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EFFECT OF CYTOKINE GENE POLYMORPHISM IN DEVELOPMENT OF FEBRILE SEIZURE

Genetics

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ABSTRACT

BACKGROUND : Among all childhood seizure patients febrile seizure accounts for more than 30% of the total. A lot of confounding factors are there in the causation of febrile seizure. We designed this study to investigate the mutation in cytokine gene to determine whether this polymorphism could be a marker of susceptibility to febrile seizure in children.

MATERIALS AND METHODS: 100 case and 100 age and sex matched healthy controls were selected. Fasting blood samples were collected. Data was analyzed statistically.

RESULTS: Genetic variations in cytokine gene was found among the febrile seizure patients. Discussion: Any mutation in the coding region of cytokines might effect their production and resulted in febrile seizure.

CONCLUSSION: Treating the patient a gene analysis may give prior information and can decrease the morbidity and future complications to the patient and the related family members.

KEYWORDS

Febrile Seizure, Cytokine gene , Polymorphism.

INTRODUCTION :

Among all childhood seizure patients febrile seizure accounts for more than 30% of the total.[1]It is being defined as a seizure in febrile children between the ages of 6 months to 60 months who don't have any intracranial infection, metabolic disturbances or history of seizures[2].7% of these seizure patients develop epilepsy by adolescence[3].The pathogenesis of febrile seizure is not well defined but there is a complex interaction of immune-inflammatory process, genetic factors and cytokine activation [4]. Cytokine is involved as an endogenous pyrogen[5]. It has a property to induce fever in experimental models and human[6]. Animal model experiments have proved the role of these cytokine in development of hyperthermia induced seizure[7]. So the similar effect can be there in childhood febrile seizure cases. Mutation in cytokine gene can increase the encoded protein of febrile seizure[8].Various studies had shown that mutation in cytokine gene can be a precipitating factor in occurrence of febrile seizures[9-14].

We designed this study to investigate the mutation in cytokine gene to determine whether these polymorphism could be a marker of susceptibility to febrile seizure in children.

MATERIALS AND METHODS:

It was a Descriptive, Cross sectional, hospital based study. 100 children were selected from patient admitted in pediatrics ward of Calcutta National Medical College & hospital Kolkata(CNMC),West Bengal,with complains of seizure with in the period between May 2018 to November 2018. The age of the patients ranged from 6 to 60 months. Diagnosis of febrile seizures followed the criteria established by The American Academy of Pediatrics[2]. The electroencephalogram (EEG) was normal for all patients with febrile seizures or showed mild non-specific abnormalities.

One hundred children with fever, of comparable age and sex; without a history of febrile or afebrile seizures or any neurologic disorders; were enrolled as a comparable group while attending the outpatient clinics/indoor.

All participants were recruited with written consents from their parents.

Exclusion criteria: Patients with febrile seizures beginning at the age of 6 years or later, evidence of intracranial infections, afebrile seizure, epileptiform EEG traits, or metabolic disturbance.

The whole process strictly followed the guidelines and regulations set by Helsinki Declaration of 1975, with all amendments and revisions. The study was approved by Institutional Ethics Committee.

Sample Collection and Handling

Fasting blood samples were collected from patients who matched the study criteria. Blood (5 mL) was withdrawn and distributed into anticoagulant free plain tube (2 mL) and EDTA tube (3 mL). The blood sample in the plain tube was centrifuged after 30 minutes of sampling and serum was isolated and stored at -20°C and sent to the laboratory for biochemical analysis. . EDTA tubes were stored properly at -20°C for DNA extraction followed by PCR- RFLP .

Molecular Analysis

DNA extraction

The venous blood, which was collected in the evacuated EDTA tubes, was used for DNA extraction.DNA from study subjects was isolated from peripheral blood (EDTA).

PCR-RFLP

PCR/RFLP : Reference sequence and details of SNPs, PCR primers' design and restriction enzymes were obtained from protocol.

STATISTICAL ANALYSIS

Data was analyzed using the SPSS 17 and graph-pad software. Continuous variables were expressed as mean (standard deviation) and the differences were accomplished by comparison via student's unpaired 2-sided t-test. The genotype distributions of SNPs were also analyzed A significant difference is considered at $p < 0.05$.

RESULTS:

The study included 200 children between 6 months to 6 years of age. Among them 100 were effected with seizure.100 age, sex and ethnicity matched control subjects were selected. 60 are male and 40 are female. Out of 100 febrile seizure patients 63 are male and 37 are female. Average age of onset of febrile seizure was 15 months (range 6- 60 months) and duration was 7 minutes (1-30 minutes). 45 patients are having family history of seizure and 8 were having family history of epilepsy. Genetic variation in cytokine gene was found among the febrile seizure patients.

Table 1: Demographic Profile of Case & Control

	Patient (n= 100)	Control (n=100)	p
Age/mon	29 (6- 60)	30 (6-60)	>0.05
Sex(M/F)	63/37	60/40	-
Age of onset at febrile seizure/month	15(6-35)	-	-
Seizure duration/minutes	7	-	-
Family history Febrile Seizure Epilepsy	45 8	- -	- -

Table 2: Genetic variation in Seizure patients & Control

	Case(n= 100)	Control(n=100)	Significance
Presence of Allele variation	89	09	P<0.0001

DISCUSSION:

Febrile seizure is the most common childhood seizure. Its etio-pathogenesis is not very clearly understood. Pro-inflammatory cytokines may play a vital role in potentiating host response to infection and induction of fever. So they may have a similar role in induction of febrile seizure[15]. Any mutation in the coding region of these cytokines may effect their production and that may results in febrile seizure[16].

In our study, we found increased mutation in febrile seizure cases than controls. Our findings are similar with Virta et al[17] & Ozen et al[18].By contrast many studies didn't find any role of gene polymorphism in febrile seizure[19-20].The difference in results between our study and the others may be due to study design, environmental variation, gene-environment or gene –gene interactions.

The family history also may play a vital role in development of the disease. A positive family history always increases the risk of febrile seizure[21]. But the sporadic cases may have a different pathophysiology. Along with the genetic predisposition ,repeated exposure to infections and environmental factors may also play a vital role in development of febrile seizure specially in the the developing countries like India.

CONCLUSSION:

A proper family history along with the ethinicity and environmental factors play a major role in development of febrile seizure.while treating the patient a gene analysis may give prior information and can decrease the morbidity and future complications to the patient and the related family members.Polymorphisms study can predict the febrile seizure in advance so that Physicians and parents can plan accordingly.

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