Original article

Lipid profile fractions responsible for non ST-Elevation myocardial infarction in unstable angina patients of south India

[1]Dr Kiran R. Bagale, [2]Dr Avinash S. Ingle, [3]Dr Rajeev Choudhary,[4] Dr Rita Venugopal

[1] (Biochemistry), Assistant professor, Department of Biochemistry, Pt JNM medical College, Raipur, CG, India.

[2](Physiology), Associate Professor, Department of Physiology, AIIMS-Raipur, CG, India.

[3]Professor, SOS Physical Education, Pt. RSU, Raipur. CG, India

[4]Professor, SOS Physical Education, Pt. RSU, Raipur. CG, India.

Corresponding author: Dr Avinash S. Ingle

Abstract:

Background: Although it is well known that dyslipidemia is the major risk factor for developing acute coronary syndrome (ACS) but its role in causing minor myocardial damage in unstable angina (UA) patients is rarely studied in south India, where obesity and dyslipidemia has been more common due to genetic and life style factors.

Objective: The aim of our study was to find out the role of lipid profile parameters predicting the occurrence of non ST-elevation myocardial infarction (NSTEMI) in the patients of UA.

Methods: In this study, the laboratory parameters like troponin-I and serum total cholesterol (TC), triacylglycerol (TAG), low density lipoproteins (LDL), high density lipoproteins (HDL) were estimated in all patients of UA by using commercially available kits whereas TC/HDL and LDL/HDL ratio were calculated manually. The lipid levels were compared between troponin-I positive i.e. NSTEMI group and troponin-I negative i.e. UA groups. The data was analyses by using unpaired T-test and forward type of logistic regression analysis.

Results: The TC/HDL was found to be the only factor predicting the occurrence of NSTEMI in UA patients, even though the mean values of serum TC, TAG, TC/HDL and LDL/HDL ratio were significantly high in NSTEMI group as compared to UA group.

Conclusion: It can be concluded that TC/HDL ratio is the best risk predictor of myocardial damage in unstable angina patients of south India.

Keywords: Dyslipidemia, non ST-elevation myocardial infarction, unstable angina, troponin-I.

Introduction

In last three decades mortality due to acute coronary syndrome (ACS) has increased in Indians which may be due to reduced physical activities associated with urbanization [1]. It has been noted that the Indian population particularly south Indians are more prone for ACS even at lower levels of known risk factors in comparison with western standard [2].

Dyslipidemia is a well-established risk factor for development of atherosclerotic coronary artery disease. In most of the studies high levels of plasma low-density lipoprotein (LDL) and very low density lipoproteins (VLDL) cholesterol, high triglycerides concentrations are directly correlated with the development of coronary artery disease whereas low high-density lipoprotein (HDL) cholesterol concentrations have been pointed out as one of the strongest independent risk factors for coronary atherosclerotic disease.

Among all the risk factors for coronary artery disease, serum lipids are perhaps least studied in south India. The south Indians being an ethnically susceptible population for coronary artery disease, the threshold for risk factors should be lower. Therefore the studies conducted on Western population for lipid guidelines may not be applicable to south Indians [2].

Patients with ischemic heart disease fall into two large groups i.e. patients with stable angina secondary to chronic coronary artery disease and patients with acute coronary syndrome (ACS). ACS encompasses the patients of myocardial infarction along with ST segment elevation on their presenting ECG (STEMI) & those with unstable angina (UA) and non-ST elevation myocardial infarction (NSTEMI).

Unstable angina is defined as angina pectoris (or equivalent type of discomfort) with at least one of the fallowing features [3].

1. Occurring at rest (minimal exertion) usually lasting >20 min (if not interrupted by nitroglycerin administration)
2. Being severe & described as frank pain or of new onset (within one month)
3. Occurring with crescendo pattern (more severe prolonged frequent than the previous one)

The diagnosis of NSTEMI is established if patient with clinical features of unstable angina develop evidence of myocardial necrosis. Recently it has been seen there is increase incidence of non ST elevation myocardial infarction with fewer patients presenting with classical acute myocardial infarction. So as per the suggestions of National Academy of Clinical Biochemists (NACB), while defining non-ST elevation myocardial infarction, ECG changes plus elevated troponin levels should be considered [4].

Presently there is enough evidence that newer cardiac markers like Troponin-I is more specific, sensitive and reliable in prediction of myocardial damage than creatine kinase-MB (CK-MB) and electrocardiogram (ECG), especially in the cases of micro infarction therefore the elevated levels of troponin-I are used to distinguish patients of NSTEMI from those of UA [5,6].

Although there is no doubt regarding the role of lipids in causation of ischemic heart disease but still the results are confusing regarding the contribution of exact lipid fractions causing the minor myocardial damage in the patients of UA in various populations may be due to ethnicity and life style habits. So our objective in this study was to find out the lipid fractions and ratios responsible for minor myocardial damage of NSTEMI in UA patients of south India.

Methods

The present study comprised of 50 patients (males-36 & females-14) admitted to intensive cardiac care

www.ijbamr.com P ISSN: 2250-284X , E ISSN : 2250-2858

unit of Narayana Medical College, Nellore. The age ranged from 35 to 65 years with mean age of 50 yrs. Informed consent was taken from the patients. The study was approved by institutional ethical committee of Narayana Medical College, Nellore.

Inclusion criteria

Patients admitted with typical symptoms and signs of UA i.e. left pericardial chest pain with duration 5 to 30 min or more radiating to the left arm, jaw, neck, right shoulder associated with sweating, nausea, vomiting and relieved by nitrates along with no ST segment elevation on ECG.

Exclusion criteria

Patients presenting with STEMI, known cases of thyroid dysfunction, liver disease, renal disease, trauma, sepsis, pulmonary embolism, myocarditis, congestive heart failure, muscular dystrophy, skeletal muscle injury were excluded from the study.

Sample collection

First blood sample was collected within 12-24 hours after the onset of chest pain in evacuated tubes and allowed to clot at room temperature. Then serum was separated by centrifugation at 3000 rpm for 10 minutes. Troponin-I was detected by hexagon Troponin plus kit. It detects cardiac Troponin-I with sensitivity limit minimum of 0.5 ng / ml [7].

Then fasting blood sample was collected next day morning after admission for laboratory evaluation of lipid profile. The serum cholesterol was estimated by Cholesterol Oxidase Phenol Aminophenazone (CHOD-PAP) method. The serum TAG was estimated by Glycerol 3 Phosphate Phenol Aminophenazone (GPO-PAP) method. All these estimations were done by using an autoanalyser Humastar 300 (Human Diagnostics).The serum LDL and HDL was measured by enzymatic colorimetric method. The ratio of TC/HDL and LDL/HDL were calculated manually. Control and calibrators were run before each batch to achieve adequate quality control. Troponin-I positivity indicates the myocardial damage, hence troponin-I was estimated to establish the diagnosis of NSTEMI where as troponin-I negative cases were included in UA group. Fasting serum TC, TAG, LDL, HDL, TC/HDL and LDL/HDL cholesterol ratio were estimated to predict the risk in both groups [8].

Data evaluation was done using statistical package for social sciences (SPSS) programme by using unpaired student's-t test. The results were expressed as a mean $\pm$ standard deviation. The p-value was used to compare between two groups. The p-value of <0.05 was considered to be significant. The evaluation of the factors predicting the myocardial injury (Troponin-I positive) the forward type of logistic regression analysis method was used [9].

Results

Out of the 50 patients studied, 17 were Troponin-I positive (14 males & 3 females) which were detected as NSTEMI and 33 patients who were Troponin-I negative (22 males & 11 females) were diagnosed as UA.

As shown in table 1, fig. 1 and 2, the means of TG, TC/HDL, TC and LDL/HDL ratio were significantly higher in troponin-I positive group as compared to troponin-I negative. The mean values of serum LDL and VLDL cholesterol were higher in troponin-I positive group as compared to troponin-I negative group but the difference was not found to be statistically significant. The mean value of serum HDL cholesterol was lower in troponin-I positive group as compared to troponin-I negative group with no significant difference.

www.ijbamr.com P ISSN: 2250-284X , E ISSN : 2250-2858

Table 1: Comparison of various lipid profile parameters, TC/HDL ratio and LDL/HDL ratio with troponin-I test results

Variables	Total number of patients (*n*=50)		
	Troponin-ve (*n*=17), 34%	Troponin +ve (*n*=33) 66%	Significance/ *P* value
TC (mg/dL)	176.18±31.86	208.18±48.09	<0.05
TG (mg/dL)	132.27±43.99	187.82±74.14	<0.01
LDL (mg/dL)	99.30±29.59	108.18±43.36	>0.05
HDL (mg/dL)	49.27±23.82	43.41±27.24	>0.05
VLDL (mg/dL)	28.58±15.42	36.00±14.07.	>0.05
TC/HDL	4.25±0.75	5.71±1.49	<0.0001
LDL/HDL	2.57±0.53	3.11±0.99	<0.05

All values are expressed as mean ± standard deviation.

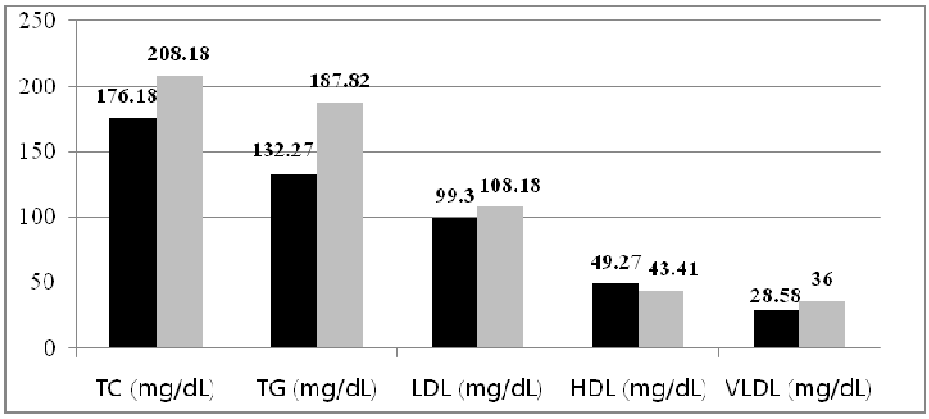

Fig 1: Lipid profile fractions in unstable angina and NSTEMI group.

Troponin-I Negative (UA group)-Black colored, Troponin-I Positive (NSTEMI group)-Grey colored.

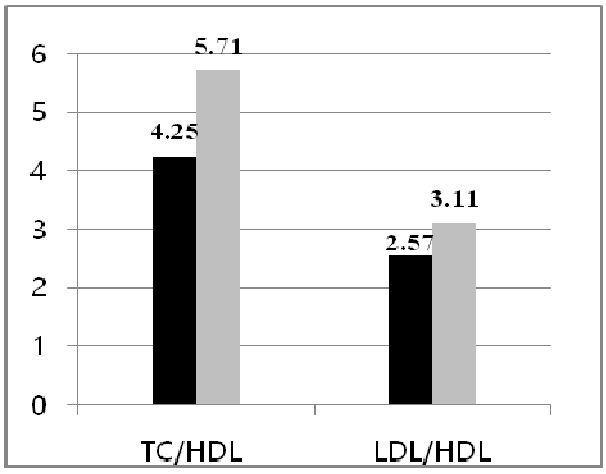

Fig 2: Lipid profile ratios in unstable angina and NSTEMI group.

Troponin-I Negative (UA group)-Black colored, Troponin-I Positive (NSTEMI group)-Grey colored.

After studying the results of unpaired T-test in this study, the logistic regression analysis was used since our interest was to determine the predictors for Trop-I which was a categorically dichotomous variable, so forward conditional method was used. At first to get the amount of variation, the Nagelkerke R square was determined the value of which was 0.44. It tells that 44.1% of the variation in the dependent variable (Troponin-I) is explained by the logistic regression model (Table.3).

Table 2 provides the overall accuracy of model to predict risk of subjects getting NSTEMI 82%. The sensitivity is given 9/10 i.e. 56.25% and the specificity by 32/34 i.e. 94.11%. The positive prediction value is equal to 9/11 i.e. 81.81% and negative predictive value will be 32/39 i.e. 82.05 %. The equation for calculating the probability of having the risk of NSTEMI (Troponin-I becoming positive) will be:

Probability (Troponin-I positive) = $1 / (1+ e^{-Z})$

Where e denotes exponential function with z= -7.976 + 1.503 x TC/HDL

Table 2: Model discrimination table.

	Observed value		**Predicted value**		
			Dependent variable		% Correct
			Troponin-I negative	Troponin-I Positive	
Step1	Dependent variable	Troponin-I Negative	32	2	94.1
		Troponin-I Positive	7	9	56.2
	Overall percentage				82.0
a. The cut value is 0.50					

In table 3, Wald estimates gives the importance of TC/HDL. The Wald value of 10.413 is higher enough to justify the importance of this model. The justification for excluded variables is given in table 4.

Table 3: Estimates of the logistic regression model.

Variables in the Equation									
		B	S.E.	Wald	df	Sig.	Exp(B)	95.0% C.I.for EXP(B)	
								Lower	Upper
Step 1	TC/HDL	1.503	0.466	10.413	1	0.001	4.494	1.804	11.194
	Constant	-7.976	2.245	12.626	1	0.000	0.000		

a. Variable(s) entered on step 1: TC/HDL.

www.ijbamr.com P ISSN: 2250-284X , E ISSN : 2250-2858

Table 4: Variables not in equation.

Variables not in the Equation					
			Score	df	Sig.
Step	Variables	TC	1.138	1	0.286
		TAG	3.502	1	0.061
		LDL	0.675	1	0.411
		VLDL	0.013	1	0.908
		HDL	0.826	1	0.363
		LDL/HDL	2.338	1	0.126
		Overall Statistics	9.323	6	0.156

Discussion

As like previous studies, our study also proved the importance of lipid profile parameters for causation of the heart disease in south Indian community as like other populations. As per our findings TC, TAG, TC/HDL and LDL/HDL ratios were significantly increased in NSTEMI group as compared to UA group. But by forward type of logistic regression analysis, this study proves that TC/HDL ratio is the only factor which has significant contribution in causing the NSTEMI in the south Indian patients of UA even though the HDL levels were not significantly low in NSTEMI patients. Other factors like LDL & VLDL, the levels of which were higher in NSTEMI group as compared to UA group but these findings were not statistically significant. So it can be predicted that even though the severity increases with rise of TC, TAG, TC/HDL and LDL/HDL ratios but the only factor which mainly contributes in causation of NSTEMI is TC/HDL ratio in patients of UA which is one of a primary stage of ACS.

TC/HDL and LDL/HDL cholesterol ratios

TC/HDL and LDL/HDL ratios were significantly higher in troponin-I positive group as compared to troponin-I negative group when compared between two means. But regression analysis proved that TC/HDL ratio was the only factor predicting the occurrence of NSTEMI. Similarly Rafaela et al [10], Amit Kumar Shrivastav et al [11], P.K. Nigam et al [12] and Vishwanathan et al[13] observed TC/HDL cholesterol and LDL/HDL cholesterol ratios significantly high in coronary artery disease as compared to controls. Rafaela Andrade Penalva et al [10] who correlated lipid profile parameters with number of vessel involvement observed that only TC/HDL ratio is the marker of severity in NSTEMI group involving more number of vessels. Regarding these ratios the only conflicting study was Haseeb A. Khan et al [14] who did not find significant changes in TC/HDL cholesterol and LDL/HDL cholesterol ratios.

Serum cholesterol

As like our observations in other studies TC levels were significantly higher in acute MI than controls [11,15]. But R. Salehi et al[16] who compared lipid profile of UA, NSTEMI and STEMI patients with controls observed that serum TC levels were borderline higher as compared to controls although no significant difference was found. Okon Ekwere Essien et al [17] in his autopsy study retrospectively noted the significant rise of TC in MI group as compared to angina group. Haseeb et

www.ijbamr.com P ISSN: 2250-284X , E ISSN : 2250-2858

al [14] studied lipid profile in STEMI and NSTEMI also couldn't find any change in TC, whereas Rafaela et al who studied lipid profile in NSTEMI patients could not found any association TC with severity of ACS [9]. Others P. K. Nigam [12] and Vessa Manninen [18] have noted no significant increase or decrease in total cholesterol after acute myocardial infarction.

Serum triacylglycerol

The mean values serum TAG were statistically highly significant ($P < 0.001$) in NSTEMI group as compare to US group. It may be due to elevated flux of fatty acids and impaired removal of VLDL from the plasma. The studies of R. Salehi et al [16], Amit Kumar Shrivastav et al [11], P. K. Nigam et al [12] and Okon Ekwere Essien et al [17] supported our findings. Jorgen Jeppesen et al [15] conducted prospective study on risk factor of ischemic heart diseases and showed that low TAG and High HDL lowers the risk of developing IHD. Other researchers have observed higher triacylglycerol values in coronary artery disease as compared to controls**.** Unlike our findings some researcher Haseeb A. Khan et al [14], Rafaela et al [10] have noted lower triacylglycerol levels after acute coronary syndrome.

Serum HDL cholesterol

The mean values of serum HDL cholesterol were low in troponin-I positive group as compare to troponin-I negative group which is statistically not significant ($p> 0.05$). Most of the researchers have found significantly lower levels of HDL cholesterol in coronary artery disease patients [12, 14, 15, 16]. But some of the workers also noted that the reduction in HDL occurs only during acute stage of ACS [12, 19].

Serum LDL and VLDL cholesterol

The mean values of LDL and VLDL cholesterol were higher in troponin-I positive group as compared to troponin-I negative group which is statistically insignificant**.** But R. Salehi et al [16], Amit Kumar Shrivastav et al [11], P. K. Nigam et al [12] and Vishwanathan et al [13] noted the significantly high LDL cholesterol values in coronary artery disease as compared to controls. Recently researchers have indicated that the LDL levels get reduced after 24 hours of myocardial damage [14]. Mouaz H. Al.-Mallah et al demonstrated that on admission low LDL levels have positive correlation with increased three year mortality [19].

Summary

This study demonstrated that significantly higher TC, TAG, TC/HDL, LDL/HDL levels were estimated in patients of NSTEMI as compare to UA patients, along with no significant difference of HDL. But by logistic regression (binary) analysis TC/HDL ratio was found to be the only factor predicting the NSTEMI occurring in UA patients. These observations support the value of calculating the TC/HDL ratio and not individual lipid parameter while predicting the risk of myocardial damage in cases of UA as stated in adult treatment panel-IV guidelines, quoted by P.G. Talwalkar et all [20]. So that early correction or close monitoring of TC and HDL could be of use to reduce the chances of future myocardial damage occurring in unstable angina patients.

Limitations of our study: We could not estimate the newer inflammatory markers (apolipoprotein-A neopterin, high sensitive C-reactive proteins) as the selection bias, less number of cases with unequal distribution of base line co-morbidities were also some of the limitations of our study.

Future scope: The larger study is required to be carried out in the future with inclusion of newer cardiac risk factors correlating with the severity of the acute coronary syndrome on UA patients of south India where there is higher prevalence of the ACS.

www.ijbamr.com P ISSN: 2250-284X , E ISSN : 2250-2858

Conclusion

From above findings it can be concluded that even though the lipid profile factors like TG, TC, TC/HDL cholesterol and LDL/HDL cholesterol ratios are risk factors for ischemic heart disease but TC/HDL ratio is the main culprit among lipid profile parameters to cause myocardial damage in unstable angina patients and converting them to NSTEMI.

Funding: The authors declare that the study was completely self funded.

References

1. KJ Raihanathul Misiriya, N.Sudhayakumar, S. Abdul Khadar, Raju George, VL Jayprakash, Joseph M. Pappachan. The clinical spectrum of Acute coronary syndrome: Experience from major center in Kerala. JAPI May 2009;57:378-83,.
2. Rajeev Gupta, Soneil Guptha, Krishna Kumar Sharma, Arvind Gupta and Prakash Deedwania. Regional variations in cardiovascular risk factors in India: India heart watch. World J Cardiol [internet] 2012 Jan24 [cited April 26]; 4(4):112-120. Available from: http://www.wjgnet.com/1949-8462/full/v4/i4/112
3. Jennifer N. Smith, Jenna M. Negrelli, Megha B. Manek, Emily M. Hawes and Anthony J. Viera. Diagnosis and management of acute coronary syndrome: an evidence-based update. JABFM March-April 2015;28:283-92,.
4. Fred S. Apple, Roberat L. Jesse, L. Cristin Newby, Alan H. B. Wu and Robert H. Christenson. National academy of clinical Biochemistry and IFCC committee for standardization of markers of cardiac damage laboratory medicine practice guidelines: Analytical issues for biochemical markers of acute coronary syndromes. Circulation 2007;115:352-5.
5. Fred S. Apple, Alireza Falahati, Pamela R. Paulsen, Elizabeth A. Miller and Scott W. Skarkey. Improved detection of minor ischemic myocardial injury with measurement of serum cardiac troponin-I. Clinical chemistry 1997;43:2047-51.
6. Jochen D. Muehlschlegel, Tjorvi E. Perry, Kuang-Yu Liu, Luigino Nascimben, Amanda A. Fox, Charles D. Collard et al. Troponin is superior to electrocardiogram and creatine kinase MB for predicting clinically significant myocardial injury after coronary artery bypass grafting. Europian Heart Journal 2009;30:1574-83.
7. Jaffe AS, Lindahl B, Katus HA. Sensitive tropinin I assay in patients with suspected acute coronary syndrome. J Am Med Assoc 2011;306:488–9.
8. National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III) Third report of the National Cholesterol Education Program (NCEP) expert panel on detection, evaluation, and treatment of high blood cholesterol in adults (Adult treatment panel III) final report. Circulation 2002;106:3143-421.
9. Y.H. Chan. Biostatistics 20. I: Linear Regression analysis. Singapore Med J 2004;45:55-61.
10. Rafaela Andrade Penalva, Markel D. Oliveira Huoya, Luis Claudio Lemos Correia, Gilson Soares Feitosa, Ana Marice Teixeira Ladeia. Lipid profile and severity of atherosclerotic disease in acute coronary syndrome. Arq Bras Cardiol 2008;90:24-9.
11. Amit Kumar Shrivastava, Harshvardhan Singh, Arun Raizada, Sanjeev Kumar Singh. Serial measurements

of lipid profile and inflammatory markers in patients with acute myocardial infarction. EXCLI Journal 2015;14:517-26.

12. P. K. Nigam, V. S. Narain and M. Hasan. Serum lipid profile in patients with acute myocardial infarction. Indian J of clinical biochemistry 2004;19:67-70.
13. Vishwanthan Mohan, Raj Deepa, Subramaniam Shanthi Rani, Gopal Premlatha. Prevalence of coronary artery disease and its relationship to lipids in selected population in south India, The Chennai urban population study. JACC2001;38:682-7.
14. Haseeb A. Khan, Abdullah S. Alhomida and Samia H. Sobki. Lipid profile of patients with acute myocardial infarction and its correlation with systemic inflammation. Biomarker insights 2013;8:1-7.
15. Jorgen Jeppesen, Hans Ole Hein, Paul Suadicani, Finn Gyntelberg. Low Triglycerides-High High-Density Cholesterol and risk of Ischemic heart disease, Arch Intern Med 2001;161:361-6.
16. R. Salehi , M. Rahbani-Nobar, N. Rashtchizadeh. Lipoprotein(A), Apo(A) phenotypes and dyslipidemia in acute coronary syndromes. Medical Journal of Islamic academy of sciences, Cardiology 2001;14:139-43.
17. Okon Ekwere Essien, Joseph Andy, Victor Ansa, Akaninyene Asuquo, Otu, Alphonsus Udoh. Coronary artery disease and the profile of and the profile of cardiovascular risk factors in south south Nigeria: A clinical and autopsy study, Cardiology and research and practice [internet], Vol. 2014 Jan2, cited 2014 March10. http://dx.doi.org/10.1155/2014/804751.
18. Vessa Manninen, Leena Tenkanen, Pekka Koskinen, Jussi K. Huttunen, Matti Mantari, Olli P. Heinonen et al Joint effects of serum triglycerides, LDL cholesterol, HDL cholesterol concentration risk on CHD: Risk in the Helsinke heart study, Circulation 1992;85:37-45.
19. Mouaz H. Al.-Mallah, Hazem Hatahet, Joao L. Cavalcante Sanjaya Khanal, Low admission LDL cholesterol is associated with increased three year all-cause mortality in patients with non-ST-elevation myocardial infarction, Cardiology 2009;16:227-33.
20. P. G. Talwalkar, C. G. Sreenivas, Ashish Gulati, Hemang Baxi. Journey in guidelines for lipid management: From adult treatment panel (ATP)-I to ATP-III and what to expect in ATP-IV. Indian Journal of Endocrinology and Metabolism 2013;17:628-35.