

**EFFECT OF INTEGRATED APPROACH OF YOGA
THERAPY(IAYT) ON ANTIOXDANT STATUS IN
SUBJECTS WITH ARTHRITIS**

Dissertation submitted by

Master of Science (Yoga Therapy)

By

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**Registration Number –
SVYASA/MSCYT/AUG18/15**

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CERTIFICATE

This is to certify that Shubhi Taneja who has got M.Sc (Yoga Therapy) registration with starting from August 18, 2018 by Swämi Vivekänanda Yoga Anusandhäna Samsthäna, Deemed to be University, has successfully completed the required training in acquiring the relevant background knowledge in Yoga Therapy and has completed the M.Sc. course of 2 years to submit this research project entitled **EFFECT OF INTEGRATED APPROACH OF YOGA THERAPY (IAYT) ON ANTIOXDANT STATUS IN ARTHRITIC SUBJECTS** as per regulations of the University.

DATE: 16-07-2020

PLACE: SVYASA University, Bangaluru

**DR. MITHILA M V
RESEARCH GUIDE**

DECLARATION

I, hereby declare that this study was conducted by me at Swami Vivekananda Yoga Anusandhana Samsthana (S-VYASA), Bengaluru, under the guidance of Dr. Mithila M V, S-VYASA University, Bengaluru.

I also declare that the subject matter of my dissertation entitled “EFFECT OF INTEGRATED APPROACH OF YOGA THERAPY(IAYT) ON ANTIOXDANT STATUS IN ARTHRITIC SUBJECTS” has not previously formed the basis of the award of any degree, diploma, associate-ship, fellowship or similar titles.

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Shubhi Taneja

**STANDARD INTERNATIONAL TRANSLITERATION CODE USED TO
TRANSLITERATE SAṂSKṚTA WORDS**

अ	आ	इ	ई	उ	ऊ	ऋ		
a	ā	i	ī	u	ū	ṛ		
ऌ	ए	ऐ	ओ	औ	A	A>		
ṛ	e	ai	o	au	aṁ	aḥ		
	क	ख	ग	घ	ङ			
	ka	kha	ga	gha	~na			
	च	छ	ज	झ	ञ			
	ca	cha	ja	jha	~na			
	ट	ठ	ड	ढ	ण			
	ṭa	ṭha	ḍa	ḍha	ṇa			
	त	थ	द	ध	न			
	ta	tha	da	dha	na			
	प	फ	ब	भ	म			
	pa	pha	ba	bha	ma			
य	र	ल	व	श	ष	स	ह	ळ
ya	ra	la	va	śa	ṣa	sa	ha	ḷa

ABBREVIATIONS

1. TC: Total Cholesterol
2. TG: Triglycerides
3. LDL: Low-density lipoprotein cholesterol
4. HDL: High-density lipoprotein cholesterol
5. ALT: Alanine aminotransferase
6. AST: Aspartate transaminase
7. ABTS: (2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid))
8. UA: Uric acid
9. TAC: Total antioxidant capacity
10. NO : Nitric oxide
11. MDA : Malondialdehyde
12. GSH : Glutathione

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1. INTRODUCTION

1.1 YOGA:

Yoga, is a form of mind-body therapy, which was developed in India thousands of years ago and consists of eight components (limbs): yamas (ethical disciplines), niyamas (rules of behaviour), asana (postures), pranayama (breathing), pratyahara (sensory control), dharana (mind concentration), dyhana (meditation), and Samadhi. Then there is relaxation with some yoga styles that also incorporate discussions of yoga philosophy and lifestyle advice. Although originating in ancient India, in the Western context Yoga has become a common way of achieving and maintaining health and wellbeing. Use of yoga is increasingly prevalent worldwide (Djalilova et al., 2018)(Lauche, Hunter, Adams, & Cramer, 2019).

1.2 OXIDATIVE STRESS:

Oxidative stress is caused by exposure to intermediate reactive oxygen such as superoxide anion ($O_2^{\cdot-}$), hydrogen peroxide (H_2O_2), and hydroxyl radical (HO^{\cdot}) that can damage the proteins, nucleic acids, and cell membranes (Lauche et al., 2019). Oxidative stress is a steady state level of reactive oxygen radicals in the biological system. It may result from over-production of oxygen radicals and/or reduced efficiency of the antioxidant system. There is growing evidence that the cumulated damage from reactive oxygen species leads to various diseases (Hegde, Adhikari, Shetty, Manjrekar, & Souza, 2013). Antioxidants are natural substances that can prevent or delay certain forms of cell damage. Free radicals are highly unstable molecules that are naturally formed when you work out and the body converts food into energy (Cyril, 2010).

Sources of oxidative stress: oxidative stress is an inevitable by-product of aerobic lifestyle, because $O_2^{\cdot-}$ and H_2O_2 are formed whenever molecular oxygen chemically oxidizes electron carriers. $O_2^{\cdot-}$ and H_2O_2 have different chemical reactivities and produce distinct types of damage within the cells (Lauche et al., 2019).

Effect of yoga on Oxidative Stress:

Oxidative stress is the product of excess free radicals inside the cells over antioxidants. The results of excess free radicals are the oxidation of lipids and proteins on the cell membrane, and the mutation of deoxyribonucleic acids within the nucleus and mitochondria.

Several studies have documented a decrease in the oxidative stress markers and an increase in antioxidants after yoga practices like Sudarshan Kriya (Form of yoga propagated by Art of living), Hatha yoga practice, Nadishodhan pranayama, and different yogasanas and pranayama modules. Malondialdehyde (MDA), NO, F2-isoprostane were the principal parameters of the oxidative stress tested. GSH, glutathione S transferase, glutathione peroxidase, vitamin C, vitamin E, superoxide dismutase, phospholipase A2 - PLA2 and protein oxidation were the antioxidantsexamined (Chakrabarty & Bhat, 2018).

1.3 ARTHRITIS:

"Arthritis" is a combinatorial word that originates from a mixture of Latin and Greek. "Arthron" in Greek means joint, and "Itis" in Latin indicates inflammation. While arthritis is generally treated as an illness caused by inflamed joints. It is necessarily not just a single disease but a set of medical problems collectively called "Arthritis". In the case of arthritis, it is mainly due to faulty joints that the suffering begins.

There are many reasons for the disease; (a) possible damage to cartilage, (b) lack of synovial fluid, (c) autoimmune attack, (d) infections. Here are some common ones: Osteoarthritis (OA), Rheumatoid Arthritis (RA), Gout, Ankylosing Spondylitis (AS), Lupus arthritis (LA), Infectious arthritis (IA), Juvenile Arthritis (JA), Psoritic arthritis (PA), fibromyalgia. The majority of outbreaks are seen for OA, RA and gout, to some extent, AS, while the remaining are less frequent (*ARTHRITIS: CLASSIFICATION, NATURE & CAUSE - A REVIEW. Sankar Mitra PhD Review Article ARTHRITIS: CLASSIFICATION, NATURE & CAUSE - A REVIEW. Sankar Mitra PhD *, 2015*).

1.4 ROLE OF OXIDATIVE STRESS IN ARTHRITIS:

Increase in reactive oxygen species (ROS) plays a very important role in the pathogenesis of rheumatoid arthritis (RA). Also antioxidants and anti oxidative enzymes have been shown to reduce cartilage damage in animal models of RA. Over the course of evolution, cells have developed complex mechanisms as defense against oxidative stress. A battery of genes encoding detoxifying and antioxidative enzymes is orchestrated upon exposure to ROS. In a study NRF2 (nuclear factor erythroid 2-related factor 2, a transcription factor that maintains the cellular defense against oxidative stress in RA) was activated in the joints of arthritic mice and of patients with RA. NRF2-knockout mice had more severe cartilage injuries and more

oxidative damage. Also there were a number of spontaneously fractured bones in NRF2-knockout mice with AIA(Antibody-induced arthritis) (Wruck et al., 2011).

Inflammation is a natural form of defense against pathogens. This exists in many pathogenic diseases (microbial and viral infections, allergen contamination, radiation and toxic chemicals, autoimmune diseases, etc.). Chronic diseases associated with higher ROS development result in OS and a number of protein oxidations. In addition, some oxidized proteins cause the release of inflammatory signal molecules. RA is one of the conditions under which OS is induced. A five-fold increase in mitochondrial ROS production in whole blood and monocytes in patients with RA compared to healthy subjects suggests that OS is a pathogenic hallmark in RA. Free radicals are indirectly involved in joint damage because they also play a role as secondary messengers of inflammatory and immune cellular response in RA (Access, n.d.).

1.5 ROLE OF YOGA IN ARTHRITIS:

In a particular study where yoga was given to participants with arthritis, among the cardinal biomarkers of neuroplasticity, there was a significant increase in BDNF, serotonin, endorphins. Among the biomarkers for cellular health & integrity – there was a significant decrease in OS as there was decrease in ROS. Yoga intervention upregulates the expression of DNA repair genes, and the levels of anti-inflammatory cytokines and decreases the levels of inflammatory cytokines. Thus decrease in the severity of RA, which further affects psychological well being by changes in biomarkers at systemic level (Delhi & Delhi, 2018).

After attention to posture, deep breathing, chanting, yoga practice often begins with a slow movement sequence to increase blood flow and warm muscles. This sequence is followed by poses that include flexion, extension, adduction, abduction, and rotation. Holding poses builds strength by engaging muscles in isometric contraction. Moving joints through their full range of motion increases flexibility, whereas standing poses promote balance by strengthening and stabilizing muscles and improving proprioception to reduce falls. Thus, yoga incorporates several elements of exercise that may be beneficial for arthritis. To cope with pain, patients with arthritis often reduce activity. Inactivity can result in muscle or tendon shortening, articular capsule contraction, and weakened ligaments. Conversely, regular activity may decrease pain and preserve stability (Haaz & Bartlett, 2011).

2. LITERATURE REVIEW

2.1 Ancient literature review:

2.1.1 Relation of pain and Vata according to Ayurveda:

According to Ayurveda pain can not exist without imbalance in Vata. This imbalance happens:

- When it is increased beyond limits.
- When it is vitiated spoiling Kapha and other Dhatus like Rakta(blood), Mansa(Ligaments and Muscles), Meda(Synovial fluid membrane), Asthi(Bones and Cartilage) and Majja(Nerves).

सन्कोचः पर्वणाम् स्तम्भो भेदोऽस्थाम् पर्वणामपि ।

लोमहर्षः प्रलापश्च पाणिपृष्ठशिरोग्रहः ॥ (*Cha.Chi.28/20*)

Vata gets aggravated because of :

- Disturbed metabolism(Agni dushti)
- Circulating endo-toxins(Ama)
- Wrong food habits(Mithya ahara)
- Wrong activities(Mithya vihar)
- Holding of nature cells(Vegavrodha)
- Wrong life style causing depletion or loss of tissues.

2.1.2 SANDHIGATA VATA:

हन्ति सन्धिगताः सन्धिन् शूलटपौ करोति च । - मधवनिदान

वातपूर्णद्रुतिस्पर्शः शोथः सन्धिगतेऽनिले ।

प्रसारणाकुञ्चनयोः प्रवृत्तिश्च सवेदना ॥ (*Cha.Chi.28/37*)

When such vitiated vata affects the joints it causes strong pain, swelling and discomfort in movement called as *Sandhigata vata*. This condition indicates the explanation similar to what we now know as *Osteoarthritis or degenerative arthritis*.

2.1.3 AMAVATA:

हस्तपादशिरोगूल्फत्रिकजानूरुसंधिषु ।

करोति सरूजं शोथं यत्र दोशः प्रपद्यते ॥ माधवनिदान-आमवात

Where Ama(endo-toxins) involved along with of vata to produce inflammatory arthritis with special mention of *Rheumatoid Arthritis* creating severe pain, swelling, redness, warmth.

2.1.4 VATA RAKTA:

स्वेदोत्थर्मम् न वा काष्ण्यम् स्पर्शाज्ञत्वम् क्षतेऽ तिरुक् ।

सन्धिशैथिल्यमालस्यम् सदनम् पिडकोद्गमः ॥

जनुजन्धोरुकटयम्सहस्तपादान्गसन्धिषु ।

निस्तोदः स्फुरणम् भेदो गुरुत्वम् सुप्तिरेव च ॥ (*Cha.Chi.21/13*)

Where the vitiated Vata along with the vitiated Rakta produce painful condition of the joints mimicking the picture of crystal arthritis or *Gouty Arthritis*.

2.1.5 ROLE OF RASAYANA (Antioxidants) ACCORDING TO AYURVEDA:

A cell is a fundamental unit of our body and so if it is healthy , the whole of our systems will be balanced in their functions .

वाता पित्ता कफा देहे सर्वं स्रोतो अनुसरिना (*Cha.Chi.28/59*)

We can say that Vata , Pitta , Kapha hae their identity in each cell of our body as each of the cells can be correlated to srotasa(Organs).

In relation to disease , Sushruta stated :

सर्वेशम च व्याधिनाम वाता पित्ता श्लेष्मनम् एव मलमुलकम् (*Su.Su.24/8*)

Thus , imbalance of Vata , Pitta , Kapha at cellular level can be the precursor of a diseased condition .

While explaining basic pathological process in any disease,

कूपितनम हि दोशनम शरिरे परिधवतम ।

यत्र सन्ग ख वैगुन्यात व्याधि तत्र उपजायते ॥ (Su.Su.24/10)

So, we land up with the conclusion that disease is nothing but cellular equilibrium disturbance .
So we can correlate *oxidative stress* with “ *Khavaigunya*” .

The cumulative effect of multiple changes by *free radicals* is the proximate cause of cell death.
“ *Uttarottara dhatu dushti*” is nothing but the same .

Ayurveda claims to balance this status again with the help of Rasayana . As, Rasayana has an important role in maintaining homeostasis .

रसायन तन्त्रम् नाम वयस्थापनम आयुर्मेधबलकरम रोगापहरन समर्थम् च । (Su. Su.1/10)

Here, according to Sushruta , Rasayana helps in arresting ageing process , lengthening of life span , improves memory , stamina and develops resistance against diseases.

Rasayana like haritaki, Amalaki, Vacha, Vidang, Haridra, Shunthi, Pippali, Guda have a specific organ related activity which controls the imbalance caused by free radicals.(Ranade, 2015).

2.2 Scientific literature review:

SUMMARY TABLE OF SCIENTIFIC RESEARCH ON ARTHRITIS, OXIDATIVE STRESS AND YOGA

Author and year	Sample size	Intervention	Variables	Result	Conclusion
(Chakrabarty & Bhat, 2018)	40 male and female volunteers	Sudarshan kriya yoga	MDA level	12% reduction of MDA levels	Many studies have reported that the markers of oxidative stress were decreased and antioxidants were increased after practicing yoga.
(Lauche et al., 2019)	640 individuals with mainly lower extremity osteoarthritis aged 50–80 years	multicomponent interventions employing postures, breathing, and/or meditation	Pain, Physical function, Stiffness	Effect sizes for these variables were moderate to large, with lower confidence boundaries in the small to moderate range.	The findings of this meta-analysis indicate that yoga may be effective for improving pain, function, and stiffness in individuals with osteoarthritis of the knee, compared to exercise and non-exercise control groups.
(Djalilova et al., 2018)	A total of 937 participants, weighted mean age of 48.54 years	Hatha yoga was the most common, kundalini yoga, “classical yoga” grounded in the principles of the Kaivalyadhama Yoga Institute.	Most common biomarkers measured were interleukin-6 (n ¼ 11), C-reactive protein (n ¼ 10), and tumor necrosis factor (n ¼ 8)	Yoga interventions reduced inflammation in the studies involving cancer, cardiovascular disease, chronic stress condition, and rheumatoid arthritis.	The combination of postures, slowbreathing techniques, and higher doses of yoga practice may be particularly advantageous in reducing inflammation in chronic conditions.
Janice k. Kiecolt-glaser, lisa christian et al. 2010 (Questionnaire & Affect, n.d.)	50 healthy women, mean age, 41.32 years	yoga, movement control, and passive-video control	Demographic, Physiological, Dietary, and Behavioral Data for Novice and	Novices had substantially less hamstring and low-back flexibility than	across the battery of inflammatory assays, 60% of novices were high

			Expert Yoga Practitioners	yoga experts, Participants' positive mood scores increased after yoga, decreased after the video, and were unchanged after movement, overall mean heart rate during the yoga condition did not differ from that in the movement condition	producers compared with only 24% of experts, and 40% of experts were low producers compared with 0% of novices. These data suggest that regular yoga practice may reduce inflammation below levels
(Wruck et al., 2011)	Arthritic mice and patients with RA.		Activation status of Nrf2, expression of vascular endothelial growth factor (VEGF)-A	Nrf2 was activated in the joints of arthritic mice and of patients with RA. Nrf2-knockout mice had more severe cartilage injuries and more oxidative damage, VEGF-A mRNA and protein expression was upregulated in Nrf2-knockout mice during AIA	These results provide strong evidence that oxidative stress is significantly involved in cartilage degradation in experimental arthritis, and indicate that the presence of a functional Nrf2 gene is a major requirement for limiting cartilage destruction.
(Hegde et al., 2013)	Twenty nine prediabetes subjects aged 30—75 years.	Participants were randomized to either 3-month yoga or wait-list control groups.	Malondialdehyde, glutathione, vitamin C, vitamin E, superoxide dismutase, plasma glucose, glycated haemoglobin	significant decline in malondialdehyde, in comparison with the control, there was a significant improvement in BMI, waist circumference, systolic blood	Yoga intervention may be helpful in control of oxidative stress in prediabetes subjects. Yoga was also helpful in reducing some risk factors for type 2diabetes —

				pressure and fasting glucose levels at follow-up.	BMI, waist circumference, systolic blood pressure and fasting glucose.
(Greysen, Hong, & Katz, 2019)	The sample included 398 adults with RA; 88% were females, 66% were white, mean age 61.8 years, mean disease duration 24.8 years; 10.6% participated in yoga.	Vinyasa, Bikram, Hatha, Iyengar, and restorative yoga styles were practiced, mostly in a group setting.	Pain, sleep quality, fatigue, depression, physical function.	Yoga participation was not associated with improved pain, sleep quality, fatigue severity, and depressive symptom severity in the population.	In this descriptive cross-sectional study, adults with RA who practice yoga are significantly more likely to be employed full-time and have better physical function than adults with RA who do not.
(Yadav, Magan, Mehta, Sharma, & Mahapatra, 2012) ⁱ	Eighty-six (86) patients (44 female, 42 male, 40.07 – 13.91 years), Patients with chronic inflammatory diseases and overweight/obese subjects	A pretested intervention program included asanas (postures), pranayama (breathing exercises), stress management, group discussions, lectures, and individualized advice.	stress (plasma cortisol and b-endorphin) and inflammation (interleukin [IL]-6 and tumor necrosis factor [TNF]-a)	mean level of cortisol decreased from baseline to day 10 (p = 0.001) while b-endorphins increased from baseline to day 10 (p = 0.024). Also, there was reduction from baseline to day 10 in mean levels of IL-6 (p = 0.036) and TNF-a (p = 0.002).	This brief yoga-based lifestyle intervention reduced the markers of stress and inflammation as early as 10 days in patients with chronic diseases
Surabhi Gautam, Madhuri Tolahunase, Uma Kumar and Rima Dada (Delhi & Delhi, 2018)	72 RA patients	8 weeks yoga based mind body intervention (MBI)	erythrocyte sedimentation rate(DAS28ESR), health assessment questionnaire disability index (HAQ-DI), depression severity	significant decrease in the severity of RA as seen by reduction in levels of various systemic inflammatory markers as well as in DAS28ESR, decline in depression symptoms over the period of 8	Yoga, a mind body intervention re-established immunological tolerance by aiding remission at molecular and cellular level along with significant reduction in depression. Thus in this inflammatory

				weeks	arthritis with a major psychosomatic component, yoga can be used as a complementary therapy
(Study, 2014)	The study comprised 718 adolescents	Physical activity(PA) measured by accelerometry and liver enzymes	liver enzymes (i.e., AST, ALT, AST/ALT, and GGT)	Meeting the PA recommendations (60 min/d of MVPA) was significantly associated with higher AST and AST/ALT, which persisted after further adjusting for sedentary time and waist circumference.	Meeting the current PA recommendations of 60 min/d is associated with higher levels of AST and AST/ALT regardless of time spent sedentary as well as total and central body fat in European adolescents.

3. AIMS AND OBJECTIVES

3.1 AIMS OF THE STUDY:

To explore/understand the effect of yoga (IAYT) in Arthritis- induced oxidative stress.

3.2 OBJECTIVES OF THE STUDY:

- To screen the Arthritis participants for study recruitment.
- To assay/quantify the Biochemical markers *invitro* pre and post yoga intervention.
- To correlate the effect of yoga intervention in managing Arthritis induced oxidative stress.

3.3 RESEARCH QUESTIONS:

Does IAYT module have any effect on antioxidant status in the subjects with Arthritis?

3.4 HYPOTHESIS:

Yoga therapy (IAYT) has an effect on antioxidant status in the subjects with Arthritis.

3.5 NULL HYPOTHESIS :

Yoga therapy (IAYT) does not have an effect on antioxidant status in the subjects with Arthritis.

4. METHODS

Blood sampling:

At both baseline and after intervention, anthropometric data were recorded, and venous blood samples (fasting) were collected for biochemical and antioxidant assessments.

Biochemical assessments:

The biochemical assessments were performed using an automated serum chemistry analyzer (BS390, Mindray, China) with standard reagents. Parameters assessed were; Fasting Blood Glucose(FBS), Low Density Lipoprotein (LDL), High Density Lipoprotein (HDL), Triglycerides (TG), Total Cholesterol (TC), Uric Acid (UA), Albumin (Alb), Aspartate transaminase (AST) and alanine transaminase (ALT)

Antioxidant measurement:

(Total antioxidant Capacity – TAC by ABTS method)

TAC assay was performed according to the method of (Arnao, Cano, & Acosta, 2001) with slight modifications. Briefly, ABTS stock solution was prepared by mixing equal volumes of 7mM ABTS with 2.45mM potassium persulfate (v/v) and allowed to react at room temperature in dark for 14hrs. Methanol was added to this mixture to obtain absorbance of 0.7 ± 0.01 at 734nm. Serum sample was then treated with the ABTS stock, incubated at room temperature for 7 min and absorbance measured at 734nm using plate reader (Enspire, Perkin Elmer). The ABTS scavenging capacity of the sample was compared against a Trolox standard curve (0.5-1.5mM) and expressed as Trolox equivalents. All reactions were performed in duplicates.

4.1 PARTICIPANTS:

- Sample size:41 (38 Females, 3 Males)
- Selection of participants: For selection of sample, convenient sampling was done where the sample was taken from a group of people easy to reach.
- Source of Participants: Section-D of Arogyadhama(Holistic Home) which deals with the intervention of yoga related to Arthritis, in Prashanti kuteeram, Bangalore.

4.2 INCLUSION CRITERIA:

- Age Group -20 to 70
- People who came for participation in Section-D of Arogyadhama .

- People with arthritic symptoms or diagnosed arthritis.
- People who were able completely participate in the all the practices.

4.3 EXCLUSION CRITERIA:

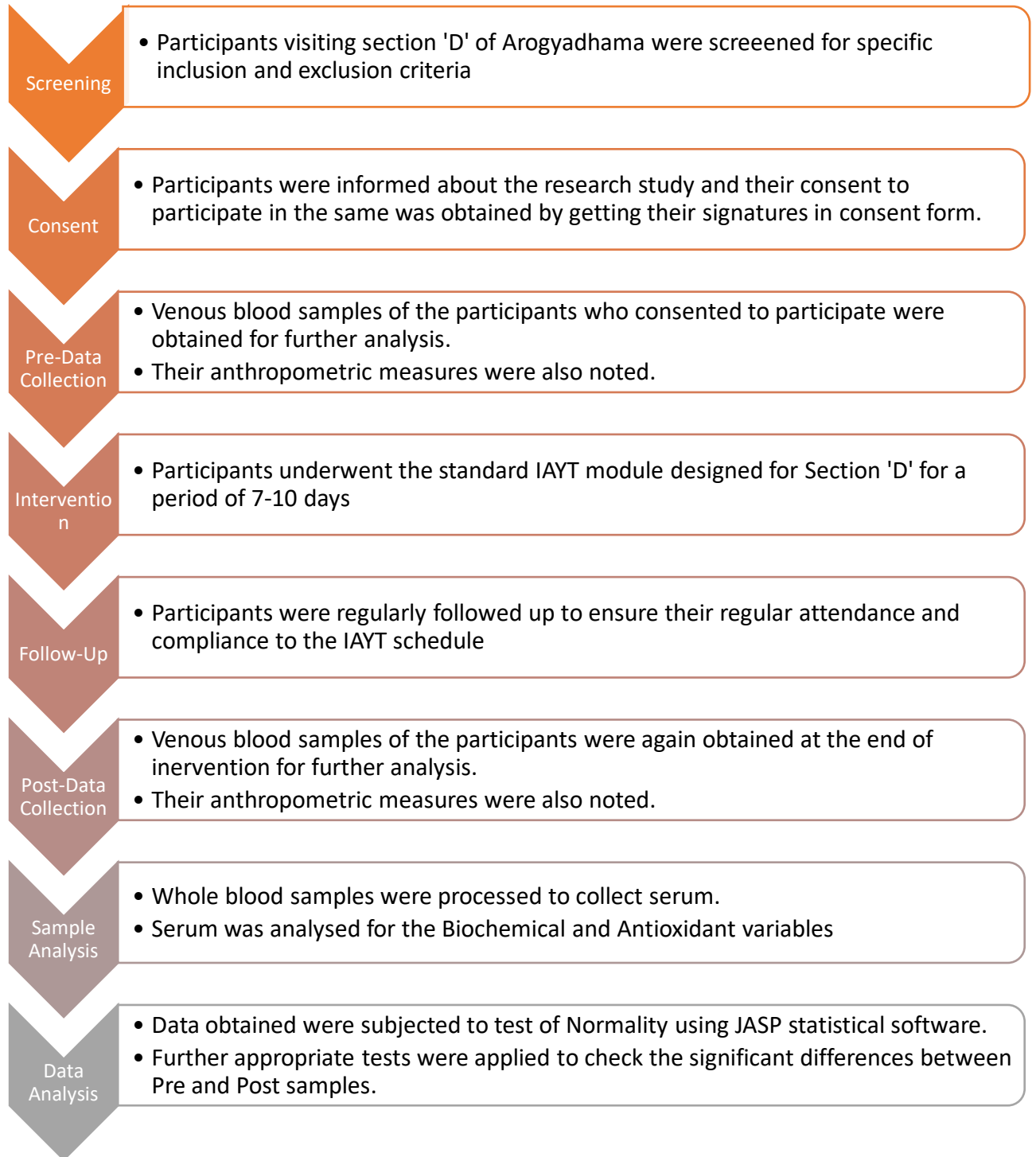
- Those who were practicing yoga prior to the intervention; at least for 6 months.
- Those with serious injury or with a history of any major surgery in the recent past.

4.4 ETHICAL CONSIDERATION:

- Subject approval with a signature on consent form before collecting their blood samples
- Maintenance of confidentiality of all the patient details.

5. DESIGN OF THE STUDY: PRE-POST DESIGN

STUDY FLOWCHART



5.1 VARIABLES STUDIED:

Primary: Antioxidants levels (Total antioxidant capacity, Albumin and Uric acid).

Secondary: Lipid profile(LDL, HDL, TC, TG), Liver function tests(ALT,AST), Weight, Blood Pressure, Pulse rate, Bhramari time, Serum glucose.















5.2 INTERVENTION:

Detailed Daily Activities of Integrated Approach of Yoga Therapy(IAYT)

Time	Schedule
5:30 AM	OM MEDITATION
6:00 AM	BREATHING AND LOOSENING PRACTICES
7:00 AM	BHAGWAD GEETA CHANTING FOLLOWED BY LECTURE ON YOGA
8:00 AM	BREAKFAST
9:00 AM	LECTURE ON YOGA
10:00 AM	DISCUSSION WITH WARD DOCTORS AND ASSESSMENT OF VITALS
11:00 AM	PRANAYAMA PRACTICES
12:00 AM	LOOSENING PRACTICES AND CHAIR SURYA NAMASKARA
1:00 PM	LUNCH
3:00 PM	CYCLIC MEDITATION
4:00 PM	BREATHING AND LOOSENING PRACTICES
5:00 PM	TUNING TO NATURE
6:00 PM	BHAJAN
6:30 PM	TRATAKA
7:00 PM	MIND SOUND RESONANCE TECHNIQUE
7:30 PM	DINNER
8:30 PM	HAPPY ASSEMBLY

List of Section-D Practices

Passive Rotation of Toes		Wrist movement	
Toe Bending		Elbow Bending	
Ankle bending & Rotation		Shoulder movement	
Loosening of Knee & Tightening		Neck Bending	
Patella Movement & Massaging		Neck twisting & rotation	
Knee bending		Straight Leg Raising Cycling	
Knee Rotation		Setubandasana	
Half butterfly		Pawan muktasana	
Butterfly		Folded Legs Lumbar stretch	
Waist rotation		Viparitkarani	
Loosening of fingers		DRT	
Hands stretch Breathing		Ardha Chakrasana	

Ankle Stretch		Bujangasana	
Anguli Sakti Vikasaka		Shalabasana	
Kara Tala Sakti Vikasaka		Cycling	
Kara Prista Sakti Vikasaka		St. Leg Raising	
Mani Bandh Sakti Vikasaka		Pavanmuktasana lumbar stretch 10	
Kaphoni Sakti Vikasaka		Setubandasana	
Bujabanda shakti Vikasaka		Vertical stretch of the knee	
Griva Sakti Vikasaka		Vipritkarni	
Ardhakati chakrasana		DRT(Deep Rexalation Technique)	

5.3 DATA EXTRACTION AND ANALYSIS:

Data was extracted manually. Data was entered in Microsoft excel. Both pre and post values had been updated in the result table.

JASP Version 0.11.1 was used for the purpose of analysis. JASP is a free and open-source graphical program for statistical analysis supported by the University of Amsterdam.

- For Assumption check Shapiro-Wilk test was used.
- For Within Group, Paired Sample T-Test was used.
- Student Test was done for Variables with data normally distributed and Wilcoxon signed rank test was done for Variables with data not normally distributed.

6. RESULT

Table 1: Test of Normality (Shapiro-Wilk) For Anthropometric variables

		W	p
weight_pre	- weight_post	0.931	0.016
systolicBP_pre	- systolicBP_post	0.965	0.243
diastolicBP_pre	- diastolicBP_post	0.980	0.692
bhramari_pre	- bhramari_post	0.987	0.915
pulse_pre	- pulse_post	0.980	0.664

Table 2: Mean± SD and within-group analysis of Anthropometric Variables

Anthropometric Variables	Pre-treatment	Post-treatment	Effect size	95% CI		P-value
				lower	upper	
Age(years)	58.049±8.888	-	-	-	-	-
Height(cm)	153.56±7.682	-	-	-	-	-
Weight(kg)	73.732±14.86	72.295±14.214	0.926	0.854	0.963	< .001
Systolic BP(mmHg)	125.659±17.046	122.39±11.638	0.218	0.094	0.526	0.171
Diastolic BP(mmHg)	78.78±10.525	77.317±10.914	0.150	0.159	0.457	0.344
Bhramari(sec)	10.366±4.097	13.902±5.324	- 0.840	1.194	0.480	< .001
Pulse Rate(bpm)	77.902±8.062	78.951±9.066	- 0.095	0.401	0.212	0.546

Table 2: P<0.05 was considered statistically significant.

Table 3: Test of Normality (Shapiro-Wilk) For Biochemical Variables

		W	p
TC_PRE	- TC_POST	0.762	< .001
TG_PRE	- TG_POST	0.832	< .001
LDL_PRE	- LDL_POST	0.792	< .001
HDL_PRE	- HDL_POST	0.986	0.876
ALT_PRE	- ALT_POST	0.798	< .001
AST_PRE	- AST_POST	0.959	0.141
GLU_PRE	- GLU_POST	0.899	0.002

Table 4: Mean± SD and within-group analysis of Biochemical Variables

Biochemical Variables	Pre-treatment	Post-treatment	Effect size	95% CI		P-value
				lower	upper	
Glucose(mg/dl)	101.878±26.988	99.390±25.835	0.120	-0.239	0.451	0.523
LDL-C(mg/dl)	159.610±52.401	166.195±54.352	-0.236	-0.541	0.123	0.207
HDL-C(mg/dl)	57.171±16.456	56.951±15.339	0.034	-0.273	0.339	0.831
TC(mg/dl)	196.85±45.336	203.317±51.183	-0.118	-0.438	0.228	0.513
TG(mg/dl)	129.463±53.141	128.024±55.341	0.159	0.189	0.471	0.378
ALT(U/L)	23.256±8.196	25.759±8.541	-0.601	-0.782	-0.327	< .001
AST(U/L)	21.636±7.162	24.194±7.230	-0.501	-0.823	-0.173	0.003

Table 4: P<0.05 was considered statistically significant.

Table 5: Test of Normality (Shapiro-Wilk) For Antioxidants

		W	p
PRE_ALB g/dl	- POST_ALB g/dl	0.913	0.004
PRE_UA	- POST_UA	0.933	0.048
PRE_ABTS	- POST_ABTS	0.985	0.840

Table 6: Mean± SD and within-group analysis of Antioxidants

Antioxidants in serum	Pre-treatment	Post-treatment	Effect size	95% CI		P-value
				lower	upper	
TAC(TE/ml)	1.65±0.57	1.438±0.681	0.261	-0.052	0.571	0.102
Albumin(g/dL)	4.534±0.653	4.736±0.475	-0.513	-0.738	-0.184	0.008
Uric Acid(mg/dL)	4.928±1.302	5.2±1.279	-0.402	-0.699	0.014	0.075

Table 6: P<0.05 was considered statistically significant.

The study included 41 participants (38 Females, 3 Males; mean age 58.04±8.888 years). Normality distribution (Shapiro-Wilk Test) given in **Table 1** for demographics showed that Systolic BP, Diastolic BP, pulse and Bhramari were normally distributed, while weight was not normally distributed.

As seen in the demographic characteristics (baseline Vs post-treatment) presented in **Table 2**, there was a significant reduction in weight and improvement in Bhramari time in the post-treatment scenario. Large effect size was also observed for both weight and Bhramari time.

Normality distribution (Shapiro-Wilk Test) given in **Table 3** for biochemical variables showed that only HDL and AST measures were normally distributed, whereas the rest of the measures (TC, TG, LDL, ALT, GLU) were not normally distributed.

Table 4 shows the mean(SD) of the biochemical variables at both pre and post treatment. As obtained by within group analysis, only the liver function tests (ALT, AST) show significant

differences at post-treatment scenario with a moderate effect size, while the rest of the biochemical measures (Glu, LDL, HD, TC, TG) have varied non-significantly with low effect size.

Normality distribution (Shapiro-Wilk Test) given in **Table 5** for Antioxidant variables shows that only ABTS (or TAC) measure was normally distributed where as both Albumin and Uric acid measures were not normally distributed.

As seen in the **Table 6** which gives the mean (SD) for the antioxidant variables, Albumin shoed significant difference, followed by uric acid, which was close to significance and TAC was the least significant when subjected to within group analysis. Although it can be seen that, both Albumin and Uric Acid exhibit moderate effect size.

7. DISCUSSION

The participants of this study spent one week in prashanti kuteeram, following whole day routine which is designed to provide yogic way of healing to them. Having seen the pre-post results in previous section, we observed slightly increased levels of AST and ALT with a moderate effect size of 0.501 and 0.601 respectively.

Mild elevations of transaminase levels are often transient.(Lazo, Sc, & Clark, 2008). More than 30% of adults with initially elevated AST and ALT levels have normal results on a repeat testing (Lazo, Selvin, & Clark, 2008).

AST is present in the liver but in good amounts also in other tissues including the muscles, So it cannot be excluded however that the observed association of total Physical Activity.and MVPA(Moderate to Vigorous Physical Activity) with AST and AST/ALT might be partially explained by increased AST released from muscles after Physical Activity.(Study, 2014)

ALT is present in the liver, muscle, and kidney. In fact, muscle has more AST and ALT when compared with that in the liver because of a larger tissue mass. Thus, transaminase levels may be elevated due to various types of muscular disorders or injury.(Pavletic, Pao, & Wright, 2015) Since it was a short term intervention because of initial muscle straining as they had not done yoga asanas before, the levels of AST and ALT had increased slightly.

There was increase in the levels of Albumin, with a moderate effect size of 0.513. Albumin is the most abundant circulating protein in the plasma, exerts important antioxidant activities. The molecule acts through its multiple-binding sites and free radical-trapping properties. If beneficial actions are exerted by native albumin, very detrimental effects can be provoked by oxidized form of the protein and acting as a biomarker of oxidative stress.(Roche, Rondeau, Singh, Tarnus, & Bourdon, 2008).

According to (Wunder et al., 2003) Because the synovium of patients with rheumatoid arthritis (RA) shares various features observed also in tumors albumin appears to be a suitable drug carrier in RA, most probably due to effects on synovial fibroblasts, which might increase the therapeutic efficacy of and reduce side effects of MTX(methotrexate).

Uric acid levels were seen to be increased with lower effect size of 0.402. According to (Kusumayanti, Dewantari, & Author, 2017) Uric acid is the metabolism result in the body, which levels should not be excessive. Everyone has uric acid in their body, due to every normal metabolism producing uric acid. The trigger is food and other compounds that consist of a lot of purines. The body provides 85% (percent) of purine compounds for daily needs. It defines that the food purine requirement is only about 15%. The factors that contribute to increased uric acid production is the foods consumption that consists of a lot of purine and uric acid, obesity, drug use, physical activity and certain diseases in the blood.

Also Uric acid is a powerful scavenger of carbon-centered and peroxy radicals in the hydrophilic environment but loses an ability to scavenge lipophilic radicals and cannot break the radical chain propagation within lipid membranes (Muraoka & Miura, 2003). uric acid is involved in a complex reaction with several oxidants and may have some protective effects under certain conditions.

As we have seen in results section the value for TAC has reduced with a low effect size of 0.261. Which is probably because of the change in place, sleeping patterns, physical activities, food etc. as initially these changes causes certain stress in the body. And as we asked the participants also they discussed the changes are taking few days to get used to.

With long term yoga practice there has been a decrease in oxidative stress and increase in antioxidant activities according to several studies. According to a study (Randomized & Study, 2015) regular practice of yoga for 12 weeks significantly decreased oxidative stress-related concerns, including serum levels of NO, MDA, and F2-isoprostane. The total GSH contents, activities of GSHPx, and GST were significantly increased after yoga classes, GSH is the most representative antioxidant protein.

In another study (Ray, 2007), the yoga group was trained in yoga for 6 months where the yoga schedule consisted of prayers, *asana*, *pranayama*, and meditation. After completion of 6-month training period reduced and oxidized glutathione, glutathione reductase activity and total antioxidant status (TAS) were estimated.

8. CONCLUSION

In this research participants underwent yoga intervention in prashanti kuteeram which included asana, pranayama, kriya, bhajan, meditation, chanting, sattwik diet. They come in batches and stay here for 1 week or 2 weeks, we have selected the participants who had stayed for 1 week. So we don't see increased levels of antioxidants because of the short term practices they underwent. But most of them indicated reduction in knee pain and reduced stress levels after the intervention. It may be concluded that yoga may upregulate the antioxidant capacity of cells to combat oxidative stress. And the anthropometric and pain reduction suggest that Yoga can be used as an adjunct therapy for lifestyle modification and management of Arthritis and related complications.

9. REFERENCES

- Access, O. (n.d.). *We are IntechOpen , the world ' s leading publisher of Open Access books Built by scientists , for scientists TOP 1 % Impact of Oxidative Stress on Inflammation in Rheumatoid and Adjuvant Arthritis : Damage to Lipids , Proteins , and Enzymatic Antioxidant Defense in Plasma and Different Tissues.*
- Arnao, M. B., Cano, A., & Acosta, M. (2001). *The hydrophilic and lipophilic contribution to total antioxidant activity.* 73, 239–244.
- ARTHRITIS : CLASSIFICATION , NATURE & CAUSE - A REVIEW . Sankar Mitra PhD Review Article ARTHRITIS : CLASSIFICATION , NATURE & CAUSE - A REVIEW . Sankar Mitra PhD *.* (2015). (December 2013).
- Chakrabarty, J., & Bhat, V. R. (2018). *Yoga Interventions for Oxidative Stress and Antioxidant Status.* (January), 2–7. <https://doi.org/10.5958/0976-5506.2018.00038.4>
- Cyril, A. (2010). *(12) United States Patent.* 2(12).
- Delhi, N., & Delhi, N. (2018). *Un co rre cte d Au tho r P Impact of yoga based mind-body roof Un co rre cte d Au tho r P roof.* <https://doi.org/10.3233/RNN-180875>
- Djalilova, D. M., Schulz, P. S., Berger, A. M., Case, A. J., Kupzyk, K. A., & Ross, A. C. (2018). *Impact of Yoga on Inflammatory Biomarkers : A Systematic Review.* (c). <https://doi.org/10.1177/1099800418820162>
- Greysen, H. M., Hong, O. S., & Katz, P. (2019). *The Association Between Yoga Use , Physical Function , and Employment in Adults With Rheumatoid Arthritis.* 71–79. <https://doi.org/10.1097/HNP.0000000000000296>
- Haaz, S., & Bartlett, S. J. (2011). *Yog a fo r Ar th r it is : A Scoping Review. Rheumatic Disease Clinics of NA,* 37(1), 33–46. <https://doi.org/10.1016/j.rdc.2010.11.001>

- Hegde, S. V., Adhikari, P., Shetty, S., Manjrekar, P., & Souza, V. D. (2013). Effect of community-based yoga intervention on oxidative stress and glyceimic parameters in prediabetes : A randomized controlled trial &. *Complementary Therapies in Medicine*, 21(6), 571–576. <https://doi.org/10.1016/j.ctim.2013.08.013>
- Kusumayanti, G. A. D., Dewantari, N. M., & Author, C. (2017). *The Influence of Low Purine Diet and Physical Activity on Changing of Uric Acid Levels in Hyperuricemia*. 1(3).
- Lauche, R., Hunter, D. J., Adams, J., & Cramer, H. (2019). *Yoga for Osteoarthritis : a Systematic Review and Meta-analysis*.
- Lazo, M., Sc, M., & Clark, J. M. (2008). *The Epidemiology of Nonalcoholic Fatty Liver Disease : A Global Perspective*. <https://doi.org/10.1055/s-0028-1091978>.
- Lazo, M., Selvin, E., & Clark, J. M. (2008). Brief communication: Clinical implications of short-term variability in liver function test results. *Annals of Internal Medicine*, 148(5), 348–352. <https://doi.org/10.7326/0003-4819-148-5-200803040-00005>
- Muraoka, S., & Miura, T. (2003). Inhibition by Uric Acid of Free Radicals that Damage Biological Molecules. *Pharmacology and Toxicology*, 93(6), 284–289. <https://doi.org/10.1111/j.1600-0773.2003.pto930606.x>
- Pavletic, A. J., Pao, M., & Wright, M. E. (2015). Letters to the Editor Exercise-induced Elevation of Liver Enzymes in a Healthy Female Research Volunteer. *Psychosomatics*, 56(5), 604–606. <https://doi.org/10.1016/j.psych.2015.03.002>
- Questionnaire, S., & Affect, N. (n.d.). *Stress, Inflammation, and Yoga Practice*. (22), 113–121. <https://doi.org/10.1097/PSY.0b013e3181cb9377>
- Ranade, A. (2015). *Concept of Rasayana w . s . r Haritakyadi Yoga as stated in Charaka Samhita International Journal Of Ayurvedic And Herbal Medicine 3 : 1 (2013) 1057 : 1064 Concept of Rasayana w . s . r Haritakyadi Yoga as stated in Charaka Samhita .*

(January 2013).

Randomized, A., & Study, C. P. (2015). *Immune Function , and Stress Hormone Releases in Young Healthy People : 00(0)*, 1–9. <https://doi.org/10.1089/acm.2014.0044>

Ray, U. S. (2007). *Improvement of Glutathione and Total Antioxidant Status with Yoga. 13(10)*, 1085–1090. <https://doi.org/10.1089/acm.2007.0567>

Roche, M., Rondeau, P., Singh, N. R., Tarnus, E., & Bourdon, E. (2008). *The antioxidant properties of serum albumin. 582*, 1783–1787. <https://doi.org/10.1016/j.febslet.2008.04.057>

Samhita, C. (2001). Charaka samhita. *Vimana Sthana. 8th*, 14.

Study, P. (2014). *adolescents : the HELENA study. 75(6)*. <https://doi.org/10.1038/pr.2014.26>

Susruta, A. (2008). Susruta Samhita. *Uttar tantra, 56(6)*, 413.

Wruck, C. J., Fragoulis, A., Gurzynski, A., Brandenburg, L., Kan, Y. W., Chan, K., ... Pufe, T. (2011). *Role of oxidative stress in rheumatoid arthritis : insights from the Nrf2-knockout mice*. <https://doi.org/10.1136/ard.2010.132720>

Wunder, A., Muller-Ladner, U., Stelzer, E., Neumann, E., Sinn, H., Gay, S., & Fiehn, C. (2003). Albumin-based drug delivery as novel therapeutic approach for rheumatoid arthritis. *Arthritis Res Ther, 5(3)*, 9. <https://doi.org/10.1186/ar810>

Yadav, R. K., Magan, D., Mehta, N., Sharma, R., & Mahapatra, S. C. (2012). *Efficacy of a Short-Term Yoga-Based Lifestyle Intervention in Reducing Stress and Inflammation : 18(7)*, 662–667. <https://doi.org/10.1089/acm.2011.0265>