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Associations of dietary protein intake with fat free mass and grip strength: cross-sectional study in 146,816 UK Biobank participants

Celis-Morales CA[1*], Petermann F[1*], Steell L[1], Anderson J[2], Welch P[1], Mackay DF[2], Iliodromiti S[3], Lyall DM[2], Lean ME[3], Pell JP[b], Sattar N[1], Gill JMR[1,] Gray SR[1]

[1] Institute of Cardiovascular and Medical Sciences, [2] Institute of Health and Wellbeing and [3] School of Medicine, University of Glasgow, Glasgow, UK

* CACM and FP contributed equally to this work and are joint-first authors.

Corresponding author. Institute of Cardiovascular and Medical Sciences, University of Glasgow, BHF Glasgow Cardiovascular Research Centre, Glasgow, G12 8TA, UK. E-mail: stuart.gray@glasgow.ac.uk

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the study and take responsibility for the integrity of the data and the accuracy of the data analysis.

Abstract

Adequate dietary protein intake is important for the maintenance of fat-free mass (FFM) and muscle strength: however optimal requirements remain unknown. The aim of the current study was to explore the associations of protein intake with FFM and grip strength. We used baseline data from the UK Biobank (146,816 participants aged 40-69 years) to examine the associations of protein intake with FFM and grip strength. Protein intake was positively associated with FFM (men 5.1% [95% CI: 5.0; 5.2] and women 7.7% [95% CI: 7.7; 7.8]) and grip strength (men 0.076 kg/kg [95% CI: 0.074; 0.078] and women 0.074 kg/kg [95% CI: 0.073; 0.076]) per 0.5 g/kg/day increment in protein intake. FFM and grip strength were higher with higher reported intakes across the full range of intakes, i.e. highest in those reporting consuming >2.0 g grams per kg body mass per day (g/kg/day) independently of socio-demographics, other dietary measures, physical activity and comorbidities. FFM and grip strength were both lower with age, but this association did not differ by category of protein intake (P>0.05). These data suggest that current recommendation for all adults (40-69 years) for protein intake (0.8 g/kg/day) may need to be increased to optimise FFM and grip strength.

Keywords: Protein, grip strength, UK Biobank, fat-free mass, age

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Introduction

Low muscle strength and muscle mass are associated with increased risk of mortality (1–3). Muscle strength and muscle mass both decrease progressively after 35-40 years of age (4–6), eventually leading to a reduction in the ability to carry out everyday tasks such as standing from a seated position, an increased risk of falls and associated fractures, and a decrease in quality of life (7,8). The exact cause of this age-related decline in muscle mass and function remains to be established but is likely multifactorial in nature – involving inactivity, genetics, hormonal changes, chronic-low grade inflammation, motor unit loss, dietary changes and alterations in anabolic responses to exercise/nutrients (9,10). Expanding on this final point, it has been suggested that older people may require a greater daily protein intake than younger people, to optimise muscle mass and function (11).

The current dietary recommendation for protein intake is for all adults to consume 0.8 grams of protein per kg body mass per day (g/kg/day) (12,13). However this recommendation is based primarily on nitrogen-balance studies (12,13), which have a number of limitations and are generally now considered inappropriate for establishing recommendations (14). For older adults (> 65 years), it has been argued that protein intake should be higher, at 1.0-1.2 g/kg/day (15,16). This assertions is made based upon recent studies using the indicator amino acid oxidation technique, which showed greater protein requirements in older people (17,18), and findings that older people require a greater protein intake (relative to body weight) to maximally stimulate muscle protein synthesis (23). There have been few studies evaluating the associations between protein intake and muscle mass/function; those that exist are generally relatively small and primarily focused only on older adults (aged > 60 years).

Their results have been conflicting (19–24) and the studies have been unable to investigate associations between muscle mass/function and defined protein intake categories across a range of ages. Such data are important to help determine optimal levels of protein intake and whether this optimum varies by age, which could potentially feed into dietary protein recommendations.

The aim of the current study, therefore, was to explore the associations of reported protein intake with FFM and grip strength, and how these varied with age and sex, in UK Biobank, a very large general population cohort study of participants aged 40-69 years.

Methods

Study design

UK Biobank (www.ukbiobank.co.uk) is a very large, general population cohort study. Between 2007 and 2010, 502,628 participants, aged 40–69 years, were recruited and participated in baseline assessments at 22 centres across England, Scotland and Wales. Detailed information was obtained via a self-completed, touch-screen questionnaire and a face-to-face interview, and trained staff undertook a series of measurements using standard operating procedures. In the current study the main outcome measures considered were fat-free mass (FFM), as a percentage of body mass, and grip strength, expressed relative to body mass (kg/kg body mass). The independent predictor variable of interest was daily protein intake (g/kg/day). Protein intake was expressed as g/kg/day as these are the units used in the current recommendations and most relevant for maintenance of muscle mass/function. Socio-demographic factors (age, ethnicity, Townsend deprivation index, professional qualifications and gross income), month of recruitment, smoking status, height, body weight, sedentary behaviour, dietary intake (total energy intake, carbohydrates, fats, alcohol, red meat, processed meat, oily fish and fruit and vegetables), diabetes, hypertension, and CVD medication were treated as potential confounders. Participants who had already been given a medical diagnosis of neurological-related diseases, depression, cancer, chronic pain, inflammatory diseases, alcohol or drug abuse and diseases or conditions that could influence dietary intake, grip strength or body composition were excluded from the analysis (n=64,231). Similarly, those with implausible total energy intake (n=12,189; e.g. extremely high) were excluded from the analysis (**Supplementary Table S1 and Figure S1**). Therefore, out of 211,047 participants with dietary, handgrip strength and fat-free mass data available 146,816 participants with full data available and who were free of comorbities listed above were included in the analyses.

Study procedures

Dietary information was collected via the Oxford WebQ; a web-based 24 h recall questionnaire which was developed specifically for use in large population studies and has been validated against an interviewer-administered 24 h recall questionnaire (25). The Oxford WebQ derives energy intake (total and from specific macronutrients) from the information recorded in McCance and Widdowson's The Composition of Food, 5th edition (26). For participants who completed more than one online dietary questionnaire (n=126,878), mean values were calculated from all of the information provided. Implausibly low or high energy intakes were defined as less than 1.1 times basal metabolic rate (calculated according to Henry equation (27)) (1.1*BMR)), and greater than 2.5 times basal metabolic rate respectively; the latter being the upper limit of sustained energy expenditure defined by the Scientific Advisory Committee for Nutrition (28).

Height was measured to the nearest centimetre (cm) using a Seca 202 height measure, and a Tanita BC-418 body composition analyser was used to measure weight to the nearest 0.1 kg and FFM to the nearest 1 g, by bio-impedance. Grip strength was assessed using a Jamar J00105 hydraulic hand dynamometer and the mean of the right hand and left hand values, expressed as kg/kg body mass, was used in the analyses. The duration of light, moderate and vigorous physical activity undertaken over the previous 24 h was self-reported using the International Physical Activity Questionnaire (IPAQ), as described previously (29). Participants were asked three questions: In a typical day, how many hours do you spend watching TV/doing PC screening/driving?, and the combined figure was used as a proxy for overall sedentary behaviour (29).

Ethnicity was self-reported and categorized into: White, South Asian, Black, Chinese, other and mixed ethnic background. Smoking status was self-reported and classified as: never, former and current. Townsend deprivation index, professional qualification and gross income were used as proxies of socio-economic status. Townsend deprivation index was derived from postcode of residence using the Townsend score (30), which generates a deprivation scores based on four census variables; unemployment, non-car ownership, non-house ownership and household overcrowding. This was calculated before participants joined the UK Biobank and was based on the preceding national census data, with each participant assigned a score corresponding to the postcode of their home dwelling. Professional qualifications were self-reported and coded as an ordinal variable; participants were asked, “Which of the following qualifications do you have? (You can select more than one),” with the options college or university degree, A levels or equivalent, O levels or GCSEs or equivalent, CSEs, NVQ/HND/HNC, professional qualifications (i.e., nursing or teaching). A categorical income variable was generated from self-reported income data; participants reported their annual household income of <£18 000 (€23 600; $25 800), £18 000 to £30 999, £31 000 to £51 999, £52 000 to £100 000, and >£100 000. Medical history (as shown in Supplementary Table 1) was collected from the baseline assessment questionnaire. Further details of these measurements can be found in the UK Biobank online protocol (http://www.ukbiobank.ac.uk). This study was performed under generic ethical approval obtained by UK Biobank from the NHS National Research Ethics Service (approval letter ref 11/NW/0382, dated 17 June 2011).

Statistical analyses

Fat-free mass and handgrip strength were our outcome variables of interest, both were treated as continuous variables. To first explore an association between protein intake and FFM and grip strength multivariable linear regression analyses were performed, protein intake was first fitted into the model as a continuous variable and changes in the outcomes of interest were estimated by 0.5 g/kg/day protein intake increments. To explore a potential non-linear dose-repose relationship between protein intake and outcomes we used visual inspection of the associations between the outcomes and protein intake in a continuous manner. In addition, likelihood ratio tests were performed to test the departure from linearity, however no evidence of non-linear association were found therefore linear regression were used. To investigate the association of levels of protein intake and the outcomes of interest protein was categorised into <0.8 g/kg/day, 0.8-1.2 g/kg/day, 1.2-1.6 g/kg/day, 1.6-2.0 g/kg/day and >2.0 g/kg/day. Associations of protein intake (continuous or categorical variable) with FFM and grip strength were investigated using multivariable linear regression analyses. The results are reported as fully-adjusted means and their 95% confidence intervals (CI), or as adjusted beta coefficients and 95% CI, as appropriate. We then investigated whether the associations of protein intake with FFM and grip strength differed by sex by performing a 2-way interaction analyses and fitting a protein*sex interaction term into our model. Finally, to investigate whether the association of protein intake with FFM and grip strength differed by age we added a protein*age interaction term into our models.

All analyses were adjusted for month of assessment, ethnicity, socioeconomic status (three markers of deprivation were used including Townsend deprivation index, professional qualification and gross income), smoking, total physical activity, discretionary sedentary time, height, body weight, dietary intake (total energy intake, carbohydrate intake, total fat intake, alcohol intake, oily fish, red meat, processed meat, fruit and vegetables), and comorbidities (diabetes, hypertension, medication for CVD and CVD illness). All analyses were performed in STATA MP 14 (College Station, Texas, USA).

Results

The baseline characteristics of the participants included and excluded in the study are presented in **Supplementary Tables 2 and 3.** There were no major differences between those included and excluded, although the individuals excluded did have slightly higher prevalence of current smokers, diabetes, CVD, high blood pressure history and were slightly heavier and had lower grip strength. The baseline characteristics of included participants by categories of reported protein intake **Tables 1 and 2**. Overall, 25,020 (17.0%) of participants reported consuming <0.8 g/kg/day of protein, 70,559 (48.1%) reported consuming 0.8-1.2 g/kg/day, 39,862 (27.2%) reported consuming 1.2-1.6 g/kg/day, 9,487 (6.5%) reported consuming 1.6-2.0 g/kg/day and 1,888 (1.3%) reported consuming >2.0 g/kg/day

As demonstrated in **Figure 1** both FFM and grip strength demonstrated a linear and positive association with reported protein intake categories up to >2.0 g/kg/day, and this was not modified by age or sex **(Supplementary Tables 4 and 5)**. When treated as a continuous variable a 0.5 g/kg/day higher reported protein intake was associated with greater FFM in both men (β-coefficient 5.1% [95% CI: 5.0; 5.2], p<0.0001) and women (β-coefficient 7.7% [95%CI: 7.7; 7.8], p<0.0001). It was also associated with grip strength in men (β-coefficient 0.076 kg/kg [95% CI: 0.074; 0.078], p<0.0001) and women (β-coefficient 0.074 kg/kg [95% CI: 0.073; 0.076], p<0.0001).

Age was negatively associated with both FFM and grip strength. The β-coefficients, per 5 year increase in age, for the association between FFM (expressed as a %) and age were -0.192 (95% CI -0.224; -0.160, p<0.0001) in women and -0.307 (95% CI -0.334; -0.279, p<0.0001) in women. The β-coefficients, per 5 year increase in age, for the association with grip strength (kg/kg) were -0.012 (95% CI -0.012; -0.011, p<0.0001] in men and -0.014 (95% CI -0.015; -0.014, p<0.0001) in women. Interestingly, the association of FFM and grip strength with age was similar regardless of reported protein intake, with no significant (p=0.673) protein intake*age interactions observed **(Supplementary Figures 2 and 3).**

Discussion

The main finding of the current study is that FFM and grip strength were higher in those with higher reported protein intakes, with FFM and grip strength highest at reported intakes of >2.0 g/kg/day, irrespective of main confounding factors including age and sex.

The importance of dietary protein intake in relation to the maintenance of muscle mass and function stems from the role of amino acids as regulators of muscle protein synthesis (31). Indeed it has been demonstrated in many studies that an increased availability of amino acids stimulates muscle protein synthesis (32–34). For this reason, dietary protein intake is thought to be of key importance in optimising muscle mass and function. The current data support a positive association between reported dietary protein intake and FFM/strength. Previous data in this area are conflicting (19–24) with studies having had relatively low participant numbers (237 – 2,675 participants) primarily of older age. Due to these limitations no previous studies have been able to investigate associations of FFM/grip strength with defined protein intake categories across a broad age range (40-69 years). The current data demonstrate a positive association between reported protein intake and FFM/grip strength, with both FFM and grip strength highest at reported protein intakes of >2.0 g/kg/day. It is important to note at this point that the positive associations we have observed cannot be extrapolated beyond the range of the intakes reported, and protein intakes higher than evaluated here may not necessarily incur great benefits for FFM/grip strength and may have negative health effects.

These data indicate that the current recommendations for protein intake of 0.8 g/kg/day (12,13) may be too low for the maintenance of FFM and strength. This is not surprising as recommendations are limited to maintaining nitrogen balance, thus avoiding deficiency, and not to maximise muscle mass and strength. It has been proposed, previously, that for healthy older people dietary protein intake should be at least 1.0-1.2 g/kg/day (15,16). Our data suggests that both the current and suggested recommendations may be too low for those in the age range, 40-69 years, and not just older people.

In the current analyses the association between protein intake and FFM/grip strength was broadly similar regardless of participant age. In other words, across the age range of our participants our data do not support the need for differential protein recommendations. This may be surprising as previous data have suggested that older people have an “anabolic resistance” to protein ingestion and so would require greater dietary protein, compared to younger people, to optimise muscle mass and function (17,18,35). However, these previous comparisons have been made between young (~20 years) and older (~70-80 years) participants, whilst the current study has a narrower age range. This means that either the so called “anabolic resistance” to protein is already present at middle age, which is possible as this is the age at which muscle mass/function begins to decline (5), or that a greater protein requirement is only required in people older than the current participants (i.e. 70 years and older). Further work is needed to clarify such speculation.

The current data have clear public health implications. Our data indicate that protein intake recommendations should potentially be similar and higher across the 40-69 year age range and that a large proportion of the population have protein intakes which are sub-optimal for the maintenance of FFM and grip strength. Highlighting this, the current data demonstrate that in the UK biobank cohort whilst 83% of participants reported protein intakes of at least 0.8 g/kg/day recommendations only 6.5% of participants reported protein intakes 1.6-2.0 g/kg/day and 1.3% reporting intakes >2.0 g/kg/day. Interventions to increase dietary protein intake may, therefore, help optimise FFM and muscle function. Clearly the causality of these associations remains to be tested in appropriated designed trials. Furthermore, whilst, due to previous data highlighting the importance of dietary protein for muscle anabolic processes (36), the focus of the current paper was the association of reported protein intake and FFM/grip strength further work should investigate whether this relationship is altered by variations in the consumption of other macronutrients.

Increasing FFM and muscle function may be of benefit as previous research has demonstrated that muscle tissue has an important role in health and disease (37). Muscle has not only a functional role, in locomotion, but also metabolic roles as the primary protein and glucose storage site in the body (38–40). As well as determining the causality in these associations further work is required to confirm the relationship between protein intake and health outcomes, as high protein (most strongly when combined with low carbohydrate) intake has been associated with higher all cause, cancer and cardiovascular disease (CVD) mortality (41,42). This area is, however, not without controversy, as other studies have found no clear association between high-protein (when combined with low-carbohydrate) intake and mortality (43). The inconsistent findings may reflect the relatively small sample size of some studies, as well as methodological inadequacy for estimating long-term dietary exposures and this is an area where further work is needed.

Strengths and limitations

UK Biobank aimed to be representative of the general population in terms of age, sex, ethnicity and socioeconomic status but is unrepresentative in terms of lifestyle, with participants less likely to be obese and have lower disease frequency – indicative of a “healthy volunteer” selection bias (44,45). In the current study such bias may be amplified as only ~36% of the UK Biobank participants were included in this analysis. Therefore, caution should be heeded in generalizing summary statistics to the general population. This does not detract from the ability to generalize estimates of the magnitude of associations. Our study benefited from a very large number of participants, recruited from the general population, across the whole of the UK. We had sufficient power to undertake analyses by age categories. The cross-sectional aspects of this study do not allow us to demonstrate causality in the observed associations. This highlights the need for robust trials to investigate the causality of these associations, which should be feasible in shorter term for FFM and muscle strength outcomes. The greatest sources of uncertainty for all nutritional epidemiology, including the present analyses lie in the estimation of long-term exposure to food and drinks intakes, and then from the application of standard food composition tables to quantify protein consumption (46). All methods of dietary assessment can incur extensive errors, and biases which are diminished, but not eliminated, by studying large numbers (46,47). Dietary intake was self-reported outside the clinic, which may encourage more truthful reporting, and was collected using a 24-h recall questionnaire which has been shown to produce more accurate results than a food frequency questionnaire (the usual approach adopted in large-scale studies) (48). Accuracy was further improved by administering the questionnaire on four occasions over the course of a year and deriving mean values. In addition, online administration of the questionnaires is expected to minimize any reporting bias due to social desirability. Any regression dilution bias due to errors in protein intake measurement would bias the association towards the null and so the strength association we have observed may be an underestimation.

In conclusion our findings demonstrated (within the range of reported protein intakes evaluated) that in healthy people aged 40-69 higher reported dietary protein intake, up to >2.0 g/kg/day, is associated with higher FFM and grip strength. An appropriately designed randomised controlled trial is required to determine whether this association is causal but our data do suggest that current dietary protein recommendations of 0.8 g/kg/day may be too low.

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Assembly Government and the British Heart Foundation. The authors have no conflicts of interest to declare.

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Figure legends

Figure 1 Handgrip strength and fat-free mass by protein intake and sex

Data presented as adjusted mean and their 95%CI. Handgrip strength is expressed as kg of grip strength divided by body weight (kg) and fat-free mass is presented as % of total body weight. Analyses were adjusted for month of assessment, age, ethnicity, socioeconomic status (Townsend deprivation index, professional qualification and gross income), smoking, total physical activity, discretionary sedentary time, height, dietary intake (total energy intake, carbohydrate intake, total fat intake, alcohol intake, oily fish, red meat, processed meat, fruit and vegetables), and comorbidities (diabetes, hypertension, medication for CVD and CVD illness).

Figure 1

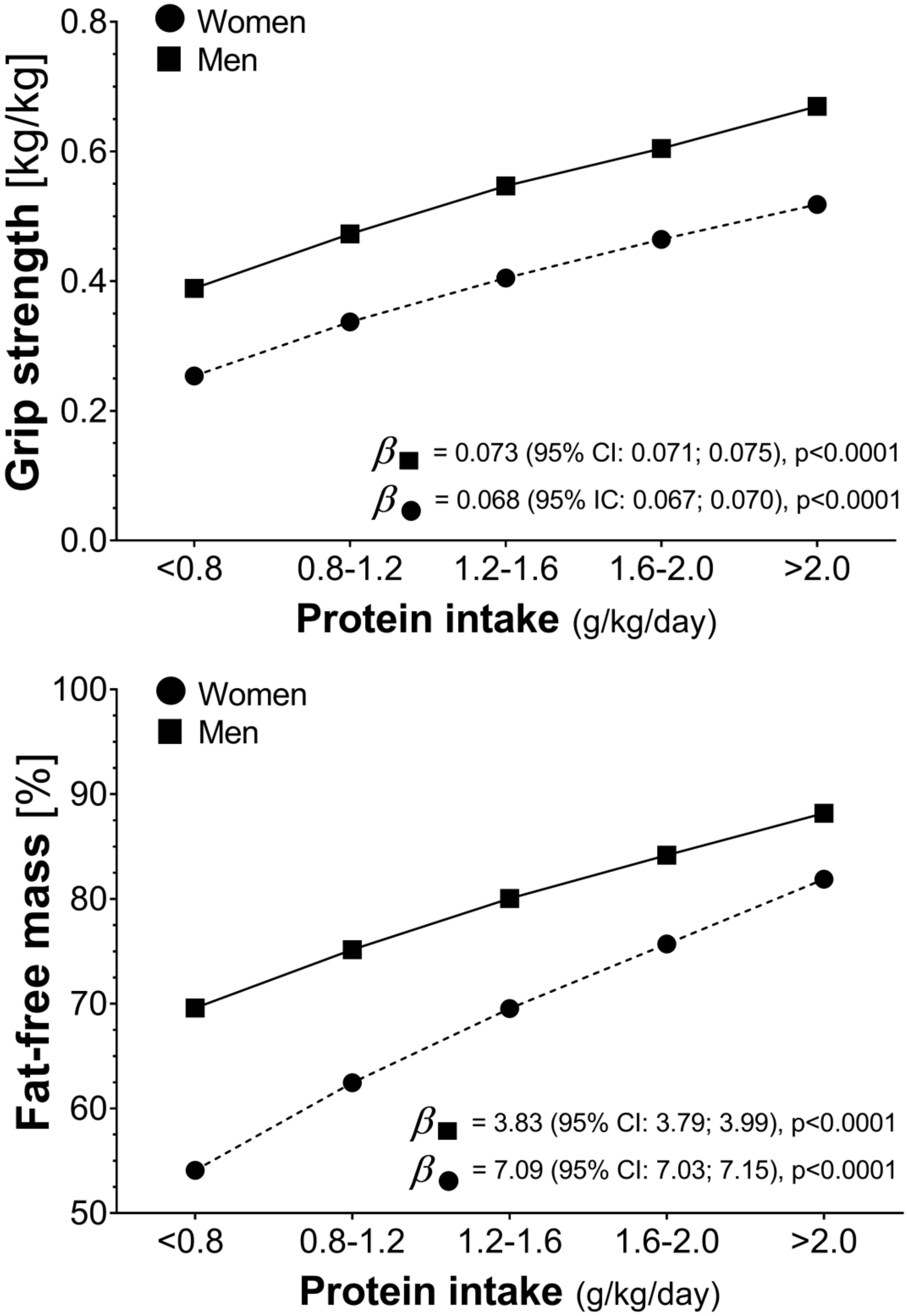
Women
Men
Grip strength [kg/kg]
0.8
0.6
0.4
0.2
0.0
β■ = 0.073 (95% CI: 0.071; 0.075), p<0.0001
β● = 0.068 (95% IC: 0.067; 0.070), p<0.0001
<0.8
0.8-1.2
1.2-1.6
1.6-2.0
>2.0
Protein intake (g/kg/day)
Women
Men
Fat-free mass [%]
100
90
80
70
60
50
β■ = 3.83 (95% CI: 3.79; 3.99), p<0.0001
β● = 7.09 (95% CI: 7.03; 7.15), p<0.0001
<0.8
0.8-1.2
1.2-1.6
1.6-2.0
>2.0
Protein intake (g/kg/day)

Table 1. Baseline characteristics of women from the UK Biobank study by protein intake categories.

	Categories of protein intake (grams of protein per kg body mass per day (g/kg/day))				
	<0.8 g/kg/day	**0.8-1.2 g/kg/day**	**1.2-1.6 g/kg/day**	**1.6-2.0 g/kg/day**	**>2.0 g/kg/day**
Socio-demographics					
Total n	12,251	36,369	24,419	6,272	1,190
Age (years), mean (SD)	54.3 (7.85)	55.0 (7.78)	55.0 (7.80)	54.4 (8.08)	53.3 (8.22)
Age categories					
<45 years	1,675 (16.5)	4,399 (43.3)	2,965 (29.2)	898 (8.8)	230 (2.2)
45-50 years	2,123 (16.4)	5,626 (43.4)	3,904 (30.1)	1,095 (8.4)	226 (1.7)
51-55 years	2,357 (16.0)	6,689 (45.6)	4,353 (29.6)	1,102 (7.5)	192 (1.3)
56-60 years	2,288 (14.7)	7,140 (45.8)	4,841 (31.0)	1,128 (7.2)	210 (1.3)
61-65 years	2,461 (14.0)	8,138 (46.2)	5,521 (31.3)	1,293 (7.3)	206 (1.2)
>65 years	1,347 (14.3)	4,377 (46.4)	2,835 (30.0)	756 (8.0)	126 (1.3)
Deprivation index, mean (SD)	-1.19 (2.98)	-1.59 (2.83)	-1.81 (2.72)	-1.74 (2.79)	-1.52 (2.87)
Deprivation					
1 (Least Deprived)	2,742 (12.8)	9,571 (44.7)	6,997 (32.7)	1,789 (8.4)	316 (1.4)
2	3,052 (14.5)	9,478 (45.0)	6,602 (31.3)	1,656 (7.9)	291 (1.3)
3	3,247 (15.5)	9,591 (45.6)	6,263 (29.8)	1,581 (7.5)	335 (1.6)
4 (Most Deprived)	3,210 (18.9)	7,729 (45.5)	4,557 (26.8)	1,246 (7.3)	248 (1.5)
Ethnicity, n (%)					
White	11,438 (14.9)	34,975 (45.5)	23,437 (30.5)	5,025 (7.7)	1,047 (1.4)
Mixed background	94 (16.3)	236 (41.0)	181 (31.4)	50 (8.7)	15 (2.6)
South Asian	250 (26.0)	355 (36.9)	237 (24.6)	94 (9.7)	27 (2.8)
Black	311 (30.2)	406 (39.4)	213 (20.7)	61 (5.9)	39 (3.8)
Chinese	20 (6.9)	76 (26.0)	104 (35.6)	52 (17.8)	40 (13.7)
Other	102 (16.0)	246 (38.6)	197 (30.9)	73 (11.4)	20 (3.1)
Smoking status, n (%)					
Never	7,132 (14.3)	22,116 (44.4)	15,664 (31.5)	4,095 (8.2)	794 (1.6)
Previous	4,075 (16.0)	11,936 (46.9)	7,321 (28.8)	1,795 (7.1)	304 (1.2)
Current	1,044 (19.8)	2,317 (44.0)	1,434 (27.2)	382 (7.3)	92 (1.7)
Obesity-related markers					
Height (meters), mean (SD)	1.64 (0.06)	1.64 (0.06)	1.63 (0.06)	1.62 (0.06)	1.61 (0.07)
Body weight (kg), mean (SD)	79.5 (16.3)	71.8 (12.2)	65.1 (8.7)	60.9 (8.7)	58.8 (9.7)
BMI, mean (SD)	29.6 (5.95)	26.8 (4.49)	24.5 (3.44)	23.2 (3.15)	22.7 (3.54)

BMI Categories, n (%)					
Under weight (<18.5 kg.m^{-2})	21 (3.3)	119 (18.5)	244 (37.9)	178 (27.6)	82 (12.7)
Normal weight (18.5-24.9 kg.m^{-2})	2,906 (7.8)	14,108 (37.6)	14,963 (39.9)	4,637 (13.4)	879 (2.3)
Overweight (25.0 to 29.9 kg.m^{-2})	4,368 (15.6)	14,680 (52.4)	7,548 (27.0)	1,232 (4.4)	183 (0.6)
Obese (≥30.0 kg.m^{-2})	4,956 (34.5)	7,462 (52.0)	1,664 (11.6)	225 (1.6)	46 (0.3)
Waist Circumference (cm)	89.9 (13.7)	84.0 (11.2)	78.7 (9.16)	75.4 (8.48)	74.1 (9.02)
Central Obesity, n (%)	6,494 (27.7)	12,313 (52.6)	3,944 (16.9)	562 (2.4)	100 (0.4)
% Body fat, mean (SD)	39.5 (6.79)	36.5 (6.27)	33.2 (5.97)	30.6 (6.17)	29.3 (6.91)
Body fat-free mass (%), mean (SD)	60.5 (6.79)	63.5 (6.27)	66.8 (5.97)	69.4 (6.17)	70.7 (6.91)
Body fat-free mass (kg), mean (SD)	47.2 (5.67)	45.0 (4.57)	43.1 (3.90)	41.9 (3.81)	41.1 (4.05)
Fitness and Physical activity					
Total PA (MET.h^{-1}.week^{-1}), mean (SD)	40.2 (49.4)	41.4 (47.5)	43.7 (48.7)	47.0 (49.2)	48.9 (54.1)
Physically active individuals n,(%)	6,094 (13.5)	19,990 (44.3)	14,426 (32.0)	3,906 (8.6)	728 (1.6)
Grip Strength (kg), mean (SD)	24.6 (6.08)	24.5 (5.88)	24.3 (5.76)	24.1 (5.78)	23.5 (5.94)
Grip strength (kg.kg body mass^{-1}), mean (SD)	0.32 (0.10)	0.35 (0.10)	0.38 (0.10)	0.40 (0.10)	0.41 (0.11)
TV viewing (h.day^{-1}), mean (SD)	2.60 (1.50)	2.45 (1.44)	2.31 (1.42)	2.21 (1.40)	2.23 (1.48)
Total Sedentary behaviour (h.day^{-1}) , mean (SD)	4.92 (2.19)	4.67 (1.99)	4.45 (1.90)	4.32 (1.89)	4.31 (2.04)
Dietary intakes					
Total energy intake (kcal.day^{-1}), mean (SD)	1,612 (389)	1,906 (410)	2,177 (430)	2,429 (446)	2,600 (428)
Carbohydrate intake (% of TE), mean (SD)	50.2 (8.79)	47.8 (7.56)	46.5 (7.40)	45.4 (7.78)	42.8 (9.26)
Fat intake (% of TE), mean (SD)	31.7 (7.52)	32.2 (6.56)	32.8 (6.27)	33.1 (6.42)	33.2 (7.03)
Protein intake (% of TE), mean (SD)	13.2 (3.18)	15.6 (3.20)	16.7 (3.24)	17.9 (3.50)	20.6 (4.56)
Alcohol intake (% of TE), mean (SD)	4.78 (6.97)	4.49 (5.65)	4.04 (4.82)	3.56 (4.45)	3.35 (4.95)
Alcohol frequency (times.week^{-1}), mean (SD)	2.79 (1.54)	3.08 (1.47)	3.17 (1.44)	3.09 (1.48)	2.87 (1.56)
Fruit and vegetable intake (g.day^{-1}), mean (SD)	343.1 (196.1)	353.3 (178.2)	356.7 (177.5)	367.2 (189.8)	378.0 (206.0)
Processed meat intake (portion.week^{-1}), mean (SD)	1.41 (1.01)	1.53 (0.99)	1.64 (0.99)	1.66 (0.99)	1.68 (1.06)
Red meat (portion.week^{-1}), mean (SD)	1.55 (1.24)	1.73 (1.25)	1.88 (1.31)	1.94 (1.33)	2.00 (1.53)
Oily fish (portion.week^{-1}), mean (SD)	0.99 (0.98)	1.09 (0.98)	1.16 (1.00)	1.24 (1.04)	1.31 (1.12)
Health status, n (%)					
Diabetes history	539 (26.0)	939 (45.3)	437 (21.1)	126 (6.1)	31 (1.5)
CVDs history	3,224 (19.8)	7,607 (46.6)	4,328 (26.5)	981 (6.0)	175 (1.1)
High blood pressure history	2,873 (19.5)	6,889 (46.7)	3,945 (26.7)	900 (6.1)	149 (1.0)

BMI body mass index; PA physical activity; MET basal metabolic-equivalent; TE total energy intake. SD standard deviation; n number

Table 2. Baseline characteristics of men from the UK Biobank study by protein intake categories.

	Categories of protein intake (grams of protein per kg body mass per day (g/kg/day))				
	<0.8 g/kg/day	**0.8-1.2 g/kg/day**	**1.2-1.6 g/kg/day**	**1.6-2.0 g/kg/day**	**>2.0 g/kg/day**
Socio-demographics					
Total n	12,769	34,190	15,443	3,215	698
Age (years), mean (SD)	56.0 (8.01)	56.4 (8.01)	55.8 (8.22)	54.9 (8.50)	52.9 (8.17)
Age categories					
<45 years	1,451 (19.0)	3,583 (46.9)	1,960 (25.6)	509 (6.7)	142 (1.8)
45-50 years	1,704 (19.0)	4,447 (49.5)	2,206 (24.5)	510 (5.7)	124 (1.3)
51-55 years	2,045 (20.2)	5,144 (50.9)	2,301 (22.8)	479 (4.7)	142 (1.4)
56-60 years	2,380 (19.5)	6,340 (52.0)	2,805 (23.0)	550 (4.5)	116 (1.0)
61-65 years	3,134 (19.5)	8,617 (53.5)	3,607 (22.4)	643 (4.0)	96 (0.6)
>65 years	2,055 (18.2)	6,059 (53.7)	2,564 (22.7)	524 (4.7)	78 (0.7)
Deprivation index, mean (SD)	-1.50 (2.91)	-1.79 (2.79)	-1.68 (2.85)	-1.30 (3.07)	-0.85 (3.28)
Deprivation					
1 (Least Deprived)	3,292 (17.8)	9,927 (53.7)	4,299 (23.3)	827 (4.4)	151 (0.8)
2	3,301 (19.0)	9,099 (52.3)	4,069 (23.4)	792 (4.5)	148 (0.8)
3	3,300 (19.6)	8,663 (51.4)	3,918 (23.3)	781 (4.6)	191 (1.1)
4 (Most Deprived)	2,876 (21.2)	6,501 (48.0)	3,157 (23.3)	815 (6.0)	208 (1.5)
Ethnicity, n (%)					
White	12,156 (19.1)	32,981 (52.0)	14,753 (23.2)	2,959 (4.7)	600 (1.0)
Mixed background	61 (19.4)	146 (46.4)	77 (24.4)	24 (7.6)	7 (2.2)
South Asian	263 (24.9)	446 (42.2)	235 (22.2)	84 (7.9)	30 (2.8)
Black	129 (20.1)	277 (43.2)	156 (24.3)	55 (8.6)	25 (3.8)
Chinese	16 (10.3)	47 (30.1)	47 (30.1)	34 (21.8)	12 (7.7)
Other	86 (20.1)	164 (38.4)	116 (27.2)	39 (9.1)	22 (5.2)
Smoking status, n (%)					
Never	6,292 (17.8)	18,081 (51.2)	8,726 (24.7)	1,825 (5.2)	392 (1.1)
Previous	5,190 (20.7)	13,220 (52.9)	5,377 (21.5)	1,029 (4.1)	210 (0.8)
Current	1,287 (21.6)	2,889 (48.4)	1,340 (22.4)	361 (6.0)	96 (1.6)
Obesity-related markers					
Height (meters), mean (SD)	1.77 (0.06)	1.77 (0.07)	1.76 (0.07)	1.75 (0.07)	1.74 (0.07)
Body weight (kg), mean (SD)	91.1 (14.4)	84.9 (11.9)	79.2 (11.1)	76.3 (11.3)	75.3 (11.7)
BMI, mean (SD)	29.0 (4.25)	27.2 (3.51)	25.7 (3.26)	25.0 (3.35)	25.0 (3.46)

BMI Categories, n (%)					
Under weight (<18.5 kg.m^{-2})	5 (3.9)	31 (24.4)	51 (40.2)	35 (27.6)	5 (3.9)
Normal weight (18.5-24.9 kg.m^{-2})	1,905 (9.4)	9,400 (46.2)	6,945 (34.1)	1,740 (8.5)	371 (1.8)
Overweight (25.0 to 29.9 kg.m^{-2})	6,396 (19.2)	18,440 (55.4	6,987 (21.0)	1,207 (3.6)	273 (0.8)
Obese (≥30.0 kg.m^{-2})	4,463 (35.6)	6,319 (50.5)	1,460 (11.7)	233 (1.9)	49 (0.3)
Waist Circumference (cm)	100.2 (11.4)	95.4 (9.71)	91.1 (9.24)	88.9 (9.48)	88.2 (9.65)
Central Obesity, n (%)	5,277 (33.5)	8,217 (52.1)	1,913 (12.1)	301 (1.9)	59 (0.4)
% Body fat, mean (SD)	26.7 (5.44)	24.5 (5.23)	22.3 (5.37)	21.2 (5.66)	20.9 (5.84)
Body fat-free mass (%), mean (SD)	73.3 (5.44)	75.5 (5.23)	77.7 (5.37)	78.8 (5.66)	79.1 (5.84)
Body fat-free mass (kg), mean (SD)	66.3 (7.74)	63.8 (7.01)	61.2 (6.74)	59.8 (7.08)	59.1 (7.36)
Fitness and Physical activity					
Total PA (MET.h^{-1}.week^{-1}), mean (SD)	44.3 (58.8)	45.1 (56.0)	50.4 (60.8)	58.3 (71.2)	71.7 (99.9)
Physically active individuals (%)	6,840 (17.5)	19,932 (51.0)	9,709 (24.9)	2,108 (5.4)	461 (1.2)
Grip Strength (kg), mean (SD)	40.3 (8.49)	40.2 (8.25)	39.8 (8.25)	39.1 (8.44)	39.4 (8.70)
Grip strength (kg.kg body mass^{-1}), mean (SD)	0.45 (0.11)	0.48 (0.11)	0.51 (0.11)	0.52 (0.12)	0.53 (0.12)
TV viewing (h.day^{-1}), mean (SD)	2.64 (1.51)	2.48 (1.45)	2.36 (1.45)	2.34 (1.47)	2.31 (1.47)
Total Sedentary behaviour (h.day^{-1}) , mean (SD)	5.67 (2.52)	5.34 (2.31)	5.13 (2.30)	5.11 (2.44)	5.12 (2.42)
Dietary intakes					
Total energy intake (kcal.day^{-1}), mean (SD)	1,955 (419)	2,303 (467)	2,718 (552)	3,090 (614)	3,328 (608)
Carbohydrate intake (% of TE), mean (SD)	48.7 (8.76)	46.6 (7.56)	45.5 (7.60)	44.1 (8.17)	41.1 (9.05)
Fat intake (% of TE), mean (SD)	30.6 (7.19)	31.8 (6.26)	32.8 (6.16)	33.5 (6.42)	33.7 (6.79)
Protein intake (% of TE), mean (SD)	12.7 (2.81)	15.0 (2.75)	16.2 (3.03)	17.7 (3.57)	20.8 (4.56)
Alcohol intake (% of TE), mean (SD)	8.05 (9.04)	6.54 (6.89)	5.50 (6.01)	4.80 (5.65)	4.37 (5.71)
Alcohol frequency (times.week^{-1}), mean (SD)	3.43 (1.41)	3.58 (1.33)	3.52 (1.38)	3.36 (1.43)	3.16 (1.46)
Fruit and vegetable intake (g.day^{-1}), mean (SD)	300.0 (191.2)	308.4 (182.4)	313.8 (179.2)	325.4 (202.1)	352.1 (253.2)
Processed meat intake (portion.week^{-1}), mean (SD)	2.01 (1.08)	2.13 (1.04)	2.24 (1.04)	2.25 (1.07)	2.28 (1.10)
Red meat (portion.week^{-1}), mean (SD)	1.87 (1.34)	2.02 (1.38)	2.13 (1.44)	2.24 (1.59)	2.47 (1.88)
Oily fish (portion.week^{-1}), mean (SD)	0.97 (0.97)	1.08 (1.01)	1.18 (1.08)	1.28 (1.18)	1.38 (1.29)
Health status, n (%)					
Diabetes history	911 (26.7)	1,669 (48.8)	651 (19.0)	147 (4.3)	41 (1.2)
CVDs history	4,470 (23.0)	10,263 (52.9)	3,814 (19.5)	720 (3.7)	169 (0.9)
High blood pressure history	3,554 (23.1)	8,127 (52.7)	3,034 (19.7)	573 (3.7)	134 (0.8)

BMI body mass index; PA physical activity; MET basal metabolic-equivalent; TE total energy intake. SD standard deviation; n number