Current Molecular Diagnostics of Cardiovascular Diseases-A Step Closer to Personalized Medicine

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ABSTRACT

Concept of study: The leading cause of death worldwide is not communicable diseases or infectious diseases, rather it is non-communicable diseases and life style related disorders. Among non-communicable diseases, cardiovascular diseases is attributable as the leading cause of mortality worldwide. Cardiovascular disease is a multifactorial disorder and thus it is more important to prevent it beforehand rather than finding a cure. It is known fact that cardiovascular diseases are more prevalent in elderly/upper age group individuals, but owing to increasing work stress and sedentary life style, younger age group under fifty years are also suffering from early cardiac diseases. This can be due to accelerated biological aging. Therefore in this study we have tried to find possible links between lifestyle disorders and cardiovascular diseases so that we can prevent the outcome of the disease at the earliest.

Key words: Biomarkers, Cardiovascular Diseases, Intervention, Risk factors, Sedentary life style.

INTRODUCTION

According to the World Health Organisation (WHO) 2014 global reports on cardiovascular disease, 17.9 million deaths occurred due to Cardiovascular diseases(CVDs) in the year 2012 worldwide CVD are the major cause of worldwide death, particularly in the elderly population presenting an increase rate of mortality and morbidity, they are consequences of genetic and epigenetic interaction.[1] microRNAs (miRNAs or miRs The genetic components include genomic instability, cellular senescence, signalling network, dietary restriction, molecular damage, in particular oxidative damage, over activity, loss of proteostasis, mitochondrial dysfunction, stem cell exhaustion, and alternation in intercellular communications.[1] microRNAs (miRNAs or miRs owing) to the broad and heterogeneous population of patients with CVDs, early risk stratification is highly needed to guide the therapeutic management and an improved outcome.

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Cardiovascular diseases

CVDs are among the major health issues in society and is expected to be the number one cause of death in worldwide in 2020.[2] There are several subtypes of CVDs which include myocardial infarction, coronary artery disease, stroke, peripheral artery disease and congestive heart failure.[3] In majority cases, these clinical conditions result from atherosclerosis which is a progressive and inflammatory disease of the arterial wall.[4] There are evidences where, oxidative-stress-induced genome instability, DNA strand breaks, altered nucleotides, micronutrient deficiencies, impaired homocysteine metabolism may be significantly responsible for developing acute atherosclerotic plaque.[5–8] An emerging field of predictive and preventive medicine involves prediction of CVDs, a number of factors are analysed such as, pathophysiology of CVD, inflammatory markers, platelet aggregation, lipoprotein or lipid-related metabolic mechanisms.[9]

Major types of cardiovascular disease most prevalent worldwide

Myocardial infarction

Myocardial infarction(MI) or acute myocardial infarction (AMI) is a medical term for an event commonly known as

a heart attack.[10] In AMI, unstable build-up of white blood cells(WBC), cholesterol and fats blocks the flow of blood to one of the coronary artery, which causes injury to a part of the heart due to lack of oxygen availability. The event is called "acute" if it is sudden and serious.[10] In many cases (about 64%), the person does not have chest pain or other symptoms, these are called "silent" myocardial infarctions.[11]

Coronary artery disease (CAD), also known as atherosclerotic heart disease, coronary heart disease (CHD), or ischemic heart disease (IHD). This is the most common type of heart disease and primary cause of heart attacks. Most individuals with CAD show no sign or symptoms, but often the person may experience a sudden heart attack. Risk of CAD increases with increasing age, smoking, high blood cholesterol, high blood pressure, diabetes and with incidence of CAD among close relatives. Other causes include coronary vasospasm,[12] a spasm of the blood vessels of the heart, it is usually called Prinzmetal's angina.[13]

Cardiac hypertrophy or ventricular hypertrophy is the thickening of the ventricular walls (lower chambers) in the heart. Enlargement can also occur in the right, or both ventricles. Healthy cardiac hypertrophy (physiologic hypertrophy or "athlete's heart") is the normal response to healthy exercise or pregnancy[14] which results in an increase in the heart's muscle mass and pumping ability. Pathological hypertrophy increases the size of the heart without enhancement of its pumping ability, and accumulates myocardial scarring (collagen). In pathological hypertrophy, the heart can increase its mass by up to 150%.

Cardiac arrhythmia

Cardiac dysrhythmia is a group of conditions in which the electrical activity of the heart is irregular i.e. faster or slower than normal.[15] Although many arrhythmias are not life-threatening, some can cause cardiac arrest. It may occur at any age. Some are barely perceptible in terms of palpitations, whereas others can be more dramatic and can even lead to sudden cardiac death.[16]

Congestive heart failure

Heart failure (HF), often used to mean chronic heart failure (CHF), occurs when the heart is unable to provide sufficient pump action to maintain blood flow to meet the needs of the body.[17] Edema along with heart failure is termed as congestive heart failure or congestive cardiac failure (CCF). Heart failure can cause a number of symptoms including shortness of breath, leg swelling, and exercise intolerance. Common causes of heart failure include myocardial infarction

(heart attack) and other forms of coronary artery disease, hypertension, valvular heart disease, and cardiomyopathy.

Sedentary life style and cardiovascular diseases

The global trends in deaths from CVD predicts a greater contribution from middle and low income countries compared with high income countries in near future. The life style of people is usually controlled by the socioeconomic status. Obesity, hypertension, sedentary life style etc., are mere reflection of the socioeconomic status.[18] Sedentary life-style can be suitably defined as activities that do not increase the energy expenditure above the resting level of 1.0-1.5 Metabolic equivalents (METs).[19] CVD is highly influenced by life-style factors, sedentary life-style is a major risk factor for CVD. Smoking is one of the most well-known and established factors for CVD, also more than 60 g/day of alcohol consumption has shown to have a greater risk of developing CVD, increased distribution of central fat is more related to cardiovascular disease then the generalized obesity. The main markers of cardiovascular diseases are limited aerobic capacity and reduced muscle strength.[20] We further discuss the impact of life style related factors in causing CVD.

Lack of physical activity (PA)

During adolescence, a fast reduction in the level of PA usually takes place, but if regular PA is continued from childhood it will help to have a healthy adulthood. Physical inactivity increases the risk of many adverse health conditions including CVD, type-2-diabetes, premature mortality etc.[21] In a study it has been shown that a transition from high to low levels of daily PA for only 5 days impairs flow-mediated dilation(FMD) in the popliteal artery, and produces an increase in circulating levels of CD31^{+}/CD42b_ Endothelial microparticles(EMPs).[22] Few data exist examining the vascular consequences of short-term reductions in daily physical activity. Thus we tested the hypothesis that popliteal and brachial artery flow-mediated dilation (FMD) Regular PA, enough sleep duration and good sleep quality are factors which lowers the risk of CVD. There are many population based studies done whose reports suggests that physically active people are less insomniac, experience less nocturnal awakenings, have shorter onset latency, less sleep insufficiency, and also feel less tired during the day than inactive persons.[23]

Addiction to smoking/caffeine/alcohol/junk food habits

According to American Heart Association (AHA) statistical fact sheet 2014 update, majority of the younger population

are getting addicted to smoking and alcohol. It is widely reported that consumption of moderate amount of alcohol lowers the risk of CVD, but people consuming more than 60 g/day of alcohol, reported to have greater risk of developing CVD. Drinkers with variable alcohol intake have an increased risk of CVD as well as total mortality.[24] Also people who possess a genetic variant of alcohol dehydrogenase type-3, have a greater chance of developing CVD on alcohol consumption.[25] sedentary lifestyle, alcohol intake and body mass index (BMI) People carrying the epsilon 4 allele of APOE gene are more vulnerable towards developing CVD.[25] Hyperhomocysteinemia is an independent risk factor for stroke and other vascular events. Studies have shown that there is an association between T-allele of MTHFR C677T with cardiovascular disease and stroke. Smoking and MTHFR C677T may interact concerning the association with homocysteine plasma levels and vascular diseases arguing for hyperhomocysteinemia as a casual risk factor for vascular disease. In a study it was observed that likelihood of being a smoker was lowest in those, who had homozygous allele for the wild-type allele 677CC of MTHFR gene, while those who had the presence of a mutant allele increased the chances of being a smoker.[26] Similar to smoking of tobacco, smokeless form of tobacco is also reported to cause CVD in very few studies, but more study is required to relate an association between addiction of smokeless tobacco and CVD.[27]

Work pressure, working hours, deadlines, stress

Work pressure or stress is the result of lack of balance between three work factors: work demands, work support, and work constraints. Studies in the United States and Japan have found positive association between over-time work, shift work, and long travelling time to work and cardiovascular diseases.[28] The physiological response of residual stress may increase sympathetic nerve activity and elevate blood catecholamine levels which may be detrimental to health over a long period of time. Elevated blood pressure responses could lead into thickening of artery walls and narrowing of the blood vessels, thus increasing the risk of developing cardiovascular diseases.[28] There has been reports showing a higher incidence of myocardial infarction in employees with low intensity of work but high responsibility.[29] Few studies have been reported on association between chronic stresses with CVD which suggests that, stress is a significant predictor of stroke. Also, waist circumference was found to be a very strong mediator for association of stressful life events and social strain with both CHD and stroke.[30] Individuals may adopt certain behaviours as a result of chronic exposure to psychosocial stressors that are related to CVD risk, including cigarette smoking, high fat and carbohydrate food intake, physical

inactivity, and heavy alcohol use.[30] Job strain is conceived as a chronic stress or that contributes to a progressive rise in blood pressure (BP) and hence hypertension. Employees with a high job strain have a higher BP at home during sleep, as well as at work. Higher BP at sleep suggests a possibility that job strain may impact the circadian rhythm. A number of studies have reported that hypertensive patients show more end-organ damage and higher cardiovascular morbidity and mortality than those who show normal BP.[31] Clinicians are also not immune to the effects of unhealthy life style and work pressure. This specific kind of shift work includes frequent stressful situations, sleep deficit, and disturbance of the circadian rhythm. Cardiac autonomic function is largely under the influence of sleep- wake cycle with a relative dominance of the sympathetic system during daytime and the parasympathetic system during night-time. Generally it is assumed, irregular sleeping patterns cause fatigue and adversely affect physiological functions.[32] Common workplace stressors include organizational stressors, in adequate communication and inter-personal conflict, task stressors, and work environment stressors, these stressors are considered to be associated with increased risk of cardiovascular diseases.[33] Job strain affects the development of cardiovascular diseases by directly altering standard biological risk factors.[34] Exposure to high concentration of heavy metals like Cadmium is reported to be detrimental to the health of exposed workers. Cadmium is identified as a risk factor for cardiovascular diseases in humans. In some conditions, a reduction in nitric oxide (NO) bioavailability contributes to the appearance of diseases such as atherosclerosis, myocardial infarction, coronary heart disease and hypertension. Oxidative stress induced by cadmium might also increase the lipid peroxidation, which in turn could induce atherosclerosis. Acute cadmium toxicity that when occurring repeatedly, may initiate an inflammatory process and atherosclerosis development in the aorta.[35]

Sleep deprivation

In developed as well as developing nations, people are working in shifts to increase the output, which actually results into poor sleep health and in turn account for sleep disorders e.g., sleep apnea, insomnia, narcolepsy, and restless legs syndrome. There are reports showing association between long sleep duration and cardiovascular outcomes such as hypertension, stroke, etc.[36] FMD is reduced after a single 24-hour work shift, also chronic stress with sleep deprivation was associated with reduced FMD. Substantial partial sleep restriction in healthy individuals leads to impaired endothelial function.[37] As per a study, mortality rates from ischemic heart disease, cancer, stroke etc., were lowest for individuals sleeping 7 or 8 hours

per night, and men sleeping 6 hours or less or 9 hours or more, had 1.7 times the total age-adjusted death rate of men sleeping 7 or 8 hours per night. Sleep deprivation also increases sympathetic nervous system activity, heart rate, and vasoconstriction as well as salt retention. These factors may be associated with hypertension caused by cardiac overdrive and volume overload. Working more than 60 hours a week was related to increased incidence of AMI.[38] Evidence suggests that short sleep duration is associated with obesity, all-cause mortality, diabetes, and cardiovascular disease. There are number of potential mechanisms that might be responsible for these associations and include effects of sleep on appetite, insulin resistance, endothelial function, inflammation and thrombotic or hemostatic factors. Damage to the endothelium can lead to activation of hemostatic, coagulation, and thrombotic pathways, which may lead to the formation of a thrombus, impaired blood flow, and possibly a stroke.[39]

Accelerated biological aging/ senescence

Cell senescence is defined as the irreversible loss of the ability of the cells to divide. There are two types of cell senescence - a) replicative senescence, and b) stress-induced premature senescence. Senescence associated β-galactosidase (SAβG), is a lysosomal enzyme seen in senescence of multiple human cell types. Increased numbers of SAβG-positive vascular smooth muscle cells (VSMCs), endothelial cells (ECs), and monocytes/macrophages are observed in aged vessels and atherosclerotic lesions when compared with their respective young and normal counterparts. This gives the idea very good support that atherosclerosis is associated with premature cellular senescence. There are reports where people have found shortened telomeres in the event of atherosclerosis, observed in plaques VSMCs and ECs, relative to normal vessel wall, and in circulating endothelial progenitor cells (EPCs).[40] The decreased number of senescence of circulating EPCs is considered markers of vascular senescence associated with aging, atherosclerosis, and CAD. There are reports published where EPCs from premature coronary artery disease (PCAD) patients showed a reduction in the telomere length and telomerase activity. PCAD patients have accelerated vascular senescence as compared to those without CVD. Aging associated numerical and functional decline of EPCs in CAD patients has been attributed to exhaustion of stem/ progenitor cells in the bone marrow due to chronic vascular injury, reduced mobilization, diminished migratory and adhesion capacity of EPCs and differentiation, and is thought to render the elderly more prone to endothelial dysfunction and cardiovascular diseases. Impaired repair mechanism of the body predisposes individuals to endothelial dysfunction at an early age.

Early-accelerated vascular senescence plays a crucial role in premature CAD in developing countries.[41] In another study, presence of the CC genotype of vascular endothelial growth factor(VEGF), the A allele of Interleukin (IL-10), and the A allele of IFN-γ resulted to be associated with an increased risk of AMI and was more frequent among offspring's with a high positive parental history of AMI. So, AMI is a multifactorial disease with a complex pathogenesis where lifestyle, individual genetic background and environmental risk factors are believed to play crucial roles.[42]

Major risk factors responsible for developing cardiovascular diseases

There are multiple reasons for death due to CVD and one of the prominent one is lack of early diagnosis. Hypertension and diabetes are among the main risk factors of CVD in global population. By the end of 2015, CVD has been predicted to be one of the leading cause for death.[43] Sometimes the risk factors may be related to pollution, such as annoyance due to noise such as noise caused by aircrafts, rails and daily traffic.[44] Reports are there which shows that chronic exposure to noise leads to increase in the blood pressure and heart rate and increase of noradrenalin levels in humans.[44] In the year 2013 it has been reported that there is significant increased risk of stroke, CHD, and CVD, especially among the population living near the airports who are affected by the highest level of daytime and night time aircraft noise.[44] The onset of pathological events occur at a very early phase of the disease, but the multifactorial nature of CVD makes it very challenging to confirm the initial pathological events. Atherosclerosis is the most common pathological process that leads to CVD including MI, heart failure, and stroke. Necrotic cores, calcification, accumulation of modified lipids, foam cells, vascular dendritic cells, T-cells, and endothelial cells are characteristic features of atherosclerotic plaques.[45] Oxidation of low density lipoproteins (LDL) is an early event in atherosclerosis. Cholesterol containing LDL particles are modified biochemically in the area of lesion in the artery wall, which induce leukocyte adhesion, also activation of macrophages add into the cause by disrupting the fibrous cap and thrombus formation. Sometimes disruption of a vulnerable plaque may lead to a complete occlusion, to plaque progression or result in an acute coronary syndrome, i.e., AMI, and sudden cardiac death or stroke in case of carotid plaque disruption.[45] Inflammation-associated oxidative stress may lead to a rapid consumption of circulating antioxidant, and low intake of antioxidant in particular vitamins and increased homocysteine levels were found to be associated with atherosclerotic outcomes and risk.[45] Single nucleotide polymorphism in the alcohol

Table 1: Parameters used to diagnose cardiovascular diseases (CVDs)

Variables	General diagnosis parameters
a) Preliminary check up by the physician	Blood pressure (BP)
	Echocardiogram(EKG)
	Echocardiography(ECG)
	Stress- ECG or Treadmill Test(TMT)
	Stress - Echo
	Stress- Thallium
	Angiogram
	Coronary Angiography/ City- angiography
b) Anthropometric measurements	Body weight
	Height
	Waist: Hip ratio (very occasional or rare)
c) Blood test is generally prescribed, mainly to know the lipid profile of the patient	% saturated fatty acid
	% mono-unsaturated fatty acid
	% poly-unsaturated fatty acids
	% low-density lipoprotein-cholesterol
	% high-density lipoprotein-cholesterol
	Lipoprotein A(LPA)
	Blood sugar
	Homocysteine
	Fibrinogen concentration

These parameters used by clinicians, only gives superficial idea about the outcome of the disease, but does not provide knowledge of molecular details, which is actually developing the disease.

dehydrogenase 1B gene (ADH1B) occurs due to heavy alcohol consumption, which encodes for the ADH1B enzyme and provides the primary pathway for the alcohol metabolism.[46] Reports are published about some potential protective effects of light to moderate alcohol consumption on risk of coronary heart disease and stroke but more work has to be done.[46] Often some physiological stress parameters are not considered as important parameters for CVD, one such parameter is sudden outbursts of anger, which have been reported to be associated with an abrupt increase in the cardiovascular events such as acute myocardial infarction, acute coronary syndrome, arrhythmia, ischemia and ischemic and hemorrhagic stroke.[47] In South Asian developing countries including India, prevalence of CVD risk factors is found higher in urban population, yet it is increasing at an alarming rate in the rural population also.[43]

New edge predictors and biomarkers of cardiovascular diseases

In developing countries of south East Asia, the diagnosis of CVD is done with the help of some known parameters which are enumerated in Table 1.

Discovery of few biomarkers related to CVDs are becoming an important area of interest, there are reports where anaemia has been shown to be associated with three-fold increase risk of death or myocardial infarction. Current life style has led to intake of junk food rich in harmful fatty acids which increase the level of cholesterol in the body. Interestingly

there are reports where the prediction of chronic diseases related to inflammation and oxidative stress is improved by consumption of long chain omega-3 polyunsaturated fatty acids(LC n-3 PUFA), especially eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA).[49] LC n-3 PUFAs modulates specific risk factors such as reduction of platelet aggregation, decrease of plasma triglycerides and BP as well as direct regulation of systemic and local inflammation underlying plaque formation, progression and instability.[49] In another context, relationship between periodontal disease and CVD was reported.[50] Studies have also shown that, subjects with severe periodontal disease had significantly higher calcium level which contributes to greater calcification of cardiac structures. Thus it might be a more important risk factor in the younger individuals for calcification of the cardiac structure.[50] Just like periodontal disease, anaemia not only decreases oxygen delivery to the injured myocardium and periphery but also associated with arrhythmias, and hypertension.[48] Another emerging topic in cardiac research is the concept of microparticles. Microparticles are small cell vesicles that are released by eukaryotic cells during cellular stress and cell-activation.[51] Within the last 1-2 decades it has been shown that microparticles are released by platelets, leukocytes and endothelial cells and are useful blood surrogate markers of different pathological conditions, such as acute and chronic vascular inflammation, endothelial dysfunction and arterial hypertension. Circulating microparticles can fuse with distinct target cells and delivers cellular components of their parental cells to the target cells. Specific microparticles

Table 2: Parameters which are known to involve in CVDs, but are not used in diagnosis of CVD

Variables	Probable diagnosis parameters	References
a) Background DNA damage measurement	Formation of micronuclei, anaphase bridges, chromosomal aberrations	[54]
b) Up-regulation/Down-regulation of a particular target gene involved in CVD	KCNAB1, KCNH2, KCNA4, RyR2, ATP2A2, KCNQ1, CLOCK, LEP.	[55,56]
c) miRNA status	miR-1, 17,210, etc.,	[1]
d) Presence of exosomes, or microparticles	Diagnosis for miRNA packed inside the exosomes or microparticles.	[57-59]
e) New biochemical parameters	Protein biomarkers :- 1) Apelin 2) Neopterin 3) Serum gamma gluteryl transferase	[52,53,60]

subtypes are increased in conditions of enhanced vascular inflammation and coagulation. Microparticles released from platelets attached to neutrophils and activate those, thus it may in fact be an additional link between vascular coagulation and inflammation in cardiovascular disease.[51] Many studies have focused on proteins, genes and micro RNA (miRNA) to be suitable candidates as biomarkers for the screening purpose of CVD, one of the protein biomarker is apelin, which is strongly expressed in heart, large conduit vessel, coronary vessels, and endothelial cells, the plasma level of Apelin decreases in cardiovascular diseases, including ST-elevated myocardial infarction and stable angina, whereas the level of apelin peptides increases in the human atherosclerosis coronary artery tissue. Plasma apelin levels could be co-related with the severity of coronary artery stenosis and the stability of atherosclerotic plaque in humans.[52] Gamma-glutamyl transferase (GGT) which is a cellular product of oxidants is also considered as a biomarker for CVD, in some recent epidemiological interpretations it has been found that serum GGT positively co-related with cardiovascular diseases such as myocardial infarction, stroke, high blood pressure, which enumerates that serum GGT may participate in oxidative and inflammatory processes.[53] Circulating oxidised low-density-lipoprotein (oxLDL) ia an important factor which was reported to be present in the CVD patients.[45] Enhanced serum levels of oxLDL are predictive for endothelial dysfunction and coronary heart disease, and thus can be used to be a predictive marker for future coronary heart disease events in apparently healthy men. High density lipoprotein is another potential biomarker with anti atherogenic properties due to its function in the reverse cholesterol transport and in decreasing lipoprotein oxidation. Plasma HDL cholesterol(HDL-C) levels are inversely associated with CVD risk in preclinical and large epidemiological studies.[45] Another recently discovered biomarker is Neopterin, whose concentration in the body fluid is considered as an indicator for the immune activation-associated oxidative stress. TNFα can directly enhance the IFNγ production, which is known to be the major stimulus for the formation of neopterin. Moreover patients with CVD were reported to have increased neopterin concentrations, supporting the involvement of the chronic immune activation, in particular of macrophages, in atherogenesis, which strengthens the use of neopterin as an independent marker of CVD, with predictive value for CVD progression.[45] Also genes like Matrix metallopeptidase-9 (MMP-9), whose expression is reported to get induced in human macrophages may affect the stability of the atherosclerotic plaques, has been considered as a marker of instability of atherosclerotic plaques.[52] Some known parameters, known to be involved in CVD, but not included in the clinical diagnosis are listed in Table 2.

These parameters were found to be related with various cardiovascular diseases, these parameters play crucial role in development of CVDs, and diagnosis of these parameters reflect the actual scenario of the disease, which will further lead into the concept of personalized medicine.

Recent studies have pointed out that use of miRNA can be useful in terms of being a potential marker for various diseases and targets for various diagnostic and therapeutic applications. miRNAs are promising targets for drug and biomarker development, they can regulate hundreds of target mRNAs containing partially complementary sequences. Binding of the miRNA containing nucleoproteins to the mRNA leads to destabilization as well as inhibition of the translation.[61] Some miRNAs which have already reported to be involved in common CVDs prevalent in Southeast Asia is listed in Table 3.

DISCUSSION

The most effective approach to cardiovascular disease prevention is risk factor reduction in apparently healthy people rather than in individuals at high risk of future CHD events. Focus must be on tobacco control, healthy diet and exercise. It is important to start at the very basal level by providing children with healthy school meals and physical education.[75] On the other hand we have also tried to look upon the interaction between life-style related disorders (CVD, diabetes, obesity, dental plaque, cirrhosis) by looking

Table 3: Common CVDs and corresponding miRNAs involved

| Most common type of CVD	miRNA involved	Source of miRNA	Increased or Decreased level of miRNA	References
CAD	miR-126, 17, 145, 155	Plasma or Serum	Decrease	62
CAD	miR-221 , 222	PBMC	Increase	63
CAD	miR-146 a/ b	PBMC	Increase	64
Premature CAD	miR-340*, 624*	Platelets	Increase	65
ACS	miR-19, 21, 146, 155, 133	Plasma MPs	Increase	66
Unstable Angina	miR-145	Plasma	Increase	67
Unstable angina pectoris	miR-135a	PBMC	Increase	68
AMI	miR-1	Plasma	Increase	69
AMI	miR-133, 208 b, 499	Plasma	Increase	70
AMI	miR-150	Plasma	Decrease	71
AMI	miR-126	Plasma	Decrease	72
Post MI	miR-1, 21	Plasma	Increases	73
Chronic heart failure	miR-133a, 423-5p, 499-5p, 21	Plasma	Increase	74
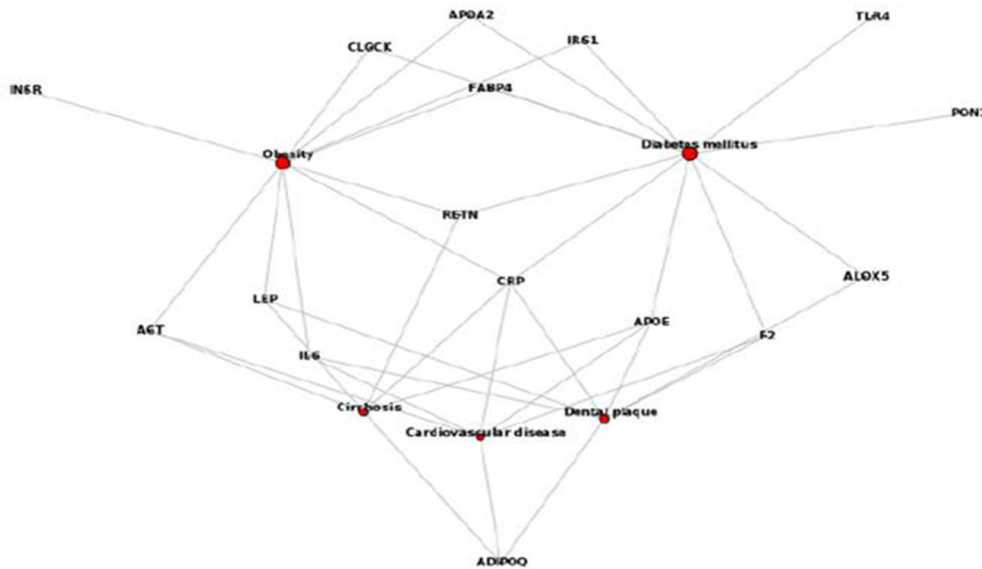
Figure 1: Exploring genes of five lifestyle disorders using functional disease ontology annotations. The size of the disease node is proportional to the number of edges

into certain genes responsible for those disorders and their degree of connectivity, we could find out a interact to me between life-style disorders (shown in Figure 1) and can thus speculate that each of the life-style disorder is responsible for becoming the cause of occurrence of another life-style disorder.[56]

Cutting edge diagnostics is developing bio-fluid based molecular test for personalized medicine.[57] In this field, other than proteins and gene expression studies, expression of miRNAs is becoming the most exciting and innovative mechanism of diagnosis in MI, heart failure and hypertrophy.[76-78] Various important micro-RNAs are eligible candidate to be used in diagnostics of CVD like miR-1, miR-

133a, miR-499 are expressed by skeletal muscle, and the over expression of these miRNAs in plasma might be due to skeletal muscle damage.[79] Over expression of miR-423-5p in plasma in the beginning of an AMI event, and this miRNA has been suggested as a potential early marker of myocardial necrosis.[80] It is also reported that miR-499 levels get significantly high in acute HF patients,[81] also miR-126 is an important regulator of angiogenesis and vascular integrity.[82,83] In figure 1, it can be speculated that other than miRNAs, certain genes are also playing major roles in developing life style disorders, it will a great boon to the society if we can speculate the probability of developing certain life-style disorders including CVD just by checking the status of certain cohort of key genes and pathways affected.
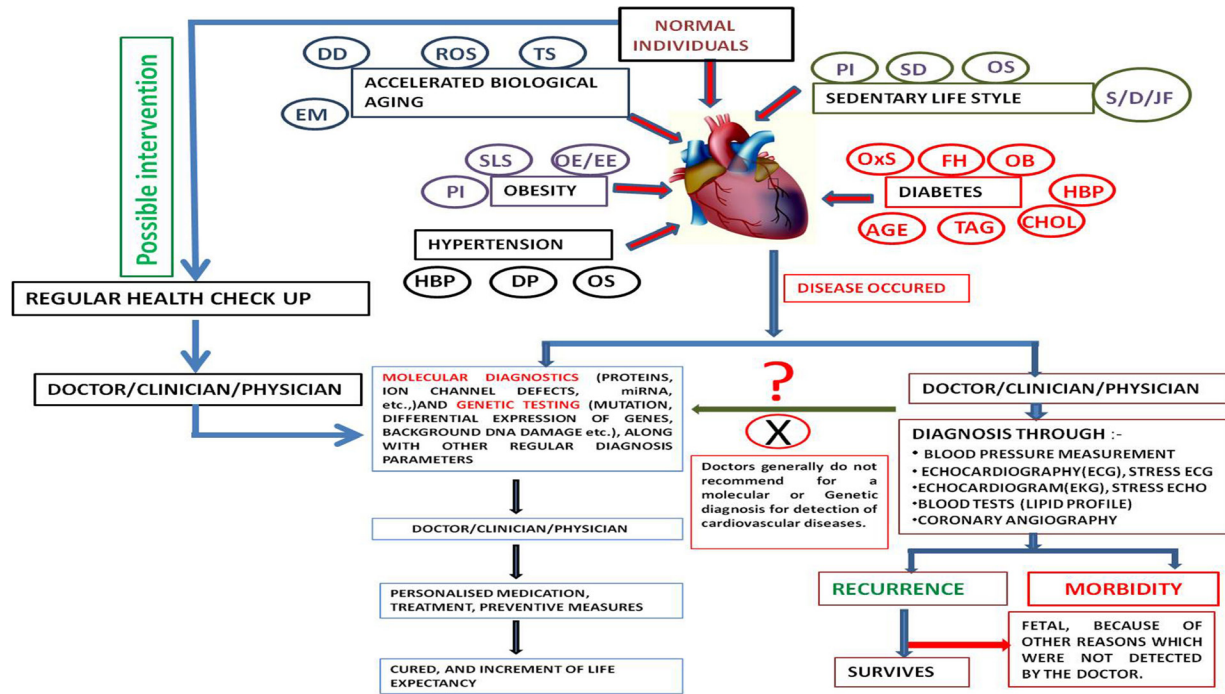
Figure 2: A schematic diagram relating various factors like hypertension, obesity, diabetes, accelerated biological aging, and sedentary life style with prevalence of cardiovascular diseases, also a probable reason for ever increasing rate of mortality from cardiovascular diseases in middle and low income countries has been speculated

CHOL-cholesterol, DD-DNA damage, DP-depression, EE-excess eating, EM-epigenetic modifications, FH-family history, HBP-high blood pressure, OB-obesity, OE-overeating, OS-occupational stress, OxS-oxidative stress, PI-physical inactivity, ROS-reactive oxygen species, SD-sleep deprivation, S/D/JF-smoking/drinking/junk food, SLS-sedentary life style, TAG-triacyl glycerol, TS-telomere shortening.

CONCLUSION

In the past decade many studies have been done in the area of developing new therapeutics and biomarkers for treatment and diagnostics of CVDs but clinical translation of the markers have not become part of routine clinical management or diagnostics of CVDs. It is high time that the approach towards treating CVD patients should be personalised. Each patient should be diagnosed thoroughly, and with availability of high throughput molecular diagnostics platform, treatment can be made personalised, thus providing effective preventive and predictive health care solutions. We have proposed an integrated approach by a schematic representation of the molecular diagnostic management of cardiovascular diseases (shown in Figure 2).

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CONFLICTS OF INTEREST

The authors declare that there are no conflicts of interest.

ABBREVIATION

AC   : Acute coronary syndrome

AMI   : Acute myocardial infarction

CAD   : Coronary artery disease

MI   : Myocardial infarction

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