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Dietary determinants of serum total cholesterol among middle-aged and older adults in Dar es Salaam, Tanzania: a population-based cross-sectional study

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ABSTRACT

 Objective: To assess the dietary determinants of serum total cholesterol.

Design: Cross-sectional population-based study.

Setting: Peri-urban region of Dar es Salaam, Tanzania.

Participants: 356 adults aged 40 years and older from the Dar es Salaam Urban Cohort Hypertension Study (DUCS-HTN).

Main outcome measure: Serum total cholesterol measured using a point of care device. Results: Mean serum total cholesterol level was 203 mg/dL (Interquartile range (IQR) 169 to 235 mg/dL) in women and 183 mg/dL (IQR 149 to 215 mg/dL) in men. After adjustment for major demographic, socioeconomic and lifestyle factors, participants who reported using palm oil as the major cooking oil had serum total cholesterol higher by 17 mg/dL (95% confidence interval 4 to 30 mg/dL) compared with those who reported using sunflower, groundnut, corn or other oils. Higher meat consumption (P for trend= 0.03) and lower consumption of fruits and vegetables were also associated (P for trend= 0.01) with higher serum total cholesterol.

Conclusions: Use of palm oil for cooking is associated with higher serum total cholesterol levels in Tanzania. Reduction of saturated fat content of edible oil may be explored as a population-based strategy for the primary prevention of cardiovascular diseases.

Key words: cholesterol, palm oil, diet, nutrition, Tanzania, Sub-Saharan Africa

STRENGTHS AND LIMITATIONS OF THIS STUDY

- This is the first population-based study to report an association between using palm oil for cooking and serum total cholesterol in East Africa.
- A reliable point-of care- lipid testing system was used to measure serum total cholesterol that has good clinical agreement with laboratory reference methods.
- Potential confounders of the association between diet and serum total cholesterol were systematically measured and adjusted for, increasing the validity of findings.
- Only serum total cholesterol was measured instead of a full lipid panel, due to logistic and financial limitations.
- The assessment of diet using any method is prone to measurement error, which may have resulted in bias, although differential misreporting of cooking oil by cholesterol level is unlikely.

INTRODUCTION

Sub-Saharan Africa is facing a burgeoning epidemic of non-communicable diseases (NCDs) due to changing demographic profiles, epidemiologic transition, urbanization, and lifestyle changes as a result of economic development as well as increasing survival of HIV patients. [1, 2] In 2015, one third of all estimated NCD deaths in Sub-Saharan Africa were due to cardiovascular diseases (CVD). [3] By 2030, the share of all deaths attributable to CVD is projected to increase by almost 40% in sub-Saharan Africa. [3]

Dyslipidemia is a well-known risk factor for ischemic heart disease (IHD), stroke, and other vascular diseases. [4, 5] In middle-aged adults, IHD mortality is lower by as much as 30 to 50% for every 1 mmol/L (38.6mg/dL) lower serum total cholesterol. [4] Sub-Saharan Africans have historically had a favorable lipid profile. [6] However, hyperlipidemia is becoming increasingly common in these countries, especially in urban areas. [7-9] It is estimated that thirty-four percent of ischemic heart disease deaths in Tanzania are attributable to dyslipidemia. [10]

Previous studies in Tanzania have reported an association between higher intake of meat and lower intake of fish and green vegetables with higher serum total cholesterol.[11-15] Two of these studies were conducted in Bantu populations whose diet differs substantially from that of other ethnic groups in Tanzania.[14, 15] The other studies either did not report the magnitude of the association,[12] or did not adjust for potential confounders of the relationship between diet and serum total cholesterol.[11, 13] A better understanding of dietary determinants of dyslipidemia in Tanzania will be valuable in designing population-level interventions to reduce the growing CVD burden. We conducted a cross-sectional population-based study to assess the determinants of serum total cholesterol in adults in Dar es Salaam.

METHODS

Study design and population

The Dar es Salaam Health and Demographic Surveillance System (HDSS) is a demographic surveillance system in Ukonga and Gongo la Mboto wards of Dar es Salaam, Tanzania with over 100,000 enumerated individuals living in 21,000 households followed since 2011/12. For the DUCS Hypertension Study (DUCS-HTN), we randomly selected one large and one small neighborhood in the HDSS and contacted adult residents aged 40 years and older, who had been registered in the HDSS database in 2011 or 2012. A random subsample of one-fifth of the eligible participants was selected for additional assessment of diet and measurement of serum total cholesterol. The size of this subsample was determined largely based upon the availability of resources. Pregnant women and those who were physically and mentally incapable of completing the interview and measurements were excluded. The participant selection flowchart is shown in figure 1. Face-to-face interviews and measurements were conducted in participant's home from March to June 2014 by six trained interviewers. All interviewers had previous experience conducting public health surveys. For this study, interviewers received five days of training and two days of field practice in a nearby neighborhood. At least three attempts were made to contact all potential participants. Written informed consent was obtained from all participants.

The study protocol was approved by the Institutional Review Board of the Harvard TH Chan School of Public Health and the Research and Publications Committee of Muhimbili University of Health and Allied Sciences.

Measurements

Non-fasting serum total cholesterol was measured from 20 micro-liters of finger-prick capillary blood samples using Cardiochek PA devices (Polymer Technology Systems Inc, IN, USA).[16, 17] Data on diet was collected using a semi-quantitative food frequency questionnaire (FFQ). The FFQ assessed intake of 179 food and drink items over the past 30 days. A shorter 85-item version of this instrument has been previously used in Tanzania.[18] Nutrient intakes were calculated by multiplying the frequency of food consumption measured from FFQ by the nutrient content of the specified

portion size using the Tanzanian food composition table. [19] An additional question was asked to assess the main type of oil used for cooking.

All participants completed a socio-demographic and lifestyle questionnaire and had their height, weight, waist circumference, hip circumference, and blood pressure measured by trained interviewers. Body mass index was calculated as weight (kg) divided by height (m) squared, and categorized as normal (<25.0), overweight (25.0 to <30.0), and obese (≥30.0).[20] Some demographic information (age, sex, neighborhood, components of wealth index) was taken from the HDSS baseline interview, which was conducted from 2011 to 2012. Physical activity was assessed using the Global Physical Activity Questionnaire in the domains of work, transportation, and leisure,[21] and categorized into tertiles of Metabolic Equivalent (MET) hours per week. To measure socioeconomic status, we created a household wealth index by principal component analysis of household characteristics (floor material, roof material, wall material, electricity), household asset ownership (television, radio, shop, sewing machine, sofa, fan, iron, stove with oven, stove without oven, dining table, cupboard, watch, mobile phone, bike, motorcycle, cart, car, motorboat), and animal ownership (goats, sheep, chicken/ducks, pigs). The household wealth index was categorized into tertiles.

Statistical analysis

 Participants with implausible dietary data (total energy intake <500 or >5000 kcal/day), and with missing data on serum total cholesterol and/or any of the potential covariates were excluded from the analysis.

Mean differences and 95% confidence intervals in serum total cholesterol across categories of selected independent variables were estimated using multivariable linear regression. Variables were included in the model based on a-priori knowledge of lifestyle and socio-demographic determinants of diet and serum total cholesterol. Chisquare tests were used to assess binary variables, F-tests were used for categorical variables and tests of linear trend based on median value within each category for ordinal variables with a linear association with serum total cholesterol (i.e. age, wealth index, and food consumption categories). We adjusted for interviewer to remove the effect of any extraneous variation in measurement of subjective risk factors across interviewers.[22, 23] We examined interactions of potential determinants of serum

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total cholesterol with sex, but no interactions were statistically significant at 0.2 level, so we did not include these product terms in the final model.

All analyses were performed using STATA software version 13.1 (STATA Corporation, College Station, Texas).

RESULTS

A total of 420 participants were interviewed for the biomarker subsample. Among these, data on serum total cholesterol and selected covariates was available for 399 participants. We excluded 43 participants due to implausible caloric intake, resulting in a sample of 356 participants (figure 1).

Slightly less than half (42%) of the participants (n=356) were men. Mean age of men was 55 years (SD 11), and that of women was 52 (SD 10). About 44% of the women (n=207) and 19% of men (n=149) were obese, and about one third of men and women were overweight. Mean energy intake per day was about 2500 kcal (table 1). About three-quarters of participants reported using palm oil as the major cooking oil and the rest reported mainly using sunflower oil (20.5%). Other less commonly used cooking oils were groundnut (0.6%), olive (0.6%), corn (0.3%), or other oils (0.6%). Thirty-six percent of men and 53% of women were hypercholesterolemic (i.e. had total cholesterol \geq 200 mg/dL). Mean serum total cholesterol in women was 203 mg/dL (95% confidence interval 175 to 192 mg/dL). Serum total cholesterol levels were higher in older age groups.

Variable	All (n=356)	Men (n=149)	Women (n=207)	Use palm oil (n=274)	Use other cooking oil (n=82)
Age					
40-49	152 (43)	57 (38)	95 (46)	123 (45)	29 (35)
50-59	117 (33)	45 (30)	72 (35)	82(30)	35 (43)
60 and above	87(24)	47 (32)	40 (19)	69 (25)	18 (22)
Employment status					
Unemployed	85(24)	14 (9)	71 (34)	70(26)	15 (18)
Employed (includes self-	244 (69)	115 (77)	129(62)	183 (67)	61(74)
Retired	27 (8)	20 (13)	7(3)	21(8)	6(7)
Household wealth index					
Tertile 1, Poorest	119(33)	50 (34)	69 (33)	109 (40)	10 (12)
Tertile 2	119(33)	54 (36)	6 5 (31)	90 (33)	29 (35)
Tertile 3, Wealthiest	118 (33)	45 (30)	73 (35)	75 (27)	43 (52)
BMI (kg/m²)					
Underweight (<18.50)	20 (6)	14 (9)	6 (3)	19 (7)	1 (1)
Normal weight (18.50-<25.00)	103 (29)	58 (39)	45 (22)	91 (33)	12 (15)
Overweight (25-<30.00)	115 (32)	49 (33)	66 (32)	81 (30)	34 (41)
Obese (≥ 30.00)	118 (33)	28 (19)	90 (44)	83 (30)	35 (43)
Physical activity					. ,
Tertile 1, (0-14 MET-hours/week)	128 (36)	47 (32)	81 (39)	96~(35)	<i>32</i> (<i>39</i>)
Tertile 2, (15-112 MET-hours/week)	111 (31)	40 (27)	71 (34)	86 (31)	<i>25</i> (30)
Tertile 3, (113-840 MET-hours/week) Alcohol drinking ª	117 (33)	62 (42)	55 (27)	92 (34)	25 (30)

Table 1. Participants' characteristics in DUCS-HTN biomarker sub-study, 2014

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Variable	All (n=356)	Men (n=149)	Women (n=207)	Use palm oil (n=274)	Use other cooking oil (n=82)
Non-drinker	294 (83)	106 (71)	188 (91)	225 (82)	69 (84)
Smoking					
Non-smoker	284 (81)	86~(59)	198 (97)	210 (78)	74 (90)
Former smoker	45 (13)	41 (28)	4(2)	38 (14)	7(9)
Current smoker	23 (7)	20 (13)	3 (1)	22 (8)	1 (1)
DIETARY VARIABLES					
Total caloric intake $(kcal/day)^b$	$2414 \\ (1703, 3216)$	2629 (1756, 3216)	2329(1617, 3209)	2369 (1654, 3150)	2775 (1861, 3606)
Palm oil as major cooking oil	274 (77)	119 (80)	155 (75)		
Meat intake					
<1 serving/week	42 (12)	22 (15)	20 (10)	36 (13)	6(7)
1-6 servings/week	177 (50)	56 (38)	121 (58)	145 (53)	<i>32</i> (<i>39</i>)
≥1 servings/day	137 (39)	71 (48)	66 (32)	93 (34)	44 (54)
Fish intake					
< 1 serving/week	42 (12)	14 (9)	28 (14)	31 (11)	11 (13)
1-7 servings/week	202 (57)	79(53)	123 (59)	154 (56)	48 (59)
> 1 servings/day	112 (31)	56(38)	56(27)	89(32)	23 (28)
Dairy intake					
< 1 serving/week	164 (46)	65 (44)	99 (48)	136 (50)	28 (34)
1-6 servings/week	137 (39)	67 (45)	70 (34)	99 (36)	38 (46)
\geq 1 servings/day	55 (15)	17 (11)	38 (18)	39 (14)	16 (20)
Fruit and vegetable					
< 5 servings/day	123 (35)	49(33)	74 (36)	101 (37)	22 (27)
5-7 servings/day	70 (20)	27 (18)	43(21)	54(20)	16 (20)

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Variable	All (n=356)	Men (n=149)	Women (n=207)	Use palm oil (n=274)	Use other cooking oil (n=82)
>7 servings/day	163 (46)	73 (49)	90 (43)	119 (43)	44 (54)
Nuts and legumes					
< 1 serving/week	29 (8)	8(5)	21 (10)	17 (6)	12 (15)
1-7 servings/week	206 (58)	90 (60)	116 (56)	162 (59)	44 (54)
> 1 servings/day	121 (34)	51 (34)	70 (34)	95 (35)	
Fat intake ^b					/ \
Total fat (grams/day)	87 (58, 121)	90 (56, 124)	86 (60, 119)	85 (54, 119)	98 (68, 125)
Saturated fat (grams/day)	49 (33, 70)	53 (31, 74)	47 (34, 66)	49 <i>(32, 69)</i>	50 (39, 71)
Polyunsaturated fat	8(5, 13)	9(5, 12)	7(4, 13)	7(4, 12)	10 (6, 17)
Cholesterol					
Serum total cholesterol (mg/dl) ^c	195 (3)	183 (4)	203 (4)	199(3)	181(5)
Hypercholesterolemia ^d	164 (46)	54 (36)	110 (53)	135 (49)	29 (35)
Cholesterol level was previously checked	30 (8)	7 (5)	23 (11)		
Notified by doctors that they had hypercholesterolemia	13 (8)	1(2)	12 (11)		
Now taking cholesterol lowering medications	9 (3)	1 (1)	8 (4)		

Number (%) reported for all variables, except as specified b and c below

^a According to report of drinking alcohol in the past 30 days

 ^b Total caloric and fat intake, which is reported as median (interquartile range)

^c Serum total cholesterol, reported as mean (standard error)

^d Hypercholesterolemia defined as total cholesterol ≥ 200 mg/dL

Those who reported using palm oil as the major cooking oil had serum total cholesterol levels higher by 14 mg/dL (95% confidence interval 1 to 26 mg/dL) compared with those who reported using sunflower or other oils (table 2, figure 2. After adjustment for major demographic, socioeconomic and lifestyle factors, participants who used palm oil for cooking had 17mg/dl (95% confidence interval 4 to 30 mg/dL) higher serum total cholesterol. Serum total cholesterol was also higher in participants who ate more meat (P for trend 0.026). Specifically, those who ate more than one serving of meat per day had 16 mg/dL (95% confidence interval -4 to 37 mg/dL) higher serum total cholesterol compared with those who ate less than one serving per week. Consumption of more than seven servings of fruits and vegetables per day was associated with 13 mg/dL (95% confidence interval -28 to 3 mg/dL) lower serum total cholesterol, compared to consumption of fewer than five servings per day (P for trend 0.011). Adjusted for all other covariates, a combination of using palm oil for cooking, eating more than one serving of meat per day and fewer than one five servings of fruits and vegetables per day was associated with 46 mg/dL (95% confidence interval 17 to 75 mg/dL) (figure 2)

Variable	Interviewer- adjusted mean difference (95% CI)	P Value ^a	Fully-adjusted [,] mean difference (95% CI)	P Valueª
Major cooking oil				
Sunflower, groundnut or corn oil	Reference		Reference	
Palm oil	14 (1, 26)	0.031	17 (4, 30)	0.011
Age		0.145		0.064
40-49	Reference		Reference	
50-59	3 (-9, 15)		7(-5, 20)	
60 and above	10 (-3, 23)		15 (0, 31)	
Sex				
Male	Reference		Reference	
Female	22 (11, 32)	< 0.001	28 (16, 39)	< 0.001
Employment status		0.503		0.616
Unemployed	Reference		Reference	
Employed ^c	-7 (-20, 5)		7(-8, 22)	
Retired	-2 (-24, 21)		9 (-15, 33)	

Table 2. Adjusted mean differences for serum total cholesterol (mg/dL) for socio-demographic, dietary and lifestyle characteristics; DUCS-HTN biomarker sub-study (n=356)

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Variable	Interviewer- adjusted mean difference (95% CI)	P Value ^a	Fully-adjusted ^{,,} mean difference (95% CI)	P Valueª
Household Wealth	/	0.616		0.814
Index				
1, Poorest	Reference		Reference	
2	- 9 (- 22, 4)		-7 (-20, 6)	
3, Wealthiest	-2 (-15, 11)		0 (-14, 14)	
Meat intake	. ,	0.559		0.026
<1 serving/week	Reference		Reference	
1-6 servings/week	-1 (-19, 16)		-1 (-19, 17)	
≥1 servings/day	3 (-16, 21)		16 (-4, 37)	
Fruit and vegetable	, ,	0.023	× ,	0.011
intake				
< 5 servings/day	Reference		Reference	
5-7 servings/day	-8 (-23, 7)		-7 (-22, 9)	
> 7 servings/day	-16 (-30, -2)		-13 (-28, 3)	
Fish intake		0.579		0.639
< 1 serving/week	Reference		Reference	
1-7 servings/week	-10 (-28, 7)		-7 (-25, 11)	
>1 servings/day	-11 (-32, 10)		-7 (-29, 15)	
Dairy intake		0.853		0.628
< 1 serving/week	Reference		Reference	
1-6 servings/week	- 9 (- 22, 4)		-8 (-21, 6)	
\geq 1 servings/day	-1 (-17, 15)		-2 (-19, 15)	
Total energy intake	-5 (-11, 1)	0.095	-5(-12, 2)	0.184
(Per 1000 kcal)				
Physical activity		0.972		0.406
tertile				
1, least active	Reference		Reference	
2	2 (-12, 15)		3 (-11, 17)	
3, most active	1 (-14, 16)		8 (-8, 24)	
Alcohol consumption d				
Non-drinker	Reference			
Drinker	-7 (-21, 7)	0.308	-2 (-17, 12)	0.752

^a F test for categorical variables (major cooking oil, sex, employment status, alcohol consumption); t test for continuous variables (total energy intake); test of trend based on median value in each category for ordinal categorical variables (age category, wealth tertile, food consumption and physical activity categories)

^b Adjusted for covariates in column 1 as well as interviewer

^c Self-employed, or government job, or job in private company

^d According to report of drinking alcohol in the past 30 days

Only 11% of women and 5% of men reported having their cholesterol levels checked at least once in their lifetime (figure 3). Among those with hypercholesterolemia (n=164), 2% of men (1 of 54), and 11% of women (12 of 110) reported being notified by doctors about it, and about 2% of men and 7% of women reported taking cholesterol-lowering medications.

DISCUSSION

In this cross-sectional study among middle-aged and older adults in Dar es Salaam, Tanzania, we found a high prevalence of dyslipidemia. About a third of men and half of women had serum total cholesterol levels greater than or equal to 200 mg/dL. Serum total cholesterol was higher by 17 mg/dL among those who used palm oil as the major cooking oil, and showed a significant increasing trend with higher consumption of meat (P=0.03) and lower consumption of fruits and vegetables (P=0.01).

Mean serum total cholesterol levels for men and women were similar to that reported in other studies done in other urban populations in Tanzania. [18, 24, 25] A repeated cross-sectional study reported an increase over time in the prevalence of hypercholesterolemia in Dar es Salaam with a prevalence of about 17% in men and 7% in women in 1987 versus 30% in men and 50% in women in 1998. [24] The prevalence of hypercholesterolemia in our sample was in agreement with this trend, and also higher than that reported by studies conducted between 2008-2014 in rural Uganda (3% men, 8% women), [9] peri-urban Nigeria (2% men, 6% women), [8] and blacks in urban South-Africa (25% men, 23% women), [26] but is lower than that reported in a study from Senegal (54% men, 61% women) that included both urban and rural participants. [27]

Our estimated association between palm oil use versus unsaturated vegetable oil use is consistent with findings from dietary observational studies and intervention trials. A recent meta-analysis of 30 dietary intervention trials comparing diets rich in palm oil

 with diets rich in vegetable oils low in saturated fatty acids showed significantly higher total (0.35 mmol/L) and LDL (0.24 mmol/L) cholesterol, and slightly higher HDL cholesterol (0.02 mmol/L) 2 to 16 weeks after intervention. [28] In addition, a large case-control study conducted in Costa Rica, found that palm oil use was independently associated with 33% higher odds of myocardial infarction as compared to soybean oil use, [29] suggesting that the potentially beneficial effects of palm oil through its high vitamin A and E and oleic acid content (about 40% of total fat), may not be sufficient to compensate for the deleterious effects of its high saturated fat content, as compared to soybean oil. Higher meat consumption has previously been reported to be related to higher serum total cholesterol in Tanzania by comparing rural, urban, and pastoral populations. [12, 13] Similar to our finding, they found that consumption of meat once per day was associated with 24 mg/dL higher serum total cholesterol levels, after adjusting for age, select dietary components and resting energy expenditure. Observational studies have reported lower LDL cholesterol by about 0.06 mmol/L for every additional serving of fruits or vegetables. [30-32] The magnitude of 13 mg/dL reduction in serum total cholesterol that we found for six or more servings of fruits and vegetables per day is consistent with the reductions in LDL-cholesterol seen in these studies. We did not find significant associations between serum total cholesterol and fish intake, as reported in previous studies. [12, 13] This could have been due to insufficient statistical power or measurement error in fish intake.

Palm oil is obtained mostly from the middle section (pulp) of the fruits of the tropical plant *Elaeis guineensis*.[33] Compared with most other vegetable oils, it contains less unsaturated fat and more saturated fat (about 40 to 50% of total fat content) with the majority being palmitic acid.[33] However, compared with saturated fat from animal sources, palm oil may have a smaller impact on serum cholesterol because almost 70% of the palmitic acid in palm oil is located in the first and third position of the triglycerides, which is less absorbable than palmitic acid in the second position of the triglycerides which is the major position of palmitic acid in animal fats.[34, 35] Despite these biological differences, evidence from both observational and experimental studies, as summarized above, points to harmful effects of palm oil on serum cholesterol and CVD when substituted for unsaturated fatty acids.

Our study has several limitations. First, due to logistic and financial limitations, we measured serum total cholesterol instead of a full lipid panel. However, data from recent meta-analysis of randomized trials suggests that the effect of substituting palm oil for vegetable oils low in saturated fat on LDL cholesterol levels is far greater than that on HDL. [28] Second, although the FFO specified portion size, the assessment of diet using any method is prone to measurement error which may have resulted in bias. However, it is quite unlikely that participants with higher cholesterol levels differentially misreport using palm oil in cooking. There are some important strengths of this study. To our knowledge, this is the first population-based study to report an association between using palm oil for cooking and serum total cholesterol in East Africa. We used a reliable point-of care- lipid testing system that has good clinical agreement with laboratory reference methods for total cholesterol. [17] We systematically collected data on and adjusted for potential confounders of the association between diet and serum total cholesterol, increasing the validity of our findings. Our findings are generalizable to other peri-urban areas in Tanzania, and possibly other East African countries.

Were our findings to be replicated in other studies in East Africa using different study designs, they could have important implications for nutritional policies aimed at CVD prevention. Palm oil is now the major edible oil in Tanzania. More than half of the palm oil is imported, [36] and it is the second most imported agricultural commodity.[37] In our study population, about three-quarters of participants used palm oil as the major cooking oil with 92% of participants (n=119) in the lowest tertile of wealth using palm oil. Policy interventions to address composition of dietary fat have previously been implemented in Seychelles and Mauritius.[38, 39] In the early 1990s, Seychelles implemented a nationwide program that included increasing use of unsaturated vegetable oils, which may have contributed to the fairly high rate of decline in age-standardized CVD mortality of 44% in men and 28% in women over a 22-year period.[38,40, 41] A national NCD prevention program was also implemented in Mauritius in the late 1980s to reduce saturated fat content of cooking oil by changing the government subsidized 'ration oil' from palm oil to soybean oil. The magnitude of

reduction in serum total cholesterol of 0.8mmol/L (31 mg/dL) over a period of 5 years, [39, 42], is compatible with the estimated impact of replacing palm oil with unsaturated vegetable oil estimated in a recent meta-analysis of dietary intervention trials. [43]

Similar policies can be considered to reduce palm oil intake in Tanzania. However, it should be noted that edible oils are viewed as necessities, [44] and increasing their price (e.g. by taxation) without providing healthier alternatives at comparable prices may have unintended adverse consequences on overall energy consumption or food security. [45-47] Reducing saturated fat in edible oil by mixing it with other unsaturated oils has been proposed as a potential intervention in Costa Rica, Singapore and India. [29, 48, 49] and could be a feasible short-term intervention in Tanzania. The long-term focus should be on incentivizing the production, import and use of healthier oils. Our results also indicate that policies to increase fruit and vegetable intake, and to reduce meat intake would be beneficial in lowering serum cholesterol. In the meantime, there is an urgent need to improve the capacity of the healthcare system in Tanzania to improve the diagnosis and treatment of dyslipidemias as recommended by the WHO. [50] Some of the first steps in this regard are: training of healthcare professionals, developing and adopting clinical guidelines for diagnosis and treatment of dyslipidemia and provision of appropriate lab equipment to district and local hospitals.

WHAT THIS PAPER ADDS

What is already known on this subject

- Dyslipidemia is a well-known risk factor for cardiovascular diseases and is becoming increasingly common in Sub-Saharan Africa, especially in urban areas.
- Diet is an established lifestyle determinant of cardiovascular diseases, and diet modification can be an effective primary prevention strategy.
- Evidence on the dietary determinants of dyslipidemia in sub-Saharan Africa is limited and unclear with regard to the magnitude of the association.

What this study adds

- This cross-sectional study in a peri-urban Tanzanian population suggests that the prevalence of undiagnosed and untreated dyslipidemia is quite high among middle aged and older adults.
- Use of cooking oil low in saturated fat, reducing meat consumption to less than 1 serving per week, and increasing fruit and vegetable consumption to 7 or more servings per day may lower mean serum total cholesterol levels up to 46 mg/dL.

CONTRIBUTORS GD, JK, GHL and RMZ conceived and designed the study. GD obtained the funding. SSK and SF analyzed the data. GD and SSK interpreted the data and drafted the article, which was revised critically for substantive content by all authors, who approved the final version for publication. GD, SSK, SF and RMZ had full access to all of the data (including statistical reports and tables) in the study and can take responsibility for the integrity of the data and the accuracy of the data analysis. GD is the study guarantor.

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COMPETING INTERESTS: All authors have completed the ICMJE uniform disclosure form at <u>www.icmje.org/coi_disclosure.pdf</u> (available on request from the corresponding author) and declare: no support from companies for the submitted work; no relationships with companies that might have an interest in the submitted work in the previous three years; their spouses, partners, or children have no financial relationships that may be relevant to the submitted work; no non-financial interests that may be relevant to the submitted work.

ETHICAL APPROVAL: The study protocol was approved by the Institutional Review Board of the Harvard TH Chan School of Public Health and the Research and Publications Committee of Muhimbili University of Health and Allied Sciences. All participants gave informed consent.

DATA SHARING: No additional data available

TRANSPARENCY: The lead authors affirm that the manuscript is an honest, accurate, and transparent account of the study being reported; that no important aspects of the study have been omitted; and that any discrepancies from the study as planned have been explained.

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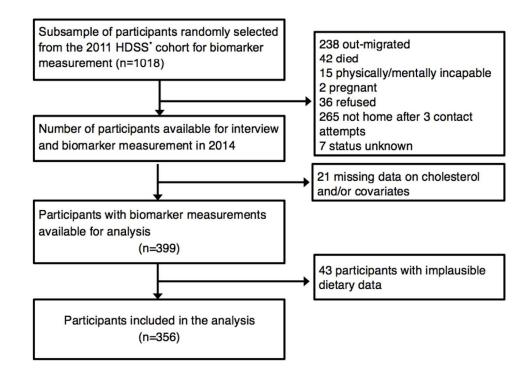
Figure legends

 Figure 1. Selection of study participants, DUCS-HTN biomarker sub-study, 2014

Figure 2. Association between diet and serum total cholesterol. Values were adjusted for age, sex, socioeconomic status, food consumption frequency, total energy intake, major cooking oil used, physical activity and alcohol consumption.

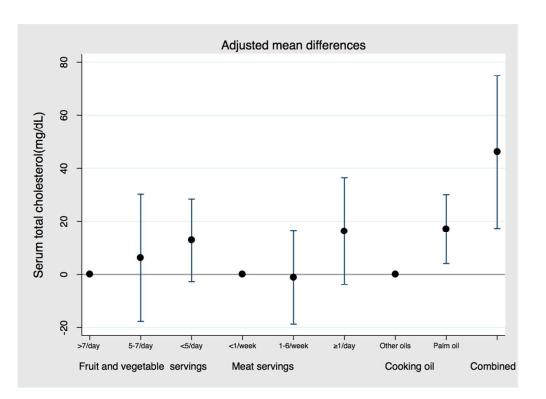
Figure 3. Diagnosis, awareness and treatment of hypercholesterolemia in men (Part A; n=54 for all percentages) and women (Part B; n=110 for all percentages) in the DUCS-HTN biomarker sub-study, 2014.

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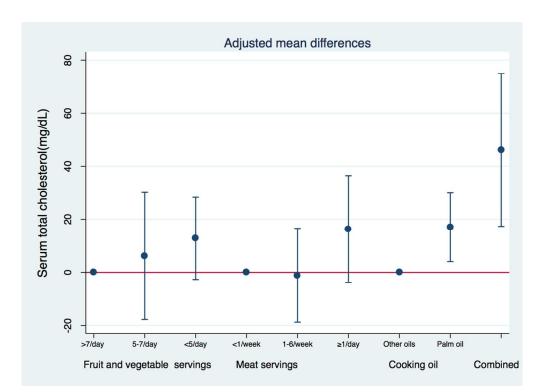


^{*}Dar es salaam Health and Demographic Surveillance System

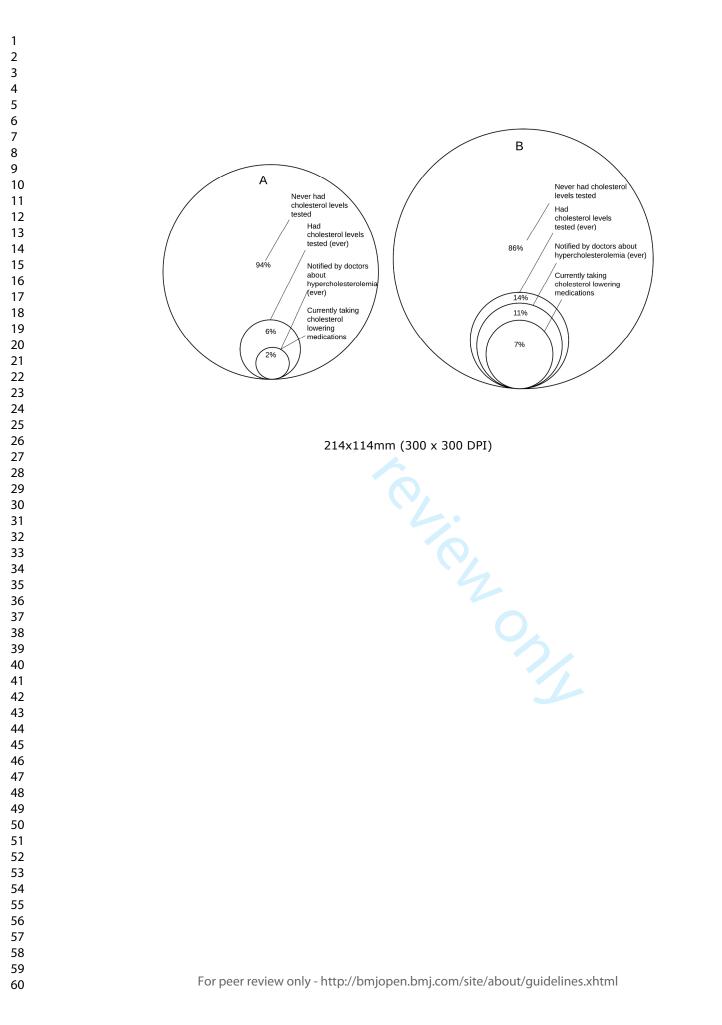
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	Item No	Recommendation
Title and abstract	1	(a) Indicate the study's design with a commonly used term in the title or the abstrac
		[Within the title page 1 and method section of the abstract page 2]
		(b) Provide in the abstract an informative and balanced summary of what was done
		and what was found [See results section of the abstract page 2]
Introduction		
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported [page 4]
Objectives	3	State specific objectives, including any prespecified hypotheses [page 4]
Methods		
Study design	4	Present key elements of study design early in the paper [page 5]
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment,
-		exposure, follow-up, and data collection [page 5]
Participants	6	(a) Give the eligibility criteria, and the sources and methods of selection of
		participants [page 5]
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effec
		modifiers. Give diagnostic criteria, if applicable [page 5-6]
Data sources/	8*	For each variable of interest, give sources of data and details of methods of
measurement		assessment (measurement). Describe comparability of assessment methods if there
		more than one group [page 5-6]
Bias	9	Describe any efforts to address potential sources of bias [page 6]
Study size	10	Explain how the study size was arrived at [page 5]
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable,
		describe which groupings were chosen and why [page 6]
Statistical methods	12	(a) Describe all statistical methods, including those used to control for confounding
		[page 6-7]
		(b) Describe any methods used to examine subgroups and interactions [page 7]
		(c) Explain how missing data were addressed [page 6]
		(d) If applicable, describe analytical methods taking account of sampling strategy
		[N/A]
		(e) Describe any sensitivity analyses [N/A]
Results		
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially
_		eligible, examined for eligibility, confirmed eligible, included in the study,
		completing follow-up, and analysed [page 7 and Figure 1]
		(b) Give reasons for non-participation at each stage [Figure 1]
		(c) Consider use of a flow diagram [Figure 1]
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and
Ł		information on exposures and potential confounders [page 7]
		(b) Indicate number of participants with missing data for each variable of interest
		[Figure 1]
Outcome data	15*	Report numbers of outcome events or summary measures [page 7-8 and Figure 3]
Main results	16	(<i>a</i>) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and
		their precision (eg, 95% confidence interval). Make clear which confounders were
		response (0, 20, 20, 20, 20, 20, 20, 20, 20, 20, 2

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		(b) Report category boundaries when continuous variables were categorized [table
		_1]
		(c) If relevant, consider translating estimates of relative risk into absolute risk for a
		meaningful time period [N/A]
Other analyses	17	Report other analyses done-eg analyses of subgroups and interactions, and
		sensitivity analyses [N/A]
Discussion		
Key results	18	Summarise key results with reference to study objectives [page 14]
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or
		imprecision. Discuss both direction and magnitude of any potential bias [page 16]
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations,
		multiplicity of analyses, results from similar studies, and other relevant evidence
		[page 14,16]
Generalisability	21	Discuss the generalisability (external validity) of the study results [page 17]
Other information		6
Funding	22	Give the source of funding and the role of the funders for the present study and, if
		applicable, for the original study on which the present article is based [page 20]

*Give information separately for exposed and unexposed groups.

Note: An Explanation and Elaboration article discusses each checklist item and gives methodological background and published examples of transparent reporting. The STROBE checklist is best used in conjunction with this article (freely available on the Web sites of PLoS Medicine at http://www.plosmedicine.org/, Annals of Internal Medicine at http://www.annals.org/, and Epidemiology at http://www.epidem.com/). Information on the STROBE Initiative is available at www.strobe-statement.org.

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Dietary determinants of serum total cholesterol among middle-aged and older adults: a population-based crosssectional study in Dar es Salaam, Tanzania

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Primary Subject Heading :	Nutrition and metabolism
Secondary Subject Heading:	Epidemiology, Public health, Global health, Cardiovascular medicine
Keywords:	cholesterol, palm oil, diet, Nutrition < TROPICAL MEDICINE, Tanzania, sub- Saharan Africa

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3 4	1	Dietary determinants of serum total cholesterol among middle-aged and older adults:
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6 7	2	a population-based cross-sectional study in Dar es Salaam, Tanzania
8 9	3	
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1 2	ABSTRACT
3	Objective: To assess the dietary determinants of serum total cholesterol.
4	Design: Cross-sectional population-based study.
5	Setting: Peri-urban region of Dar es Salaam, Tanzania.
6	Participants: 347 adults aged 40 years and older from the Dar es Salaam Urban Coho
7	Hypertension Study (DUCS-HTN).
8	Main outcome measure: Serum total cholesterol measured using a point-of-care de
9	Results: Mean serum total cholesterol level was 204 mg/dL (Interquartile range (IQR
10	169 to 236 mg/dL) in women and 185 mg/dL (IQR 152 to 216 mg/dL) in men. After
11	adjusting for demographic, socioeconomic, lifestyle and dietary factors, participants
12	who reported using palm oil as the major cooking oil had serum total cholesterol hig
13	by 15 mg/dL (95% confidence interval 1 to 29 mg/dL) compared with those who
14	reported using sunflower oil. Consumption of one or more servings of meat per day
15	for trend= 0.017) and less than five servings of fruits and vegetables per day (P for
16	trend= 0.024) were also associated with higher serum total cholesterol. A combination
17	of using palm oil for cooking, eating more than one serving of meat per day and few
18	than five servings of fruits and vegetables per day was associated with 46 mg/dL (95
19	confidence interval 16 to 76 mg/dL) higher serum total cholesterol.
20	Conclusions: Using palm oil for cooking was associated with higher serum total
21	cholesterol levels in this peri-urban population in Dar es Salaam. Reduction of satura
22	fat content of edible oil may be considered as a population-based strategy for prima
23	prevention of cardiovascular diseases.
24	Key words: cholesterol, palm oil, diet, nutrition, Tanzania, Sub-Saharan Africa
25	
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29	STRENGTHS AND LIMITATIONS OF THIS STUDY

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•	This is the first population-based study to report an association between using palm		
	oil for cooking and serum total cholesterol in East Africa.		
•	A reliable point-of care lipid testing system was used to measure serum total		
	cholesterol that has good clinical agreement with laboratory reference methods.		
•	Potential confounders of the association between diet and serum total cholesterol		
	were systematically measured and adjusted for.		
•	Only serum total cholesterol was measured instead of a full lipid panel, due to logistic		
	and financial limitations.		
•	The relatively small sample size may have resulted in insufficient statistical power to		
	detect some associations.		
•	The assessment of diet using any method is prone to measurement error, which may		
	have resulted in bias, although differential misreporting of cooking oil by cholesterol		
	level is unlikely.		
	•		

1 INTRODUCTION

Sub-Saharan Africa is facing a burgeoning epidemic of non-communicable diseases
(NCDs) due to changing demographic profiles, epidemiologic transition, urbanization,
and lifestyle changes as a result of economic development as well as increasing survival
of HIV patients.[1, 2] In 2015, one third of all estimated NCD deaths in sub-Saharan
Africa were due to cardiovascular diseases (CVD).[3] By 2030, the share of all deaths
attributable to CVD is projected to increase by nearly 40% in this region.[3]

9 Dyslipidaemia is a well-known risk factor for ischemic heart disease (IHD), stroke, and 10 other vascular diseases.[4, 5] In middle-aged adults, IHD mortality is lower by as much 11 as 30 to 50% for every 1 mmol/L (38.6mg/dL) lower serum total cholesterol.[5] Sub-12 Saharan Africans have historically had a favorable lipid profile [6] but hyperlipidaemia is 13 becoming increasingly common in these countries, especially in urban areas.[7-9] It is 14 estimated that thirty-four percent of ischemic heart disease deaths in Tanzania are 15 attributable to dyslipidaemia.[10]

A few studies have already explored the relationship between diet and serum total cholesterol in eastern sub-Saharan Africa and reported an association between higher intake of meat and lower intake of fish and green vegetables with higher serum total cholesterol. [11-15] However, two of these studies compared rural and coastal Bantu populations that are known to have, respectively, vegetarian and fish-based diets and did not measure diet in each participant and based their conclusions on known dietary differences between ethnic populations. [14, 15] Another study included participants from rural, urban and pastoral regions of Tanzania but did not report the magnitude of the association[12] The other two studies conducted in urban Tanzania did not adjust for potential confounders of the relationship between diet and serum total cholesterol. [11, 13] There are a few studies from other parts of sub-Saharan Africa that have looked at the association between diet and serum cholesterol. A study conducted in a rural population in Uganda found that eating fewer than 5 servings of fruits and

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1	vegetables daily was associated with higher odds of low HDL cholesterol among men,
2	but lower odds among women.[16] Another study in urban Uganda found lower mean
3	serum total cholesterol in lacto-ovo-vegetarians compared to non-vegetarians.[17]
4	
5	Another potential dietary determinant of serum cholesterol is the type of oil used in
6	cooking.[18,19] Using cooking oils with higher saturated fat content such as coconut
7	and palm oil has been associated with higher serum total cholesterol levels in clinical
8	trials and observational studies.[20, 21] Population-level differences in CVD morbidity
9	and mortality is associated with differences in type of cooking oil in eastern
10	European, [22] Costa Rica, [23] and Singapore. [24] Mauritius, Seychelles and Singapore
11	have already implemented policy schemes to reduce saturated fat content of cooking oil
12	as population-level interventions to reduce burden of CVD.[25-27]
13	A thorough understanding of the role of dietary determinants of serum cholesterol will
14	be invaluable in designing population-level interventions to address the growing CVD
15	burden especially considering that treatment for dyslipidaemia is either unavailable or
16	prohibitively expensive for most patients in low-income countries.[28] Tanzanian diet
17	consists predominantly of carbohydrates (mainly maize, sweet potatoes, cassava and
18	rice) consumed with green leafy vegetables, beans or peas and occasionally with meat
19	or fish.[29] Palm oil is the major cooking oil, followed by sunflower oil.
20	We conducted a cross-sectional population-based study to assess the association of
21	type of cooking oil (palm vs sunflower oil) and diet (meat, fish, fruit and vegetable
22	consumption) with serum total cholesterol in an adult population in a peri-urban area of
23	Dar es Salaam. We hypothesized that use of palm oil for cooking and higher intake of
24	meat and lower intake of fruits and vegetables will be associated with higher serum
25	total cholesterol.
26	METHODS

27 Study design and population

The Dar es Salaam Health and Demographic Surveillance System (HDSS) is a demographic surveillance system in Ukonga and Gongo la Mboto wards of Dar es Salaam, Tanzania with over 100,000 enumerated individuals living in 21,000 households followed since 2011/12. For the Dar es Salaam Urban Cohort Hypertension Study (DUCS-HTN), we randomly selected one large and one small neighborhood in the HDSS and contacted all adult residents aged 40 years and older, who had been registered in the HDSS database in 2011 or 2012. A random subsample of one-fifth of the eligible participants was selected for additional assessment of diet and measurement of serum total cholesterol. The size of this subsample was determined largely based upon the availability of resources. Pregnant women and those who were physically and mentally incapable of completing the interview and measurements were excluded. The participant selection flowchart is shown in Figure 1. Face-to-face interviews and measurements were conducted in participant's home from March to June 2014 by six trained interviewers. All interviewers had previous experience conducting public health surveys. For this study, interviewers received five days of training and two days of field practice in a nearby neighborhood. At least three attempts were made to contact all potential participants. Written informed consent was obtained from all participants. The study protocol was approved by the Institutional Review Board of the Harvard TH Chan School of Public Health and the Research and Publications Committee of Muhimbili University of Health and Allied Sciences. Measurements Non-fasting serum total cholesterol was measured from 30 micro-liters of finger-prick capillary blood samples using Cardiochek PA devices (Polymer Technology Systems Inc, IN, USA).[30, 31] The coefficient of variation for this device has been determined to be in the range of 1.3 to 2.9%.[30]

26 Data on diet was collected using a semi-quantitative food frequency questionnaire
27 (FFQ). The FFQ assessed intake of 179 food and drink items over the past 30 days. A

28 shorter 85-item version of this instrument has been previously used in Tanzania.[32]

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Nutrient intakes were calculated by multiplying the frequency of food consumption measured from FFQ by the nutrient content of the specified portion size using the Tanzanian food composition table. [33] Meat intake was defined to include all red and white meat consumption, excluding fish consumption. Fruit and vegetable intake was defined to include consumption of all fruits and vegetables, except for root vegetables and fruit juice. An additional question was asked to assess the main type of cooking oil: "What is the kind of oil used most often in your home for frying food?". Frying is the most common cooking technique involving oil use in this region. The reference to a cooking technique was made to distinguish it from other oils that may be used in a household, such as fuels.

All participants completed a socioeconomic, demographic and lifestyle questionnaire and had their height, weight, waist circumference, hip circumference, and blood pressure measured by trained interviewers. Some demographic information (age, sex, neighborhood, components of wealth index) was taken from the HDSS baseline interview. Body mass index was calculated as weight (kg) divided by height (m) squared, and categorized as underweight (<18.5), normal (18.5 to <25.0), overweight (25.0 to <30.0), and obese (≥30.0).[34] Physical activity was assessed using the Global Physical Activity Questionnaire in the domains of work, transportation, and leisure, [35] and categorized into tertiles of Metabolic Equivalent (MET) hours per week. To measure socioeconomic status, we created a household wealth index using a principal component analysis of household characteristics (floor material, roof material, wall material, electricity), household assets (television, radio, shop, sewing machine, sofa, fan, iron, stove with oven, stove without oven, dining table, cupboard, watch, mobile phone, bike, motorcycle, cart, car, motorboat), and animal ownership (goats, sheep, chicken/ducks, pigs).[36] The household wealth index was categorized into tertiles.

26 Statistical analysis

We excluded participants with implausible dietary data (total energy intake <500 or
>5000 kcal/day), and with missing data on serum total cholesterol, diet and/or any of
the potential covariates.

Chi-square tests were used to assess binary variables, F-tests were used for categorical variables and tests of linear trend based on median value within each category for ordinal variables with a linear association with serum total cholesterol (i.e. age, wealth index, and food consumption categories). Mean differences and 95% confidence intervals in serum total cholesterol across categories of dietary factors, including cooking oil were estimated using multivariable linear regression. Potential confounders were included in the model based on a-priori knowledge of lifestyle and socio-demographic determinants of diet and serum total cholesterol. The fully adjusted model included age, sex, total energy intake, physical activity, alcohol consumption, wealth index and employment status as covariates. We also adjusted for interviewer to remove the effect of any extraneous variation in measurement of subjective risk factors across interviewers. [37, 38] We examined interactions of potential determinants of serum total cholesterol with sex, but no interactions were statistically significant at 0.2 level, therefore we did not include these product terms in the final model. We conducted sensitivity analyses by excluding 16 participants who reported either being previously diagnosed with hypercholesterolaemia, or currently taking cholesterol-lowering medications or being advised by a health professional to modify diet to lower cholesterol. All analyses were performed using STATA software version 13.1 (STATA Corporation, College Station, Texas). RESULTS A total of 420 participants were enrolled for the random subsample with diet and cholesterol measurements. Among these, data on serum total cholesterol and selected covariates was available for 399 participants. We excluded 43 participants due to implausible caloric intake. In addition, after descriptive analyses we excluded 9 (2%) participants who reported using cooking oils other than palm or sunflower oil. The final

analytic sample included 347 participants (FIGURE 1).

29 Forty-two percent of the participants (n=347) were men (TABLE 1). Mean age of men

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1	was 55 years (SD 11), and that of women was 52 (SD 10). 44% of the women (n=202)
2	and 19% of men (n=145) were obese, and one third of men and women were
3	overweight. Mean energy intake per day was 2413 kcal. Thirty-seven percent of men
4	and 54% of women were hypercholesterolemic (i.e. had total cholesterol ≥200 mg/dL).
5	Mean serum total cholesterol in women was 204 mg/dL (95% confidence interval 197 to
6	211 mg/dL) and was significantly higher than that of men at 185 mg/dL (177 to 193
7	mg/dL). Serum total cholesterol levels were higher in older age groups.
8	79% of participants reported using palm oil as the major cooking oil and the rest
9	reported using sunflower oil (TABLE 1). Those who reported using palm oil were most
10	likely to be in the poorest tertile of wealth, whereas those who reported using
11	sunflower oil were most likely to be in the wealthiest tertile (p<0.001). Those who
12	reported using sunflower oil had a higher energy intake (p=0.04), and reported

consuming meat (p=0.004) and dairy (p=0.02) more frequently than those who reported

using palm oil. Mean serum total cholesterol in those who reported using palm oil was

199 mg/dL (193 to 205) and was significantly higher than those who reported using

sunflower oil 184 mg/dL (173 to 195).

	All (n=347)	Men (n=145)	Women (n=202)	Use palm oil	Use sunflower oil	
Variable				(n=274)	(n=73)	
Age						
40-49	150 (43)	56 (39)	94 (47)	123 (45)	27 (37)	
50-59	113 (33)	43 (30)	70 (35)	82 (30)	31 (42)	
60 and above	84 (24)	46 (32)	38 (19)	69 (25)	15 (21)	
Employment status						
Unemployed	83 (24)	14 (10)	69 (34)	70 (26)	13 (18)	
Employed (includes self-employed)	238 (69)	112 (77)	126 (62)	183 (67)	55 (75)	
Retired	26 (7)	19 (13)	7 (3)	21 (8)	5 (7)	
Household wealth index						
Tertile 1, Poorest	115 (33)	49 (34)	66 (33)	109 (40)	6 (8)	
Tertile 2	117 (34)	53 (37)	64 (32)	90 (33)	27 (37)	
Tertile 3, Wealthiest	115 (33)	43 (30)	72 (36)	75 (27)	40 (55)	
BMI (kg/m²)						
Underweight (<18.50)	19 (5)	13 (9)	6 (3)	19 (7)	0 (0)	
Normal weight (18.50-<25.00)	100 (29)	56 (39)	44 (22)	91 (33)	9 (12)	
Overweight (25-<30.00)	112 (32)	48 (33)	64 (32)	81 (30)	31 (42)	
Obese (≥ 30.00)	116 (33)	28 (19)	88 (44)	83 (30)	33 (45)	
Physical activity						
Tertile 1,	123 (35)	46 (32)	77 (38)	96 (35)	27 (37)	
(0-14 MET-hours/week)	123 (33)	+0 (JZ)	(30)	50 (55)	27 (37)	
Tertile 2,	108 (31)	38 (27)	70 (35)	86 (31)	22 (30)	
(15-112 MET-hours/week) Tertile 3,						
(113-840 MET-hours/week)	116 (33)	61 (42)	55 (27)	92 (34)	24 (33)	

Table 1. Participants' characteristics in DUCS-HTN biomarker sub-study, 2014

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Variable	All (n=347)	Men (n=145)	Women (n=202)	Use palm oil (n=274)	Use sunflower oi (n=73)
Alcohol drinking ^a					
Non-drinker	286 (82)	103 (71)	183 (91)	225 (82)	61 (84)
Smoking					
Non-smoker	278 (81)	85 (59)	193 (97)	210 (78)	68 (93)
Former smoker	43 (13)	39 (27)	4 (2)	38 (14)	5 (7)
Current smoker	22 (6)	19 (13)	3 (2)	22 (8)	0 (0)
DIETARY VARIABLES					
Palm oil as major cooking oil	274 (79)	119 (82)	155 (77)		
Meat intake					
<1 serving/week	41 (12)	21 (14)	20 (10)	36 (13)	5 (7)
1-6 servings/week	173 (50)	55 (38)	118 (58)	145 (53)	28 (38)
≥1 servings/day	133 (38)	69 (48)	64 (32)	93 (34)	40 (55)
Fish intake					
< 1 serving/week	41 (12)	13 (9)	28 (14)	31 (11)	10 (14)
1-7 servings/week	196 (56)	76 (52)	120 (59)	154 (56)	42 (58)
> 1 servings/day	110 (32)	56 (39)	54 (27)	89 (32)	21 (29)
Dairy intake					
< 1 serving/week	159 (46)	63 (43)	96 (48)	136 (50)	23 (32)
1-6 servings/week	133 (38)	65 (45)	68 (34)	99 (36)	34 (47)
≥ 1 servings/day	55 (16)	17 (12)	38 (19)	39 (14)	16 (22)
Fruit and vegetable					
< 5 servings/day	121 (35)	49 (34)	72 (36)	101 (37)	20 (27)
5-7 servings/day	68 (20)	26 (20)	42 (21)	54 (20)	14 (19)

Variable	All (n=347)	Men (n=145)	Women (n=202)	Use palm oil (n=274)	Use sunflower oi (n=73)
>7 servings/day	158 (46)	70 (48)	88 (44)	119 (43)	39 (53)
Nuts and legumes					
< 1 serving/week	27 (8)	7 (5)	20 (10)	17 (6)	10 (14)
1-7 servings/week	200 (58)	87 (60)	113 (56)	162 (59)	38 (52)
> 1 servings/day	120 (35)	51 (35)	69 (34)	95 (35)	25 (34)
Total energy intake (kcal/day) ^b	2413 (1701, 3215)	2629 (1780, 3216)	2313 (1617, 3207)	2369 (1654 <i>,</i> 3150)	2743 (1865, 3606)
Fat intake ^b					
Total fat (% of energy)	17 (15, 20)	17 (15, 21)	17 (15, 20)	17 (14, 20)	20 (15, 22)
Saturated fat (% energy)	10 (8, 13)	10 (8, 13)	10 (8, 12)	10 (8, 13)	10 (8, 11)
Monounsaturated fat (% of energy)	4 (3, 5)	4 (3, 5)	4 (3, 5)	4 (3 <i>,</i> 5)	4 (4, 5)
Polyunsaturated fat (% of energy)	2 (1, 2)	2 (1, 2))	2 (1, 2)	1 (1, 2)	2 (1, 2)
Cholesterol					
Serum total cholesterol (mg/dl) ^c	196 (3)	185 (4)	204 (4)	199 (3)	184 (6)
Hypercholesterolaemia ^d	164 (47)	54 (37)	110 (54)	135 (49)	29 (40)
Cholesterol level was previously checked	29 (18)	7 (13)	22 (20)	13 (10)	16 (55)
Previously diagnosed hypercholesterolaemia	13 (8)	1 (2)	12 (11)	5 (4)	8 (28)
Currently taking cholesterol lowering medications	9 (5)	1 (2)	8 (7)	3 (2)	6 (21)

Number (%) reported for all variables, except as specified b and c below ^a According to report of drinking alcohol in the past 30 days ^b Total energy and fat intake, which is reported as median (interquartile range)

. a (standard error) J. al cholesterol 2 200 mg/dL ^c Serum total cholesterol, reported as mean (standard error) ^d Hypercholesterolaemia defined as total cholesterol \geq 200 mg/dL

In the minimally adjusted model (with interviewer as the only covariate), those who reported using palm oil as the major cooking oil had serum total cholesterol levels higher by 11 mg/dL (-2 to 24 mg/dL) compared with those who reported using sunflower oil (TABLE 2). After adjusting for covariates, participants who used palm oil for cooking had 15 mg/dL (1 to 29 mg/dL) higher serum total cholesterol (FIGURE 2). The negative confounding was mostly due to the negative correlation between female gender and meat intake with palm oil use (both variables were positively correlated with serum total cholesterol). Serum total cholesterol was also higher in participants who ate meat more frequently (P for trend 0.017) after adjusting for potential confounders. Specifically, those who ate more than one serving of meat per day had 18 mg/dL (-3 to 39 mg/dL) higher serum total cholesterol compared with those who ate less than one serving per week. In this relationship, higher total energy intake was the major confounder with a positive correlation with meat intake but negative correlation with serum total cholesterol. Consumption of more than seven servings of fruits and vegetables per day was associated with 13 mg/dL (-29 to 3 mg/dL) lower serum total cholesterol, compared to consumption of fewer than five servings per day (P for trend 0.024). A combination of using palm oil for cooking, eating more than one serving of meat per day and fewer than five servings of fruits and vegetables per day was associated with 46 mg/dL (16 to 76 mg/dL) higher serum total cholesterol. We found similar associations after excluding 16 participants who reported either being previously diagnosed with hypercholesterolaemia, or currently taking cholesterol lowering medications or being advised by a health professional to modify diet to lower

- cholesterol. Those who used palm oil had 17 mg/dL (2 to 32 mg/dL) higher serum total
 cholesterol compared with those who used sunflower oil (TABLE 2).

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Table 2. Adjusted mean differences for serum total cholesterol (mg/dL) by dietary factors; DUCS-HTN biomarker sub-study

Variable	Interviewer- adjusted mean difference (95% Cl)	P Value ^a	Fully-adjusted ^b mean difference (95% CI) (n=347)	P Value ^ª	Fully-adjusted ^c mean difference after excluding 16 participants diagnosed or treated for dyslipidaemia (95% CI) (n=331)	P Value ^ª
Major cooking oil		0.092		0.033		0.028
Sunflower oil	Reference		Reference		Reference	
Palm oil	11 (-2, 24)		15 (1, 29)		17 (2, 32)	
Meat intake		0.485		0.017		0.031
<1 serving/week	Reference		Reference		Reference	
1-6 servings/week	-1 (-19, 16)		-1 (-19, 17)		-2 (-20, 17)	
≥1 servings/day	4 (-15, 22)		18 (-3, 39)		16 (-5, 37)	
Fruit and vegetable		0.028		0.011		0.024
intake						
< 5 servings/day	16 (2,30)		13 (-3, 29)		12 (-5, 29)	
5-7 servings/day	10 (-6, 25)		8 (-8, 24)		10 (-7, 26)	
> 7 servings/day	Reference		Reference		Reference	
Fish intake		0.399		0.446		0.309
< 1 serving/week	14 (-8, 36)		10 (-13, 33)		14 (-9, 38)	
1-7 servings/week	3 (-11, 17)		2 (-13, 17)		4 (-11, 20)	
>1 servings/day	Reference		Reference		Reference	
Dairy intake		0.802		0.572		0.690
< 1 serving/week	Reference		Reference		Reference	
1-6 servings/week	-9 (-22,4)		-7 (-21, 6)		-7 (-21, 7)	

Variable	Interviewer- adjusted mean difference (95% CI)	P Value ^ª	Fully-adjusted ^b mean difference (95% Cl) (n=347)	P Value ^a	Fully-adjusted ^c mean difference after excluding 16 participants diagnosed or treated for dyslipidaemia (95% CI) (n=331)	P Value ^ª
≥ 1 servings/day	-2 (-18, 15)		-2 (-20, 15)		-1 (-20, 17)	
Total energy intake (Per 1000 kcal)	-8 (-21, 5)	0.226	-5 (-12, 3)	0.209	-4 (-11, 4)	0.365
Age		0.106		0.071		0.099
40-49	Reference		Reference		Reference	
50-59	4 (-9, 16)		6 (-6, 19)		5 (-9, 18)	
60 and above	11 (-2, 25)		16 (0, 31)		15 (-1, 31)	
Sex		<0.001		<0.001		<0.001
Male	Reference		Reference		Reference	
Female	21 (10, 32)		27 (15, 39)		24 (11, 36)	
Employment status		0.432		0.567		0.581
Unemployed	Reference		Reference		Reference	
Employed ^d	-7 (-20, 6)		7 (-9, 22)		5 (-12, 21)	
Retired	2 (-21, 24)		12 (-13, 36)		13 (-12, 39)	
Household Wealth		0.692		0.754		0.952
Index						
1, Poorest	Reference		Reference		Reference	
2	-9 (-22, 4)		-8 (-21, 5)		-7 (-20, 7)	
3, Wealthiest	-1 (-15, 12)		0 (-14, 14)		2 (-13, 16)	
Physical activity tertile		0.953		0.430		0.599
1, least active	Reference		Reference		Reference	

Variable	Interviewer- adjusted mean difference (95% CI)	P Value ^ª	Fully-adjusted ^b mean difference (95% CI) (n=347)	P Value ^a	Fully-adjusted ^c mean difference after excluding 16 participants diagnosed or treated for dyslipidaemia (95% Cl) (n=331)	P Value ^a
2	1 (-13, 15)		2 (-12, 16)		4 (-11, 18)	
3, most active	0 (-15, 15)		7 (-9, 24)		6 (-11, 24)	
Alcohol consumption ^e		0.315		0.666		0.641
Non-drinker	Reference		Reference		Reference	
Drinker	-7 (-21,7)		-3 (-18, 11)		-4 (-18, 11)	

^a F2test for categorical variables (cooking oil, sex, employment status, alcohol consumption); t test for continuous variables (total energy intake); test of trend based on median value in each category for ordinal categorical variables (age category, wealth index tertile, food consumption and physical activity categories)

 $^{\text{b}}$ AB justed for covariates in column 1 as well as interviewer

^c Excluding those previously diagnosed with hypercholesterolaemia; adjusted for covariates in column 1 as well as interviewer

^d S*e*lf-employed, or government job, or job in private company

^e A8 cording to report of drinking alcohol in the past 30 days

1 Only 11% of women and 5% of men reported having ever had their cholesterol levels

2 checked (TABLE 1; FIGURE 3). Among those with hypercholesterolaemia (n=164), 2% of

3 men (1 of 54), and 11% of women (12 of 110) reported being notified by doctors about

4 it, and 2% of men and 7% of women reported taking cholesterol-lowering medications.

DISCUSSION

In this cross-sectional study among middle-aged and older adults in a peri-urban ward in Dar es Salaam, Tanzania, we found a high prevalence of hypercholesterolaemia. Slightly more than a third of men and half of women had serum total cholesterol levels greater than or equal to 200mg/dL. After adjusting for major confounders, serum total cholesterol was higher by 15 mg/dL among those who used palm oil compared to those who used sunflower oil as the major cooking oil, and showed a significant increasing trend with higher consumption of meat (P=0.017) and lower consumption of fruits and vegetables (P=0.01).

Mean serum total cholesterol levels for men and women were similar to that reported in other studies conducted in other urban Tanzanian populations. [32, 39, 40] A repeated cross-sectional study reported an increase over time in the prevalence of hypercholesterolaemia in Dar es Salaam with a prevalence of 17% in 1987 versus 30% in 1998 in men, and 7% versus 50% in women. [39] The prevalence of hypercholesterolaemia in our sample was higher than that reported by studies conducted between 2008-2014 in rural Uganda (3% men, 8% women),[9] peri-urban Nigeria (2% men, 6% women),[8] and blacks in urban South-Africa (25% men, 23% women),[41] but is lower than that reported in a study from Senegal (54% men, 61% women) that included both urban and rural participants.[42] Our estimated association between palm oil versus sunflower oil use and serum total cholesterol is consistent with findings from dietary observational studies and intervention trials. A recent meta-analysis of 30 dietary intervention trials comparing

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diets rich in palm oil with diets rich in vegetable oils low in saturated fatty acids showed
significantly higher total (13.5 mg/dL) and LDL (9.3 mg/dL) cholesterol, and slightly
higher HDL cholesterol (0.8 mg/dL) 2 to 16 weeks after intervention.[21] In addition, a
large case-control study conducted in Costa Rica found that palm oil use compared to
soybean oil use was independently associated with 33% higher odds of myocardial
infarction. [23].

Higher meat consumption has previously been reported to be associated with higher serum total cholesterol in Tanzania. [12, 13] Similar to our finding, daily consumption of meat was found to be associated with 24 mg/dL higher serum total cholesterol levels, after adjusting for age, select dietary components and resting energy expenditure.[13] Observational studies have also reported lower LDL cholesterol by 2.3mg/dL for every additional serving of fruits or vegetables.[43-45] The magnitude of 13 mg/dL reduction in serum total cholesterol that we found for six or more servings of fruits and vegetables per day is consistent with the reductions in LDL-cholesterol seen in these studies. We did not find significant associations between serum total cholesterol and fish intake in men, as reported in previous studies. [12, 13] This could have been due to insufficient statistical power due to smaller expected effect size for fish intake or higher measurement error.

Palm oil is obtained mostly from the middle section (pulp) of the fruits of the tropical plant *Elaeis quineensis*.[46] Compared with most other vegetable oils, it contains less unsaturated fat and more saturated fat (nearly 40 to 50% of total fat content) with the majority being palmitic acid. [46] However, compared with saturated fat from animal sources, palm oil may have a smaller impact on serum cholesterol because almost 70% of the palmitic acid is located in the first and third position of the triglycerides, which is less absorbable than those in the second position (which is the most common position of palmitic acid in animal fats). [47, 48] Despite these biological differences, evidence from both observational and experimental studies, as summarized above, points to

harmful effects of palm oil on serum cholesterol and CVD when substituted for
 unsaturated fatty acids.

Our study has several limitations. First, due to logistic and financial limitations, we measured serum total cholesterol instead of a full lipid panel. However, data from recent meta-analysis of randomized trials suggests that the effect of substituting palm oil for vegetable oils low in saturated fat on LDL cholesterol levels is larger than that on HDL.[21] Second, the assessment of diet using any method is prone to measurement error which may have resulted in bias. However, it is unlikely that participants with higher serum cholesterol differentially misreport using palm oil in cooking, limiting the impact of such measurement error on our results. Thirdly, the relatively small sample size may have resulted in insufficient statistical power to detect some associations (e.g. with fish intake). There are some important strengths of this study. To our knowledge, this is the first population-based study to report an association between using palm oil for cooking and serum total cholesterol in East Africa. We used a reliable point-of-care lipid testing system that has good clinical agreement with laboratory reference methods.[31] We systematically collected data on and adjusted for potential confounders of the association between diet and serum total cholesterol. Were our findings to be replicated in other studies, they could have important

implications for nutritional policies aimed at CVD prevention. Palm oil is now the major edible oil in Tanzania and many other developing countries. [49] In Tanzania, more than half of the palm oil is imported, [50] and it is the second most imported agricultural commodity.[51] In our study population, more than three-guarters of participants used palm oil (cost 2500 Tanzanian Shillings ≈ USD1.1 per litre as the major cooking oil with 94% of participants (n=119) in the lowest tertile of wealth using palm oil. Even in the wealthiest tertile, only 34% (n=123) used sunflower oil (cost 4000 Tanzanian Shillings ≈ USD1.8 per litre). These socioeconomic differentials in palm oil use may affect

1 2		
3 4	1	socioeconomic disparities in cardiovascular disease in the coming years with the burden
5 6	2	shifting more swiftly to poorer households.
7 8	3	
9	4	Policy interventions to address composition of dietary fat have previously been
10 11	5	implemented in Seychelles, Mauritius and Singapore.[27, 52, 53] In the early 1990s,
12 13	6	Seychelles implemented a nationwide program that included increasing use of
14 15	7	unsaturated vegetable oils, which may have contributed to the fairly high rate of decline
16 17	8	in age-standardized CVD mortality of 44% in men and 28% in women over a 22-year
18 19	9	period.[25,52,54] A national NCD prevention program was also implemented in
20 21	10	Mauritius in the late 1980s to reduce saturated fat content of cooking oil by changing
22 23	11	the government subsidized ration oil from palm oil to soybean oil. The observed 31
23 24 25	12	mg/dL reduction in serum total cholesterol over a period of 5 years, [26, 53] is
26	13	compatible with the estimated impact of replacing palm oil with unsaturated vegetable
27 28 29 30	14	oil estimated in a recent meta-analysis of dietary intervention trials.[55] Similar policies
	15	can be considered to reduce palm oil intake in Tanzania. However, it should be noted
31 32	16	that edible oils are viewed as necessities, [56] and increasing their price (e.g. by taxation)
33 34	17	without providing healthier alternatives at comparable prices may have unintended
35 36	18	adverse consequences on overall energy consumption or food security.[57-59] Reducing
37 38	19	saturated fat in edible oil by mixing it with other unsaturated oils has been proposed as
39 40	20	a potential intervention in Costa Rica and India, [23, 60] and could be a feasible short-
41	21	term intervention in Tanzania.
42 43	22	
44 45	23	In conclusion, we found that using palm oil for cooking versus sunflower oil, lower
46 47	24	intake of fruits and vegetables and higher intake of meat was strongly associated with
48 49	25	higher serum total cholesterol in this peri-urban population in Tanzania. Dietary policies
50 51	26	aimed at altering these targets can be used to improve lipid profiles and therefore
52 53	27	prevent cardiovascular diseases in Tanzania and other low-income countries with a
54 55	28	similar dietary profile.
55 56	29	

29

CONTRIBUTORS GD, JK, GHL and RMZ conceived and designed the study. GD obtained the funding. SSK and SF analyzed the data. GD and SSK interpreted the data and drafted the article, which was revised critically for substantive content by all authors, who approved the final version for publication. GD, SSK, SF and RMZ had full access to all of the data (including statistical reports and tables) in the study and can take responsibility for the integrity of the data and the accuracy of the data analysis. GD is the study guarantor.

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COMPETING INTERESTS: All authors have completed the ICMJE uniform disclosure form

23 COMPETING INTERESTS: All authors have completed the ICMJE uniform disclosure form
 at www.icmje.org/coi_disclosure.pdf (available on request from the corresponding
 author) and declare: no support from companies for the submitted work; no
 relationships with companies that might have an interest in the submitted work in the
 previous three years; their spouses, partners, or children have no financial relationships
 that may be relevant to the submitted work; no non-financial interests that may be
 relevant to the submitted work.

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3 4	1	ETHICAL APPROVAL: The study protocol was approved by the Institutional Review Board
5 6	2	of the Harvard TH Chan School of Public Health and the Research and Publications
7 8	3	Committee of Muhimbili University of Health and Allied Sciences. All participants gave
9 10	4	informed consent.
11	5	
12 13	6 7	DATA SHARING: No additional data available
14 15	8	TRANSPARENCY: The lead authors affirm that the manuscript is an honest, accurate,
16 17	9	and transparent account of the study being reported; that no important aspects of the
18 19	10	study have been omitted; and that any discrepancies from the study as planned have
20 21	11	been explained.
22 23	12 13	study have been omitted; and that any discrepancies from the study as planned have been explained.
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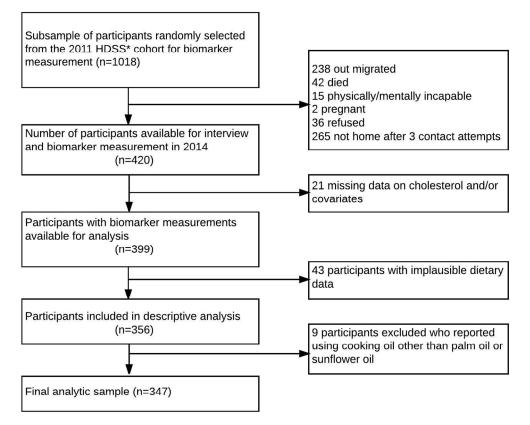
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1 Figure legends

Figure 1. Selection of study participants, DUCS-HTN biomarker sub-study, 2014

- Figure 2. Association between diet and serum total cholesterol. Values were adjusted
- 6 for age, sex, socioeconomic status, food consumption frequency, total energy intake,
- 7 major cooking oil used, physical activity and alcohol consumption.8
- 9 Figure 3. Diagnosis, awareness and treatment of hypercholesterolaemia in men (Part A;
- 10 n=54 for all percentages) and women (Part B; n=110 for all percentages) in the DUCS-
- HTN biomarker sub-study, 2014. If SUD-Store



*Dar Es Salaam Health and Demographic Surveillance System

Figure 1. Selection of study participants, DUCS-HTN biomarker sub-study, 2014

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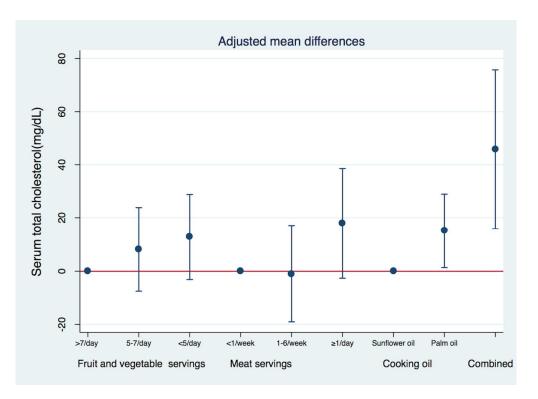
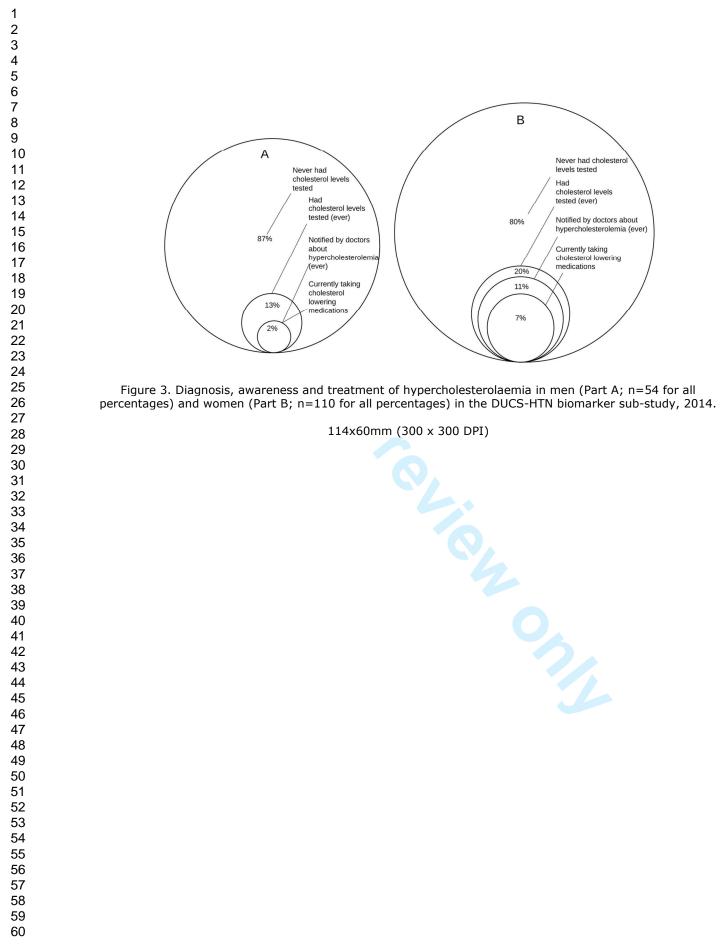
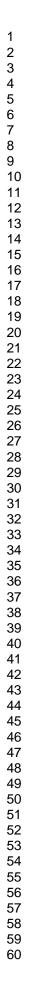


Figure 2. Association between diet and serum total cholesterol. Values were adjusted for age, sex, socioeconomic status, food consumption frequency, total energy intake, cooking oil, physical activity and alcohol consumption.

155x113mm (300 x 300 DPI)

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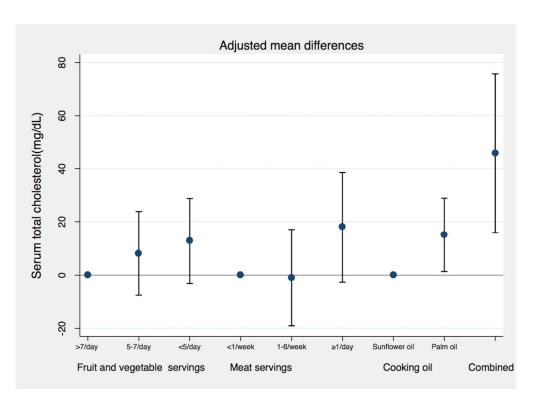


Figure 2. Association between diet and serum total cholesterol. Values were adjusted for age, sex, socioeconomic status, food consumption frequency, total energy intake, cooking oil, physical activity and alcohol consumption

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	Item No	Recommendation
Title and abstract	1	(a) Indicate the study's design with a commonly used term in the title or the abstract
		[Within the title page 1 and method section of the abstract page 2]
		(b) Provide in the abstract an informative and balanced summary of what was done
		and what was found [See results section of the abstract page 2]
Introduction		· · · · · · · · · · · · · · · · · · ·
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported [page 4]
Objectives	3	State specific objectives, including any prespecified hypotheses [page 4]
Methods		
Study design	4	Present key elements of study design early in the paper [page 5]
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment,
-		exposure, follow-up, and data collection [page 5]
Participants	6	(a) Give the eligibility criteria, and the sources and methods of selection of
		participants [page 5]
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect
		modifiers. Give diagnostic criteria, if applicable [page 5-6]
Data sources/	8*	For each variable of interest, give sources of data and details of methods of
measurement		assessment (measurement). Describe comparability of assessment methods if there i
		more than one group [page 5-6]
Bias	9	Describe any efforts to address potential sources of bias [page 6]
Study size	10	Explain how the study size was arrived at [page 5]
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable,
		describe which groupings were chosen and why [page 6]
Statistical methods	12	(a) Describe all statistical methods, including those used to control for confounding
		[page 6-7]
		(b) Describe any methods used to examine subgroups and interactions [page 7]
		(c) Explain how missing data were addressed [page 6]
		(d) If applicable, describe analytical methods taking account of sampling strategy
		[N/A]
		(e) Describe any sensitivity analyses [N/A]
Results		
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially
		eligible, examined for eligibility, confirmed eligible, included in the study,
		completing follow-up, and analysed [page 7 and Figure 1]
		(b) Give reasons for non-participation at each stage [Figure 1]
		(c) Consider use of a flow diagram [Figure 1]
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and
		information on exposures and potential confounders [page 7]
		(b) Indicate number of participants with missing data for each variable of interest
		[Figure 1]
Outcome data	15*	Report numbers of outcome events or summary measures [page 7-8 and Figure 3]
Main results	16	(a) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and
		their precision (eg, 95% confidence interval). Make clear which confounders were
		adjusted for and why they were included [page 12 and Table 2]

		(b) Report category boundaries when continuous variables were categorized [table
		1]
		(c) If relevant, consider translating estimates of relative risk into absolute risk for a
0.1 1	17	meaningful time period [N/A]
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses [N/A]
		Sensitivity analyses [N/A]
Discussion		
Key results	18	Summarise key results with reference to study objectives [page 14]
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or
		imprecision. Discuss both direction and magnitude of any potential bias [page 16]
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations,
		multiplicity of analyses, results from similar studies, and other relevant evidence
		[page 14,16]
Generalisability	21	Discuss the generalisability (external validity) of the study results [page 17]
Other information		
Funding	22	Give the source of funding and the role of the funders for the present study and, if
		applicable, for the original study on which the present article is based [page 20]

*Give information separately for exposed and unexposed groups.

Note: An Explanation and Elaboration article discusses each checklist item and gives methodological background and published examples of transparent reporting. The STROBE checklist is best used in conjunction with this article (freely available on the Web sites of PLoS Medicine at http://www.plosmedicine.org/, Annals of Internal Medicine at http://www.annals.org/, and Epidemiology at http://www.epidem.com/). Information on the STROBE Initiative is available at www.strobe-statement.org.

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Dietary determinants of serum total cholesterol among middle-aged and older adults: a population-based crosssectional study in Dar es Salaam, Tanzania

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3 4	1	Dietary determinants of serum total cholesterol among middle-aged and older adults:
5 6	2	a population-based cross-sectional study in Dar es Salaam, Tanzania
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9 10		
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1 2	ABSTRACT
3	Objective: To assess the dietary determinants of serum total cholesterol.
4	Design: Cross-sectional population-based study.
5	Setting: Peri-urban region of Dar es Salaam, Tanzania.
6	Participants: 347 adults aged 40 years and older from the Dar es Salaam Urban Cohe
7	Hypertension Study (DUCS-HTN).
8	Main outcome measure: Serum total cholesterol measured using a point-of-care de
9	Results: Mean serum total cholesterol level was 204 mg/dL (Interquartile range (IQR
10	169 to 236 mg/dL) in women and 185 mg/dL (IQR 152 to 216 mg/dL) in men. After
11	adjusting for demographic, socioeconomic, lifestyle and dietary factors, participants
12	who reported using palm oil as the major cooking oil had serum total cholesterol hig
13	by 15 mg/dL (95% confidence interval 1 to 29 mg/dL) compared with those who
14	reported using sunflower oil. Consumption of one or more servings of meat per day
15	for trend= 0.017) and less than five servings of fruits and vegetables per day (P for
16	trend= 0.024) were also associated with higher serum total cholesterol. A combinat
17	of using palm oil for cooking, eating more than one serving of meat per day and few
18	than five servings of fruits and vegetables per day was associated with 46 mg/dL (95
19	confidence interval 16 to 76 mg/dL) higher serum total cholesterol.
20	Conclusions: Using palm oil for cooking was associated with higher serum total
21	cholesterol levels in this peri-urban population in Dar es Salaam. Reduction of satura
22	fat content of edible oil may be considered as a population-based strategy for prima
23	prevention of cardiovascular diseases.
24	Key words: cholesterol, palm oil, diet, nutrition, Tanzania, Sub-Saharan Africa
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29	STRENGTHS AND LIMITATIONS OF THIS STUDY

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1						
2 3 4	1	•	This is the first population-based study to report an association between using palm			
5	2		oil for cooking and serum total cholesterol in East Africa.			
6 7	3	•	A reliable point-of care lipid testing system was used to measure serum total			
8 9	4		cholesterol that has good clinical agreement with laboratory reference methods.			
10 11	5	•	Potential confounders of the association between diet and serum total cholesterol			
12 13	6		were systematically measured and adjusted for.			
14 15	7	•	Only serum total cholesterol was measured instead of a full lipid panel, due to logistic			
16 17	8		and financial limitations.			
18 19	9	•	The relatively small sample size may have resulted in insufficient statistical power to			
20 21	10		detect some associations.			
22	11	•	The assessment of diet using any method is prone to measurement error, which may			
23 24	12		have resulted in bias, although differential misreporting of cooking oil by cholesterol			
25 26	13		level is unlikely.			
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1 INTRODUCTION

Sub-Saharan Africa is facing a burgeoning epidemic of non-communicable diseases
(NCDs) due to changing demographic profiles, epidemiologic transition, urbanization,
and lifestyle changes as a result of economic development as well as increasing survival
of HIV patients.[1, 2] In 2015, one third of all estimated NCD deaths in sub-Saharan
Africa were due to cardiovascular diseases (CVD).[3] By 2030, the share of all deaths
attributable to CVD is projected to increase by nearly 40% in this region.[3]

9 Dyslipidaemia is a well-known risk factor for ischemic heart disease (IHD), stroke, and 10 other vascular diseases.[4, 5] In middle-aged adults, IHD mortality is lower by as much 11 as 30 to 50% for every 1 mmol/L (38.6mg/dL) lower serum total cholesterol.[5] Sub-12 Saharan Africans have historically had a favorable lipid profile [6] but hyperlipidaemia is 13 becoming increasingly common in these countries, especially in urban areas.[7-9] It is 14 estimated that thirty-four percent of ischemic heart disease deaths in Tanzania are 15 attributable to dyslipidaemia.[10]

A few studies have already explored the relationship between diet and serum total cholesterol in eastern sub-Saharan Africa and reported an association between higher intake of meat and lower intake of fish and green vegetables with higher serum total cholesterol. [11-15] However, two of these studies compared rural and coastal Bantu populations that are known to have, respectively, vegetarian and fish-based diets and did not measure diet in each participant and based their conclusions on known dietary differences between ethnic populations. [14, 15] Another study included participants from rural, urban and pastoral regions of Tanzania but did not report the magnitude of the association[12] The other two studies conducted in urban Tanzania did not adjust for potential confounders of the relationship between diet and serum total cholesterol. [11, 13] There are a few studies from other parts of sub-Saharan Africa that have looked at the association between diet and serum cholesterol. A study conducted in a rural population in Uganda found that eating fewer than 5 servings of fruits and

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1	vegetables daily was associated with higher odds of low HDL cholesterol among men,
2	but lower odds among women.[16] Another study in urban Uganda found lower mean
3	serum total cholesterol in lacto-ovo-vegetarians compared to non-vegetarians.[17]
4	
5	Another potential dietary determinant of serum cholesterol is the type of oil used in
6	cooking.[18,19] Using cooking oils with higher saturated fat content such as coconut
7	and palm oil has been associated with higher serum total cholesterol levels in clinical
8	trials and observational studies.[20, 21] Population-level differences in CVD morbidity
9	and mortality is associated with differences in type of cooking oil in eastern
10	European, [22] Costa Rica, [23] and Singapore. [24] Mauritius, Seychelles and Singapore
11	have already implemented policy schemes to reduce saturated fat content of cooking oil
12	as population-level interventions to reduce burden of CVD.[25-27]
13	A thorough understanding of the role of dietary determinants of serum cholesterol will
14	be invaluable in designing population-level interventions to address the growing CVD
15	burden especially considering that treatment for dyslipidaemia is either unavailable or
16	prohibitively expensive for most patients in low-income countries.[28] Tanzanian diet
17	consists predominantly of carbohydrates (mainly maize, sweet potatoes, cassava and
18	rice) consumed with green leafy vegetables, beans or peas and occasionally with meat
19	or fish.[29] Palm oil is the major cooking oil, followed by sunflower oil.
20	We conducted a cross-sectional population-based study to assess the association of
21	type of cooking oil (palm vs sunflower oil) and diet (meat, fish, fruit and vegetable
22	consumption) with serum total cholesterol in an adult population in a peri-urban area of
23	Dar es Salaam. We hypothesized that use of palm oil for cooking and higher intake of
24	meat and lower intake of fruits and vegetables will be associated with higher serum
25	total cholesterol.
26	METHODS

27 Study design and population

The Dar es Salaam Health and Demographic Surveillance System (HDSS) is a demographic surveillance system in Ukonga and Gongo la Mboto wards of Dar es Salaam, Tanzania with over 100,000 enumerated individuals living in 21,000 households followed since 2011/12. For the Dar es Salaam Urban Cohort Hypertension Study (DUCS-HTN), we randomly selected one large and one small neighborhood in the HDSS and contacted all adult residents aged 40 years and older, who had been registered in the HDSS database in 2011 or 2012. A random subsample of one-fifth of the eligible participants was selected for additional assessment of diet and measurement of serum total cholesterol. The size of this subsample was determined largely based upon the availability of resources. Pregnant women and those who were physically and mentally incapable of completing the interview and measurements were excluded. The participant selection flowchart is shown in Figure 1. Face-to-face interviews and measurements were conducted in participant's home from March to June 2014 by six trained interviewers. All interviewers had previous experience conducting public health surveys. For this study, interviewers received five days of training and two days of field practice in a nearby neighborhood. At least three attempts were made to contact all potential participants. Written informed consent was obtained from all participants. The study protocol was approved by the Institutional Review Board of the Harvard TH Chan School of Public Health and the Research and Publications Committee of Muhimbili University of Health and Allied Sciences. Measurements Non-fasting serum total cholesterol was measured from 30 micro-liters of finger-prick capillary blood samples using Cardiochek PA devices (Polymer Technology Systems Inc, IN, USA).[30, 31] The overall coefficient of variation for this device for total cholesterol has been determined to be in the range of 1.3 to 2.9%.[30] Data on diet was collected using a semi-quantitative food frequency questionnaire (FFQ). The FFQ assessed intake of 179 food and drink items over the past 30 days. A shorter 85-item version of this instrument has been previously used in Tanzania.[32]

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Nutrient intakes were calculated by multiplying the frequency of food consumption measured from FFQ by the nutrient content of the specified portion size using the Tanzanian food composition table. [33] Meat intake was defined to include all red and white meat consumption, excluding fish consumption. Fruit and vegetable intake was defined to include consumption of all fruits and vegetables, except for root vegetables and fruit juice. An additional question was asked to assess the main type of cooking oil: "What is the kind of oil used most often in your home for frying food?". Frying is the most common cooking technique involving oil use in this region. The reference to a cooking technique was made to distinguish it from other oils that may be used in a household, such as fuels.

All participants completed a socioeconomic, demographic and lifestyle questionnaire and had their height, weight, waist circumference, hip circumference, and blood pressure measured by trained interviewers. Some demographic information (age, sex, neighborhood, components of wealth index) was taken from the HDSS baseline interview. Body mass index was calculated as weight (kg) divided by height (m) squared, and categorized as underweight (<18.5), normal (18.5 to <25.0), overweight (25.0 to <30.0), and obese (≥30.0).[34] Physical activity was assessed using the Global Physical Activity Questionnaire in the domains of work, transportation, and leisure, [35] and categorized into tertiles of Metabolic Equivalent (MET) hours per week. To measure socioeconomic status, we created a household wealth index using a principal component analysis of household characteristics (floor material, roof material, wall material, electricity), household assets (television, radio, shop, sewing machine, sofa, fan, iron, stove with oven, stove without oven, dining table, cupboard, watch, mobile phone, bike, motorcycle, cart, car, motorboat), and animal ownership (goats, sheep, chicken/ducks, pigs).[36] The household wealth index was categorized into tertiles.

26 Statistical analysis

We excluded participants with implausible dietary data (total energy intake <500 or
>5000 kcal/day), and with missing data on serum total cholesterol, diet and/or any of
the potential covariates.

Chi-square tests were used to assess binary variables, F-tests were used for categorical variables and tests of linear trend based on median value within each category for ordinal variables with a linear association with serum total cholesterol (i.e. age, wealth index, and food consumption categories). Mean differences and 95% confidence intervals in serum total cholesterol across categories of dietary factors, including cooking oil were estimated using multivariable linear regression. Potential confounders were included in the model based on a-priori knowledge of lifestyle and socio-demographic determinants of diet and serum total cholesterol. The fully adjusted model included age, sex, total energy intake, physical activity, alcohol consumption, wealth index and employment status as covariates. We also adjusted for interviewer to remove the effect of any extraneous variation in measurement of subjective risk factors across interviewers. [37, 38] We examined interactions of potential determinants of serum total cholesterol with sex, but no interactions were statistically significant at 0.2 level, therefore we did not include these product terms in the final model. We conducted sensitivity analyses by excluding 16 participants who reported either being previously diagnosed with hypercholesterolaemia, or currently taking cholesterol-lowering medications or being advised by a health professional to modify diet to lower cholesterol. All analyses were performed using STATA software version 13.1 (STATA Corporation, College Station, Texas). RESULTS A total of 420 participants were enrolled for the random subsample with diet and cholesterol measurements. Among these, data on serum total cholesterol and selected covariates was available for 399 participants. We excluded 43 participants due to implausible caloric intake. In addition, after descriptive analyses we excluded 9 (2%) participants who reported using cooking oils other than palm or sunflower oil. The final

analytic sample included 347 participants (FIGURE 1).

29 Forty-two percent of the participants (n=347) were men (TABLE 1). Mean age of men

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1	was 55 years (SD 11), and that of women was 52 (SD 10). 44% of the women (n=202)
2	and 19% of men (n=145) were obese, and one third of men and women were
3	overweight. Mean energy intake per day was 2413 kcal. Thirty-seven percent of men
4	and 54% of women were hypercholesterolemic (i.e. had total cholesterol ≥200 mg/dL).
5	Mean serum total cholesterol in women was 204 mg/dL (95% confidence interval 197 to
6	211 mg/dL) and was significantly higher than that of men at 185 mg/dL (177 to 193
7	mg/dL). Serum total cholesterol levels were higher in older age groups.
8	79% of participants reported using palm oil as the major cooking oil and the rest
9	reported using sunflower oil (TABLE 1). Those who reported using palm oil were most
10	likely to be in the poorest tertile of wealth, whereas those who reported using
11	sunflower oil were most likely to be in the wealthiest tertile (p<0.001). Those who
12	reported using sunflower oil had a higher energy intake (p=0.04), and reported

consuming meat (p=0.004) and dairy (p=0.02) more frequently than those who reported

using palm oil. Mean serum total cholesterol in those who reported using palm oil was

199 mg/dL (193 to 205) and was significantly higher than those who reported using

sunflower oil 184 mg/dL (173 to 195).

	All (n=347)	Men (n=145)	Women (n=202)	Use palm oil	Use sunflower oi	
Variable				(n=274)	(n=73)	
Age						
40-49	150 (43)	56 (39)	94 (47)	123 (45)	27 (37)	
50-59	113 (33)	43 (30)	70 (35)	82 (30)	31 (42)	
60 and above	84 (24)	46 (32)	38 (19)	69 (25)	15 (21)	
Employment status						
Unemployed	83 (24)	14 (10)	69 (34)	70 (26)	13 (18)	
Employed (includes self-employed)	238 (69)	112 (77)	126 (62)	183 (67)	55 (75)	
Retired	26 (7)	19 (13)	7 (3)	21 (8)	5 (7)	
Household wealth index						
Tertile 1, Poorest	115 (33)	49 (34)	66 (33)	109 (40)	6 (8)	
Tertile 2	117 (34)	53 (37)	64 (32)	90 (33)	27 (37)	
Tertile 3, Wealthiest	115 (33)	43 (30)	72 (36)	75 (27)	40 (55)	
BMI (kg/m²)						
Underweight (<18.50)	19 (5)	13 (9)	6 (3)	19 (7)	0 (0)	
Normal weight (18.50-<25.00)	100 (29)	56 (39)	44 (22)	91 (33)	9 (12)	
Overweight (25-<30.00)	112 (32)	48 (33)	64 (32)	81 (30)	31 (42)	
Obese (≥ 30.00)	116 (33)	28 (19)	88 (44)	83 (30)	33 (45)	
Physical activity						
Tertile 1,	123 (35)	46 (32)	77 (38)	96 (35)	27 (37)	
(0-14 MET-hours/week)	125 (55)	40 (32)	// (50)	50 (55)	27 (37)	
Tertile 2, (15, 112 MET hours (wook)	108 (31)	38 (27)	70 (35)	86 (31)	22 (30)	
(15-112 MET-hours/week) Tertile 3,						
(113-840 MET-hours/week)	116 (33)	61 (42)	55 (27)	92 (34)	24 (33)	

Table 1. Participants' characteristics in DUCS-HTN biomarker sub-study, 2014

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Variable	All (n=347)	Men (n=145)	Women (n=202)	Use palm oil (n=274)	Use sunflower oil (n=73)
Alcohol drinking ^a					
Non-drinker	286 (82)	103 (71)	183 (91)	225 (82)	61 (84)
Smoking					
Non-smoker	278 (81)	85 (59)	193 (97)	210 (78)	68 (93)
Former smoker	43 (13)	39 (27)	4 (2)	38 (14)	5 (7)
Current smoker	22 (6)	19 (13)	3 (2)	22 (8)	0 (0)
DIETARY VARIABLES					
Palm oil as major cooking oil	274 (79)	119 (82)	155 (77)		
Meat intake					
<1 serving/week	41 (12)	21 (14)	20 (10)	36 (13)	5 (7)
1-6 servings/week	173 (50)	55 (38)	118 (58)	145 (53)	28 (38)
≥1 servings/day	133 (38)	69 (48)	64 (32)	93 (34)	40 (55)
Fish intake					
< 1 serving/week	41 (12)	13 (9)	28 (14)	31 (11)	10 (14)
1-7 servings/week	196 (56)	76 (52)	120 (59)	154 (56)	42 (58)
> 1 servings/day	110 (32)	56 (39)	54 (27)	89 (32)	21 (29)
Dairy intake					
< 1 serving/week	159 (46)	63 (43)	96 (48)	136 (50)	23 (32)
1-6 servings/week	133 (38)	65 (45)	68 (34)	99 (36)	34 (47)
≥ 1 servings/day	55 (16)	17 (12)	38 (19)	39 (14)	16 (22)
Fruit and vegetable					
< 5 servings/day	121 (35)	49 (34)	72 (36)	101 (37)	20 (27)
5-7 servings/day	68 (20)	26 (20)	42 (21)	54 (20)	14 (19)

Variable	All (n=347)	Men (n=145)	Women (n=202)	Use palm oil (n=274)	Use sunflower oi (n=73)
>7 servings/day	158 (46)	70 (48)	88 (44)	119 (43)	39 (53)
Nuts and legumes					
< 1 serving/week	27 (8)	7 (5)	20 (10)	17 (6)	10 (14)
1-7 servings/week	200 (58)	87 (60)	113 (56)	162 (59)	38 (52)
> 1 servings/day	120 (35)	51 (35)	69 (34)	95 (35)	25 (34)
Total energy intake (kcal/day) ^b	2413 (1701, 3215)	2629 (1780, 3216)	2313 (1617, 3207)	2369 (1654, 3150)	2743 (1865, 3606)
Fat intake ^b					
Total fat (% of energy)	17 (15, 20)	17 (15, 21)	17 (15, 20)	17 (14, 20)	20 (15, 22)
Saturated fat (% energy)	10 (8, 13)	10 (8, 13)	10 (8, 12)	10 (8, 13)	10 (8, 11)
Monounsaturated fat (% of energy)	4 (3, 5)	4 (3, 5)	4 (3, 5)	4 (3 <i>,</i> 5)	4 (4, 5)
Polyunsaturated fat (% of energy)	2 (1, 2)	2 (1, 2))	2 (1, 2)	1 (1, 2)	2 (1, 2)
Cholesterol					
Serum total cholesterol (mg/dl) ^c	196 (3)	185 (4)	204 (4)	199 (3)	184 (6)
Hypercholesterolaemia ^d	164 (47)	54 (37)	110 (54)	135 (49)	29 (40)
Cholesterol level was previously checked	29 (18)	7 (13)	22 (20)	13 (10)	16 (55)
Previously diagnosed hypercholesterolaemia	13 (8)	1 (2)	12 (11)	5 (4)	8 (28)
Currently taking cholesterol lowering medications	9 (5)	1 (2)	8 (7)	3 (2)	6 (21)

Number (%) reported for all variables, except as specified b and c below ^a According to report of drinking alcohol in the past 30 days ^b Total energy and fat intake, which is reported as median (interquartile range)

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. al cholesterol 2 200 mg/dL ^c Serum total cholesterol, reported as mean (standard error) ^d Hypercholesterolaemia defined as total cholesterol \geq 200 mg/dL

In the minimally adjusted model (with interviewer as the only covariate), those who reported using palm oil as the major cooking oil had serum total cholesterol levels higher by 11 mg/dL (-2 to 24 mg/dL) compared with those who reported using sunflower oil (TABLE 2). After adjusting for covariates, participants who used palm oil for cooking had 15 mg/dL (1 to 29 mg/dL) higher serum total cholesterol (FIGURE 2). The negative confounding was mostly due to the negative correlation between female gender and meat intake with palm oil use (both variables were positively correlated with serum total cholesterol). Serum total cholesterol was also higher in participants who ate meat more frequently (P for trend 0.017) after adjusting for potential confounders. Specifically, those who ate more than one serving of meat per day had 18 mg/dL (-3 to 39 mg/dL) higher serum total cholesterol compared with those who ate less than one serving per week. In this relationship, higher total energy intake was the major confounder with a positive correlation with meat intake but negative correlation with serum total cholesterol. Consumption of more than seven servings of fruits and vegetables per day was associated with 13 mg/dL (-29 to 3 mg/dL) lower serum total cholesterol, compared to consumption of fewer than five servings per day (P for trend 0.024). A combination of using palm oil for cooking, eating more than one serving of meat per day and fewer than five servings of fruits and vegetables per day was associated with 46 mg/dL (16 to 76 mg/dL) higher serum total cholesterol. We found similar associations after excluding 16 participants who reported either being previously diagnosed with hypercholesterolaemia, or currently taking cholesterol lowering medications or being advised by a health professional to modify diet to lower

cholesterol. Those who used palm oil had 17 mg/dL (2 to 32 mg/dL) higher serum total
cholesterol compared with those who used sunflower oil (TABLE 2). There was no

- 26 significant association between percent energy from saturated fat and total serum
- cholesterol after adjusting for total fat, total protein and total carbohydrate (Adjusted
 mean difference 1mg/dL (-3 to 5 mg/dL))

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Table 2. Adjusted mean differences for serum total cholesterol (mg/dL) by dietary factors; DUCS-HTN biomarker sub-study

Variable	Interviewer- adjusted mean difference (95% CI)	P Value ^a	Fully-adjusted ^b mean difference (95% Cl) (n=347)	P Value ^a	Fully-adjusted ^c mean difference after excluding 16 participants diagnosed or treated for dyslipidaemia (95% CI) (n=331)	P Value ^ª
Major cooking oil		0.092		0.033		0.028
Sunflower oil	Reference		Reference		Reference	
Palm oil	11 (-2, 24)		15 (1, 29)		17 (2, 32)	
Meat intake		0.485		0.017		0.031
<1 serving/week	Reference		Reference		Reference	
1-6 servings/week	-1 (-19, 16)		-1 (-19, 17)		-2 (-20, 17)	
≥1 servings/day	4 (-15, 22)		18 (-3, 39)		16 (-5, 37)	
Fruit and vegetable		0.028		0.011		0.024
intake						
< 5 servings/day	16 (2,30)		13 (-3, 29)		12 (-5, 29)	
5-7 servings/day	10 (-6, 25)		8 (-8, 24)		10 (-7, 26)	
> 7 servings/day	Reference		Reference		Reference	
Fish intake		0.399		0.446		0.309
< 1 serving/week	14 (-8, 36)		10 (-13, 33)		14 (-9, 38)	
1-7 servings/week	3 (-11, 17)		2 (-13, 17)		4 (-11, 20)	
>1 servings/day	Reference		Reference		Reference	
Dairy intake		0.802		0.572		0.690
< 1 serving/week	Reference		Reference		Reference	
1-6 servings/week	-9 (-22, 4)		-7 (-21, 6)		-7 (-21, 7)	

Variable	Interviewer- adjusted mean difference (95% Cl)	P Value ^a	Fully-adjusted ^b mean difference (95% CI) (n=347)	P Value ^a	Fully-adjusted ^c mean difference after excluding 16 participants diagnosed or treated for dyslipidaemia (95% CI) (n=331)	P Value ^a
≥ 1 servings/day	-2 (-18, 15)		-2 (-20, 15)		-1 (-20, 17)	
Total energy intake (Per 1000 kcal)	-8 (-21, 5)	0.226	-5 (-12, 3)	0.209	-4 (-11, 4)	0.365
Age		0.106		0.071		0.099
40-49	Reference		Reference		Reference	
50-59	4 (-9, 16)		6 (-6, 19)		5 (-9, 18)	
60 and above	11 (-2 <i>,</i> 25)		16 (0, 31)		15 (-1, 31)	
Sex		< 0.001		< 0.001		<0.001
Male	Reference		Reference		Reference	
Female	21 (10, 32)		27 (15, 39)		24 (11, 36)	
Employment status		0.432		0.567		0.581
Unemployed	Reference		Reference		Reference	
Employed ^d	-7 (-20, 6)		7 (-9, 22)		5 (-12, 21)	
Retired	2 (-21, 24)		12 (-13, 36)		13 (-12, 39)	
Household Wealth Index		0.692		0.754		0.952
1, Poorest	Reference		Reference		Reference	
2	-9 (-22, 4)		-8 (-21, 5)		-7 (-20, 7)	
3, Wealthiest	-1 (-15, 12)		0 (-14, 14)		2 (-13, 16)	
Physical activity tertile		0.953		0.430		0.599
1, least active	Reference		Reference		Reference	

Variable	Interviewer- adjusted mean difference (95% CI)	P Value ^ª	Fully-adjusted ^b mean difference (95% CI) (n=347)	P Value ^a	Fully-adjusted ^c mean difference after excluding 16 participants diagnosed or treated for dyslipidaemia (95% Cl) (n=331)	P Value ^a
2	1 (-13, 15)		2 (-12, 16)		4 (-11, 18)	
3, most active	0 (-15, 15)		7 (-9, 24)		6 (-11, 24)	
Alcohol consumption ^e		0.315		0.666		0.641
Non-drinker	Reference		Reference		Reference	
Drinker	-7 (-21,7)		-3 (-18, 11)		-4 (-18, 11)	

^a F2test for categorical variables (cooking oil, sex, employment status, alcohol consumption); t test for continuous variables (total energy intake); test of trend based on median value in each category for ordinal categorical variables (age category, wealth index tertile, food consumption and physical activity categories)

 $^{\text{b}}$ AB justed for covariates in column 1 as well as interviewer

^c Excluding those previously diagnosed with hypercholesterolaemia; adjusted for covariates in column 1 as well as interviewer

^d S*e*lf-employed, or government job, or job in private company

^e A8 cording to report of drinking alcohol in the past 30 days

1 Only 11% of women and 5% of men reported having ever had their cholesterol levels

2 checked (TABLE 1; FIGURE 3). Among those with hypercholesterolaemia (n=164), 2% of

3 men (1 of 54), and 11% of women (12 of 110) reported being notified by doctors about

4 it, and 2% of men and 7% of women reported taking cholesterol-lowering medications.

DISCUSSION

In this cross-sectional study among middle-aged and older adults in a peri-urban ward in Dar es Salaam, Tanzania, we found a high prevalence of hypercholesterolaemia. Slightly more than a third of men and half of women had serum total cholesterol levels greater than or equal to 200mg/dL. After adjusting for major confounders, serum total cholesterol was higher by 15 mg/dL among those who used palm oil compared to those who used sunflower oil as the major cooking oil, and showed a significant increasing trend with higher consumption of meat (P=0.017) and lower consumption of fruits and vegetables (P=0.01).

Mean serum total cholesterol levels for men and women were similar to that reported in other studies conducted in other urban Tanzanian populations. [32, 39, 40] A repeated cross-sectional study reported an increase over time in the prevalence of hypercholesterolaemia in Dar es Salaam with a prevalence of 17% in 1987 versus 30% in 1998 in men, and 7% versus 50% in women. [39] The prevalence of hypercholesterolaemia in our sample was higher than that reported by studies conducted between 2008-2014 in rural Uganda (3% men, 8% women),[9] peri-urban Nigeria (2% men, 6% women),[8] and blacks in urban South-Africa (25% men, 23% women),[41] but is lower than that reported in a study from Senegal (54% men, 61% women) that included both urban and rural participants.[42] Our estimated association between palm oil versus sunflower oil use and serum total cholesterol is consistent with findings from dietary observational studies and intervention trials. A recent meta-analysis of 30 dietary intervention trials comparing

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diets rich in palm oil with diets rich in vegetable oils low in saturated fatty acids showed
significantly higher total (13.5 mg/dL) and LDL (9.3 mg/dL) cholesterol, and slightly
higher HDL cholesterol (0.8 mg/dL) 2 to 16 weeks after intervention.[21] In addition, a
large case-control study conducted in Costa Rica found that palm oil use compared to
soybean oil use was independently associated with 33% higher odds of myocardial
infarction. [23].

Higher meat consumption has previously been reported to be associated with higher serum total cholesterol in Tanzania. [12, 13] Similar to our finding, daily consumption of meat was found to be associated with 24 mg/dL higher serum total cholesterol levels, after adjusting for age, select dietary components and resting energy expenditure.[13] Observational studies have also reported lower LDL cholesterol by 2.3mg/dL for every additional serving of fruits or vegetables.[43-45] The magnitude of 13 mg/dL reduction in serum total cholesterol that we found for six or more servings of fruits and vegetables per day is consistent with the reductions in LDL-cholesterol seen in these studies. We did not find significant associations between serum total cholesterol and fish intake in men, as reported in previous studies. [12, 13] This could have been due to insufficient statistical power due to smaller expected effect size for fish intake or higher measurement error.

Palm oil is obtained mostly from the middle section (pulp) of the fruits of the tropical plant *Elaeis quineensis*.[46] Compared with most other vegetable oils, it contains less unsaturated fat and more saturated fat (nearly 40 to 50% of total fat content) with the majority being palmitic acid. [46] However, compared with saturated fat from animal sources, palm oil may have a smaller impact on serum cholesterol because almost 70% of the palmitic acid is located in the first and third position of the triglycerides, which is less absorbable than those in the second position (which is the most common position of palmitic acid in animal fats). [47, 48] Despite these biological differences, evidence from both observational and experimental studies, as summarized above, points to

harmful effects of palm oil on serum cholesterol and CVD when substituted for
 unsaturated fatty acids.

Our study has several limitations. First, due to logistic and financial limitations, we measured serum total cholesterol instead of a full lipid panel. However, data from recent meta-analysis of randomized trials suggests that the effect of substituting palm oil for vegetable oils low in saturated fat on LDL cholesterol levels is larger than that on HDL.[21] Second, the assessment of diet using any method is prone to measurement error which may have resulted in bias. However, it is unlikely that participants with higher serum cholesterol differentially misreport using palm oil in cooking, limiting the impact of such measurement error on our results. Thirdly, the relatively small sample size may have resulted in insufficient statistical power to detect some associations (e.g. with fish intake). There are some important strengths of this study. To our knowledge, this is the first population-based study to report an association between using palm oil for cooking and serum total cholesterol in East Africa. We used a reliable point-of-care lipid testing system that has good clinical agreement with laboratory reference methods.[31] We systematically collected data on and adjusted for potential confounders of the association between diet and serum total cholesterol. Were our findings to be replicated in other studies, they could have important

implications for nutritional policies aimed at CVD prevention. Palm oil is now the major edible oil in Tanzania and many other developing countries. [49] In Tanzania, more than half of the palm oil is imported, [50] and it is the second most imported agricultural commodity.[51] In our study population, more than three-quarters of participants used palm oil (cost 2500 Tanzanian Shillings ≈ USD1.1 per litre as the major cooking oil with 94% of participants (n=119) in the lowest tertile of wealth using palm oil. Even in the wealthiest tertile, only 34% (n=123) used sunflower oil (cost 4000 Tanzanian Shillings ≈ USD1.8 per litre). These socioeconomic differentials in palm oil use may affect

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3 4	1	socioeconomic disparities in cardiovascular disease in the coming years with the burden
5 6	2	shifting more swiftly to poorer households.
7 8	3	
9 10	4	Policy interventions to address composition of dietary fat have previously been
11	5	implemented in Seychelles, Mauritius and Singapore.[27, 52, 53] In the early 1990s,
12 13	6	Seychelles implemented a nationwide program that included increasing use of
14 15	7	unsaturated vegetable oils, which may have contributed to the fairly high rate of decline
16 17	8	in age-standardized CVD mortality of 44% in men and 28% in women over a 22-year
18 19	9	period.[25,52,54] A national NCD prevention program was also implemented in
20 21	10	Mauritius in the late 1980s to reduce saturated fat content of cooking oil by changing
22 23	11	the government subsidized ration oil from palm oil to soybean oil. The observed 31
24 25	12	mg/dL reduction in serum total cholesterol over a period of 5 years,[26, 53] is
26 27	13	compatible with the estimated impact of replacing palm oil with unsaturated vegetable
28	14	oil estimated in a recent meta-analysis of dietary intervention trials.[55] Similar policies
29 30	15	can be considered to reduce palm oil intake in Tanzania. However, it should be noted
31 32	16	that edible oils are viewed as necessities, [56] and increasing their price (e.g. by taxation)
33 34	17	without providing healthier alternatives at comparable prices may have unintended
35 36	18	adverse consequences on overall energy consumption or food security.[57-59] Reducing
37 38	19	saturated fat in edible oil by mixing it with other unsaturated oils has been proposed as
39 40	20	a potential intervention in Costa Rica and India,[23, 60] and could be a feasible short-
41 42	21	term intervention in Tanzania.
43	22	
44 45	23	In conclusion, our results indicate that using palm oil for cooking, lower intake of fruits
46 47	24	and vegetables and higher intake of meat is strongly associated with higher serum total
48	~ -	

cholesterol in this peri-urban population in Tanzania. Dietary policies aimed at altering
these targets can be used to improve lipid profiles and therefore prevent cardiovascular

27 diseases in Tanzania and other low-income countries with a similar dietary profile.

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CONTRIBUTORS GD, JK, GHL and RMZ conceived and designed the study. GD obtained the funding. SSK and SF analyzed the data. GD and SSK interpreted the data and drafted the article, which was revised critically for substantive content by all authors, who approved the final version for publication. GD, SSK, SF and RMZ had full access to all of the data (including statistical reports and tables) in the study and can take responsibility for the integrity of the data and the accuracy of the data analysis. GD is the study guarantor.

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relevant to the submitted work.

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3 4	1	ETHICAL APPROVAL: The study protocol was approved by the Institutional Review Board
F	2	of the Harvard TH Chan School of Public Health and the Research and Publications
-	3	Committee of Muhimbili University of Health and Allied Sciences. All participants gave
9	4	informed consent.
12 13	5 6 7	DATA SHARING: No additional data available
14	, 8	TRANSPARENCY : The lead authors affirm that the manuscript is an honest, accurate,
16 17	9	and transparent account of the study being reported; that no important aspects of the
18 19 10	0	study have been omitted; and that any discrepancies from the study as planned have
20 21 1	1	been explained.
22 12 23 12 24 14 25	3	
$ \begin{array}{ccccccccccccccccccccccccccccccccc$	5	and transparent account of the study being reported; that no important aspects of the study have been omitted; and that any discrepancies from the study as planned have been explained.

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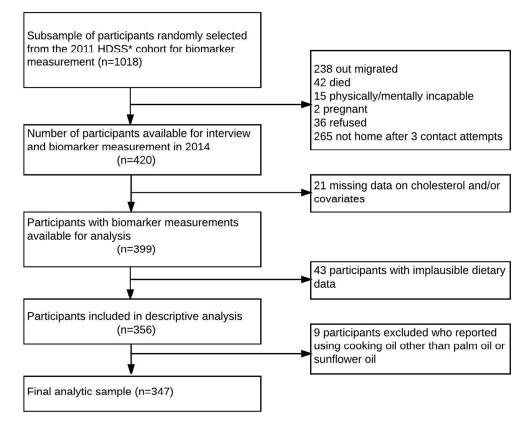
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1 Figure legends

Figure 1. Selection of study participants, DUCS-HTN biomarker sub-study, 2014

- Figure 2. Association between diet and serum total cholesterol. Values were adjusted
- 6 for age, sex, socioeconomic status, food consumption frequency, total energy intake,
- 7 major cooking oil used, physical activity and alcohol consumption.

- Figure 3. Diagnosis, awareness and treatment of hypercholesterolaemia in men (Part A;
- 10 n=54 for all percentages) and women (Part B; n=110 for all percentages) in the DUCS-
- HTN biomarker sub-study, 2014. If SUD-Store



*Dar Es Salaam Health and Demographic Surveillance System

Figure 1. Selection of study participants, DUCS-HTN biomarker sub-study, 2014

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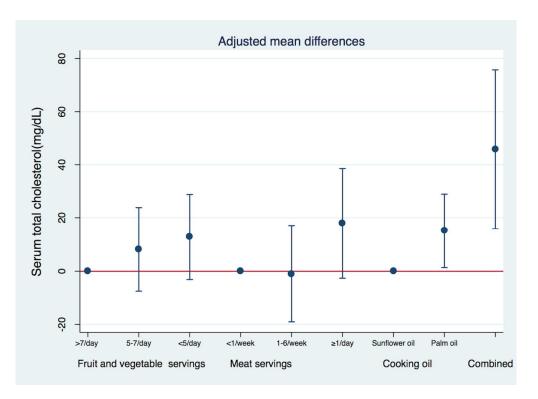
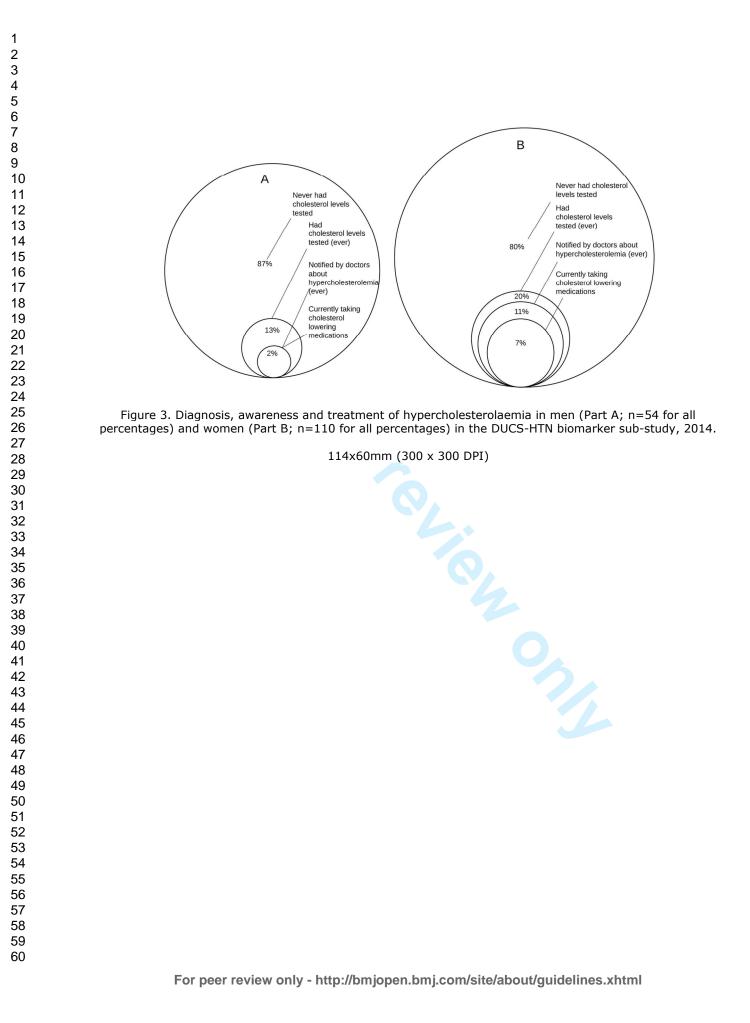


Figure 2. Association between diet and serum total cholesterol. Values were adjusted for age, sex, socioeconomic status, food consumption frequency, total energy intake, cooking oil, physical activity and alcohol consumption.

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STROBE Statement—Checklist of items that should be included in reports of *cross-sectional studies*

	Item No	Recommendation
Title and abstract	1	(a) Indicate the study's design with a commonly used term in the title or the abstract
		[Within the title page 1 and method section of the abstract page 2]
		(b) Provide in the abstract an informative and balanced summary of what was done
		and what was found [See results section of the abstract page 2]
Introduction		
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported
Dackground/rationale	2	[page 4]
Objectives	3	State specific objectives, including any prespecified hypotheses [page 4]
	3	state specific objectives, menualing any prespectifical hypotheses [page 4]
Methods	1	Descent have also and a fature design contrain the namer (name 5)
Study design	4	Present key elements of study design early in the paper [page 5]
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment,
		exposure, follow-up, and data collection [page 5]
Participants	6	(a) Give the eligibility criteria, and the sources and methods of selection of
· · · · ·		participants [page 5]
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect
		modifiers. Give diagnostic criteria, if applicable [page 5-6]
Data sources/	8*	For each variable of interest, give sources of data and details of methods of
measurement		assessment (measurement). Describe comparability of assessment methods if there is
		more than one group [page 5-6]
Bias	9	Describe any efforts to address potential sources of bias [page 6]
Study size	10	Explain how the study size was arrived at [page 5]
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable,
		describe which groupings were chosen and why [page 6]
Statistical methods	12	(a) Describe all statistical methods, including those used to control for confounding
		[page 6-7]
		(b) Describe any methods used to examine subgroups and interactions [page 7]
		(c) Explain how missing data were addressed [page 6]
		(d) If applicable, describe analytical methods taking account of sampling strategy
		[N/A]
		(e) Describe any sensitivity analyses [N/A]
Results		
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially
		eligible, examined for eligibility, confirmed eligible, included in the study,
		completing follow-up, and analysed [page 7 and Figure 1]
		(b) Give reasons for non-participation at each stage [Figure 1]
		(c) Consider use of a flow diagram [Figure 1]
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and
		information on exposures and potential confounders [page 7]
		(b) Indicate number of participants with missing data for each variable of interest
		[Figure 1]
Outcome data	15*	Report numbers of outcome events or summary measures [page 7-8 and Figure 3]
Main results	16	(a) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and
		their precision (eg, 95% confidence interval). Make clear which confounders were

		(b) Report category boundaries when continuous variables were categorized [table
		1]
		(c) If relevant, consider translating estimates of relative risk into absolute risk for a
		meaningful time period [N/A]
Other analyses	17	Report other analyses done-eg analyses of subgroups and interactions, and
		sensitivity analyses [N/A]
Discussion		
Key results	18	Summarise key results with reference to study objectives [page 14]
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or
		imprecision. Discuss both direction and magnitude of any potential bias [page 16]
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations,
		multiplicity of analyses, results from similar studies, and other relevant evidence
		[page 14,16]
Generalisability	21	Discuss the generalisability (external validity) of the study results [page 17]
Other information		
Funding	22	Give the source of funding and the role of the funders for the present study and, if
		applicable, for the original study on which the present article is based [page 20]

*Give information separately for exposed and unexposed groups.

Note: An Explanation and Elaboration article discusses each checklist item and gives methodological background and published examples of transparent reporting. The STROBE checklist is best used in conjunction with this article (freely available on the Web sites of PLoS Medicine at http://www.plosmedicine.org/, Annals of Internal Medicine at http://www.annals.org/, and Epidemiology at http://www.epidem.com/). Information on the STROBE Initiative is available at www.strobe-statement.org.

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Dietary determinants of serum total cholesterol among middle-aged and older adults: a population-based crosssectional study in Dar es Salaam, Tanzania

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Keywords:	cholesterol, palm oil, diet, Nutrition < TROPICAL MEDICINE, Tanzania, sub Saharan Africa

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1 2	ABSTRACT
3	Objective: To assess the dietary determinants of serum total cholesterol.
4	Design: Cross-sectional population-based study.
5	Setting: Peri-urban region of Dar es Salaam, Tanzania.
6	Participants: 347 adults aged 40 years and older from the Dar es Salaam Urban Cohor
7	Hypertension Study (DUCS-HTN).
8	Main outcome measure: Serum total cholesterol measured using a point-of-care device
9	Results: Mean serum total cholesterol level was 204 mg/dL (Interquartile range (IQR)
10	169 to 236 mg/dL) in women and 185 mg/dL (IQR 152 to 216 mg/dL) in men. After
11	adjusting for demographic, socioeconomic, lifestyle and dietary factors, participants whether the second se
12	reported using palm oil as the major cooking oil had serum total cholesterol higher by
13	mg/dL (95% confidence interval 1 to 29 mg/dL) compared with those who reported usi
14	sunflower oil. Consumption of one or more servings of meat per day (P for trend= 0.01
15	and less than five servings of fruits and vegetables per day (P for trend= 0.024) were al
16	associated with higher serum total cholesterol. A combination of using palm oil for
17	cooking, eating more than one serving of meat per day and fewer than five servings of
18	fruits and vegetables per day was associated with 46 mg/dL (95% confidence interval 1
19	to 76 mg/dL) higher serum total cholesterol.
20	Conclusions: Using palm oil for cooking was associated with higher serum total
21	cholesterol levels in this peri-urban population in Dar es Salaam. Reduction of saturate
22	fat content of edible oil may be considered as a population-based strategy for primary
23	prevention of cardiovascular diseases.
24	Key words: cholesterol, palm oil, diet, nutrition, Tanzania, Sub-Saharan Africa
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5	2	STRENGTHS AND LIMITATIONS OF THIS STUDY
6 7	3	• A reliable point-of care lipid testing system was used to measure serum total
8 9	4	cholesterol that has good clinical agreement with laboratory reference methods.
10 11	5	• Potential confounders of the association between diet and serum total cholesterol were
12	6	systematically measured and adjusted for.
13 14	7	• Only serum total cholesterol was measured instead of a full lipid panel, due to logistic
15 16	8	and financial limitations.
17 18	9	• The relatively small sample size may have resulted in insufficient statistical power to
19 20	10	detect smaller associations.
21	11	• The assessment of diet using any method is prone to measurement error, which may
22 23	12	have resulted in bias, although differential misreporting of cooking oil by cholesterol
24 25	13	level is unlikely.
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INTRODUCTION

Sub-Saharan Africa is facing a burgeoning epidemic of non-communicable diseases
(NCDs) due to changing demographic profiles, epidemiologic transition, urbanization,
and lifestyle changes as a result of economic development as well as increasing survival
of HIV patients.[1, 2] In 2015, one third of all estimated NCD deaths in sub-Saharan
Africa were due to cardiovascular diseases (CVD).[3] By 2030, the share of all deaths
attributable to CVD is projected to increase by nearly 40% in this region.[3]

9 Dyslipidaemia is a well-known risk factor for ischemic heart disease (IHD), stroke, and
10 other vascular diseases.[4, 5] In middle-aged adults, IHD mortality is lower by as much
11 as 30 to 50% for every 1 mmol/L (38.6mg/dL) lower serum total cholesterol.[5] Sub12 Saharan Africans have historically had a favorable lipid profile [6] but hyperlipidaemia is
13 becoming increasingly common in these countries, especially in urban areas.[7-9] It is
14 estimated that thirty-four percent of ischemic heart disease deaths in Tanzania are
15 attributable to dyslipidaemia.[10]

A few studies have already explored the relationship between diet and serum total cholesterol in eastern sub-Saharan Africa and reported an association between higher intake of meat and lower intake of fish and green vegetables with higher serum total cholesterol. [11-15] However, two of these studies compared rural and coastal Bantu populations that are known to have, respectively, vegetarian and fish-based diets and did not measure diet in each participant and based their conclusions on known dietary differences between ethnic populations. [14, 15] Another study included participants from rural, urban and pastoral regions of Tanzania but did not report the magnitude of the association[12] The other two studies conducted in urban Tanzania did not adjust for potential confounders of the relationship between diet and serum total cholesterol.[11, 13] There are a few studies from other parts of sub-Saharan Africa that have looked at the association between diet and serum cholesterol. A study conducted in a rural population in Uganda found that eating fewer than 5 servings of fruits and vegetables daily was associated with higher odds of low HDL cholesterol among men, but lower odds among

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1	women.[16] Another study in urban Uganda found lower mean serum total cholesterol in
2	lacto-ovo-vegetarians compared to non-vegetarians.[17]
3	
4	Another potential dietary determinant of serum cholesterol is the type of oil used in
5	cooking.[18,19] Using cooking oils with higher saturated fat content such as coconut and
6	palm oil has been associated with higher serum total cholesterol levels in clinical trials
7	and observational studies.[20, 21] Population-level differences in CVD morbidity and
8	mortality is associated with differences in type of cooking oil in eastern European,[22]
9	Costa Rica,[23] and Singapore.[24] Mauritius, Seychelles and Singapore have already
10	implemented policy schemes to reduce saturated fat content of cooking oil as population-
11	level interventions to reduce burden of CVD.[25-27]
12	A thorough understanding of the role of dietary determinants of serum cholesterol will be
13	invaluable in designing population-level interventions to address the growing CVD
14	burden especially considering that treatment for dyslipidaemia is either unavailable or
15	prohibitively expensive for most patients in low-income countries.[28] Tanzanian diet
16	consists predominantly of carbohydrates (mainly maize, sweet potatoes, cassava and rice)
17	consumed with green leafy vegetables, beans or peas and occasionally with meat or
18	fish.[29] Palm oil is the major cooking oil, followed by sunflower oil.
10	
19	We conducted a cross-sectional population-based study to assess the association of type
20	of cooking oil (palm vs sunflower oil) and diet (meat, fish, fruit and vegetable
21	consumption) with serum total cholesterol in an adult population in a peri-urban area of
22	Dar es Salaam. We hypothesized that use of palm oil for cooking and higher intake of
23	meat and lower intake of fruits and vegetables will be associated with higher serum total
24	cholesterol.
25	

1 METHODS

2 Study design and population

The Dar es Salaam Health and Demographic Surveillance System (HDSS) is a demographic surveillance system in Ukonga and Gongo la Mboto wards of Dar es Salaam, Tanzania with over 100,000 enumerated individuals living in 21,000 households followed since 2011/12. For the Dar es Salaam Urban Cohort Hypertension Study (DUCS-HTN), we randomly selected one large and one small neighborhood in the HDSS and contacted all adult residents aged 40 years and older, who had been registered in the HDSS database in 2011 or 2012. A random subsample of one-fifth of the eligible participants was selected for additional assessment of diet and measurement of serum total cholesterol. The size of this subsample was determined largely based upon the availability of resources. Pregnant women and those who were physically and mentally incapable of completing the interview and measurements were excluded. The participant selection flowchart is shown in Figure 1. Face-to-face interviews and measurements were conducted in participant's home from March to June 2014 by six trained interviewers. All interviewers had previous experience conducting public health surveys. For this study, interviewers received five days of training and two days of field practice in a nearby neighborhood. At least three attempts were made to contact all potential participants. Written informed consent was obtained from all participants. The study protocol was approved by the Institutional Review Board of the Harvard TH Chan School of Public Health and the Research and Publications Committee of Muhimbili University of Health and Allied Sciences.

23 Measurements

Non-fasting serum total cholesterol was measured from 30 micro-liters of finger-prick
capillary blood samples using Cardiochek PA devices (Polymer Technology Systems Inc,
IN, USA).[30, 31] The overall coefficient of variation for this device for total cholesterol
has been determined to be in the range of 1.3 to 2.9%.[30]

28 Data on diet was collected using a semi-quantitative food frequency questionnaire (FFQ).

29 The FFQ assessed intake of 179 food and drink items over the past 30 days. A shorter 85-

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item version of this instrument has been previously used in Tanzania.[32] Nutrient intakes were calculated by multiplying the frequency of food consumption measured from FFQ by the nutrient content of the specified portion size using the Tanzanian food composition table.[33] Meat intake was defined to include all red and white meat consumption, excluding fish consumption. Fruit and vegetable intake was defined to include consumption of all fruits and vegetables, except for root vegetables and fruit juice. An additional question was asked to assess the main type of cooking oil: "What is the kind of oil used most often in your home for frying food?". Frying is the most common cooking technique involving oil use in this region. The reference to a cooking technique was made to distinguish it from other oils that may be used in a household, such as fuels.

All participants completed a socioeconomic, demographic and lifestyle questionnaire and had their height, weight, waist circumference, hip circumference, and blood pressure measured by trained interviewers. Some demographic information (age, sex, neighborhood, components of wealth index) was taken from the HDSS baseline interview. Body mass index was calculated as weight (kg) divided by height (m) squared, and categorized as underweight (<18.5), normal (18.5 to <25.0), overweight (25.0 to <30.0), and obese (≥ 30.0).[34] Physical activity was assessed using the Global Physical Activity Ouestionnaire in the domains of work, transportation, and leisure, [35] and categorized into tertiles of Metabolic Equivalent (MET) hours per week. To measure socioeconomic status, we created a household wealth index using a principal component analysis of household characteristics (floor material, roof material, wall material, electricity), household assets (television, radio, shop, sewing machine, sofa, fan, iron, stove with oven, stove without oven, dining table, cupboard, watch, mobile phone, bike, motorcycle, cart, car, motorboat), and animal ownership (goats, sheep, chicken/ducks, pigs).[36] The household wealth index was categorized into tertiles.

28 Statistical analysis

We excluded participants with implausible dietary data (total energy intake <500 or
 >5000 kcal/day), and with missing data on serum total cholesterol, diet and/or any of the
 potential covariates.

Chi-square tests were used to assess binary variables, F-tests were used for categorical variables and tests of linear trend based on median value within each category for ordinal variables with a linear association with serum total cholesterol (i.e. age, wealth index, and food consumption categories). Mean differences and 95% confidence intervals in serum total cholesterol across categories of dietary factors, including cooking oil were estimated using multivariable linear regression. Potential confounders were included in the model based on a-priori knowledge of lifestyle and socio-demographic determinants of diet and serum total cholesterol. The fully adjusted model included age, sex, total energy intake, physical activity, alcohol consumption, wealth index and employment status as covariates. We also adjusted for interviewer to remove the effect of any extraneous variation in measurement of subjective risk factors across interviewers. [37, 38] We examined interactions of potential determinants of serum total cholesterol with sex, but no interactions were statistically significant at 0.2 level, therefore we did not include these product terms in the final model. We conducted sensitivity analyses by excluding 16 participants who reported either being previously diagnosed with hypercholesterolaemia, or currently taking cholesterol-lowering medications or being advised by a health professional to modify diet to lower cholesterol. All analyses were performed using STATA software version 13.1 (STATA Corporation, College Station, Texas).

	1	
	2	RESULTS
	3	A total of 420 participants were enrolled for the random subsample with diet and
	4	cholesterol measurements. Among these, data on serum total cholesterol and selected
)	5	covariates was available for 399 participants. We excluded 43 participants due to
<u> </u> 2	6	implausible caloric intake. In addition, after descriptive analyses we excluded 9 (2%)
3 1	7	participants who reported using cooking oils other than palm or sunflower oil. The final
5	8	analytic sample included 347 participants (FIGURE 1).
7 }	9	Forty-two percent of the participants (n=347) were men (TABLE 1). Mean age of men
)	10	was 55 years (SD 11), and that of women was 52 (SD 10). 44% of the women (n=202)
	11	and 19% of men (n=145) were obese, and one third of men and women were overweight.
<u>2</u> 3	12	Mean energy intake per day was 2413 kcal. Thirty-seven percent of men and 54% of
4 5	13	women were hypercholesterolemic (i.e. had total cholesterol \geq 200 mg/dL). Mean serum
) 7	14	total cholesterol in women was 204 mg/dL (95% confidence interval 197 to 211 mg/dL)
3	15	and was significantly higher than that of men at 185 mg/dL (177 to 193 mg/dL). Serum
) 	16	total cholesterol levels were higher in older age groups.
2 2 3	17	79% of participants reported using palm oil as the major cooking oil and the rest reported
1	18	using sunflower oil (TABLE 1). Those who reported using palm oil were most likely to
) }	19	be in the poorest tertile of wealth, whereas those who reported using sunflower oil were
3	20	most likely to be in the wealthiest tertile ($p < 0.001$). Those who reported using sunflower
)	21	oil had a higher energy intake (p=0.04), and reported consuming meat (p=0.004) and
	22	dairy (p=0.02) more frequently than those who reported using palm oil. Mean serum total
<u>-</u> 3	23	cholesterol in those who reported using palm oil was 199 mg/dL (193 to 205) and was
4 5 6 7	24	significantly higher than those who reported using sunflower oil 184 mg/dL (173 to 195).

Variable	All (n=347)	Men (n=145)	Women (n=202)	Use palm oil (n=274)	Use sunflower oil (n=73)
Age					
40-49	150 (43)	56 (39)	94 (47)	123 (45)	27 (37)
50-59	113 (33)	43 (30)	70 (35)	82 (30)	31 (42)
60 and above	84 (24)	46 (32)	38 (19)	69 (25)	15 (21)
Employment status					
Unemployed	83 (24)	14 (10)	69 (34)	70 (26)	13 (18)
Employed (includes self-	238 (69)	112 (77)	126 (62)	183 (67)	55 (75)
Retired	26 (7)	19 (13)	7 (3)	21 (8)	5 (7)
Household wealth index					
Tertile 1, Poorest	115 (33)	49 (34)	66 (33)	109 (40)	6 (8)
Tertile 2	117 (34)	53 (37)	64 (32)	90 (33)	27 (37)
Tertile 3, Wealthiest	115 (33)	43 (30)	72 (36)	75 (27)	40 (55)
BMI (kg/m ²)					
Underweight (<18.50)	19 (5)	13 (9)	6 (3)	19 (7)	0 (0)
Normal weight (18.50-<25.00)	100 (29)	56 (39)	44 (22)	91 (33)	9 (12)
Overweight (25-<30.00)	112 (32)	48 (33)	64 (32)	81 (30)	31 (42)
Obese (≥ 30.00)	116 (33)	28 (19)	88 (44)	83 (30)	33 (45)
Physical activity					
Tertile 1, (0-14 MET-hours/week)	123 (35)	46 (32)	77 (38)	96 (35)	27 (37)
Tertile 2, (15-112 MET-hours/week)	108 (31)	38 (27)	70 (35)	86 (31)	22 (30)
Tertile 3, (113-840 MET-hours/week) Alcohol drinking ^a	116 (33)	61 (42)	55 (27)	92 (34)	24 (33)

Table 1. Participants' characteristics in DUCS-HTN biomarker sub-study, 2014

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Variable	All (n=347)	Men (n=145)	Women (n=202)	Use palm oil (n=274)	Use sunflower oil (n=73)
Non-drinker	286 (82)	103 (71)	183 (91)	225 (82)	61 (84)
Smoking					
Non-smoker	278 (81)	85 (59)	193 (97)	210 (78)	68 (93)
Former smoker	43 (13)	39 (27)	4 (2)	38 (14)	5 (7)
Current smoker	22 (6)	19 (13)	3 (2)	22 (8)	0 (0)
DIETARY VARIABLES					
Palm oil as major cooking oil	274 (79)	119 (82)	155 (77)		
Meat intake					
<1 serving/week	41 (12)	21 (14)	20 (10)	36 (13)	5 (7)
1-6 servings/week	173 (50)	55 (38)	118 (58)	145 (53)	28 (38)
≥1 servings/day	133 (38)	69 (48)	64 (32)	93 (34)	40 (55)
Fish intake					
< 1 serving/week	41 (12)	13 (9)	28 (14)	31 (11)	10 (14)
1-7 servings/week	196 (56)	76 (52)	120 (59)	154 (56)	42 (58)
> 1 servings/day	110 (32)	56 (39)	54 (27)	89 (32)	21 (29)
Dairy intake					
< 1 serving/week	159 (46)	63 (43)	96 (48)	136 (50)	23 (32)
1-6 servings/week	133 (38)	65 (45)	68 (34)	99 (36)	34 (47)
\geq 1 servings/day	55 (16)	17 (12)	38 (19)	39 (14)	16 (22)
Fruit and vegetable					
< 5 servings/day	121 (35)	49 (34)	72 (36)	101 (37)	20 (27)
5-7 servings/day	68 (20)	26 (20)	42 (21)	54 (20)	14 (19)
>7 servings/day	158 (46)	70 (48)	88 (44)	119 (43)	39 (53)

Variable	All (n=347)	Men (n=145)	Women (n=202)	Use palm oil (n=274)	Use sunflower oil (n=73)	
Nuts and legumes						
< 1 serving/week	27 (8)	7 (5)	20 (10)	17 (6)	10 (14)	
1-7 servings/week	200 (58)	87 (60)	113 (56)	162 (59)	38 (52)	
> 1 servings/day	120 (35)	51 (35)	69 (34)	95 (35)	25 (34)	
Total energy intake (kcal/day) ^b	2413 (1701, 3215)	2629 (1780, 3216)	2313 (1617, 3207)	2369 (1654, 3150)	2743 (1865, 3606)	
Fat intake ^b						
Total fat (% of energy)	17 (15, 20)	17 (15, 21)	17 (15, 20)	17 (14, 20)	20 (15, 22)	
Saturated fat (% energy)	10 (8, 13)	10 (8, 13)	10 (8, 12)	10 (8, 13)	10 (8, 11)	
Monounsaturated fat (% of energy)	4 (3, 5)	4 (3, 5)	4 (3, 5)	4 (3, 5)	4 (4, 5)	
Polyunsaturated fat (% of energy)	2 (1, 2)	2 (1, 2))	2 (1, 2)	1 (1, 2)	2 (1, 2)	
Cholesterol						
Serum total cholesterol (mg/dl) ^c	196 (3)	185 (4)	204 (4)	199 (3)	184 (6)	
Hypercholesterolaemia ^d	164 (47)	54 (37)	110 (54)	135 (49)	29 (40)	
Cholesterol level was previously checked	29 (18)	7 (13)	22 (20)	13 (10)	16 (55)	
Previously diagnosed hypercholesterolaemia	13 (8)	1 (2)	12 (11)	5 (4)	8 (28)	
Currently taking cholesterol lowering medications	9 (5)	1 (2)	8 (7)	3 (2)	6 (21)	

Number (%) reported for all variables, except as specified b and c below

2 3

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^a According to report of drinking alcohol in the past 30 days ^b Total energy and fat intake, which is reported as median (interquartile range) ^c Serum total cholesterol, reported as mean (standard error)

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.slesterol ≥ 200 mg/dl. ^d Hypercholesterolaemia defined as total cholesterol $\geq 200 \text{ mg/dL}$

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In the minimally adjusted model (with interviewer as the only covariate), those who reported using palm oil as the major cooking oil had serum total cholesterol levels higher by 11 mg/dL (-2 to 24 mg/dL) compared with those who reported using sunflower oil (TABLE 2). After adjusting for covariates, participants who used palm oil for cooking had 15 mg/dL (1 to 29 mg/dL) higher serum total cholesterol (FIGURE 2). The negative confounding was mostly due to the negative correlation between female gender and meat intake with palm oil use (both variables were positively correlated with serum total cholesterol). Serum total cholesterol was also higher in participants who ate meat more frequently (P for trend 0.017) after adjusting for potential confounders. Specifically, those who ate more than one serving of meat per day had 18 mg/dL (-3 to 39 mg/dL) higher serum total cholesterol compared with those who ate less than one serving per week. In this relationship, higher total energy intake was the major confounder with a positive correlation with meat intake but negative correlation with serum total cholesterol. Consumption of more than seven servings of fruits and vegetables per day was associated with 13 mg/dL (-29 to 3 mg/dL) lower serum total cholesterol, compared to consumption of fewer than five servings per day (P for trend 0.024). A combination of using palm oil for cooking, eating more than one serving of meat per day and fewer than five servings of fruits and vegetables per day was associated with 46 mg/dL (16 to 76 mg/dL) higher serum total cholesterol. We found similar associations after excluding 16 participants who reported either being previously diagnosed with hypercholesterolaemia, or currently taking cholesterol lowering medications or being advised by a health professional to modify diet to lower cholesterol. Those who used palm oil had 17 mg/dL (2 to 32 mg/dL) higher serum total cholesterol compared with those who used sunflower oil (TABLE 2). There was no significant association between percent energy from saturated fat and total serum cholesterol after adjusting for total fat, total protein and total carbohydrate (Adjusted mean difference 1mg/dL (-3 to 5 mg/dL))

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Table 2. Adjusted mean differences for serum total cholest	erol (mg/dL) by dietary factors; DUCS-HTN biomarker sub-study
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Variable	Interviewer- adjusted mean difference (95% CI)	P Value ^a	Fully-adjusted ^b mean difference (95% CI) (n=347)	P Value ^a	Fully-adjusted ^c mean difference after excluding 16 participants diagnosed or treated for dyslipidaemia (95% CI) (n=331)	P Value ^a
Major cooking oil		0.092		0.033		0.028
Sunflower oil	Reference		Reference		Reference	
Palm oil	11 (-2, 24)		15 (1, 29)		17 (2, 32)	
Meat intake		0.485		0.017		0.031
<1 serving/week	Reference		Reference		Reference	
1-6 servings/week	-1 (-19, 16)		-1 (-19, 17)		-2 (-20, 17)	
≥ 1 servings/day	4 (-15, 22)		18 (-3, 39)		16 (-5, 37)	
Fruit and vegetable		0.028		0.011		0.024
intake						
< 5 servings/day	16 (2,30)		13 (-3, 29)		12 (-5, 29)	
5-7 servings/day	10 (-6, 25)		8 (-8, 24)		10 (-7, 26)	
> 7 servings/day	Reference		Reference		Reference	
Fish intake		0.399		0.446		0.309
< 1 serving/week	14 (-8, 36)		10 (-13, 33)		14 (-9, 38)	
1-7 servings/week	3 (-11, 17)		2 (-13, 17)		4 (-11, 20)	
>1 servings/day	Reference		Reference		Reference	
Dairy intake		0.802		0.572		0.690
< 1 serving/week	Reference		Reference		Reference	
1-6 servings/week	-9 (-22, 4)		-7 (-21, 6)		-7 (-21, 7)	
\geq 1 servings/day	-2 (-18, 15)		-2 (-20, 15)		-1 (-20, 17)	

Variable	Interviewer- adjusted mean difference (95% CI)	P Value ^a	Fully-adjusted ^b mean difference (95% CI) (n=347)	P Value ^a	Fully-adjusted ^c mean difference after excluding 16 participants diagnosed or treated for dyslipidaemia (95% CI) (n=331)	P Value ^a
Total energy intake	-8 (-21, 5)	0.226	-5 (-12, 3)	0.209	-4 (-11, 4)	0.365
(Per 1000 kcal)						
Age		0.106		0.071		0.099
40-49	Reference		Reference		Reference	
50-59	4 (-9, 16)		6 (-6, 19)		5 (-9, 18)	
60 and above	11 (-2, 25)		16 (0, 31)		15 (-1, 31)	
Sex		< 0.001		< 0.001		< 0.001
Male	Reference		Reference		Reference	
Female	21 (10, 32)		27 (15, 39)		24 (11, 36)	
Employment status		0.432		0.567		0.581
Unemployed	Reference		Reference		Reference	
Employed ^d	-7 (-20, 6)		7 (-9, 22)		5 (-12, 21)	
Retired	2 (-21, 24)		12 (-13, 36)		13 (-12, 39)	
Household Wealth		0.692		0.754		0.952
Index						
1, Poorest	Reference		Reference		Reference	
2	-9 (-22, 4)		-8 (-21, 5)		-7 (-20, 7)	
3, Wealthiest	-1 (-15, 12)		0 (-14, 14)		2 (-13, 16)	
Physical activity tertile		0.953		0.430		0.599
1, least active	Reference		Reference		Reference	
2	1 (-13, 15)		2 (-12, 16)		4 (-11, 18)	
3, most active	0 (-15, 15)		7 (-9, 24)		6 (-11, 24)	

Variable	Interviewer- adjusted mean difference (95% CI)	P Value ^a	Fully-adjusted ^b mean difference (95% CI) (n=347)	P Value ^a	Fully-adjusted ^c mean difference after excluding 16 participants diagnosed or treated for dyslipidaemia (95% CI) (n=331)	P Value ^a
Alcohol consumption ^e		0.315		0.666		0.641
Non-drinker	Reference		Reference		Reference	
Drinker	-7 (-21, 7)		-3 (-18, 11)		-4 (-18, 11)	
for categorical variables	(cooking oil, sex, en	ployment sta	tus, alcohol consumption)	; t test fo	r continuous variables (total	energy intak

cat**4**gories)

^b Afaljusted for covariates in column 1 as well as interviewer

^c Excluding those previously diagnosed with hypercholesterolaemia; adjusted for covariates in column 1 as well as interviewer

^d S**∂**If-employed, or government job, or job in private company

^e According to report of drinking alcohol in the past 30 days

1 Only 11% of women and 5% of men reported having ever had their cholesterol levels

2 checked (TABLE 1; FIGURE 3). Among those with hypercholesterolaemia (n=164), 2%

3 of men (1 of 54), and 11% of women (12 of 110) reported being notified by doctors about

4 it, and 2% of men and 7% of women reported taking cholesterol-lowering medications.

DISCUSSION

In this cross-sectional study among middle-aged and older adults in a peri-urban ward in Dar es Salaam, Tanzania, we found a high prevalence of hypercholesterolaemia. Slightly more than a third of men and half of women had serum total cholesterol levels greater than or equal to 200mg/dL. After adjusting for major confounders, serum total cholesterol was higher by 15 mg/dL among those who used palm oil compared to those who used sunflower oil as the major cooking oil, and showed a significant increasing trend with higher consumption of meat (P=0.017) and lower consumption of fruits and vegetables (P=0.01).

Mean serum total cholesterol levels for men and women were similar to that reported in other studies conducted in other urban Tanzanian populations.[32, 39, 40] A repeated cross-sectional study reported an increase over time in the prevalence of hypercholesterolaemia in Dar es Salaam with a prevalence of 17% in 1987 versus 30% in 1998 in men, and 7% versus 50% in women.[39] The prevalence of hypercholesterolaemia in our sample was higher than that reported by studies conducted between 2008-2014 in rural Uganda (3% men, 8% women),[9] peri-urban Nigeria (2% men, 6% women).[8] and blacks in urban South-Africa (25% men, 23% women).[41] but is lower than that reported in a study from Senegal (54% men, 61% women) that included both urban and rural participants.[42] Our estimated association between palm oil versus sunflower oil use and serum total

cholesterol is consistent with findings from dietary observational studies and intervention
trials. A recent meta-analysis of 30 dietary intervention trials comparing diets rich in

30 palm oil with diets rich in vegetable oils low in saturated fatty acids showed significantly

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higher total (13.5 mg/dL) and LDL (9.3 mg/dL) cholesterol, and slightly higher HDL
cholesterol (0.8 mg/dL) 2 to 16 weeks after intervention.[21] In addition, a large casecontrol study conducted in Costa Rica found that palm oil use compared to soybean oil
use was independently associated with 33% higher odds of myocardial infarction. [23].

6 Higher meat consumption has previously been reported to be associated with higher 7 serum total cholesterol in Tanzania. [12, 13] Similar to our finding, daily consumption of 8 meat was found to be associated with 24 mg/dL higher serum total cholesterol levels, 9 after adjusting for age, select dietary components and resting energy expenditure.[13] 10 Observational studies have also reported lower LDL cholesterol by 2.3mg/dL for every 11 additional serving of fruits or vegetables.[43-45] The magnitude of 13 mg/dL reduction 12 in serum total cholesterol that we found for six or more servings of fruits and vegetables 13 per day is consistent with the reductions in LDL-cholesterol seen in these studies. We did 14 not find significant associations between serum total cholesterol and fish intake in men. 15 as reported in previous studies. [12, 13] This could have been due to insufficient 16 statistical power due to smaller expected effect size for fish intake or higher measurement 17 error.

18

19 Palm oil is obtained mostly from the middle section (pulp) of the fruits of the tropical 20 plant *Elaeis guineensis*.[46] Compared with most other vegetable oils, it contains less 21 unsaturated fat and more saturated fat (nearly 40 to 50% of total fat content) with the 22 majority being palmitic acid. [46] However, compared with saturated fat from animal 23 sources, palm oil may have a smaller impact on serum cholesterol because almost 70% of 24 the palmitic acid is located in the first and third position of the triglycerides, which is less 25 absorbable than those in the second position (which is the most common position of 26 palmitic acid in animal fats). [47, 48] Despite these biological differences, evidence from 27 both observational and experimental studies, as summarized above, points to harmful 28 effects of palm oil on serum cholesterol and CVD when substituted for unsaturated fatty 29 acids.

30

Our study has several limitations. First, due to logistic and financial limitations, we measured serum total cholesterol instead of a full lipid panel. However, data from recent meta-analysis of randomized trials suggests that the effect of substituting palm oil for vegetable oils low in saturated fat on LDL cholesterol levels is larger than that on HDL.[21] Second, the assessment of diet using any method is prone to measurement error which may have resulted in bias. However, it is unlikely that participants with higher serum cholesterol differentially misreport using palm oil in cooking, limiting the impact of such measurement error on our results. Thirdly, the relatively small sample size may have resulted in insufficient statistical power to detect some associations (e.g. with fish intake). There are some important strengths of this study. To our knowledge, this is the first population-based study to report an association between using palm oil for cooking and serum total cholesterol in East Africa. We used a reliable point-of-care lipid testing system that has good clinical agreement with laboratory reference methods.[31] We systematically collected data on and adjusted for potential confounders of the association between diet and serum total cholesterol.

Were our findings to be replicated in other studies, they could have important implications for nutritional policies aimed at CVD prevention. Palm oil is now the major edible oil in Tanzania and many other developing countries.[49] In Tanzania, more than half of the palm oil is imported, [50] and it is the second most imported agricultural commodity.[51] In our study population, more than three-quarters of participants used palm oil (cost 2500 Tanzanian Shillings \approx USD1.1 per litre as the major cooking oil with 94% of participants (n=119) in the lowest tertile of wealth using palm oil. Even in the wealthiest tertile, only 34% (n=123) used sunflower oil (cost 4000 Tanzanian Shillings \approx USD1.8 per litre). These socioeconomic differentials in palm oil use may affect socioeconomic disparities in cardiovascular disease in the coming years with the burden shifting more swiftly to poorer households.

29 Policy interventions to address composition of dietary fat have previously been

- 30 implemented in Seychelles, Mauritius and Singapore. [27, 52, 53] In the early 1990s,
- 31 Seychelles implemented a nationwide program that included increasing use of

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unsaturated vegetable oils, which may have contributed to the fairly high rate of decline in age-standardized CVD mortality of 44% in men and 28% in women over a 22-year period.[25,52,54] A national NCD prevention program was also implemented in Mauritius in the late 1980s to reduce saturated fat content of cooking oil by changing the government subsidized ration oil from palm oil to soybean oil. The observed 31 mg/dL reduction in serum total cholesterol over a period of 5 years [26, 53] is compatible with the estimated impact of replacing palm oil with unsaturated vegetable oil estimated in a recent meta-analysis of dietary intervention trials.[55] Similar policies can be considered to reduce palm oil intake in Tanzania. However, it should be noted that edible oils are viewed as necessities, [56] and increasing their price (e.g. by taxation) without providing healthier alternatives at comparable prices may have unintended adverse consequences on overall energy consumption or food security.[57-59] Reducing saturated fat in edible oil by mixing it with other unsaturated oils has been proposed as a potential intervention in Costa Rica and India, [23, 60] and could be a feasible short-term intervention in Tanzania.

In conclusion, our results indicate that using palm oil for cooking, lower intake of fruits and vegetables and higher intake of meat is strongly associated with higher serum total cholesterol in this peri-urban population in Tanzania. Our findings are generalisable to other peri-urban populations in Tanzania and possibly to populations in low-income countries with a similar dietary profile. Dietary policies aimed at altering the targets identified in this study can be used to improve lipid profiles and therefore prevent cardiovascular diseases in these countries.

CONTRIBUTORS GD, JK, GHL and RMZ conceived and designed the study. GD obtained the funding. SSK and SF analyzed the data. GD and SSK interpreted the data and drafted the article, which was revised critically for substantive content by all authors, who approved the final version for publication. GD, SSK, SF and RMZ had full access to all of the data (including statistical reports and tables) in the study and can take responsibility for the integrity of the data and the accuracy of the data analysis. GD is the study guarantor.

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no relationships with companies that might have an interest in the submitted work in the
previous three years; their spouses, partners, or children have no financial relationships
that may be relevant to the submitted work; no non-financial interests that may be
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ETHICAL APPROVAL: The study protocol was approved by the Institutional Review
Board of the Harvard TH Chan School of Public Health and the Research and

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TRANSPARENCY: The lead authors affirm that the manuscript is an honest, accurate,

study have been omitted; and that any discrepancies from the study as planned have been

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DATA SHARING: No additional data available

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1 Figure legends

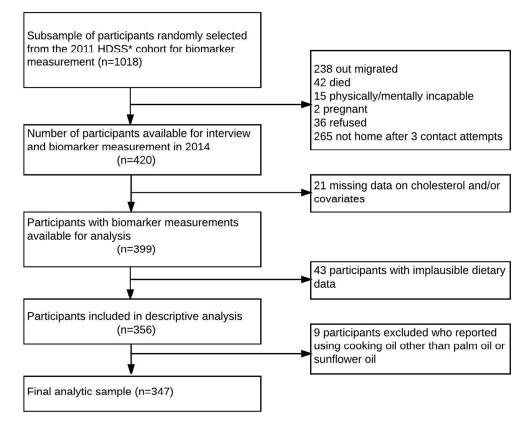
Figure 1. Selection of study participants, DUCS-HTN biomarker sub-study, 2014

Figure 2. Association between diet and serum total cholesterol. Values were adjusted for
age, sex, socioeconomic status, food consumption frequency, total energy intake, major
cooking oil used, physical activity and alcohol consumption.

9 Figure 3. Diagnosis, awareness and treatment of hypercholesterolaemia in men (Part A;

10 n=54 for all percentages) and women (Part B; n=110 for all percentages) in the DUCS-

11 HTN biomarker sub-study, 2014.



*Dar Es Salaam Health and Demographic Surveillance System

Figure 1. Selection of study participants, DUCS-HTN biomarker sub-study, 2014

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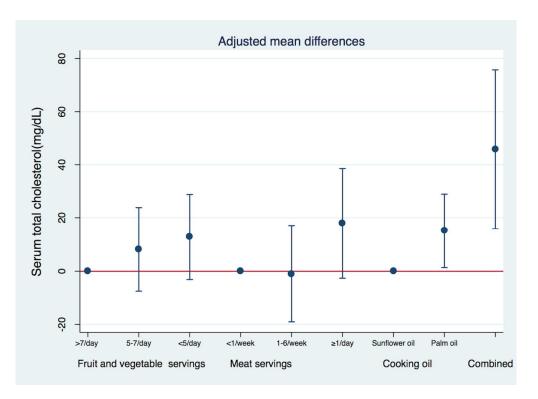
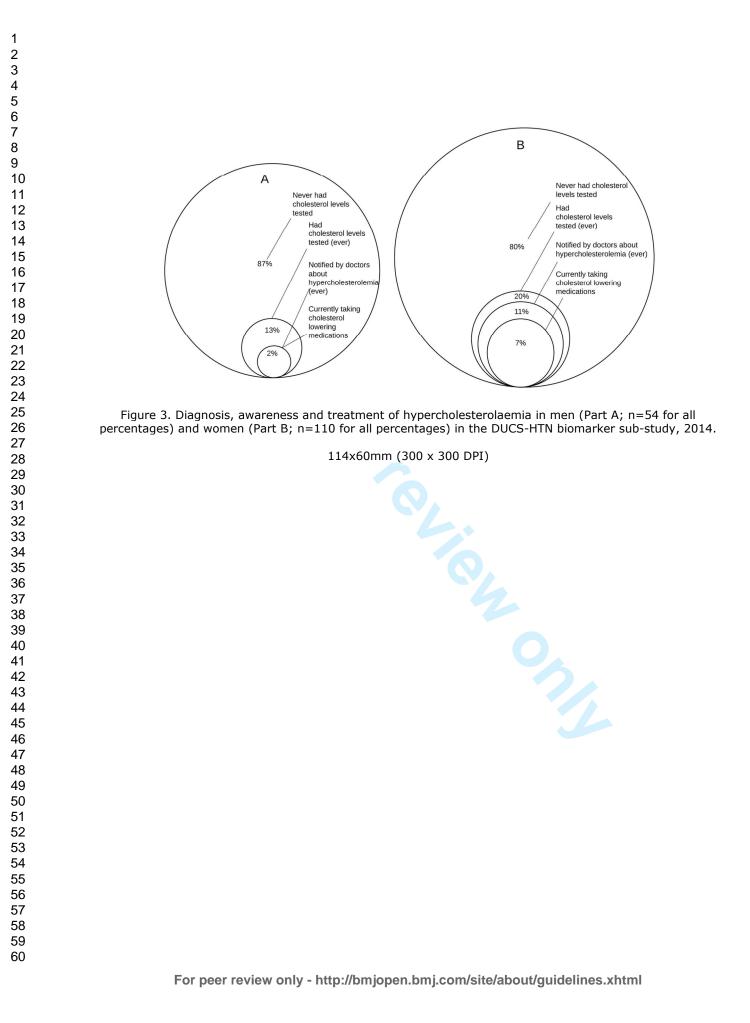


Figure 2. Association between diet and serum total cholesterol. Values were adjusted for age, sex, socioeconomic status, food consumption frequency, total energy intake, cooking oil, physical activity and alcohol consumption.

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STROBE Statement—Checklist of items that should be included in reports of *cross-sectional studies*

	Item No	Recommendation
Title and abstract	1	(<i>a</i>) Indicate the study's design with a commonly used term in the title or the abstract
		[Within the title page 1 and method section of the abstract page 2]
		(b) Provide in the abstract an informative and balanced summary of what was done
		and what was found [See results section of the abstract page 2]
Introduction		ma num na coma [secondation of the second page -]
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported
Duckground/rationale	2	[pages 4,5]
Objectives	3	State specific objectives, including any prespecified hypotheses [page 5]
Methods		Sum specific selection and many properties all periods and
Study design	4	Present key elements of study design early in the paper [page 6]
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment,
Setting	5	exposure, follow-up, and data collection [page 6]
Participants	6	(a) Give the eligibility criteria, and the sources and methods of selection of
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Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect
		modifiers. Give diagnostic criteria, if applicable [page 6-8]
Data sources/	8*	For each variable of interest, give sources of data and details of methods of
measurement		assessment (measurement). Describe comparability of assessment methods if there is
		more than one group [page 6-8]
Bias	9	Describe any efforts to address potential sources of bias [page 8]
Study size	10	Explain how the study size was arrived at [page 6]
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable,
		describe which groupings were chosen and why [pages 7, 8]
Statistical methods	12	(a) Describe all statistical methods, including those used to control for confounding
		[page 8]
		(b) Describe any methods used to examine subgroups and interactions [page 8]
		(c) Explain how missing data were addressed [page 8]
		(d) If applicable, describe analytical methods taking account of sampling strategy
		[N/A]
		(e) Describe any sensitivity analyses [page 8]
Results		
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially
-		eligible, examined for eligibility, confirmed eligible, included in the study,
		completing follow-up, and analysed [pages 6, 9 and Figure 1]
		(b) Give reasons for non-participation at each stage [Figure 1]
		(c) Consider use of a flow diagram [Figure 1]
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and
-		information on exposures and potential confounders [page 9 and Table 1]
		(b) Indicate number of participants with missing data for each variable of interest
		[Figure 1]
Outcome data	15*	Report numbers of outcome events or summary measures [pages 9, 18 and Figure
		3]
Main results	16	(a) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and

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		adjusted for and why they were included [page 14 and Table 2]
		(b) Report category boundaries when continuous variables were categorized [table
		(c) If relevant, consider translating estimates of relative risk into absolute risk for a
		meaningful time period [N/A]
Other analyses	17	Report other analyses done-eg analyses of subgroups and interactions, and
		sensitivity analyses [page 14]
Discussion		
Key results	18	Summarise key results with reference to study objectives [pages 18, 19]
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or
		imprecision. Discuss both direction and magnitude of any potential bias [page 20]
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations,
		multiplicity of analyses, results from similar studies, and other relevant evidence
		[pages 21]
Generalisability	21	Discuss the generalisability (external validity) of the study results [pages 21]
Other information		
Funding	22 <	Give the source of funding and the role of the funders for the present study and, if
		applicable, for the original study on which the present article is based [page 22]

*Give information separately for exposed and unexposed groups.

Note: An Explanation and Elaboration article discusses each checklist item and gives methodological background and published examples of transparent reporting. The STROBE checklist is best used in conjunction with this article (freely available on the Web sites of PLoS Medicine at http://www.plosmedicine.org/, Annals of Internal Medicine at http://www.annals.org/, and Epidemiology at http://www.epidem.com/). Information on the STROBE Initiative is available at www.strobe-statement.org.