IMPACT OF EXERCISE ON ENDOTHELIUM-DEPENDENT VASODILATOR SERUM NITRIC OXIDE

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Abstracts: The vascular endothelium produces a large number of vasodilator and vasoconstrictor compounds which regulate vascular tone. Nitric Oxide (NO) is recognised to be the most important and best characterised mediator; its intrinsic vasodilator function is commonly used as a surrogate index of endothelial function. Various studies have demonstrated that exercise training improves endothelium-dependent vasodilator function. Yoga is a group of physical, mental, and spiritual practices or disciplines which originated in ancient India. The poses and stretches are thought to have physical and mental health benefits. Optimum exercise training schedules in the form of Yogasanas & pranayam may result in possible sequential changes in the endovascular milieu. Yoga exercise mediated improvement in endothelial function may provides plausible cardio protective benefits of exercise training. We evaluated the impact of exercise in the form of yoga v/s sedentary life on serum Nitric Oxide (NO). This prospective study consisted of 70 volunteer subjects in the age range of 18 to 73 years, mean age 33 yrs, from a small locality in the north of Bangalore. Serum Nitric Oxide levels was measured using Cadmium reduction method. The subject's history of regular exercise, type & duration of exercise was evaluated; those who did not perform regular exercise of at least 2 days a week were classified as sedentary. 40 subjects who performed regular exercise in the form of Yoga and 30 subjects who did not perform regular exercises were classified as sedentary. Sedentary group was significantly younger with a mean age of 18.5 yrs compared to the exercise group whose mean age was 44yrs. 22% of the subjects in the exercise group had some coexisting disease with 70% requiring medication, compared to no coexisting disease in sedentary group who were much younger. BMI & Waist Hip ratio was significantly higher in exercise group. The exercise group had been performing Yoga for a mean duration of 3.64±5.40 years. Comparison of subject who practiced yoga for and those who were sedentary revealed a stastically significant higher serum Nitric Oxide. Serum Nitric Oxide (NO µmol/l) was 77.26±37.71 in those who performed regular yoga exercise compared to 23.57±5.83 in Sedentary subjects <0.001**. It also showed a significant correlation with age. Hence our observation in this study was even though Subjects in the exercise group was were two Decades older with higher BMI and FBS and Co-morbidities the serum Nitric Oxide levels were comparatively higher compared to younger sedentary subjects. The presence of significantly higher levels of Nitric Oxide NO can be attributed to the continued regular exercise in the form of Yoga practices.

Key Words: Nitric oxide, NO, exercise, yoga, vascular endothelial factor.

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Introduction:

In the last two decades, it has been well recognised that a normal endothelial function is integral to vascular health. The vascular endothelium produces a large number of vasodilator and vasoconstrictor compounds which regulate vascular tone. It is recognised that the vasodilator, nitric oxide (NO) also has additional anti-atherogenic properties. Nitric Oxide (NO) is recognised to be the most important and best characterised mediator, its intrinsic vasodilator function is commonly used as a surrogate index of endothelial function (14,17).

Many age related conditions, including those commonly associated as risk factors for atherosclerosis such as hypertension, hypercholesterolemia, smoking, diabetes mellitus and heart failure are associated with diminished release of nitric oxide into the arterial wall either because of impaired synthesis or excessive oxidative degradation, which, in turn, correlates with increased cardiovascular mortality. The decreased production of NO in these

pathophysiological states results in serious impairment of endothelial equilibrium.

It is well recognised clinically that improved endothelial function tend to be associated in response to interventions. Numerous therapies have been investigated to assess the possibility of reversing endothelial dysfunction by enhancing the release of nitric oxide from the endothelium Furthermore (11,16).

It has now been recognised that stress on endothelial cells is a potent stimulus for NO production. Though the role of endothelium-derived NO in acute exercise has not been fully resolved, exercise training involving repetitive bouts of exercise over weeks or months up-regulates endothelial NO bioactivity. Increase in blood flow, resulting in change in hemodynamic which occur during acute exercise may, provide a stimulus for both acute and chronic changes in vascular function (15,18).

Various studies have demonstrated that exercise training improves endothelium-dependent vasodilator function, not only as a localised phenomenon in the active muscle group, but also as a systemic response when a relatively large mass of muscle is activated regularly during an exercise training programme. Individuals with initially impaired endothelial function at baseline appear to be more responsive to exercise training than healthy individuals. While improvement is reflected in increased NO bioactivity, the detail of mechanisms, the relative importance of up-regulation of mediators and antioxidant effects, is unclear (2,9,12,13).

Yoga is a group of physical, mental, and spiritual practices or disciplines which originated in ancient India. Yogaasana gained attention of the west the mid-19th century along with other topics of Indian philosophy. Asana`s collectively constitute the physical aspect of worship in ancient Yoga and various stretches and meditative poses of modern Yoga. The poses and stretches are thought to have physical and mental health benefits

Optimum exercise training schedules in the form of Yogasanas & pranayam will result in possible sequential changes in the endovascular milieu. Yoga exercise mediated improvement in endothelial function may provides plausible cardio protective benefits of exercise training.

Objective of study was To evaluate the impact of exercise in the form of yoga v/s sedentary life on serum Nitric Oxide (NO)

Material and Methods:

The study was approved by Scientific Review and Intuitional Ethics Committee. With informed consent as per ICH GCP Guidelines, 70 normal volunteer subjects from a small locality in the north of Bangalore were enrolled for the study. All the subjects were clinically evaluated, medical history recorded and their blood Serum collected and stored for further evaluation. Serum Nitric Oxide levels was measured using Cadmium reduction method. The subject's history of regular exercise, type & duration of exercise was evaluated; those who did not perform regular exercise of at least 2 days a week were classified as sedentary. The Data was analysed using SPSS .

Result:

This prospective study consisted of 70 volunteer subjects in the age range of 18 to 73 years, mean age 33 yrs, Males 49: Female 31. In the study subjects there were 40 who performed regular exercise in the form of Yoga and 30 subjects who did not perform regular exercises and were classified as sedentary.

Subjects in the Sedentary group were significantly in the younger age mean age 18.5 yrs compared to the exercise group whose mean age was 44yrs. M:F ratio was balanced between the two group. Of the exercise group had a family history of 1st degree relative with T2DM and Hypertension in 37 % & 32% respectively. A significant number (87%) of Sedentary subjects consumed non vegetarian diet regularly (Table 1). 22% of the subjects in the exercise group had some coexisting disease with 70% requiring medication, compared to no coexisting disease in sedentary group who were much younger (Table 2). BMI & Waist Hip ratio was significantly higher in exercise group.

It was observed that in the exercise group had been performing Yoga for a mean duration of 3.64±5.40 years. 70% of these subjects were less than 50 years of age. 45% of the subjects were Vegetarians. 60% of the subjects were practicing Yoga for more than 1year.

Comparison of subject who practiced yoga for and those who were sedentary revealed a stastically significant higher serum Nitric Oxide. Serum Nitric Oxide (NO µmol/l) was 77.26±37.71 in those who performed regular yoga exercise compared to 23.57±5.83 in Sedentary subjects <0.001**. It also showed a significant correlation with age (Table 4 & 5).

Discussion: Regular physical exercise has been shown to improve endothelium-dependent vasodilatation in both experimental models and humans. One of the most important molecular consequences of exercise is the absolute increase of vascular nitric oxide (NO) concentration (4). Impairment in NO production can contribute to limitations in exercise capacity through inadequate coronary or peripheral perfusion or via its metabolic effects. Exercise training in individuals with elevated cardiovascular risk or established disease can increase NO bioavailability and may represent an important mechanism by which exercise training conveys benefit in the setting of secondary prevention (8). Our study demonstrated the effect of long term exercise in the form of yoga promotes serum NO.

The production of NO is catalyzed by NOS. Three isoforms of NOS, termed neuronal NOS (nNOS), inducible NOS (iNOS) and endothelial NOS (eNOS) have been recognized. A large number of studies demonstrated that the presence of iNOS is minimal under physiological conditions. Increase in the production of NO by iNOS is observed in response to the inflammation. Inflammation-induced iNOS expression in the endothelium may contribute to vascular dysfunction by limiting the availability of tetrahydrobiopterin ($BH_{4)}$ for eNOS (4).

Tanaka et al. examined the regulation of NO on acute adaptations to exercise. It was found that acute exercise improved endothelium-dependent vasodilatation in rat aorta by increased eNOS activation. These results shows that one bout of moderate aerobic exercise improves endothelial function by increasing NO bioavailability (Tanaka, Bechara et al. 2015). Roque et al. demonstrated that 12 weeks of aerobic exercise improved endothelial function and vascular stiffness in coronary and mesenteric arteries in spontaneously hypertensive rats (Roque, Briones et al. 2013) (2).

Li-Wei Chien et al investigated the effect of 12 weeks yoga training on headache frequency, severity, duration and blood nitric oxide levels as well as headache impacts on female migraineurs' lives. Thirty-two female patients with migraine were randomly divided into two groups. The control group (*n* = 14) received medication and the yoga group (*n* = 18) participated in 12 weeks yoga training in addition to receiving the same medication as that of the control group. the metabolite of NO also was measured. After 3 months intervention, in the yoga group, there was a significant reduction in the impact of headache on patients' lives, headache frequency, and severity and a non-significant reduction in headache duration in the yoga group. There was no significant difference in the plasma levels of NO between yoga and control groups before and after the study (3). In our study a significantly higher NO levels were recorded in Exercise group compared to sedentary subject.

Li-Wei Chien et al, explored the effect of yoga on the serum nitric oxide (NO) in subjects with primary dysmenorrhoea and normal healthy controls. Yoga intervention was found to be associated with reductions in severity of dysmenorrhoea and may be effective in lowering serum homocysteine levels after an intervention period of 8 weeks. These observations suggest that yoga may have therapeutic effects in women by restoring endothelial function there were no statistically significant differences in NO levels between the two groups at baseline and after intervention(3).

Conclusion:

Hence our observation in this study was even though Subjects in the exercise group was were two Decades older with higher BMI and FBS and Co-morbidities the serum Nitric Oxide levels were comparatively higher compared to younger sedentary subjects. The presence of significantly higher levels of Nitric Oxide NO can be attributed to the continued regular exercise in the form of Yoga practices.

In conclusion, the current study provides some evidence that a simple and regular exercise in the form of Yoga increases the basal production of NO. Such effects may have contributed to the lower blood pressure observed in this study.

References:

1. AMBRING, ANNELI; BENTHIN, GUENTHER; PETERSSON, ANN SOFI; JUNGERSTEN, LENNART; WENNMALM, AKE (1994). Indirect evidence of increased expression of NO synthase in marathon runners, and upregulation of NO synthase activity during running. Circulation 90(4 PART 2): I137
2. Bode-Boger, S. M., Boger, R. H., Schroder, E. P., & Frolich, J. C. (1994). Exercise Increases Systemic Nitric Oxide Production in Men. *European Journal of Cardiovascular Prevention & Rehabilitation, 1*(2), 173-178. doi:10.1177/174182679400100212
3. Chien, L., Chang, H., & Liu, C. (2013). Effect of Yoga on Serum Homocysteine and Nitric Oxide Levels in Adolescent Women With and Without Dysmenorrhea. *The Journal of Alternative and Complementary Medicine, 19*(1), 20-23. doi:10.1089/acm.2011.0113
4. Cortese-Krott, M. M., Kulakov, L., Opländer, C., Kolb-Bachofen, V., Kröncke, K., & Suschek, C. V. (2014). Zinc regulates iNOS-derived nitric oxide formation in endothelial cells. *Redox Biology, 2*, 945-954. doi:10.1016/j.redox.2014.06.011
5. Green, D. J., Maiorana, A., O'driscoll, G., & Taylor, R. (2004). Effect of exercise training on endothelium-derived nitric oxide function in humans. *The Journal of Physiology, 561*(1), 1-25. doi:10.1113/jphysiol.2004.068197
6. Hunter, S. D., Dhindsa, M. S., Cunningham, E., Tarumi, T., Alkatan, M., Nualnim, N., & Tanaka, H. (2013). The Effect of Bikram Yoga on Arterial Stiffness in Young and Older Adults. *The Journal of Alternative and Complementary Medicine, 19*(12), 930-934. doi:10.1089/acm.2012.0709
7. Javanmard, S., Boroujeni, M., Marandi, S., Esfarjani, F., Sattar, M., & Shaygannejad, V. (2015). Yoga intervention on blood NO in female migraineurs. *Advanced Biomedical Research, 4*(1), 259. doi:10.4103/2277-9175.172995
8. Kingwell, B. A. (2000). Nitric oxide-mediated metabolic regulation during exercise: Effects of training in health and cardiovascular disease. *The FASEB Journal, 14*(12), 1685-1696. doi:10.1096/fj.99-0896rev
9. Lewis, T. V., Dart, A. M., Chin-Dusting, J. P., & Kingwell, B. A. (1999). Exercise Training Increases Basal Nitric Oxide Production From the Forearm in Hypercholesterolemic Patients. *Arteriosclerosis, Thrombosis, and Vascular Biology, 19*(11), 2782-2787. doi:10.1161/01.atv.19.11.2782
10. Lind, L., Hall, J., Larsson, A., Annuk, M., Fellstrom, B., & Lithell, H. (2000). Evaluation of endothelium-dependent vasodilation in the human peripheral circulation. *Clinical Physiology, 20*(6), 440-448. doi:10.1046/j.1365-2281.2000.00281.x
11. Maiorana, A., O driscoll, G., Taylor, R., & Green, D. (2003). Exercise and the Nitric Oxide Vasodilator System. *Sports Medicine, 33*(14), 1013-1035. doi:10.2165/00007256-200333140-00001
12. Maiorana A, O'Driscoll G, Dembo L, Cheetham C, Goodman C, Taylor R, Green D. (2000). Effect of aerobic and resistance exercise training on vascular function in heart failure. Am J Physiol Heart Circ Physiol. Oct;279 (4):H1999-2005
13. Node, K., Kitakaze, M., Sato, H., Koretsune, Y., Karita, M., Kosaka, H., & Hori, M. (1998). Increased release of nitric oxide in ischemic hearts after exercise in patients with effort angina. *Journal of the American College of Cardiology, 32*(1), 63-68. doi:10.1016/s0735-1097(98)00196-x
14. Palmer, R. M., Rees, D. D., Ashton, D. S., & Moncada, S. (1988). L-arginine is the physiological precursor for the formation of nitric oxide in endothelium-dependent relaxation. *Biochemical and Biophysical Research Communications, 153*(3), 1251-1256. doi:10.1016/s0006-291x(88)81362-7
15. Prior, B. M., Lloyd, P. G., Yang, H. T., & Terjung, R. L. (2003). Exercise-Induced Vascular Remodeling. *Exercise and Sport Sciences Reviews, 31*(1), 26-33. doi:10.1097/00003677-200301000-00006
16. Tousoulis, D., Kampoli, A., Papageorgiou, C. T., & Stefanadis, C. (2012). The Role of Nitric Oxide on Endothelial Function. *Current Vascular Pharmacology, 10*(1), 4-18. doi:10.2174/157016112798829760

17. Vallance, P., Collier, J., & Moncada, S. (1989). Effects Of Endothelium-Derived Nitric Oxide On Peripheral Arteriolar Tone In Man. *The Lancet, 334*(8670), 997-1000. doi:10.1016/s0140-6736(89)91013-1
18. Vallance, P., Bhagat, K., Macallister, R., Patton, S., Malinski, T., Radomski, M., & Moncada, S. (1995). Direct measurement of nitric oxide in human beings. *The Lancet, 346*(8968), 153-154. doi:10.1016/s0140-6736(95)91211-8

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Tables Captions

Table 1: **Study design**: A Comparative two group study those who performed regular exercise in the form of Yoga v/s Sedentary subjects

	Regular Exercise Group (Yoga (n=40))	Sedentary Group (n=30)	P value
Age in years			
<20	0(0%)	30(100%)	
21-30	7(17.5%)	0(0%)	
31-40	9(22.5%)	0(0%)	
41-50	12(30%)	0(0%)	
51-60	6(15%)	0(0%)	
61-70	6(15%)	0(0%)	
Mean ± SD	44.33±13.78	18.50±0.51	P<0.001**
Gender			
Female	18(45%)	13(43.3%)	
Male	22(55%)	17(56.7%)	
Family History of Diabetes Mellitus			
1st degree	15(37.5%)	8(26.7%)	
2nd Degree	3(7.5%)	14(46.7%)	
Family History of Hypertension			
1st degree	13(32.5%)	0(0%)	
2nd Degree	1(2.5%)	0(0%)	
Diet			
Non Vegetarian	22(55%)	26(86.7%)	
Vegetarian	18(45%)	4(13.3%)	P=0.005**

Table 2: Associated Disease in two groups studied

	Regular Exercise Group - Yoga Exercise (n=40)	Sedentary (n=30)	P value
Associated Disease			
Nil	31(77.5%)	30(100%)	
Yes	9(22.5%)	0(0%)	
• Hypertension	7(17.5%)	0(0%)	
• Respiratory	1(2.5%)	0(0%)	
• Thyroid	1(2.5%)	0(0%)	
Medications for associated disease			
No	28(70%)	30(100%)	
Yes	12(30%)	0(0%)	P=0.001**

Table 3: Comparison of Clinical risk factors BMI (kg/m^2) & WHR in two groups studied

	Regular Exercise Group-Yoga	Sedentary Group	P value
BMI (kg/m^2)			
<18.5	2(5%)	3(10%)	
18.5-25	18(45%)	20(66.7%)	
25-30	16(40%)	7(23.3%)	
>30	4(10%)	0(0%)	
Mean ± SD	25.39±3.95	22.33±3.63	P=0.001**
Waist hip ratio			
<0.8	2(5%)	15(50%)	
0.8-1	36(90%)	15(50%)	
>1	2(5%)	0(0%)	
Mean ± SD	0.91±0.07	0.80±0.07	P<0.001**
Blood Pressure			
SBP (mm Hg)	125.40±12.97	120.80±6.40	0.079+
DBP (mm Hg)	81.00±9.69	72.87±4.89	<0.001**

Table 4: Duration of Yoga (years) in two groups studied and its impact on serum Nitric Oxide

	Regular Exercise Group-Yoga	Sedentary	P value
Duration of Yoga (years)			
<1.5	17(42.5%)	30(100%)	
1.5-2.5	7(17.5%)	0(0%)	
2.5-5	11(27.5%)	0(0%)	
>5	5(12.5%)	0(0%)	
Mean ± SD	3.64±5.40	0.00±0.00	P<0.001**
Serum Nitric Oxide (NO) µmol/l	**77.26±37.71**	**23.57±5.83**	**<0.001****

Table 5: **Correlation serum nitric oxide with other variables**

Correlation serum nitric oxide with other variables	r value	P value
Serum Nitric Oxide (NO) µmol/l **v/s** FBS in mg/dl	-0.257	0.109
Serum Nitric Oxide (NO) µmol/l **v/s** BP systolic	0.251	0.119
Serum Nitric Oxide (NO) µmol/l **v/s** BP diastolic	-0.111	0.497
Serum Nitric Oxide (NO) µmol/l **v/s** Waist Hip	0.039	0.810
Serum Nitric Oxide (NO) µmol/l **v/s** BMI (kg/m^2)	0.074	0.652
Serum Nitric Oxide (NO) µmol/l **v/s** Duration of Yoga (yrs)	-0.142	0.382
Serum Nitric Oxide (NO) µmol/l v/s Age in years	**-0.362**	**0.022***