

Epidemiology and risk factors of retinoblastoma in Chongqing area

Yu-Qiong Yang, Jia Li, Hong-Feng Yuan

Department of Ophthalmology, Institute of Surgery Research, Daping Hospital, Third Military Medical University, Chongqing 400042, China

Co-first authors: Yu-Qiong Yang and Jia Li

Correspondence to: Hong-Feng Yuan. Department of Ophthalmology, Institute of Surgery Research, Daping Hospital, Third Military Medical University, Chongqing 400042, China. yhf871@sina.cn

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Abstract

• **AIM:** To investigate the risk factors of retinoblastoma in Chongqing and offer the bases for scientific measures.

• **METHODS:** One hundred and thirty-three retinoblastoma patients were taken as case group, and 133 children were taken as control group in 1:1 ratio. The relevant factors were collected from a questionnaire survey which was made by our research group. First, Chi-square and t -test were used to discuss the various factors, and then the logistic regression analysis was made by statistics software SPSS17.0 based on the result of 266 people.

• **RESULTS:** Single factor analysis results showed the differences between the two groups were statistically significant ($P < 0.05$) in 17 factors which were education level of their parents, occupation of their parents, exposure to harmful chemicals of their parents 6mo before pregnancy, the history of pelvic inflammatory disease of mother before pregnancy, childbearing history of their parents, pregnant age of their parents, the medication history of father 6mo before pregnancy, living place of their parents, the pollution living place of mother, hobbies of their parents. Multivariate analysis showed that the living place of parents, mother who feed pets before pregnancy, and exposure to harmful chemicals of father before pregnancy were the independent risk factors of retinoblastoma ($P < 0.05$).

• **CONCLUSION:** The living place of parents, mother who feed pets before pregnancy, exposure to harmful chemicals of father before pregnancy were the risk factors of retinoblastoma which affects the occurrence of retinoblastoma in a certain extent.

• **KEYWORDS:** Chongqing area; retinoblastoma; epidemiology; risk factors

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INTRODUCTION

Retinoblastoma (RB) is the most common primary intraocular malignancy usually occurring in childhood, which lead to blind of children by 5%^[1-3]. It's autosomal dominant genetic disease arise from the inner neuroepithelial layers of the embryonic optic cup. RB occurs in 4 to 8wk of gestation that is the formation stage of retina, continues grow to infancy, and presents clinical feature in 6mo^[4].

Knudson^[5] proposed secondary mutation theory as the mechanisms of retinoblastoma, is now widely accepted. Little is known about the risk factors of the life-style and environment related to retinoblastoma. Orjuela *et al*^[6] reported that decreased intake of vegetables and fruits during pregnancy and the consequent decreased intake of nutrients such as folate and lutein/zeaxanthin, necessary for DNA methylation, synthesis, and retinal function, may increase risk for having a child with sporadic retinoblastoma. Other risk factors of retinoblastoma is unknown, so that in order to provide a scientific basis for prevention and treatment of retinoblastoma, a case control research had been used to detect the related risk factors of retinoblastoma.

SUBJECTS AND METHODS

Subjects A case-control study was conducted in Chongqing. A total of 133 patients with retinoblastoma were recruited to the study when they entered the Department of Ophthalmology at Daping Hospital of Third Military Medical University, Xi'nan Hospital of Third Military Medical University, the Children's Hospital Affiliated to Chongqing Medical University. One hundred and thirty-three cases of healthy children with same gender and age were taken as control group in 1:1 ratio. Informed consent was obtained from all individual participants included in the study. The study protocol and informed consent documents were approved by the Ethics Committee of Research Institute of Surgery and Daping Hospital, Third Military Medical University.

Diagnostic Criteria 1) All the patients were examined by Retcam II, the result of fundus examination conforms to the Intraocular International Retinoblastoma Classify (IIRC). 2)

Color Doppler suggests blood flow signals in the tumor. Computed tomography (CT) or magnetic resonance imaging (MRI) suggests calcification in tumor.

Data Collection The medical records of case group and control group were obtained from unified questionnaire from their parents. All the investigation through face-to-face or telephone interviews were done by one trained investigator.

Analysis The following items obtained from questionnaire were analyzed as risk factors: family general condition classified into groups of incomes, education level, occupation of parents; family history classified into groups of retinoblastoma history, genetic disease, tumor; past and contact history of parents classified as radiation exposure before pregnancy, harmful chemicals exposure before pregnancy, pet contact during pregnancy, human papilloma virus (HPV) infection, menstrual, cervicitis, pelvic infection; obstetrical history of parents classified as marital history, childbearing; the situation of parents during pregnancy classified as age, abortion, pregnancy reaction, medication history, cold history; habits of parents classified as residence (city or village), noise, pollution, diet, hobbies, cigarette smoking, alcohol drinking.

Statistical Analysis Univariate logistic regression analysis was performed to examine the predictive effect of each factor on the risk for retinoblastoma. The 19 potential risk factors for case group were compared with those of control group using Fisher's exact test, with the software package SPSS 13.0; $P < 0.05$ was considered statistically significant. Significant risk factors were entered into a forward selection multivariate logistic regression analysis. The involvement of the various factors in the development of retinoblastoma was analyzed using multiple logistic regression analysis with the SPSS statistical software package. The logistic model is a multivariate analysis model by which the odd ratio (OR, a good approximate value for the relative risk) of risk factors is calculated by logit transformation of the probability of the development of the end point. Using this model, the degree of involvement of each factor in the occurrence of RB could be estimated. When the 95% confidence interval (CI) of the relative risk of a given factor does not include, the value is significant ($P < 0.05$).

RESULTS

Demographic Characteristic of the Two Groups The design was comply to the requirement in both group which is comparable.

In the research, the age was not significantly difference between case group and control group [51.51 ± 55.98 (SD) vs 64.89 ± 48.27 (SD)mo, $P > 0.05$].

Analysis of Risk Factors Risk factors of retinoblastoma included education level of their parents, occupation of their parents, exposure to harmful chemicals of their parents 6mo before pregnancy, pelvic inflammatory history of mother

before pregnancy, childbearing history of their parents, pregnant age of their parents, the medication history of father 6mo before pregnancy, residence of their parents, the pollution living place of mother, hobbies of their parents. Multivariate analysis showed that the residence of parents, mother who feed pets before pregnancy, exposure to harmful chemicals of father before pregnancy were significantly difference between case group and the controls ($P < 0.05$). The results are shown in Table 1.

Multivariate Logistic Regression Analysis of Risk Factors

On multivariate analysis, living place (OR=0.262, 95% CI: 0.146 to 0.469), mother who feed pets before pregnancy (OR=0.428, 95%CI: 0.207 to 0.887), exposure to harmful chemicals of father before pregnancy (OR=0.165, 95%CI: 0.076 to 0.357) were the independent risk factors of retinoblastoma ($P < 0.05$). The results are shown in Table 2.

DISCUSSION

This study demonstrates that retinoblastoma results for the interaction of many factors, which includes residence of parents, mother who feed pets during pregnancy, exposure to harmful chemicals of father 6mo before pregnancy. Their OR values were 0.262, 0.428, 0.165, which indicated that the incidence of RB respectively increased by 3.82 (1/0.262) times, 2.34 (1/0.428) times and 6.06 (1/0.165) times. Father exposed to harmful chemicals 6mo before pregnancy has the greatest effect on the incidence of RB, second for the parents' living place, third for mother who feed pets during pregnancy. The results showed that the occurrence of retinoblastoma is closely related to the living environment. The analysis of risk factors and preventive strategy are shown as follows.

Father Exposed to Harmful Chemicals 6mo Before Pregnancy

About 80% tumors are caused by the external environment, two-thirds are chemical carcinogens. The research showed that father of the patients were contact with paint, leather equipment, decoration, electronic accessories at work, which included 18 (13.53%) cases exposed on benzene, 8 (6.02%) cases exposed on formaldehyde, 5 (3.76%) cases contact with zinc, manganese and other heavy metals. The WHO has classified Benzene (BZ) as a known human carcinogen based upon both human and animal evidence, used in the manufacture of plastics, detergents, pesticides, and other chemicals [7-10]. BZ is able to induce many different kinds of genetic damage such as point mutations, DNA adducts, oxidative DNA damage, structural chromosomal aberrations (CA), numerical CA, DNA methylation, which may cause cancer [11-13]. Formaldehyde is another carcinogen which is genotoxic and induces both DNA damage and chromosome changes, frequently expressed as DNA-protein crosslinks (DPCs), CA, sister chromatid exchanges (SCEs), and micronuclei (MN). A large number of studies have demonstrated that these

Risk factors of retinoblastoma

Table 1 Risk factors of retinoblastoma

Items	Classification	Case group (n=133)	Control group (n=133)	χ^2	<i>n</i> (%) <i>P</i>
Basic situation of families					
Education level of father	≥High school	51 (38.35)	102 (76.69)	40.0177	<0.001
	<High school	82 (61.65)	31 (23.31)		
Education level of mother	≥High school	51 (38.35)	99 (74.44)	32.5299	<0.001
	<High school	82 (61.65)	34 (25.56)		
Occupation of father	Worker	17 (12.78)	43 (32.33)	79.3578	<0.001
	Farmer	112 (84.21)	55 (41.35)		
	Other	4 (3.01)	35 (26.32)		
Occupation of mother	Worker	21 (15.79)	54 (40.60)	56.3142	<0.001
	Farmer	94 (70.68)	43 (32.33)		
	Other	18 (13.53)	36 (27.07)		
Past and contact history					
Exposure to harmful chemicals of father 6mo before pregnancy	N	56 (42.11)	10 (7.52)	42.6406	<0.001
	Y	77 (57.89)	123 (92.48)		
Exposure to harmful chemicals of mother 6mo before pregnancy	N	89 (66.92)	10 (7.52)	26.8602	<0.001
	Y	44 (33.08)	123 (92.48)		
Pelvic inflammatory history of mother before pregnancy	N	119 (89.47)	130 (97.74)	7.6036	0.0058
	Y	14 (10.53)	3 (2.26)		
Obstetrical history					
Father	1	61 (45.86)	93 (69.92)	15.7922	0.0001
	≥2	72 (54.14)	40 (30.08)		
Mother	1	64 (48.12)	96 (72.18)	16.0604	0.0001
	≥2	69 (51.88)	37 (27.82)		
Characteristics of pregnancy					
Pregnant age of father	≤20	5 (3.76)	2 (1.50)	5.0180	0.0251
	20-35	75 (56.39)	61 (45.86)		
	≥35	53 (39.85)	70 (52.63)		
Pregnant age of mother	≤20	23 (17.29)	9 (6.77)	7.1077	0.0286
	20-30	81 (60.90)	94 (70.68)		
	≥30	29 (21.80)	30 (22.56)		
Medication history of father before pregnancy	N	121 (90.98)	131 (98.50)	7.5397	0.006
	Y	12 (9.02)	2 (1.50)		
Living, eating and habits					
Living place of father	City	51 (38.35)	103 (77.44)	43.8063	<0.001
	Suburban or village	82 (61.65)	30 (22.56)		
Living place of mother	City	49 (36.84)	105 (78.95)	49.8884	<0.001
	Suburban or village	84 (63.16)	28 (21.05)		
Mother who live in polluted area	No pollution	115 (86.47)	124 (93.23)	4.1814	0.0409
	Pollution	18 (13.53)	9 (6.77)		
	Table games	25 (18.80)	12 (9.02)		
Habit of father	Reading	2 (1.50)	9 (6.77)	32.2861	<0.001
	Surfing on line	24 (18.05)	56 (42.11)		
	Open field activity	74 (55.64)	42 (31.58)		
Habit of mother	Table games	12 (9.02)	2 (1.50)	24.0151	0.0001
	Reading	7 (5.26)	8 (6.02)		
	Surfing on line	18 (13.53)	48 (36.09)		
Father who feed pets	Open field activity	89 (66.92)	67 (50.38)	11.333	0.001
	N	105 (78.95)	124 (93.23)		
Mother who feed pets	Y	28 (21.05)	9 (6.77)	13.029	0.000
	N	93 (69.92)	117 (87.97)		
	Y	40 (30.08)	16 (12.03)		

Y: Yes; N: No.

alterations could cause cancer [14-16]. A report demonstrated that certain environment caused DNA methylation which led

to changes in epigenetics and then passed on to offspring [17-19]. In our survey, father exposed to chemicals is one of the risk

Table 2 Logistic regression analysis to identify risk factors associated with retinoblastoma

Risk factors	B	S.E.	Wald	df	Sig.	Exp (B) (95%CI)
Living place	-1.339	0.297	20.324	1	0.000	0.262 (0.146 to 0.469)
Mother who feed pets before pregnancy	-0.848	0.372	5.204	1	0.023	0.428 (0.207 to 0.887)
Exposure to harmful chemicals of father before pregnancy	-1.803	0.395	20.865	1	0.000	0.165 (0.076 to 0.357)
Constant	1.083	0.198	29.963	1	0.000	

B: Regression coefficient; S.E.: Standard error; Wald: Chi-square value; df: Degree of freedom; Sig.: Significance conspicuousness; Exp (B) (95%CI): Odd ratio values (dominance ratio).

factors of retinoblastoma that may because father exposed to chemicals induce to genetic damage which destroyed the normal transcription activity of the genes that cause epigenetic changes and pass on to offspring.

The Living Place of Parents In our survey, 51 cases of patients' father and 49 cases of patients' mother living in the city, accounted for 38.35% and 36.84% respectively, which is lower than the urbanization rate of permanent resident population (56.98%) in Chongqing. Eleven cases of father and 10 cases of mother living in the noise pollution areas, accounted for 8.8% and 8.0% respectively. Seventeen cases of father and 18 cases of mothers living in the pollution area, accounted for 13.6% and 13.53% respectively. The pollution sources are mainly from types of mines, waste dumps (station), high tension line and satellite signals. From the point of parents' occupation, more than 50% were engaged in primary industry, and at the end of 2012 first industry professionals accounted for only 4.48% in Chongqing. The rate of respondents engaged in the primary industry is significantly higher than the average level of Chongqing, so was the urbanization rate. The result reveals that respondents were derived from the underdeveloped areas, the morbidity of RB is related to the economic level. The more backward areas, the higher the incidence of RB will happen; the more developed economic, the less incidence of RB occurs. This may be related to the lack of nutritional supplement, medical care and service that pregnant women got during pregnancy.

Mother Feed Pets During Pregnancy Our survey shows that 30.08% patients' mother raising pets during pregnancy, mainly for cats and dogs. Cats are carriers of toxoplasma. Pregnant women who infected with toxoplasma would cause miscarriage, premature birth, stillbirth and teras. The average infection rate of toxoplasma in domestic pregnant women is about 8.16%^[20], which will cause congenital malformation. These abnormalities including anencephaly, anophthalmos, acheiropodia, no anus, auricle defect, genital defect and toes hypoplasia. Until recently, there is no report about toxoplasma infect will cause retinoblastoma. Shetty *et al*^[21] gave evidence that HPV is one of the nosogenesis of sporadic retinoblastoma, and in our research there is one case whose mother infected HPV during pregnancy. Mother who feed pets during pregnancy is one of the risk factors in our survey, that may due to the infection of viruses or parasite while contacting with pets which is the carrier of

various of viruses or parasite, ultimately causing chromosome loss or mutations appeared in the process of fetal development.

In conclusion, due to the high risk factors of retinoblastoma, we should adopt integrated interventions. A targeted prevention health education of retinoblastoma can improve the health care knowledge and the awareness of cancer of people, changing their lifestyle and habits in order to reduce morbidity, mortality and improve the quality of life. 1) In everyday life or work, we should avoid exposure to toxic chemicals; if it must be, protective measures should be taken, such as wearing protective clothing to avoid direct contact with toxic chemicals, reduce exposure time, regularly body check and so on. 2) In diet, expert suggests that less pickled vegetables, leftovers and fried foods should be taken; more green vegetables, fruit and other food which could help prevent cancer should be taken. Take enough folic acid and necessary nutrition that fetal development needed during pregnancy. Keep fresh light diet, good mood, no smoking, less drinking, fit exercise and make regular prenatals check. 3) Protecting the environment, avoiding the emission of harmful substances. For polluting areas, strategies should be taken to solve the problems. 4) For pregnant women, direct or indirect contact with pets should be avoided, prenatal examination with various of virus should be taken conventionally.

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