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Sero-positivity rate of rubella antibodies in Iraqi autistic children

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Abstract

Background: Autism Spectrum Disorder (ASD) is a complex neurodevelopment disease resulting from complex interaction between genetic and environmental factors.
Aim: This study aimed to detect if there is association between rubella virus and ASD.
Materials and methods: This case control study conducted from November 2016 to June 2017, blood samples were collected from forty ASD patients whom attended to the central teaching hospital for children in Al-Eskan city also this study included forty apparently healthy children as a control group. Enzyme Linked Immuno-Sorbent Assay (ELISA) technique was used for investigation of anti rubella antibodies in sera samples.
Results: The present results demonstrated significant difference regarding IgG sero-positivity of rubella virus between studied groups, there was 6 cases among 40 autistic patients with IgG sero-negative while all the controls have IgG positive (P value =0.025).
Conclusion: Individuals with sero-negative of anti-rubella IgG may consider at risk for ASD.

Key words: Autism, IgG, Rubella.

INTRODUCTION

Autism spectrum disorder (ASD) is invasive developmental disorder characterized by deficits in social communication skills with repetitive, restricted or stereotyped behaviors (1). ASD apparently have a multifactorial etio-pathogenesis, resulting from a very complex interaction between genetic and environmental factors (2).

Adverse intrauterine environment resulting from maternal bacterial and viral infections during pregnancy represent a significant risk factor for some neuropsychiatric disorders including ASD (3). Many but not all population or case control studies have shown a slight to moderate association of maternal infections with ASD. The controversy might be related to the fact that only several specific maternal infections are associated with ASD. These are mainly rubella, cytomegalovirus (CMV) and possibly influenza. There were, however, case control studies that did not find any association of ASD with either maternal viral or bacterial infections (4).

Maternal fever during pregnancy has been linked to increased risk of ASD (5). Maternal antibodies increase in response to viruses or bacteria and may cross the placenta and disrupt fetal neurodevelopment by cross-reacting with fetal brain antigens via molecular mimicry (6).

Rubella infection in pregnant women can lead to fetal death or severe life-long disabilities as congenital rubella syndrome (7). In spite of the availability of an effective vaccine for rubella since the 1960s, the virus is still a global health concern with over 100,000 babies born with congenital rubella syndrome every year (8).

Children with congenital rubella syndrome have 200 times the prevalence of ASD at a time when the diagnosis of ASD was much more limited. Although rubella is one of many maternal infections with possible links to ASD, no other infection boasts such an increased prevalence (9). In Iraq there is increasing in diagnosis of autism on the other hand data on rubella infection and autism are lacking, so this prompted us to conduct this study.

MATERIALS AND METHODS

A total of 40 Iraqi children with ASD (7 females and 33males) were included in this study. They were among patients attending to central teaching hospital for children, during the period from December 2016 till June 2017, their age ranged from 3 to12 years. All patients were diagnosed by psychiatric physician according to the criteria for diagnosis of American Psychiatric Association's Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition (DSM-5).

In addition, forty apparently healthy children, consist of 9 females and 31males, their age ranged from (3-12) years were involved in the current study as control group.

Two milliliters of venous blood was drawn from each subject under aseptic technique. The sample was transferred to gel tube, then centrifuged at 2500 rpm for 10 minutes and the separated serum was divided into several aliquots in an eppendorf tube, and immediately frozen at -20 c◦ till further use to avoid repeated thawing and freezing.

Detection of IgG/IgM antibodies to rubella virus By Combo Rapid test cassette:-

The Rubella IgG/IgM Combo Rapid test Device (serum /plasma) is a qualitative, lateral flow immunoassay for the detection of IgG and IgM antibodies to rubella virus in serum or plasma specimens.

Assay Procedure: according to manufacturer's instruction.

Detection of IgG antibodies to rubella virus by ELISA

The RUBG01 Rubella IgG kit is based on the ELISA technique.

Assay Procedure: according to manufacturer's instruction.

RESULTS

In this study the age of patients with autism spectrum disorder ranged between 3-12 years with a mean age 4.75±1.71 years and the mean age of healthy control was 4.70±2.23 as shown in table (1). In addition, there was male predominance among patients, about 33(82.5%) of patients were males and 7(17.5%) were females as explained in Table (2).A comparison between autistic patients and disease free children according to rubella IgG and IgM showed statistically significant difference in sero-positivity of IgG but not IgM antibodies as in table (3)

Table (1) Comparison between patients and control group according to the age by t- test

Parameters		Study groups		p value
		Patients No.40	Controls No.40	
Age (years)	Mean	4.75	4.70	0.321^{NS}
	Standard Deviation	1.71	2.23	
	Range	9.00	9.00	
	Minimum	2.00	3.00	
	Maximum	11.00	12.00	

NS: None statistical significance (p>0.05).
Comparison done by independent sample t-test

Table (2) Statistical description of sex for patients and control group by Fisher exact test

Sex	Study groups			P value
	Patients	Controls	Total	
Female	7	9	16	0.227^{NS}
	17.5%	22.5%	20.0%	
Male	33	31	64	
	82.5%	77.5%	80.0%	

NS: None statistical significance (p>0.05).
Comparison done by Fisher exact test

Table 3 Statistical description of anti-rubella IgM and IgG in patients and control group by Fisher exact test

Parameter		Study groups		Total	P value
		Patients	Controls		
Anti-rubella-IgM by rapid test#	Negative	39	40	79	0.248^{NS}
	%	97.5%	100.0%	98.8%	
	Positive	1	0	1	
	%	2.5%	0.0%	1.3%	
Anti-rubella-IgG by rapid test#	Negative	6	0	12	0.025*
	%	15.0%	0.0%	7.5%	
	Positive	34	40	68	
	%	85.0%	100.0%	92.5%	
Anti-rubella IgG by ELISA#	Negative	6	0	6	0.025*
	%	15.0%	0.0%	7.5%	
	Positive	34	40	74	
	%	85.0%	100.0%	92.5%	
Total		40	40	80	
		100.0%	100.0%	100.0 %	

NS: None statistical Significant difference (p>0.05).
*: Statistical significant difference (≤0.05)
#: Comparison done by Fisher exact test.

DISCUSSION

In this study forty Iraqi Arab patients were enrolled with age ranged from (3-12) years, mean of age was (4.75±1.71) This finding was slightly different than that reported by Sipos et al (2012) (10) who found that the children's age ranging between 2 and 14 years, with a mean = 6.46. The differences may be due to the selection criteria of the current study which consists of children aged 3-12 years old.

The American Psychiatric Association (APA) (2014) reported that autism appearing within the first three years of life (11). This result was congruent with Karst and Van Hecke (2012) who reported that the parents are informed of an autistic disorder diagnosis for their child often comes much later, when the child is at 3 years of age (12).

In current study, thirty three 33(82.5%) of patients were males and 7(17.5%) were females. This sex distribution is supported by Sipos et al. (2012) who found that their sample included 73.7% boys, and 26.3% girls (144).The result of the current study was congruent with the previous studies and the CDC (2013) which reported that the autism is five times more likely to occur in boys than in girls (13). Other studies reported that the ASDs are almost 5 times more common among boys than girls. Girls are less likely to be diagnosed with autism than boys, unless they also have intellectual or behavioral problems (14). Girls with high-functioning ASD tend to be clinically identified later than boys (15).

The prevalence of diagnosed ASDs in 2011–2012 was estimated to be 2 % in the united state of America (USA). The male to female ratio is approximately 4:1, suggesting a possible imprinting effect and/or involvement of the sex chromosomes (16).

The interpretation of higher incidence in males may be attributed to the sex chromosomal genes which have been proposed to be key players in molecular mechanisms driving females' protection from ASDs liability conferred by specific risk loci and/or by genome-wide mutational load. An early theory proposed that ASDs might be a X- linked disorder, in which females are protected from deleterious effects of X chromosomal mutations by compensatory transcription from their intact second X chromosome. Although ASD may not be X-linked in the Mendelian sense, sex chromosome complement may still modulate ASDs risk. Sex chromosome aneuploidies provide test cases for this hypothesis, with an increased rate of ASDs diagnosis in Turner syndrome (17), Klinefelter syndrome (18) and 47,XYY syndrome, but no increased rate in X chromosome trisomy (19), (20).

Regarding prenatal viral infection rubella virus have important role in activation the mother's immune response and greatly increases the risk for ASD (21).

In this study there was statistically significant difference of anti-rubella IgG. There were 6 cases of patients have no IgG against rubella virus in comparison with healthy control that all of them appeared with IgG +ve by combo rapid test and also by ELISA technique. On the other hand, there was no significant difference regarding IgM antibodies, all cases of patients and controls showed IgM -ve. These data agree with other studies that yielded compelling results of antibodies in children with autism. One of this studies noticed of 15 children with ASD, 5 of 13 had undetectable antibodies to rubella despite vaccination; in comparison, 8 neurologically typical (NT) children all had antibodies. Another study by (Libbey *et al.,* 2007), which included 33 classic autism children and 26 regressive autism children, found 15 of these 59 combined children had low or no antibodies to rubella; as compared to 2 of 49 in the control group (22) (23).

The result of current study inconsistent with Gentile *et al.,* 2013 who studied 31 children with ASD and 29 NT children that found the antibodies to rubella were similar in both groups (24).

In utero, the fetus has an immature immune system, and if CRI occurs early, then the fetus will not recognize the virus as foreign. CRI individuals develop a tolerance to rubella, in particular the E1 epitope (25), (22). Interestingly, in 10–20% of CRI infants, the rubella-specific IgG will drop to undetectable levels, and cannot be induced by boosters of rubella vaccine (26).This immune tolerance may contribute to viral persistence (23).

Two studies of antibodies in children with ASD yielded compelling results. One study showed undetectable antibodies to rubella despite vaccination; in comparison, the neurologically typical (NT) children all had antibodies (24).

Another study showed low or no antibodies to rubella; as opposed to control group, (27). Yet, in another study included children with ASD and children with NT, the antibodies to rubella were similar in both groups (28). Conclusion: Individuals with anti-rubella IgG sero-negative may consider at risk for ASD.

REFERENCES

1- American Psychological Association (APA). Diagnostic and Statistical Manual of Mental Disorders. 5th ed Washington, DC: APA; (2013).
2- Lyall K, Schmidt RJ, and Hertz-Picciotto I, Maternal lifestyle and environmental risk factors for autism spectrum disorders Int J Epidemiol, 43 (2014); 443–464.
3- Fox E, Amaral D, Van de Water J. (Maternal and fetal antibrain antibodies in development and disease. Dev. Neurobiol. 2012. 72, 1327–1334.
4- Bantavala J E, Brown D W G. Rubella. Lancet, (2004); 363, 1127–1137.
5- Boyson JE, Erskine R, Whitman MC, etal. Proc Natl Acad Sci U S A, 2002; 10;99(25):16180-5.
6- Shiroishi M, Kuroki K, Ose T. 2006; 14;281(15):10439-47.
7- Battisti AJ, Yoder JD, Plevka P, et al. Cryo-electron tomography of rubella virus. J Virol. 2012; 86 (20):11078–85.
8- Oker-Blom C, Kalkkinen N, Kaariainen L, etal. Rubella virus contains one capsid protein and three envelope glycoproteins, E1, E2a, and E2b. J Virol. 1983; 46(3):964–73.
9- Hutton J. Does Rubella Cause Autism: A 2015 Reappraisal? Front. Hum. Neurosci, 2016;10:25.
10- Sipos R, Predescu E, Muresan G, et al. The evaluation of family quality of life of children with autism spectrum disorder and attention deficit hyperactive disorder, Applied Medical Informatics. 2012;30:(1),1-8.
11- American Psychiatric Association (APA): Autism, 2014.
12- Karst SK, Van Hecke AV. Parents and family impact of autism spectrum disorder: A review and proposed model for intervention evaluation, Clin Child FamPsychol Rev. 2012; 15 (3): 247- 277.
13- Centers for Disease Control and Prevention (CDC): Data &Statistics, AutismSpectrum Disorders, 2013.
14- Dworzynski K, Ronald A, Bolton P, et al. How different are girls and boys above and below the diagnostic threshold for autism spectrum disorders? Journal of the American Academy of Child and Adolescent Psychiatry. 2012; 51 (8): 788-97.
15- Giarelli E, Wiggins LD, Rice CE, *et al*. Sex differences in the evaluation and diagnosis of autism spectrum disorders among children. Disability and Health Journal. 2010;3 (2), 107- 16
16- Nygren G, Cederlund M, Sandberg E et al. The prevalence of autism spectrum and Developmental Disorders. Journal of Autism and Developmental Disorders. 2012;42(7):1491–1497.
17- Donnelly SL, Wolport CM, Menold MM, et al. Female with autistic disorder and monosomy X (Turner syndrome): parent-of-origin effect of the X chromosome. Am J Med Genet. 2000;96(3):312–6.
18- van Rijn S, Bierman M, Bruining H, et al. Vulnerability for autism traits in boys and men with an extra X chromosome (47,XXY): the mediating role of cognitive flexibility. J Psychiatr Res. 2012;46(10):1300–6.
19- Bishop DV, Jacobs PA, Lachlan K, et al. Autism, language and communication in children with sex chromosome trisomies. Arch Dis Child. 2011;96(10):954–9.
20- Ross JL, Roeltgen DP, Kushner H, et al. Behavioral and social phenotypes in boys with 47,XYY syndrome or 47,XXY Klinefelter syndrome. Pediatrics. 2012;129(4):769–78.
21- Libbey JE, Sweeten TL, McMahon WM, etal. Autistic disorder and viral infections. J Neurovirol. 2005;11(1):1–10.
22- Stubbs E G. Autistic children exhibit undetectable hemagglutination-inhibition antibody titers despite previous rubella vaccination. J. Autism Child. Schizophr, (1976); 6, 269–274.
23- Libbey J E, Coon H H, Kirkman N J, et al. Are there altered antibody responses to measles, mumps, or rubella viruses in autism? J. Neurovirol. 2007. 13, 252–259.
24- Gentile I, Bravaccio C, Bonavolta R, et al. Response to measles-mumps-rubella vaccine in children with autism spectrum disorders. In Vivo, 2013; 27, 377–382.
25- Adams Waldorf K M, McAdams, R M. Influence of infection during pregnancy on fetal development. Reproduction,(2013); 146, R151–R162.
26- Gardener H, Spiegelman D, Buka S L. (Perinatal and neonatal risk factors for autism: a comprehensive meta-analysis. Pediatrics, 2011; 128, 344–355. 155-Gardener, H., Spiegelman, D., and Buka, S. LPerinatal and neonatal risk factors for autism: a comprehensive meta-analysis. Pediatrics ,2011; 128, 344–355.
27- Cordero E A, Veit T D, da Silva M A,et al. HLA-G polymorphism influences the susceptibility to HCV infection in sickle cell disease patients. Tissue Antigens. 2009;74(4):308–313.
28- Eskandari-Nasab E, Hashemi M, Hasani S S, et al. Association between HLA-G 3′UTR 14-bp ins/del polymorphism and susceptibility to breast cancer. Cancer Biomarkers. 2013;13(4):253–259.