
S-Hepta-*O*-benzoyl maltosyl-1-arylisothiocarbamides I (a-f)





1-Aryl-2-*o*-tolyl-5-*S*-hepta-*O*-benzoyl maltosyl-1, 2-dihydro-1, 2, 4-triazol-3-ones II(a-f)

Scheme II



S-Hepta-*O*-benzoyl maltosyl-1-arylisothiocarbamides I (a-f)

4-Aryl-5-phenylimino-3-*S*-Hepta-*O*-benzoyl maltosyl-1, 2, 4-thiadiazolines III (a-f)

Where,

Bz = COC_6H_5

R = a) Phenyl, b) *o*-Cl Phenyl, c) *m*-Cl Phenyl, d) *p*-Cl Phenyl, e) *o*-tolyl, f) *p*-tolyl.

Experimental:-

All the melting points recorded were found to be uncorrected. The structures of newly synthesized compound were confirmed on the basis of elemental and IR, ^{1}H NMR and Mass spectral analysis[12-13]. IR spectra were recorded in KBr on a FTIR Perkin-Elmer (4000-450cm^{-1}) spectrophotometer and in KBr disks on SHIMADZU IR affinity-1 FTIR spectrometer. ^{1}H NMR spectra are run on Brucker DRX-300 instrument operating at 300 MHz using $CDCl_3$ solution with TMS at internal standard and Mass spectra on JEOL-AccuTof JMS-T100LC Mass spectrometer.

All the compounds have been screened for antifungal activity against phytopathogenic fungi using poisoned food technique by calculating the percent inhibition. The percent inhibition of fungal growth by each compound was assessed by poisoned food technique[14] on PDA containing 20mg/ml solution of different compounds in DMSO (dimethyl sulphoxide). Bavistin is used as standard. The compounds were screened for antifungal activity against *Drechslera tetramera, Curvularia ergostidies, Bipolaris sorokenia, Rhizopus sp.* and *Xamthomonas campestris* in potato dextrose agar medium. These sterilized agar media containing compounds were poured in to Petri dishes and allowed to solidify. Fungal disc of 5mm diameter was kept at center of each petriplate containing poisoned medium. The petriplate of PDA without chemical compounds were simultaneously inoculated with fungal disc. The percent inhibitions of fungal growth were calculated using vicent formula and are shown in Table 3 and Table 4.

$$I = \frac{C - T \times 100}{C}$$

Where, I – Percent inhibition of growth.

C – Growth of fungus in control.

T – Growth of fungus in tretement.

Material and Method:

Synthesis of* 1-Aryl-2-*o*-tolyl-5-*S*-hepta-*O*-benzoyl maltosyl-1, 2-dihydro-1, 2, 4-triazol-3-ones *II (a-f)

S-Hepta-*O*-benzoyl maltosyl-1-aryl-5-*o*-tolyl-2-isothiobiurets were synthesized by the interaction of *S*-Hepta-*O*-benzoyl maltosyl-1-arylisothiocarbamides I(a-f) with *o*-tolyl isocyanate followed by oxidative cyclization. *S*-Hepta-*O*-benzoyl maltosyl-1-aryl-5-*o*-tolyl-2-isothiobiurets were made into a paste with chloroform and to it was added bromine in chloroform (20% bromine solution in chloroform, v/v) drop by drop with stirring. The bromine in chloroform was added till orange red sticky masses were

obtained. It was then allowed to stand for 5-6 hr. The sticky masses were washed several times with petroleum ether (60-80ºC) and then dissolved in ethanol and was basified by using dil. ammonia the products were isolated as free bases. It were purified by ethanol-water.

Synthesis of* 4-Aryl-5-phenylimino-3-*S*-hepta-*O*-benzoyl maltosyl-1, 2, 4-thiadiazolines *III (a-f)

Mixtures of *N*-Phenyl-*S*-Chloro isothiocarbomoyl chloride in chloroform and *S*-Hepta-*O*-benzoyl maltosyl-1-phenylisothiocarbamide in chloroform was refluxed for 3 hr. The resultant solution was allowed to stand for several hours but no solid was separated. The chloroform was distilled off. The sticky mass thus obtained was triturated several times with petroleum ether (60-80ºC). It was furnished a granular solid. It was purified by ethanol-water and solid was obtained. Condensation of *S*-hepta-*O*-benzoyl maltosyl-1-arylisothiocarbamides II (a-f) with phenyl isothiocyanate in benzene was carried out for 9 h. The benzene was distilled off. The sticky masses obtained were triturated several times with petroleum ether (60-80 ºC) furnishes as granular solid. It was purified by ethanol-water.

The IR, ^{1}H NMR and Mass spectral analysis (Table 1 and Table 2) indicates the products and designs the structures of the compounds.

Table 1:- Characterization of Several 1-Aryl-2-*o*-tolyl-5-*S*-hepta-*O*-benzoyl maltosyl-1, 2-dihydro-1, 2, 4-triazol-3-ones ***II (a-f)***

Comp.	m.p. (ºC)	Mol. Formula	IR(KBr cm^{-1})	^{1}H NMR ($CDCl_3$, ppm)	Mass (m/z)
IIa	174	$C_{76}H_{61}O_{18}N_3S$	3062 (Ar C-H), 2974 (Ali C-H) 1730 (C=O), 1600 (C=N), 1450 (C-N), 1271 (C-O), 1095, 1026 and 937 (Characteristics of maltose), 709 (C-S),	δ 8.04-7.25 (44H, m, Ar. proton), 6.2-3.7 (14H, m, maltosyl proton), 1.17 (3H, s, CH_3)	1335 (M^+), 1199, 1053, 931, 579, 475
IIb	139	$C_{76}H_{60}O_{18}N_3SCl$	-	-	-
IIc	141	$C_{76}H_{60}O_{18}N_3SCl$	-	-	-
IId	132	$C_{76}H_{60}O_{18}N_3SCl$	3062 (Ar C-H), 2960 (Ali C-H) 1730 (C=O), 1600 (C=N), 1452 (C-N), 1269 (C-O), 1093, 1026 and 937 (Characteristics of maltose), 709 (C-S), 601 (C-Cl)	δ 8.12-7.18 (43H, m, Ar. proton), 6.16-3.87(14H, m, maltosylated proton), 1.88 (3H, s, CH_3)	1371 (M^+), 1221, 1139, 1053, 948, 931, 579
IIe	166	$C_{77}H_{63}O_{18}N_3S$	-	-	-
IIf	142	$C_{77}H_{63}O_{18}N_3S$	3062 (Ar C-H), 2960 (Ali C-H) 1730 (C=O), 1452 (C-N), 1269 (C-O), 1093, 1041and 937 (Characteristics of maltose), 709 (C-S)	δ 8.1-7.1 (43H, m, Ar. proton), 5.9-3.8 (14H, m, maltosylated proton), 1.8 (3H, s, CH_3), 1.6 (3H, s, CH_3)	1349 (M^+), 1273, 1053, 931, 794, 579

Table 2:- Characterization of Several 4-Aryl-5-phenylimino-3-*S*-hepta-*O*-benzoyl maltosyl-1, 2, 4-thiadiazolines *III (a-f)*

Comp.	m.p. (ºC)	Mol. Formula	IR(KBr cm^{-1})	^{1}H NMR ($CDCl_3$, ppm)	Mass (m/z)
IIIa	132-135	$C_{75}H_{59}O_{17}N_3S_2$	3062 (Ar C-H), 2962 (Ali C-H), 1730 (C=O), 1600 (C=N), 1452 (C-N), 1271 (C-O), 1095, 1026and 937 (Characteristics of maltose), 709 (C-S)	δ 8.1-7.15 (45H, m, Ar. proton), 6.24-3.82 (14H, m, maltosylated proton)	1338 (M+ not located), 1276, 1234, 931, 579
IIIb	134-140	$C_{75}H_{58}O_{17}N_3S_2Cl$	3062 (Ar C-H), 2962 (Ali C-H) 1730 (C=O), 1600 (C=N), 1450 (C-N), 1269 (C-O), 1095, 1026and 937 (Characteristics of maltose), 709 (C-S)	δ 8.095-7.14 (44H, m, Ar. proton), 6.20-3.81(14H, m, maltosylated proton)	1371 (M+ not located), 1309, 931, 579
IIIc	125	$C_{75}H_{58}O_{17}N_3S_2Cl$	-	-	-
IIId	130	$C_{75}H_{58}O_{17}N_3S_2Cl$	-	-	-
IIIe	140-145	$C_{76}H_{61}O_{17}N_3S_2$	3062 (Ar C-H), 2972 (Ali C-H) 1730 (C=O), 1452 (C-N), 1269 (C-O), 1095, 1026 and 937 (Characteristics of maltose), 709 (C-S)	δ 5.731 (2H, s, 2N-H), 8.094-7.240 (44H, m, Ar. proton), 4.832-4.285 (14H, m, maltosylated proton), 2.311 (3H, s, CH_3)	1353 (M+ not located), 1290, 1198, 1157, 1031, 990, 927, 579
IIIf	143	$C_{76}H_{61}O_{17}N_3S_2$	-	-	-

Table 3: Percent inhibition of 1-Aryl-2-*o*-tolyl-5-*S*-hepta-*O*-benzoyl maltosyl-1, 2-dihydro-1, 2, 4-triazol-3-ones *II (a-f)* against phytopathogenic fungi (by poisoned food technique).

Sr. No.	Compds	Phytopathogens				
		Drechslera tetramera	*Curvularia ergrostidis*	*Bipolaris sorokiniana*	*Rhizopus sp.*	*Xanthomonas campestris*
1	IIa	15.38	24.24	44.44	34.48	32.26
2	IIb	19.23	24.24	29.62	37.93	25.80
3	IIc	26.92	30.30	22.22	37.93	54.83
4	IId	30.77	33.33	33.33	41.38	32.26
5	IIe	34.61	30.30	7.41	31.03	22.58
6	IIf	42.31	36.36	14.81	41.38	22.58
7	Bavistin	85.00	57.15	33.33	66.45	51.61

Note: Simultaneously separate controls are used without compounds.

Table 4: Percent inhibition of 4-Aryl-5-phenylimino-3-*S*-hepta-*O*-benzoyl maltosyl-1, 2, 4-thiadiazolines *III (a-f)* against phytopathogenic fungi (by poisoned food technique).

Sr. No.	Compds	Phytopathogens				
		Drechslera tetramera	*Curvularia ergrostidis*	*Bipolaris sorokiniana*	*Rhizopus sp.*	*Xanthomonas campestris*
1	IIIa	26.92	39.39	18.52	41.38	25.80
2	IIIb	30.77	33.33	25.92	37.93	35.48
3	IIIc	23.08	33.33	28.92	37.93	32.26
4	IIId	19.23	39.39	18.52	48.15	29.03
5	IIIe	19.23	27.27	40.74	34.48	45.16
6	IIIf	15.38	30.30	33.33	3.48	41.93
7	Bavistin	85.00	57.15	33.33	66.45	51.61

Note: Simultaneously separate controls are used without compounds.

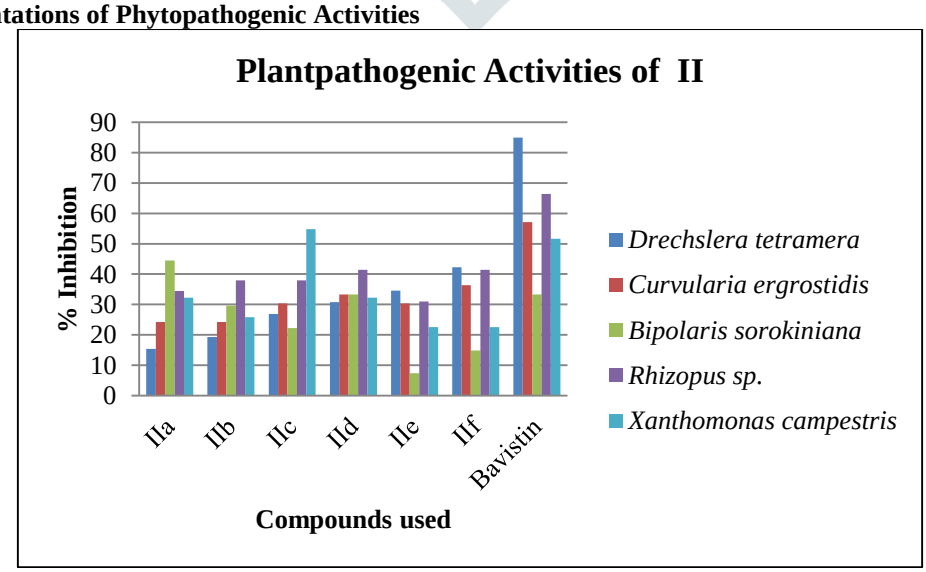
Result and Discussion:

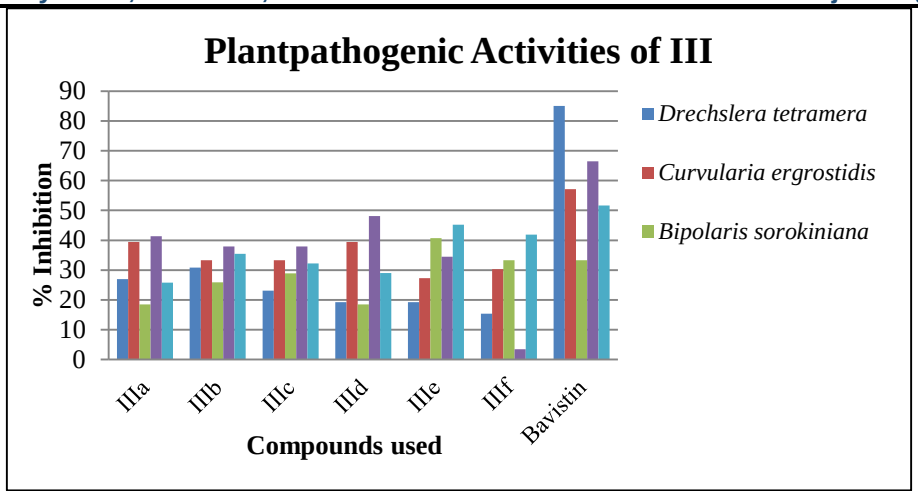
It has been observed that some of these compounds exhibited interesting microbial activities. IIe, IIf, IIIa and IIIb exhibited most significant activity against *Drechslera tetramera.* IId, IIf, IIIa and IIId exhibited most significant activity against *Curvularia ergostidies,* IIa, IId, IIIe and IIIf exhibited most significant activity against *Bipolaris sorokenia,* IId, IIf, IIIa and IIId exhibited most significant activity against *Rhizopus sp.* and IIa, IIc, IIIe and IIf exhibited most significant against *Xamthomonas campestris* respectively. All other compounds exhibited low to moderate activity.

Among the four test pathogens, *Bipolaris sorokenia* appeared less sensitive to all test compounds including bavistin whereas *Drechslera tetramera* appeared more sensitive to all test compounds including bavistin.

The results of antifungal activity are tabulated in Table 3 and Table 4.

Graphical Representations of Phytopathogenic Activities





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