IN VITRO CYTOTOXIC POTENCY OF BIS-THIAZOLIDINONES AND ITS DERIVATIVES AGAINST BRINE SHRIMP

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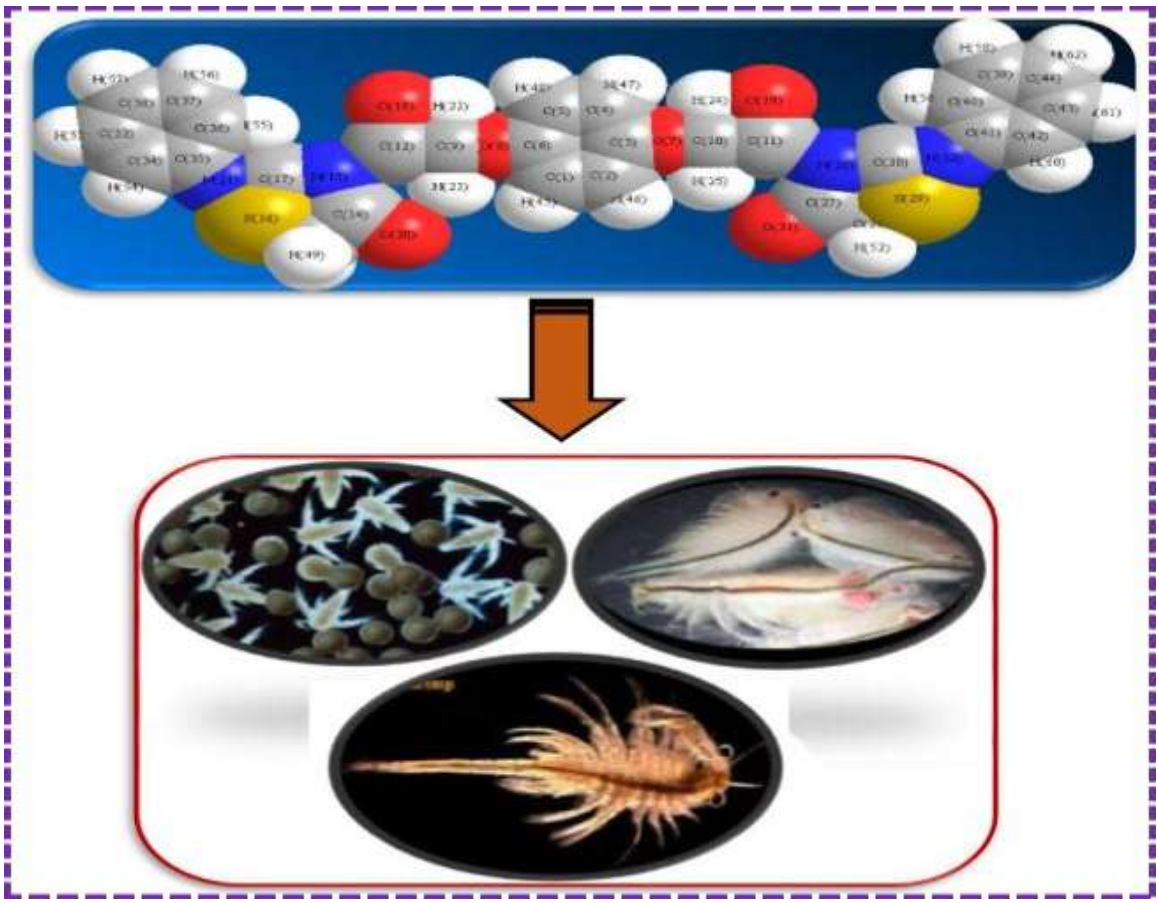
ABSTRACT

In the present manuscript the Brine shrimp bioassay lethality was carried out for in vitro cytotoxic measurements of synthesized Bis-Thiazolidinones and its derivatives compounds against Artemia salina. The LD50 values were calculated after probit transformation of the resulting mortality data. All the synthesized compounds are found to be promising bioactive compounds by registering their LD50 of 1.030 X 10^{-4}, 1.179 X 10^{-4} and 1.472 X 10^{-4} which correspond to compounds 1, 2 and 3 respectively.

GRAPHICAL ABSTRACT:

In vitro cytotoxic potency of Bis-Thiazolidinones and its derivatives against Brine Shrimp.



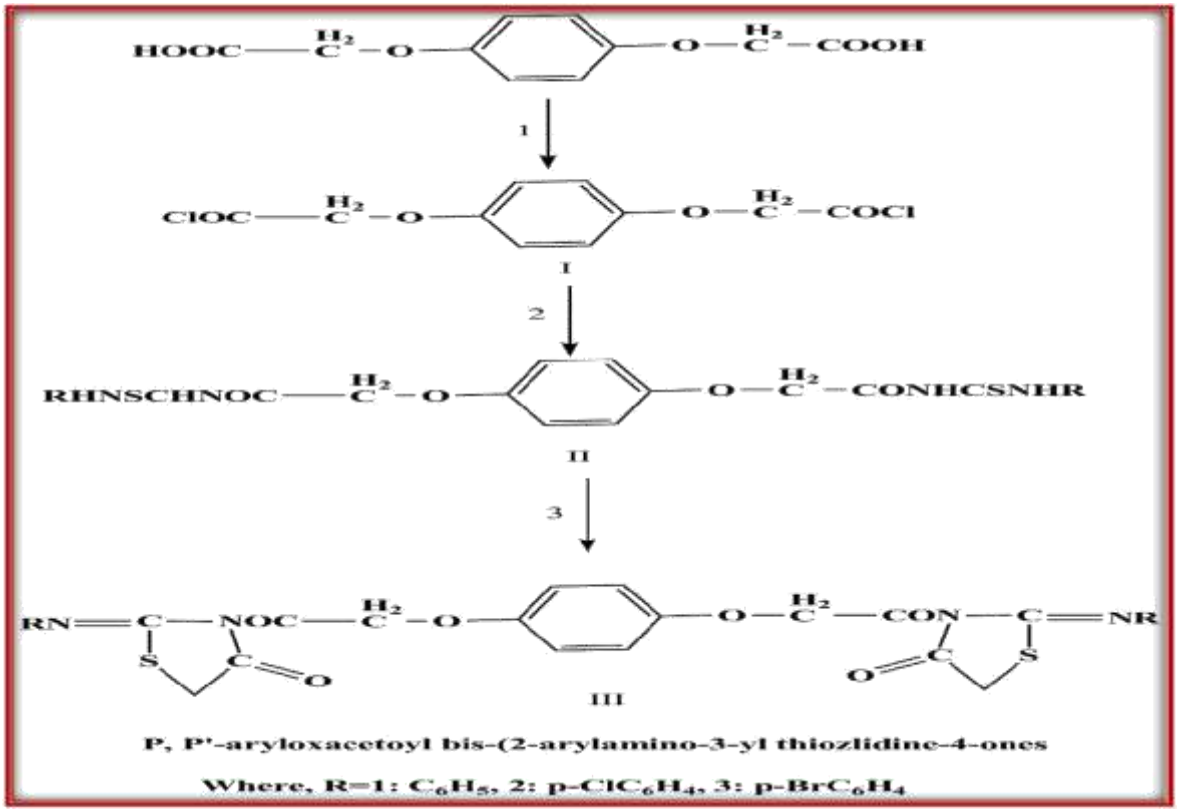
1. INTRODUCTION:

Sulfur containing heterocycles have been under investigation for a long time because of their important medicinal properties. Among these type of molecules, 4-thiazolidinones have been shown to have various important biological activities such as antimicrobial, antiviral, antituberculostatic, anticancer, anti-inflammatory and analgesic properties [1].

Thiazolidinones are derivatives of thiazolidine and are significant group of heterocyclic compounds. There are three types of thiazolidinones based on 2nd, 4th and 5th position of carbonyl group. The most significant one is thiozolidinone with carbonyl group at 4th position, which is also known as 4-thiazolidinone or 4-oxo-thiazolidine. Thiazolidin- 4-ones and their derivatives have attracted much attention due to diverse biological activities such as antidiabetic, antihistaminic, Ca^{2+-} channel blocker, anti-platelet activating factor6, antidiarrheal, platelet activating factor (PAF) antagonist, cardioprotective, anti-ischemic, anticancer activities [2].

Hence, the available literature survey on diverse application of Bis-Thiazolidinones and its derivatives in extension of our enduring work, the present investigation deals with the study of cytotoxic activity evaluation of p, p'-aryloxacetoyl bis-(2-arylamino-3-yl thiozlidine-4-ones against brine shrimp (Artemia salina) to ensure their Cytotoxic potency.

The synthesis, spectral characterization such as IR, ^1H NMR and Mass along with Antimicrobial activities of p, p'-aryloxacetoyl bis-(2-arylamino-3-yl thiozlidine-4-ones were already reported in our earlier protocol as shown in Scheme 1 and elemental analysis data in



The antimicrobial activity results were shown as supplementary Fig S1 and S2.

Scheme 1: Synthesis of Bis-Thiazolidinones and its derivatives.

Table 1: Elemental analysis data of p, p'-aryloxacetoyl bis-(2-arylamino-3-yl thiozlidine-

4-ones (III, 1-3).

Compd. No	R	Molecular formal	M.P (º C)	C	H	N	O	S	Cl	Br
1	C6H5	C28H22N4O6S2	135	58.52 (58.40)	3.86 (3.71)	9.45 (9.75)	16.71 (16.52)	11.18 (11.14)	-	-
2	p-ClC6H4	C28H20Cl2N4O6S2	151	52.26 (52.02)	3.13 (3.04)	8.41 (8.72)	14.92 (14.69)	9.53 (9.96)	11.02 (10.86)	-
3	p-BrC6H4	C28H20Br2N4O6S2	115	45.92 (45.81)	2.75 (2.57)	7.94 (7.65)	13.11 (13.02)	8.92 (8.74)	-	21.82 (21.67)

2. RESULT AND DISCUSSION:

2.1. Cytotoxic activity method:

The synthesized compounds were screened for their cytotoxicity by brine-shrimp bioassay lethality method using standard Mayer protocol [4]. The brine shrimp, Artemia salina, was used as a convenient monitor for the screening. Since bioactive compounds are often toxic to shrimp larvae. Hence, shrimp larvae have been extensively used as rapid and simple preliminary test for cytotoxicity [5, 6]. In addition, this bioassay is a recent development in the assay procedure of bioactive compounds, which shows cytotoxicity as well as a wide range of pharmacological activities (e.g. anticancer, antiviral, insecticidal, pesticidal, AIDS, etc.) of the compounds [7].

The brine shrimp (*Artemia salina*) eggs were placed in one side of a locally fabricated small tank alienated by net containing artificial seawater, which was initially prepared with a commercial salt mixture and double distilled water (3.8% NaCl solution) for hatching. However, a light source was located to catch the attention of nauplii in the other side of the tank. About 50 mg of eggs were sprinkled into the big compartment, which was kept darkened whereas the small compartment was open to normal light. After 2 days of hatching period the nauplii were ready for the experiment, collected by a pipette from the lighted side.

A sample of the synthesized compounds (20 mg) was accurately measured and dissolved in 2 mL of Dimethyl sulfoxide (DMSO) to get a solution of varying concentrations 100, 50, and 25 µg/mL then, transferred to nine vials (three for each dilutions were used for each test sample and LD50 is the mean of three values) and also one vial was kept as control having 2 mL of DMSO only.

The solvent was allowed to evaporate overnight. After two days, when shrimp larvae were ready, 1 mL of seawater and 10 shrimps were added to each vial (30 shrimps / dilution) and allowed to stay there for 24 hours finally, the volume was adjusted with seawater to 10 ml per vial. After that, the mortality was observed and recorded after 24 hour of incubation, using a magnifying glass and the number of survivors in each vial was counted, noted and the data

were analyzed by a Finney computer program to determine the LD50 values [5].

From these data, the percentage of mortality of the nauplii was calculated for each concentration.

In the present investigation, from the **Table 2 and Figure 1**, it is clearly revealed that the p-ClC6H4 and p-BrC6H4 derivatives exhibit promising activity whereas, the simple -C6H5 exhibit moderate activity as indicated by their LD50 values. The structure activity relationship studies of these compounds revealed that R with functional groups –Cl, –Br showed good cytotoxicity. Thus, from the results discussed above it is clear that the synthesized compounds are biologically active and these findings may help to serve as a basis for future direction towards the development of bacteriostatic agents of lower cytotoxicity.

Table 2: Brine shrimp bioassay data of the Synthesized Bis-Thiazolidinones and its DERIVATIVES

Compounds	LD50 (M/mL)
C6H5	1.030×10^{-4}
p-ClC6H4	1.179×10^{-4}
p-BrC6H4	1.472×10^{-4}

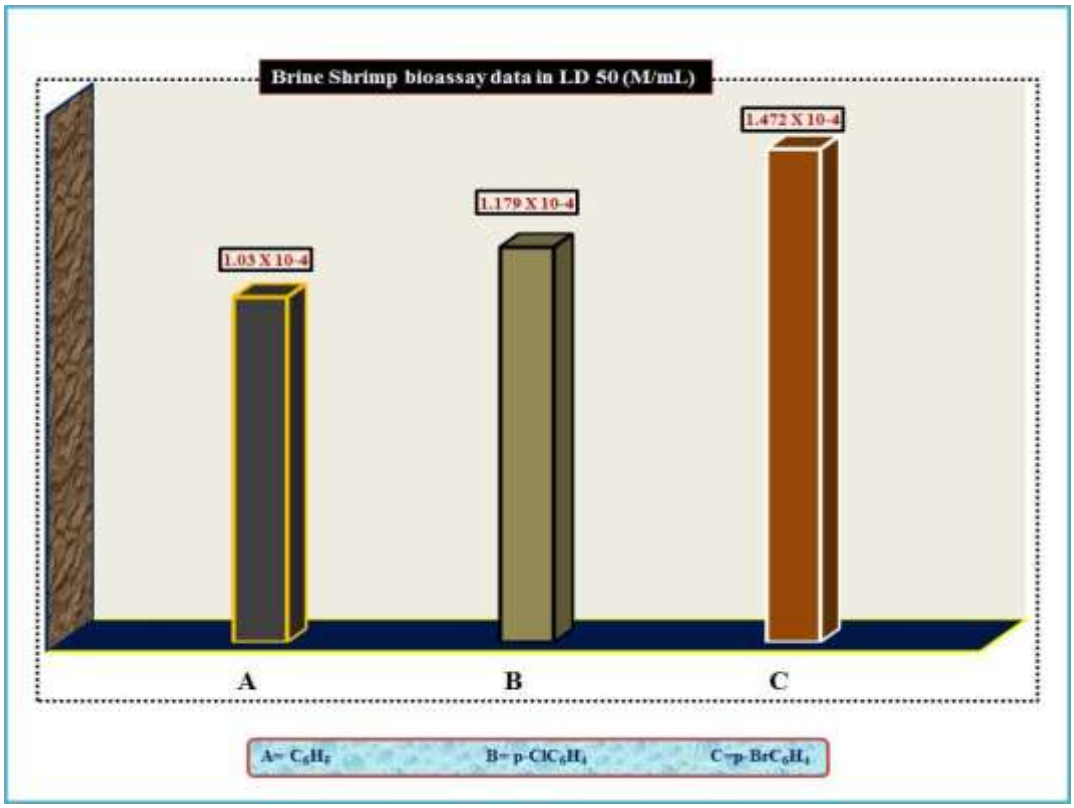


Fig. 1: Brine shrimp bioassay data of the synthesized Bis-Thiazolidinones derivatives.

3. CONCLUSION:

The main aim of the present work is to synthesize Bis-thiazolidinone derivatives and investigate for various bioassays with the hope of discovering new structure leads serving as

potential broad spectrum pharmacological agents.

In vitro cytotoxic activity results clearly revealed that all the synthesized compounds are found to be promising bioactive compounds by registering their LD50 of 1.030 X 10^{-4}, 1.179 X 10^{-4} and 1.472 X 10^{-4} correspond to compounds 1, 2 and 3 respectively.

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REFERENCEES:

1. M. Jain and M. Agarwal. International *Journal of Scientific and Research Publications*, 3, 1-4, **2013.**
2. P. B. Rathna Kumar, M. S. Murthy and K.N Jayaveera. *Journal of Pharmacy and Biological Sciences.* 10, 30-36, **2015.**
3. S. M. Kudari, W. Muneera and S. M. Beede. *Oriental Journal of Chemistry*, 12, 167-171, **1996**.
4. B. N. Meyer, N. R. Ferrigni, J. E. Putnam, J. B. Jacobsen, D. E. Nicholsand and J. L. Mclaughlin. *Planta Medica*. 45, 31-34, **1982**.
5. W. L. F. Armarego and C. L. L. Chai, *Purification of Laboratory Chemicals*, 5th Ed; Butterworth-Heinemann: London, **2004**
6. A. Rehman, M. I. Choudhary, *Bioassay Techniques for Drug Development*; Harwood Academic Publishers: Amsterdam, **2001**; pp. 9-14.
7. K. E. Zahan, M. S. Hossain, S. Sarkar, M. M. Rahman, M. A. Farooque, M. N. Karim, L. Nahar and M. A. Hossain. Dhaka University Journal of Pharmaceutical Sciences 3: 43-47, **2004**.

Fig. S1: Histogram showing the antibacterial comparative activity of Bis-Thiazolidinones and its derivatives at 50, 100 and 200 µg/mL.

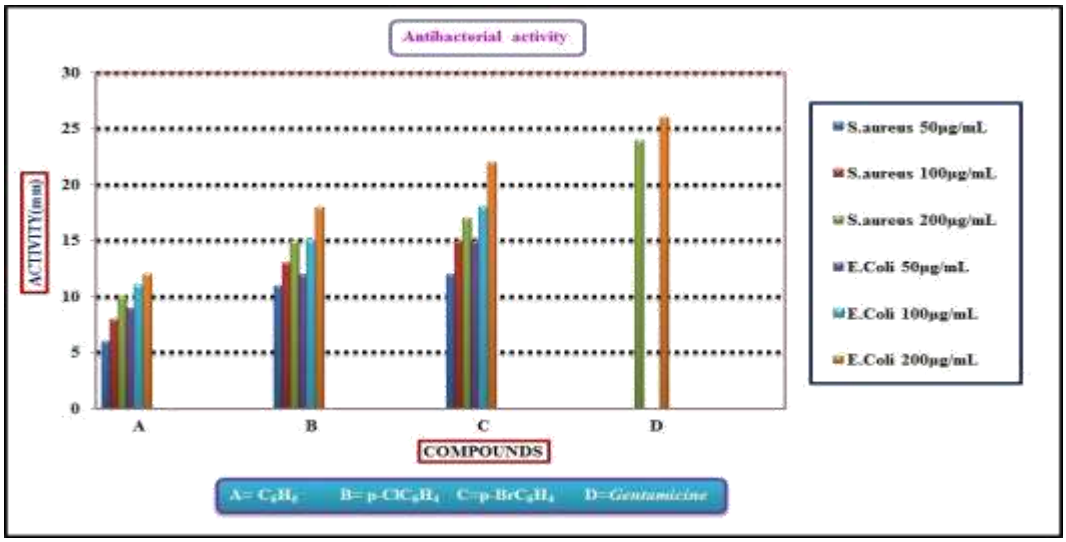

Fig. S2: Histogram showing the antifungal comparative activity of Bis-Thiazolidinones

and its derivatives at 50, 100 and 200 µg/mL.