JSIANE : Vol. 3, Issue 1, 2023, Page 15-21 ISSN (Online) 2583-2093

Journal of Science Innovations and Nature of Earth

Journal homepage : www.jsiane.com

ESTIMATION OF TOTAL LEUCOCYTE COUNT AND ERYTHROCYTE SEDIMENTATION RATE IN *Channa punctatus* (BLOCH.) UNDER TOXIC STRESS OF POTTERY CHEMICALS

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Abstract

The pottery and ceramic industry release toxic heavy metals during preparation and other steps which in turn go to environment and finally to aquatic ecosystem through leaching and runoff water. In the view of above facts it is mandatory to explore this non target effect of ceramic industry for aquatic ecosystem. A dye is a coloured material that chemically attaches to the substrate it is applied on. Dyes are distinguished from pigments by the fact that they do not chemically attach to the substrate they colour. In most cases, the dye is administered in an aqueous solution. Keeping these points in view, the present study is undertaken to assess impact of pottery industry chemical tin oxide on total leucocyte count and erythrocyte sedimentation rate of *Channa punctatus* (Bloch.). The TLC and ESR have been found to be increased after treatment due to hematotoxic effect of pottery chemicals and adverse effect on hemopoietic system of fish.

Keywords: *Channa punctatus* (Bloch.), Pottery chemicals, Pottery industry, TLC, ESR

Received 10.12.2022 **Revised 15.02.202 3** **Accepted 26.03.2023**

Introduction

Khurja is an important industrial and commercial town in Uttar Pradesh's Bulundshahr region. Pottery and agriculture account for nearly half of Khurja's employment and livelihood. The pottery business directly employs around 25,000 people, with an additional 5000-7000 people involved in different support services and linked activities. Trade and commerce, as well as a limited amount of other manufacturing, are other sources of employment in Khurja. Lead leaking into food and drink from ceramics heated with lead glazes is a serious hazard. The Food and Drug Administration (FDA) of the United States and the Canadian Consumer and Corporate Affairs have both set limits on how much lead may leak from food ware into food and drink. Acidic solutions are very dangerous. Similarly, continuous microwave warming (for example, a coffee mug at work) might result in higher lead glaze leaching. The leaching of lead from lead-glazed ceramics has resulted in several incidents of lead poisoning, including some fatalities. Tin oxide, or stannic oxide, is an inorganic substance having the formula SnO_2. Cassiterite is the mineral form of SnO_2, and it is the primary resource of tin. This tin oxide, also known by several other names, is an essential substance in tin chemistry. It is a solid that is colourless, diamagnetic, and amphoteric. Tin oxide has long been used in ceramic glazes as an opacifier and a white colourant. Nickel carbonate is an inorganic chemical or a compound combination comprising nickel and carbonate. Nickel hydroxycarbonate is a chemical intermediate that is used to make nickel metal powder or other nickel salts. Another important application is as an intermediary in nickel catalysts. Nickel hydroxycarbonate can also be found in nickel plating solutions, as a pH regulator, and as an ingredient in tri-cation phosphating baths. Green powder is the appearance of nickel hydroxycarbonate.

Channa punctatus (bloch.) is a pond, swamp, brackish water, ditch, and beel species. Adults love stagnant water in muddy streams and are a snakehead species. It may be found in Afghanistan, Pakistan, India, Sri Lanka, Nepal, Bangladesh, Myanmar, and Tibet, as well as other parts of the Indian Subcontinent. Swamps, ponds, and brackish water systems are its native habitats. It is a fish with great food value but low aquarium value.

Materials and Methods

About forty adult fresh water air breathing teleost, *Channa punctatus* (Bloch.), representing both sexes will be collected alive from the local fish market. They will carefully examine for injury & kept in 1 percent solution of potassium permanganate for a few minutes before transferring them into large aquaria measuring 75cmx37.5cmx37.5cm. The fishes will be collected in the season when room temperature ranged from 30.0^C to 35.0^C & that of the water

from 25.0^{C} to 28.0^{C}. Dechlorinated water will be used in the aquaria, which was changed every alternate day. The fishes will feed daily two times with flour pallets and small pieces of boiled eggs albumin. Tin oxide and nickel hydroxyl-carbonate will be used as experimental compound to assess the effect of ceramic dyes. Total leucocytes were counted using an upgraded standard Neubaur chamber haemocytometer, as reported by Dacie and Lewis (1975) (Dacie and Lewis, 1975). The erythrocyte1sedimentation rate was estimated by Wintrobe's method (1981). All the statistical calculation for data has been conducted using computer software KY plot version 3.0.

Results and Discussion

The fishes of group 5-9 treated with 1/10th of LC_{50} of TO, revealed significant increase (P<0.05) in TLC in whole duration of the experiment, which increased frequently with increased duration of the experiment. A significant increase in TLC was observed at 11th day (12.00 percent), at 22nd day (17.33 percent), at 33rd day (38.67 percent), at 44th day (44.00 percent) and at 56th day (28.00 percent) as compared to control group (1-4), which is highly significant (P<0.05, Table 1 and Fig.1). The group 27-31 treated with 1/10th of LC_{50} of NHC, revealed gradual increase in TLC in the whole duration of experiment, which increased frequently with increased duration of experiment. A significant increase (P<0.05) in TLC of experimental fishes was observed at 11th day (22.67 percent), at 22nd day (31.20 percent), at 33rd day (37.33 percent), at 44th day (38.67 percent) and at 56th day (86.67 percent) as compared to control, which is highly significant (Table 1 and Fig.1).

In the fishes of group 15-20 treated with 1/20th of LC50 of TO, revealed significant increase in TLC in whole duration of experiment with exception at 15th day of treatment, where the experimental fishes showed mild increase (P<0.01) of 1.70 per cent. A significant increase (P<0.05) in TLC of experimental fishes was observed at 30th day (44.00 percent), at 45th day (54.67 percent), at 60th day (38.67 percent), at 75th day (44.00%) and at 90th day (65.33 percent) as compared to control group 1-4 (Table 2 and Fig.2).

The group 37-42 treated with 1/20th of LC50 of NHC, revealed gradual increase in TLC in the whole duration of experimental period, which increased frequently with increased duration of experiment. A significant increase in TLC of experimental fishes was observed at 15th day (22.667 percent), at 30th day (33.333 percent), at 45th day (60.467 percent), at 60th day (70.667 percent), at 75th day (76.00 percent) and at 90th day (92.00 percent), as compared to control group (Table 2 and Fig. 2).

The higher and lower doses of TO produced leucocytosis in treated fishes. Saxena and Tripathi [2009] observed leucocytosis in the blood of C. punctatus under the stress of tin. Some other workers also observed increased TLC of fishes treated with heavy metals (Akarte et al., [2009] and Karuppasamy et al., [2008]). Significant leucocytosis was also observed in the blood of fishes exposed to both higher and lower concentration of NHC. The results indicates infection in exposed fishes. Both the toxicants i.e., TO and NHC increased the number of lymphocytes and monocytes while number of neutrophils decreased in both high and low doses. According to Chadha [1981] the loss of neutrophils shows the loss of diapedesis system which may create serious pathological condition in animal. The elevated lymphocytic count has been observed in man due to Cd toxicity (Harada et al., [1979]) and experimental mammal (Oshawa and Kawai, [1981]) and even a cytological shift resulting in increased large lymphocytes population (Oshawa and Kawai, [1981]) has been reported.

Group no. 5-9 treated with 1/10th of LC50 of TO revealed significant increase (P<0.05) in ESR value of the experimental fishes. A significant increase in ESR were 100.00 percent at 11th day, 25.00 percent at 22nd day, 50.00 per cent at 33rd day, l00.00 percent at 44th day and 150.00 per cent at 56th day as compared to the control group 1-4 (Table 3 and Fig.3). The exposure of NHC in fishes of group 27-31 (1/10th of LC50) revealed more significant increase (P<0.05) in ESR value throughout the experimental period. The significant increase in ESR value of experimental fishes were recorded at 11th day (50.00 per cent), at 22nd day (50.00 per cent), at 33rd day (200.00 per cent), at 44th day (50.00 per cent) and at 56th day (100.00 per cent) as compared to the control group 1-4 (table 3 and Fig.3).

The chronic exposure of TO (1/20th of LC50) in group 15-20 showed more significant increase (P<0.05) in ESR value in the whole duration of the experiment. The increases were 50.00 percent at 15th day and 30th day, 350.00 percent at 45th day, 100.00 percent at 60th day, 350.00 percent at 75th day and 100.00 percent at 90th day as compared to control group 1-4 (table 4 and Fig.4). The chronically NHC treated (I/20th of LC_{50}) fishes of group 37-42 showed no changed in ESR value at 11th day of exposure, while more significant increase (P<0.05) in the ESR value of experimental fi shes were recorded at 30th day (50.00 per cent), at 45th day (100.00 per cent), at 60th day (100.00 per cent), at 75th day (100.00 per cent) and at 90th day (150.00 percent) as compared to control group 1-4 (Table 4 and Fig.4).

TO and NHC produced significant elevation in ESR value in toxicated fishes at high and low level dose, which indicates that the TO and NHC produced infection and change in tissue fluid of fishes. The similar elevation in ESR value of fishes and mammals exposed to chromium was observed by Saxena and Tripathi [2009] and Agrawal et al.,[1985]. Sengupta [2006] observed elevated ESR value in NHC exposed fish H. fossilis. The present finding confirm the findings of aforesaid authors. The higher and lower doses of TO produced leucocytosis in treated fishes. Saxena and Tripathi [2009] observed leucocytosis in the blood of C. punctatus under the stress of tin. Some other workers also observed increased TLC of fishes treated with heavy metals (Akarte et al.,[2009] and Karuppasamy et al., [2005]). Significant leucocytosis was also observed in the blood of

fishes exposed to both higher and lower concentration of NHC. The results indicate infection in exposed fishes.

Table 1: Effect of pottery chemicals (1/10thof LC_{50}) on total leucocytic count (x10^6/mm^3) of fish-*Channa punctatus* (Bloch.)

Treatment (mg/lit.)	Exposed period in days										
	0	**11**	**±**	**22**	**±**	**33**	**±**	**44**	**±**	**56**	**±**
TIN OXIDE	59.02 ±0.32	64.21± 0.04^c	-1.82	66.04 ±0.15 ^{a}	+5.46	80.89± 0.05^c	- 10.91	82.02± 0.15^c	- 10.64	75.10 ±0.10 ^{c}	-5.46
NHC	59.02 ±0.32	75.41± 0.0 2^a	- 4.55	78.03 ±0.03 ^{c}	-6.27	80.01± 0.31^c	- 10.64	81.02± 0.32^c	- 10.46	108.0 2±0.3 8^c	-5.55

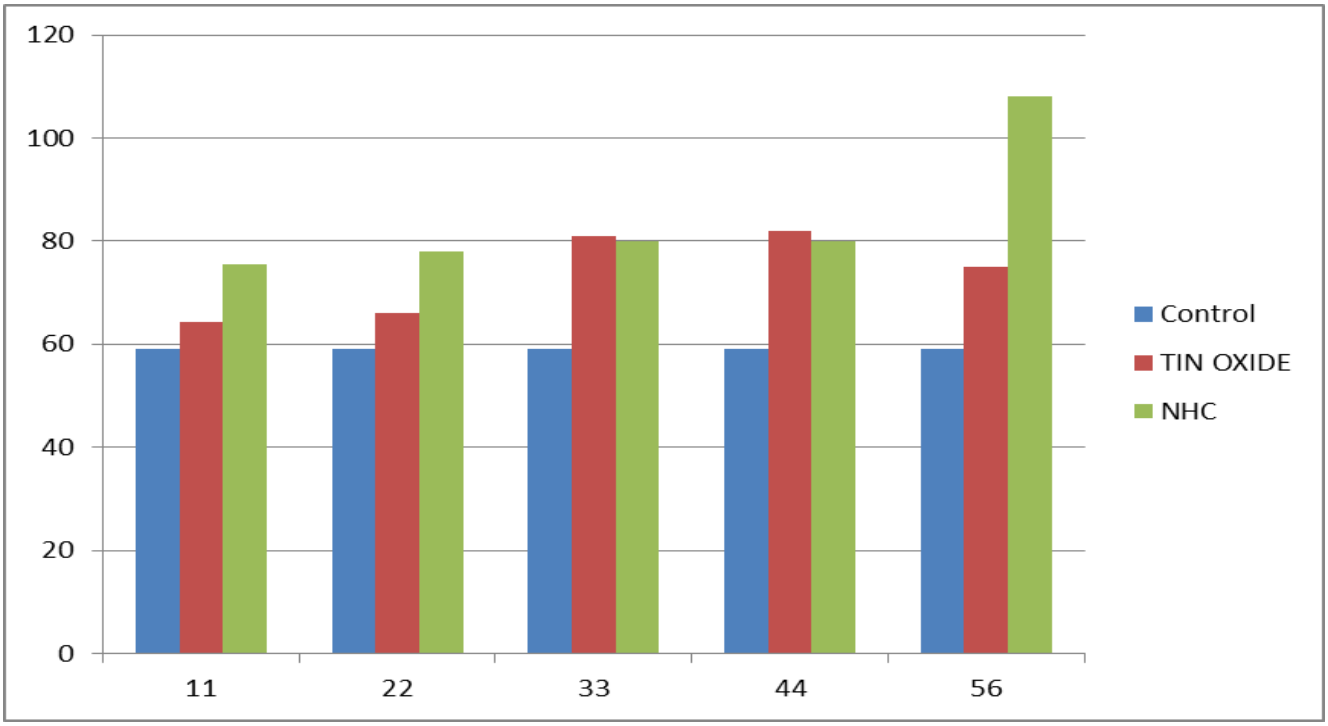

Fig. 1: Effect of pottery chemicals (1/10th of LC_{50}) on TLC (x10^6/mm^3) of fish-*Channa punctatus* (Bloch.)

Table 2: Effect of pottery chemicals (1/20thof LC_{50}) on total leucocytic count (x10^6/mm^3) of fish-*Channa punctatus* (Bloch.)

Treatment (mg/lit.)	Exposed period in days												
	0	**15**	**±**	**30**	**±**	**45**	**±**	**60**	**±**	**75**	**±**	**90**	**±**
TO	59.02 ±0.32	60.00 ±0.43 b	0.00	83.02 ±0.14 c	-3.64	90.31±0.12 c	-15.46	81.04±0.26 c	-17.18	83.64±0.02 c	-1.82	89.32 ±0.08 c	-20.73
NHC	59.02 ±0.32	71.21 ±0.49 b	+ 1.62	78.05 ±0.06 c	-9.10	93.03±0.11 c	-12.73	100.21±0.20 c	-25.32	102.31±0.16 c	-21.55	112.62±0.31 c	-25.09

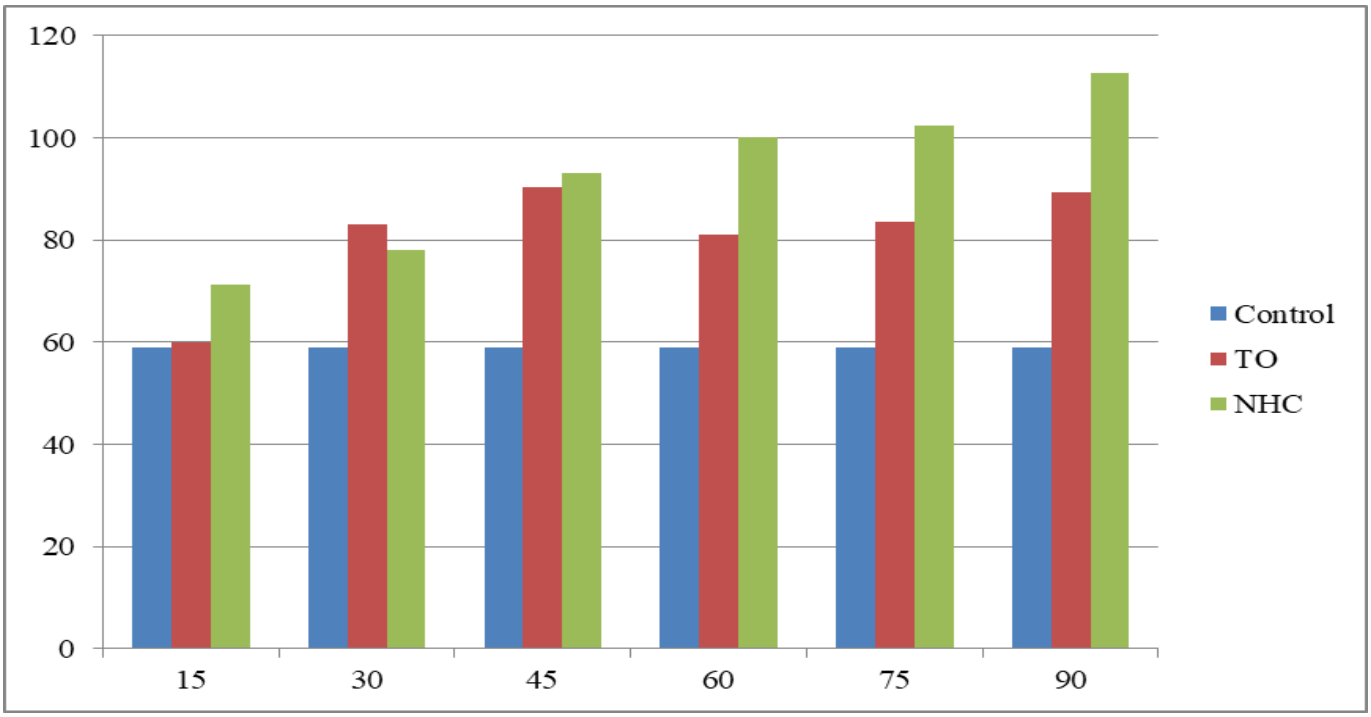

Fig. 2: Effect of pottery chemicals (1/20th of LC_{50}) on TLC (x10^6/mm^3) of fish-*Channa punctatus* (Bloch.)

Table 3: Effect of pottery chemicals (1/10th of LC_{50}) on erythrocytes sedimentation rate (mm/hrs) of fish- *Channa punctatus* (Bloch.)

Treatment (mg/lit.)	Exposed period in days										
	0	**11**	**±**	**22**	**±**	**33**	**±**	**44**	**±**	**56**	**±**
TIN OXIDE	02.01 ±0.17	04.01± 0.17[a]	+100.01	02.4± 0.21[a]	+25.01	03.01+ 0.10[a]	+50.01	04.01± 0.19[a]	+100.01	05.01 ±0.16[a]	+150.01
NHC	02.01 ±0.17	03.01± 0.12	+50.01	03.01 ±0.15[a]	+50.01	06.01± 0.13[a]	+200.01	03.01± 0.10[a]	+50.01	04.01 ±0.15[a]	+100.01

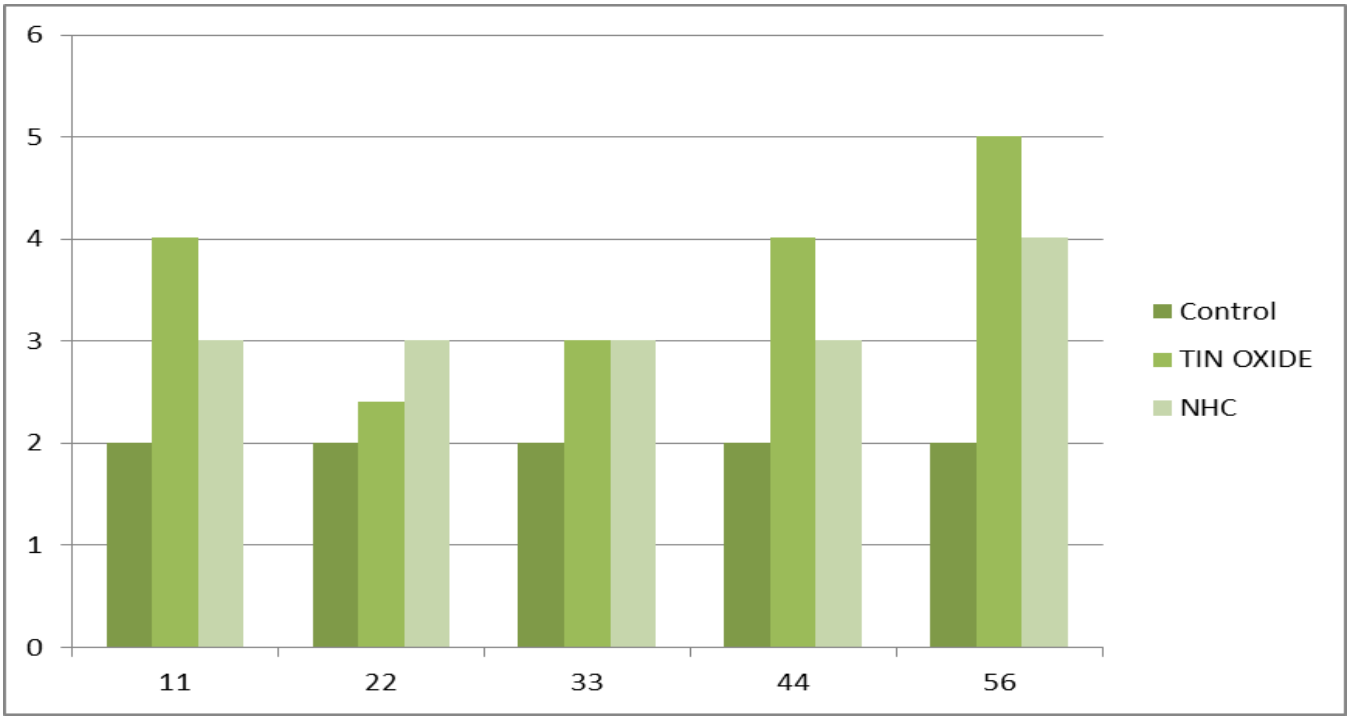

Fig. 3: Effect of pottery chemicals (1/10th of LC_{50}) on ESR (mm/hrs) of fish-*Channa punctatus* (Bloch.)

Table 4: Effect of pottery chemicals (1/20th of LC_{50}) on erythrocytes sedimentation rate (mm/hrs) of fish- *Channa punctatus* (Bloch.)

Treatment (mg/lit.)	Exposed period in days												
	0	**15**	**±**	**30**	**±**	**45**	**±**	**60**	**±**	**75**	**±**	**90**	**±**
TIN OXIDE	02.01 ±17	03.01 ±16[a]	+50 .01	03.01 ±15[a]	+50. 01	09.0 1±0. 34[a]	+35 0.0 1	04.0 1±17 [a]	+10 0.0 1	09.0 1±0. 32	+35 0.0 1	04.01 ±0.16	+10 0.0 1
NHC	02.01 ±17	02.01 ±17[c]	+00 .01	03.01 ±0.13 [a]	+50. 01	04.0 1±0. 13[a]	+10 0.0 1	04.0 1±0. 14	+10 0.0 1	04.0 1±0. 17[a]	+10 0.0 1	05.01 ±0.17 [a]	+15 0.0 1

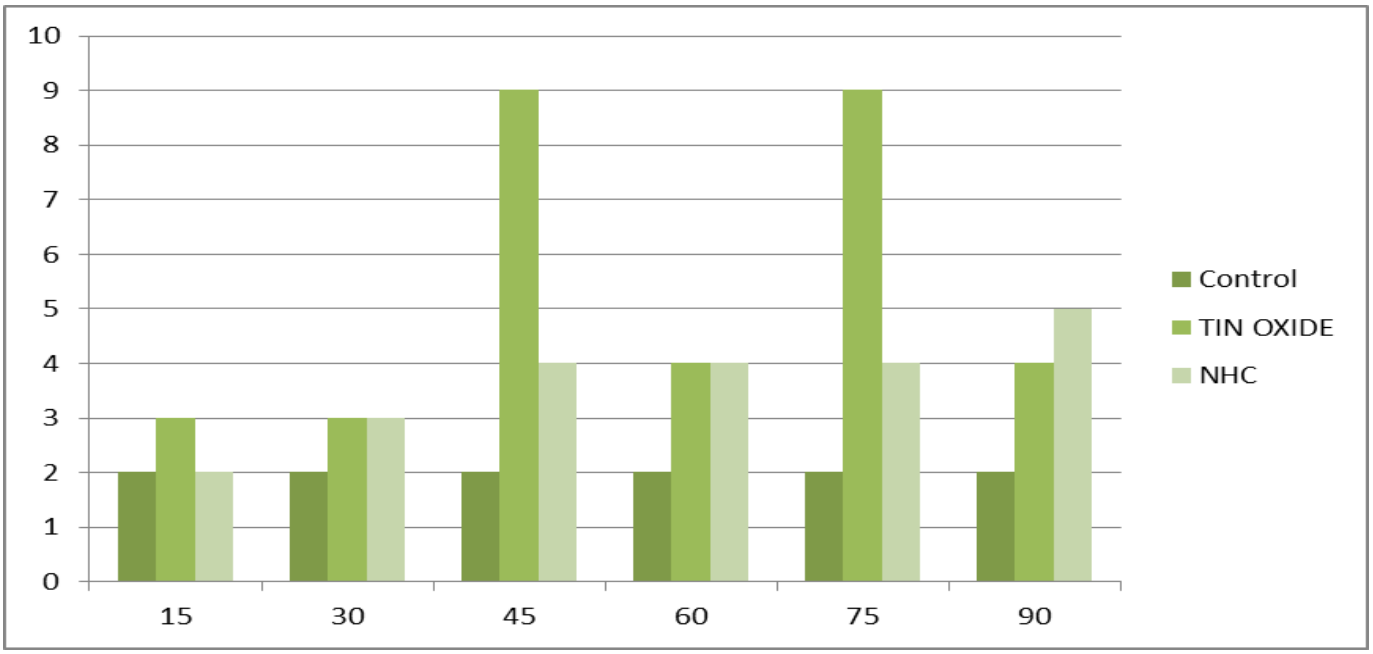

Fig. 4: Effect of pottery chemicals (1/20th of LC_{50}) on ESR (mm/hrs) of fish-*Channa punctatus* (Bloch.)

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