Evaluation of the Endothelial Cell Antibodies in Serum and Perilymphatic Fluid of Cochlear Implanted Children with Sensorineural Hearing Loss

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ABSTRACT

Introduction: Serum Anti endothelial Cell Antibodies (AECAs) play a prominent role in idiopathic Sensorineural Hearing Loss (SNHL) in that they induce vascular damage (immune mediated).The of the current study is To compare AECAs in serum and perilymphatic fluid of idiopathic SNHL children (<15y) undergoing cochlear implant surgery.

Methods: This was a cross sectional study performed in the cochlear implant ward in Rasoul Akram hospital, Tehran, Iran (2008 -2010) on 99 SNHL children undergoing cochlear implant surgery. The data collected from47 idiopathic and 52 non-idiopathic SNHL cases. AECAs were measured by indirect immuno fluorescence assay and compared in sera and perilymphatic fluids between the two groups. P-value<0.05 was considered significant.

Results: Idiopathic SNHL was diagnosed in 47.5% of cases. Positive AECA results in serum and perilymphatic fluid were 10% and 12%, respectively. Although AECA results in perilymphatic fluids were different between idiopathic and non-Idiopathic SNHL patients (PV<0.05), AECAs in serum showed no significant difference between the two (PV=0.1). No significant difference was detected between the mean age of idiopathic and non-idiopathic SNHL patients with positive AECAs in serum and perilymphatic fluids (PV=0.2; PV=0.2).

Discussion: Idiopathic SNHL was diagnosed in 47.5 % of studied cases. Idiopathic SNHL has a poor out come in children. In cases with idiopathic SNHL, finding AECAs in perilymphatic fluids are more valuable than in the serum. We suggest that serum and perilymphatic fluids testing for AECAs would be helpful in management of idiopathic SNHL cases.

Specific immunosuppressive treatments for selected cases suffering from Idiopathic SNHL (only in those older than 5) might be successful in disease management. However, this theory should first be validated by randomized clinical trials.

1. Introduction

The incidence of unilateral hearing loss in children is approximately 0.1 %. In 7.5% of cases unilateral deafness is diagnosed accidentally, usually between the age of 7 and 10 (Olusanya & Okolo,2006). Neither children nor their parents could precisely determine the time of its onset, especially when it is not accompanied by other symptoms, such as dizziness or tinnitus (Walch et al., 2009). The etiology of most of these cases remains unknown (Adams, 2002). Risk factors for hearing loss in neonates have been explained by some authors (Martínez-Cruz , Poblano & Fernández-Carroc-

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era,2008; Kountakis,2002). Idiopathic sudden sensorineural hearing loss and its prognostic factors have also been discussed in many references (Cadoni et al.,1996; Vasama & Linthicum,2000 ; Merchant , Adams & Nadol,2005). AECAs are of prognostic importance in these diseases and can be considered as a useful clinical tool to differentiate patients with idiopathic hearing loss(Vasama & Linthicum,2000; Merchant et al.,2005).

Autoimmune hearing loss is a plausible explanation for a certain percentage of the group categorized as the idiopathic type. SNHL in children can be caused by autoimmune disorders localized to the inner ear or secondary to systemic immune diseases (Merchant et al., 2005; Cadoni et al., 2003).

Cadoni et al. (2003) investigated the presence of AECAs and its role in causing striavascularis damage in immune-mediated sensorineural deafness.

Many studies established the non-specific auto antibodies vs. the inner ear, such as anti endothelial cell antibodies(Cvorović , Deric , Probst & Hegemann,2008; Xenellis & Karapatsas,2006; Solares , Hughes & Tuohy,2003; Naumann , Hempel & Schorn,2001; Ceylan et al.,2007; Agrup & Luxon,2006).

The appearance of antiendothelial cell antibody is related to poor outcome in hearing loss. AECAs detection could be helpful in the selection of particular patients with sensorineural hearing loss for specific immunosuppressive treatments(Plontke et al.,2005; Banerjee & Parnes,2005; Westerlaken , Stokroos , Dhooge , Wit & Albers,2003; Tucci , Farmer , Kitch & Witsell,2002; Fowler & Boppana,2006).

SNHL due to various etiologies is common in Iranian children (Verbeeck et al.,2008; Foulon, Naessens , Foulon , Casteels & Gordts,2008). Cochlear implant surgery is needed for some cases (Noorbakhsh et al., 2008; Noorbakhsh , Memari , Farhadi & Tabatabaei,2008 ; Noorbakhsh et al., 2006; Noorbakhsh, Farhadi & Tabatabae,2008; Noorbakhsh, Farhadi & Tabatabaei,2005; Noorbakhsh, Siadati & Farhadi,2006).

Serum AECA might play some role in idiopathic SNHL in that they induce vascular damage (immune mediated).

Aim of study: To compare AECA in serum and perilymphatic fluid of idiopathic SNHL children (<15y) undergoing cochlear implant surgery. The outcome suggests possible clinical relevance for assessment of AECA in serum and perilymphatic fluid of children with suspected ISNHL and clinical significance.

2. Methods

This was a cross sectional study performed in the cochlear implant ward in Rasoul Akram hospital, Tehran, Iran (2008 -2010).This study was approved by the Ethical Committee in the ENT and head &Neck surgery Research Center affiliated by Tehran University of Medical Sciences. The parents (or patients) signed the consent letter.

Initially, a questionnaire was completed by an authorized physician for each case. Audio logic screenings (Auditory Brainstem Response, Evoked Otto-acoustic Emissions and Pure Tone Audiometry) appropriate for patients` age were performed in all cases. 99 children undergoing cochlear implant surgery entered the trial. All cases were candidates for cochlear implant surgery due to severe SNHL (>95db).They were between 2.5-12 years old with a mean age of 5.22.6±1.7years old. 61% of the patients were male and 39% were female. 47 idiopathic and 52 non-idiopathic SNHL cases were diagnosed by specialists based on AAO (American academy of Otolaryngology) criteria for distinguishing the type of SNHL (idiopathic and non- idiopathic). Blood samples (2 ml) were taken, then centrifuged and transferred to our research laboratory. Perilymphatic fluids were taken by ENT specialist during surgery in operation room. All samples were kept frozen at -80°C until usage. We looked for AECAs (IgG) in sera and perilymphatic fluids by indirect fluorescent antibody test (KMI diagnostics, USA). The results were calculated qualitatively as suggested by the AECAs manufacturer. AECAs were measured and compared in sera and perilymphatic fluids between the two groups.

In order to minimize the false-positive interferences with AECAs, titers of rheumatoid factors (RFs) and antinuclear antibodies (ANAs) were measured in serum samples. All patients with positive RFs and ANAs (5 Idiopathic cases and 3 non-idiopathic) were excluded.

Statistical analysis: Student t-test was used to determine differences between the means of all continuous variables. Chi-square values were calculated for all categorical variables. P value less than 0.05 was con-

sidered significant. All analysis was conducted using SPSS version 11.5.

3. Results

Idiopathic type of SNHL was diagnosed in 47.5% (n= 47) of cases, and non-idiopathic type in 52.5% (n=52). Known causes of SNHL include familial 16%, infectious causes 14%, convulsion 13.3%, mental retardation 4.5%,Trauma 1.5%, prematurity1.5%, hypoxic ischemic 6.5% and fetal radiation 3%.

There was no meaningful difference between the age of patients and idiopathic and non-idiopathic types of SNHL (Mean age 5.6±1.4 vs. 5±1.9 years; P-value =0.2).

Serologic results: Positive AECAs were detected in 10% of serum samples and 12% of perilymphatic fluids in SNHL cases.

AECAs detection in perilymphatic fluids showed different results between idiopathic and non-idiopathic types of SNHL (P-value=0.04) (Table1, Fig.1).

However, positive AECAs in serum was not significantly different between the two types of SNHL (P-value=0.1) (Table2, Fig.2).

The mean age of cases with positive AECAs in serum and perilymphatic fluid had no significant difference between idiopathic and non-idiopathic type of SNHL (P-value=0.2, P-value=0.2).

Table 1. Comparison between positive perilymphatic AECAs in the two types of SNHL

Total	Idiopathic		Perilymphatic AECA
	Negative	Positive	
12	3	9	Positive
87	49	38	Negative
99	52	47	Total

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Table 2. Comparison between serum AECAs results in the two types of SNHL

Total	Negative	Positive	
10	3	7	AEC
89	49	40	A
99	52	47	Total

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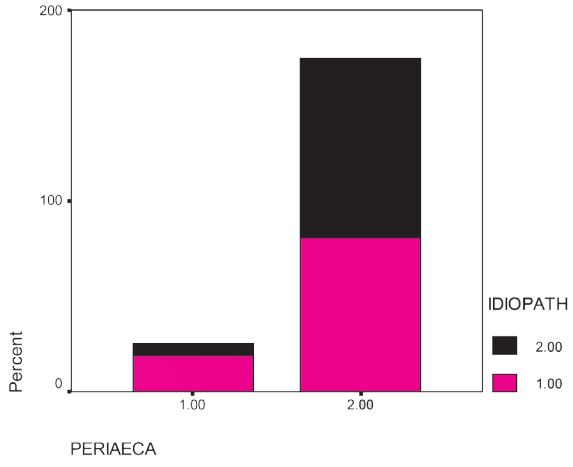

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Figure 1. Positive perilymphatic AECA in the two types of SNHL

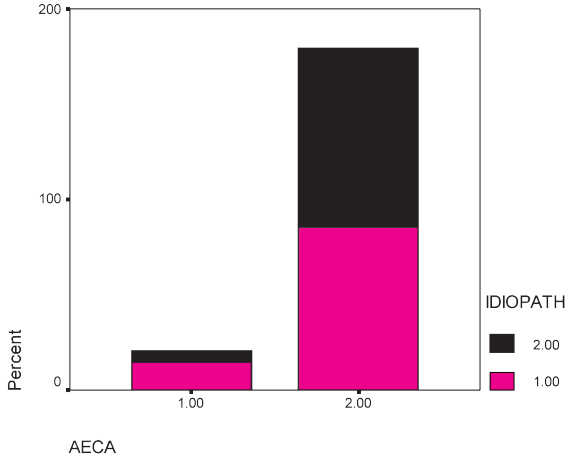

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Figure 2. Positive serum AECA in the two types of SNHL

4. Discussion

In this study, Idiopathic SNHL was diagnosed in 47.5% of children undergoing cochlear implant surgery. At least one etiologic factor was recognized for profound SNHL in 52.5% (n= 52) of cases (age: 2.5-12 years old).

Familial SNHL (16%), infectious causes (14%) and convulsive disorders (13.3%) were the 3 most common causes. Incidence of idiopathic type of SNHL in our study was very close to that reported by other studies (38.7%)(Olusanya & Okolo,2006; Walch et al.2009; Adams,2002; Martínez-Cruz et al., 2008).

Idiopathic hearing loss basically means hearing loss without any perceivable reason. A more likely scenario would be that the person's hearing loss actually takes place over a few hours(Cadoni et al.,2003; Cvorović et al.,2008; Xenellis et al.,2006).

Positive AECAs were observed in serum of 10% (10/99) of cases between 3.5-5.5 years old, without any meaningful differences between idiopathic and non-idiopathic cases (P-value=0.1).

This number is much lower than the 54% reported by Cadonni et al.(2003) in adult cases suffering from SNHL.6The results of a previous study in our center determined that there is no difference between cases with SNHL and normal controls in regard to positive serum AECAs (14.5% vs. 21%, P-value=0.36), but cases with positive serum AECAs were older than those with negative results (mean=50 vs. 32 months, P-value=0.047). But in this study, no such difference was observed (P-value=0.2).

Cadoni et al. (2003) investigated the presence of AECAs and their role in causing damage to the striavascularis in immune-mediated sensorineural deafness. Cvorovićet al. (2008) reported a prognostic model for predicting hearing recovery in patients with idiopathic sudden sensorineural hearing loss. Xenelliset al.(2006) described prognostic factors for idiopathic sudden sensorineural hearing loss .The appearance of endothelial cell antibody is related to the poor outcome of hearing loss (Solares , Hughes & Tuohy ,2003; Naumann , Hempel & Schorn,2001; Ceylan et al.,2007; Agrup & Luxon,2006; Chen , Emmerling , Ilgner & Westhofen,2005).

Positive AECAs in older idiopathic SNHL cases (> 5years old) could define the clinical associations of AECAs with immune-mediated inner-ear disorders. Probably, AECAs play a prominent role in causing damage to the striavascularis after infancy in immune-mediated SNHL. Production of serum AECAs would act as a marker of disease activity. The association between AECAs and endothelial injury in the course of these diseases prompted us to develop assays for said antibodies in clinical practice.

Positive AECAs in perilymphatic fluid was reported in 12% of cases (3.5 -5.7 years old) and more frequently in idiopathic type of SNHL (P-value=0.04). No significant difference was observed between positive and negative results in regard to the age of patients (P-value= 0.3).

Cvorovićet al.(2008) reported that the appearance of AECAs is related to poor outcome and recovery of the adults. Prognostic factors for Idiopathic SNHL in adults have been reported by many authors(Cvorović et al,.2008; ; Xenellis et al.,2006; Solares et al.,2003; Naumann et al.,2001; Ceylan et al.,2007). Multiple potential mechanisms can result in immune-mediated inner ear disease in children. All previous studies, but for one, were carried out in adults (Herr & Marzo, 2005).

Many authors recommendsystemic or intra tympanic steroids as a treatment for immune-mediated SNHL in adults (Agrup & Luxon,2006; Chen , Emmerling , Ilgner & Westhofen n,2005; Herr & Marzo,2005 ; Gouveris , Selivanova & Mann,2005; Plontke et al.,2005; Banerjee & ParnesL, 2005).

Westerlaken et al. (2003) and Tucci et al. (2002) even treated the Idiopathic SNHL cases with a combination of steroids and antiviral drugs.

Not enough studies have been performed previously on the correlation between infections and AECAs in children. These studies were mostly done in adults rather than children, especially the Idiopathic SNHL cases.

The most important limitation of the study is the small study sample especially in younger patients (<2 years). To determine the clinical outcome and possible clinical relevance of AECA assessment in serum and perilymphatic fluid of children with suspected Idiopathic SNHL, follow up studies are recommended.

Conclusion: Idiopathic SNHL was diagnosed in 47.5 % of studied cases. Idiopathic SNHL has a poor outcome in children. In cases with idiopathic SNHL, finding AECAs in perilymphatic fluids are more valuable

than in the serum. We suggest that serum and perilymphatic fluids testing for AECAs would be helpful in management of idiopathic SNHL cases.

Specific immunosuppressive treatments for selected cases suffering from idiopathic SNHL (only in those older than 5) might be successful in disease management; however this theory should first be validated by randomized clinical trials.

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Ethical Considerations

Ethical Committee in the ENT and head &Neck Research Center inTehran University of Medical Scienceshas reviewed and approved the Waiver of Authorization for use of protected health information (PHI) for research purposes for the following study.

Principal Investigator: DrSamilehNoorbakhsh MD; Professor in Pediatric Infectious Diseases; Research Center of Pediatric Infectious Diseases, Tehran University of Medical Sciences.

Title: Searching the Antiendothelial cell antibody (AECA) in perilymphatic fluid and serum of cochlear implanted children"

Date of Approval: May 2007

The following PHI for which use or access is requested has been determined to be necessary for the conduct of the study by the ENT &CPID Research centers.

[Insert the patient information to be used or disclosed, or attach documentation of the information.]

In approving this Waiver of Authorization, the ENT &CPID Research centers have made the following determinations:

1. The use or disclosure of PHI involves no more than minimal risk.

• Granting of waiver will not adversely affect privacy rights and welfare of the individuals whose records will be used.

• The project could not practicably be conducted without a waiver.

• The project could not practicably be conducted without use of PHI.

• The privacy risks are reasonable relative to the anticipated benefits of research.

• An adequate plan to protect identifiers from improper use and disclosure is included in the research proposal.

• An adequate plan to destroy the identifiers at the earliest opportunity, or justification for retaining identifiers, is included in the research proposal.

• the project plan includes written assurances that PHI will not be re-used or disclosed for other purposes.

• whenever appropriate, the subjects will be provided with additional pertinent information after participation.

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