AN EPIDEMIOLOGICAL STUDY ON HEALTH EFFECTS BY DIOXIN IN VIETNAM INTRODUCTION OF NEW "HOT SPOT" AREA

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Abstract

Introduction This study is to assess the relationship between AO/dioxin exposure and human health effects in a dioxin hot spot and non-exposed areas in Vietnam. *Materials and Methods* The epidemiological study has been carried out in both areas. The subjects consist of 58 lactating females in hot spot area and 53 lactating females in non-exposed, aged 20-30 years old and their infants aged 4-16 weeks old in each area. Information about risk factors for disease was obtained through interviews to mothers. These mothers have taken saliva to analyze hormone. The measurement data on bodies of mothers and their infants were compared between two areas. *Results and Discussion* The results show that prevalence of low hormone values in mothers between hot spot and non-exposed areas are not significant. The cortisone in saliva is good relation with that in breast milk samples of Vietnamese mothers. Significant differences were not shown between the salivary hormone levels of mothers in hot spot and non-exposed areas. However, the prevalence of low estradiol level was detected higher in hot spot area than in non-exposed area. Further research should be carried out to clarify the association between dioxin exposure and human health effects.

Introduction

During the Vietnam war, between 1962 and 1971 the US military initiated use of herbicides in Viet Nam for general defoliation and crop destruction through a program codenamed Operation Ranch Hand[1]. Over 80 million litres of herbicide were used in Viet Nam. Sixty-one percent of the chemical herbicide used was Agent Orange from 1965 to 1970. Agent Orange mixture contained the highly toxic chemical 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD). It was estimated that Southern Vietnam has been contaminated to perhaps over 600 kg of dioxin[2]. TCDD is one of 75 congeners of Dioxin that are created by change position and number of Chlorine atoms around in Dioxin structure. Several TCDD contamination episodes have occurred, resulting in identification of health effects attributed to high-dose exposure to TCDD, including the skin disorders and liver damage. Adverse reproductive effects observed in these populations included increases in spontaneous abortions, low birth weight, growth retardation, and hypoplastic deformed nails[3]. TCDD is known as risk factors for cancer[4], birth defect at micro responsible dose. Hormones can be altered after low-dose exposure to TCDD and fetal lethality can occur after high-dose exposure. There is an association between maternal TCDD exposure in animal studies and adverse birth outcomes including increased fetal mortality[5], immune deficiency decreased estradiol and progesterone levels[6], altered serum testosterone level[7] and reduced fertility and fecundity[8]. 10-80 Division, Vietnam and Hatfield Consultants Ltd., a Canadian environmental consulting company, have demonstrated that aerially sprayed regions of the Aluoi Valley do not retain high levels of TCDD, given years of tropical rains, erosion, chemical breakdown, and other environmental factors[9]. However, areas of concern include those where Agent Orange and other defoliants were spilled, loaded onto aircraft, applied by truck-mounted sprayers, or transported. The resultant dioxin loading to soils near former military installations was significantly higher than that resulting from aerial applications, and continue to exist as dioxin hot spots or dioxin reservoirs to this day[10]. From October 2002 to December 2005, a project for searching dioxin hot spots was carried out in provinces of southern Vietnam. The results showed that dioxin contaminations in soil and sediment were higher than standards, and US bases in Bien Hoa, Da Nang and Phu Cat were specifically identified as hot spots. The significant hot spots of Da Nang, Phu Cat and Bien Hoa

exceed soil/sediment guidelines for many western countries/jurisdictions. Bien Hoa had the highest level of dioxin toxicity (833 pg/g toxic equivalents - TEQ), with a 2,3,7,8-TCDD reading of 797 pg/g (sediment)[11]. In this project, Hatfield consultant and 10-80 Division carried out analysis 18 samples in Phu Cat. Highest sediment sample of dioxin level is 201 pg/g and soil sample is 169 pg/g. The highest sediment TCDD level was recorded at one location (194 pg/g). This location was downstream of a dioxin mitigation site established by Vietnamese authorities. Many current studies reporting dioxin contamination in humans or in the environment in herbicide sprayed and dioxin hot spot areas, but there were not many studies relate to hormone levels in the saliva of mothers. In this paper, our aim was to assess the relationship between AO/dioxin exposure and human health effects in a hot spot and non-exposed areas in Vietnam.

Materials and methods

The study was implemented in Phu Cat district in Binh Dinh province and Kim Bang district in Ha Nam province. Phu Cat airbase was a Ranch Hand site during the war, it is one of the three dioxin hot spots in Vietnam. There was confirmed herbicide storage, loading and plane washing at Phu Cat. The population lives in and around the Phu Cat airbase before and end of the American war. The control site was Kim Bang district in northern Vietnam, which did not experience herbicide operations during the war. The subjects consist of 58 lactating females in hot spot area and 53 lactating females in non-exposed, aged 20-30 years old and their infants aged 4-16 weeks old in each area almost all mothers lactating were selected. The mothers have interviewed to determine parity, reproductive and residential history and other specific risk factors for pollution exposure. Salivary samples were collected from volunteer donors in two areas. The mouth of mothers was rinsed with water and then saliva was directly placed in a Bakelite test tube (15 mL) and stored at -70°C until analysis. Analyses were conducted on 60 samples; 41 from Phu Cat district and 19 from Kim Bang district. Mothers were asked to provide information on age, number of infants and number of infants which they have breastfeed (including children of relatives, etc.). The measurement data on bodies of mothers and their infants were compared between two areas. The medical ethical committee of Kanazawa University approved this study, and informed consent was obtained from each participant.

Statistical method

The statistical comparisons were made using chi-square and Wilcoxon signed rank test statistics for categorical variables and Turkey-Kramer HSD test for continuous variables (α=0.05 level). Odds ratios (OR) and 95% confidence intervals (95% CI) based on the chi-square and Wilcoxon signed rank test were calculated on epidemiology data. All statistical analyses were performed using JMP®6 software (SAS Institute, Japan) for analysis.

Results and discussion

In this paper, we present results of epidemiology study and salivary hormone level of lactating females who live in a dioxin hot spot and non-exposed area. We also present that the measurement results on bodies of mothers and their infants were compared between two areas. The results of research on 58 infants in hot spot area and 53 infants in non-exposed area show that mean subject age was 10.7±3.1 weeks for infants in hot spot area and 12.0±2.7 weeks for infants in non-exposed. The results in Table 1 showed that weight and chest circumferences of infants in hot spot area were significantly lower than those in non-exposed area. There was no significant difference between infant height, head circumference, abdomen circumference and born weight in both areas. Dioxins are known to affect infant growth and neurodevelopment in both humans and animals. A recent study in Japan also showed that decreased infant head circumference at birth without decreased weight was associated with maternal exposure to 2,3,7,8-TCDD[12]. Three infants birth defect were detected (5.3%) in hot spot area but no significant difference was shown. The present and past diseases showed no significant difference in both areas. Either general characteristic of mothers in this study are presented in Table 2. The maternal height, weight, BMI, alcohol habit, smoking habit, reproductive failure, past disease and family birth defect were not significantly different in both areas. Present maternal diseases in hot spot area were 21 mothers (36.2%) higher than non-exposed with 9 mothers (17%), statistically significant differences with $p<0.05$. Present family diseases in hot spot area with 19 mothers (32.8%) were significantly higher than those in non-

exposed area with 6 mothers (11.3%), statistically significant differences with $p<0.01$. Several current decades, researchers have had many studies on humans and other vertebrates and made clear that dioxins have been shown to be risk factors for reproductive and developmental abnormalities[13], spontaneous abortions, low birth weight, increased fetal mortality. The maternal eyesight of both eyes was not significantly different in both areas. The results of research salivary hormone levels on 41 mothers in dioxin hot spot area and 19 mothers in non-exposed area are presented in Table 3. Prevalence of low hormone values in mothers between hot spot and non-exposed areas is not significant. However, the prevalence of low estradiol level was detected higher in hot spot area than in non-exposed area. In several current study showed that TCDD decreases estradiol production by human luteinized granulose cell (hLGC) has been reported[14,15,16], and a study previously demonstrated that this TCDD effect on E2 production was not due to a direct effect on TCDD on the cytochrome P450 aromatase (P450arom)[15]. Figure 1 presents the cortisone in breast milk samples is good relation with saliva of Vietnamese mothers. Figure 2 shows that significant differences were not shown between the salivary hormone levels of mothers in hot spot and non-exposed areas. Further research should be carried out to clarify the association between dioxin expose and human health effects. Hot spot areas may be an especially advantageous location for such studies because of the extremes in dioxin levels.

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Table 1: Comparison of characteristics of infants between hot spot and non-exposed areas

		Hot spot area		Non-exposed area		p-value
		N	Mean ± SD (%)	N	Mean ± SD (%)	
Infant age	(Weeks)	58	10.7 ± 3.1	53	12.0 ± 2.7	*[1)]
Infant height	(cm)	58	60.7 ± 3.1	53	60.8 ± 2.8	n.s[1)]
Infant weight	(g)	58	5648.4 ± 941.6	53	5874.3 ± 885.0	*[1)]
Head circumference	(cm)	58	39.7 ± 1.7	53	40.0 ± 1.9	n.s[1)]
Chest circumference	(cm)	58	39.8 ± 2.8	53	40.7 ± 2.2	*[1)]
Abdomen circumference	(cm)	58	38.6 ± 2.7	53	39.3 ± 2.8	n.s[1)]
Infant born weight	(g)	57	3178.9 ± 462.4	52	3252.9 ± 399.7	n.s[1)]
Birth defect	Yes	58	3 (5.2%)	53	0 (0.0%)	n.s[2)]
Present disease	Yes	58	6 (10.3%)	53	9 (17%)	n.s[2)]
Past disease	Yes	58	2 (3.45%)	53	2 (3.8%)	n.s[2)]

Data are means ± SD or number (%)

[1)]Wilcoxon signed rank test, [2)]Chi-square, n.s: not significant, *: $p<0.05$

Table 2: Comparison of characteristics of mothers between hot spot and non-exposed areas

		Hot spot area		Non-exposed area		p-value
		N	Mean ± SD (%)	N	Mean ± SD (%)	
Maternal age	(years)	58	25.8 ± 3.7	53	25.1 ± 2.8	n.s[1)]
Maternal hieght	(cm)	58	152.0 ± 6.0	53	152.5 ± 5.0	n.s[1)]
Maternal weight	(kg)	58	48.4 ± 6.5	53	48.7 ± 5.0	n.s[1)]
Maternal BMI	(kg/m^2)	58	20.9 ± 2.2	53	20.9 ± 1.8	n.s[1)]
Maternal right eye		58	1.2 ± 0.4	53	1.2 ± 0.4	n.s[1)]
Maternal left eye		58	1.2 ± 0.4	53	1.3 ± 0.4	n.s[1)]
Family income	(VND)	55	2358181.8 ± 2180272.7	51	2086274.5 ± 1126058.5	n.s[1)]
Alcohol habit	Yes	58	2 (3.45%)	53	0 (0.0%)	n.s[2)]
Smoking habit	Yes	58	0 (0.0%)	53	2 (3.8%)	n.s[2)]
Present disease	Yes	58	21 (36.2%)	53	9 (17%)	*[2)]
Past disease	Yes	56	20 (35.7%)	53	13 (24.5%)	n.s[2)]
Reproductive failure	Yes	58	9 (15.5%)	53	10 (18.9%)	n.s[2)]
Present family disease	Yes	58	19 (32.8%)	53	6 (11.3%)	**[1)]
Family birth defect	Yes	58	2 (3.45%)	53	0 (0.0%)	n.s[2)]

Data are means ± SD or number (%), BMI = body mass index

[1)]Wilcoxon signed rank test, [2)]Chi-square, n.s: not significant, *: $p<0.05$, **: $p<0.01$

Table 3: Comparison of prevalence of low hormone values in mothers between hot spot and non-exposed areas

		Hot spot area (n=41)	Non-exposed area (n=19)	p-value
Cortisol (pg/ml)	A (>1000)	34 (82.9%)	18 (94.7%)	0.21
	B (<1000)	7 (17.1%)	1 (5.3%)	
Cortisone (pg/ml)	A (>8000)	33 (80.5%)	14 (73.7%)	0.55
	B (<8000)	8 (19.5%)	5 (26.3%)	
Cortisol/Cortisone (pg/ml)	A (>0.12)	34 (82.9%)	17 (89.5%)	0.5
	B (<0.12)	7 (17.1%)	2 (10.5%)	
Dehydroepiandrosterone (pg/ml)	A (>50)	37 (90.2%)	18 (94.7%)	0.55
	B (<50)	4 (9.8%)	1 (5.3%)	
Androstenedione (pg/ml)	A (>20)	38 (92.7%)	18 (94.7%)	0.76
	B (<20)	3 (7.3%)	1 (5.3%)	
Estradiol (pg/ml)	A (>0.05)	32 (78.05%)	18 (94.7%)	0.1
	B (<0.05)	9 (21.95%)	1 (5.3%)	
Progesterone (pg/ml)	A (>0.01)	41 (100%)	18 (94.7%)	0.13
	B (<0.01)	0 (0.0%)	1 (5.3%)	
Testosterone (pg/ml)	A (>1.0)	40 (97.6%)	18 (94.7%)	0.57
	B (<1.0)	1 (2.4%)	1 (5.3%)	

[1)] Chi-square

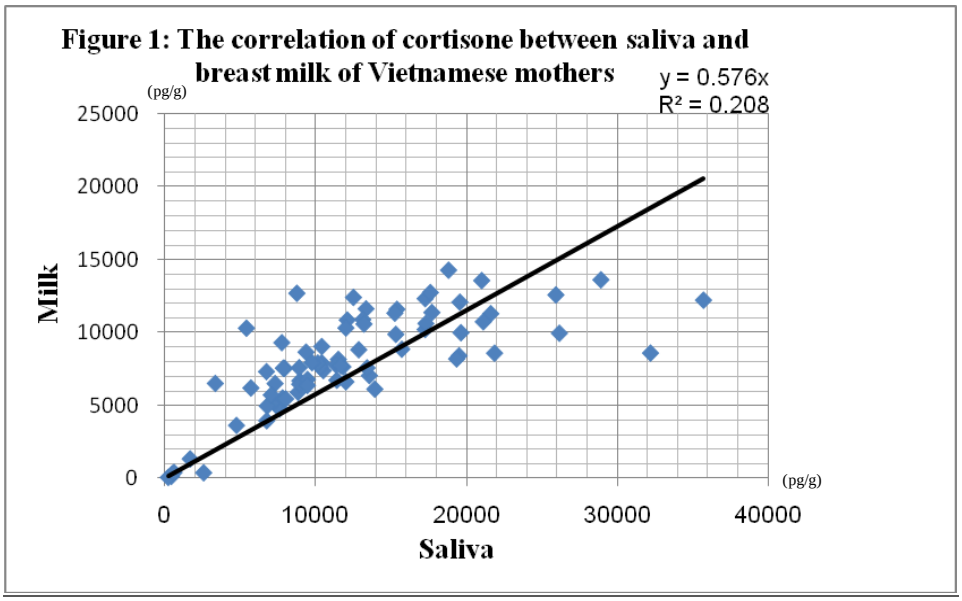

Figure 2: Comparison of the salivary hormone levels in mothers between hot spot and non-exposed areas