The Relationship Between Urinary Total Polyphenols and the Frailty Phenotype in a Community-Dwelling Older Population: The InCHIANTI Study

Mireia Urpi-Sarda,[1,2] Cristina Andres-Lacueva,[1,2] Montserrat Rabassa,[1,2] Carmelinda Ruggiero,[3] Raul Zamora-Ros,[4] Stefania Bandinelli,[5] Luigi Ferrucci,[6] and Antonio Cherubini[7]

[1]Biomarkers and Nutrimetabolomic Lab., Nutrition and Food Science Department, XaRTA, INSA, Pharmacy School, University of Barcelona, Spain. [2]Ingenio-CONSOLIDER Program, FUN-C-FOOD, Barcelona, Spain. [3]Department of Medicine, Institute of Gerontology and Geriatrics, Perugia University, Italy [4]Biomarkers Group, Nutrition and Metabolism Section, International Agency for Research on Cancer (IARC), Lyon, France. 5Geriatric Unit, Azienda Sanitaria Firenze, Florence, Italy. [6]Intramural Program, National Institute on Aging, Baltimore, Maryland. [7]Geriatrics and Geriatric Emergency Room, IRCCS-INRCA, Ancona, Italy.

Address correspondence to Cristina Andres-Lacueva, PhD, Department of Nutrition and Food Science, Faculty of Pharmacy, University of Barcelona, 08028 Barcelona, Spain. Email: candres@ub.edu

Abstract

Background. Frailty, an age-related state of increased vulnerability, is associated with a higher risk of multiple adverse events. Studies have suggested that the quality of dietary intake may affect the development of frailty. We hypothesized that frailty in older subjects would be associated with dietary total polyphenols (DTP) intake and its biomarker, urinary total polyphenols (UTP).

Methods. The Invecchiare in Chianti (InCHIANTI) Study is a prospective cohort study set in the Chianti area (Italy). We used data at baseline from 811 participants aged 65 years and older. UTP was determined using the Folin–Ciocalteu assay after solid-phase extraction. DTP was estimated using a validated Food Frequency Questionnaire and our own polyphenol database. The frailty, prefrailty, and nonfrailty states were defined according to the Fried and colleagues' criteria. Multinomial logistic regressions adjusted for potential confounders were used to assess the relationship between polyphenols and frailty.

Results. Both DTP and UTP concentrations progressively decrease from nonfrail to frail participants. Participants in the highest UTP tertile compared to those in the lowest tertile were significantly less likely to be both frail (odds ratio [OR] = 0.36 [0.14–0.88], p = .025) and prefrail (OR = 0.64 [0.42–0.98], p = .038). Exhaustion and slowness were the only individual frailty criteria significantly associated with UTP tertiles. No significant association was observed between frailty and DTP, after adjustment for covariates.

Conclusions. High concentrations of UTP were associated with lower prevalence of frailty and prefrailty in an older community-dwelling population. A polyphenol-rich diet may protect against frailty in older persons. Our findings should be confirmed in longitudinal studies.

Key Words: Frailty—Nutrition—Biomarkers—Polyphenols —Elderly people—InCHIANTI.

Polyphenols are phytochemical compounds present in plantbased foods, such as fruits and vegetables and derived foods (1). Epidemiological studies reported an inverse association between diets rich in polyphenols and risk of cardiovascular diseases, some cancers, diabetes, premature death (2‑4), and

functional limitations and disabilities (5). Many scientists believe that the effects of polyphenols on health depend on the amount of polyphenols consumed, but also on their bioavailability (including inter- and intravariability) and bioactivity, which cannot be assessed by simply evaluating dietary intake of total polyphenols (DTP) (6,7). In this respect, urinary total polyphenols (UTP) take into account all the above-mentioned factors and might be a better measure of exposure to polyphenols than dietary assessment (6). Frailty is a condition characterized by an extreme vulnerability due to a decline across multiple physiological systems that increases the risk of adverse health events, such as disability, falls, dependency, need for long-term care, and death (8). Several instruments exist for measuring the level of frailty as reviewed by de Vries and coworkers (9). The frailty phenotype of Fried and coworkers is probably the most widely used definition of frailty. According to Fried and coworkers, frailty is defined as the presence of at least three of the following criteria: reduced physical activity, muscle weakness, slow walking speed, fatigue or poor endurance, and unintentional weight loss (8). Epidemiological studies have found that high plasma concentrations of antioxidants, such as carotenoids, vitamin E, and D, are associated with lower prevalence of frailty and related conditions (10–13). Indeed, low serum carotenoid concentrations have been considered a risk factor for accelerated muscle strength decline, and high levels of carotenoids were associated with reduced risk of walking disabilities and sarcopenia (12,14). The relationship between dietary intake of polyphenols and frailty has been studied very little. A recent cross-sectional study in the National Health and Nutrition Examination Survey (NHANES) showed that urinary concentrations of an isoflavone metabolite were inversely associated with frailty in women (15). In addition, in the Invecchiare in Chianti (InCHIANTI) study, a greater adherence to a Mediterranean diet, which is a polyphenol-rich diet, was associated with a lower risk of developing frailty (16).

In the current study, we aimed at investigating whether polyphenol exposure, measured either as UTP or as DTP, was associated with the frailty syndrome and its individual criteria in an older community-dwelling population.

Methods

Study Population

The InCHIANTI is a prospective population-based study conducted in two towns (Greve in Chianti and Bagno a Ripoli) in the Chianti geographic area (Tuscany, Italy) aimed at evaluating factors affecting mobility in an older community-dwelling population. A detailed description of the study design, sampling procedure, and data collection method has been previously published elsewhere (17). Baseline data collection started in September 1998 and was completed in March 2000. The Italian National Research Council of Aging ethical committee examined and approved the study protocol, and all participants gave informed consent. The study randomly sampled 1,260 men and women, of whom 1,155 (91.6%) were enrolled. Of these, 811 (70.2%) participants had 24-hour urine measures and complete data for the evaluation of frailty. At baseline, data collection included: a home interview followed by a full medical and functional evaluation performed by trained personnel, and 24-hour urine collection and blood drawing that were coded and stored at −80°C until analysis.

Determination of Total Polyphenols in Urine Samples

The analysis of total polyphenols in baseline urine samples was performed using the Folin‐Ciocalteu reagent (Sigma-Aldrich, St. Louis, MO) after solid-phase extraction using Oasis MAX 96-well plates (Waters, Milford, MA) according to our previous methodology (6). Results were expressed as UTP concentrations in mg gallic acid equivalents (Sigma-Aldrich) in 24-hour urine.

Frailty Syndrome

Frailty was defined according to the following criteria proposed by Fried and colleagues (8): (a) shrinking or weight loss, (b) exhaustion or lack of energy; (c) low physical activity or sedentariness, (d) weakness or poor muscle strength; (e) slowness or low walking speed (see Supplementary Data). Participants who had one or two of these criteria were considered prefrail, those with three to five criteria were considered frail, and participants with no criteria were considered not frail and were called nonfrail. Covariates Data on sociodemographics (age and gender) and lifestyle variables were collected by standardized questionnaires. Food intake (g/d) at baseline was estimated using the Italian

version of the food frequency questionnaire (FFQ) developed and validated in the European Prospective Study into Cancer and Nutrition (EPIC) study (18). DTP intake was estimated using a custom-made food composition database on polyphenols (7), based on the USDA and Phenol-Explorer databases. Individual polyphenols, expressed as aglycones, were summed to calculate DTP (7). A description of other covariates used in the study is included in the Supplementary Data.

Statistical Analysis

Variables with symmetric distribution were reported as means and *SD*. Variables with asymmetric distribution were summarized as medians and interquartile ranges. Categorical variables were summarized as number of participants and percentages. UTP and DTP concentrations were not normally distributed and were normalized using a Box-Cox transformation ($\alpha = .00001$; $\lambda = .25$) (6) and as a log2-transformation, respectively. Spearman's correlation test was used to evaluate the associations between UTP and fruits and vegetables consumption and DTP.

UTP and DTP were analyzed as tertiles according to the following cutoff points: 123.6 and 173.4 mg gallic acid equivalent/24 h and 509.2 and 645.2 mg/d, respectively. Differences between frail, prefrail, and nonfrail participants, as well as differences across UTP and DTP tertiles, were estimated using analysis of variance, Kruskal-Wallis or chi-square tests, as appropriate. The association between UTP and DTP as tertiles and as continuous variables and both prefrailty and frailty (using nonfrail as the reference group) was tested using a multivariate multinomial logistic regression in three additive models. The interaction between gender and UTP and DTP as tertiles and as continuous variables was evaluated by using the models with and without an interaction term. Additionally, logistic regression was used to associate each frailty criteria with UTP and DTP as tertiles and as continuous variables. *p* values less than .05 (two-tailed) were considered to be significant. The statistical analyses were performed using the SPSS package program version 18.0 (SPSS Inc., Chicago, IL).

Results

The study included a total of 811 participants, 44.9% of whom were men, with a mean age of 74.3 } 6.9 years. In the overall sample, the prevalence of prefrailty and frailty was 39.6% and 8.9%, respectively. In comparison to those excluded due to incomplete data (n = 344), those included in the study were significantly younger (mean } *SD*, 74.3 } 6.9 vs 78.0 } 8.5 years, $p < .001$), had lower rates of activities of daily living (ADL) (5.4% vs 20.9%) and instrumental ADL (21.9% vs 42.7%) disabilities (both, $p < .001$), and had a lower prevalence of frailty (8.9% vs 12.2%) and dementia (3.8% vs 14.8%) ($p < .05$). From the nonfrail to the frail group, participants were less physically active and had a higher prevalence of disability in more than one ADL and instrumental ADL as well as of physical performance impairment. In addition, from the nonfrail to the frail group, participants had lower DTP, lower plasma antioxidants such as α-tocopherol and lower urinary UTP (see Supplementary Table 1). The characteristics of study participants across UTP and DTP tertiles are displayed in Table 1. From the lowest to the highest UTP tertiles, participants had a higher Mediterranean Diet score and higher energy intake. The prevalence of participants with disability in more than one ADL and instrumental ADL, and with physical performance impairment, decreased with increasing UTP or DTP tertiles. While the prevalence of participants with frailty progressively decreased from the lowest to the highest UTP and DTP tertiles, prefrailty prevalence only decreased significantly through the UTP tertiles. The association between frailty and UTP or DTP concentrations through the tertiles and as continuous variables is shown in Table 2. Frailty was significantly associated with UTP levels, independent of age, gender, creatinine clearance and other factors such as body mass index, total energy intake, alcohol consumption, smoking habit, and activity level (Table 2, Model 1). This association was unchanged after further adjustment for inflammatory markers, that is, IL-6 and CRP (Table 2, Model 2), and for chronic diseases (Table 2, Model 3). Participants in the highest UTP tertile were 64% (p = .025) and 36% (p = .038) less likely to be frail and prefrail, respectively, than those in the lowest UTP tertile. In addition, there were significant inverse associations between frailty and prefrailty and UTP (odds ratio [OR] = 0.69 [0.54‑0.88], p = .003; OR = 0.85 [0.76‑0.96], p = .011, respectively). Frailty and prefrailty status were not associated with DTP tertiles or continuous DTP variable (Table 2). Moreover, when the interaction

between gender and UTP and DTP as tertiles or as continuous variables was evaluated, no statistically significant interactions were observed (data not shown). The association of UTP with individual frailty criteria is shown in Table 3. After adjustment for covariates, the participants in the highest UTP tertile were less likely to report exhaustion (OR = 0.42 [0.24‑0.72], p = .002) and to walk slowly (OR = 0.55 [0.33‑0.93], p = .026) than those in the lowest UTP tertile. Furthermore, those in the middle UTP tertile were also less likely to walk slowly (OR = 0.58 [0.35‑0.95], p = .029) than those in the lowest UTP tertile. In addition, there were significant inverse associations between UTP and exhaustion and slowness criteria (OR = 0.81 [0.70‑0.94], p = .005; OR = 0.86 [0.74‑0.99], p = .036, respectively). Although two of the five criteria (weight loss and low physical activity) showed no significant association with UTP tertiles, weakness showed a borderline significant association (p = .059) between participants in the highest and in the lowest UTP tertiles in the fully adjusted model (Table 3, Model 3). This association became significant when the lineal variable was considered (OR = 0.84 [0.73‑0.96], p = .01).

Discussion

This cross-sectional study shows that high concentrations of UTP were associated with lower prevalence of prefrailty and frailty in a community- dwelling older population. Indeed, participants in the highest UTP tertile were 36% and 64% less likely to be prefrail and frail, respectively, than those in the lowest UTP tertile. On the contrary, DTP was not significantly associated with frailty status. In a previous study, a significant association was observed between UTP and total mortality, while no significant association was found with DTP (7). To our knowledge, only one epidemiological study, the NHANES study, has investigated the association between polyphenol biomarkers and frailty. It showed an inverse association between the urinary concentration of *O*-desmethylangolensin, an isoflavone metabolite, and frailty in women (15). Moreover, two recent epidemiological studies showed inverse associations between a greater adherence to the Mediterranean diet and frailty risk (16,19). In addition, not consuming fruits and vegetables daily was associated with frailty and prefrailty in the participants of the Whitehall II prospective cohort study

(20). Finally, several observational studies, which evaluated other antioxidant micronutrients, reported that frailty was associated with low intakes of vitamins D, E, C, and folate, low concentrations of plasma carotenoids and α-tocopherol, and a low diet quality index (12,21–23). In the present study, a statistically significant inverse association was observed between UTP concentrations and some frailty criteria, particularly exhaustion and slowness, and, although with borderline significance, with the weakness criterion. To our knowledge, no epidemiological studies have evaluated these associations so far, although similar results were reported for other antioxidant micronutrients. Thus, both high consumption of carotenoid-rich foods and high plasma levels of carotenoids were associated with a decline in muscle strength, impaired lower extremity performance, and walking disability in older adults (12,14,24,25). In addition, low intake and low serum concentrations of vitamin E were associated with reduced physical performance, frailty, and disability (21,24,26). Moreover, a poor nutritional score determined as the sum of several vitamins, minerals, and proteins was associated with weakness and exhaustion, although this last association lost its significance after adjustment for energy intake (21). Taking into account the weight loss criterion, the number of older subjects reporting weight loss is low, due to the good health of participants at baseline, and therefore there could be insufficient power to test the association between this variable and UTP. Moreover, in a previous InCHIANTI study, authors suggested that weight loss may not be a sensitive proxy measure of inadequate diet or undernutrition (21).

The protective effect of polyphenols towards frailty might be accounted for by their anti-inflammatory and antioxidant activity (27–29). Greater adherence to the Mediterranean diet was associated with lower levels of inflammatory biomarkers, for example, IL-6 and CRP, and particularly those related to endothelial function (ICAM-1 and VCAM-1) (29,30). Moreover, it has been hypothesized that diets rich in antioxidants (polyphenols and some vitamins) could prevent sarcopenia, a major component of frailty (27). Antioxidant containing diets resulted in less oxidative stress, elevated activities of glutathione, and a reductive shift in the glutathione redox state in muscle homogenates and mitochondria in mice (31), as well as protected skeletal muscle from oxidative damage (32). The different results obtained from the dietary measure (DTP) and the urinary

biomarkers (UTP) are probably mainly due to the fact that the latter measure also takes into account the bioavailability of food polyphenols. After absorption and metabolism, compounds reach target tissues where they will deliver the biological activity. UTP is an unspecific polyphenol biomarker that is associated with the consumption of fruits and vegetables in our population ($\rho = .15$; $p < .001$) and in other populations such as the Spanish Predimed study (33). In addition, UTP is also associated with DTP ($\rho = .134$; $p < .001$) which was evaluated in depth in a previous study in the whole InCHIANTI cohort ($r = .211$, $p < .001$) (6). Although the use of FFQ by itself has limitations in the dietary assessment measure and in capturing data of food consumed, the FFQ used in this study has been validated for this specific population and in the EPIC Italian population (6). This FFQ has been used to correlate plasma carotenoids with the consumption of fruit and vegetables (6). Therefore, the main strength of this study was the use of a validated biomarker, UTP concentrations in 24-hour urine samples, to measure polyphenol exposure which is a more reliable and accurate measure of intake than DTP and takes into account the bioavailability of each type of polyphenol (6). A further advantage was that two of the criteria (slowness and weakness) for evaluating frailty were objectively measured. These criteria were those inversely associated with higher UTP concentrations. Finally, our models were built considering potential confounders of the relationship between frailty and polyphenol exposure. The main limitation of this study is the cross-sectional design, which did not allow a causal role of low UTP in increasing frailty risk to be established. We could not assess whether the lower excretion of total polyphenols, related to their consumption, represented the initial point of the pathway causing the frailty syndrome or whether the frailty syndrome led to less consumption of polyphenolrich foods. In addition to this point, it is impossible to differentiate which polyphenols are associated with frailty using this measure. Second, three of the criteria (exhaustion, shrinking, and sedentariness) for evaluating frailty were self-reported measures, and these may have been influenced by the memory of participants, especially in older subjects. Therefore, a potential misclassification in frailty status cannot be excluded. Despite this, assessment of frailty is standardized in geriatrics and the information was collected by trained personnel in a home interview in order to reduce memory biases. Third, the DTP intake may be underestimated, although the dietary

intake was optimized by using a validated Italian FFQ and a comprehensive database on polyphenols (6,7). In conclusion, the present study showed that UTP concentrations, a marker of polyphenol intake, were inversely associated with frailty and prefrailty status in a community-dwelling older population in Italy. Participants in the highest UTP tertile had lower risks of frailty and prefrailty by 64% and 36%, respectively, when compared with those in the lowest UTP tertile. Furthermore, when frailty was explored by individual criteria, participants in the highest UTP tertile had significantly lower risk of suffering exhaustion and slowness and a borderline significantly lower risk of weakness. Our data provide new evidence suggesting that polyphenol-rich diets may protect against the occurrence of frailty in older subjects.

Supplementary Material

Supplementary material can be found at: http://biomedgerontology. oxfordjournals.org/

Funding

The InCHIANTI study was supported by the Italian Ministry of Health and by the U.S. National Institute on Aging (contracts 263 MD 916413 and 263 MD 821336). Special thanks to the MAPFRE FOUNDATION grant in collaboration with the Bosch i Gimpera Foundation (FBG306776). This research was also supported by the Spanish National Grants from the Ministry of Economy and Competitiveness (MINECO) and co-founded by FEDER (Fondo Europeo de Desarrollo Regional): AGL2009-13906-C02-01, the CONSOLIDER INGENIO 2010 Programme, FUN-C-FOOD (CSD2007-063), as well as the PI13/01172 Project (Plan N de I+D+i 2013-2016) by ISCII-Subdireccion General de Evaluacion y Fomento de la Investigacion. We thank the award of 2014SGR1566 from the Generalitat de Catalunya’s Agency AGAUR.

Acknowledgments

M.U.-S. thanks the Ramon y Cajal program of the Spanish Ministry and Fondo Social Europeo (RYC-2011-09677). We also thank the participants for their collaboration in the study.

Conflict of Interest

No conflicts of interest are declared.

References

1. Perez-Jimenez J, Fezeu L, Touvier M, et al. Dietary intake of 337 polyphenols in French adults. *Am J Clin Nutr*. 2011;93:1220‑1228. doi:10.3945/ ajcn.110.007096

2. Crowe FL, Roddam AW, Key TJ, et al. Fruit and vegetable intake and mortality from ischaemic heart disease: results from the European Prospective Investigation into Cancer and Nutrition (EPIC)-Heart study. *Eur Heart J*. 2011;32:1235‑1243. doi:10.1093/eurheartj/ehq465

3. van Dam RM, Naidoo N, Landberg R. Dietary flavonoids and the development of type 2 diabetes and cardiovascular diseases: review of recent findings. *Curr Opin Lipidol*. 2013;24:25‑33. doi:10.1097/ MOL.0b013e32835bcdff

4. Geybels MS, Verhage BA, Arts IC, et al. Dietary flavonoid intake, black tea consumption, and risk of overall and advanced stage prostate cancer. *Am J Epidemiol*. 2013;177:1388‑1398. doi:10.1093/aje/kws419

5. Houston DK, Stevens J, Cai J, Haines PS. Dairy, fruit, and vegetable intakes and functional limitations and disability in a biracial cohort: the Atherosclerosis Risk in Communities Study. *Am J Clin Nutr*. 2005;81:515‑522.

6. Zamora-Ros R, Rabassa M, Cherubini A, et al. Comparison of 24-h volume and creatinine-corrected total urinary polyphenol as a biomarker of total dietary polyphenols in the Invecchiare InCHIANTI study. *Anal Chim Acta*. 2011;704:110‑115. doi:10.1016/j.aca.2011.07.035

7. Zamora-Ros R, Rabassa M, Cherubini A, et al. High concentrations of a urinary biomarker of polyphenol intake are associated with decreased mortality in older adults. *J Nutr*. 2013;143:1445‑1450. doi:10.3945/ jn.113.177121

8. Fried LP, Tangen CM, Walston J, et al. Frailty in older adults: evidence for a phenotype. *J Gerontol A Biol Sci Med Sci*. 2001;56:M146‑M156. doi:http://dx.doi.org/10.1093/gerona/56.3.M146

9. de Vries NM, Staal JB, van Ravensberg CD, et al. Outcome instruments to measure frailty: a systematic review. *Ageing Res Rev*. 2011;10:104‑114. doi:10.1016/j.arr.2010.09.001.

10. Hirani V, Naganathan V, Cumming RG, et al. Associations between frailty and serum 25-hydroxyvitamin D and 1,25-dihydroxyvitamin D concentrations in older Australian men: the Concord Health and Ageing in Men Project. *J Gerontol A Biol Sci Med Sci*. 2013;68:1112‑1121. doi:10.1093/ gerona/glt059

11. Semba RD, Bartali B, Zhou J, et al. Low serum micronutrient concentrations predict frailty among older women living in the community. *J Gerontol A Biol Sci Med Sci*. 2006;61:594‑599. doi:http://dx.doi.org/10.1093/ gerona/61.6.594

12. Lauretani F, Semba RD, Bandinelli S, et al. Low plasma carotenoids and skeletal muscle strength decline over 6 years. *J Gerontol A Biol Sci Med Sci*. 2008;63:376‑383. doi:http://dx.doi.org/10.1093/gerona/63.4.376

13. Ble A, Cherubini A, Volpato S, et al. Lower plasma vitamin E levels are associated with the frailty syndrome: the InCHIANTI study. *J Gerontol A Biol Sci Med Sci*. 2006;61:278‑283. doi:http://dx.doi.org/10.1093/ gerona/61.3.278

14. Semba RD, Lauretani F, Ferrucci L. Carotenoids as protection against sarcopenia in older adults. *Arch Biochem Biophys*. 2007;458:141‑145. doi:10.1016/j.abb.2006.11.025

15. Eichholzer M, Richard A, Walser-Domjan E, Linseisen J, Rohrmann S. Urinary phytoestrogen levels and frailty in older American women of the National Health and Nutrition Examination Survey (NHANES) 1999-2002: a cross-sectional study. *Ann Nutr Metab*. 2013;63:269‑276. doi:10.1159/000356453

296 16. Talegawkar SA, Bandinelli S, Bandeen-Roche K, et al. A higher adherence to a Mediterranean-
297 style diet is inversely associated with the development of frailty in community-dwelling elderly men
298 and women. *J Nutr*. 2012;142:2161‑2166. doi:10.3945/jn.112.165498

299 17. Ferrucci L, Bandinelli S, Benvenuti E, et al. Subsystems contributing to the decline in ability to
300 walk: bridging the gap between epidemiology and geriatric practice in the InCHIANTI study. *J Am*
301 *Geriatr Soc*. 2000;48:1618‑ 1625.

302 18. Pisani P, Faggiano F, Krogh V, et al. Relative validity and reproducibility of a food frequency
303 dietary questionnaire for use in the Italian EPIC centres. *Int J Epidemiol*. 1997;26(suppl 1):S152‑
304 S160. doi:http://dx.doi. org/10.1093/ije/26.suppl_1.S152

305 19. Bollwein J, Diekmann R, Kaiser MJ, et al. Dietary quality is related to frailty in community-
306 dwelling older adults. *J Gerontol A Biol Sci Med Sci*. 2013;68:483‑489. doi:10.1093/gerona/gls204

307 20. Bouillon K, Kivimaki M, Hamer M, et al. Diabetes risk factors, diabetes risk algorithms, and the
308 prediction of future frailty: the Whitehall II prospective cohort study. *J Am Med Dir Assoc*. 2013;
309 14:851.e1‑e6. doi:10.1016/j.jamda.2013.08.016

310 21. Bartali B, Frongillo EA, Bandinelli S, et al. Low nutrient intake is an essential component of
311 frailty in older persons. *J Gerontol A Biol Sci Med Sci*. 2006;61:589‑593.
312 doi:http://dx.doi.org/10.1093/gerona/61.6.589

313 22. Michelon E, Blaum C, Semba RD, et al. Vitamin and carotenoid status in older women:
314 associations with the frailty syndrome. *J Gerontol A Biol Sci Med Sci*. 2006;61:600‑607.
315 doi:http://dx.doi.org/10.1093/gerona/61.6.600

316 23. Shikany JM, Barrett-Connor E, Ensrud KE, et al. Macronutrients, diet quality, and frailty in older
317 men. *J Gerontol A Biol Sci Med Sci*. 2014;69: 695‑701. doi:http://dx.doi.org/10.1093/gerona/glt196

318 24. Cesari M, Pahor M, Bartali B, et al. Antioxidants and physical performance in elderly persons:
319 the Invecchiare in Chianti (InCHIANTI) study. *Am J Clin Nutr*. 2004;79:289‑294.

320 25. Lauretani F, Semba RD, Bandinelli S, et al. Carotenoids as protection against disability in older
321 persons. *Rejuvenation Res*. 2008;11:557‑563. doi:10.1089/rej.2007.0581

322 26. Bartali B, Frongillo EA, Guralnik JM, et al. Serum micronutrient concentrations and decline in
323 physical function among older persons. *JAMA*. 2008;299:308‑315. doi:10.1001/jama.299.3.308

324 27. Kim JS, Wilson JM, Lee SR. Dietary implications on mechanisms of sarcopenia: roles of protein,
325 amino acids and antioxidants. *J Nutr Biochem*. 2010;21:1‑13. doi:10.1016/j.jnutbio.2009.06.014

326 28. Kolehmainen M, Mykkanen O, Kirjavainen PV, et al. Bilberries reduce lowgrade inflammation
327 in individuals with features of metabolic syndrome. *Mol Nutr Food Res*. 2012;56:1501‑1510.
328 doi:10.1002/mnfr.201200195

329 29. Urpi-Sarda M, Casas R, Chiva-Blanch G, et al. Virgin olive oil and nuts as key foods of the
330 Mediterranean diet effects on inflammatory biomarkers related to atherosclerosis. *Pharmacol Res*.
331 2012;65:577‑583. doi:10.1016/j.phrs.2012.03.006

332 30. Panagiotakos DB, Dimakopoulou K, Katsouyanni K, et al. Mediterranean diet and inflammatory
333 response in myocardial infarction survivors. *Int J Epidemiol*. 2009;38:856‑866.
334 doi:10.1093/ije/dyp142

335 31. Rebrin I, Zicker S, Wedekind KJ, et al. Effect of antioxidant-enriched diets on glutathione redox
336 status in tissue homogenates and mitochondria of the senescence-accelerated mouse. *Free Radic Biol*
337 *Med*. 2005;39:549‑557. doi:10.1016/j.freeradbiomed.2005.04.008

338 32. Charles AL, Meyer A, Dal-Ros S, et al. Polyphenols prevent ageing-related impairment in skeletal
339 muscle mitochondrial function through decreased reactive oxygen species production. *Exp Physiol*.
340 2013;98:536‑545. doi:10.1113/expphysiol.2012.067496.

341 33. Medina-Remon A, Barrionuevo-Gonzalez A, Zamora-Ros R, et al. Rapid Folin-Ciocalteu method
342 using microtiter 96-well plate cartridges for solid phase extraction to assess urinary total phenolic
343 compounds, as a biomarker of total polyphenols intake. *Anal Chim Acta*. 2009;634:54‑60.
344 doi:10.1016/j.aca.2008.12.012

345

346

TABLES

Table 1. Baseline Characteristics of InCHIANTI Participants Belonging to the Tertiles of UTP and to the Tertiles of DTP

Characteristic	UTP (tertiles)			DTP (tertiles)		
	1 (n = 270) (<123.6 mg GAE/24 h)	2 (n = 271) (123.6–173.4 mg GAE/24 h)	3 (n = 270) (>173.4 mg GAE/24 h)	1 (n = 268) (<509.2 mg/d)	2 (n = 267) (509.2–645.2 mg/d)	3 (n = 268) (>645.2 mg/d)
Demographics						
Age, y, mean ± *SD*	76.2 ± 7.4	74.3 ± 6.8	72.5 ± 5.9‡	75.9 ± 7.4	74.3 ± 6.7	72.8 ± 6.2‡
Men, n (%)	99 (36.7)	124 (45.8)	141 (52.2)‡	83 (31.0)	124 (46.4)	154 (57.5)‡
Behavior-related variables						
Body mass index, kg/m², mean ± *SD*	27.0 ± 4.2	27.5 ± 3.9	28.0 ± 3.8*	27.4 ± 4.3	27.5 ± 3.8	27.7 ± 3.9
Smoking, packs/y, median (IQR)	0.0 (0.0–14.6)	0.0 (0.0–16.8)	0.0 (0.0–28.0)*	0.0 (0.0–7.4)	0.0 (0.0–20.4)	0.0 (0.0–19.3)†
Energy intake, kcal/d, mean ± *SD*	1826.5 ± 542.8	1911.0 ± 536.5	2013.0 ± 572.1‡	1601.1 ± 441.9	1902.0 ± 439.5	2247.7 ± 573.2‡
Mediterranean diet score, mean ± *SD*	4.0 ± 1.6	4.0 ± 1.6	4.4 ± 1.7*	3.3 ± 1.5	4.3 ± 1.5	4.9 ± 1.6‡
Alcohol intake, g/d, median (IQR)	5.7 (0.0–17.4)	6.6 (0.0–17.5)	7.9 (0.0–26.8)	1.9 (0.0–13.4)	7.6 (0.2–19.3)	13.4 (0.5–28.3)†
Activity level in the last year, h/wk, mean ± *SD*	3.2 ± 1.1	3.3 ± 1.0	3.3 ± 0.9*	3.1 ± 1.1	3.3 ± 0.9	3.4 ± 1.0‡
Disability in ≥ 1 ADL, n (%)	25 (9.3)	13 (4.8)	6 (2.2)†	19 (7.1)	14 (5.2)	7 (2.6)§
Disability in ≥ 1 IADL, n (%)	86 (31.9)	51 (18.8)	41 (15.2)†	80 (29.9)	60 (22.5)	33 (12.3)‡
Physical performance impairment, SPPB ≤ 9, n (%)	93 (36.0)	59 (23.1)	43 (16.5)‡	82 (32.5)	64 (24.5)	49 (18.8)†
DTP, mg/d, mean ± *SD*	570.1 ± 202.1	595.0 ± 200.7	620.4 ± 180.9*	407.4 ± 77.5	574.9 ± 38.9	803.2 ± 167.9†
Plasma parameters						
IL-6, pg/mL, median (IQR)	1.6 (0.8–2.7)	1.3 (0.9–2.1)	1.5 (0.9–2.0)	1.6 (1.0–2.7)	1.5 (0.9–2.2)	1.4 (0.8–1.9)*
CRP, μg/mL, median (IQR)	2.6 (1.3–5.8)	2.5 (1.3–5.6)	2.8 (1.4–5.5)	2.8 (1.5–5.9)	2.6 (1.4–5.8)	2.5 (1.2–5.1)
Total cholesterol, mg/dL, mean ± *SD*	219.7 ± 38.0	220.4 ± 40.3	217.4 ± 39.2	220.1 ± 39.9	221.7 ± 39.7	215.9 ± 37.6
Uric acid, mg/dL, mean ± *SD*	5.1 ± 1.3	5.2 ± 1.4	5.2 ± 1.3	5.1 ± 1.4	5.1 ± 1.2	5.3 ± 1.4*
α-Tocopherol, μmol/L, median (IQR)	29.0 (24.3–33.8)	29.7 (24.8–35.3)	30.5 (25.3–36.2)*	28.7 (24.8–34.7)	30.4 (25.1–36.1)	29.5 (24.4–34.8)
Creatinine clearance, mL/min, mean ± *SD*	59.1 ± 20.6	63.8 ± 18.0	68.6 ± 18.1‡	59.0 ± 19.0	64.7 ± 18.6	68.1 ± 19.3‡
Urinary parameters						
UTP, mg GAE/24 h, mean ± *SD*	94.0 ± 21.7	147.9 ± 14.7	229.5 ± 48.7‡	145.6 ± 61.2	158.5 ± 65.5	167.4 ± 64.6‡
Normalized UTP, mg GAE/24 h, mean ± *SD*	8.4 ± 0.8	9.9 ± 0.3	11.5 ± 0.8‡	9.7 ± 1.4	10.0 ± 1.5	10.2 ± 1.4‡
Frailty phenotype						
Frailty, n (%)	37 (24.5)	22 (13.6)	13 (7.3)†	30 (19.9)	24 (14.0)	24 (8.7)*
Prefrailty, n (%)	119 (51.1)	109 (43.8)	93 (36.2)†	117 (49.2)	95 (39.1)	107 (42.1)§
Diseases and conditions						
Dementia, n (%)	18 (6.7)	8 (3.0)	5 (1.9)†	16 (6.0)	9 (3.4)	5 (1.9)*
Diabetes, n (%)	27 (10.0)	32 (11.8)	45 (16.7)*	37 (13.8)	35 (13.1)	31 (11.6)
Hypertension, n (%)	131 (49.6)	134 (50.2)	117 (43.8)	137 (52.7)	118 (44.5)	123 (46.2)
Renal failure, n (%)	188 (71.5)	159 (60.9)	142 (54.0)‡	175 (68.6)	166 (63.4)	144 (54.3)†

Notes: Statistical comparisons are from analysis of variance, Kruskal–Wallis, or chi-square, as appropriate. ADL = activities of daily living; DTP = dietary total polyphenols; GAE = gallic acid equivalents; IADL = instrumental activities of daily living; IQR = interquartile range; SPPB = Short Physical Performance Battery; UTP = urinary total polyphenols.

*p < .05, †p < .01, ‡p < .001, §p = .06–.07.

Table 2. Multinomial Logistic Model Describing the Association Between Tertiles of UTP or DTP and Prefrailty and Frailty Status in the InCHIANTI Study

	Prefrail (1–2 characteristics)			Frail (3–5 characteristics)		
Models	*n*	OR (95% CI)	*p*	*n*	OR (95% CI)	*p*
UTP						
Model 1						
Tertiles						
Lowest	119	Reference		37	Reference	
Middle	109	0.82 (0.54–1.22)	.326	22	0.64 (0.30–1.37)	.248
Highest	93	0.65 (0.43–0.99)	.042	13	0.33 (0.13–0.80)	.014
Continuous	321	0.86 (0.76–0.97)	.013	72	0.68 (0.54–0.86)	.001
Model 2						
Tertiles						
Lowest	119	Reference		37	Reference	
Middle	109	0.83 (0.55–1.25)	.365	22	0.66 (0.31–1.43)	.291
Highest	93	0.64 (0.42–0.97)	.034	13	0.35 (0.14–0.86)	.022
Continuous	321	0.85 (0.76–0.96)	.010	72	0.69 (0.54–0.87)	.002
Model 3						
Tertiles						
Lowest	119	Reference		37	Reference	
Middle	109	0.83 (0.55–1.26)	.382	22	0.68 (0.31–1.50)	.340
Highest	93	0.64 (0.42–0.98)	.038	13	0.36 (0.14–0.88)	.025
Continuous	321	0.85 (0.76–0.96)	.011	72	0.69 (0.54–0.88)	.003
DTP						
Model 1						
Tertiles						
Lowest	117	Reference		30	Reference	
Middle	95	0.74 (0.49–1.13)	.164	24	1.13 (0.51–2.48)	.769
Highest	107	1.03 (0.66–1.62)	.885	14	1.16 (0.45–2.99)	.764
Continuous	319	0.97 (0.65–1.44)	.877	68	1.48 (0.63–3.48)	.365
Model 2						
Tertiles						
Lowest	117	Reference		30	Reference	
Middle	95	0.80 (0.52–1.22)	.298	24	1.11 (0.50–2.46)	.796
Highest	107	1.16 (0.73–1.83)	.536	14	1.24 (0.47–3.28)	.662
Continuous	319	1.03 (0.69–1.55)	.875	68	1.59 (0.68–3.76)	.287
Model 3						
Tertiles						
Lowest	117	Reference		30	Reference	
Middle	95	0.81 (0.53–1.24)	.331	24	1.10 (0.49–2.47)	.811
Highest	107	1.19 (0.75–1.88)	.466	14	1.26 (0.47–3.34)	.645
Continuous	319	1.05 (0.70–1.58)	.811	68	1.60 (0.67–3.81)	.288

Notes: Tertiles and continuous variable of UTP are expressed as mg GAE/24 h. Tertiles and continuous variable of DTP are expressed as mg/d. Number of cases (*n*), OR, CI, and *p* test are estimated in Models 1–3. Model 1 adjusting for age, gender, body mass index, total energy intake, alcohol consumption, smoking habit, activity level, and creatinine clearance. Model 2 adjusting for all variables present in Model 1 plus IL-6 and CRP. Model 3 adjusting for all variables present in Model 2 plus diabetes, hypertension, dementia, and renal failure. CI = confidence interval; DTP = dietary total polyphenols; GAE = gallic acid equivalents; OR = odds ratio; UTP = urinary total polyphenols.

Table 3. Covariate-Adjusted Associations of UTP Tertiles With Individual Frailty Criteria in the InCHIANTI Study

Models	Weight Loss		Exhaustion		Low Physical Activity*		Weakness		Slowness	
	n	OR (95% CI)	*n*	OR (95% CI)	*n*	OR (95% CI)	*n*	OR (95% CI)	*n*	OR (95% CI)
		p		*p*		*p*		*p*		*p*
Model 1										
Tertiles										
Lowest	12	Reference	58	Reference	70	Reference	75	Reference	81	Reference
Middle	18	1.59 (0.71–3.61) .263	53	0.94 (0.60–1.47) .769	44	0.69 (0.42–1.11) .127	55	0.76 (0.49–1.18) .225	48	0.55 (0.34–0.90) .016
Highest	10	1.01 (0.40–2.55) .983	29	0.43 (0.26–0.73) .002	38	0.78 (0.47–1.30) .344	39	0.60 (0.37–0.97) .036	41	0.55 (0.33–0.92) .021
Continuous	40	1.00 (0.78–1.27) .993	140	0.82 (0.71–0.94) .006	152	0.91 (0.79–1.05) .208	169	0.83 (0.73–0.95) .007	170	0.86 (0.74–0.99) .034
Model 2										
Tertiles										
Lowest	12	Reference	58	Reference	70	Reference	75	Reference	81	Reference
Middle	18	1.62 (0.71–3.68) .252	53	0.98 (0.62–1.54) .935	44	0.69 (0.42–1.12) .134	55	0.78 (0.50–1.22) .273	48	0.56 (0.35–0.92) .021
Highest	10	0.93 (0.36–2.40) .878	29	0.42 (0.24–0.72) .001	38	0.78 (0.46–1.31) .348	39	0.62 (0.38–1.01) .054	41	0.56 (0.34–0.94) .027
Continuous	40	0.97 (0.76–1.25) .848	140	0.81 (0.70–0.94) .005	152	0.91 (0.78–1.05) .176	169	0.83 (0.73–0.96) .009	170	0.86 (0.74–0.99) .035
Model 3										
Tertiles										
Lowest	12	Reference	58	Reference	70	Reference	75	Reference	81	Reference
Middle	18	1.79 (0.77–4.11) .173	53	0.99 (0.63–1.56) .967	44	0.67 (0.41–1.11) .121	55	0.75 (0.48–1.18) .218	48	0.58 (0.35–0.95) .029
Highest	10	0.92 (0.35–2.42) .869	29	0.42 (0.24–0.72) .002	38	0.81 (0.48–1.37) .441	39	0.62 (0.38–1.02) .059	41	0.55 (0.33–0.93) .026
Continuous	40	0.97 (0.76–1.25) .809	140	0.81 (0.70–0.94) .005	152	0.92 (0.79–1.06) .241	169	0.84 (0.73–0.96) .010	170	0.86 (0.74–0.99) .036

Notes: Tertiles and continuous variable of UTP are expressed as mg GAE/24 h. Number of cases (*n*), OR, CI, and *p* test are estimated in Models 1–3. Prefrail and frail were jointly modeled using binary logistic regression. Model 1 adjusting for age, gender, body mass index, total energy intake, alcohol consumption, smoking habit, activity level, and creatinine clearance. Model 2 adjusting for all variables present in Model 1 plus IL-6 and CRP. Model 3 adjusting for all variables present in Model 2 plus diabetes, hypertension, dementia, and renal failure. CI = confidence interval; GAE = gallic acid equivalents; OR = odds ratio; UTP = urinary total polyphenols.

*The activity level has been excluded from the models.