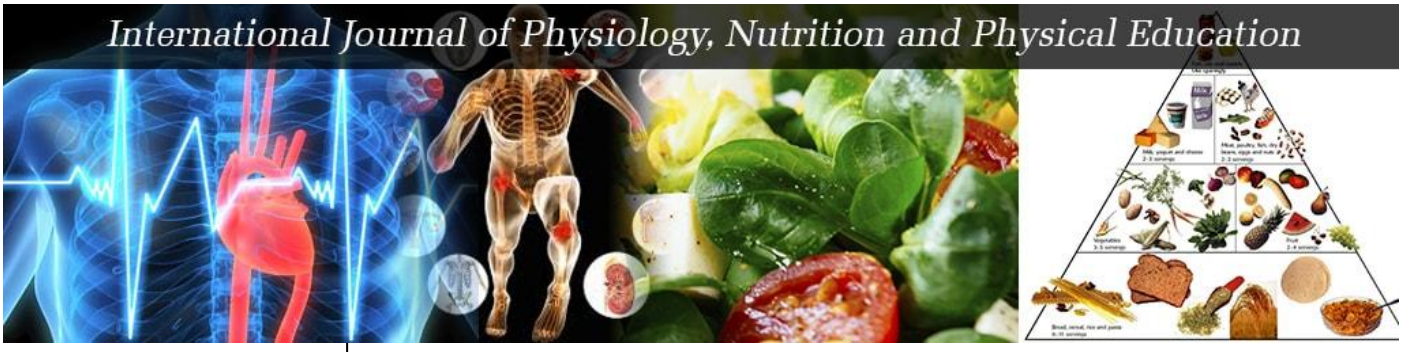

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Some important metabolic markers in blood of Trained Endomorph, Mesomorph and Ectomorph male athletes

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Abstract
Physical training improves muscle mass and different metabolic activities. Metabolic need is dependent on one's body type and dimension even in resting condition. The normal values of different metabolic markers might not match with that of trained athletes as they were not estimated according to body type. So, the purpose behind this study was to focus on the relationship between somatotypes and two important metabolic markers which represent the renal and cardiac functions of trained athletes. This cross-sectional study was carried out in total 34 trained male athletes (mean age 12.7 years). They were kinantropometrically categorized into three groups-dominant endomorph, mesomorph and ectomorph. In resting condition, blood samples were withdrawn for the analysis of creatinine and creatine kinase-MB. One-way ANOVA was performed ($p<0.05$) to analyze the results.
Significant difference in Serum Creatinine ($F=4.14$) among the three groups had been found. There was insignificant difference in Creatine Kinase-MB among the dominant endomorph, mesomorph and ectomorph. It can be concluded that the reference values for normal individual were not alike in trained athletes and it will be more accurate if we consider these important metabolic biomarkers according to their body constituent. This may help to prevent sports injury and help in proper quantification of training programme by reflecting more accurate internal metabolic status of the trained athletes.

Keywords: Somatotypes, Metabolic markers, Muscle activity

Introduction
Athletic performance is influenced by the structure and shape of the athlete. Athletes with different somatotypes exhibit outstanding performance during exercise and physical training [1-5]. Proper physical exercise and training mobilize the development of new muscle mass which ultimately change the shape and structure of the athlete. External changes can be easily identified by classifying the athlete according to their somatotype. These external changes can only be possible by modifying the internal metabolic activities of athlete. So, athletes not only distinguish themselves from sedentary person by their external structural component, but also by their metabolic activities. Therefore, it is quite expected that so called normal or reference values of different important clinical biomarkers of blood will not be identical in athlete and sedentary person. It is always more accurate and safe if the reference values can be measured according to somatotype. Athletes are more susceptible to different organ failure as they perform heavy exercise during their training session without knowing their metabolic status.
Heath and Carter [6] introduced the method for somatotype which is almost identical to measurements of anthropometrically determined body composition. After that many studies have been carried out to relate body composition variables and somatotype [7, 5, 8, 9]. All these studies suggested that endomorph is more fatty and heavier than mesomorph and ectomorph, mesomorph is comparatively more muscular than endomorphs and ectomorph, and ectomorph is the most lean and thin among the three groups. Thus, these evidences suggest that there is a definite relationship between structural components and somatotype. From all these evidences it is inferred that somatotype influences performance in athletes. Moreover, physiological functions of different tissues and organs regulate the performances of athletes by influencing the metabolic activities according to their energy demands. So, it is implied that somatotype and metabolic activities are highly related to each other, though a few studies have been carried out.

There is evidence of relationship between total serum cholesterol and somatotype [10, 5].

Allard and Goulet [11] reported that body build and increased serum cholesterol concentrations are interrelated. There are very few scientific reporting in metabolic responses to exercise in association with somatotype. Schreiber [12] reported that ectomorph, as compared to endomorph and mesomorph, metabolically had more glycoltic dependence during anaerobic function test.

This study examined the association between somatotype and metabolic activities of trained male young players. We have tried to find out the relationship between few important metabolic biomarkers in blood as determinant of physiological responses according to their structural component of body. It would be helpful to know their current metabolic status according to their metabolic demand as well as adjust the training programme according to their need. We have selected creatine kinase, creatinine and calcium as biomarkers because all these blood markers are highly related to muscular activities. Moreover, this study would help us to evaluate any deviation from existing reference values which are not determined according to structural aspect as well as metabolic demands of the trained male players.

2. Materials and methods

2.1. Participants

Thirty four trained male players (mean age 12.7 years) participated in this cross sectional study. Individual NFHS (National Standard of Living Index) and SCAT (Sports Competition Anxiety Test) are carried out in each participant. Prior permission was obtained from Institutional Ethical Committee (SCHEC/02/2015). All participants are free from cardiovascular, metabolic or orthopedic disorders and did not use any medication that might affect muscle function. They are trained for minimum 2 years and maximum 6 years and participated in district or regional competition.

2.2. Anthropometry All measurements are taken on the same day to avoid technical error by the Level 1 Anthropometrics accredited by International society for advancement of Kinanthropometry (ISAK), according to the procedure stated on the ISAK manual [13]. Skin fold thickness (biceps, triceps, sub scapular, and supraspinale) for determining the fat% and total fat content are measured with a skin fold caliper which requires a constant closing compression of 10 $g.mm^{-2}$ throughout the range of measurements. The subject is asked to assume a relaxed standing position with the right arm hanging by the side and the hand in mid-prone position. Biceps skin fold is measured from the point on the anterior surface of the arm at the level of mid-acromiale-radiale land mark, in the middle of the muscle belly. Triceps skin fold is measured from the point on the posterior surface of the arm, in the mid-line, at the level of marked mid-acromiale-radial landmark. Sub scapular skin fold measurement is taken with the fold running obliquely downwards from the subscapulare landmark at 45^0 angles. Supraspinale skin fold measurement is taken from the intersecting point of two lines, the line from the marked iliospinale and iliocristale border. Anthropometric tape which is non-extensible, flexible, not wider than 7 mm and have a stub (blank area) of at least 4 cm before zero line is used to measure girth (arm and calf) and circumferences (chest, waist, hip, mid-thigh, upper thigh). The arm girth is taken from the circumference of the arm at the level of mid-acromiale-radiale site, perpendicular to the long axis of arm. Similarly calf girth is taken from the circumference of leg at the level of medial calf skin fold site, perpendicular to its long axis. The measurement of the circumference of the thorax at the level of the mesosternale site, perpendicular to the long axis of the thorax is the chest circumference. Waist circumference is the circumference of the abdomen at this narrowest point between the lower coastal (10^{th} rib) border and the top of the iliac crest, perpendicular to the long axis of the trunk. The circumference measured from the buttocks at the level of their greatest posterior protuberance, perpendicular to the long axis of the trunk. The circumference of the mid-thigh is measured at the level of the mid-trochanterion-tibialelaterale site, perpendicular to its long axis. Upper thigh circumference is measured from the site of the thigh, 1 cm distal to the gluteal fold site, perpendicular to its long axis.

Sliding caliper has a branch length of at least 10 cm, an application face width of 1.5 cm and accurate within 0.05 cm is used for measuring breadth (biepicondylar humerus and femur). Biepicondylar humerus and femur are measured from the distance between the most lateral aspect of the lateral humeral and femoral epicondyle and the most medial humeral and femoral epicondyle respectively.

Heath-Carter's [6] equations are used for calculating somatotype -

Endomorph = $-0.7182 + 0.1454 \times \sum SF - 0.00068 \times \sum Sf^2 + 0.0000014 \times \sum SF^3$ where $\sum SF$ = (triceps + biceps + sub scapular + supraspinale skin fold) x (170.18/height in cm.).

Mesomorph = 0.858 x humerus breadth + 0.601 x femur breadth + 0.188 x corrected arm girth + 0.161 x corrected calf girth – height x 0.131 + 4.5.

Three different equations are used to calculate ectomorph according to the height-weight ratio (HWR):

If HWR is greater than or equal to 40.75 then, Ectomorph = 0.732 x HWR -28.58.

If HWR is less than 40.75 and greater than 38.25 then, Ectomorph = 0.463 x HWR – 17.63.

If HWR is equal to or less than 38.25 then, Ectomorph = 0.1.

Body Fat % and Total Fat Content (kg) From the four different skin fold thickness, the Body Density (kg/mm^3) is measured using the Durnin &Woomersley's (1974) generalized equation for body density and the Total Body Fat Percentage (%) is calculated using the equation derived by Brozek *et al*., (1963) and Siri (1956).The Total Fat Mass (kg) is evaluated by using the values of Total Body Fat Percentage (%) and Body Mass i.e. Weight (kg). Equations for calculating Body Density (kg/mm^3), Total Fat percentage (%), Total Fat Mass (kg) are given below. Body Density -

For Male =1.1620-0.0630log (Biceps + Triceps + Sub scapular + Supraspinale) for 17-19 years

For Female = 1.1549-0.0678log (Biceps + Triceps + Sub scapular + Supraspinale) for 16-19 year

Total Body Fat Percentage (%) = ((4.45/Body density)-4.142) x 100

Total Body Fat Mass (kg) = (% of Body Fat/100) x Bodyweight in kg.

2.3. Clinical measurement

Quantitative estimation of biochemical parameters like creatinine (kinetic method) [14], calcium (Arsenazo-III method) [15], creatine kinase (modified IFCC method) [16] is done by using kits manufactured by Avecon Healthcare Private Limited, Transasia Biomedicals Limited respectively.

2.3.1. Serum creatinine - Kinetic method

Serum Creatinine is determined by Jaffe's method. The

principle states that in alkaline medium picric acid reacts with creatinine and an orange coloured complex is formed with the alkaline picrate. During the fixed time, the intensity of colour formed is directly proportional to the amount of creatinine present in the sample.

2.3.2. Calcium-Arsenazo iii method

The carrier buffer is aspirated into the syringe. After that Arsenazo III and standard/ sample solution was introduced into the holding coil. Arsenazo III reagent was aspirated again. Reversal of the flow direction resulted in displacement of the reaction mixture to the flow-through cuvette. A blue-purple complex formed. Through spectrometric detector, chemical reaction between Arsenazo III and calcium is quantified. A solution of 1 M HCl was injected and washed with carrier buffer to avoid contamination from other substances.

2.3.3. Creatine kinase- Modified IFCC method

Substrate Start Assay

In a dry test tube labelled as T, 0.8 ml enzyme reagent and 0.02 ml sample is added. It is then incubated at 37 ºC for 1 min and 0.2 ml of starter reagent was added. It is mixed well and the initial absorbance is read at 340 nm and the absorbance was read after every 1, 2 and 3 min.

Sample Start Assay

In a dry test tube labelled as T, 1 ml of working reagent is added. It is then incubated at 37 ºC for 1 min and 0.02 ml of starter reagent is added. It is mixed well and the initial absorbance is read at 340 nm and the absorbance was read after every 1, 2 and 3 min. The mean absorbance change per minute is calculated.

2.4. Statistical analysis

Mean values and Standard deviations of each mentioned parameters of three groups (endomorph, mesomorph and ectomorph) were calculated. One way ANOVA (analysis of variance) is done to compare each of the parameters among the three groups. Probability of error due to random sampling is rejected at the level of $p<0.05$.

3. Results

The mean values, standard deviations, ranges and F values with level of significance of age (years), height (cm), weight (kg),total body fat percentage (%), resting heart rate (beats/minutes), creatinine (µmol/l), calcium (µmol/l) and creatine kinase (U/L) of trained male endomorph, mesomorph and ectomorph are shown in Table1.

Table 1: The mean values, standard deviation, ranges, F values and level of significance of different measured variables in dominant endomorph, mesomorph and ectomorph.

Variables	Kinanthropometric Group	Endomorph (N = 14)	Mesomorph (N = 9)	Ectomorph (N = 11)
Age (years)	Mean ± SD	12.21±2.46	13±1.41	12.9±2.07
	F Values	0.016		
	Significance of F	NS		
Height (cm)	Mean ± SD	148.37±12.56	157.7±11.21	159.57±14.21
	F Values	0.0018		
	Significance of F	NS		
Weight (kg)	Mean ± SD	45.93±11.15	51.56±9.89	42.45±8.21
	F Values	0.00079		
	Significance of F	NS		
Body Fat Percentage	Mean ± SD	20.25±2.82	13.59±3.10	10.22±3.5
	F Values	0.13		
	Significance of F	NS		
Heart Rate(beats/min)	Mean ± SD	73±1.19	67.33±14.76	78.09±19.73
	F Values	45.47		
	Significance of F	P<0.05		
Creatinine (µmol/l)	Mean ± SD	81.45±13.51	97.24 ±14.66	94.83 ±15.86
	F Values	4.11		
	Significance of F	**P<0.05**		
Creatine Kinase(U/l)	Mean ± SD	92.57±32.52	105.67±44.99	105.18±33.65
	F Values	0.11		
	Significance of F	NS		

Insignificant differences are found in age (years), height (cm), weight (kg), body fat percentage (%) and creatine kinase (U/L) among the three groups. Significant differences are observed in resting heart rate (beats/minute), creatinine (µmol/L) and calcium (µmol/L) among the three groups. Resting heart rate is lowest (67.33±14.76) in dominant mesomorph and highest (78.09± 19.73) in dominant ectomorph. Creatinine level is highest (97.24 ±14.66) in dominant mesomorph and lowest (81.45 ±13.51) in dominant endomorphs. Blood calcium level is highest in dominant endomorph (2.77 U/L) and lowest in dominant ectomorph (2.59 U/L).

4. Discussion

The mean values of age (years), stature (cm), body weight (kg) and body fat percentage (%) are almost same in dominant endomorphs, mesomorph and ectomorph as all the values are statistically insignificant. So it can be claimed that there are no variations among the three groups in age, sex, ethnicity, stature, bodyweight and fat percentage. Though body fat percentage values are found statistically insignificant among the three groups, but it is highest in dominant endomorphs (20.25 ± 2.82) followed by mesomorph (13.59 ±3.10) and ectomorph (10.22 ±3.5) which suggest determination of somatotype is correct and authentic. Moreover, body weight is maximum in mesomorph (51.56 ±9.89) indicating muscular development after training is more prominent in them than the endomorph and ectomorph. The significant difference of mean values of resting heart rate among the three groups may be due to many reasons as heart rate is influenced by different factors. But lowest heart rate in dominant mesomorph (67.33 ±14.76) indicates advantage over other two groups in physical

ability.

Each day, 1-2% of muscle creatine is converted to creatinine [17] and the reference range is 70-120 µmol/l (0.8-1.4mg/dl). We have found highest creatinine in mesomorph (97.24±14.66) which ensures that metabolic activity can be modified by somatotype in trained player. The training method which massively improve the muscle mass of the player and they become mesomorphic by performing heavy exercise, may lead to excessive production of creatinine. Enhanced creatinine may cause renal damage to the player. Though mesomorphic feature is very essential for better performance but it should not be forgotten that excess metabolic activities cause renal dysfunction. So it is very important to modulate the physical training and exercise of players by observing their morphometric changes.

Creatine kinase-MB (CK-MB) is an important biomarker for cardiac muscle damage. The reference range of Creatine kinase in serum is 20-215 U/L. The level of creatine kinase in mesomorph and ectomorph is almost same and lowest in endomorph though the values are statistically insignificant. Moreover, the most reliable test in the diagnosis of rhabdomyolysis is the level of creatine kinase (CK) in the blood. This enzyme is released by damaged muscle, and levels above 5 times the upper limit of normal indicate rhabdomyolysis. Insignificant difference and range of creatine kinase among the three groups indicate no muscle damage.

The role of calcium in cardiac as well as skeletal muscle activities is very crucial. Total calcium present in blood reflects the availability of calcium for muscular activities. In our study the total calcium level of blood in dominant endomorph is highest (2.77±0.21U/L) among the three groups and it is higher than the reference value (2.18-2.58U/L or 8.7-10.3 mg/dL) [18]. As significant differences are observed in the three groups, so it can be claimed that morphometric analysis may help to evaluate the proper status of cardiac and skeletal muscle in players. Once we estimate the proper requirement and demand according to their somatotype, quantification of training and sports injuries related to cardiac and skeletal muscle activities may be properly regulated.

This study includes three important biomarkers creatinine, Creatine kinase and calcium because these are metabolically related to the activities of cardiac and skeletal muscle directly to maintain physiological functions of the body. All three biomarkers have potential clinical importance to diagnose the current status of different major organs and systems of the body. However, more clinical research is required based on somatotype of the trained players to prevent sports hazards and to ensure safe physical training programme.

5. Conclusion

It has been observed in this study that values of metabolic markers differ among normal and trained athletes. If the metabolic markers are studied with reference to body constitution, it will help to prevent sports injury and improve training programmers.

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