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Effect of a Walnut Diet on Office and 24-Hour Ambulatory Blood Pressure in Elderly Individuals

Findings From the WAHA Randomized Trial

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Abstract—Nut consumption lowers blood cholesterol and is associated with reduced cardiovascular disease, but effects on blood pressure (BP) are inconsistent. We assessed the 2-year effects of a walnut diet versus a control diet on office BP and 24-hours ambulatory BP in free-living elders participating in the Walnuts and Healthy Aging study, a randomized trial testing the effects of walnuts at ≈15% energy on age-related disorders. In a prespecified analysis, we enrolled 305 participants, of whom 236 (75%) completed the study (65% women; age, 69 years; 60% with mild hypertension). Walnuts were well tolerated, and compliance was >98%. Mean baseline office BP was 128/79 mm Hg. Adjusted changes from baseline in mean office systolic BP were −4.61 mm Hg (95% CI, −7.43 to −1.79 mm Hg) in the walnut group and −0.59 mm Hg (−3.38 to 2.21 mm Hg) in controls (P=0.051). Respective changes in mean systolic 24-hour ambulatory BP were −3.86 mm Hg (CI, −5.45 to −2.26 mm Hg) and −2.00 mm Hg (CI, −3.58 to −0.42 mm Hg; P=0.111). No changes in diastolic BP were observed. In participants in the upper tertile of baseline 24-hour ambulatory systolic BP (>125 mm Hg), mean 2-year systolic 24-hour BP was −8.5 mm Hg (CI, −12 to −5.0 mm Hg) in the walnut group and −2.5 mm Hg (CI, −6.3 to 1.3 mm Hg) in controls (P=0.034). During the trial, participants in the walnut group required less uptitration of antihypertensive medication and had better overall BP regulation than controls. Walnut consumption reduces systolic BP in elderly subjects, particularly in those with mild hypertension.

Clinical Trial Registration—URL: http://www.clinicaltrials.gov. Unique identifier: NCT01634841. **(*Hypertension*. 2019;73:1049-1057. DOI: 10.1161/HYPERTENSIONAHA.118.12766.)** ● **Online Data Supplement**

Key Words: blood pressure ■ clinical trial ■ hypertension ■ nuts ■ nutrition therapy

High blood pressure (BP) and hypertension are major global risk factors for cardiovascular disease (CVD).[1] The rapid pace of population aging in conjunction with the obesity epidemic is associated with a raising prevalence of metabolic disorders, which is expected to increase CVD rates in older populations.[2,3] Given the social and economic burden of treating CVD and related comorbidities, identifying simple strategies to prevent CVD or delay its onset is a major public health concern.[4]

The PREvención con DIeta MEDiterránea (PREDIMED) trial has provided high-level scientific evidence that Mediterranean diets enriched with extra-virgin olive oil or mixed nuts reduce rates of incident CVD, including stroke, in individuals at high cardiovascular risk.[5] A sub-study of that trial showed that, compared with a control diet, both supplemented Mediterranean diets reduced BP as measured with 24-hour ambulatory BP monitoring.[6] Besides the Mediterranean diet, other dietary patterns with high consumption of vegetables, fruit, legumes, nuts, whole grains, dairy products, and seafood and low consumption of total meat, processed meat, and sweets are associated with BP reductions in clinical trials.[7] Among these foods, the consumption of nuts in prospective studies has been strongly and consistently associated with reduced rates of CVD and all-cause mortality.[8,9] Nuts contain a variety of nutrients, including unsaturated fatty acids, fiber, tocopherols, folate, nonsodium minerals (potassium, magnesium, calcium, and selenium) and other bioactives, such as phytosterols and polyphenols.[9] The healthy nutritional components of nuts may help explain the inverse association of their consumption with CVD. Regarding cardiovascular risk factors, there is robust clinical trial evidence that nuts reduce blood cholesterol in a dose-related manner.[10,11] However, nut consumption appears

Received February 1, 2019; first decision February 9, 2019; revision accepted February 20, 2019.

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The online-only Data Supplement is available with this article at https://www.ahajournals.org/doi/suppl/10.1161/HYPERTENSIONAHA.118.12766.

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to have little effect on office BP in clinical trials,[11,12] although 24-hour ambulatory BP monitoring, the gold standard to define normal or abnormal BP values in clinical studies, have not been performed in nut studies using unrestricted-calorie diets.

The WAHA study (Walnuts and Healthy Aging) is a randomized, 2-year clinical trial conducted in free-living elders to evaluate the effects of walnut consumption on cognitive function and retinal health as primary outcomes and on systolic and diastolic office BP and 24-hour ambulatory BP (24-hour systolic and diastolic, and daytime and nighttime BP) as prespecified secondary outcomes in a sub-sample at the Barcelona site.[13] We hypothesized that regular walnut consumption for 2 years would reduce BP in this elderly population. The findings of this WAHA sub-study are reported here.

Material and Methods

The data that support the findings of this study are available from the corresponding author on reasonable request. The WAHA study (ISRNCT01634841) is a dual center (Hospital Clínic, Barcelona, Spain; and Loma Linda University, CA), randomized, parallel-group, observer-blinded, controlled clinical trial aimed to assess whether the consumption of ≈15% of daily energy as walnuts for 2 years would prevent or slow down age-related cognitive decline and macular degeneration compared with a control diet (abstention from walnuts) in older, cognitively healthy individuals aged 65 to 75 years. To expedite recruitment, on April 9, 2014, the age range for inclusion was expanded from 63 to 79 years. The protocol has been reported in detail elsewhere.[13] The present study was conducted only in participants recruited in the Barcelona site, where expertise and 24-hour ambulatory BP measurement devices were available.[6] The study protocol complied with the Declaration of Helsinki, institutional review boards at each center approved the study, and all participants provided written informed consent.

Study Population

Participants were men and women aged 63 to 79 years and free of recent CVD, cancer, or neurodegenerative disorders. Exclusion criteria were: morbid obesity (body mass index ≥40 kg/m^2), uncontrolled diabetes mellitus (HbA1c >8%), uncontrolled hypertension (on-treatment BP ≥150/100 mm Hg), prior stroke or major head trauma, any relevant psychiatric illness, advanced cognitive deterioration (mild cognitive impairment or frank dementia), other neurodegenerative diseases (ie, Parkinson disease), any chronic illness expected to shorten survival, bereavement, allergy to walnuts, and customary use of fish oil, flaxseed oil, or soy lecithin supplements. Thus, by study design, participants were normotensive or had mild hypertension (office BP <150/100 mm Hg) and all were at low cardiovascular risk because of absent comorbidities.

Randomization and Masking

We randomly assigned participants to either the walnut group (WG, consuming ≈15% of daily energy intake as walnuts on top of their habitual diet) or the control group (CG, following their usual diet with abstention from walnuts) using a computerized, web-based, random number table with stratification by sex and age range in a 1:1 ratio. Pairs of individuals (couples, members of the same household, and partners) entering the trial were allocated to the same group using the same stratification criteria. Walnut doses in the WG ranged from 30 to 60 g/d depending on energy requirements. All study clinicians and investigators were blind to participants' intervention group, except for the dietitians in charge of dietary evaluations and walnut supply. All study data were recorded in a dedicated online database developed by Costaisa (Barcelona, Spain).

Procedures During Follow-up

After treatment allocation, participants were scheduled for visits with the study dietitians every 2 months for the duration of the trial. At baseline and 2 years we collected data on medical history, medication use and lifestyle, including dietary habits and physical activity, and performed anthropometric measurements, office BP and 24-hour ambulatory blood pressure monitoring, and urinary albumin excretion determinations. Throughout the duration of the study, participants were under the care of their primary care physicians, who maintained or changed medications, including antihypertensive drug treatment, according to their evaluation of risk factor levels.

Assessment of Risk Factors

Presence of hypertension, hyperlipidemia, or diabetes mellitus was diagnosed by clinical history, BP measurements or blood tests, and if participants were receiving drug treatment for these conditions. Smoking status was categorized as never, current or past smoking according to self-reports. Physical activity was assessed by a validated short version of the Minnesota questionnaire[14] and expressed in minutes at a given metabolic equivalent (MET-min) per week. Height, weight, and waist circumference were measured with standard methods. Urinary albumin excretion was measured in morning spot urine samples by turbidimetry. Albuminuria was expressed as albumin (mg)/creatinine (g) ratio; an elevated ratio was defined by values ranging between 30 and 300 mg/g.

Diets

Participants followed their self-selected diets, and no specific dietary recommendations were given except to consume the daily allotments of walnuts in the WG and to refrain from eating nuts in the CG. No advice on salt intake was provided. Every 2 months until study completion, dietitians collected and revised a 7-day weighed food record, of which 3 days (2 working days and 1 week-end day) were randomly selected. Food Processor Plus 10.0 (ESHA Research, Salem, OR) was used to translate food items into nutrients, using the built-in database of the software adapted to local foods, with data based on Spanish food composition tables.[15] Compliance with walnuts in the WG was evaluated by recount of empty packages. As an additional measure of adherence, we assessed changes in the α-linolenic acid (ALA) proportion of red blood cell (RBC) membranes, as ALA is a fatty acid characteristic of walnuts, and its blood membrane content is an objective biomarker of consumption.[16] Participants in the WG who had difficulty chewing because of dental problems were provided with a coffee grinder at no cost and instructed on how to eat the ground walnuts incorporated into semiliquid foods such as yogurt. All participants were advised to maintain their usual level of physical activity.

RBC Membrane Fatty Acid Analyses

Fasting blood samples were collected by venipuncture at baseline and 2 years, and aliquots of whole blood were stored at −80ºC until fatty acids analysis. The RBC fatty acid profile was determined as described.[17] In brief, cells contained in a 100 µL aliquot of EDTA-collected blood were hemolyzed and spun. The pellet (>99% RBC membranes) was dissolved in 1 mL BF3 methanol solution and heated to hydrolyze and methylate glycerophospholipid fatty acids. The fatty acid methyl esters were isolated by adding n-hexane and were separated by gas-chromatography using an Agilent HP 7890 Gas Chromatograph equipped with a 30 m×0.25 µm×0.25 mm SupraWAX-280 capillary column (Teknokroma, Barcelona, Spain), an autosampler, and a flame ionization detector. The amount of ALA was expressed as the proportion of the total fatty acids identified in the sample.

BP Measurements

Participants attended the clinic visit on a weekday between 8:00 to 10:00 AM. Office BP was measured according to current guidelines[18] with a validated semiautomatic oscillometer (Omron HEM-705CP; Hoofddorp, the Netherlands).

Ambulatory BP monitoring was performed using Spacelabs 90207/90217 devices (SpacelabsW Inc, Richmond, WA), with readings scheduled every 20 minutes during the 24-hour time frame. Periods of activity and rest were determined on an individual basis

according to a diary recording hours of sleep and waking time. The duration of the procedure in hours, the percentage of valid readings, and mean systolic BP (SBP)and diastolic BP during periods of activity and rest, and for the whole 24-hour period, were determined. We included all ambulatory BP recordings lasting 24 hour±30 minutes and having >70% measurements, including ≥1 valid measurement/h. Average 24-hour BP was defined as normal when values were <130/80 mm Hg according to the standard definition of the 7th Report of the Joint National Committee (JNC7) guidelines[19] or <125/75 mm Hg according to the new 2017 guidelines proposed by the American College of Cardiology (ACC)/American Heart Association (AHA) and other societies.[20]

Statistical Analyses

The prespecified primary outcomes were the 2-year differences in office BP and 24-hour ambulatory blood pressure monitoring between the WG and CG. A target sample size of 77 individuals per group provided >80% power to detect a mean between-diet difference of 4 mm Hg (SD, 8.3) in 24-hour systolic ambulatory BP.[21] Normal distribution of data was assessed using graphical methods and the Shapiro–Wilk test. Data are expressed as mean and SD for quantitative variables and absolute numbers (percentages) for qualitative variables. Between-group differences in cardiovascular risk factors at baseline were assessed using the χ^2 test or ANOVA, as appropriate. The effect of the intervention on changes in food and nutrient consumption, anthropometric variables, urinary albumin excretion, and office BP and 24-hour ambulatory BP was assessed using ANOVA. Mean BP values by group were calculated by ANOVA and, for differences, they were obtained by ANCOVA with multivariable adjustment by age, sex, body mass index, smoking, energy intake, changes in energy expenditure, baseline BP values, and use of antihypertensive medication and their on-treatment changes. Subgroup analyses were performed considering the effects of the intervention by tertiles of baseline office BP and 24-hour ambulatory BP. The same adjustments applied in the primary analysis were used for these secondary analyses. The differences in the proportions of individuals with well-controlled office BP and 24-hour ambulatory BP at baseline and at the end of the study by group assignment were calculated by the McNemar test using both the standard JNC7 cutoffs[19] and the new 2017 ACC/AHA normality thresholds.[20] Statistical significance was established at $P<0.05$ (2-tailed). Analyses were performed using SPSS software, release 19.0 (IBM Corp, Armonk, NY).

Results

From April 2012 to December 2013, a total of 642 potential candidates were prescreened for eligibility. Eligible participants were recruited through nonprofit local organizations, advertisements in the study center, investigators contacts, and word of mouth (Figure 1). After they completed a short questionnaire, 198 candidates were excluded for various reasons, mainly for not meeting inclusion criteria. The remaining 444 candidates were formally assessed for eligibility in a face-to-face interview with the study clinician, who explained the protocol in detail, assessed potential compliance and reviewed the medical history, inclusion and exclusion criteria, and recent blood work and use of medication or supplements. This led to further exclusion of 139 candidates, leaving a final number of 305 participants who were recruited and randomly allocated to one of the 2 interventions. Because of logistic constraints, including availability of monitoring devices, ambulatory BP was not measured in 28 participants. From the 305 individuals considered for 24-hour BP measurement, 69 (23%) dropped out for various reasons, as shown in Figure 1, leaving 116 and 120 evaluable participants in the WG and CG, respectively. The characteristics of dropouts were similar to those of the whole cohort

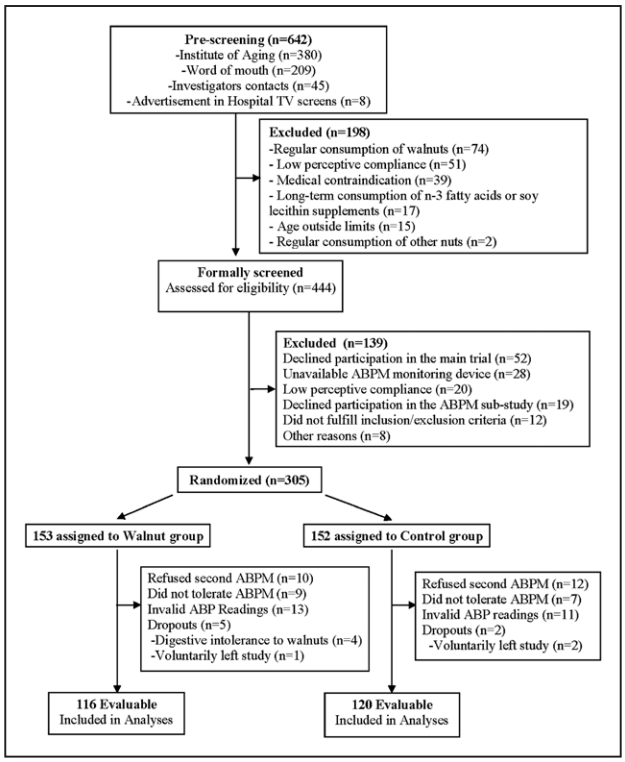

Figure 1. Flow chart of participants through the trial. ABPM, ambulatory blood pressure monitoring.

(data not shown). Follow-up was terminated after intervention for 2 years between April 2014 and December 2015.

The baseline characteristics of participants completing intervention for 2 years were well balanced between the 2 groups, except for body mass index, which was higher in the CG (Table 1). The mean age was 68.9 years, 41% were men, 53.4% had dyslipidemia, 11.1% were diabetic, and 60% had a JNC7 diagnosis of hypertension (72% by 2017 AHA cutoffs), with mean office BP values at baseline of 128.8/78.3 mm Hg. Among the 142 participants with a diagnosis of hypertension, 74 (52%) had well-controlled BP values by JNC7 standards (office BP <140/90 mm Hg).

Tolerance and Side Effects

Supplemental walnuts were generally well tolerated. There were 4 dropouts in the WG because of severe dyspepsia attributed to walnuts, whereas 20 participants had milder dyspepsia, which was solved by temporarily reducing walnut doses. Additionally, 16 participants required grinding the walnuts because of difficulty chewing because of bad dentures. Concerning bowel habit, of 116 participants in the WG, 64 (55%), 44 (38%), and 8 (7%) reported no change, improvement (softening of previously hard stools), or worsening (harder or inconveniently soft stools), respectively. Respective values in 120 CG participants were 113 (94%), 4 (3.5%), and 3 (2·5%).

Energy and Nutrient Intake

All participants followed a typical Mediterranean dietary pattern characterized by high-fat and high-monounsaturated fatty acid intake at baseline because of customary use of olive

Table 1. Baseline Characteristics of Participants Completing Intervention for 2 Years by Study Group

Variable	Walnut Group (n=116)	Control Group (n=120)	*P* Value*
Age, y	69.2 (3.5)	68.5 (3.1)	0.090
Men, n (%)	40 (34.5)	42 (35.0)	0.934
Current smokers, n (%)	8 (6.9)	2 (1.7)	0.056
Body weight, kg	69.3 (12.0)	71.6 (12.9)	0.152
Body mass index, kg/m^2	26.3 (3.4)	27.5 (4.1)	0.022
Waist circumference, cm	96.6 (10.1)	99.2 (11.9)	0.076
Energy expenditure in physical activity, MET-min/wk	2950 (1773)	2959 (1972)	0.918
Hypertension, n (%)†	68 (58.6)	74 (61.7)	0.633
Hypertension, n (%)‡	82 (70.7)	87 (72.5)	0.758
Office systolic BP, mm Hg	128.9 (15.7)	127.0 (17.8)	0.385
Office diastolic BP, mm Hg	79.3 (8.1)	78.3 (9.1)	0.350
Dyslipidemia, n (%)	61 (52.6)	65 (54.2)	0.808
Type 2 diabetes mellitus, n (%)	15 (12.9)	11 (9.2)	0.356
Educational level, n (%)			0.635
Primary education	44 (37.9)	52 (43.3)	
Secondary education	28 (24.1)	24 (20.0)	
Academic/graduate	44 (37.9)	44 (36.7)	
Medication use, n (%)			
Antihypertensive agents	60 (51.7)	60 (50.0)	0.791
ACE inhibitors/ARB	40 (34.5)	36 (30)	
Diuretics	19 (16.4)	15 (12.5)	
Calcium channel blockers	4 (3.4)	10 (8.3)	
Other antihypertensive drugs	10 (8.6)	14 (11.7)	
Antidiabetic medication	11 (9.5)	7 (5.8)	0.291
Oral hypoglycemic agents	8 (6.9)	6 (5.0)	
Insulin	4 (3.4)	1 (0.8)	
Hypolipidemic drugs	49 (42.2)	44 (36.7)	0.381
Statins	48 (41.4)	40 (33.3)	
Other lipid-lowering drugs	2 (1.7)	7 (5.8)	
Antiplatelet therapy	16 (13.8)	17 (14.2)	0.934

Values are means (SD) except for qualitative variables, expressed as n (%). ACE indicates angiotensin-converting enzyme; ARB, aldosterone receptor blockers; BP, blood pressure; and MET-min, minutes at a given metabolic equivalent level (units of energy expenditure in physical activity 1 MET-min is roughly equivalent to 1 kcal).

**P* value for comparisons between groups by χ^2 test for categorical variables and 1-way ANOVA for continuous variables.

†According to the JNC7 definition (BP ≥140/90 mm Hg or antihypertensive medication).

‡According to the 2017 ACC definition (BP ≥130/80 mm Hg or antihypertensive medication).

oil, without between-group differences except for total energy and potassium, which were higher in the WG (Table 2). After 2 years of walnut supplementation, WG participants increased significantly the intake of energy and total fat and reciprocally decreased carbohydrate, including simple sugars, compared with the control diet. Besides the increase in total fat, increases in polyunsaturated fatty acids, magnesium, and calcium in the WG reflected the energy, nutrient and mineral composition of walnuts. No between-group differences were observed in saturated fatty acids, monounsaturated fatty acids, fiber or sodium, and potassium intake at the end of the study.

Walnuts (median dose 42.5 g/d, equivalent to one-and-half 28-g servings/d) were well tolerated, and compliance was >98% (median compliance was 99 [IQ range, 96.5–99.7]) according to recount of empty packages. The analysis of RBC ALA disclosed no between-group differences at baseline (Figure S1 in the online-only Data Supplement). In contrast, at the end of intervention, 2-year changes in RBC ALA increased significantly ($P<0.001$) in the WG compared with controls, confirming good adherence to the intervention.

Energy Expenditure and Adiposity

Small changes of energy expenditure in self-reported physical activity were observed in the 2 groups: −394 MET-min/wk (95% CI, −647 to −140) in the WG and −207 MET-min/wk (−459 to 44) in the CG ($P=0.305$ for between-group comparison). Respective changes in body weight were 0.27 kg (−0.22 to 0.75) and −0.29 kg (−0.77 to 0.19; $P=0.112$), although waist circumference increased slightly and to a similar extent in the 2 groups: 0.31 cm (0.14–0.76) in the WG and 0.47 cm (0.02–0.93) in the CG ($P=0.613$).

Changes in Medication

There were few antihypertensive medication changes during the study, but they differed between the 2 groups, with 7 and 16 participants having add-on medication prescribed by their primary care physicians in the WG and CG, respectively ($P=0.046$).

Urinary Albumin Excretion

Similar urinary albumin excretion values were observed in the 2 groups at baseline and at the end of the study (Table S1). At baseline, 8 and 9 participants in WG and CG, respectively, disclosed an elevated urinary albumin-creatinine ratio, although at the end of the study there were 3 less participants per group, without between-group differences.

Changes in BP

In analyses were done by originally assigned group, baseline office BP and 24-hour BP values were similar between the 2 groups. Table 3 shows that office SBP and diastolic BP decreased significantly from baseline in the WG, but not in the CG, and between-group differences approached statistical significance for SBP ($P=0.051$). Twenty-four-hour and daytime ambulatory BP decreased significantly from baseline in the 2 groups, although nighttime ambulatory BP decreased significantly from baseline only in the WG. Whereas the changes in 24-hour, daytime and nighttime ambulatory BP were more pronounced in the WG compared with the CG (−3.86 versus −2.00 mm Hg; −3.87 versus −2.60 mm Hg; and −2.29 versus 0.08 mm Hg, respectively), between-group differences were nonsignificant.

To ascertain whether diet effects on BP related to baseline values, we categorized both office and ambulatory BP in

Table 2. Baseline levels and Changes in Daily Energy and Nutrient Intake by Group Assignment

Variable	Walnut Group (n=116)	Control Group (n=120)	*P* Value*
Energy, kcal			
Baseline	1772 (1698 to 1846)	1676 (1616 to 1736)	0.048
Change	149 (84 to 213)	–1 (–61 to 58)	0.001
Energy from protein, %			
Baseline	17.8 (17.3 to 18.3)	18.5 (17.9 to 19)	0.071
Change	–0.61 (–1.15 to –0.08)	–0.12 (–0.79 to 0.55)	0.259
Energy from carbohydrate, %			
Baseline	42.5 (41.1 to 43.8)	41.8 (40.7 to 43.0)	0.479
Change	–4.3 (–5.6 to –3.1)	2.0 (0.6 to 3.4)	<0.001
Energy from simple sugars, %			
Baseline	15.5 (14.4 to 16.6)	14.6 (13.7 to 15.5)	0.228
Change	–1.3 (–2.3 to –0.3)	1.3 (0.3 to 2.3)	<0.001
Fiber, g/1000 kcal			
Baseline	19.1 (17.7 to 20.3)	17.6 (16.5 to 18.6)	0.090
Change	1.48 (0.37 to 2.58)	0.70 (–0.76 to 2.16)	0.403
Energy from fat, %			
Baseline	39.1 (38.0 to 40.3)	39.2 (38.2 to 40.2)	0.941
Change	6.2 (5.0 to 7.5)	–1.6 (–2.9 to –0.3)	<0.001
Energy from saturated fatty acids, %			
Baseline	9.9 (9.5 to 10.3)	9.9 (9.4 to 10.3)	0.812
Change	–0.47 (–0.92 to –0.02)	–0.39 (–0.87 to 0.09)	0.797
Energy from monounsaturated fatty acids, %			
Baseline	20.5 (19.7 to 21.2)	20.8 (20.1 to 21.5)	0.519
Change	–1.7 (–2.5 to –0.9)	–1.1 (–2.0 to –1.1)	0.323
Energy from polyunsaturated fatty acids, %			
Baseline	5.2 (4.8 to 5.5)	5.3 (5.0 to 5.7)	0.521
Change	8.4 (7.8 to 9.0)	–0.6 (–1.0 to –0.2)	<0.001
Sodium, mmol			
Baseline	85.7 (80.7 to 90,6)	80.1 (75.2 to 85)	0.115
Change	–7.1 (–12.6 to –1,6)	–1.7 (–6.7 to 3.3)	0.150
Potassium, mmol			
Baseline	72.3 (68.7 to 75.9)	67.2 (64.5 to 69.9)	0.026
Change	3.2 (–0.3 to 6.6)	2.6 (–0.7 to 5.9)	0.823
Magnesium, mmol			
Baseline	11.1 (10.5 to 11.8)	10.3 (9.8 to 10.8)	0.054
Change	2 (1.4 to 2.6)	0.3 (–0.3 to 0.9)	<0.001
Calcium, mmol			
Baseline	18.3 (16.2 to 19.5)	17 (16.1 to 17.9)	0.092
Change	1.7 (0.4 to 3)	–0.1 (–1 to 0.8)	0.032

Data are expressed as mean (95% CI). Changes for which the 95% CI does not include zero are significantly different from baseline values.

**P* value for comparisons between groups by 1-way ANOVA.

tertiles. The results of this exploratory analyses (Table S2) show that, in the 2 groups, all measures of both systolic and diastolic BP tended to increase in the bottom tertile and to decrease in the middle tertile, although there was generally a marked reduction in the top tertile. Adjusted changes usually favored the WG and, for 24-hour ambulatory BP, were significantly different

Table 3. Office and Ambulatory Blood Pressure at Baseline and Changes at 2 y by Study Group

Blood pressure, mm Hg		Walnuts (n=116)	Control Diet (n=120)	*P* Value*
Systolic				
Office	Baseline	128.9 (125.9 to 132.0)	127.0 (124.0 to 130.1)	0.385
	Change	−4.61 (−7.43 to −1.79)	−0.59 (−3.38 to 2.21)	0.051
24-Hour	Baseline	121.4 (119.4 to 123.3)	120.8 (118.9 to 122.8)	0.714
	Change	−3.86 (−5.45 to −2.26)	−2.00 (−3.58 to −0.42)	0.111
Daytime	Baseline	124.9 (122.9 to 126.9)	124.2 (122.2 to 126.2)	0.632
	Change	−3.87 (−5.55 to −2.20)	−2.60 (−4.26 to −0.94)	0.296
Nighttime	Baseline	113.0 (110.8 to 115.3)	113.2 (110.9 to115.4)	0.939
	Change	−2.29 (−4.33 to −0.25)	0.08 (−1.97 to 2.13)	0.114
Diastolic				
Office	Baseline	79.3 (77.7 to 80.9)	78.3 (76.7 to 79.8)	0.350
	Change	−1.76 (−3.34 to −0.18)	0.02 (−1.54 to 1.59)	0.121
24-Hour	Baseline	70.3 (69.1 to 71.6)	70.3 (69.0 to 71.5)	0.945
	Change	−2.86 (−3.88 to −1.85)	−2.59 (−3.59 to −1.58)	0.707
Daytime	Baseline	73.4 (72.1 to 74.8)	73.1 (71.8 to 74.5)	0.764
	Change	−2.99 (−4.08 to −1.91)	−2.96 (−4.03 to −1.88)	0.962
Nighttime	Baseline	63.7 (62.3 to 65.2)	63.8 (62.4 to 65.2)	0.928
	Change	−1.84 (−3.16 to −0.52)	−0.97 (−2.29 to 0.36)	0.366

Data are expressed as mean (95% CI). Changes for which the 95% CI does not include zero are significantly different from baseline values.

**P* value for comparisons between groups by 1-way ANOVA and for comparisons of changes by ANCOVA with multivariable adjustment by age, sex, BMI, smoking, energy intake, changes in energy expenditure, and use of antihypertensive medication and their on-treatment changes.

(*P*=0.034) from the CG for mean top tertile systolic ambulatory BP (−8.5 versus −4.2 mm Hg) but not mean diastolic ambulatory BP (−5.8 versus -5.3 mm Hg), as shown in Figure 2. Table S2 shows that nighttime ambulatory BP in the top tertile also decreased to a significantly greater extent (*P*=0.023) in the WG than in the CG (−8.3 versus −2.5 mm Hg).

Control rates of BP at baseline, defined by either JNC7 or 2017 AHA cutoffs, were similar between the 2 intervention groups according to both office BP and 24-hour BP (Tables S3a and S3b). By JNC7 standards, 24-hour and daytime ambulatory BP regulation improved significantly (*P*<0.005) at 2 years in the WG but not in the CG, whereas office BP and nighttime ambulatory BP control were similar between groups (Table S3a). When considering 2017 AHA thresholds, the control of office BP improved significantly (*P*=0.004) in the WG but not in the CG, whereas control by ambulatory BP improved similarly in the 2 intervention groups (Table S3b). White-coat hypertension, defined by elevated office BP with normal 24-hour ambulatory BP values, was present in <5% of participants at baseline and up to 10.4% at 2 years by JNC7 cutoffs, with similar rates in the 2 groups (Table S3a). The proportion of participants with white-coat hypertension was more than doubled at both baseline and study's end when considering the new ACC/AHA thresholds (Table S3b).

Discussion

In this prespecified sub-study of the WAHA trial, a diet supplemented with walnuts at ≈15% energy during 2 years resulted in a nearly significant (*P*=0.051) mean 4.6 mm Hg reduction in systolic office BP and nonsignificant reductions in office diastolic BP or 24-hour ambulatory BP values compared with a control diet in fit elders (mean age 69 years), of whom 60% had mild hypertension, treated pharmacologically in most of them and well controlled in 52%. Given the relatively low baseline office BP values in the cohort (mean 129/78 mm Hg) and the difficulty to detect net differences when the baseline level is low,[22] we categorized BP in tertiles and found significant reductions of ≈8 mm Hg in mean systolic 24-hour and nighttime ambulatory BP in the top tertile (mean 24-hour systolic ambulatory BP >125 mm Hg) of the WG compared with the CG. Importantly, during the trial, less participants in the WG required uptitration of antihypertensive medication for BP control than those in the CG and BP control by 24-hour and daytime ambulatory BP monitoring improved significantly in the WG but not in the CG. No other lifestyle recommendations were given, and at the end of the study the 2 groups had similar body weight and levels of physical activity and sodium intake, albeit WG participants increased intakes of energy, total and polyunsaturated fat, and nonsodium minerals, reflecting the energy and nutrient composition of walnuts. Thus, the beneficial effect of the intervention on BP can be reasonably ascribed to walnut consumption in itself.

Of note, walnut supplementation for 2 years did not lead to unwanted weight gain in spite of the extra energy they contributed to the diet. The lack of a weight-promoting effect of nuts has been attributed in part to their satiating effect, which leads to a reduction in energy intake and to a deficit of metabolizable energy because of inefficient energy absorption leading to

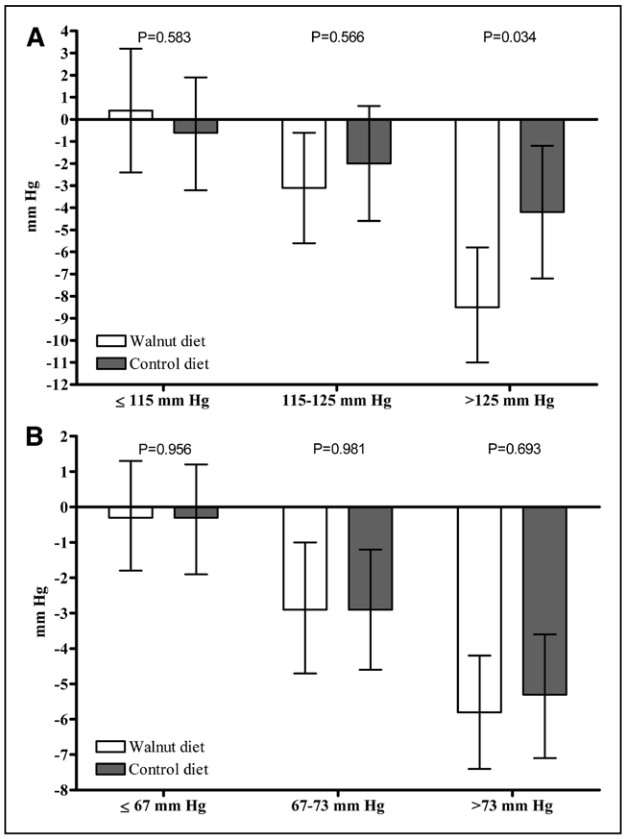

Figure 2. Changes in systolic (**A**) and diastolic (**B**) 24-hour ambulatory blood pressure by tertiles of baseline ambulatory blood pressure. Values are means; error bars are 95% CIs. **P* value for comparisons between groups by 1-way ANOVA and for comparisons of changes by ANCOVA with multivariable adjustment by age, sex, body mass index (BMI), smoking, energy intake, changes in energy expenditure, and use of antihypertensive medication and their on-trial changes.

increased fecal fat excretion.[9] Also, in our study, the decrease in total carbohydrate reciprocal to the increased fat intake from walnuts was associated with a significant reduction in intake of simple sugars, which may help curb weight gain. A recent report from the Loma Linda site of the WAHA study confirms the lack of fattening effect of the walnut doses used in the trial.[23]

The present results provide high-level scientific evidence on the effects of walnuts on BP in the elderly. Till now, the evidence from meta-analyses of randomized feeding studies has been that nuts in general[11,12] or walnuts in particular[24] have no effect on office BP. However, BP results in prior studies were usually derived from post hoc analyses, and most trials had low statistical power. To our knowledge, only 2 prior controlled trials assessed 24-hour ambulatory BP in response to nut feeding. A study by Doménech et al[6] conducted within the frame of the PREvención con DIeta MEDiterránea (PREDIMED) trial in 235 individuals at high cardiovascular risk, most with treated hypertension, showed that Mediterranean diets supplemented with extra-virgin olive oil or mixed nuts (30 g per day: 15 g walnuts, 7.5 g almonds, and 7.5 g hazelnuts) reduced 24-hour ambulatory BP compared with the control diet after intervention for 1 year. However, as there were other dietary changes in the PREvención con DIeta MEDiterránea (PREDIMED) study, this beneficial effect cannot be solely ascribed to nut consumption. Dhillon et al[25] conducted a 12-week study in overweight but otherwise healthy middle-aged adults undergoing an energy-restricted diet with or without almond supplementation and detected no changes in 24-hour ambulatory BP, but participants in the almond group who were good compliers with the intervention had a significant reduction in office diastolic BP compared with the CG.

Among nuts, walnuts are particularly well suited to have BP effects because they contain little sodium and possess a complex nutrient matrix, including sizable amounts of bioactive molecules: ALA, the vegetable n-3 fatty acid, the metabolism of which gives rise to vasodilatory and anti-inflammatory oxylipins; γ-tocopherol, a form of vitamin E active in reducing oxidation and inflammation; nonsodium minerals with BP-lowering effects such as potassium, magnesium and calcium; arginine, the amino acid precursor of the endogenous vasodilator nitrous oxide; and characteristic polyphenols disclosing potent antioxidative and anti-inflammatory actions.[26] Indeed, walnuts are the only nut type shown to consistently improve endothelial function in controlled trials testing nut diets for effects on vascular reactivity.[27]

Presently compelling evidence has accumulated that the lowest incidence of cardiovascular complications is observed among individuals with normal SBP and even slightly elevated SBP significantly increase CVD risk and mortality, although the protective effect of antihypertensive therapy directly relates to the achieved SBP lowering.[28] Such level of evidence underlies the lower thresholds to define hypertension for office BP and ambulatory BP monitoring proposed by the new ACC/AHA guidelines.[20] Hence, the SBP-lowering effect of a walnut diet demonstrated by 24-hour ambulatory BP monitoring in participants with elevated BP or mild hypertension is particularly important. Recent data from a large registry indicating that 24-hour systolic ambulatory BP is a stronger predictor of all-cause and cardiovascular mortality than office SBP[29] further support the clinical relevance of our findings. Together with their cholesterol-lowering effect,[24] the antihypertensive effect of walnuts also helps explain the consistent reduction in CVD rates and mortality observed in prospective studies.[8,9] That a safe, low-cost, nonpharmacological intervention such as regular walnut consumption helps lower SBP among individuals at low cardiovascular risk with elevated BP and mild hypertension is important, given that antihypertensive drug treatment in this situation appears to have little impact on CVD outcomes or mortality and could be associated with an increased risk of adverse events.[30]

Of note, the reduction in incident stroke observed with a Mediterranean diet supplemented with nuts in the PREvención con DIeta MEDiterránea (PREDIMED) trial[5] supported a recommendation of the 2014 AHA/American Stroke Association guideline for the primary prevention of stroke.[31] The benefit on stroke of such a diet, together with their demonstrated BP-lowering effect[6] also informed the recent lifestyle recommendations of the ACC/AHA hypertension guidelines[20] and the 2018 European Guidelines[18] as the first attempt to reduce BP at high-normal range (SBP, 120–139 mm Hg) in individuals at low cardiovascular risk.

Our study has limitations. First, participants could not be blinded to the intervention, since it was made up of a whole food. Second, except for their age, participants were at low cardiovascular risk and their BP values were not elevated,

thus compromising the detection of BP changes because of the intervention. Finally, the study sample was an elderly cohort, thus the results cannot be easily extrapolated to younger individuals. The study has also strengths, such as the randomized design with a sizable population and long follow-up; the relatively good retention rate for such an elderly sample; the use of objective biomarkers to confirm adherence to the intervention; and the evaluation of BP outcomes with 24-hour ambulatory BP monitoring, a technique that minimizes imprecision and is of choice for clinical trials.

In conclusion, the results of our randomized feeding study provide good level scientific evidence that walnut consumption has BP-lowering effect and can be used safely in the dietary management of hypertension. The data support the recommendation to incorporate walnuts to a healthy dietary pattern to help control BP and reduce cardiovascular risk in individuals with elevated BP.

Perspectives

In a randomized controlled clinical trial conducted in 236 elderly individuals at low cardiovascular risk, we showed that supplementation of the usual diet with walnuts at ≈15% of energy during 2 years reduced office SBP in the whole cohort and mean systolic 24-hour ambulatory BP in those with elevated BP and mild hypertension compared with a control diet without nuts. That regular consumption of a single whole food appears to be a useful adjunct to dietary and pharmacological approaches for improving the control of high BP has public health implications because it might help reduce the burden and attendant side-effects of pharmacological treatment of mild hypertension. These results should be confirmed in younger individuals and in non-Mediterranean cohorts.

Acknowledgments

We thank the participants in the WAHA trial for their enthusiastic collaboration and Emili Corbella for expert assistance with statistical analyses. Authors' contributions to the article: E. Ros and A. Sala-Vila designed research; T.-M. Freitas-Simoes, M. Cofán, M. Serra-Mir, I. Roth, C. Calvo, C. Valls-Pedret, M. Domènech, conducted the research; M. Domènech and E. Ros wrote the article; J. Sabaté and E. Ros had primary responsibility for the final content. All authors read and approved the final article. CIBEROBN is an initiative of ISCIII, Spain.

Sources of Funding

This work was supported by a grant from the California Walnut Commission, Sacramento, CA. The funding agency had no input in the study design, data collection, analyses, or writing and submission of the article. An external overseeing committee monitored the study to ensure quality control, data integrity, and participants' safety. A. Sala-Vila holds a Miguel Servet I fellowship from the Ministry of Economy and Competitiveness through ISCIII, Spain.

Disclosures

J. Sabaté and E. Ros have received research funding through their institutions from the California Walnut Commission (CWC) and are nonpaid members of its Scientific Advisory Committee. ER has also received honoraria from the CWC for preparation of scientific presentations and other activities. The other authors report no conflicts.

References

1. Lawes CM, Vander Hoorn S, Rodgers A. Global burden of blood pressure related disease, 2001. *Lancet*. 2008;37:1513–1518.
2. Afshin A, Forouzanfar MH, Reitsma MB, et al; GBD 2015 Obesity Collaborators. Health effects of overweight and obesity in 195 countries over 25 years. *N Engl J Med*. 2017;377:13–27. doi: 10.1056/NEJMoa1614362
3. North BJ, Sinclair DA. The intersection between aging and cardiovascular disease. *Circ Res*. 2012;110:1097–1108. doi: 10.1161/CIRCRESAHA.111.246876
4. Bloom DE, Chatterji S, Kowal P, Lloyd-Sherlock P, McKee M, Rechel B, Rosenberg L, Smith JP. Macroeconomic implications of population ageing and selected policy responses. *Lancet*. 2015;385:649–657. doi: 10.1016/S0140-6736(14)61464-1
5. Estruch R, Ros E, Salas-Salvadó J, et al; PREDIMED Study Investigators. Primary prevention of cardiovascular disease with a mediterranean diet supplemented with extra-virgin olive oil or nuts. *N Engl J Med*. 2018;378:e34. doi: 10.1056/NEJMoa1800389
6. Doménech M, Roman P, Lapetra J, García de la Corte FJ, Sala-Vila A, de la Torre R, Corella D, Salas-Salvadó J, Ruiz-Gutiérrez V, Lamuela-Raventós RM, Toledo E, Estruch R, Coca A, Ros E. Mediterranean diet reduces 24-hour ambulatory blood pressure, blood glucose, and lipids: one-year randomized, clinical trial. *Hypertension*. 2014;64:69–76. doi: 10.1161/HYPERTENSIONAHA.113.03353
7. Ndanuko RN, Tapsell LC, Charlton KE, Neale EP, Batterham MJ. Dietary patterns and blood pressure in adults: a systematic review and meta-analysis of randomized controlled trials. *Adv Nutr*. 2016;7:76–89. doi: 10.3945/an.115.009753
8. Aune D, Keum N, Giovannucci E, Fadnes LT, Boffetta P, Greenwood DC, Tonstad S, Vatten LJ, Riboli E, Norat T. Nut consumption and risk of cardiovascular disease, total cancer, all-cause and cause-specific mortality: a systematic review and dose-response meta-analysis of prospective studies. *BMC Med*. 2016;14:207. doi: 10.1186/s12916-016-0730-3
9. Ros E. Nuts and CVD. *Br J Nutr*. 2015;113(suppl 2):S111–S120. doi: 10.1017/S0007114514003924
10. Sabaté J, Oda K, Ros E. Nut consumption and blood lipid levels: a pooled analysis of 25 intervention trials. *Arch Intern Med*. 2010;170:821–827. doi: 10.1001/archinternmed.2010.79
11. Del Gobbo LC, Falk MC, Feldman R, Lewis K, Mozaffarian D. Effects of tree nuts on blood lipids, apolipoproteins, and blood pressure: systematic review, meta-analysis, and dose-response of 61 controlled intervention trials. *Am J Clin Nutr*. 2015;102:1347–1356. doi: 10.3945/ajcn.115.110965
12. Mohammadifard N, Salehi-Abargouei A, Salas-Salvadó J, Guasch-Ferré M, Humphries K, Sarrafzadegan N. The effect of tree nut, peanut, and soy nut consumption on blood pressure: a systematic review and meta-analysis of randomized controlled clinical trials. *Am J Clin Nutr*. 2015;101:966–982. doi: 10.3945/ajcn.114.091595
13. Rajaram S, Valls-Pedret C, Cofán M, et al. The walnuts and healthy aging study (WAHA): protocol for a nutritional intervention trial with walnuts on brain aging. *Front Aging Neurosci*. 2016;8:333. doi: 10.3389/fnagi.2016.00333
14. Molina L, Sarmiento M, Peñafiel J, Donaire D, Garcia-Aymerich J, Gomez M, Ble M, Ruiz S, Frances A, Schröder H, Marrugat J, Elosua R. Validation of the regicor short physical activity questionnaire for the adult population. *PLoS One*. 2017;12:e0168148. doi: 10.1371/journal.pone.0168148
15. Mataix Verdú J. *Tabla de Composición de Alimentos*. 3ed. Granada: Universidad de Granada, Instituto de Nutrición y Tecnología de Alimentos; 2003.
16. Zambón D, Sabaté J, Muñoz S, Campero B, Casals E, Merlos M, Laguna JC, Ros E. Substituting walnuts for monounsaturated fat improves the serum lipid profile of hypercholesterolemic men and women. A randomized crossover trial. *Ann Intern Med*. 2000;132:538–546.
17. Sala-Vila A, Harris WS, Cofán M, Pérez-Heras AM, Pintó X, Lamuela-Raventós RM, Covas MI, Estruch R, Ros E. Determinants of the omega-3 index in a Mediterranean population at increased risk for CHD. *Br J Nutr*. 2011;106:425–431. doi: 10.1017/S0007114511000171
18. Williams B, Mancia G, Spiering W, et al; Authors/Task Force Members. 2018 ESC/ESH guidelines for the management of arterial hypertension: the task force for the management of arterial hypertension of the European Society of Cardiology and the European Society of Hypertension: the task force for the management of arterial hypertension of the European Society of Cardiology and the European Society of Hypertension. *J Hypertens*. 2018;36:1953–2041. doi: 10.1097/HJH.0000000000001940
19. Chobanian AV, Bakris GL, Black HR, Cushman WC, Green LA, Izzo JL Jr, Jones DW, Materson BJ, Oparil S, Wright JT Jr, Roccella EJ; Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure. National Heart, Lung, and Blood Institute; National High Blood Pressure Education Program Coordinating Committee.

Seventh report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure. *Hypertension*. 2003;42:1206–1252. doi: 10.1161/01.HYP.0000107251.49515.c2
20. Whelton PK, Carey RM, Aronow WS, et al. 2017 ACC/AHA/AAPA/ABC/ACPM/AGS/APhA/ASH/ASPC/NMA/PCNA guideline for the prevention, detection, evaluation, and management of high blood pressure in adults: a report of the American College of Cardiology/American Heart Association task force on clinical practice guidelines. *Hypertension*. 2018;71:1269–1324. Erratum: *Hypertension*. 2018;71:e136–e139.
21. Stergiou GS, Baibas NM, Gantzarou AP, Skeva II, Kalkana CB, Roussias LG, Mountokalakis TD. Reproducibility of home, ambulatory, and clinic blood pressure: implications for the design of trials for the assessment of antihypertensive drug efficacy. *Am J Hypertens*. 2002;15(2 pt 1):101–104.
22. Vollmer WM, Sacks FM, Ard J, Appel LJ, Bray GA, Simons-Morton DG, Conlin PR, Svetkey LP, Erlinger TP, Moore TJ, Karanja N; DASH-Sodium Trial Collaborative Research Group. Effects of diet and sodium intake on blood pressure: subgroup analysis of the DASH-sodium trial. *Ann Intern Med*. 2001;135:1019–1028.
23. Bitok E, Rajaram S, Jaceldo-Siegl K, Oda K, Sala-Vila A, Serra-Mir M, Ros E, Sabaté J. Effects of long-term walnut supplementation on body weight in free-living elderly: results of a randomized controlled trial. *Nutrients*. 2018;10:E1317.
24. Guasch-Ferré M, Li J, Hu FB, Salas-Salvadó J, Tobias DK. Effects of walnut consumption on blood lipids and other cardiovascular risk factors: an updated meta-analysis and systematic review of controlled trials. *Am J Clin Nutr*. 2018;108:174–187. doi: 10.1093/ajcn/nqy091
25. Dhillon J, Tan SY, Mattes RD. Almond consumption during energy restriction lowers truncal fat and blood pressure in compliant overweight or obese adults. *J Nutr*. 2016;146:2513–2519. doi: 10.3945/jn.116.238444
26. Ros E, Izquierdo-Pulido M, Sala-Vila A. Beneficial effects of walnut consumption on human health: role of micronutrients. *Curr Opin Clin Nutr Metab Care*. 2018;21:498–504. doi: 10.1097/MCO.0000000000000508
27. Xiao Y, Huang W, Peng C, Zhang J, Wong C, Kim JH, Yeoh EK, Su X. Effect of nut consumption on vascular endothelial function: a systematic review and meta-analysis of randomized controlled trials. *Clin Nutr*. 2018;37:831–839. doi: 10.1016/j.clnu.2017.04.011
28. Bundy JD, Li C, Stuchlik P, Bu X, Kelly TN, Mills KT, He H, Chen J, Whelton PK, He J. Systolic blood pressure reduction and risk of cardiovascular disease and mortality: a systematic review and network meta-analysis. *JAMA Cardiol*. 2017;2:775–781. doi: 10.1001/jamacardio.2017.1421
29. Banegas JR, Ruilope LM, de la Sierra A, Vinyoles E, Gorostidi M, de la Cruz JJ, Ruiz-Hurtado G, Segura J, Rodríguez-Artalejo F, Williams B. Relationship between clinic and ambulatory blood-pressure measurements and mortality. *N Engl J Med*. 2018;378:1509–1520. doi: 10.1056/NEJMoa1712231
30. Sheppard JP, Stevens S, Stevens R, Martin U, Mant J, Hobbs FDR, McManus RJ. Benefits and harms of antihypertensive treatment in low-risk patients with mild hypertension. *JAMA Intern Med*. 2018;178:1626–1634. doi: 10.1001/jamainternmed.2018.4684
31. Meschia JF, Bushnell C, Boden-Albala B, et al; American Heart Association Stroke Council; Council on Cardiovascular and Stroke Nursing; Council on Clinical Cardiology; Council on Functional Genomics and Translational Biology; Council on Hypertension. Guidelines for the primary prevention of stroke: a statement for healthcare professionals from the American Heart Association/American Stroke Association. *Stroke*. 2014;45:3754–3832. doi: 10.1161/STR.0000000000000046

Novelty and Significance

What Is New?

- The results of our randomized feeding trial provide high-level scientific evidence that regular walnut consumption has a blood pressure (BP)-lowering effect in elderly individuals at low cardiovascular risk.

What Is Relevant?

- Long-term consumption of a single whole food with a rich nutrient composition such as walnuts can help control BP and reduce the need of antihypertensive medication among individuals with elevated BP and mild hypertension, and might thus be used safely in the dietary management of hypertension.

Summary

Our data support the recommendation to incorporate walnuts to a healthy dietary pattern to reduce BP and increase BP control rates, particularly among individuals with elevated BP and low overall cardiovascular risk.