ESTIMATION OF HERITABILITY OF FAMILIAL HYPERCHOLESTEROLEMIA AMONG 335 FAMILY MEMBERS OF FIVE HYPERCHOLESTEROLEMIC PROBANDS OF PAKISTANI POPULATION

Fauzia Imtiaz

Department of Biochemistry, Dow International Medical College, Dow University of Health Sciences, Karachi.

Background: Familial hypercholesterolemia is an autosomal dominant disorder, caused by mutation in Low-density lipoprotein receptor (LDL-R) gene. **Methods:** Cross-sectional study conducted to recruit the population of Karachi-Pakistan, screened for familial hypercholesterolemia. A total of 1523 hyper-cholesterolemic individuals have taken part in the study, five were found to be familial hyper-cholesterolemia. Their lipids profile was estimated and a family pedigree was drawn. **Results:** Parent-offspring correlation, coefficient of linear regression, and heritability is calculated by using SPSS 12.0. A significant positive correlation of cholesterol was found among parents and their offspring (r=0.511, p=0.01, n=76). Coefficient of linear regression analysis also showed that parents-offspring relationship was highly significant at p<0.01 with b=0.438. Relationship between Father-Son, Father-Daughter, Mother-Son and Mother-Daughter were highly significant with b=0.794, 0.41, 0.766 and 0.56 respectively. **Conclusion:** The heritability among the parents and their offspring showed that genetic factors are major determinant of the familial resemblance in serum cholesterol among the Pakistani population living in the metropolitan area of Karachi.

Keywords: Familial hypercholesterolemia, heritability, pedigree analysis

INTRODUCTION

Familial hypercholesterolemia (FH) is an autosomal dominant disorder, of cholesterol metabolism related to a qualitative and/or quantitative defect of the LDL receptor gene. Elevated plasma concentration of the total cholesterol (TC) and Low density lipoprotein cholesterol (LDL-C) are major risk factors for coronary heart disease (CHD). Accordingly, much effort has been focused on the factors that determine plasma total cholesterol and Low-density lipoprotein cholesterol concentrations. Likewise, Triglyceride (TG) and high density lipoprotein cholesterol (HDL-c) are also important components. Shared gene and environmental factors were reported to influence HDL-c and TG levels simultaneously.[1] In addition, pleiotrophic effects on low HDL-c and high TG were evident among families.[2,3] Many factors were associated with high TC level such as age, gender, lifestyle activities and obesity. Study illustrates that the relationship of the FH genotype to the FH phenotype is not straightforward.[4] Genetic factors considered as a tool in determination of serum levels.[5] Data obtained from different families have indicated that genetic factors account for about 50% of the inter-individual variation in serum total cholesterol concentration.[6] Coronary heart disease particularly at a young age, largely influence by genetic variance. The influence of the genetic variance on serum lipids is of great interest. Numerous studies have demonstrated that the increased cholesterol levels predict coronary heart disease, stroke and are associated with features of the metabolic syndrome.[7] Lowering LDL-cholesterol concentrations results in a large decrease in cardiovascular morbidity and mortality, especially in patients at highest risk.[8] To our knowledge, estimation of heritability of FH with the risk of inheritance to their offspring has not been done. In this study, we therefore, estimate the magnitude of genetic influence on cholesterol level. The heritability index of serum cholesterol levels estimated by linear regression, the average of the offspring-serum cholesterol value on the mid-parent value, using weighted least-squares method.[9]

MATERIALS AND METHODS

This is a cross-sectional study, started in 1999, and designed to recruit the population living in metropolitan area of Karachi-Pakistan screened for familial hypercholesterolemia. One thousand, five hundred and twenty-three (1523), hyper-cholesterolemic individuals have taken part in the study, out of which, five individuals found to be familial hypercholesterolemia. Further investigations performed on these five individuals and their family members for determination of the true cases of FH. Confounders like hypothyroidism, diabetes mellitus, kidney disease, liver diseases were excluded by taking an extensive history. Selected families include 335 members in which 76 pairs of parents with their offspring were present. The selection criterion of having familial hypercholesterolemia is TC>300 mg/dl, LDL-c>200 mg/dl, and family history of premature heart disease (CHD). Furthermore, they examine for anthropometric measures, blood pressure and lipid profiles. All relatives of probands (1st, 2nd, and 3rd degree) were included in the study.

Blood samples were collected at the overnight fasting (12–14 hrs). Serum total cholesterol levels measured using the CHOD-PAP method (Boehringer

Mannheim, Germany). Lipid profile having Cholesterol, Triglyceride, LDL, and LDL-c measured following precipitation of apolipoprotein B-containing lipoproteins with phosphotungestic acid and magnesium ions (Boehringer Mannheim, Germany). Triglyceride concentrations measured by the GPO-DAOS method (Wako Co., Japan). All the lipid measurements were CDC standardized and performed on Hitachi 901, automated analyzer (Hitachi, Japan). LDL concentrations calculated using the Friedewald formula.[10] The serum collected by centrifugation, immediately stored at 20 ºC before it as transported in dry ice to the clinical laboratory for lipid measurements. The samples were then stored at 70 ºC until analysis.

The collected data were analyzed for correlation, linear regression and heritability was calculated by using SPSS-12.0, and pedigree was made by using the computer based software Cyrillic version 2.10 (Oxford, UK).

RESULTS

Five hypercholesterolemic proband cholesterol level >300 mg/dl (normal range 140–240 mg/dl), were selected with 335 family members of different ethnic groups of Pakistani population living in the metropolitan area of Karachi. The study comprises of 76 pairs of parents and their offspring's. Heritability of the cholesterol was estimate by fitting the regression modal. Heritability estimation reveals regression of mean offspring on mid parent TC, indicated that genetic factors accounted for 43.8% of the variance in cholesterol concentration b=0.438 at p<0.01 (Figure-1).

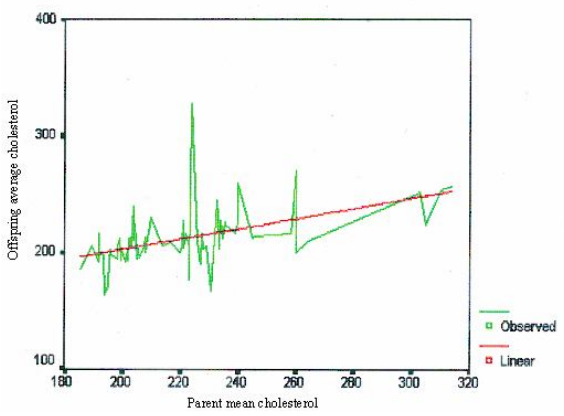


Figure-1: Linear Regression Model of cholesterol concentration between parent and their offspring

The Pearson correlation coefficient modal applied to determine the significance level of offspring depends on their parent's cholesterol levels. Results were found to be positively correlated with r=0.511 at p<0.01 (Table-1).

Table-1: Pearson correlation between parent and offspring mean cholesterol

	Parent mean cholesterol	Offspring mean cholesterol
Parent mean Cholesterol	1.000	0.511**
Offspring average Cholesterol		1.000

**p<0.01

Pearson correlation analysis applies in order to evaluate the differences in son and daughter, inherited from their parent. A positive significant differences found in the son and daughter cholesterol levels at p<0.01 (Table-2). The heritability in different combinations showed in Table-2. According to our study, the son inherited more cholesterol concentration from their parent as compared to daughter. Mother-son, father-son are calculated as 0.766 and 0.766, while mother-daughter and father-daughter correlation coefficients is found to be 0.56 and 0.41 respectively at p<0.01 (Table-2). The study enables us to conclude that the risk of inheritance of high cholesterol from their parent was considerably higher in son as compare to daughter. The familial aggregation can also be seen in the pedigree of probands (Pedigree-1).

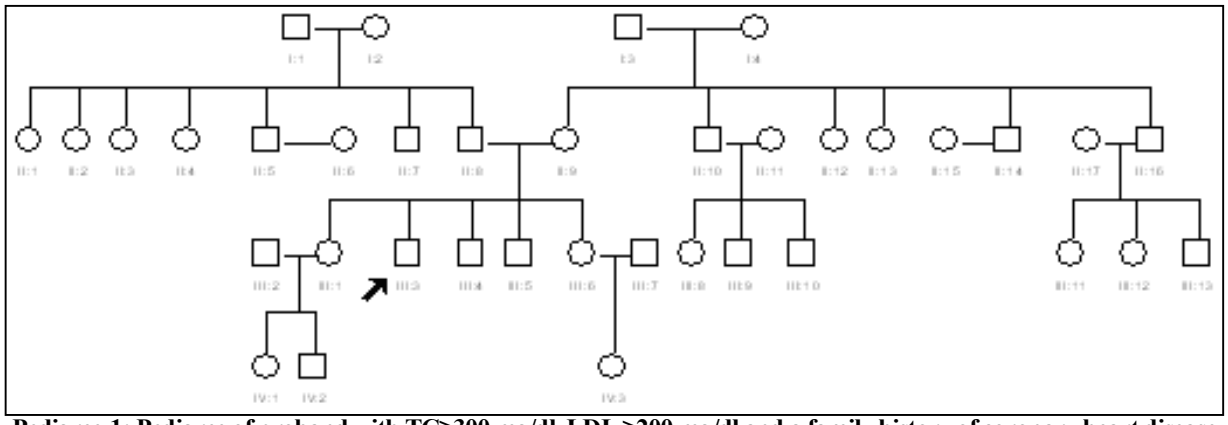
Table-2: Heritability among different relations

Relation	Value of linear regression (b)	t-Value	Heritability (h)
Parent–offspring	0.438	5.117**	0.438
Father–offspring	0.312	4.489**	0.624
Mother–offspring	0.295	3.426**	0.590
Mother–Son	0.383	3.239**	0.766
Father–Son	0.397	4.670**	0.766
Mother–Daughter	0.280	5.036**	0.560
Father–Daughter	0.207	3.347**	0.410

**p<0.01

DISCUSSION

Present study is the first in Pakistani population to proved heritability estimates of familial hypercholesterolemia using families randomly ascertained concerning their lipid and lipoprotein profile. The results suggest that these phenotypes strongly aggregate in families and characterized by significant maximal heritability estimates of 43.8%. In addition, the lack of significance spouse correlation, combined with significant parent-offspring and sibling correlations, suggests that genetic factors are likely the major determinants of the familial aggregation. Life expectancy of patients with familial hypercholesterolemia is decreased. Some untreated patients reach a normal life span and, therefore, additional risk factors and the type of mutation in the low-density lipoprotein (LDL) receptor gene are likely to influence the clinical outcome.[11]



Pedigree-1: Pedigree of proband with TC≥300 mg/dl, LDL ≥200 mg/dl and a family history of coronary heart disease

Genetic analysis frequently focuses on correlation or covariance among the relatives and attempts to partition these observed correlations or co-variances in linear regression (*b*) components attributable to shared genes and shared environments, for these components heritability can be calculated.[12] In 1986, scientist use univariate and bivariate analysis to see the familial aggregations of cholesterol in 95 pedigrees. They observed that in univariate as well as bivariate analysis familial aggregation of serum cholesterol was strongly influenced by both shared gene and shared environmental factors.[13] Father-offspring and mother-offspring correlation were also found to be significant at $p<0.01$.[14]

In comparison with the general population, the study found a similar 2 to 3-fold higher coronary mortality both in patients with treated definite FH diagnosed on the basis of elevated cholesterol concentrations and the presence of TX, and in patients with a presumptive diagnosis of FH based on elevated cholesterol concentrations and a dominant pattern of transmission of premature CHD.[15]

Analysis on 139 White families and demonstrated that the familial aggregation of serum cholesterol was found to be highly correlated with the average parental value r=0.43 at $p<0.01$.[16] Familial aggregation for serum cholesterol, correlation and multiple regression analysis on the 242 family members of Columbian population, also the significant results.[17] The population of North America and Israel showed the substantial correlation between the parent and their offspring.[18] The pooled mother and child correlation was also significantly higher than the father and child values in the North American population[18]. The general population of 1431 individuals' inhabitants over 10 years of age, estimated the correlation coefficient of the serum TC between parent and child and found to be as 0.26004, while the heritability of the cholesterol was found to be 0.5995.[19] A heritage family study also showed the influence of genetic factors in the adaptation to exercise training and its relationship with cardiovascular disease risk factor and studied the familial aggregation of lipids and lipoproteins among 86 Caucasian families. The results showed that the pattern of familial correlations was significant between parent and their offspring.[20]

In this study, the heritability (h) of serum cholesterol in our population calculated as 0.438 (43.8%). Estimation of heritability on a Finnish population reveals significant positive familial correlation of cholesterol was found for the pairs of mother-offspring (r=0.35), father-offspring (r=0.29), mother-daughters (r=0.46), mother-sons (r=0.27), and father-daughters (r=0.29). The consistent cholesterol associations between mother and offspring indicated that the key role of the mother for the primary prevention of hypercholesterolemia.[21] In our study, when different relations were considered both the parents showed more significant role in transferring the trait to their son as compared to the daughter at $p<0.01$ (Table-2). The family resemblance for lipids and lipoprotein, according to them; probands were selected form the Princeton School district, included 160 White, and 59 Black families and the estimated familial correlation by the method of maximum likelihood, Father and child correlation was of larger magnitude in Whites as compare to Blacks for each lipid and lipoproteins and estimation of genetic heritability was larger in Whites than Blacks families. Likewise the correlation (r) in our study was observed as 0.511 at $p<0.01$ (Table-1), which showed the positive correlation among parents and their offspring.[22] The lipid profile in 115 Blacks and 99 Whites who participated in the heritage family study, and the heritability ranges from 25% to 38%.[23]

CONCLUSION

Heritability is the proportion of variance due to additive familial effects, including both genetic and non-genetic sources of variance. Although, the pattern of familial correlations in the Pakistani population, suggested that the familial resemblance equally contributed from the genes as well as

environment for the disease progression of the hypercholesterolemia.

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Address for Correspondence:

Dr. Fauzia Imtiaz, B**-237, Block-N, North Nazimabad, Karachi-75850, Pakistan. Tel: +92-21-36677782**

Email: fauziaku@yahoo.com