# Epidemiological study of the patients referred for thalassemia diagnosis using chorionic villous sampling (CVS) in Genetic Laboratory of Dastgheib Hospital, Shiraz, 2011

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## Abstract

**Objective:** Pre-natal diagnosis is the most effected way to prevent genetic diseases in a society. The aim of this research was to show the prevention level of the birth of the children with major thalassemia disorder and the demographic condition of the people referring to the Shahid Dastgheib Genetic Center in Shiraz for the pre natal diagnosis.

**Materials and methods:** The present research was a cross- sectional (descriptive, analytical) one. In this study, the amount of sampling was done by census in a way that all the case (372 cases) related to the year 2010. The questionnaire was prepared based on the information present in the files. In order to compare the quantitative and qualitative variables, two sample t - test and K sample t- test were used.

**Results:** Out of 372 fetuses tested, 25.5% had major thalassemia, 48.7% minor thalassemia, 0.8% intermediate, 1.3% sickle cell, and 23.7% were healthy. All the cases diagnosed with thalassemia were introduced for abortion, and abortion was carried out. Major thalassemia was more prevalent in Lore tribes (32.9%), which was more in comparison to the members of others tribes.

**Conclusion:** In order to prevent major thalassemia, it is important to identify the gene carriers and prevent their marriage. Nevertheless, in many places in the country, especially in the villages and rural areas, the couples do the experiment after they have already gotten emotionally involved and made the arrangements to get married; therefore they're unwilling to stop the marriage. As a result, post-nuptial CVS during pregnancy is crucial.

**Keywords:** Thahlassemia, Genetic, Chorionic Villous Sampling, Laboratory

## Introduction

Pre-natal diagnosis is the one of the best choice to prevent genetic diseases in a society. With on the time prenatal diagnosis, one can prevent the birth of the children with different inherited, congenital, and genetic diseases. In general, post-natal genetic diseases can not be cured. Many genetic syndromes and diseases which appear at birth, such as Down syndrome, and the ones which can be detected after

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birth, such as thalassemia and hemophilia, are easily diagnosed before birth. Depending on their intensity, these diseases can be fatal during the first few months after birth or continue to exist for many years. In the latter case, not only does the person afflicted suffer for a long time but also burdens the society or the family with a huge amount of expense (1).

Pre- natal diagnosis involves access to the fetus or a fetal sample which can be done with different methods: invasive or non-invasive. CVS or chorionic villus sampling is an invasive method (2). Abdominal CVS was first done by the biologists at Biocell Center in Italy (3).

The most important advantage of fetal tissues is the early diagnosis on the fetus. The mentioned tissue can be obtained from the fetus using a special catheter through Sonography in the $10^{th}$ to $12^{th}$ weeks of pregnancy. The level of accuracy and certainty of both CVS and Amniocentesis methods is almost the same.

In addition to the molecular experiments, the tissue can be used for chromosome analysis. There is also the possibility of villi culture for further diagnosis. Studies show that sampling in the ninth week of pregnancy or earlier show facial and limb disorders or disabilities which should be taken into consideration during the sampling (4,5,6).

One of the problems encountered during CVS is the possibility of the mother's tissue contamination with the fatal cells which can lead to errors in the result of the experiment (7). Another problem that can occur in CVS and amniocentesis is RH sensitivity which happens in cases where the mother's RH is negative, while the fetus's RH is positive. This problem can be solved by using immunoglobulin during the sampling (8).

CVS is normally done 10 to 12 weeks after the last menstrual period. This is earlier than the time amniocentesis is done (9). Whenever it is necessary to be informed of the fetus's condition sooner than 14 weeks, CVS method is preferably used, which makes it possible to a timely decision about the fetus (10).

These days early diagnosis of some of the fetal genetic diseases, such as thalassemia, which is done by CVS, trans-abdominal methods is accepted all over the world because low risk of miscarriages which helps improve the public the health and on the other hand removes the parents' worries in case the results show that the fetus is healthy (11).

Major thalassemia is one of the most prevalent genetic diseases in Iran; therefore, it stays in priority in congenital disease prevention plans (12). Iran is considered as one of the countries on thalassemia belt in the world. This disease is mostly prevalent in the provinces along side the Caspian Sea, Persian golf, and Oman Sea, such as Mazandaran, Gilan, Khoozestan, Fars, Bushehr, Hormozgan, Sistan Baloochestan, and Kerman (13). Moreover Beta thalassemia is the most prevalent genetic disease in Iran and Fars province is the most affected (14).

Although major thalassemia gene prevalence is 4 to 8 percent in Iran, it sometimes reaches 10% in Sistan-Baloochestan and Fars provinces (15). Thalassemia and Hemophilia Genetic and PND center in south of Iran is located in Shahid dastgheib Shiraz affiliated to Hematology research center of Shiraz University of Medical Sciences has started genetic counseling and major thalassemia diagnosis using CVS through trans-abdominal method since 2001, and did genetic experiments on 2200 couples up to the end of 2010. Up to now, no studies have been conducted on the people who referred to this laboratory. The purpose of this research was to show the prevention level of the birth of the children with major thalassemia disorder and the demographic condition of the people referring to the clinic for the PND. The findings of this research can be used in educational planning for controlling as well as preventing this disease. In addition, the researcher plans to carry out another research on the subjects who participated in this study in order to check the specificity as well as sensitivity of the CVS experiments.

## Materials and methods

The present research was a cross- sectional (descriptive, analytical) one. The participants of this study were the individuals who had referred to our genetic center to test the fetus for major thalassemia genetic condition. From 2001 up to the end of 2010, 2200 CVS tests were done in this center. In this study, the amount of sampling was done by senses in a way that all the case related to the year 2010, 372 cases was studied. The questionnaire was prepared based on the information present in the files and genetic experts and lab Technicians supervision, and its validity was confirmed. The questions of the questionnaire were related to the demographic as well as the epidemiological factors of the participants, such as the mother's age, gravity, age of fetal, place of resistance, parents' nationality, and major thalassemia record in the family. The data were analyzed through the SPSS statistical software (version 17). In addition, Descriptive results were

presented in graphs and charts in the form of numbers and percents. Moreover, in order to compare the quantitative variables, two sample t- test were run. Also K sample t- test were used for comparing the qualitative variables.

## Results

Out of 372 fetuses tested at Shahid Dastgheib Hospital genetic center, 25.5% had major thalassemia, 48.7% minor thalassemia, 0.8% intermediate, 1.3% homozygote sickle cell, and 23.7% were healthy. The above figures are close to the law of probability related to the percentage of major thalassemia birth rate and minor thalassemia birth rate in each pregnancy in couples with minor thalassemia (Table 1).

**Table1:** CVS findings on tested fetuses in Shahid Dastgheyb Hospital Genetic Center, Shiraz, 2010

| | result | Number | percent |
|---|---|---|---|
| 1 | Healthy | 88 | 23.7 |
| 2 | Minor thalassemia | 101 | 48.7 |
| 3 | Major thalassemia | 95 | 25.5 |
| 4 | Intermediate | 3 | .8 |
| 5 | Homozygote sickle cell | 5 | 1.3 |
| 6 | total | 372 | 100 |

Out of the 372 samplings, one miscarriage was observed.The average age of the mothers referring to the center was 26 years and the average of the gestational age was 11 weeks. Besides, the gestational age at the time of test ranged from 7 to 14 weeks. All the cases diagnosed with thalassemia were introduced for abortion, and abortion was carried out.

In the present study 42.7% of mothers in the first pregnancy, 35% in the second, 14% in the third and 7% in their fourth pregnancy had referred to the center. In addition, 60% of the mothers lived in urban area while 40% lived in rural areas. Moreover, 43.4% of the mothers who referring to the center had high school education and 14.6% had university education. Also 27.7% of the mothers had babies with major thalassemia in their previous pregnancies. Blood relationship of the couples was not significant in major thalassemia, while it was significant regarding the couples having minor thalassemia. Furthermore, Major thalassemia was more prevalent in Lore tribes (32.9%), which was more in comparison to the members of others tribes. Moreover, the Statistical difference between the Lore and the members of other tribes was significant (Table2) (p=.02).

**Table2:** The incidence of thalassemia in different ethnic groups attending in Shahid Dastgheyb Hospital Genetic Center, Shiraz, 2010

| ethnic | total | MajorThalassemia | percent |
|---|---|---|---|
| Fars | 258 | 61 | 23.7 |
| Turk | 20 | 5 | 25 |
| Lore | 82 | 27 | 32.5 |
| Arab | 11 | 2 | 18.2 |
| Balooch | 1 | 0 | 0 |

Out of the 372 chorionic villus samplings, one miscarriage was seen (0.2%). The correlation between the mother's blood type and thalassemia major existence in the foetus was statistically significant (p=0.013). Besides, major Thalassemia was more prevalent among the foetuses whose mother's blood type was $O^+$ or $A^+$. Existence of major thalassemia in foetuses whose mothers, blood types were O, A, B, and AB 28.6%, 28.3%, 20%, and 10% respectively. All the mothers with negative blood RH received Rhogam injection.

## Discussion

In a study carried out in Tehran, the side effect of using this method was not significant (16). In addition Another study showed that 97% of the couples who used CVS wanted to make sure there was no danger of major thalassemia and that abdominal CVS was the best method to early diagnose foetal disorders which was low risk of dangerous for the mother so for the foetus (17).

Using the CVS method in the first trimester of pregnancy gives accurate results. However, in amniocentesis of 12-14 weeks of pregnancy, there are more false negative results and after 15-18 weeks of pregnancy real positive results increases (18). Knowledge of the mutations helps a lot in diagnosing the prenatal major thalassemia and, as a result, leads to saving time and experiment expenses (19).

In another study performed in Sari, it was found that 20.6% of the major thalassemia patients' parents were blood relatives (20). However in this study this proportion was 50%.

In many places in the country, especially in the villages and rural areas, the couples do the experiment after they have already gotten emotionally involved and made the arrangements to get married; therefore they're unwilling to stop the

marriage (21). As a result, post-nuptial CVS during pregnancy is crucial.

Considering the high annual expense of taking care of a patient with thalassemia (about 100,000,000 Rails per year), low cost of pre-nuptial test (about 200,000 Rials) as well as CVS (1,500,000 Rials), and low side effects of these tests, they are recommended for minor thalassemia couples. In which addition to the expenses, the patients with thalassemia suffer a lot both mentally and emotionally. A Study was carried out in Shiraz in 1998 showed that the patients with major thalassemia had a worse mental and physical condition in comparison to healthy individuals. Moreover, this difference was more significant in the patients who suffered more (22). Our aim is improving quality of life for alive patients with best care of them, and prevent of new born affected cases.

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