Original Article

Research Design for a Randomized Control Trial to Assess the Effects of Almond Supplementation on Insulin Resistance, Glycemic Markers, and Inflammation Among Overweight Asian Indians

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Abstract

Background: Fatty acids play an important role in health and well-being; almonds have the highest amount of monounsaturated fatty acids (MUFAs) among the nuts. Western studies have shown positive health effects of almonds. However, well-designed studies are sparse on Asian Indians who have a unique phenotype with higher predisposition to diabetes and cardiovascular disease (CVD). Hence, the present study describes the design and methods of a clinical trial to assess the effect of almond supplementation on insulin resistance, glycemic markers, and inflammation in overweight Asian Indians. **Methods and Outcome Assessments:** Parallel-arm open-labeled, randomized controlled trial was conducted in Chennai, India. The study included 400 overweight and obese volunteers of age 25–65 years with a body mass index ≥23 kg/m^2 and with some having cardiometabolic risks. The participants in the intervention group received 43 g of almonds per day as recommended by the American Heart Association for 12 weeks, whereas the participants in the control arm followed their habitual dietary patterns and were advised not to consume any nuts. All other lifestyle habits were similar. The anthropometric, clinical, biochemical, and diet data of the participants were assessed periodically. Dietary 24-hour recalls and plasma percent fatty acid of the participants were assessed at the baseline and end of the study as a measure of participant compliance to protocol. This study also assessed gut hormone levels as a marker for satiety. The effects of almonds supplementation on anti-inflammatory and inflammatory markers such as adiponectin, monocyte chemoattractant protein-1, and tumor necrosis factor-α were also assessed. **Discussion:** The study findings, if benefits are found, would help to improve the MUFAs intake by a single supplementation of almonds daily to meet the dietary guidelines of 15% of total calories of MUFAs. In addition, it might aid in the prevention of obesity-related chronic diseases such as diabetes and CVDs by reducing the cardiometabolic risk factors. **Trial Registration:** The trial was registered in the clinical trial registry of India CTRI201710010251.

Keywords: Almonds, insulin resistance, insulin sensitivity, MUFA, obesity

BACKGROUND

Consumption of nuts, a rich source of monounsaturated fatty acids (MUFAs), has historically been low in India (8 g/day in 1975–1979 and 17 g/day in 1996–1997).[1] Various

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clinical trials on nut consumption have shown health benefits in improving the overall metabolic outcome. Mediterranean diets enriched with mixed nuts (30 g/day) have shown to be helpful in the management of metabolic syndrome in the Prevención con Dieta Mediterránea study.[2] Our recent randomized controlled trial (RCT) in Asian Indians with type 2 diabetes (T2DM) on cashew nut supplementation has shown improved systolic blood pressure and high-density lipoprotein cholesterol (HDL-c) levels with no significant deleterious effects on body weight, glycemic, or other lipid parameters.[3]

The prevalence of diabetes and prediabetes continues to be on the rise. Currently, 463 million people in the world have diabetes and of this 77 million people with diabetes reside in India[4] and 80 million people in India have prediabetes.[5] Asian Indian phenotype and thrifty genotype combined with altered lifestyles (poor dietary practices and physical inactivity) are the key drivers for the escalating prevalence of obesity further fuelling the diabetes epidemic in India.[6,7] Suboptimal diet is attributed to more deaths globally even when compared to smoking. Good quality of a diet can prevent one in every five deaths.[8]

Diets high in refined grains showed a positive association with insulin resistance, low HDL levels, metabolic syndrome, and T2DM.[9-11] Replacement of refined grains with healthy fats such as MUFAs may have beneficial effects on HDL-c.[3,12] MUFA-rich diet prevented central fat redistribution and the postprandial decrease in peripheral adiponectin gene expression and insulin resistance induced by a carbohydrate-rich diet.[13] Studies have also reported the beneficial role of MUFA intake in the prevention of T2DM.[14] The inclusion of natural whole nuts like almonds offers promise in terms of delivering MUFA in the healthiest form in addition to MUFA-rich cooking oils or as a standalone dietary supplement.

Almonds are good sources of protein (17%), dietary fiber (13%) and fat (58%), and magnesium (318 mg/100g), and remain to be a very rich source of MUFA (38%).[15,16] Almonds have been well studied in the western population and are known to have positive health effects on satiety (appropriate changes in gut hormones), insulin sensitivity, improved glycemic, lipids, and inflammatory markers. Poor dietary practices of Asian Indians are associated with cardiometabolic risks and include a high intake of refined carbohydrates such as white rice, low intake of whole grains, fruits, vegetables, MUFA, and micronutrients.[17,18] Therefore, MUFA-rich food supplements like almonds could offer a strategy to improve MUFA intake in Asian Indians. Very few studies in India have shown the beneficial effects of almonds on cardiometabolic risk factors, dyslipidemia, and glucose responses in T2DM. Almonds have also known to promote the growth of healthy microbiota.[19-21] However, the role of almonds on insulin resistance, inflammation, and satiety in Asian Indians especially in overweight and obese individuals, who are at risk for T2DM and cardiovascular disease (CVD), remains unexplored. Studies on the 24-hour glycemic response to nuts (a rich source of MUFA) supplementation adjusted to maintain total calorie intake may provide unique insights about postprandial glycemic excursions especially in "at risk population." Therefore, there is an unmet need to evolve dietary strategies from well-conducted randomized clinical trials to mitigate the health and economic burden of diabetes. Thus, this article explains the study design and methods of a randomized trial to evaluate the effect of 43 g almond supplementation as per the guidelines of the American Heart Association (AHA, 2010) for 12 weeks among overweight and obese adults with the following objectives:

1. **Primary:** To determine the effects of almond supplementation on Homeostasis Model Assessment-insulin resistance (HOMA-IR) and beta-cell function assessed by oral disposition index (DIo), C-peptide levels, and adiposity.
2. **Secondary:** To study the effect on the biomarkers of glucose metabolism (i.e., fasting glucose and hemoglobin A1c), lipid profile (i.e., triglycerides, total cholesterol, low-density lipoprotein cholesterol (LDL-c), and HDL-cholesterol), plasma fatty acid profile, satiety-related gut hormones using a multiplex panel (glucagon-like peptide-1 [GLP-1], gastric inhibitory polypeptide [GIP], peptide YY [PYY], and ghrelin), adiponectin, and selected inflammatory markers (i.e., hs-C reactive protein [hs-CRP], tumor necrosis factor-α [TNF-α], monocyte chemoattractant protein-1 [MCP-1]).

MATERIALS AND METHODS

Study design and ethics approval
The study was a randomized, parallel-arm 12 weeks trial. The participants recruited for the study were randomized either into the control group or the almond (43 g/day) intervention group. The study was approved by the Institutional Ethics Committee at Madras Diabetes Research Foundation (MDRF) and registered in the clinical trial registry of India, reference number CTRI201710010251. The study was conducted in accordance with guidelines in the Declaration of Helsinki.

Study population
Sample size
The sample size was calculated with the primary outcome measure of HOMA-IR with the mean difference of 0.72 considered from the reported studies in literature since data on almond supplementation in Asian Indians were unavailable. The study was planned with 80% power and an alpha of 0.05 to detect a difference of 20% change in

HOMA-IR with 200 participants per group and a dropout rate of 20%.

Participant recruitment and consent

The present study was conducted among 400 overweight/obese Asian Indian adults selected from the volunteer registry maintained at our research center, the MDRF. Potential participants were invited for screening at our center and interviewed by trained study staff who explained the background, objectives, and procedures of the trial, sought and obtained written informed consent, confirmed eligibility, and completed the baseline assessments. Participants then completed a baseline glucose tolerance test to rule out any new diabetes. Eligible participants meeting the inclusion and exclusion criteria were recruited for the study.

Inclusion and exclusion criteria employed to screen participants were as follows:

Inclusion criteria

- 25–60 years (men and women)
- Body mass index (BMI) $\geq 23\,\mathrm{kg/m^2}$
- Central obesity (waist circumference $\geq 90\,\mathrm{cm}$ for men and $\geq 80\,\mathrm{cm}$ for women)
- Dyslipidemia – low HDL ($\leq 40\,\mathrm{mg/dl}$ men and $50\,\mathrm{mg/dl}$ women) and (or) high triglyceride ($\geq 150\,\mathrm{mg/dl}$)
- Positive family history of diabetes
- Hypertension ($\geq 140/90\,\mathrm{mmHg}$)
- Snacking (mid-morning)

Exclusion criteria

- Self-reported diabetes
- History of allergy to nuts
- Use of nutritional supplements
- Bad dentures
- Weight loss diets
- Pregnant/lactating women
- Liver, kidney, thyroid, or other endocrine diseases
- Any condition that hinders compliance to the intervention
- Possible eating disorder

Randomization

After screening, randomization was done centrally by an independent biostatistician using a computer-generated random number sequence. Participants were randomized with stratification by day-to-day screening based on a 1:1 allocation ratio to either the intervention or to the control group.

Intervention and control group

Participants assigned to the intervention group were given sachets containing ≈43 g of raw almonds daily for 12 weeks. A 1-week run-in period was conducted before intervention to ensure participants' acquiescence to the study protocol. The participants in the intervention group were advised to adjust the almond calories for either carbohydrate or fat calories in their routine standard Indian diets by trained research dietitians. Further, the participants were instructed not to share the almonds with family members, and to ensure this, per month 200 g of almonds were provided for their family members. To preserve the quality and freshness of almonds, sufficient sachets of almonds were provided to the participants monthly once. Table 1 depicts the nutrient composition of almonds. Almonds are rich in MUFAs (38 g/100 g), dietary fiber (13 g/100 g), and calcium (228 mg/100 g), magnesium (318 mg/100g), vitamin E (13 g/100 g) and have minimal carbohydrate (3 g/100 g) content. This is especially important in the Indian context wherein the diets are high in carbohydrates and deficient in MUFA and fiber. The present study was carried out using the nonpareil almond variety which is the most common and popular form of almonds known for its versatility and texture. The nutrient composition of nonpareil versus other common almond varieties is given in Table 2. The nutrient content of nonpareil almonds was more or less similar to the other almond varieties.

Participants in the control group continued with their usual diet and were further instructed to refrain from intake of nuts in any form during the 12-week study period. All the participants recruited for the study were advised to refrain from fasting and feasting and also requested not to make any major lifestyle modifications during the study period.

Table 1: Selected nutrition profile of almond

S. No.	Nutrients	Almond (*Prunusamygdalus*)
1	Energy (kcal)	609 ± 1
2	Carbohydrate (g)	3.04 ± 0.24
3	Protein (g)	18.41 ± 0.04
4	Fat (g)	58.49 ± 0.04
5	Total SFA (g)	4.4 ± 0.2
6	Total MUFA (mg)	38.3 ± 0.16
7	Total PUFA (mg)	13.2 ± 0.15
8	Dietary fibre (g)	13.06 ± 0.31
9	Calcium (mg)	228 ± 10.2
10	Iron (mg)	4.59 ± 0.61
11	Magnesium (mg)	318 ± 49.5
12	Potassium (mg)	699 ± 43.4
13	Selenium (µg)	3.61 ± 1.30
14	Sodium (mg)	1.50 ± 0.51
15	Zinc (mg)	3.50 ± 0.10
16	Moisture (g)	4.37 ± 0.31
17	Total polyphenols (mg)	84.94 ± 1.88

Source: Longvah T, Anantan I, Bhaskarachary K, Venkaiah K, Longvah T. Indian food composition tables. Hyderabad: National Institute of Nutrition, Indian Council of Medical Research; 2017 May.

Table 2: Nutrient profile of nonpareil almond vs other almond varieties							
Nuts	Fritz	Mission	Carmel	Butte	Monterey	Nonpareil	Sonora
Moisture (g)	4.6	4.5	3.6	4.7	3.9	4.0	4.1
Protein (g)	22.5	22.1	20.4	20.5	21.3	20.6	22.4
Fat (g)	48.4	46.9	48.8	50.0	49.4	46.5	50.2
Total dietary fiber (g)	11.0	13.5	12.5	12.2	11.8	12.9	11.8
Carbohydrate (g)		23.6	25.0				
Calcium (mg)	290.0	330.0	279.0	288.0	252.0	261.0	234.0
Iron (mg)	3.6	3.3	3.3	3.3	3.6	3.5	3.8
Magnesium (mg)	260.0	272.0	262.0	263.0	278.0	275.0	256.0
Potassium (mg)	664.0	724.0	679.0	664.0	766.0	762.0	773.0
Zinc (mg)	2.8	2.8	2.8	3.0	2.8	3.2	3.8
Total saturated fatty acids (g)	3.4	3.7	3.9	4.1	3.7	3.8	3.9
Total Monounsaturated fatty acids (mg)	30.5	31.6	29.7	29.4	32.3	31.3	31.4
Total polyunsaturated fatty acids (mg)	12.0	11.6	13.8	13.9	11.2	11.7	12.4

Source: Yada, S.; Huang, G.; Lapsley, K. Natural variability in the nutrient composition of california-grown almonds.
J. Food Compos. Anal. **2013**, 30, 80–85.

Data collection and follow-up
Anthropometric and clinical assessments
Bodyweight (kg) (electronic OMRON machine; Omron HBF 212, Tokyo, Japan), and waist circumference (cm) were measured using standard protocols. BMI was calculated as weight (kg) divided by height squared (m²). Blood pressure was measured twice on each occasion with an interval of 5 minutes using an electronic OMRON machine (Omron HEM 7120, Tokyo, Japan). Anthropometric and clinical assessments were done at baseline and followed up monthly once during the study period.

Biochemical assessments
Glucose tolerance test (0, 30, and 120), HbA1c, fasting insulin, hs-CRP, and lipid profile were assessed on the first day of biochemical assessments. In addition, after 2 days interval another blood sample was collected at fasting for assessment of inflammatory markers such as TNF-α, MCP-1, adiponectin, gut hormones such as GLP-1, GIP, PYY, and ghrelin and post-standardized breakfast blood samples were collected for assessment of GLP-1 (four pointers: 0, 30, 60, and 120 min), and postprandial C-peptide. The plasma fatty acid profiles of the participants were also measured as a marker of compliance at baseline and post-intervention with almonds for 12 weeks. All the specified biochemical assessments were done at baseline and end of 12 weeks study period. In addition, fasting blood glucose and fasting insulin were assessed at midpoint of the study (45 days). The blood samples were collected by a trained phlebotomist.

Plasma glucose was estimated using the glucose oxidase–peroxidase method (Roche Diagnostics, Basel, Switzerland). Glycemic and lipid profiles were assessed using a Hitachi 912 Autoanalyzer (Hitachi, Mannheim, Germany). HbA1c was estimated by high-pressure liquid chromatography using the Variant machine (Bio-Rad, Hercules, and CA), and

serum insulin by electrochemiluminescence assay (Roche Diagnostics, Basel, Switzerland). The β cell function has been evaluated using DIo, which is a measure of ratio of the difference in fasting to 30-min insulin (I) to glucose (G) values = $(\delta I0–30/\delta G0–30)$, and insulin resistance calculated by the HOMA-IR using the formula: fasting insulin (uIU/ml) × fasting glucose (mmol/l)/22.5.

Serum cholesterol (CHOD-PAP method), serum triglycerides (GPO-PAP method), and HDL cholesterol (Direct method–polyethylene glycol-pre-treated enzymes) have been measured, and LDL cholesterol calculated using the Friedwald formula. The coefficients of variation for the biochemical assays ranged from 3.1% to 7.6%. Gut hormones and inflammatory markers assessments were done using Enzyme-Linked Immunosorbent Assays. Plasma fatty acid was assessed using gas chromatography, lipids were extracted in chloroform–methanol mixture and converted to fatty acid methyl esters for quantification of plasma fatty acids using gas chromatography. Appropriate fatty acid methyl ester standards were used for the identification and quantification of peaks.

Diet and lifestyle
Data collection was done by trained research staff and baseline demographics, medical history, and lifestyle factors of the participants were assessed using a screening questionnaire. Participants' physical activity (PA) level was assessed at baseline and end of every month using a validated PA questionnaire. Dietary 24-hour recalls were administered during screening, run-in period, at baseline blood collection, and monthly twice (1 weekday and 1 weekend) during the study. Further, adverse event and satiety score questionnaires were assessed at baseline and at the end of every month.

Participants' compliance to the study protocol was assessed at three levels: (1) return of empty sachet of almonds collected at end of every month, (2) change in the

nutrient profile of the participants from 24-hour dietary recall data, and (3) change in plasma fatty acid levels

The study parameters and timeline of the assessments are shown in Table 3 and Figure 1.

Statistical analysis

Statistical analysis was performed using SAS software, version 9.0 (SAS Institute Inc., Cary, NC). Between-group differences for change in health measurements and dietary intake between baseline and 12 weeks will be analyzed using generalized linear models. The outcome data will be controlled for baseline outcome as a sensitivity analysis. The baseline demographic, biochemical, clinical, and dietary data of participants recruited for the study are reported as mean ± SD.

RESULTS

The proposed trial was designed as an intervention trial with almonds that are of late commonly consumed in different parts of India.

Table 3: Study protocol assessments and schedule

Description	Pre-study visits			During the study visits							
				Visits							
	1	2		3	4	5	6	7	8	9	10
		End of run-in period		Day 0	Day 1	Day 14	Day 30	Day 45	Day 75	Days 88–89	Days 91–92
Screening^	√		Washout period – 2 weeks								
Informed consent	√										
Almond supply for run-in period	√										
Adverse effect Questionnaire		√					√	√		√	
Compliance Questionnaire		√									
Blood pressure				√			√	√		√	
Anthropometry assessments-Height, weight, body fat, waist circumference, and BMI				√			√	√		√	
Dietary 24-hour Recall	√			√	√		√	√		√	
Physical activity Questionnaire				√			√	√		√	
Satiety Score		√					√	√		√	
Glucose tolerance test (0, 30, and 90 min)				√							√
Serum insulin (0, 30, and 90 min) (µIU/ml)				√							√
AGP assessments				√$	√#				√$		√#
Fasting plasma glucose (mg/dl)								√			
Fasting Insulin (µIU/ml)								√			
HOMA-IR				√				√			√
Oral disposition index (DIo)				√							√
Glycosylated hemoglobin (HbA1c%)				√							√
Triglycerides (mg/dl)				√							√
Total cholesterol (mg/dl)				√							√
HDL cholesterol (mg/dl)				√							√
LDL cholesterol (mg/dl)				√							√
Apo A (mg/dl)				√							√
Apo B (mg/dl)				√							√
hs-CRP (mg/l)				√							√
TNF-α (pg/ml)				√							√
MCP-1 (pg/ml)				√							√
Adiponectin (µg/ml)				√							√
Standard iso-caloric breakfast 1.*C Peptide (2 pointer: 0, and 120 min) 2.*Gut hormones panel (GLP-1 (4 pointer- 0, 30, 60 and 120 min, GIP, CCK, PYY and ghrelin)				√						√	
Participants perception (Feed Back)											√

BMI = body mass index, DIo = oral disposition index, HOMA-IR = Homeostasis Model Assessment insulin resistance, hs-CRP = hs-C reactive protein, GLP-1 = glucagon-like peptide-1, GIP = gastric inhibitory polypeptide, HDL-c = high-density lipoprotein cholesterol, LDL-c = low-density lipoprotein cholesterol, MCP-1 = monocyte chemoattractant protein-1, PYY = peptide YY, TNF-α = tumor necrosis factor-α.
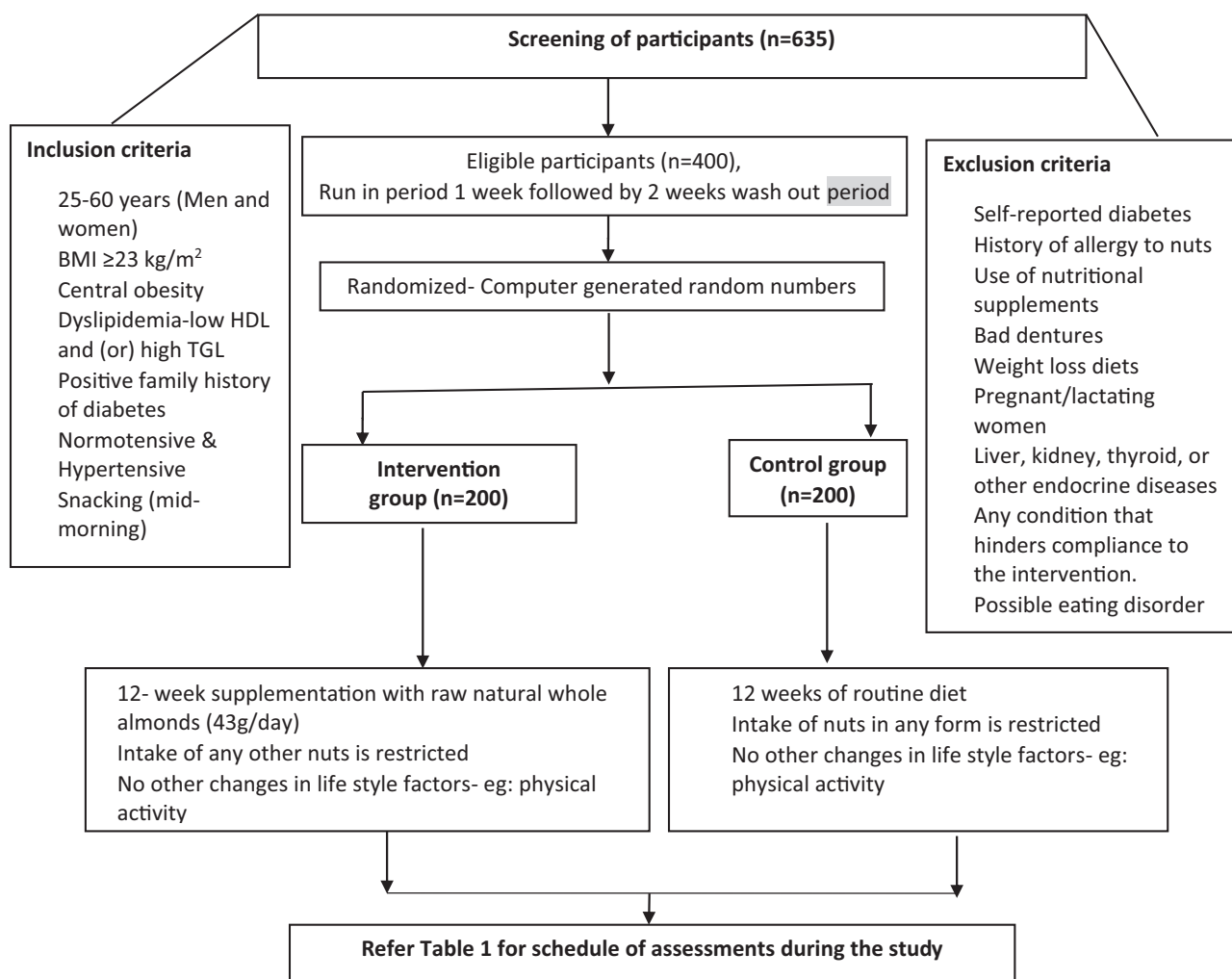
Figure 1: Study design.

Participants (*n* = 635) willing to participate in the study were screened and underwent an oral glucose tolerance test. Participants with newly diagnosed diabetes, not willing to participate, refused to give a blood sample and wear flash glucose monitors, etc (*n* = 235) were excluded. Participants (*n* = 400) were then assigned to intervention group (*n* = 200) and control group (*n* = 200) using computer-generated randomized numbers.

The baseline characteristics of the participants recruited for the study are given in Table 4. The mean age of the participants recruited for the study was 37± 9 years. About 68% of participants in the control group and 57% in the intervention group were women. The systolic blood pressure, 30 min and 120 min insulin assays were observed to be higher in the intervention group compared with the control group.

The baseline dietary characteristics of the study participants based on an average of three dietary recalls (24 hours) collected are shown in Table 5. The total energy, carbohydrate, protein, fat, and dietary fiber in the intervention group were higher, whereas the calorie from carbohydrate was lower compared with control group.

DISCUSSION

This study successfully recruited 400 participants suggesting that the approach developed can efficiently and rapidly recruit participants at risk for developing cardiometabolic disorders from the urban population in India to evaluate the effect of almond supplementation on insulin resistance, glycemic markers, and inflammation among overweight Asian Indian adults. The evaluation of the acceptability and adherence to long-term almond supplementation depended on the complete, unbiased follow-up, and outcome assessment of participants during the study period.

This study was designed to encourage future RCT that can be conducted for longer terms and in greater strengths of cohorts that will test the effect of almond supplementation in a more diverse population with

Table 4: Demographic, anthropometric, and clinical characteristics of the study participants at baseline (*n* = 400)		
Variables	Intervention group $ (*n* = 200)	Control group (*n* = 200)
Age (years)	37.6 ± 9.2	37.8 ± 8.0
Sex		
Male, n (%)	86.0 (43.0)	65.0 (32.5)
Female, n (%)	114.0 (57.0)	135.0 (67.5)
Weight (kg)	75.0 ± 10.4	71.9 ± 11.9
BMI (kg/m²)	28.7 ± 3.7	28.2 ± 3.8
WC (cm)	94.7 ± 9.6	93.0 ± 10.5
SBP (mmHg)	115.7 ± 13.0	114.7 ± 14.2
DBP (mmHg)	78.4 ± 9.2	77.8 ± 9.7
FBS (mg/dl)	94.3 ± 11.1	94.5 ± 12.1
HOMA IR	3.3 ± 1.7	3.3 ± 1.8
DIo (mmol/L)	3.1 ± 4.4	3.1 ± 4.7
Insulin assay (0 mins) μIU/ml	14.0 ± 6.4	14.1 ± 6.9
Insulin assay (30 mins) μIU/ml	114.0 ± 67.2	109 ± 71.5
Insulin assay (120 mins) μIU/ml	92.1 ± 85.2	72.8 ± 67.2
HbA1c (%)	5.5 ± 0.4	5.6 ± 0.4
C-reactive protein mg/l	4.2 ± 3.1	4.0 ± 3.1
C-peptide (0 min) pmol/ml	0.9 ± 0.3	1.0 ± 0.4
C-peptide (120 min) pmol/ml	2.3 ± 1.0	2.4 ± 1.0
PAL	1.5 ± 0.1	1.6 ± 0.2
Adiponectin (μg/ml)	0.89 ± 8.2	0.24 ± 2.0
Tumor necrosis factor alpha (pg/ml)	418.2 ± 425	421.5 ± 389.9
Monocyte chemoattractant protein-1 (pg/ml)	5.6 ± 3.6	3.2 ± 3.6

BMI = body mass index, DBP = diastolic blood pressure, DIo = oral disposition index, FBS = fasting blood sugar, HOMA-IR = Homeostasis Model Assessment insulin resistance, PAL = physical activity level, SBP = systolic blood pressure, WC = waist circumference.

^Data presented as mean ± SD.

$Raw almond 43 g/day (intervention) for 12 weeks.

Table 5: Dietary characteristics of the study participants at baseline (*n* = 400)		
Descriptive	Intervention group	Control group
Total energy (kcal)	1619 ± 477	1560 ±442
Carbohydrates (g)	242.5 ± 69	237.4 ± 64.6
Protein (g)	48.9 ± 16.3	46.7 ± 15.1
Total fat (g)	48.1 ± 19.2	44.8 ± 18.4
Total saturated fatty acid (g)	15 ± 9	15 ± 9.2
Total mono unsaturated fatty acid (g)	12.2 ± 5.1	11.5 ± 5
Total poly unsaturated fatty acid (g)	17.4 ± 7	16.3 ± 6.8
Total dietary fibre (g)	25.6 ± 8.6	24.1 ± 8.5
Carbohydrate (%E)	60.6 ± 5.3	61.8 ± 5.4
Protein (%E)	12 ± 1.5	11.9 ± 1.5
Total fat (%E)	25.9 ± 4.8	24.9 ± 5.2
Total saturated fatty acid (%E)	8.2 ± 3.1	8.3 ± 3.3
Total monounsaturated fatty acid (%E)	6.6 ± 1.5	6.4 ± 1.6
Total polyunsaturated fatty acid (%E)	9.4 ± 2.1	9 ± 2.2
Total linoleic acid (%E)	89.2 ± 21.5	86.6 ± 22.9
Total linolenic acid (%E)	2.0 ± 0.8	1.9 ± 0.6

respect to socio-demographical differences. Evidence-based medicine seeks the best use of available evidence in making sound clinical decisions while caring for patients. Only RCTs offer the kind of confirmation for dietary suggestion better than observational study designs.[22] RCTs have proven that medical nutrition therapy is beneficial and cost-effective in improving the metabolic outcomes of patients.[23] There is a growing recognition that nutrition science will benefit from more adequately powered and well-conducted RCTs to evaluate the health effects of nutrients and foods on clinical outcomes, which can complement findings from prospective cohorts

and metabolic studies. These research approaches have different strengths and weaknesses, and the consistency of findings can significantly enhance the development of dietary guidelines and their effective implementation.[24] In a country like India where CVD and diabetes are known to increase disability-adjusted life years,[25] it is important to establish nutrition interventions, which will aid in the prevention of the disease especially through its effects on the predominant modifiable risk factor dyslipidemia and nuts consumption falls in line with this need of the hour.[26]

Dietary guidelines have been developed for Asian Indians for the prevention of obesity and dyslipidemia and include reduction in the intake of carbohydrates, preferential intake of complex carbohydrates and low glycemic index foods, higher intake of fiber, lower intake of saturated fats, the optimal ratio of essential fatty acids, and reduction of trans fatty acids.[27,28] Besides this, the Chennai Urban Rural Epidemiological Studies identified the combination of obesity, physical inactivity, unfavorable diet risk score, hypertriglyceridemia, and low-HDL cholesterol could explain 80.7% of all incident diabetes and concluded that modifying these easily identifiable risk factors could therefore prevent the majority of cases of non-communicable diseases beginning with diabetes.[9] Therefore, almonds which are a rich source of minerals like calcium, copper, iron, magnesium, phosphorus, potassium, zinc, manganese, and vitamins such as thiamine, riboflavin, niacin, and vitamin E with their healthy fatty acid composition are greatly beneficial in mitigating the risks of CVDs by reducing the levels of LDL-C and triglycerides while improving HDL levels.[26] Besides these, omega-3 fatty acids, especially alpha-linolenic acid, which are present in almonds reduce CVD risk, by changing vascular inflammation and improving endothelial dysfunction[29] and the authors also pointed out that sufficient studies to prove the efficacy of nuts especially almonds and walnuts were sparse. Therefore, design and execution of this study seem to contribute science-based pieces of evidence for the prevention of non-communicable disease through simple lifestyle changes like consuming almonds as a potent dietary supplement.

There are several notable strengths of the study. First, the study 2-arm RCT allowed testing for a potential effect of almond consumption. Second, the sample size of the study provides a piece of stronger evidence compared with previously conducted RCT on almond supplementation. Third, using inflammatory markers such as hs-CRP, TNF-α, MCP-1; adiponectin, gut hormones- GLP-1, GIP, PYY, and ghrelin and plasma fatty acids to assess compliance minimized the risk of measurement bias and errors associated with self-reported dietary intake.

The limitations of the study include

1. It was not possible to blind the participants to their randomized group.
2. Long-term effects may not be evaluated as this study is up to 12 weeks of supplementation in a community trial.
3. The baseline plasma fatty acid and gut hormones are yet to be analyzed
4. Though we advised the participants to adhere to the study protocol and not to make major lifestyle and dietary changes, in a community-based trial such as this, family and environmental influences (like festivals, marriage feasts, and parties) may have had some impact on the outcomes.
5. These effects would be likely to occur in both groups, biasing results toward the null.

CONCLUSION

The study results if found to be positive data could help to devise dietary strategies which can potentially reduce insulin resistance even among overweight and obese adults and thereby help prevent diabetes. Public health message to improve the MUFA intake by a single supplementation of almonds daily which would help Asian Indians not only to meet the dietary guidelines of 15% of total calories of MUFA, but also aids in prevention of obesity-related chronic diseases such as diabetes and CVDs.

Authors contribution
GR initiated the article, KN supported in data collection and editing the article. VSM assisted in writing article. VS, SS, RGJ, RMA, RU, GK, KK, AB, GP, LMJ, JSS, WW, and VM reviewed and edited the article.

Conflicts of interest
There are no conflicts of interest.

REFERENCES

1. Vijayaraghavan K, Venkaiah K, Damayanthi K, Nayak MU. NNMB Report on diet & nutritional status of adolescents. Report No. 20. National Institute of Nutrition, India 2000. p. 59-62.
2. Salas-Salvadó J, Fernández-Ballart J, Ros E, Martínez-González MA, Fitó M, Estruch R, *et al.*; PREDIMED Study Investigators. Effect of a mediterranean diet supplemented with nuts on metabolic syndrome status: One-year results of the PREDIMED randomized trial. Arch Intern Med 2008;168:2449-58.

3. Mohan V, Gayathri R, Jaacks LM, Lakshmipriya N, Anjana RM, Spiegelman D, *et al*. Cashew nut consumption increases HDL cholesterol and reduces systolic blood pressure in Asian Indians with type 2 diabetes: A 12-week randomized controlled trial. J Nutr 2018;148:63-9.

4. Atlas D. International Diabetes Federation. IDF Diabetes Atlas. 9th ed. Brussels: International Diabetes Federation; 2020.

5. Anjana RM, Pradeepa R, Deepa M, Datta M, Sudha V, Unnikrishnan R, *et al*.; ICMR–INDIAB Collaborative Study Group. Prevalence of diabetes and prediabetes (impaired fasting glucose and/or impaired glucose tolerance) in urban and rural India: Phase I results of the Indian Council of Medical Research-India Diabetes (ICMR-INDIAB) study. Diabetologia 2011;54:3022-7.

6. Unnikrishnan R, Anjana RM, Mohan V. Diabetes in south Asians: Is the phenotype different? Diabetes 2014;63:53-5.

7. Mohan V, Sandeep S, Deepa R, Shah B, Varghese C. Epidemiology of type 2 diabetes: Indian scenario. Indian J Med Res 2007;125:217-30.

8. Afshin A, Sur PJ, Fay KA, Cornaby L, Ferrara G, Salama JS, *et al*. Health effects of dietary risks in 195 countries, 1990–2017: A systematic analysis for the Global Burden of Disease Study 2017. The Lancet 2019;393:1958-72.

9. Anjana RM, Sudha V, Nair DH, Lakshmipriya N, Deepa M, Pradeepa R, *et al*. Diabetes in Asian Indians: How much is preventable? Ten-year follow-up of the Chennai Urban Rural Epidemiology Study (CURES-142). Diabetes Res Clin Pract 2015;109:253-61.

10. Narasimhan S, Nagarajan L, Vaidya R, Gunasekaran G, Rajagopal G, Parthasarathy V, *et al*. Dietary fat intake and its association with risk of selected components of the metabolic syndrome among rural south Indians. Indian J Endocrinol Metab 2016;20:47-54.

11. Sowmya N, Lakshmipriya N, Arumugam K, Venkatachalam S, Vijayalakshmi P, Ruchi V, *et al*. Comparison of dietary profile of a rural south Indian population with the current dietary recommendations for prevention of non-communicable diseases (CURES 147). Indian J Med Res 2016;144:112-9.

12. Li SC, Liu YH, Liu JF, Chang WH, Chen CM, Chen CY. Almond consumption improved glycemic control and lipid profiles in patients with type 2 diabetes mellitus. Metabolism 2011;60:474-9.

13. Paniagua JA, Gallego de la Sacristana A, Romero I, Vidal-Puig A, Latre JM, Sanchez E, *et al*. Monounsaturated fat-rich diet prevents central body fat distribution and decreases postprandial adiponectin expression induced by a carbohydrate-rich diet in insulin-resistant subjects. Diabetes Care 2007;30:1717-23.

14. Thomas T, Pfeiffer AF. Foods for the prevention of diabetes: How do they work? Diabetes Metab Res Rev 2012;28:25-49.

15. Longvah T, Ananthan R, Bhaskarachary K, Venkaiah K. Indian food composition tables 2017. Hyderabad: National Institute of Nutrition; 2017.

16. Yada S, Huang G, Lapsley K. Natural variability in the nutrient composition of California-grown almonds. J Food Compos Anal 2013;30:80-5.

17. Lakshmipriya N, Gayathri R, Praseena K, Vijayalakshmi P, Geetha G, Sudha V, *et al*. Type of vegetable oils used in cooking and risk of metabolic syndrome among Asian Indians. Int J Food Sci Nutr 2013;64:131-9.

18. Radhika G, Sumathi C, Ganesan A, Sudha V, Jeya Kumar Henry C, Mohan V. Glycaemic index of Indian flatbreads (rotis) prepared using whole wheat flour and 'atta mix'-added whole wheat flour. Br J Nutr 2010;103:1642-7.

19. Gulati S, Misra A, Pandey RM. Effect of almond supplementation on glycemia and cardiovascular risk factors in Asian Indians in north India with type 2 diabetes mellitus: A 24-week study. Metab Syndr Relat Disord 2017;15:98-105.

20. Beatrice DA, Shivaji G. Effect of almond supplementation on the anthropometric measurements, biochemical parameters and blood pressure levels of men with metabolic syndrome. Indian J Nutr Diet 2015;52:184-91.

21. Dreher ML. A comprehensive review of almond clinical trials on weight measures, metabolic health biomarkers and outcomes, and the gut microbiota. Nutrients 2021;13:1968.

22. Ahuja AS. Should RCT'S be used as the gold standard for evidence based medicine? Integr Med Res 2019;8:31-2.

23. Pastors JG, Warshaw H, Daly A, Franz M, Kulkarni K. The evidence for the effectiveness of medical nutrition therapy in diabetes management. Diabetes Care 2002;25:608-13.

24. Liu Y, Li N, Yan N, Pan XF, Li Q, Micha R, *et al*. Protocol for a randomized controlled trial to test the acceptability and adherence to 6-months of walnut supplementation in Chinese adults at high risk of cardiovascular disease. Nutr J 2021;20:3.

25. Vos T, Lim SS, Abbafati C, Abbas KM, Abbasi M, Abbasifard M, *et al*. Global burden of 369 diseases and injuries in 204 countries and territories, 1990–2019: A systematic analysis for the Global Burden of Disease Study 2019. The Lancet 2020;396:1204-22.

26. Kalita S, Khandelwal S, Madan J, Pandya H, Sesikeran B, Krishnaswamy K. Almonds and cardiovascular health: A review. Nutrients 2018;10:468.

27. Misra A, Sharma R, Gulati S, Joshi SR, Sharma V, Ghafoorunissa, *et al*.; National Dietary Guidelines Consensus Group. Consensus dietary guidelines for healthy living and prevention of obesity, the metabolic syndrome, diabetes, and related disorders in Asian Indians. Diabetes Technol Ther 2011;13:683-94.

28. Misra A, Shrivastava U. Obesity and dyslipidemia in South Asians. Nutrients 2013;5:2708-33.

29. Bhardwaj R, Dod H, Sandhu MS, Bedi R, Dod S, Konat G, *et al*. Acute effects of diets rich in almonds and walnuts on endothelial function. Indian Heart J 2018;70:497-501.