Comparative effect of treadmill exercise and yoga on Superoxide dismutase level in normal young adults

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Abstract
Fifty male subjects of age group 18-20 years and were divided into two groups, treadmill exercise (Group A) and yoga (Group B) consisting of 25 each in Department of Physiology at Exercise Physiology Lab (JNMC, DMIMS, Sawangi Meghe, Wardha). On regular performance of exercise, subjects were assessed at the start of and after 3 months of the training schedule (45 min/ 5 days/ week). Blood samples were collected to assess the SOD levels. Results were compared using student paired "t" test and unpaired "t" test & age-wise distribution by Chi-Square test.

Results showed that the pre-exercise levels of SOD in Group A (11.56±1.0) and Group B (11.56±1.0) and the post-exercise levels in Group A (12.01±1.09) and Group B (13.31±0.9). Thus the above study shows that yoga has more beneficial effect than the treadmill exercise which leads to decrease in the free radical production.

Keywords: Treadmill exercise and yoga, oxidative stress, superoxide dismutase (SOD)

1. Introduction
In recent years there has been a growing appreciation of the issues of quality of life and stresses involved medical training as this may affect their learning and academic performance [1]. Medical courses in India are very demanding for the students thus making career in medical education very stressful [2]. Fear of failure, vast amount of content that has to be mastered, inability to cope with the high expectations of parents and peers are found to be the most commonly observed sources of stress. A study conducted on 238 students undergoing Medical course showed that the stress in medical students is common and is process oriented. This indicates the importance of such studies in the Indian scenario [3]. Stress is a term that refers to the sum of physical mental and emotional strains or tensions on a person. The term 'stress' was first employed in the 1930's by the endocrinologist Hans Selye [4]. Stress also indicates the consequence of the failure of an organism-human or animal to respond appropriately to emotional or physical threats [5]. Production of reactive oxygen species is particularly destructive aspect of stress [6]. The science of free radicals in living organisms entered a second era after McCord and Fridovich discovered the enzyme superoxide dismutase an antioxidant having first line defense against free radicals in body [7]. Cells continuously produce free radicals and ROS as part of metabolic processes. These free radicals are neutralized by an elaborate antioxidant defense system consisting of enzymes such as catalase, superoxide dismutase, glutathione peroxidase, and numerous non-enzymatic antioxidants, including vitamins A, E and C, glutathione, ubiquinone, and flavonoids. While regular exercise training is associated with numerous health benefits, it can be viewed as an intense physical stressor leading to increased oxidative cellular damage, likely due to enhanced production of reactive oxygen species (ROS) [8]. An array of mind-body therapies are being used presently as adjuncts to conventional treatment for a number of common clinical conditions such as asthma, post-traumatic stress disorder, addiction treatment, stress management, anxiety, Obsessive victims Compulsive Disorder (OCD) and various cardiovascular dysfunctions. Yoga, an ancient system of life style helps to achieve an overall harmonious state of well-being and is one such mind body medicine approach which is highly appreciated, researched and recommended as a practice towards better health, adjuvant in various disease conditions and for life fulfillment [9]. As a trend, youth of today have opted for strenuous physical exercises.
The well-equipped gym schools of today are very lucrative and attractive. Yogic exercises are more relaxing and cost effective. To the best of our knowledge there are only few studies to see the comparative effect of anaerobic exercise with relaxation techniques i.e yoga. So this study aims to compare the effects of relaxation technique and anaerobic exercises on medical students, who are one of the groups most commonly encountered by stress.

2. Materials and methods

2.1 Type of study: Comparative interventional study

2.2 Study area: The selection and screening of subjects were conducted at Exercise Physiology Laboratory, Department of Physiology, Jawaharlal Nehru Medical College, Sawangi (Meghe), Wardha, Maharashtra, India. Treadmill tests were conducted at the Electrophysiology Laboratory at Acharya Vinoba Bhave Hospital, Sawangi (Meghe), Wardha, Maharashtra, India.

2.3 Selection of subjects: After obtaining approvals from the Institutional Ethics Committee and the institutional authorities, students aged 18 years and above, of either sex, who had enrolled for the first-year of the Bachelor of Medicine, Bachelor of Surgery (MBBS) course at Jawaharlal Nehru Medical College were explained about the study. Those with history of any type of medical illness and those below 18 years of age were excluded. From among those students (all males between 18 and 20 years of age) who gave written informed consent, subjects were randomly allocated by blind-chit (lottery) method into two groups A and B, each group comprising 25 subjects.

2.4.1 Intervention for Group A: After giving information, all subjects in Group A underwent treadmill exercise as per Bruce Protocol [11]. Bruce Protocol is a maximal exercise test, where, the subject works to complete exhaustion as the treadmill speed and incline is increased every three minutes. The total duration of workout was 45min/day for 5 days/week for 12 weeks (initial warm up for 5min followed by intervention of 35min and ending the procedure with 5min relaxation). 30 settings of 45min were given each on alternate day for 12 weeks. This was followed by once a week interview during which, the method followed by the patients was checked and any queries related to the methodology were answered. During the test, heart rate and blood pressure were recorded. During this time, subjects were instructed to sit or rest, refrain from eating and were only allowed to drink up to 1L of water. A person was made to walk on the treadmill (RMS, Ambala) thus implying the Standard Bruce Protocol for the subjects, who are active and devoid of any diseases. Subjects were instructed to press the red knob on the treadmill or raise hand to stop the test, in case of any discomfort. All tests were done at 1.74mph walking with an increment of grade by 2% at every 3minutes interval. The average grades achieved by all the students are 16-18%. Electrocardiogram was continuously monitored and the treadmill test was terminated when the subject reached 75-80% of their estimated heart rate reserve.

[10] Subjects performed the exercise for 30min/day for 5days a week for 3 months, after which their blood samples from capillaries were collected, mostly in the morning (8am to 11am). All subjects remained in the electrophysiology lab during their post-exercise sample collection for 1 hour to keep control on their food and water intake.

2.4.2 Intervention for Group B: After providing information, subjects in Group B underwent yogic exercise program comprising of pranayam (breathing exercises), asana (physical postures) and rajyoga meditation (mental relaxation exercise) lasting 45 min/day for a total duration of 12 weeks. They were made to relax for 5min and then they were allowed to practice asanas for 15min. Asana [12] (Sanskrit word meaning “sitting down”) is a body position, typically associated with the practice of yoga, originally identified as a mastery of sitting still. Three types of asanas were included Suryanamaskar [13] trikonasana, tadasana [14]. The asanas were followed by pranayam (anulom-vilom and kapalbhati) [15]. After rest for 5min, rajyoga meditation was practiced for 15min in a quiet, dimly lit room. A total of 7 orientation classes each of 45 minutes were delivered by the Raj yoga specialist, of which last 20 minutes are devoted to a guided audio clip. All of them were required to note about the experience of the meditation practice in their diary [16]. Review was taken in the department after every 15 days. The yogic exercise ended with 5min relaxation. Subjects were closely monitored and exercise was put on hold in case of slight discomfort. Subjects were called every week and compliance was noted.

2.5 Blood sample collection: Blood samples (2ml of capillary blood) were collected aseptically in plain bulb in a quiet room after the subject had 10 minutes of adjustment in the supine position. The samples were collected before the commencement of intervention and after 12 weeks of intervention. Any blood samples displaying evidences of hemolysis were discarded and not stored for assay.

2.6. Parameters: 2.6.1 Body Mass Index (BMI): The body mass index (BMI) was calculated by dividing the weight (in kilograms) by the square of height (in meters) [17]. Height was measured in centimeters using a measuring tape (Medscope Ltd, Marsden H-630, Cirencester, UK) fixed on wall with the subject standing barefoot. Weight was measured in kilograms on a digital weighing scale (Omron HN-286 Digital weight scale, Haryana), with subject in minimum clothing.

2.6.2 Pre & Post WHO-QOL Assessment: Subjects of both the groups were given WHO-QOL to assess the quality of life and health. The questions were read out to the subjects, along with the response options and were told to tick the desired option that appears most appropriate. The WHOQOL-BREF (Field Trial Version) [10], containing 26 questions, was used to calculate four domain scores. The mean score of items within each domain was used to calculate the domain score. Mean scores were then multiplied by 4 in order to make domain scores comparable with the scores used in the WHO-QOL-100. The first transformation method converts scores ranging from 4-20, compared with the WHO-QOL-100. The second transformation method converts domain scores to a 0-100 scale. Where more than 20% of data is missing from assessment, the assessment was discarded.
2.6.3 Superoxide dismutase: Two new test tubes named as blank and test were used. 2.7ml of Tris buffer was added in blank tube. 2.699ml of tris buffer was added in test tube and 1µl of hemolysate added to each of it. At the time of reading 300µl of pyrogallol was added to both blank and test tubes and finally both the tubes consisted of 3ml solution in it. Stopwatch was started and readings were noted in 1min. Mixture was vortexed for 20sec. After that the mixture was put in the cuvette, cuvette was placed in spectrophotometer. By pressing enter after 60sec and 90sec respectively, 10 readings were noted which were being displayed on the screen at the wavelength of 420nm [18, 19].

2.6.4 Statistical Analysis: Results were analyzed by SPSS 22.0 Windows version (IBM Corporation, Armonk, NY, USA). All the data were analyzed group-wise by descriptive statistics using mean and standard deviation. For age-wise distribution of both the groups, Chi-square test was used. The intra-group pre- and post- intervention data were analyzed using Student’s paired t-test. Inter-group data were analyzed using Student’s unpaired t-test. The statistical significance was considered at probability value less than 0.05.

3. Results
50 male participated in this study. The entire group had a mean age of 20±1.02 years, mean BMI for Group A was 1.63±0.1 kg/m² and Group B was 0.92±0.1 kg/m². The total duration of the exercise protocol was 45min/day for 5 days a week for 3 months.

WHO-QOL responses in Table B shows that in Group A there was more progressive data in social relationship (10.20±3.9) than those in physical (8.56±2.3), psychological (9.48±3.3) and environmental domains(8.52±3.6). Whereas, Group B subjects showed significant increase in physical (14.52±2.4), followed by psychological (13.88±2.4), relationship (12.72±4.2) and environmental (10.96±5.4) domains. Superoxide Dismutase (SOD) response showed in Table C, that there was an increase in SOD in Group A (12.01±1.09) level than its pre (11.56±1.0). Whereas, in Group B post (13.31±0.9) intervention than its pre (11.56±1.0) intervention levels. When compared Group B (13.31±0.9) showed considerable increase in SOD as compared to the post intervention values (12.01±1.09) of Group A.

4. Discussion
Chitnis P et al. (2013) studied the effect of stress on obesity on 174 students of mean age group 17-21 years. Perceived Stress Scale score is higher in obese and overweight as compared to normal and underweight subjects. Stress is one of the contributing factors for obesity. Effective counseling for management of stress helps to reduce obesity and its related complications [20]. Findings of table A, are parallel to Patil CH et al. (2012), Shinde N et al. (2013), whereas findings of Telles S et al. 2014 that both yoga and walking groups showed a significant decrease in BMI, waist circumference, hip circumference, lean mass, body water and total cholesterol. Exercise has a negative linear relationship with body mass index. [21, 22, 23]

The findings of table B, are in line with Manocha et al. (2012) and Bankar M et al. (2013), thus the above studies conclude that relaxing exercise have physical as well as mental effect in positive direction which reduces the stress on once social and environmental surroundings. [24, 25] Thus, long-term practitioners of yoga or meditation experience better functional health, especially mental health, compared to the general population.

Thus according to table C, regular and moderate intensity exercise has less stressful effect on once body rather than a strenuous and intense work out. This oxidative stress is closely linked to the progress of pathogenesis and it leads to impairment of homeostasis in human body. Excessive oxidative stress is related to the initiation and development of various disorders including neurodegenerative diseases, chronic fatigue syndrome, cardiac dysfunction, metabolic syndromes and cancer occurrences [26, 27]. To defend from oxidative stress most of the mammals well equipped of the antioxidant systems in enzymatic and non-enzymatic components. The antioxidant systems including superoxide radical (SOD) and catalase efficiently work to prevent from these harmful oxidative stressors [28, 29]. Pranayama (breathing exercise) is documented to produce an intense calming effect on the mind and is responsible to relieve psychological stress [30]. Which might have enhanced led to the chain of reactions relieving oxidative stress. By reducing the activation and reactivity of the sympatno-adrenal system and the hypothalamic pituitary adrenal (HPA) axis and promoting feelings of well-being, yoga may alleviate the effects of stress and foster multiple positive downstream effects on neuroendocrine status, metabolic function and related inflammatory responses. Also, directly stimulating the vagus nerve, yoga may enhance parasympathetic output and thereby shift the autonomic nervous system balance from primarily sympathetic to parasympathetic leading to positive changes in cardiac-vagal function, in mood and energy state, and in related neuroendocrine, metabolic, and inflammatory responses [31].

The purpose of current study was designed to acquire scientific and clinical evidences for explaining properties of regular yoga training in healthy university populations. Herein, we confirmed results by looking for antioxidant components in the serum level of participants.

Table 1: Mean and Standard Deviation of Body Mass Index (kg/m²) in Groups A and B

<table>
<thead>
<tr>
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<th>Group A (n=25)</th>
<th>Group B (n=25)</th>
<th>p value</th>
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<tbody>
<tr>
<td>Pre</td>
<td>28.33±1.9</td>
<td>28.33±1.9</td>
<td>0.02 *</td>
</tr>
<tr>
<td>Post</td>
<td>26.70±2.0</td>
<td>27.41±2.0</td>
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Table 3: Comparison of blood levels of superoxide dismutase (U/mHb) in Groups A and B

<table>
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<tr>
<th></th>
<th>Group A (n=25)</th>
<th>Group B (n=25)</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pre</td>
<td>11.56±1.0</td>
<td>13.31±0.9</td>
<td>0.002 *</td>
</tr>
<tr>
<td>Post</td>
<td>12.01±1.09</td>
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*statistically significant

5. Conclusion
Our study thus, conclude that yoga has more beneficial effect than the strenuous treadmill exercise in combating oxidative stress. We recommend that similar studies to be taken up on large population and with increased intervention period.
6. Limitations
Due to the availability of subjects for limited time the study
was not able to continue for longer duration.
Study was focused on the male subjects as female subjects
were not willing to give their informed consent.

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