

**BENEFICIAL EFFECT OF *TAMARINDUS INDICA* ON THE KIDNEY OF ALBINO RAT AFTER FLUORIDE INTOXICATION****P.K. SINGH\* AND S. A. MIR***Department of Zoology, School of Life Sciences, Khandari Campus, Dr. B.R. Ambedkar University, Agra – 282 002, India***ABSTRACT**

The toxic effects were evaluated of fluoride ingestion on the kidney of the albino rats, and the possible reversal beneficial effect by use of *Tamarindus indica*. The effect of fluoride intoxication on lipid peroxidation and antioxidant system (SOD, CAT, GST and GR) in the kidney of albino rats were also studied. The body weight, organ weight and body weight – organ weight ratio were decreased significantly due to the side effect of ground water fluoride, while increased due to beneficial effect of *T. indica*. The lipid peroxidation increased due to the side effect of ground water fluoride; while decreased due to beneficial effect of *T. indica*. while, SOD, CAT, GST and GR decreased significantly due to the effect of ground water fluoride, while increased due to beneficial effect *T. indica*. Increased lipid peroxidation and generation of Reactive Oxygen Species (ROS) thus, exerting a toxic effect on proximal tubular cells of the kidney, The effect of fluoride on kidney has two aspects: one is direct action; the other is indirect – disturbances of free radical balance and abnormal of some inorganic elements.

**KEYWORDS** : Albino rat, Antioxidants, CAT, Fluoride intoxication, GST, GR, Lipid peroxidation, ROS, *Tamarindus indica***P.K. SINGH**

Department of Zoology, School of Life Sciences, Khandari Campus, Dr. B.R. Ambedkar University, Agra – 282 002, India

\*Corresponding author

## INTRODUCTION

Ground water is one of the most important sources of drinking water and contamination of ground water with fluoride is one of the serious problems encountered in India. People have been long exposed to fluoride through drinking water, food, and air<sup>1</sup>. Fluorosis has been regarded in the past as disease affecting only bone and teeth<sup>1</sup>. Fluoride is also known to cross the cell membrane and to enter soft tissue<sup>2</sup>. Recent studies have given sample evidence that fluoride toxicity adversely affect most of the soft organ. Generation of free radicals, lipid peroxidation, and altered antioxidant defense system are considered to play an important role in the toxic effects of fluoride<sup>3-4</sup>. The pharmacological investigation on *T. indica* extracts have reported them to have antibacterial, antifungal, hypoglycemic, cholestolemic, cytotoxic, anti-inflammatory, gastrointestinal, and hypolipomic. The antioxidant activity of *Tamarind* has been found by many researchers<sup>5</sup>. The *Tamerind* intake appears to have are additional beneficial effect on mobilization of deposited fluoride from bone by enhancing urinary excretion of fluoride<sup>6</sup>. In the present work, the possibility of preventing fluoride toxicity in albino rats with *Tamarind* pulp has been investigated by analyzing various biochemicals.

## MATERIALS AND METHODS

### (i) EXPERIMENTAL ANIMAL

Thirty five male albino rats (*Rattus norvegicus*) wistar strain of almost equal size and weight  $120 \pm 25$  gm and eight weeks aged were obtained from the animal house of Zoology Department, School of Life Sciences, Khandari Campus, Agra. The rats were randomly divided into seven groups of five rats of each. They were maintained on standard with Gold mohar brand feed and water given *ad libitum*.

### (ii) COLLECTION OF FLUORIDE WATER

The fluoride water was collected from fluoride region at villages panchgai, Khera, Sikandra and Trans Yamuna, in Agra. The water samples were collected directly from different as usual water sources like hand pumps in polypropylene bottles. The selection of the fluoride water samples of panchgai village near Agra which has maximum unit of fluoride 14.29 mg/L. The concentration

of fluoride ions in water sample was measured by method given ion selective method.

### (iii) COLLECTION AND DOSING OF PLANT EXTRACT

The fruits of *Tamarindus indica* were collected from local market. All the materials were taxonomically identified by Department of Botany, School of Sciences Khandari Campus, Agra. The aqueous pulp extract of *Tamarindus indica* was prepared by extraction with hot water in order to simulated the local procedure<sup>6</sup>. The selected therapeutic dose of *Tamarindus indica* for entire research was 10gm/kg body weight given to albino rat<sup>7-8</sup>. The selection of dose of *Tamarindus indica* has not been determined yet but however, in some previous studies the *Tamarindus indica* was administered orally to dogs at 10mg/kg body weight in juice formulation<sup>8</sup>. All treatments were given orally using a hypodermic syringe and a bent tip canula. The one group of albino rats were treated as control group for 7, 14 and 21 days and was treated with distilled water in which the fluoride ion concentration was 0 mg/L. The next three groups of albino rats were given the fluoride water for 7, 14 and 21 days. The remaining three groups of albino rats were treated with fluoride water in the way and then given *T. indica* dose for 3, 7 and 14 days respectively. The albino rats of both groups were sacrificed under light ether anaesthesia. Body weight was measured before and after the experimental period.

### (iv) ESTIMATION OF ANTIOXIDANT INDICES

The organ weight of kidney was measured after dissected out carefully, blotted free of blood, and weighed on a Roller Smith Torison Balance (USA) to the nearest mg and utilized for the study. The kidney were homogenized and centrifuged. The supernatant was then assayed for the activities of lipid peroxidation<sup>9</sup>, superoxide dismutase, (SOD)<sup>10</sup>, chloramphenicol acetyltransferase (CAT)<sup>11</sup>, glutathione S-transferase (GSTS)<sup>12</sup> and GR<sup>13</sup>. The data were statistically analysed by students 't' test and Analysis of variance (ANOVA).

## RESULTS AND DISCUSSION

Body weight, organ weight, ratio, lipid peroxidation, Superoxide dismutase (SOD), Chloramphenicol Acetyl Transferase (CAT), Glutathione S-Transferases (GST), Glutathione Reductase (GR) of the control, fluoride water (F.W.) and the fluoride water, *Tamarindus indica* (TI) treated rats were given in Table 1 and 2.

**Table 1**  
**Changes in body weight, organ weight and their ratio: values are expressed in gram**

Group	Body weight	Organ weight	Organ – body weight ratio
Control	135.8 ± 1.428	1.726 ± 0.0378	0.0127 ± 0.000295
7 days F.W.I.	130 ± 1.4142	1.468 ± 0.0344	0.0113 ± 0.000362
3 days T.i. treatment after 7 days F.W.I.	134 ± 1.4142	1.746 ± 0.0365	0.0130 ± 0.000340
14 days F.W.I.	120 ± 1.4142	1.34 ± 0.0447	0.0111 ± 0.000409
7 days T.i. treatment after 14 days F.W.I.	130.8 ± 1.019	1.644 ± 0.0278	0.0122 ± 0.000290
21 days F.W.I.	114.6 ± 1.326	1.128 ± 0.0287	0.00987 ± 0.000246
14 days T.i. treatment after 21 days F.W.I.	128.4 ± 1.166	1.662 ± 0.0405	0.0129 ± 0.000330

Data are expressed as Mean ± S.E. \*P < 0.05; \*\*P < 0.01; \*\*\*P < 0.001. Comparison of control with fluoride water ingestion (F.W.I.) and also *Tamarindus indica* (Ti) with fluoride water ingestion F.W.I. (n = 5)

Table 2

**Changes in lipid peroxidation, Superoxide dismutase (SOD), Chloramphenicol Acetyl Transferase (CAT), Glutathione S-Transferases (GST), Glutathione Reductase (GR) with liver. Values are expressed in  $\mu\text{mol/mL}$**

Group	Lipid peroxidation	SOD	CAT	GST	GR
Control	3.592 $\pm$ 0.0844	2.876 $\pm$ 0.0556	2.689 $\pm$ 0.0458	0.348 $\pm$ 0.02477	13.53 $\pm$ 0.1143
7 days F.W.I.	4.34 $\pm$ 0.0761***	2.228 $\pm$ 0.0496***	2.222 $\pm$ 0.0424***	0.24 $\pm$ 0.01949**	12.84 $\pm$ 0.0394***
3 days T.i. treatment after 7 days F.W.I.	3.512 $\pm$ 0.0491***	2.806 $\pm$ 0.0444***	2.668 $\pm$ 0.0548***	0.384 $\pm$ 0.0163***	13.57 $\pm$ 0.06767***
14 days F.W.I.	5.794 $\pm$ 0.0467***	1.914 $\pm$ 0.0472***	1.96 $\pm$ 0.0484***	0.196 $\pm$ 0.0136***	12.194 $\pm$ 0.0428***
7 days T.i. treatment after 14 days F.W.I.	3.734 $\pm$ 0.571**	2.744 $\pm$ 0.03906***	2.506 $\pm$ 0.04118**	0.434 $\pm$ 0.0224***	12.744 $\pm$ 0.06087***
21 days F.W.I.	7.02 $\pm$ 0.0533***	1.494 $\pm$ 0.04489***	1.63 $\pm$ 0.0327***	0.108 $\pm$ 0.01067***	11.37 $\pm$ 0.03209***
14 days T.i. treatment after 21 days F.W.I.	3.296 $\pm$ 0.0444***	2.564 $\pm$ 0.0581***	2.508 $\pm$ 0.02956***	0.468 $\pm$ 0.03826***	13.43 $\pm$ 0.0349***

Data are expressed as Mean  $\pm$  S.E. \*P < 0.05; \*\*P < 0.01; \*\*\*P < 0.001. Comparison of control with fluoride water ingestion (F.W.I.) and also *Tamarindus indica* (Ti) with fluoride water ingestion F.W.I. (n = 5)

Intracellular fluoride concentrations are lower, but they change proportionately and simultaneously with those of plasma with the exception of the kidney, which concentrates fluoride within the renal tubules, tissue-to-plasma fluoride ratios are less than 1.0. The renal clearance of fluoride is directly related to urinary pH and, under some conditions, to urinary flow rate. Recent data from stop-flow studies with dogs indicate that fluoride reabsorption is greatest from the distal nephron, the site where the tubular fluid is acidified<sup>13</sup>. In the present study the body weight, organ weight and body weight – organ weight ratio decreased due to side effect of ground water fluoride; while increased due to beneficial effect of *Tamarindus indica*. Similar findings have been reported in dogs<sup>9</sup>, and in rats<sup>15</sup> after beneficial effects of amino acid, glycine; while in mice due to side effects of chronic fluorosis<sup>16</sup>, due to side effects of fluoride and aluminum in the muscle and liver in male mice<sup>17</sup>. Therefore in the present study an increased lipid peroxidation in the kidney due to side effect of ground water fluoride, and decreased due to the beneficial effect of *Tamarindus indica*. Similar findings have also been reported in rat after adverse effect of fluoride due to the antagonistic effect on antioxidant<sup>18</sup>, while in rats due to side effect of fluoride<sup>19</sup> and in rats due to fluoride intoxication and increased lipid peroxidation in liver, kidney and decrease other antioxidant indices<sup>20</sup>. In the present study the SOD, GST, CAT and GR significantly decreased due to side effect of ground water fluoride; while increased due to the beneficial effect of *Tamarindus indica*. Similar observations have been reported in kidney of rat due to side effect of fluoride and also observed the efficiency of *Tamarindus indica* to protect against fluoride intoxication to ameliorate the oxidative stress in rats due to side effect of fluoride intoxication and also due to decrease ability of the tissue to handle oxygen radicals in the muscle of mice due to side effect of fluoride<sup>21</sup> in the liver and kidney of mice due to fluoride induce toxicity in the ovary of mice<sup>22</sup> influence of fluoride on the biological free radical, while in the liver and kidney of rats due to adverse effect of fluoride.

Likewise the decrease in the activity of the antioxidant enzyme in the liver decrease fluoride intoxication<sup>23</sup>, thus fluoride abusively stimulated the respiratory burst and produces superoxide and hydroxyl radicals and decreases these antioxidant enzymes possibly by forming a strong hydrogen bond with the amide group<sup>4</sup>. On the other hand after *Tamarindus indica* treatment showed a increase an antioxidant enzymes due to a protection antioxidant effect and significant lipid lowering effect of *Tamarind* plant constituent present in the pulp extract because of their free radical scavenging ability, antioxidants have an important role in ameliorating fluoride toxicity. As seen in the experiment here, the free radical scavenging effect *Tamarind* pulp is authenticated by the decrease lipid peroxidation content in the liver, kidney and by the increased level of polyphenols and flavonoids<sup>10</sup>.

## CONCLUSION

From this investigation it could be concluded that the *Tamarindus indica* is beneficial in the medicinal perspective against the toxic effects of fluoride by disrupting the normal function of antioxidant indices in the kidney of the concerned ground water toxicity. The conflict of interest of this research to aware the people about the various effects of fluoride and also beneficial effects of *Tamarindus indica* to manage the fluoride effect on the body.

## ACKNOWLEDGEMENT

We thank to the Univerty Grants Commission (UGC) New Delhi, for providing funds and facilities for conducting this research work.

## CONFLICT OF INTEREST

None.

## REFERENCES

1. Ando M, Tandon M, Liang C, Cao S. Health effects of indoor fluoride pollution from coal burning in China. *Environmental Health Perspectives*, 1998;106(5):239-42.
2. Bezerre de, Menezes LM, Volpato MC, Rasalen PL, Cury JA. Bone as a biomarker of acute fluoride toxicity. *Forensic Sci Int* 2003;137:209-14.
3. Neurath C. Tooth decay trends for 12 year old in nonfluoridated and fluoridated countries. *Fluoride* 2005;38:324-25.
4. Jacyszyn K, Marut A. Fluoride in blood and urine in human administered fluoride and exposed to fluoride-polluted air. *Fluoride* 1986;19:26-32.
5. Shashi A. *In vivo* studies concerning toxic effect of sodium fluoride on hepatic function in rabbits. *Fluoride* 2003;36(1):30-7.
6. Kolodziejczyk L, Grygial WK, Mysliwiec Z. Protective effect of chrysin in rats subchronically exposed to sodium fluoride. *Fluoride* 2004;37(3):209-12.
7. Cicek E, Aydin G, Akdogan M, Okutan H. Effect of chronic ingestion of sodium fluoride on myocardium in a second generation of rats. *Hum Exp Toxicol* 2005;24:79-7.
8. Khandare AL, Rao, GS Lakshmaiah N. Effect of Tamarind ingestion on fluoride excretion in humans. *Eur. J. Clin. Nutr.*, 2002; 56(1): 82-85.
9. Rzeuski R, Chlubek D, Machoy Z. Interactions between fluoride and biological free radical reactions. *Fluoride* 1998;31:43-5.
10. Shrama A, Chinoy NJ. Role of free radicals in fluoride induced toxicity in liver of mice and its reversal. *Fluoride* 1998;31: 26-2
11. Parvez SS, Parvez MM, Nishihara E, Fujii Y. *Tamarindus indica* leaf is a source of allelopathic substance. *Plant growth Regulation* 2003;40(2):107-15.
12. Khandare AL, Kumar PU, Lakshmaiah N. Beneficial effect of *Tamarindus ingestion* on fluoride toxicity in human. *Eur J Clin Nutr* 2002;56(1):82-5.
13. Harwood JE. The use of an ion-selective electrode for routine analysis of water samples. *Water Res* 1969;3:273-5.
14. Nebot C, Moutet M, Huet P, J-Z Xu JC, Yadav Chandiere J. Spectrophotometric assay of superoxide dismutase activity based on the activated auto-oxidation of a tetralycol catachol. *Anal biochem* 1993;214:442-57.
15. Callaway JK. The analysis of free radicals antioxidant enzymes and CAT assay in the tissue. *Biotechniques* : 1998;488-99.
16. Mannervik BP, Guthenberg C. Identification of three classes of cytosolic glutathione transferase common to several mammalian species : correlation between structural data and enzymatic properties. *Proc Natl Acad Sci USA* 1985;82:7202-6.
17. Bompport GJ, Prerof DS, Basconds JS. Rapid automated analysis of glutathione reductase, peroxidase, and S-transferase activity. *Clin Biochem* 1990;23:501-4.
18. Whiford GM, Pashley DH. Fluoride reabsorption by non-ionic diffusion in the distal nephron of the dog. *Proc Soc Exp Biol Med* 1991;196(2):178-3.
19. Chinoy NJ, Metha D. Beneficial effects of the amino acid glycine and glutamine on testis of mice treated with sodium fluoride. *Fluoride* 1999;3 62-70.
20. Ersan Y, Evren KOC, Ismail ARI, Basaran K. Histopathological effects of chronic fluorosis on the liver of mice. *Trunk J Med Sci* 2010;40(4):619-2.
21. Chinoy NJ, Memon MR. Beneficial effects of some vitamins and calcium on fluoride and aluminum toxicity on gestronemius muscle and liver of male mice. *Fluoride* 2001;34: 21-33.
22. Vani ML, Reddy KP. Effects of fluoride accumulation on some enzymes of brain and gastrocnemius muscle of mice. *Fluoride* 2000;33:17-26.
23. Ranjan R, Swarup D, Patra RC. Oxidative stress indices in erythrocytes, liver and kidney of fluoride-exposed. *Fluoride* 2009;42:88-3.