



Original Research

## Association of life style and dietary habits with blood choline and cardiovascular outcome

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**Abstract:** Whole-blood choline, plasma choline and serum choline are emerging biomarkers in cardiovascular diseases (CVD). To examine the association of Whole-blood choline is an early predictor for cardiac events. In case control study, we enrolled 240 individuals including 120 normal (39 females and 82 males) and 120 cases (49 females and 71 males) where age limit was >40 years) Information through interviews, family disease history, 24 recall diet assessment and blood sampling. Odds ratios express the associated risks with CVD and without CVD patients. In healthy populations, good dietary habits and active lifestyle were observed. The number of participants with CVD were smokers than normal. In men, and women the risk was observed highly significant. ( $p=0.0049$ ) Different blood parameters like Triglycerides, Uric Acid, Urea, Creatinine, CRP and ESR were non-significant observed. In females the low carbohydrates and high protein and frequent salad vegetable consumption observed. On the other hand, men consume more carbohydrates. Body mass index was significantly with  $p=0.036$  (OD 1.12 95% 1.00-1.26). The total fats ( $p=0.017$ ) (OD 1.3301 95% 1.05-1.69) total carbohydrate ( $p=0.076$ ) (OD 1.1536 95% 0.98-1.35) and total proteins ( $p=0.287$ ) (OD 1.1456 95% 0.89-1.47) effecting respectively. The Blood choline level was significant observed between cases ( $p=0.026$ ) OD (0.944 95% 0.89- 0.99).

**Key words:** Blood choline; Cardiovascular diseases; Dietary habits.

### Introduction

Cardiovascular diseases (CVD) are the major cause of worldwide morbidity and mortality (1). The World Health Organization (WHO) estimated that roughly 17 million people die every year from CVDs (2). Developing countries like Pakistan contributes a greater share to the CVD global burden. In Pakistan, CVD accounts for over 200,000 deaths per year (3).

The role of life style and dietary habits in the prevention and control of morbidity and premature mortality due to CVD diseases has been well established by the vast population-based epidemiological studies carried out during the last decade (4-6). Cardiovascular disease getting common in general population because of inactive life styles (7). Conversely, people who preferred physical working, walking, cycling, running, swimming habitually they have more capacity to resist vascular illness or chronic heart problem (8). Furthermore, the patients with cardiovascular disease intervention with the exercise improve their muscular tolerance,

got better quality of life, improved blood profile; lower the hemostasis and inflammation (9). Physical exercise effect the individual's daily life applies numerous positive effects on community health like reduce the burden obesity (lower the BMI) (10), and helpful in diabetic patients that improve their insulin resistance (11,12). On the other hands environmental factors got impotence like tobacco smoking, physical inactivity, low education status, low socio-economic status such all things modified the genetic susceptibility, reduce the immunity against pathological condition. Therefore, Manolio et al. (13) found significant association of genetic variants with cardiovascular disease that is modified by environmental conditions moreover genetic susceptibility important for the cardiovascular disease risk estimate and inhibition (14-16). The role of diet and nutrition is pivotal in the development and prevention of CVDs (17,18). Dietary habits can be modified therefore it become one of the main target for interventions that will impact cardiovascular risk factors (19).

Comparisons of different diet plan showed that a

diet include fresh fruit and vegetables has 73% less risk to CVDs (19,20). Fruits and vegetables are rich source of antioxidants vitamins which have a protective role against CVDs (20,21). A case control study showed that increased consumption of fruits and vegetables significantly lowers the plasma lipids and BMI (22). Different epidemiological studies associate consumption of saturated fats including Tran's fats and limited intake of polyunsaturated fatty acids disturb the blood lipid profile and promote the oxidation process; all these attributes lead to higher risk of coronary heart disease and mortality (23). On the other hands, foods have good quality of nutrients, they improve immune system of body bioactive ingredients reduce the risk of any disease. Healthy diets contain more bioactive nutrients and antioxidants also (24).

Choline is a quaternary amines found in a variety of foods and can also be endogenously synthesized by liver (25,26). It has several biological function and involved in many physiological processes such as including transport of lipids, methylation reactions, normal metabolism and neurotransmitter synthesis.

Recently, consumption of choline has been inversely linked with inflammatory markers related to atherosclerosis and serum total homocysteine (tHcy) (27,28). An adverse cardiovascular risk factor profile is associated with high plasma homocysteine. Association of choline consumption with risk of cardiovascular disease is still in debate and little is known about this relationship. Several cardiovascular risk factors are associated with high plasma choline concentrations (29).

The purpose of the existing study was to observe the pooled effect of multiple low-risk lifestyle factors: diet habits, physically activity, BMI, smoking and education, lipid profile, choline level and inflammatory biomarkers on occurrence of cardiovascular diseases.

## Materials and Methods

### Study design and anthropometric measurements

The present study was conducted in the local hospitals of Lahore city. Lahore is the 2<sup>nd</sup> largest city of Pakistan and capital of province Punjab between April 2013 to January 2014, convenient sample aged  $\geq 40$  years male and female were selected for this study. Study participants were interviewed in person using a structured questionnaire to obtain information on demographic background, dietary habits, socioeconomic status, medical history, physical activity and smoking history. Height and weight were measured while the subjects were barefoot and wearing only their cloths without any upper. Body mass index (BMI) was calculated as adult weight (kilograms)/adult height (inches) (30).

### Dietary assessment

Dietary intakes were assessed using 24 Hour Recall method and assessed twice a week. A structured questionnaire was developed to interview the participants about their food, beverage and supplements consumption during the preceding 24 hours (31). All records were checked at return and any incomplete information was clarified. The daily average of energy consumption and nutrients intake such as total carbohydrates, protein, and fat were calculated.

## Measurement of serum lipids

For biochemical analysis 9 ml of venous blood sample was collected from each participant with proper aseptic precaution to measure serum lipids, total cholesterol (TC), low-density lipoprotein cholesterol (LDL-C), triglycerides (TG) and high density lipoprotein cholesterol (HDL-C); random blood glucose (RBG); cardiac bio marker including troponin, CPK and choline; inflammatory bio markers such as uric acid, urea, ESR, CRP, creatinine. Briefly, blood specimens were drawn and instantly centrifuged for 15 minutes at 3000 rpm and 4°C for plasma separation, and stored at -40°C until assayed. Total cholesterol, Triglyceride, HDL-C and LDL-C were measured enzymatically with specific commercially available kits. Random serum glucose was measured by the glucose-oxidase method by means of commercial kits (Nanjing jiancheng biotechnology Co).

## Statistical analyses

A multivariate logistic-regression analysis under modal PCA by using R: software (R:A language and environment for statistical computing. R: Foundation for Statistical Computing, Vienna, Austria) was applied on the data obtained at the end of each analysis of cases and controls (32). The adjusted mean, SD and risk level (OD) with 95% confidence interval were calculated and are presented for each variable. The statistical significance will be defined as  $P \leq 0.05$ .

## Results

Comparison of established risk factors, blood lipids, inflammatory biomarkers, Cardiac-biomarkers, Gender, Age, dietary habits and BMI score in cases and controls are shown in table 1. Body mass index was significant with  $p = 0.036$ . Similar studies emphasize Body mass index which relates weight to height, is the most widely used and simple measure of body size, and is frequently used to estimate the prevalence of obesity within a population (33). On the other hand Age is was also found to have association with CVD ( $p = 0.059$ ).

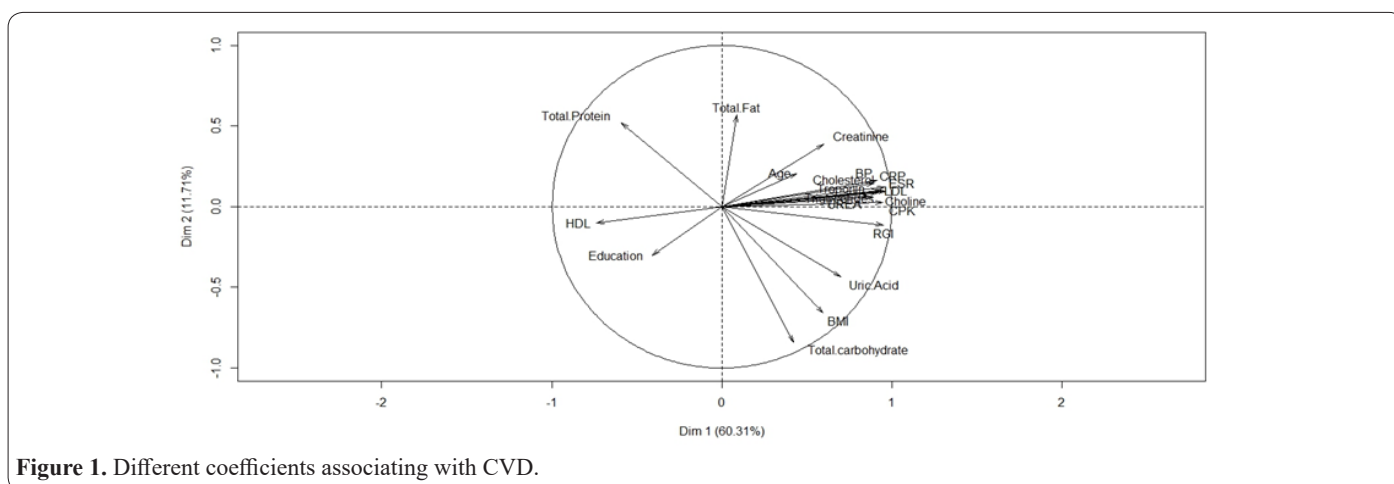
Data showed that dietary pattern have a significant role in the development of cardiovascular disease; the total fats ( $p = 0.017$ ), total carbohydrate ( $p = 0.076$ ) and total proteins ( $p = 0.287$ ). Moreover, cardiac biomarker choline ( $p = 0.026$ ) and Troponin ( $p = 0.001$ ) concentration were high in the cases than normal. The prevalence of high glucose, high blood pressure, Cholesterol ( $p = 0.025$ ), HDL ( $p = 0.023$ ), LDL ( $p = 0.011$ ) were significantly higher among cases than in controls.

Results revealed that the anthropometric and biochemical risk factors are inter-correlated with each other and contribute to CVD prevalence. Therefore a different approach was used to identify the major risk factor of CVDs. Principal Component Analysis (PCFA) was used to analyze the relation of various risk factors to CVDs (Figure 1). Total carbohydrate, BMI, Uric acid, RGI, Age, LDL, Choline, BMI, creatinine, blood pressure, and total fat, total are major risk factors for CVDs. In addition increase of HDL and education decrease the occurrence of CVDs.

Choline is an important component of phospholipids, plasma lipoproteins and cell membranes, and is

**Table 1.** Risk Estimation between Cases and Control Groups.

Coefficients	95% confidence Level			p-Value
	OR	Lower CL	Upper CL	
Blood Pressure	1.0563	1.0176	1.098	0.004 **
Random Glucose Level	1.011	0.9995	1.0229	0.062
Cholesterol	1.0115	1.0015	1.0219	0.025 *
Triglycerides	1.0104	0.9896	1.0321	0.332
HDL	0.9454	0.8997	0.9915	0.023 *
LDL	0.9871	0.977	0.9969	0.011*
Uric Acid	1.0294	0.7635	1.3887	0.848
UREA	1.0348	0.9885	1.0847	0.146
Creatinine	0.5664	0.105	3.005	0.503
CRP	0.9866	0.9465	1.0281	0.522
ESR	1.011	0.979	1.0443	0.506
Troponin	0.9662	0.9459	0.9859	0.001 **
CPK	0.9865	0.9698	1.0029	0.110
Choline	0.944	0.8964	0.9927	0.026 *
Total. Carbohydrate	1.1536	0.9866	1.3553	0.076
Total. Protein	1.1456	0.8933	1.4778	0.287
Total. Fat	1.3301	1.0552	1.6948	0.017 *
Age	1.0347	0.9989	1.0726	0.059 .
BMI	1.1284	1.0092	1.2674	0.036 *



**Figure 1.** Different coefficients associating with CVD.

essential for the formation of the neurotransmitter acetylcholine. Blood Choline concentration is dependent on dietary source and metabolic disorder, for example Ischemic membrane injury, phospholipid breakdown, mitochondrial dysfunction and atherosclerotic plaque progression. Furthermore, data suggest that high blood LDL and total fat consumption increase the blood choline (Table 2). Other major contributing factors for elevated blood choline were CRP, ESR and BMI.

**Discussion**

Odds ratios express the associated risks with CVD and without CVD patients. In normal people groups, comparatively better dietary habits and active lifestyle observed. Increasing age has been described as a cardiovascular risk factor from the start of cardiovascular epidemiology (34). The prevalence of atherosclerosis (35), metabolic dysfunction (36), and immune dysregulation (37) all increase with age, resulting in an increased incidence of hypertension, diabetes, dyslipidemia, and

vascular dysfunction (38). These phenomena have been described in both sexes, although the relevance of these risk factors is more prominent in women compared to men (39).

Cardiac-biomarkers, Gender, Age, dietary habits and BMI levels in cases and controls. Body mass index was very significantly with  $p= 0.036$ . BMI has been found to be consistently associated with an increased risk of cardiovascular disease (CVD) and type 2 diabetes (40), yet this measurement does not account for variation in body fat distribution and abdominal fat mass, which can differ greatly across populations and can vary sub-

**Table 2.** Estimated risk of following parameters with blood choline.

Parameters	Std. Error	P value
LDL	0.012	0.0062**
CRP	0.055	0.036 *
ESR	0.043	0.097
BMI	0.148	0.087
Total Fats	0.272	0.005 **

tantially within a narrow range of BMI (41). Excess intra-abdominal fat is associated with greater risk of obesity-related morbidity than is overall adiposity (42). High blood pressure is usually associated with certain cardiovascular disease (CVD) risk factors conjointly resulting in increased CVD morbidity (43). Both, systolic and diastolic blood pressure correlated positively to a statistically significant extent with BMI. This is not surprising and has been the experience in several studies. Dudina *et al.* (44) expressed that a single unit rise in BMI was associated with a 1.14 mmHg rise in SBP. Doll *et al.* (45) also found that both SBP and DBP rose with BMI across all populations. Individuals who are obese tend to have hyperinsulinaemia which in various ways is related to elevated blood pressure. There was also a positive significant correlation with total cholesterol, an experience shared by (46).

Present study showed that the total fats and total carbohydrate significantly correlate with CVDs occurrence. Previous studies suggest that dietary pattern have significant role in cardiovascular disease (47). Cardiac biomarker choline concentration was high in CVD patients. It has been suggested that elevated levels of blood choline is markers of ischemia (48).

The prevalence of high glucose, high blood pressure, Cholesterol, HDL, LDL was significantly higher among cases than in controls. Different blood parameters like Triglycerides, Uric Acid, UREA, Creatinine, CRP and ESR were non-significant observed. Lane *et al.* (49) studied that baseline circulating concentrations of C-reactive protein (CRP) are significantly associated with cardiovascular disease risk in general populations. This modest association has been inappropriately conflated with causality, and it has been claimed that CRP is pro-atherogenic. C-reactive protein (CRP), the classical acute phase protein, is the most extensively studied systemic marker of inflammation (50). There is also uncertainty about the separate issue of the value of measurement of CRP in the long-term prediction of cardiovascular outcomes (51,52). In females the low carbohydrates and high protein and frequent salad vegetable consumption observed. On the other hand, men consume more carbohydrates. Body mass index was very significantly with  $p=0.036$  (OD 1.12 95% 1.00-1.26). The total fats ( $p=0.017$ ) (OD 1.3301 95% 1.05-1.69) total carbohydrate ( $p=0.076$ ) (OD 1.1536 95% 0.98-1.35) and total proteins ( $p=0.287$ ) (OD 1.1456 95% 0.89-1.47).

The present study is the first evaluation of lifestyle, dietary habits and choline biomarker risk estimation for cardiovascular diseases in the local population. To reduce cardiovascular morbidity in these communities, BMI, TC, UA and salt intake in the diet should be targeted for reduction. Physical activity should be encouraged. Interestingly, these fall into the sphere of a healthy lifestyle which should be encouraged and reinforced.

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