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Evaluation of Association of Cord Blood Fluoride Levels with Visible Congenital Anomalies in Newborns

Abhra Ghosh[1], Simmi Kharb[2], Smiti Nanda[3]

[1]*Junior Resident, Department of Biochemistry, Pt. B. D. Sharma PGIMS, Rohtak, Haryana*
[2] *Professor, Department of Biochemistry, Pt. B. D. Sharma PGIMS, Rohtak, Haryana*
[3] *Professor, Department of Obstetrics and Gynecology, Pt. B. D. Sharma PGIMS, Rohtak, Haryana*

ABSTRACT

Studies have suggested that fluoride might be associated with alteration in reproductive hormone levels, infertility, Down's syndrome, increased rate of congenital anomalies and cancers. There is an increased concentration of congenital anomalies in north-west India, however, no studies are available regarding the possible association with fluoride. In the present study, serum fluoride levels were explored in cord blood of newborns with visible congenital anomalies (n=30) and in healthy newborns (n=30). The detailed history of present pregnancy, the status of the baby (birth order, previous stillbirth, details of defects and handicaps), nutritional status of parents (deficiency, if any), history of pesticides or heavy metal exposure of parents, history of use of any medication (prior or during pregnancy). Detailed examination of babies, in terms of site of the defect, the neurological deficit was also recorded. Five mL cord blood was collected from the placental end of the umbilical cord after delivery of baby and serum was separated and routine biochemistry investigation was performed by autoanalyzer. Serum fluoride levels were analyzed by ISE in cord blood of both the groups. In the present study, cord blood fluoride levels were higher in babies with congenital anomalies as compared to healthy controls. Fluoride crosses the placenta and gets incorporated into tissues of developing conceptus. Chronic exposure to high fluoride levels may produce harmful effects on developing fetus especially brain and heart, thyroid and increased future risk of cancer. Thus, findings of the present study suggest that fluoride in pollution exposures (agricultural and industrial) in this region might be a causative factor in congenital anomalies in this region.

Keyword: *Cord Blood, Congenital Anomalies, Newborns, Fluoride.*

1. INTRODUCTION

Environmental threats to children's health are wide-spread and are of burgeoning concern in a country like India, where rapid, largely unchecked industrialization is leading to dangerous levels of contamination in many elements. According to WHO classification of five mortality strata, India is classified under Sear-D category and is among the countries in the South-East Asian region with high child and adult mortality rates.

1.1 Effects of Fluoride on Human Health

Many heavy metals have been reported to move freely across human placenta, but the potential health effects of heavy metal transfer from mothers to young infants cannot be discounted [1]. Recently introduced dangers are asbestos construction materials, fluoride in groundwater; untreated manufacturing wastes released to landfills; chlorinated hydrocarbons and pesticides [1-3]. A number of studies have shed light on the degree to which genetic and environmental factors play extremely important roles in human health [1-5].

Drinking water is another major issue of concern in the context of children's health. They include heavy metals, arsenic, and fluoride with the last one being the most toxic to the fetus, infants, and children below 12 years of age. In India, excess fluoride problems have been detected in 20 states and arsenic in two states [3, 5] A study has been reported no significant difference in levels of fluoride in neural tube defects in comparison to the normal babies [6, 7]. Also, few studies have observed significantly higher fluoride levels in osteosarcoma [8, 9]. Studies conducted in Brazil and China have shown that children whose parents use pesticides in and around the home are more likely to get leukemia, brain cancer, non-Hodgkin's lymphoma and soft tissue sarcoma [10].

Environmental Health Prospective Journal study has shown that children exposed prenatally to household insecticides had an increased risk of brain cancer later on. A few studies of human populations have suggested that fluoride might be associated with alterations in reproductive hormones, fertility and Down's syndrome [11]. Fluorides also inhibit the activity of cholinesterases, including acetylcholinesterase.

Toxic metals have been documented to be reproductive and developmental toxins causing birth defects and damaging fetal development, as well as, neurological effects, developmental delays, learning disabilities and behavioral abnormalities in otherwise normal appearing children. A National Academy of Sciences (NAS) report found that 50% of US pregnancies result in birth defects or neurological conditions or other chromosomal developmental problems and a majority of these are due to toxic exposures. Lead has also been shown to be the most potent promoter of cancer and birth defects. Municipal waste and coal contain a large number of toxic metals like mercury, lead, cadmium, etc.; large volumes of toxic metal emissions are occurring where stringent controls on incinerator fuel sources and stack emissions are not in effect [12].

The fetus is not well protected against industrial chemicals. The placenta does not block the passage of many environmental toxicants from the maternal to the fetal circulation, and more than 200 foreign chemicals have been detected in umbilical cord blood. Moreover, the developing human brain is exceptionally sensitive to injury caused by toxic chemicals and several developmental processes have been shown to be highly vulnerable to chemical toxicity. With respect to developmental neurotoxicity of organophosphates (OPs) in humans, knowledge is still relatively sparse, and most studies reflect exposures to more than one pesticide.

Punjab is turning into a hotspot of environmental toxicity and genocide and Haryana is following its footsteps. Highly toxic chemicals have been added to the soil, water, air, and food chain of Punjab for the last many decades under the name of development. For the last few decades dangerously toxic agricultural inputs - pesticides and fertilizers are being used blindly, mindlessly and without any scientific evidence of their actual need. But recently added radioactive toxicity, the toxicity of genetically modified crops (Bt. Toxin) has the potential of causing disaster in itself. The prevalence of congenital abnormalities has increased manifold [4-7]. Increase incidence of congenital abnormalities in north-west India suggests the role of some environmental factor as N-W India is predominantly an agricultural community and fluoride and pesticides exposure are hypothesized to be one of the causative agents. If such a cause is found, preventive measures can be taken. In western studies, heavy metals, pesticides have been implicated [1-3].

1.2 Aim and objectives

However, no such studies are available in the Indian context, especially in Haryana. Hence the present study was planned to evaluate association of cord blood fluoride levels with visible congenital anomalies in Indian population to collect baseline data so that preventive and diagnostic measures can be planned in environmental health studies for Indian children.

2. MATERIALS AND METHODS

The present prospective observational case-control study was conducted in the Department of Biochemistry in collaboration with Department of Obstetrics & Gynecology, Pandit Bhagwat Dayal Sharma PGIMS, and Rohtak.

2.1 Grouping

Fifty babies delivered in the labour room were selected for the study and grouped as:

a. Group 1: Newborn infants with visible congenital anomalies (n=30).
b. Group 2: Healthy newborn infants without congenital anomalies (n=30).

All the patients were subjected to detailed history and clinical examination: a detailed history of both present and during pregnancy; status of baby: birth order, previous stillbirth, details of defects and handicaps. Also, examination of parents for nutritional deficiency, pesticide/heavy metal exposure was done. Examination of a baby was carried out for the site of the defect, neurological deficit if any, detailed radiological investigation.

2.2 Sampling

Five mL cord blood was collected from the placental end of the umbilical cord after delivery of the baby. The serum was separated by centrifugation.

2.3 Analysis

Routine investigations were done as per standard methods by autoanalyzer. Serum (cord blood) fluoride level was analysed by ISE [11].

2.4 Statistics

The data so obtained was subjected to appropriate statistical analysis namely Student’s t-test.

3. RESULTS

3.1 Distribution of cases in different congenital anomalies

Cleft lip (n=7) was the most common congenital anomaly in the present study, while CTEV with 6 cases and hypospadias, polydactyly, spina bifida showed the similar distribution with 3 cases each; limb defect was seen only in 2 cases (Table 1).

Table 1. Distribution of Cases in Different Congenital Anomalies

CONGENITAL ANOMALY	NO. OF CASES
Cleft lip	7
CTEV	6
Hypospadias	3
Polydactyly	3
Spina Bifida	3
Limb defect	2
Anencephaly	1
Gastroschisis	1
Encephalocele	1
Omphalocele	1
Cleft palate	1
Imperforate anus	1

3.2 Cord Blood Fluoride Levels in Both Groups

4. Table 2. Cord blood fluoride levels in both groups, p=0.000

	CASES	CONTROLS
Cord blood fluoride levels (mg/L)	0.176 ± 0.058	0.055 ± 0.029

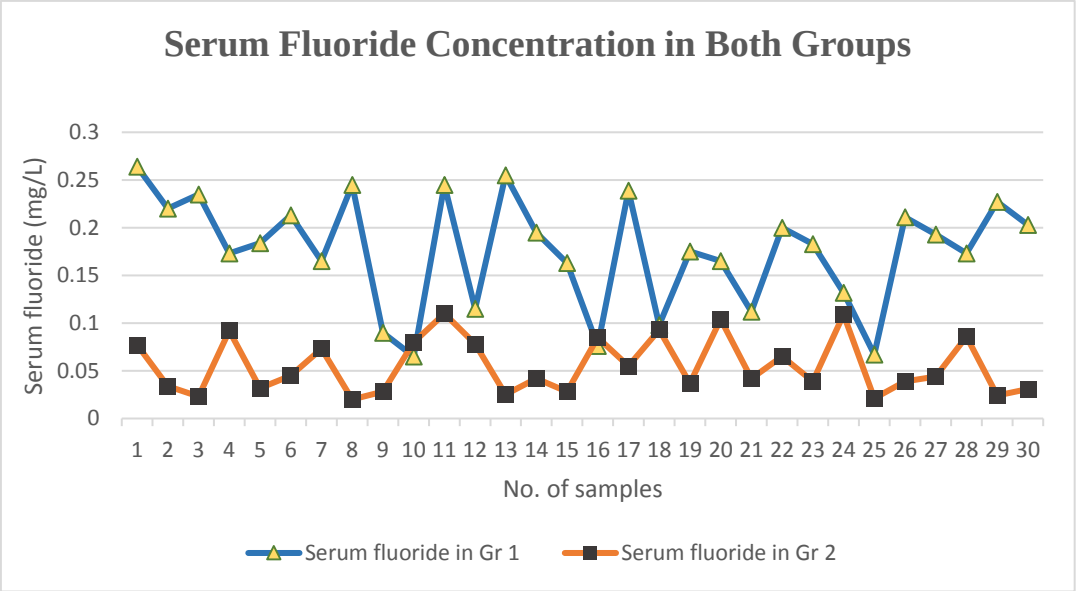

Fig.1. Serum Fluoride Concentration in Both Groups

In the present study, cord blood fluoride levels were higher were significantly raised in babies with congenital anomalies as compared to healthy counterparts (p=0.000, Table 2, Fig.1).

3.3 Fluoride Levels According to Occupation

On analyzing cord blood fluoride levels according to occupation, it was observed that there was no correlation with occupation as there was no history of any occupation hazard or exposure to heavy metals and/or pesticides to parents, since only three parents were farmers in the present study (Table 3).

Table 3: Fluoride levels According to Occupation

Parameters	Farmers (n=7)	Housewives (n=16)	Service women (n=7)
Serum fluoride (mg/L)	0.201 ± 0.06	0.175 ± 0.06	0.151 ± 0.02

3.4 District-Wise Distribution of Cases and Fluoride Levels

Serum fluoride levels in group I was highest in babies from Panipat district followed by babies from Sonepat district. Babies from Rohtak had the lowest concentration of fluoride in group 1 (Table 4).

Table 4. District Wise Distribution of Cases and Fluoride Levels

District (no. of cases)	Serum Fluoride (mg/L)
Sonipat (7)	0.206 ± 0.044
Panipat (6)	0.224 ± 0.027
Bhiwani (5)	0.162 ± 0.056
Jind (5)	0.162 ± 0.061
Rohtak (3)	0.101 ± 0.054
Hisar (3)	0.125 ± 0.035
Jhajjar (1)	0.193 ± 0.000

3.5 Fluoride Levels in Different Types of Cases

Babies with congenital defects namely anencephaly, omphalocele, encephalocele and limb defect had significantly higher fluoride levels as compared to other congenital anomalies (0.264 ± 0.000, 0.245 ± 0.000, 0.245 ± 0.000 and 0.212 ± 0.039 mg/L respectively). On the other hand, other congenital anomalies also had higher fluoride levels compared to healthy controls (Table 5).

Table 5. Fluoride Levels in Different Types of Cases

CONGENITAL ANOMALY	FLUORIDE (mg/L)
Cleft lip (7)	0.169 ± 0.073
CTEV (6)	0.172 ± 0.060
Hypospadias (6)	0.196 ± 0.019
Spina Bifida (3)	0.129 ± 0.073
Limb defect (DDH) (2)	0.212 ± 0.039
Anencephaly (1)	0.264 ± 0.000
Omphalocele (1)	0.245 ± 0.000
Gastroschisis (1)	0.163 ± 0.000
Encephalocele (1)	0.245 ± 0.000
Cleft palate (1)	0.132 ± 0.000
Imperforate anus (1)	0.1650.000

4. DISCUSSION

Fluoride functions primarily as protection against dental cavities and plays a role in bone mineralization. However, excess fluoride can be harmful to organisms. In recent years, researchers have noted that fluoride poisoning appears to begin in the fetal stage [12, 13].

Fluoride crosses the placenta and gets incorporated into tissues of developing conceptus. Chronic exposure to high fluoride levels may produce harmful effects on developing fetus especially brain and heart, thyroid and increased future risk of cancer [12-14]. In recent years, several investigations demonstrated that fluoride can induce oxidative stress and modulate intracellular redox homeostasis, lipid peroxidation and protein carbonyl content, as well as alter gene expression and cause apoptosis. Genes modulated by fluoride include those related to the stress response, metabolic enzymes, the cell cycle, cell-cell communications and signal transduction [14].

Cord blood fluoride levels were significantly raised in the present study in the babies with congenital anomalies as compared to healthy counterparts (p=0.000, Table 2, Fig.1).

No data are available in the literature regarding cord or blood fluoride levels in newborns with congenital anomalies. A higher concentration of fluoride has been found in embryonic brain tissue obtained from the termination of pregnancy in China in areas where fluorosis due to coal burning was prevalent [15]. NaF has been reported to have genotoxic and toxic effects in Drosophila melanogaster [16].

Association between water fluoride level and incidence of Down syndrome is inconclusive [17]. NRC committee report on fluoride in drinking water 2006 reports that a number of human studies have linked developmental/ reproductive effects and fluoride in drinking water. No significant increase in the frequency of chromosomal aberrations of human foreskin fibroblast cells induced by sodium fluoride treatment was reported by Tsutsui [18]. However, the significant increase in chromosomal aberrations have been reported in rat vertebral body-derived cells exposed to sodium fluoride [19]. The cytotoxic and clastogenic effects of sodium fluoride during various phases of cell cycle of human cultured diploid fibroblasts have been reported indicating that cytotoxicity and clastogenicity of sodium fluoride to cultured human diploid fibroblasts are cell phase dependent and that the cells in early and middle S phases are more sensitive to these effects, NaF inhibits DNA synthesis/repair, suggesting that NaF-induced aberrations may occur by an indirect mechanism involving the inhibition of DNA synthesis/repair [20].

Exposure to fluoride during pregnancy results in increased incidence of spontaneous abortions, congenital cardiac disease or late adverse pregnancy outcomes including congenital anomalies, stillbirths and deaths. An ecological study in the US has reported that at least 3 mg fluoride per liter in drinking water showed significant associations between annual fertility rate and fluoride exposure compared with positive associations [21].

Area wise distribution of cord blood fluoride levels showed that fluoride levels were higher in Panipat and Sonepat districts and lower in Jhajjar, Bhiwani and Jind districts and lowest in Rohtak district. Though the sample size is less, still it can be commented here that Panipat district is an industrial area and Bhiwani and Jind districts are agricultural areas. Finding of high cord blood fluoride levels in these areas are possibly due to contamination of the drinking water by toxic chemicals of industrial waste or pesticides used in the fields.

5. CONCLUSION

Thus, findings of the present study suggest that fluoride in pollution exposures (agricultural and industrial) in this region might be a causative factor in congenital anomalies in this region. The data presented may serve as the baseline in planning precautionary approaches to reduce community exposure to environmental pollution.

6. REFERENCES

[1] Ramirez GB, Cruz MC, Pagulayan O, Ostrea E, Dalisay C. The Tagum study I: analysis and clinical correlates of mercury in maternal and cord blood, breast milk, meconium, and infants' hair. Pediatrics 2000; 106: 774-81.

[2] Muckle G, Ayotte P, Dewailly E E, Jacobson SW, Jacobson JL. Prenatal exposure of the northern Québec Inuit infants to environmental contaminants. Environ Health Perspect 2001; 109: 1291-9.

[3] Carpenter DO, Chew FT, Damstra T, Lam LH, Landrigan PJ, Makalinao IR, et al. Environmental threats to the health of children: the Asian perspective. Environ Health Perspect 2000; 108: 989–92.

[4] Suk WA, Ruchirawat KM, Balakrishnan K, Berger M, Carpenter D, Damstra T, et al. Environmental threats to children's health in Southeast Asia and the Western Pacific. Environ Health Perspect 2003; 111:1340-7.

[5] Kalantzi OI, Alcock RE, Johnston PA, Santillo D, Stringer RL, Thomas GO, et al. The global distribution of PCBs and organochlorine pesticides in butter. Environ Sci Technol 2001; 35: 1013–8.

[6] Ratan SK, Rattan KN, Pandey RM, Singhal S, Kharab S, Bala M, et al. Evaluation of parental folate, vitamin B_{12}, fluoride and homocysteine levels in the newborns with neural tube defects (NTD). 20th Congress of Asian Association of Pediatric Surgeons (AAPS) with WOFAPS Executive Meet, New Delhi. November 12-15, 2006, p59.

[7] Ratan SK, Rattan KN, Pandey RM, Singhal S, Kharab S, Bala M, et al. Evaluation of the levels of folate, vitamin B_{12}, homocysteine and fluoride in the parents and the affected neonates with neural tube defect and their matched controls. Pediatr Surg Int 2008; 24: 803-8.
[8] Sandhu R, Lal H, Kundu ZS, Kharb S. Serum fluoride and sialic acid levels in osteosarcoma. Biol Trace Elem Res 2011; 144: 1-5.
[9] Kharb S, Sandhu R, Kundu ZS. Fluoride levels and osteosarcoma. South Asian J Cancer 2012; 1: 76–7.
[10] Huo Daijei. Further observation of radiological changes of endemic food-borne skeletal fluorosis. Fluoride 1984; 17: 9-14.
[11] Hanley TR Jr, Calhoun LL, Kociba RJ, Greene JA. The effects of inhalation exposure to sulfuryl fluoride on Fetal development in rats and rabbits. Fundam Appl Toxicol 1989; 13: 79-86.
[12] Venkateswarlu P. Evaluation of an analytical method for fluorine in biological and related materials. J Dent Res 1990; 69: 514–21.
[13] Guan Zhongzhong. Research into the DNA and RNA content of the cerebellum of chronically fluoride poisoned rats. J Guizhou Medical College 1987; 12: 104.
[14] Barbier O, Arreola-Mendoza L, Del Razo LM. Molecular mechanisms of fluoride toxicity. Chem Biol Interact 2010; 188: 319-33.
[15] He H, Chen ZS, Liu XM. The influence of fluoride on the human embryo. Chinese J Control Epidemic Diseases1989; 4:136-7.
[16] Erciyas .K, Sarikaya .R. Genotoxic evaluation of sodium fluoride in the Somatic Mutation and Recombination Test (SMART). Food Chem Toxicol 2009; 47: 2860-2.
[17] Whiting P, McDonagh M, Kleijnen J. Association of Down's syndrome and water fluoride level: a systematic review of the evidence. BMC Public Health 2001; 1:6. DOI: 10.1186/1471-2458-1-6.
[18] Tsutsui T, Tanaka Y. Matsudo Y. Takahashi M. No increases in chromosome aberrations in human diploid fibroblasts following exposure to low concentrations of sodium fluoride for long times. Mutat Res 1995; 335: 15-20.
[19] Tsutsui .T, Suzuki N, Ohmori .M, Maizumi .H. Cytotoxicity, chromosome aberrations and unscheduled DNA synthesis in cultured human diploid fibroblasts induced by sodium fluoride. Mutat Res 1984; 139: 193-8.
[20] Aardema MJ1, Gibson DP, LeBoeuf RA. Sodium fluoride-induced chromosome aberrations in different stages of the cell cycle: a proposed mechanism. Mutat Res 1989; 223:191-203.
[21] Freni SC. Exposure to high fluoride concentrations in drinking water is associated with decreased birth rates. J Toxicol Environ Health 1994; 42:109-21.

