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Title: Interplay between dietary phenolic compound intake and the human gut microbiome in hypertension: a cross-sectional study

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Abstract: In the present study, potential associations between dietary phenolic compounds (PCs), gut microbiota composition and targeted faecal metabolites were identified in a cross-sectional study including grade 1 hypertensive (HT) and normotensive (NT) subjects. We performed comprehensive quantification of PC intake, together with 16S rRNA gene sequencing of the gut microbiota, and faecal SCFAs determination. The results showed multiple-way relationships between PCs from several plant-based foods and 25 bacterial taxa previously defined as discriminant biomarkers among groups. Coffee PCs were positively associated with systolic and diastolic blood pressure, SCFAs and Bacteroides plebeius and Bacteroides coprocola in HT and negatively associated with Faecalibacterium prausnitzii and Christensenellaceae R-7 in NT. Olive fruit PCs were positively associated with Ruminococcaceae UCG-010 and Christensenellaceae R-7 in NT. These interplays with discriminant bacterial taxa in HT and NT highlight the potential role of specific PCs as gut microbiome modulators in either the pathogenesis or prevention of hypertension.

Reus, August 11, 2020

Dear Editor in Chief,

On behalf of all authors, the manuscript entitled " **Interplay between dietary phenolic compound intake and the human gut microbiome in hypertension: a cross-sectional study**" is submitted as an *Original Research paper* for the consideration of the Editorial Board of *Food Chemistry*.

The present work emerges as a new developing field where potential associations between dietary phenolic compounds (PCs), the faecal microbiota composition and short-chain fatty acids (SCFAs), as targeted metabolites, are assessed in a cross-sectional study involving non-treated grade I hypertensive (HT) and normotensive subjects (NT). In the frame of our previous study (Calderón-Pérez et al., 2020), we hypothesize that specific dietary PCs from habitual diet could be precursors for the occurrence of particular bacterial taxa that differ between HT and NT subjects. After a comprehensive quantification of individual PC intake by food source and phenolic class and the application of 16S rRNA gene sequencing and targeted metabolomics, we were able to estimate habitual dietary PC intake and to determine gut microbiota composition and its derived metabolites.

Interestingly, we identified multiple-way relationships between PCs from several plant-based foods and discriminant bacterial taxa in both groups. In particular, coffee PCs were positively associated with systolic and diastolic blood pressure, faecal SCFAs, and *Bacteroides plebeius* and *Bacteroides coprocola* in HT, and negatively associated with *Faecalibacterium Prausnitzii* in NT. Additionally, olive fruit PCs were positively associated with *Ruminococcaceae_UCG-010* and *Christensenellaceae_R-7* only in NT. These interplays highlight the potential role of specific PCs as precursors for gut microbiome modulation and its effect in blood pressure regulation. Nevertheless, given the complexity of the reported relationships, future trials are needed to better understand the mechanisms involved in the plant-food PCs mediated effects in hypertension.

Given the relevance of our research topic in the understanding of deeply connections among key food phytochemicals, such as phenolic compounds, with gut microbiome composition and targeted metabolites in humans, we consider our work could fit in *Food Chemistry* as Original Research and would be of great interest to the readers.

All authors warrant that neither the entire manuscript nor any part of its content has been published previously or is being submitted to another journal, in English or in any other language. All authors have contributed to the study according to international consensus on authorship and have approved the final draft, agreeing with the analyses of the data and the conclusions reached in the manuscript. The corresponding authors formally state, on behalf of all authors, that there are no competing interests to declare.

We look forward to receiving your opinion as to the suitability of our manuscript for inclusion in *Food Chemistry*.

Yours sincerely,

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Dear Editor and Reviewers,

Thank you for your letter and the reviewers' comments on our manuscript entitled "**Interplay between dietary phenolic compound intake and the human gut microbiome in hypertension: a cross-sectional study"**. We appreciate the time and effort that you and the reviewers have dedicated to providing your valuable feedback on our manuscript and we are grateful to the reviewers for their insightful comments on our paper.

We have been able to address all the issues suggested by the reviewers as well as incorporate all the proposed changes into the original manuscript. Accordingly, we have added the appropriate clarifications and some new references to the revised manuscript, which has allowed us to improve the manuscript content. We have highlighted the changes suggested by the reviewers in yellow within the revised manuscript.

POINT-BY-POINT RESPONSES:

Comments from Reviewer #2:

The work presented in this manuscript is important and relevant, particularly in the interest of prebiotic potential of phenolic compounds.

Suggestions to improve the paper

- **Comment 1.** Subjects and study design: Were vegetarians and vegan excluded from study?

Response 1: As stated by the reviewer, subjects following a vegetarian/vegan diet were excluded from the study because of their evident differences in gut microbiota composition (*Tomova A et al., 2019*). Moreover, plant-based diets could result in greater intake of phenolic compounds than omnivore diets. Notably, we did not find any vegetarian individuals at the time of recruitment.

Accordingly, we have included further explanation on this point in the *Subjects and study design* section of the revised manuscript:

(Page 5, Line 128 - 130): "Further, subjects with chronic alcohol consumption, use of either antibiotics or probiotics, or a vegetarian or vegan diet at the time of inclusion were also excluded."

Reference:

Tomova A, Bukovsky I, Rembert E, Yonas W, Alwarith J, Barnard ND, Kahleova H. The Effects of Vegetarian and Vegan Diets on Gut Microbiota. Front Nutr. 2019;6:47.

- **Comment 2.** Determination of fecal SCFAs- Why was serum SCFA not performed (mention also in limitation).

Response 2: In accordance with the reviewer suggestion, we have considered adding new data on circulating SCFAs to the revised manuscript. In fact, plasma SCFAs were fully assessed in our previous work in relation to blood pressure (*Calderón-Pérez L et al., 2020*).

As stated, the potential usefulness of faecal SCFAs as an indicator of the gut microbiota ecosystem has been previously reported in humans (*Yamamura, R et al., 2019*). In addition, phenolic compounds can be associated with gut microbiota composition and contribute to faecal SCFA production (*Fernandez-Navarro, T et al., 2018*). Colonic SCFAs represent the major end products of microbial activity and are highly affected by diet composition. Thus, in the present work we initially chose faecal SCFAs as the best indicators for studying correlations with dietary phenols. However, given the reviewer suggestion, we have included plasma SCFA correlations since they could provide additional explanation on how phenolic compounds affect SCFA absorption in both HT and NT subjects.

Accordingly, we have made the following changes within the revised manuscript:

(Supplementary material, *Supplementary table 4*): Correlations between phenolic compound food sources and classes with plasma SCFA concentrations in HT and NT subjects.

(Page 2, Lines 37 and 43): In the *Abstract,* we have pointed out the plasma SCFA assessment and correlations.

(Page 8, Line 203 - 208): In the *Material and Methods* section, we have briefly described the procedure used for plasma SCFA assessment.

(Page 14, Line 352 - 356): In the *Results* section, we have added significant correlations between phenolic compounds and plasma SCFAs as follows: "On the other hand, in the NT group, significant positive correlations were noted for fresh fruit and olive fruit PCs with plasma butyrate ($p = 0.042$ and 0.049, respectively) and valerate ($p = 0.001$ and 0.020, respectively) levels. Additionally, anthocyanins were positively correlated with plasma valerate ($p = 0.047$)."

(Page 16, Line 411 - 413): In the *Discussion* section, we have provided a new argument highlighting the notable interrelationships found among olive fruit PCs and plasma SCFAs as follows: "…, discriminant ASV biomarkers, such as Ruminococcaceae UCG-010 and Christensenellaceae R-7, and plasma butyrate and valerate. These interrelationships could reflect a favoured butyrate and valerate production and absorption in the colon of NT group mediated by olive fruit PCs."

(Page 21, Line 540): In the *Conclusion*, we have mentioned the positive association for olive fruit PCs with plasma SCFAs.

(Figure 2): We have specified faecal (FE) and plasma (PL) SCFA interrelationships with phenolic compounds in the figure.

(Graphical Abstract): We have included new interrelationships for olive fruit PCs with plasma SCFAs.

References:

Calderón-Pérez, L., Gosalbes, M. J., Yuste, S., Valls, R. M., Pedret, A., Llauradó, E.,… & Solà, R. (2020). Gut metagenomic and short chain fatty acids signature in hypertension: a cross-sectional study. Scientific Reports, 10(1), 6436.

Yamamura, R., Nakamura, K., Kitada, N., Aizawa, T., Shimizu, Y., Nakamura, K., … & Tamakoshi, A. (2019). Associations of gut microbiota, dietary intake, and serum short-chain fatty acids with fecal short-chain fatty acids. Bioscience of microbiota, food and health, 19-010.

Fernandez-Navarro, T., Salazar, N., Gutierrez-Diaz, I., Sanchez, B., Ruas-Madiedo, P., de Los Reyes-Gavilan, C. G., … Gonzalez, S. (2018). Bioactive compounds from regular diet and faecal microbial metabolites. European Journal of Nutrition, 57(2), 487–497.

- **Comment 3.** Line 200-201; rephrase to improve clarity.

Response 3: The sentence has been rephrased as follows: "…, and their quantification was determined with reference to the peak side of the internal standard (4-methyl valeric acid)." (Page 8, Line 201)

- **Comment 4.** Line 252: Show evidence that sample size will provide significant power in study.

Response 4: Thank you for pointing this out. The authors would like to note that at the time of the study design (2016), to provide significant power between the two groups, we considered a sample size of 30 hypertensive subjects and their respective healthy controls (1:1 ratio) based on a study performed by *Yang T et al. (2015). Yang T et al.*

reported for the first time that the human intestinal microbiome in 7 hypertensive patients harboured lower microbial diversity than the microbiome of 10 healthy controls. Taking into account the study by *Yang T et al.*, in our study, we decided to expand the sample size to 30 untreated grade 1 hypertensive and 30 normotensive subjects.

Accordingly, we have detailed the sampling strategy we relied on in the statistical analysis section (Page 10, Line 256 - 260): "Sample size was estimated based on a study by Yang T et al. (*Yang et al., 2015*) where significant differences were observed in the gut microbiota composition between 7 hypertensive patients and 10 healthy controls. Thus, we assumed that an expanded sample size of 30 untreated grade 1 HT subjects and their respective healthy controls (1:1 ratio) was enough to achieve significant power."

Reference:

Yang T, Santisteban MM, Rodriguez V, Li E, Ahmari N, Carvajal JM, Zadeh M, Gong M, Qi Y, Zubcevic J, Sahay B, Pepine CJ, Raizada MK, Mohamadzadeh M. Gut dysbiosis is linked to hypertension. Hypertension. 2015 Jun;65(6):1331-40.

- **Comment 5.**

  Comment 5 a. Discussion: it is important that the authors also mention how their results correlate with serum SCFA- *eg. Yamamura, R., Nakamura, K., Kitada, N., Aizawa, T., Shimizu, Y., Nakamura, K., ... & Tamakoshi, A. (2019). Associations of gut microbiota, dietary intake, and serum short-chain fatty acids with fecal short-chain fatty acids. Bioscience of microbiota, food and health, 19-010.*

  Comment 5 b. Also the authors should also mention about ellagitannins (present wine, fruits and some nuts) and its effect of Gut microbs, SCFA and hypertension.

  Comment 5 c. Figure 2 is rather confusing and difficult to understand, please improve its clarity.

Response 5 a: In response to the reviewer proposal and given the forthcoming relation of circulating SCFAs to metabolic health in humans (*Müller M et al., 2019)*, we have mentioned the extent of our noted correlations with faecal and plasma SCFAs (new results) in the *Discussion* section as follows:

(Page 20, Line 505 – 513): "Furthermore, the opposite trends observed in associations among faecal and plasma SCFAs with particular dietary PCs in HT and NT subjects could explain different absorption modes depending on the hypertension stage. In

addition, it will be helpful to better understand the dynamics of SCFAs in the human body (*Yamamura et al., 2020*). Specifically, fresh fruit and olive fruit PCs seem to favour SCFA absorption, leading to higher plasma concentrations of butyrate and valerate in NT, while coffee PCs are related to higher faecal SCFA levels in HT, suggesting a lower efficiency in their intestinal absorption, which could be a causative factor for the higher BP, as hypothesized in our previous work *(Calderón-Pérez et al., 2020)*."

Response 5 b: On the other hand, in response to the reviewer suggestion about ellagitannin relationships with gut microbes, SCFAs and hypertension, the authors are aware of the prebiotic properties of ellagitannin and their modulatory effect on gut microbiota *(Kawabata K et al., 2019)*. Indeed, the microbial-derived metabolites of ellagitannins and ellagic acid have been conferred vasoprotective effects, including blood pressure reduction, in several clinical studies (*Wang D et al., 2018*). It is important to note that there is no accurate information available on ellagitannins in food databases. Indeed, in the Phenol Explorer Data Base used in the present study, they are not considered an independent phenolic class, and they have been included in the hydroxybenzoic acid phenol class. Consequently, in the present work, the specific correlation between the estimated intake of ellagitannins and all the parameters studied was not possible to analyse, as they are included in the hydroxybenzoic phenolic class, which comprises very diverse phenolic compounds.

However, in our study, the strong positive associations noted in NT subjects between dried fruits, nuts, natural juices, and hydroxybenzoic acids class, with the Ruminococcaceae and Christensenellaceae families, point out a possible beneficial effect of ellagitannins included in this phenol class on blood pressure reduction. Despite this, given the lack of correlations for hydroxybenzoic acids with SCFAs, we cannot suggest that the speculated hypotensive effect is mediated by SCFA action. Additional explanation regarding this topic has been included in the *Discussion* section as follows:

(Page 19, Line 473 - 478): "It should be noted that the correlations found in NT subjects between the nuts, dried fruits, natural juices PCs, and hydroxybenzoic acids class with Ruminococcaceae and Christensenellaceae families could explain a protective effect of ellagitannins on BP (Wang et al., 2018), as they appear to be included in this phenol class on the Phenol Explorer data base. However, given the lack of correlations with

SCFAs, we cannot suggest that the supposed ellagitannin hypotensive effect is mediated by SCFA action."

Response 5 c: Finally, the clarity of *Figure 2* has been improved through the removal of less significant associations. We have left the multiple-way associations among phenolic compounds, bacterial family and SCFAs. Moreover, the figure caption has been completed (Page 29, Line 746).

References:

Müller M, Hernández MAG, Goossens GH, Reijnders D, Holst JJ, Jocken JWE, van Eijk H, Canfora EE, Blaak EE. Circulating but not faecal short-chain fatty acids are related to insulin sensitivity, lipolysis and GLP-1 concentrations in humans. Sci Rep. 2019 Aug 29;9(1):12515.

Yamamura, R., Nakamura, K., Kitada, N., Aizawa, T., Shimizu, Y., Nakamura, K.,…& Tamakoshi, A. (2020). Associations of gut microbiota, dietary intake, and serum short-chain fatty acids with fecal short-chain fatty acids. Bioscience of Microbiota, Food and Health, 39(1), 11–17.

Kawabata K, Yoshioka Y, Terao J. Role of Intestinal Microbiota in the Bioavailability and Physiological Functions of Dietary Polyphenols. Molecules. 2019 Jan 21;24(2):370.

Wang D, Özen C, Abu-Reidah IM, Chigurupati S, Patra JK, Horbanczuk JO, Jóźwik A, Tzvetkov NT, Uhrin P, Atanasov AG. Vasculoprotective Effects of Pomegranate (Punica granatum L.). Front Pharmacol. 2018 May 24;9:544.

Comments from Reviewer #3:

The manuscript addresses the correlation of phenolic compounds intake and gut microbiota in hypertension, the study is very interesting in my opinion. The data collection is carefully described, however, I have some doubts about possible correlations:

- **Comment 1.** How about the analysis of dairy products and blood pressure (Supplementary table 1)?

Response 1: As the reviewer mentioned in the initial comment, our manuscript is mainly focused on the potential associations between phenolic compound intake and gut microbiota in hypertension. However, the authors would like to clarify that the results shown in *Supplementary table 1* refer to the mean daily intake of overall food groups assessed in the validated food-frequency questionnaire (FFQ) as we mention in *Material and Methods* (Page 6, Line 155).

Despite the recognized relationship between dairy product intake, particularly low-fat dairy, and blood pressure lowering (*Aljuraiban GS et al., 2018*), these correlations are outside our study topic. However, we do not rule out considering dairy food associations in future studies beyond phenolic compounds.

Reference:

Aljuraiban GS, Stamler J, Chan Q, Van Horn L, Daviglus ML, Elliott P, Oude Griep LM; INTERMAP Research Group. Relations between dairy product intake and blood pressure: the INTERnational study on MAcro/micronutrients and blood Pressure. J Hypertens. 2018 Oct;36(10):2049-2058.

- **Comment 2.** How about the correlation of SCFA and blood pressure (https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5584783/)?

Response 2: As suggested by the reviewer, there appears to be a link between SCFAs and blood pressure. Although part of the evidence points to the interaction of SCFAs with products of gut microbial metabolism that are known to affect host physiology via G protein-coupled receptors (GPCRs), including Gpr41 and Olfr78, as possible pathways involved in blood pressure regulation, the details and intricacies of these interactions are not yet fully understood (*Pluznick JL et al., 2017*).

As we mention in the *Introduction* section (Page 4, Line 95 - 97), our previous results in the same cohort of HT and NT subjects supported the hypothesis that greater faecal excretion together with lower plasma circulating levels of SCFAs could be a marker of hypertensive state due to poor gut health (*Calderón-Pérez et al., 2020*). On the basis of this previous study, in the present work, we provide further knowledge on the relation of SCFAs, blood pressure and gut microbiota composition with dietary phenolic compounds (PCs). In this sense, in the *Discussion* section of the revised manuscript, we have mentioned the novelty of our observed correlations of dietary PCs with faecal and plasma SCFAs in relation to blood pressure, as follows:

(Page 20, Line 502 - 513): "Our findings extend previous knowledge on the correlation between the gut microbiota and hypertension in humans (Calderón-Pérez et al., 2020) and provide a new way in which dietary PCs could induce changes in gut bacterial composition to affect SCFA production and absorption. Furthermore, the opposite trends observed in associations among faecal and plasma SCFAs with particular dietary PCs in HT and NT subjects could explain different absorption modes depending on the hypertension stage. In addition, it will be helpful to better understand the dynamics of

SCFAs in the human body (Yamamura et al., 2020). Specifically, fresh fruit and olive fruit PCs seem to favour SCFA absorption, leading to higher plasma concentrations of butyrate and valerate in NT, while coffee PCs are related to higher faecal SCFA levels in HT, suggesting a lower efficiency in their intestinal absorption, which could be a causative factor for the higher BP, as hypothesized in our previous work (Calderón-Pérez et al., 2020).

However, intervention studies are needed to determine the metabolic fate of the SCFAs produced from PC action on gut microbiota and to determine whether the kinetics of SCFA production and metabolism differ in HT and NT humans to better understand the potential connection between hypertension and SCFAs.

References:

Pluznick JL. Microbial Short-Chain Fatty Acids and Blood Pressure Regulation. Curr Hypertens Rep. 2017 Apr;19(4):25.

Yamamura, R., Nakamura, K., Kitada, N., Aizawa, T., Shimizu, Y., Nakamura, K., … Tamakoshi, A. (2020). Associations of gut microbiota, dietary intake, and serum short-chain fatty acids with fecal short-chain fatty acids. Bioscience of Microbiota, Food and Health, 39(1), 11–17. https://doi.org/10.12938/bmfh.19-010

Calderón-Pérez, L., Gosalbes, M. J., Yuste, S., Valls, R. M., Pedret, A., Llauradó, E.,… & Solà, R. (2020). Gut metagenomic and short chain fatty acids signature in hypertension: a cross-sectional study. Scientific Reports, 10(1), 6436.

- **Comment 3.** How the authors addressed the role of slight overweight of the hypertensive group on the results?

Response 3: The reviewer has raised an important point here. First, we would note that despite the significantly higher baseline BMI values in HT compared to NT subjects (26.2 ± 2.5 and 23.8 ± 2.7 kg/m^2, respectively) ($p<0.001$), the HT group did not reach the obesity BMI limit (BMI ≥ 30 kg/m^2). Thus, we assumed that the observed microbial changes between groups might be related to hypertension, and the slight overweight by itself would not contribute to the changes. In addition, the associations reported in our previous work (*Calderón-Pérez et al., 2020*) between microbial composition and blood pressure remained significant after adjustment for BMI among other variables, indicating that the differences seem to be intrinsically related to the hypertensive condition rather than other external factors.

Accordingly, we have provided further explanation of the *limitations* as follows:

(Page 20, Line 520 - 525): "…, and the existence of unmeasured human factors, such as inter-individual variability in gut microbiota composition or different clinical parameters at baseline, such as BMI, may have influenced the results. However, the associations between microbial composition and BP remained significant after adjustment for BMI among other variables, as reported in our previous work *(Calderón-Pérez et al., 2020)*, indicating that the differences were intrinsically related to the hypertensive condition.

- **Comment 4.** How about the correlations of dietary fiber and blood pressure? A mutual correlation of phenolic + fibers could be convenient?

Response 4: The authors are aware of the impact of dietary fibre intake on blood pressure regulation. Indeed, several studies have indicated a relationship between dietary fibre and decreased systolic and diastolic blood pressure when administered to hypertensive and pre-hypertensive subjects (*Aleixandre, A et al., 2016*). Moreover, the Dietary Approaches to Stop Hypertension (DASH) study already indicated that diets high in fibre and low in salt and fat decreased arterial blood pressure (*Sacks FM et al., 1995*). However, the controversy about the possible antihypertensive effects of dietary fibre remains debatable, and particular components such as polyphenols containing in the same food matrices could explain their effects on blood pressure.

In our study, food sources that explain at least 80% of phenol intake were selected to analyse their associations with gut microbiota composition and metabolites. Notably, all the selected foods, such as tubers, whole-grain cereals, legumes, fresh fruits and vegetables, also contained soluble and insoluble fibre, used by colonic microorganisms to form SCFAs, which have been related to blood pressure lowering (*Natarajan N et al., 2016*). To carry out with the evident mutual link of PCs and dietary fibre, as we mention on Page 10, Lines 240 - 242 of the manuscript, we considered fibre intake to be a confounding factor and controlled for it in the regression analysis. This adjustment ensured that the identified interactions between PCs and blood pressure were attributable only to the intrinsic effect of PCs.

Additionally, we would like to note that despite the significantly higher dietary fibre intake in NT subjects compared to HT subjects (25.90 ± 11.63 and 20.95 ± 9.90 g/day,

respectively) (p=0.029), no significant correlations were found with systolic and diastolic blood pressure. See the attached table:

	Systolic blood pressure *(mmHg)*				**Diastolic blood pressure** *(mmHg)*			
	Hypertensive		**Normotensive**		**Hypertensive**		**Normotensive**	
	r	**p**	**r**	**p**	**r**	**p**	**r**	**p**
Dietary fibre, *g/day*	0.342	0.075	0.281	0.120	0.357	0.072	-0.021	0.910

References:

Aleixandre, A., & Miguel, M. (2016). Dietary fiber and blood pressure control. Food & Function, 7(4), 1864–1871.

Sacks FM, Obarzanek E, Windhauser MM, Svetkey LP, Vollmer WM, McCullough M, Karanja N, Lin PH, Steele P, Proschan MA, et al. Rationale and design of the Dietary Approaches to Stop Hypertension trial (DASH). A multicenter controlled- feeding study of dietary patterns to lower blood pressure. Ann Epidemiol. 1995 Mar;5(2):108-18. PMID: 7795829.

Natarajan, N., Hori, D., Flavahan, S., Steppan, J., Flavahan, N. A., Berkowitz, D. E., & Pluznick, J. L. (2016). Microbial short chain fatty acid metabolites lower blood pressure via endothelial G protein-coupled receptor 41. Physiological Genomics, 48(11), 826–834.

The authors look forward to hearing from you in due time regarding our submission and to respond to any further questions or comments you may have. We hope that this improved version will meet the journal expectations.

Yours sincerely,

Anna PEDRET, PhD

and

Laura RUBIÓ, PhD
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Interplay between dietary phenolic compound intake and the human gut microbiome in hypertension: a cross-sectional study

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Word count: 6426

ABSTRACT

In the present study, potential associations between dietary phenolic compounds (PCs), gut microbiota composition and targeted faecal metabolites were identified in a cross-sectional study including grade 1 hypertensive (HT) and normotensive (NT) subjects. We performed comprehensive quantification of PC intake, together with 16S rRNA gene sequencing of the gut microbiota, and faecal and plasma short-chain fatty acids (SCFAs) determination. The results showed multiple-way relationships between PCs from several plant-based foods and 25 bacterial taxa previously defined as discriminant biomarkers among groups. Remarkably, coffee PCs were positively associated with systolic and diastolic blood pressure, faecal SCFAs, *Bacteroides plebeius* and *Bacteroides coprocola* in HT and negatively associated with *Faecalibacterium prausnitzii* and Christensenellaceae R-7 in NT. Olive fruit PCs were positively associated with Ruminococcaceae UCG-010, Christensenellaceae R-7 and plasma SCFAs in NT. These interplays with discriminant bacterial taxa in HT and NT subjects highlight the potential role of specific PCs as gut microbiome modulators in either the pathogenesis or prevention of hypertension.

Keywords: Phenolic compound; Coffee polyphenols; Olive polyphenols; Hypertension; Blood pressure; Gut microbiota; Short-chain fatty acids

1. INTRODUCTION

Diet can modulate the composition and functional capacity of the human gut microbiota, an effect that depends largely on individual dietary choices (Danneskiold-Samsøe et al., 2019). The consumption of different food components, such as animal protein, digestible and non-digestible carbohydrates, fats, and probiotics, induces shifts in host microbial diversity, with secondary effects on immunological, biochemical and metabolic markers (Danneskiold-Samsøe et al., 2019).

Phenolic compounds (PCs) are phytochemicals mainly present in fruits, vegetables, olive oil, cocoa products and wine, and their regular dietary intake has protective effects against several chronic diseases, such as cardiovascular diseases (CVDs); in addition, PCs have anti-carcinogenic and anti-inflammatory properties (Costa et al., 2017). Recent findings suggest that dietary PCs could help alleviate CVDs biomarkers by altering the gut microbiota (Moorthy, Chaiyakunapruk, Jacob, & Palanisamy, 2020). It is estimated that 90-95% of total PC intake is not absorbed in the small intestine and accumulates in the large intestinal lumen, where the gut microbiota has the metabolizing capacity to convert the PCs into active metabolites, which could be responsible for the observed health effects (Selma, Espín, & Tomás-Barberán, 2009). In turn, PCs can modulate gut microbial composition through a 'prebiotic-like effect', in which they are able to promote the growth of certain beneficial bacterial taxa (Tomas-Barberan, Selma, & Espin, 2016). Other beneficial commensal bacteria, such as mucin-degrading *Akkermansia* spp. and butyrate-producing *Faecalibacterium* spp., were recently shown to increase after administration of proanthocyanidin-rich extracts and foods containing dihydroflavonols in animal models (Tomas-Barberan et al., 2016).

Preclinical and clinical human studies have also shown a positive influence of dietary PC intake on the relative abundance of gut microbes. A reduction in the number of potential pathogens, including *Clostridium perfringens* and *Clostridium histolyticum* and certain gram-negative *Bacteroides* spp., and an increase in the growth of beneficial Clostridia, Bifidobacteria and Lactobacilli have been detected after PC intake (Duenas et al., 2015).

In most cases, human studies have focused on the administration of PC-enriched diets and controlled supplementation with PC-rich extracts or nutraceuticals with high doses of PCs (Most, Penders, Lucchesi, Goossens, & Blaak, 2017; Nash et al., 2018; Xie et al., 2016). However, none of these studies have assessed the interplay between the gut microbiome and the individual PC intake provided by foods in the context of habitual diet.

Hypertension is one of the most common chronic diseases and remains the major preventable cause of CVD globally (Esh et al., 2018). Dietary intake of fruit, vegetables and fibre has been inversely associated with blood pressure (BP) levels (Marques, Mackay, & Kaye, 2018), and has shown a long-term effect of reducing systolic and diastolic BP in subjects with normal BP or mild hypertension (Nissensohn, Roman-Vinas, Sanchez-Villegas, Piscopo, & Serra-Majem, 2016). Likewise, the Mediterranean diet has emerged as an optimal strategy for promoting gut microbial diversity and stability and preventing dysbiosis (Rinninella et al., 2019). Negative associations of total polyphenol intake (TPI), assessed by urinary total polyphenol excretion (TPE), with BP levels and the prevalence of hypertension in a Mediterranean population at high cardiovascular risk were also reported (A Medina-Remon et al., 2011). Recently, the role of gut microbiota-derived metabolites in the regulation of BP has been suggested (Marques et al., 2018). For instance, SCFAs, which are the major end products of faecal bacterial activity after dietary fibre fermentation, can help lower BP levels (Natarajan et al., 2016). Moreover, our previous results supported the hypothesis that greater faecal excretion together with lower circulating levels of SCFAs could be a marker of a hypertensive state due to poor gut health (Calderón-Pérez et al., 2020). Dietary intake of PCs such as anthocyanins and lignans has been positively associated with faecal concentrations of SCFAs (Fernandez-Navarro et al., 2018), which highlights the importance of investigating the bacterial modulatory effects of PCs and their links with bacterially derived metabolites.

Thus, dietary PCs may represent promising candidates for preventing or delaying the onset of hypertension, and the investigation of whether dietary PC intake attenuates BP through a beneficial impact on the gut microbiome is a new developing field (de Brito Alves et al.,

2016). On the basis of our previous study (Calderón-Pérez et al., 2020) reporting a particular bacterial signature in the guts of non-treated grade 1 hypertensive (HT) and normotensive (NT) subjects, the present study provides further knowledge of the relation of this effect with diet. Specifically, we hypothesize that specific dietary PCs from habitual diet could be precursors for the occurrence of particular bacterial taxa that differ between HT and NT subjects. Therefore, the main purpose of the present study is to identify potential associations between dietary PC intake, faecal microbiota composition and SCFAs as target faecal metabolites in non-treated grade 1 HT subjects compared to NT subjects.

2. MATERIAL AND METHODS

2.1. Subjects and study design

All the individuals included in the present study were recruited between 9 June 2016 and 28 November 2017. Participants were recruited by using tableaux advertisements in the *Hospital Universitari Sant Joan de Reus* (HUSJ, Spain) and databases of volunteers who had previously participated in studies carried out by our research group. HT participants were included in the study if they exhibited grade 1 hypertension, defined as systolic BP between 140 and 159 mm Hg and without major complications, according to the *ESC/ESC Guidelines (2018)* (Esh et al., 2018), and not using antihypertensive medication. NT participants presented optimal systolic BP below 120 mm Hg. All subjects fulfilled the following criteria: aged from 18 to 65 years, without a family history of cardiovascular disease or evidence of chronic disease, and willing to provide informed consent before the initial screening visit. Subjects with BMI ≥ 30 kg/m^2, fasting glucose > 126 mg/dL, low-density lipoprotein (LDL) cholesterol ≥ 190 mg/dL, triglycerides > 350 mg/dL, a history of smoking, and anaemia or intestinal disorders were excluded. Individuals were also excluded if they were using anti-hypertensive or lipid-lowering medication, as well as if they were menopausal, pregnant or breastfeeding. Further, subjects with chronic alcohol consumption, use of either antibiotics or probiotics, or a vegetarian or vegan diet at the time of inclusion were also excluded.

Participants visited the HUSJ and Eurecat-Reus, where the study was performed, twice. In a first pre-selection visit, a clinical interview verifying that participants met all the eligibility criteria was performed. In addition, blood extraction was performed, BP was measured, and anthropometric and physical activity data were collected. In the second visit, in addition to the previous measures, urine and stool samples were collected. A 3-day dietary record and a food-frequency questionnaire (FFQ) were provided to assess dietary habits.

The study protocol was approved by the local ethics committee (Clinical Research Ethical Committee of HUSJ, Reus with the 15-11-26/11obs4 reference) prior to the study beginning, and informed consent was obtained from all subjects. The protocol was conducted in accordance to the Helsinki Declaration and Good Clinical Practice Guidelines of the International Conference of Harmonization (ICH GCP).

2.2. Clinical and nutritional data collection

All clinical information was collected according to standard procedures. BP was monitored with participants in a seated position after resting for 2-5 minutes by a using multiple automated sphygmomanometer (OMRON HEM-907; Peroxfarma, Barcelona, Spain). Two repeated readings were recorded with a 1-min interval, and the average value was used for statistical analyses. To minimize white-coat/masked hypertension, it was ensured that participants rested alone and were unobserved in a quiet environment during the monitoring (Esh et al., 2018). Anthropometric parameters, including weight, height and body composition, were measured with a body composition analyser (Tanita SC 330-S; Tanita Corp., Barcelona, Spain). Waist circumference was measured at the umbilicus level using a 150-cm anthropometric steel measuring tape. Physical activity was evaluated by completion of the "Physical Activity Questionnaire Class AF" validated questionnaire (Vallbona Calbó, Roure Cuspinera, Violan Fors, & Alegre Martín, 2007).

Habitual dietary intake was assessed by using a validated, semi-quantitative, food-frequency questionnaire (FFQ) including 137 food items related to the Mediterranean diet (Fernández-Ballart et al., 2010). Mean daily energy and nutrient intakes were assessed through a 3-day

158 dietary record (2 labour days and 1 week-end day) and calculated by Spanish food composition
159 tables (Tablas de composición de alimentos del Cesnid, 2004).

160 2.3. Biological sample collection

161 Fasting blood samples were obtained in the second visit to determine lipid profiles. Total
162 cholesterol, LDL cholesterol, high-density lipoprotein (HDL) cholesterol and triglyceride
163 concentrations were measured in serum by standardized, automated enzymatic methods in an
164 autoanalyzer (Beckman Coulter-Synchron, Galway, Ireland).

165 Faecal samples were obtained with a Protocult™ stool collection device (ABC, Minnesota,
166 EEUU) with two different containers: a sterile pot and a specimen container with a spoon
167 containing 7 mL of RNAlater® storage solution (Sigma-Aldrich Quimica SL; Madrid, Spain).
168 The samples were frozen at home and transported to the laboratory with an ice pack. The
169 samples were stored then at -80ºC until the analysis of gut microbiota composition and SCFAs
170 determination.

171 Urine samples were collected in a special container over a 24-hour period, and the supernatants
172 of centrifuged samples were kept at -80ºC.

173 All samples were stored in the central laboratory's biobank at HUSJ-Eurecat
174 (biobanc.reus@iispv.cat) until they were required for batch analyses.

175 2.4. Estimation of dietary phenolic compound intake

176 Three-day dietary records provided by participants were used to obtain information about
177 the consumption of PC-rich foods and ingredients. Dietary PC intake was estimated according
178 to the phenolic content of the foods listed in the Phenol-Explorer database (Neveu et al., 2010)
179 by using individual PC values determined by high-performance liquid chromatography (HPLC).
180 The daily PC intake values were expressed as mg/day in two distinct ways: i) individually by
181 phenolic class and subclass and ii) by the sum of total PCs from major food sources. To
182 establish associations, the main PC-rich food sources were selected and grouped into categories
183 (tubers, cereals, vegetables, legumes, fresh fruits, nuts, oils, sweets, non-alcoholic beverages,

184 alcoholic beverages, and seasonings) and subcategories (refined cereals, whole grains, berries,
185 olive fruits, olive oil, chocolate, natural juices, coffee, tea, and wine).
186 TPI was calculated by matching food consumption data and the total polyphenol content
187 quantified by the Folin assay method from Phenol-Explorer.

188 ## 2.5. Total polyphenol excretion in urine

189 TPE was determined in 24 h urine samples by means of the Folin-Ciocalteu method
190 using an Oasis® MAX 96-well plate cartridge for solid-phase extraction, as described by
191 *Medina-Remón et al., 2009* (Alexander Medina-Remon et al., 2009). Creatinine measurements
192 were used to adjust for variations in analyte concentration in urine. To analyze creatinine in
193 urine, a reaction method in plates was applied with picric acid as the reagent, followed by
194 spectrophotometric measurement. TPE was expressed as mg gallic acid equivalent (GAE)/g of
195 creatinine.

196 ## 2.6. Determination of faecal and plasma SCFAs

197 Faecal SCFAs were analyzed in lyophilized feces by using an integrated system
198 including a gas chromatographer coupled to a flame ionization detector (GC-FID), as previously
199 described (Calderón-Pérez et al., 2020). Identification of the SCFAs was carried out according
200 to the retention time of standard compounds (acetic acid, propionic acid, butyric acid, and
201 valeric acid; Sigma-Aldrich), and their quantification was determined with reference to the peak
202 side of the internal standard (4-methyl valeric acid). All samples were analysed in triplicate.

203 Plasma SCFAs were quantified by a derivatization procedure and analysed by gas
204 chromatography (Agilent 6890N-MSD 5973) using a DB5 MS-UI column (30 m, 0.25 mm,
205 0.25 μm; J&W, Agilent Tech), coupled to a mass detector using SIM mode, as previously
206 described (Calderón-Pérez et al., 2020). The contents of SCFAs were calculated with internal
207 standard method. All samples were analyzed in duplicate, reporting coefficient of variability
208 values lower than 10%.

209 ## 2.7. Faecal microbiota composition analysis

DNA purification and sequencing

DNA was extracted from faecal samples stored in RNAlater® as previously described (Calderón-Pérez et al., 2020). The V3-V4 region of the 16S rRNA gene was amplified, and amplicon libraries were constructed according to the manufacturer's instructions (Illumina). Sequencing was performed with the MiSeq V3 Kit (2x230 cycles) (Illumina, Eindhoven, Netherlands) at the Centre for Public Health Research (FISABIO-Salud Pública, Valencia, Spain). All sequences were deposited on the public European Nucleotide. Archive server under accession number PRJEB32411.

Sequence analysis

16S rRNA gene reads with low-quality scores (<30) and reads shorter than 100 nucleotides as well as potential chimeras and human sequences were removed using the DADA2 (v1.8.0) pipeline (Callahan et al., 2016) in R package. The error rates for each base transition were estimated, and dereplication was carried out to combine all identical reads into unique sequences, with abundance equal to the number of reads combined. Taking the dereplicated reads and the error estimates, amplicon sequence variants (ASVs) were inferred. The SILVA database (v.132) (Quast et al., 2013) was set as the reference for taxonomic classification of each ASV. This ASVs were classified to the genus level, applying exact matching (100% identity) to assign a unique species to each ASV sequence. ASVs with an assigned genus but without exact matching at the species level, were mapped against the same reference database with a minimum identity of 97%.

2.8. Statistical analysis

Statistical analysis was performed using IBM SPSS version 25.0 (IBM SPSS, Inc, Chicago, IL, USA). The normality of variables was analysed by means of the Kolmogorov-Smirnov test. The Mann-Whitney test was used to compare non-normally distributed variables. Student's t-test was used to compare normally distributed variables. Fisher's exact test was used

for categorical variable comparisons. Descriptive data are expressed as the mean ± SD, with percentages for categorical variables.

In the first step, we analysed the Pearson correlations of TPI, urinary TPE and PC intake from major dietary sources (mg/day) and their classes and subclasses (mg/day) with systolic and diastolic BP values in all subjects. Stepwise multiple linear regression analysis was subsequently used to predict the strength of the associations. Given the possible confounding effect of dietary fibre and assuming that it is usually present in the same food matrix of PCs, this component was controlled for in the regression analysis. Energy intake and age were also controlled to ensure that the identified interactions between PCs and the gut microbiota were attributable only to the intrinsic effect of PCs.

In a second step, to identify specific bacterial taxa as biomarkers in both the HT and NT groups, the linear discriminant analysis (LDA) effect size (LEfSe) method was performed at the ASV level. The associations of ASVs with an LDA score > 2 with TPI, TPE, PC dietary sources and the main phenolic classes were analysed. Correlations were first tested with Pearson correlation analysis. Then, multivariate regression analysis was used, setting ASV biomarkers as predictors and PC variables as response variables. We also adjusted for potential confounding factors. The statistical parameters employed were β (standardized regression coefficient) and R^2 (coefficient of multiple determination). Additionally, linear regression analysis was used for graphical representation of significant relationships. Only significant results are presented. The conventional probability value for significance of 0.05 was used in the interpretation of the results.

Sample size was estimated based on a study by *Yang T et al.* (Yang et al., 2015) where significant differences were observed in the gut microbiota composition between 7 hypertensive patients and 10 healthy controls. Thus, we assumed that an expanded sample size of 30 untreated grade 1 HT subjects and their respective healthy controls (1:1 ratio) was enough to achieve significant power.

RESULTS

3.1. Clinical and lifestyle characteristics of the study participants

A total of 61 participants, including 29 HT and 32 NT participants, were enrolled in the study *(**Supplementary Fig. 1**)*. The general characteristics of the HT and NT subjects are shown in ***Table 1***. The HT subjects were older ($p < 0.001$) and presented a higher BMI ($p < 0.001$), waist circumference ($p < 0.001$) and fat mass ($p = 0.037$) than the NT subjects. Systolic BP and diastolic BP expressed as the mean ± SD were 153.1 ± 14.6 and 91.0 ± 8.8 mm Hg in HT subjects and 109.7 ± 7.1 and 65.7 ± 6.7 mm Hg in NT subjects, respectively. With respect to biochemical parameters, HT subjects showed higher fasting blood glucose ($p = 0.001$), total cholesterol ($p = 0.017$) and LDL cholesterol ($p = 0.002$) levels.

In relation to faecal SCFAs, HT subjects showed higher concentrations of acetate ($p = 0.004$), propionate ($p = 0.005$), butyrate ($p = 0.002$) and valerate ($p = 0.003$) than NT subjects (***Table 1***).

Daily energy and nutrient intakes were similar between the two groups, with the exception of total dietary fibre intake, which was greater in NT subjects ($p = 0.029$) (***Table 1***). In relation to habitual dietary intake, data reported via the FFQ revealed significantly greater daily mean intakes of processed meat, tubers, natural juices and coffee and a significantly lower intake of whole-grain in HT subjects than in NT (***Supplementary Table 1***). No differences were found in other lifestyle factors, such as physical activity.

3.2. Phenolic compound intake and its association with systolic and diastolic blood pressure

Daily PC intake expressed by dietary sources and by phenolic classes and subclasses, together with TPI and TPE in urine, is reported separately for the HT and NT groups in ***Table 2***. PCs from vegetables, legumes, fresh fruits, chocolate, coffee and wine, together with flavonoids and phenolic acids, were the major contributors to TPI in both groups. We found no significant differences between groups in terms of TPI and urinary TPE. However, HT subjects had a

significantly higher mean daily PC intake from non-alcoholic beverages, mainly coffee, and from alcoholic fermented beverages, such as wine and beer, and a lower PC intake from vegetables and whole-grain cereals than NT subjects ($p < 0.05$). With respect to phenolic classes, HT subjects consumed more hydroxycinnamic acids, alkylmethoxyphenols and methoxyphenols than NT subjects ($p < 0.05$) (***Table 2***).

To test the relationships between PCs that could contribute to the modulation of systolic and diastolic BP, we performed Pearson correlations in all populations (***Supplementary Table 2***). Based on these correlations, we subsequently conducted stepwise multiple linear regression analysis to assess major associations independent of age, dietary fibre intake and energy intake (***Table 3***). PCs from coffee and hydroxycinnamic acids showed a positive association with systolic BP (***Supplementary Table 2***), which was reinforced by multiple linear regression analysis (β = 0.278 and β = 0.310, respectively) ($p < 0.05$). Coffee and beer PCs and alkylmethoxyphenols and chalcones were also positively related to diastolic BP (β = 0.259, β = 0.259, β = 0.274 and β = 0.268, respectively) ($p < 0.05$) (***Table 3***).

3.3. Associations between phenolic compound intake and gut microbiota composition in hypertensive and normotensive subjects

With the LEfSe approach and an LDA score > 2, we detected a total of 67 ASV biomarkers that had significantly different abundances between HT and NT subjects (***Fig. 1-A***) (Calderón-Pérez et al., 2020). Of these ASVs, 14 were selected in the NT group, and 11 were selected in the HT group, showing the strongest Pearson correlations ($r \geq 0.30$) with PC dietary sources and classes (data not shown). Stepwise multiple linear regression analysis was conducted to corroborate major associations between PC dietary sources and bacterial the abundance of selected bacterial ASVs in both groups (***Table 4***). In addition, the most significant associations from the multiple linear regression analysis are shown in ***Fig. 1-B*** (NT) and ***1-C*** (HT).

In the NT group, several positive associations were observed regarding olive phenols. Specifically, PCs from olive fruits were positively associated with Ruminococcaceae UCG-010 (s408 and s1019), Christensenellaceae R-7 (s430) and *Bilophila wadsworthia* from the Desulfovibionaceae family (s1147) ($p < 0.001$) (***Table 4; Fig. 1-B***), whereas olive oil PC intake was associated with taxa s347 and s529 from the Ruminococcaceae and Rikenellaceae families, respectively ($p < 0.05$). Additionally, a strong positive association was found between some of the aforementioned taxa (s1019 and s430) and anthocyanins present in olive fruits ($p < 0.001$) (***Table 4***). Moreover, PCs from dried fruits (including dried apricots, plums, dates, figs and raisins) showed a positive correlation with the abundance of Ruminococcaceae NK4A214 (s127) ($p = 0.002$) and Christensenellaceae R-7 (s3173) ($p < 0.001$), both of which were strongly associated with hydroxybenzoic acids ($p < 0.001$) (***Table 4***). Other minor positive associations were found for PCs from fresh fruit, natural fruit juices and nuts with taxa s408, s1019, s1147, s430, s897 and s3187 in the Ruminococcaceae, Christensenellaceae, Tannerellaceae and Erysipelotrichaceae families ($p < 0.05$) (***Table 4; Fig. 1-B***). PC intake from tubers and lignans was associated with *Bacteroides xylanisolvens* (s230) ($p = 0.020$) from the Bacteroidetes phylum (***Table 4***). Conversely, coffee PCs and hydroxycinnamic acids presented negative associations with the abundances of *Faecalibacterium prausnitzii* (s372) and Christensenellaceae R-7 (s3173) ($p < 0.05$) (***Table 4***, ***Fig. 1-B***).

In the HT group, PCs from vegetables and lignans contributed positively to *Ruminococcaceae* taxa abundance (s728) ($p < 0.001$ and $p = 0.003$, respectively) (***Table 4***, ***Fig. 1-C***). In addition, PC intake from fresh fruit and dried fruits, and flavanones, the major phenolic class, showed positive associations with Ruminiclostridium 5 (s2600), *Streptococcus thermophilus* (s224) and Lachnospiraceae (s142) ($p < 0.05$), all from the Firmicutes phylum (***Table 4, Fig. 1-C***). PC intake from whole-grain cereals and flavanols was positively related to the abundance of *Bacteroides vulgatus* (s651) ($p < 0.001$) (***Fig. 1-C***). Other positive associations were found between PCs from wine and taxa s3042 in the Clostridiaceae family and between PC intake from berries and taxa s278 in the Muribaculaceae family ($p < 0.05$).

Coffee PCs, together with alkylmethoxyphenols and methoxyphenols, were identified as positive contributors to *Bacteroides plebeius* (s92) and *Bacteroides coprocola* (s16 and s41) abundance ($p < 0.05$) (***Fig. 1-C***), both defined in our previous work (Calderón-Pérez et al., 2020) as specific taxa with higher discriminatory power in the HT group.

3.4. Associations between phenolic compound intake and faecal and plasma short-chain fatty acids in hypertensive and normotensive subjects

Additionally, we tested for a possible association between PC intake and faecal and plasma SCFA concentrations by Pearson correlation (***Supplementary Tables 3 and 4***). Interestingly, in the HT group, coffee PCs and their main classes (hydroxycinnamic acids, alkylmethoxyphenols and methoxyphenols) were positively correlated with faecal levels of propionate ($p = 0.006$), acetate ($p = 0.023$), and valerate ($p = 0.003$), whereas dried fruit PCs also showed a positive correlation with faecal propionate and acetate in this group ($p = 0.049$ and 0.043, respectively). Additionally, positive correlations were found for chalcones and dihydrochalcones with faecal valerate in NT ($p = 0.003$ and 0.039, respectively). On the other hand, in the NT group, significant positive correlations were noted for fresh fruit and olive fruit PCs with plasma butyrate ($p = 0.042$ and 0.049, respectively) and valerate ($p = 0.001$ and 0.020, respectively) levels. Additionally, anthocyanins were positively correlated with plasma valerate ($p = 0.047$).

A schematic of major interrelationships between PC intake by food source and phenolic class, gut bacterial taxa at the family and ASV levels and SCFAs in both groups is provided in ***Fig. 2***.

DISCUSSION

The present study provides new insights into the interplay of dietary PCs with human gut microbiota composition and its derived metabolites in grade 1 hypertension. Our results confirm the hypothesis that specific dietary PCs could be precursors for the occurrence of particular

bacterial taxa that differs in the guts of non-treated grade 1 HT and NT subjects. We identified potential associations between PCs from plant-based foods, such as vegetables, fresh fruits, dried fruits, whole-grain cereals, tubers, olive oil, olive fruits, natural fruit juices and coffee, and the abundance of several bacterial taxa in the Ruminococcaceae, Bacteroidaceae, Lachnospiraceae, Christensenellaceae, Streptococcaceae, Erysipelotrichaceae and Clostridiaceae families in HT and NT subjects. In addition, we found interesting associations of PCs with faecal and plasma SCFA concentrations, which suggested a concomitant involvement of these colonic metabolites in the PC-mediated changes in gut bacterial composition.

A novel finding of our study was the multiple-way relationship among coffee PCs, discriminant bacterial taxa in HT and NT subjects, faecal SCFAs, and BP. Initially, in HT subjects, we observed positive associations between coffee PC intake and its chlorogenic acids, such as hydroxycinnamic acids and alkylmethoxyphenols, with particular *Bacteroides* spp. such as *B. plebeius* and *B. coprocola*. Previous studies reported an impact of moderate coffee consumption, defined as three cups of coffee/day, on particular members of the human colonic microbiota, principally species of *Bifidobacterium*, a genus with reputed beneficial effects for human health (Jaquet, Rochat, Moulin, Cavin, & Bibiloni, 2009). However, the impact of coffee intake on other dominant bacterial genera, such as *Bacteroides,* has not yet been reported, and whether the modulatory action of coffee on the gut microbiota can be attributed to PC content is still not clear. Moreover, our present data revealed positive associations of the hydroxycinnamic acid and alkylmethoxyphenol contents in coffee with systolic and diastolic BP in all subjects, even when multiple linear regression was applied. These results highlight the overall predictive capacity of coffee PCs for systolic and diastolic BP levels and, therefore, the potential involvement of specific coffee PCs in BP regulation. Additionally, in HT subjects, we found concomitant positive correlations of coffee PCs with faecal propionate, acetate and valerate levels ($r > 0.4$), which suggested the modulatory capacity of coffee PCs on SCFA-producing bacteria (Jaquet et al., 2009). Accordingly, we hypothesize that an increase in the growth of the species *B. plebeius* and *B. coprocola* induced by coffee PCs could contribute to the weakening

392 of the intestinal absorption of SCFAs, leading to higher faecal levels in HT subjects, as reported
393 in our previous study (Calderón-Pérez et al., 2020). Conversely, the negative associations found
394 in NT subjects for coffee PCs with *F. prausnitzii* and Christensenellaceae R-7, among the most
395 discriminant ASV biomarkers in NT subjects, indicated that a decrease in these specific bacteria
396 could precede the alteration of BP. Both *F. prausnitzii* and Christensenellaceae R-7, are well-
397 known butyrate producers in the human gut with potent anti-inflammatory effects (Zhang et al.,
398 2019), so depletion of these specific bacteria may have functional consequences on SCFA
399 production in the gut and, therefore, on the host's ability to repair epithelium and regulate
400 inflammation, a risk factor for hypertension (Yan et al., 2017). Although the habitual coffee
401 intake reported in the HT group (average of 60.11 mL/day) was significantly higher than that in
402 the NT group (average of 45.30 mL/day), the observed opposite relationships between coffee
403 PCs and discriminant ASV biomarkers in HT and NT subjects may not be explained only by the
404 amount ingested. Therefore, we hypothesize that specific coffee PCs could also modulate the
405 gut microbiome in different ways depending on the stage of disease, either increasing particular
406 bacterial taxa in HT subjects, or decreasing beneficial bacteria in NT subjects. Thus, given the
407 overall negative impact of coffee PCs on the gut microbiome in both groups, our results suggest
408 a new pattern in which coffee PCs could precede the rise in BP.

409 Another remarkable finding in the present study was the strong positive associations
410 reported in NT subjects between PCs from olive fruits, including those in the anthocyanin class,
411 discriminant ASV biomarkers, such as Ruminococcaceae UCG-010 and Christensenellaceae R-
412 7, and plasma butyrate and valerate. These interrelationships could reflect a favoured butyrate
413 and valerate production and absorption in the colon of NT group mediated by olive fruit PCs.
414 Commercial table olive fruits are the most widely consumed fermented food in Mediterranean
415 countries and are particularly rich in hydroxytyrosol which is found at a high abundance in
416 Greek-style natural black olives and Spanish-style green olives (approximately 250 - 760 mg
417 hydroxytyrosol/kg) (Kiritsakis & Shahidi, 2017). In our study, the total intake of PCs from olive
418 fruits ranged from 13.95 mg/day in HT subjects to 24.79 mg/day in NT subjects. This intake

was specifically provided by the high average consumption of olive fruits in NT (25 g/day), which was about three times over the median consumption of 7 g/capita/day reported in the Spanish population. In addition, the negative correlations found in our previous work for Christensenellaceae R-7 with systolic and diastolic BP (Calderón-Pérez et al., 2020), highlight the possible interplay of olive fruit PCs with the gut microbiota and reinforce the role of these compounds in the prevention of hypertension.

In addition to olive fruit PCs, positive relations were detected in NT subjects between PCs from olive oil, including flavones, the main flavonoids present in virgin olive oil, and specific taxa in the Ruminoccocaceae and Rikenellaceae families. Recently, the intake of extra-virgin olive oil was reported to be associated with greater promotion of gut bacterial diversity, mediated by an increase in the Firmicutes/Bacteroidetes ratio, in spontaneously hypertensive rats (Hidalgo et al., 2018). A similar effect has also been found in humans, in which PC-enriched olive oils are able to induce antioxidant activities and, consequently, decrease BP (Martin-Pelaez et al., 2017; Moreno-Luna et al., 2012).

Secondarily, from our data, positive associations between PCs from fresh fruits and vegetables, including flavanones and lignans, and several species of the Ruminococcaceae family were observed in both HT and NT subjects. The Ruminococcaceae family is mainly composed of numerous species able to act as primary degraders of insoluble plant cell walls in the lower gastrointestinal tract (Flint, Bayer, Rincon, Lamed, & White, 2008). Members of this family also consume hydrogen and produce acetate that can be utilized by other butyrate-producing xylanolytic species (Chassard & Bernalier- Donadille, 2006). Similarly, the positive associations found between PCs from whole-grain cereals and *B. vulgatus* abundance in HT subjects and between PCs from tubers and *B. xylanisolvens* abundance in NT subjects support the role of these bacterial strains in the colonic catabolism of soluble polysaccharides, such as amylose, amylopectin and xylans, contained in whole grains and tubers. All these positive associations of whole-grain cereals and tubers with *Bacteroides* spp. abundance remained significant after adjusting for dietary fibre intake, suggesting an independent contribution of

446 dietary PCs contained in the same polysaccharide-rich food matrix to the growth of *Bacteroides*
447 spp. Moreover, the weak positive correlation found between whole-grain cereal PCs and faecal
448 acetate in HT subjects reinforces their interplay with the gut microbiota.

449 On the other hand, in NT subjects, we observed significant positive relationships of PCs
450 from dried fruits and their main phenolic classes, such as phenolic acids and flavonoids, with
451 Ruminococcaceae NK4A214 and Christensenellaceae R-7, which are among the most
452 discriminant bacteria in NT subjects. In contrast, in HT subjects, dried fruit PCs were
453 significantly positively related to *S. thermophiles*, pointing out the role of dried fruit PCs in gut
454 microbial modulation, probably due to their coexistence with lactic acid probiotic bacteria
455 (Kumar Singh et al., 2019). Additionally, the positive correlations reported in HT subjects for
456 dried fruit PCs with faecal propionate and acetate levels led us to propose dried fruit PCs as
457 possible precursors to SCFAs synthesis with a metabolic fate mediated by the gut microbial
458 community. Nevertheless, given the low daily intake of dried fruits in both groups
459 (approximately 11 g/day), we cannot verify their impact on SCFAs synthesis, so more studies
460 are needed to confirm the possible mechanism linking dried fruit sources of PCs with gut
461 bacteria involved in SCFAs synthesis and the connection of this relationship with hypertension.

462 Interestingly, in the NT group, we also found PCs from natural fruit juices, mainly citrus
463 juices, as possible contributors to the abundance of genus *Dielma* in the Erysipelotrichaceae
464 family, which is able to acidify glucose and other dietary sugars and has been recently
465 associated with inflammation-related gastrointestinal diseases and metabolic disorders in
466 humans (Kaakoush, 2015). Moreover, a positive correlation was found between natural fruit
467 juice PCs, particularly flavanones, and systolic BP in all subjects. Given previous evidence
468 concerning the beneficial effect of citrus juice PCs on BP (Valls et al., 2020), we speculate that
469 the increase in the abundance of the *Dielma* genus after citrus juice intake would be strongly
470 related to the sugar content of the juice. In this sense, an increase in this particular bacterial
471 genus could induce a gut inflammatory response being as a precursor to the hypertensive state
472 (Yan et al., 2017).

473 It should be noted that the correlations found in NT subjects between the nuts, dried
474 fruits, natural juices PCs, and hydroxybenzoic acids class with Ruminococcaceae and
475 Christensenellaceae families could explain a protective effect of ellagitannins on BP (Wang et
476 al., 2018), as they appear to be included in this phenol class on the Phenol Explorer data base.
477 However, given the lack of correlations with SCFAs, we cannot suggest that the supposed
478 ellagitannin hypotensive effect is mediated by SCFA action.

479 In all participants, positive correlations were observed for fermented alcoholic beverage
480 PCs, such as wine and beer PCs, with systolic and diastolic BP. This finding reinforces the
481 observed dose-response alcohol-induced increases in BP levels of approximately 5 to 10 mm Hg
482 after regular alcohol consumption at a threshold of three drinks per day (30 g of ethanol/day),
483 with systolic increases nearly always greater than diastolic increases (Husain, Ansari, & Ferder,
484 2014). We specifically noted a positive contribution of beer PCs to diastolic BP levels after
485 linear regression. Nevertheless, given the high ethanol intake in the HT group, at approximately
486 11 g/day, we suppose that the positive association with diastolic BP was related to the alcohol
487 content in beer. Moreover, in a previous dietary study (Chiva-Blanch et al., 2015), after 4
488 weeks of moderate administration of a non-alcoholic beer rich in PCs, a decrease in BP levels
489 and improvements in other cardiovascular risk factors were observed compared to those under
490 the administration of a standard alcoholic beer, suggesting an alcohol-independent effect on BP
491 of the PCs contained in the non-alcoholic fraction of the beer.

492 In the present work, the fact that no significant associations were reported for TPI and
493 urinary TPE, an objective marker of exposure to PC intake, with BP values could be attributed
494 to both the small sample size and the high inter-individual variations in gut microbiota affecting
495 PC intrinsic activity, metabolization and absorption through the gut barrier (Tomas-Barberan et
496 al., 2016).

497 The relationship between the gut microbiota and BP is complex, and current evidence is
498 controversial in relation to the mechanisms involved. Most of the hypotheses point to dysbiosis
499 as a precipitant factor for BP alteration (Al Khodor, Reichert, & Shatat, 2017). However, the

observed interplays in the present study of plant-derived dietary PCs consumed voluntarily could explain their role as precursors for gut bacterial modifications and concomitant associations with faecal and plasma SCFA levels. Our findings extend previous knowledge on the correlation between the gut microbiota and hypertension in humans (Calderón-Pérez et al., 2020) and provide a new way in which dietary PCs could induce changes in gut bacterial composition to affect SCFA production and absorption. Furthermore, the opposite trends observed in associations among faecal and plasma SCFAs with particular dietary PCs in HT and NT subjects could explain different absorption modes depending on the hypertension stage. In addition, it will be helpful to better understand the dynamics of SCFAs in the human body (Yamamura et al., 2020). Specifically, fresh fruit and olive fruit PCs seem to favour SCFA absorption, leading to higher plasma concentrations of butyrate and valerate in NT, while coffee PCs are related to higher faecal SCFA levels in HT, suggesting a lower efficiency in their intestinal absorption, which could be a causative factor for the higher BP, as hypothesized in our previous work (Calderón-Pérez et al., 2020).

One distinctive point of the present study was the application of suitable tools to report PC intake, such as the FFQ and 3-day dietary records, which provided a complete picture of participants' habitual dietary PC intake. Furthermore, the large dataset on diet composition allowed us to control for potential confounders such as fibre intake. Nonetheless, the study had some limitations. The small sample size and the heterogeneity in clinical variables may have hampered the detection of other significant associations. Additionally, the cross-sectional design may have limited the establishment of causal relationships, and the existence of unmeasured human factors, such as inter-individual variability in gut microbiota composition or different clinical parameters at baseline, such as BMI, may have influenced the results. However, the associations between microbial composition and BP remained significant after adjustment for BMI among other variables, as reported in our previous work (Calderón-Pérez et al., 2020), indicating that the differences were intrinsically related to the hypertensive condition. Also PC bioavailability might be influenced by uncontrolled external factors, such as food processing

and reciprocal interactions with the gut microbiota, affecting the intestinal biotransformation of PCs into their metabolites (Ozdal et al., 2016). It would be of great interest in the future to design in vitro culture models simulating lower-intestinal-tract conditions in order to test whether the reported associations indicate an effect or reflect a possible mechanism of action of PCs from plants.

CONCLUSION

In summary, plant-derived dietary PCs appear to be associated with discriminant gut bacterial taxa and faecal metabolites in the hypertensive and healthy states. The most remarkable findings are the multiple-way positive relationships found in HT subjects among coffee PCs, systolic and diastolic BP, faecal SCFAs and the most discriminant bacterial taxa, *B. plebeius* and *B. coprocola*, and the negative associations in NT subjects between coffee PCs and the beneficial bacteria *F. prausnitzii* and Christensenellaceae R-7. These results suggest a new pattern in which coffee PCs could precede the rise in BP. Additionally, olive fruit PCs were positively associated with Ruminococcaceae UCG-010, Christensenellaceae R-7 and plasma SCFAs in NT subjects, reinforcing the previously reported role of this PCs in the prevention of hypertension.

Finally, the overall results suggested the potential role of specific food phenolic sources as precursors for human gut microbiome modulation involved in hypertension pathogenesis or its prevention. However, further studies on the causal relationship between plant-derived PCs and the gut microbiome will lead to a better understanding of their mutual interaction effects on BP.

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Figure Captions

Figure 1 | Relationships between phenolic compound intake from major food sources and gut microbial signatures in hypertensive (HT) and normotensive (NT) subjects. (A) LefSe analysis of ASVs between NT (green) and HT (red) subjects. LDA scores (log10) for the most prevalent ASVs in the NT group are represented on a positive scale, whereas negative LDA scores indicate enriched ASVs in the HT group. **(B)** Multiple linear regression analyses representing major PC food sources associated with the absolute abundance of faecal microbial ASVs in the NT group (green) and **(C)** HT group (red); R^2, coefficient of multiple determination; β, standardized regression coefficient; [a, b, c] indexes representing regressions for individual food sources in food categories; *p value ≤ 0.05; **p value ≤ 0.001.

Figure 2 | Schematic of the major interrelationships between PC intake by food sources and phenolic classes, microbial taxa (expressed at the family and ASV levels) and SCFAs in hypertensive (HT) and normotensive (NT) subjects. The colour key in green (NT) and red (HT) represents the strength of the relation. Color arrows represent the relation among PCs and microbial taxa. Doted arrows represent the relation among PCs and faecal or plasma SCFA levels.

Supplementary files

Supplementary Table 1 | Mean daily intake according to food groups.

Supplementary Table 2 | Correlations of total phenolic intake, total polyphenol excretion in urine and phenolic compounds from food sources and phenolic classes with systolic and diastolic blood pressure in all subjects.

Supplementary Table 3 | Correlations of phenolic compounds from food sources and phenolic classes with faecal short-chain fatty acid concentrations in hypertensive and normotensive subjects.

Supplementary Table 4 | Correlations of phenolic compounds from food sources and phenolic classes with plasma short-chain fatty acid concentrations in hypertensive and normotensive subjects.

Supplementary Figure 1| Flowchart based on the STROBE (Strengthening the Reporting of Observational Studies in Epidemiology) statement.

763 **TABLE 1 | Baseline characteristics of study participants**

Variables	Hypertensive	Normotensive	P-value
N	29	32	
Age, *y*	53.7 ± 9.6	41.1 ± 9.1	<0.001
Gender, *(F/M)*	(10/19)	(16/16)	0.301
Anthropometry			
Weight, *kg*	75.3 ± 9.3	68.9 ± 10.8	0.017
BMI, kg/m^2	26.2 ± 2.5	23.8 ± 2.7	<0.001
Fat mass, *%*	26.6 ± 7.9	22.1 ± 7.8	0.037
Waist circumference, *cm*	94.4 ± 8.3	84.0 ± 9.0	<0.001
Blood chemistry *(mg/dL)*			
Total cholesterol	199.6 ± 43.9	181.7 ± 34.7	0.017
LDL cholesterol	123.7 ± 21.3	100.7 ± 33.2	0.002
HDL cholesterol	62.6 ± 14.0	64.9 ± 18.0	0.580
Triglycerides	97.3 ± 38.8	80.7 ± 42.6	0.067
FBG	91.2 ± 11.3	81.1 ± 7.5	0.001
Blood pressure *(mm Hg)*			
Systolic BP	153.1 ± 14.6	109.7 ± 7.1	<0.001
Diastolic BP	91.0 ± 8.8	65.7 ± 6.7	<0.001
Faecal SCFAs concentration *(mg/g feces)*			
Acetate	22.11 ± 9.70	15.05 ± 8.74	0.004
Propionate	7.78 ± 3.38	5.26 ± 3.41	0.005
Butyrate	8.80 ± 4.71	5.62 ± 2.93	0.002
Valerate	1.57 ± 0.94	1.04 ± 0.50	0.003
Physical activity *(%)*			
Inactive	6.9	0.0	
Very low activity	10.3	10.0	
Low activity	10.3	6.7	0.729
Moderate activity	20.7	20.0	
High activity	51.7	63.3	
Diet			
Energy intake, *Kcal/day*	2089.74 ± 543.43	2208.23 ± 654.06	0.452
Protein, *g/day*	88.28 ± 21.28	94.10 ± 29.87	0.395
(% DEI)	*(17.2)*	*(17.5)*	0.741
Total carbohydrates, *g/day*	197.03 ± 61.41	212.65 ± 60.90	0.328
(% DEI)	*(38)*	*(39.3)*	0.441
Complex carbohydrates, *g/day*	110.35 ± 42.29	116.00 ± 27.31	0.548
(% DEI)	*(21)*	*(21.8)*	0.917
Simple carbohydrates, *g/day*	87.87 ± 29.64	96.62 ± 40.38	0.767
(% DEI)	*(17)*	*(17.6)*	0.611

Total fat, *g/day*	96.95 ± 31.67	102.70 ± 41.93	0.556
(% DEI)	*(41.3)*	*(40.8)*	0.782
SFA, *g/day*	27.54 ± 10.54	27.75 ± 12.47	0.944
(% DEI)	*(11.5)*	*(11.4)*	0.843
MUFA, *g/day*	45.40 ± 15.63	49.67 ± 21.92	0.395
(% DEI)	*(19.4)*	*(19.7)*	0.779
PUFA, *g/day*	15.92 ± 7.29	17.22 ± 8.58	0.563
(% DEI)	*(6.8)*	*(6.9)*	0.711
Total cholesterol, *mg/day*	328.25 ± 130.77	358.22 ± 187.89	0.482
Dietary fibre, *g/day*	20.95 ± 9.90	25.90 ± 11.63	0.029
Ethanol, *g/day*	11.37 ± 13.04	7.76 ± 10.11	0.131

Data expressed as mean ± standard deviation or percentage. Abbreviations: BP, blood pressure; BMI, body mass index; FBG, fasting blood glucose; LDL. low density lipoproteins; HDL, high density lipoproteins; SFA, saturated fatty acids; MUFA, monounsaturated fatty acid; PUFA, polyunsaturated fatty acid. % DEI, mean percentage of daily energy intake. P-value for gender and physical activity was calculated by Fisher's exact test. P-value for the rest of the variables was calculated by Student's t-test and Mann-Whitney U test.

TABLE 2 | Daily intake of phenolic compounds expressed by major dietary food sources and by phenolic classes and subclasses in hypertensive and normotensive subjects

	Hypertensive (n = 29)	Normotensive (n = 32)	P-value
PC intake by dietary source (mg/day)			
Tubers	57.45 ± 33.03	43.95 ± 37.52	0.147
Cereals	92.07 ± 109.05	113.98 ± 142.13	0.224
Whole-grain cereals	29.08 ± 45.23	44.91 ± 41.71	**0.012**
Vegetables	244.56 ± 253.40	380.66 ± 362.88	**0.042**
Legumes	168.36 ± 431,38	248.23 ± 464.33	0.352
Fresh fruits	319.09 ± 249.22	417.63 ± 365.33	0.237
Berries	38.80 ± 54.72	42.59 ± 81.43	0.525
Dried fruit	30.40 ± 130.10	27.84 ± 69.38	0.631
Nuts	98.33 ± 135.08	112.39 ± 179.49	0.812
Olive fruit	13.95 ± 21.71	24.79 ± 53.68	0.928
Oils	6.88 ± 3.15	7.49 ± 4.12	0.527
Olive oil	6.44 ± 3.48	7.68 ± 3.79	0.239
Sweets	115.35 ± 149.24	222.83 ± 335.53	0.440
Chocolate	113.82 ± 149.13	220.16 ± 333.91	0.300
Non-alcoholic beverages	254.17 ± 151.61	159.45 ± 150.98	**0.006**
Natural fruit juice	22.90 ± 38.38	22.60 ± 52.34	0.577
Coffee	203.45 ± 150.12	115.52 ± 128.44	**0.013**
Tea	24.74 ± 51.04	29.23 ± 82.87	0.889
Alcoholic beverages	163.92 ± 169.88	96.32 ± 151.14	**0.040**
Wine	127.51 ± 153.42	74.10 ± 132.40	0.074
Beer	20.30 ± 31.34	32.58 ± 40.27	0.120
Seasonings	6.85 ± 15.01	8.63 ± 23.27	0.683
PC intake by class and subclass (mg/day)			
Total flavonoids	224.32 ± 137.94	209.68 ± 117.86	0.824
Anthocyanins	30.11 ± 28.05	20.82 ± 31.77	0.112
Chalcones	0.002 ± 0.005	0.0007 ± 0.011	0.349
Dihydrochalcones	0.88 ± 1.73	2.10 ± 3.59	0.369
Dihydroflavonols	3.20 ± 3.87	1.84 ± 3.34	0.074
Flavanols	82.66 ± 70.08	88.42 ± 73.08	0.614
Flavanones	51.66 ± 58.11	30.08 ± 38.43	0.208
Flavones	15.18 ± 11.09	23.03 ± 19.35	0.159
Flavonols	28.66 ± 18.33	32.18 ± 21.25	0.604
Isoflavonoids	11.95 ± 31.55	11.18 ± 21.52	0.233

Total phenolic acids	319.66 ± 120.44	262.83 ± 131.28	0.088
Hydroxibenzoic acids	38.72 ± 43.89	38.88 ± 49.51	0.882
Hydroxycinnamic acids	277.35 ± 115.29	220.25 ± 104.95	**0.042**
Stilbenes	2.07 ± 2.43	1.21 ± 2.09	0.120
Lignans	45.51 ± 65.32	47.37 ± 41.08	0.279
Other polyphenols	61.75 ± 48.72	67.82 ± 46.51	0.424
Alkylmethoxyphenols	1.03 ± 0.63	0.61 ± 0.61	**0.012**
Alkylphenols	21.87 ± 41.09	29.33 ± 40.34	0.106
Curcuminoids	0.77 ± 1.80	0.62 ± 2.18	0.373
Methoxyphenols	0.12 ± 0.08	0.07 ± 0.07	**0.010**
Tyrosols	34.16 ± 23.28	33.98 ± 29.84	0.625
Total polyphenol intake	1616.49 ± 768.10	1634.91 ± 641.37	0.922
Total polyphenol excretion	156.74 ± 43.41	154.30 ± 33.02	0.822

772 Data expressed as mean ± standard deviation. Total polyphenol intake from PC dietary food sources and classes
773 expressed as mg/day. Total polyphenol excretion expressed as mg GAE/g creatinine. P-value was calculated by
774 Student's t-test and Mann-Whitney U test.

TABLE 3 | Multiple stepwise linear regression results for associations of phenolic compound dietary sources and *classes* with systolic and diastolic blood pressure levels (mm Hg) for all subjects

	Systolic Blood Pressure				Diastolic Blood Pressure		
Variables	**R^2**	***β***	***p***	**Variables**	**R^2**	***β***	***p***
Coffee	0.077	0.278	0.032*	Coffee	0.153	0.259	0.040*
Hydroxycinnamic	0.096	0.310	0.016*	Beer		0.259	0.040*
Acids				*Alkylmethoxyphenols*	0.187	0.274	0.032*
				Chalcones		0.268	0.035*

R^2, coefficient of multiple determination; percentage of the variance in the dependent variable explained by the independent variables in the model.

β, standardized regression coefficient.

p, two-sided test of significance.* *p* value ≤ 0.05

Independent variables included in the model: only phenolic compound significant correlations from Pearson analysis.

Analysis was adjusted for age, total fibre and energy intake. Only statistically significant results are shown.

TABLE 4 | Multivariate linear regression analysis with differential faecal ASV biomarkers in normotensive and hypertensive subjects and the intake of phenolic compounds (mg/day) from major dietary sources and classes

ASV	Bacterial group	Phenolic intake by dietary source				Phenolic intake by class			
		Variables	R^2	β	p	Variables	R^2	β	p
Normotensive (n=32)									
s372[†]	Ruminococcaceae, *Faecalibacterium prausnitzii*	Coffee	0.263	-0.338	0.047	Hydroxycinnamic acids	0.149	-0.385	0.029
s127[†]	Ruminococcaceae NK4A214	Dried fruit	0.285	0.534	0.002	Hydroxybenzoic acids	0.393	0.627	<0.001*
s230	Bacteroidaceae, *Bacteroides xylanisolvens*	Tubers	0.172	0.415	0.020	Lignans	0.168	0.410	0.020
s408	Ruminococcaceae UCG-010	Olive fruit	0.793	0.624	<0.001*	Anthocyanins	0.630	0.366	0.032
		Nuts		0.346	0.005	Hydroxybenzoic acids		0.490	0.005
s529	Rikenellaceae_RC9_gut_group	Olive oil	0.187	0.432	0.015	Flavones	0.178	0.422	0.016
s897	Tannerellaceae, *Parabacteroides distasonis*	Natural juices	0.444	0.326	0.032	Flavanones	0.157	0.396	0.025
		Fresh fruits		0.312	0.039				
s347	Ruminococcaceae	Olive oil	0.170	0.413	0.021	Flavones	0.161	0.401	0.023
S1019	Ruminococcaceae UCG-010	Olive fruit	0.837	0.553	<0.001*	Anthocyanins	0.766	0.932	<0.001*
		Fresh fruits		0.407	<0.001*	Stilbenes		-0.385	<0.001*
		Olive oil		-0.239	0.006				
s1147	Desulfovibrionaceae**,** *Bilophila wadsworthia*	Olive fruit	0.842	0.442	<0.001*	Anthocyanins	0.821	0.735	<0.001*
		Fresh fruits		0.337	0.002	Hydroxybenzoketones		0.425	<0.001*
		Nuts		0.285	0.011				
s430	Christensenellaceae R-7	Olive fruit	0.799	0.477	0.001*	Anthocyanins	0.765	0.927	<0.001*
		Fresh fruits		0.279	0.019	Stilbenes		-0.426	<0.001*
		Nuts		0.276	0.027				

s1683	Ruminococcaceae, *Butyricicoccus faecihominis*	Tea	0.525	0.724	<0.001*	Flavonols	0.177	0.421	0.017
s1296	Clostridiales vadin	Olive fruit	0.226	0.475	0.007	Anthocyanins	0.173	0.416	0.018
s3173	Christensenellaceae R-7	Dried fruit	0.762	0.825	<0.001*	Hydroxybenzoic acids	0.678	0.603	<0.001*
		Coffee		-0.259	0.009	Tyrosols		0.391	0.002
s3187	Erysipelotrichaceae, *Dielma*	Natural juices	0.550	0.742	<0.001*	Hydroxybenzoic acids	0.175	0.418	0.017
Hypertensive (n=29)									
s3042	Clostridiales, Family_XIII	Wine	0.150	0.387	0.042	Stilbenes	0.156	0.395	0.037
s2600	Ruminococcaceae, Ruminiclostridium 5	Fresh fruits	0.483	0.466	0.005	Flavanones	0.510	0.714	<0.001*
		Wine		0.403	0.012				
s728	Ruminococcaceae	Vegetables	0.451	0.672	<0.001*	Lignans	0.289	0.538	0.003
s651	Bacteroidaceae, *Bacteroides vulgatus*	Whole grain cereals	0.416	0.645	<0.001*	Flavanols	0.309	0.555	0.002
s740	Lachnospiraceae, *Lachnoclostridium pacaense*	Coffee	0.148	0.385	0.043	Methoxyphenols	0.184	0.429	0.023
s224	Streptococcaceae, *Streptococcus thermophilus*	Dried fruit	0.795	0.892	<0.001*	Isoflavonoids	0.249	0.499	0.007
s278	Muribaculaceae	Berries	0.159	0.399	0.035	Anthocyanins	0.162	0.403	0.033
s142	Lachnospiraceae	Fresh fruits	0.401	0.516	0.003	Flavanones	0.288	0.537	0.003
		Olive oil		-0.490	0.005				
s92†	Bacteroidaceae, *Bacteroides plebeius*	Coffee	0.179	0.423	0.025	Alkymethoxyphenols	0.227	0.476	0.010
s16†	Bacteroidaceae, *Bacteroides coprocola*	Coffee	0.199	0.446	0.017	Methoxyphenols	0.395	0.356	0.033
s41†	Bacteroidaceae, *Bacteroides coprocola*	Coffee	0.182	0.427	0.023	Methoxyphenols	0.169	0.411	0.030

784 R^2, coefficient of multiple determination; β, standardized regression coefficient; p, two-sided test of significance.

785 *p value ≤ 0.001

786 †ASVs with the highest discriminant power in NT and HT group (LDA score > 2.9).

787 Values from multiple stepwise regression analysis. Analysis adjusted by age, daily energy intake and total fibre intake.

788 Only statistically significant results are shown.

789

Highlights

- Dietary phenolic compounds (PCs) interplay with the gut microbiota in the hypertensive state.
- Certain PCs show multiple-way relationships with microbiota and short chain fatty acids.
- Coffee PCs correlate positively with the Bacteroides genus and faecal SCFAs in hypertense.
- Olive fruit PCs correlate positively with SCFA-producers and plasma SCFA in normotense.
- Different SCFAs absorption modes could be explained by the interplays with PCs.
- PCs could precede hypertension or its prevention through the gut microbiota modulation.

Figure(s)

Figure 1

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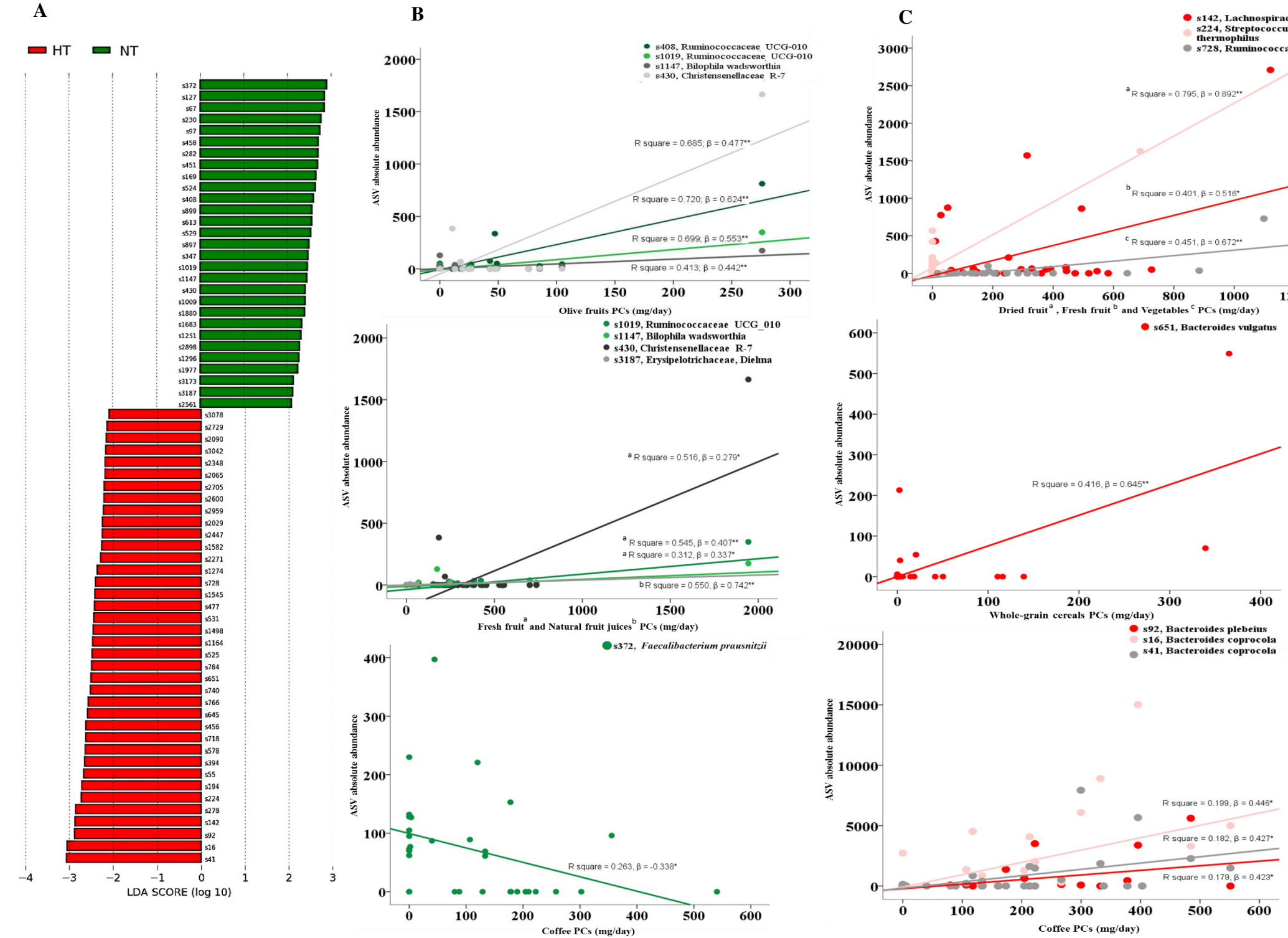

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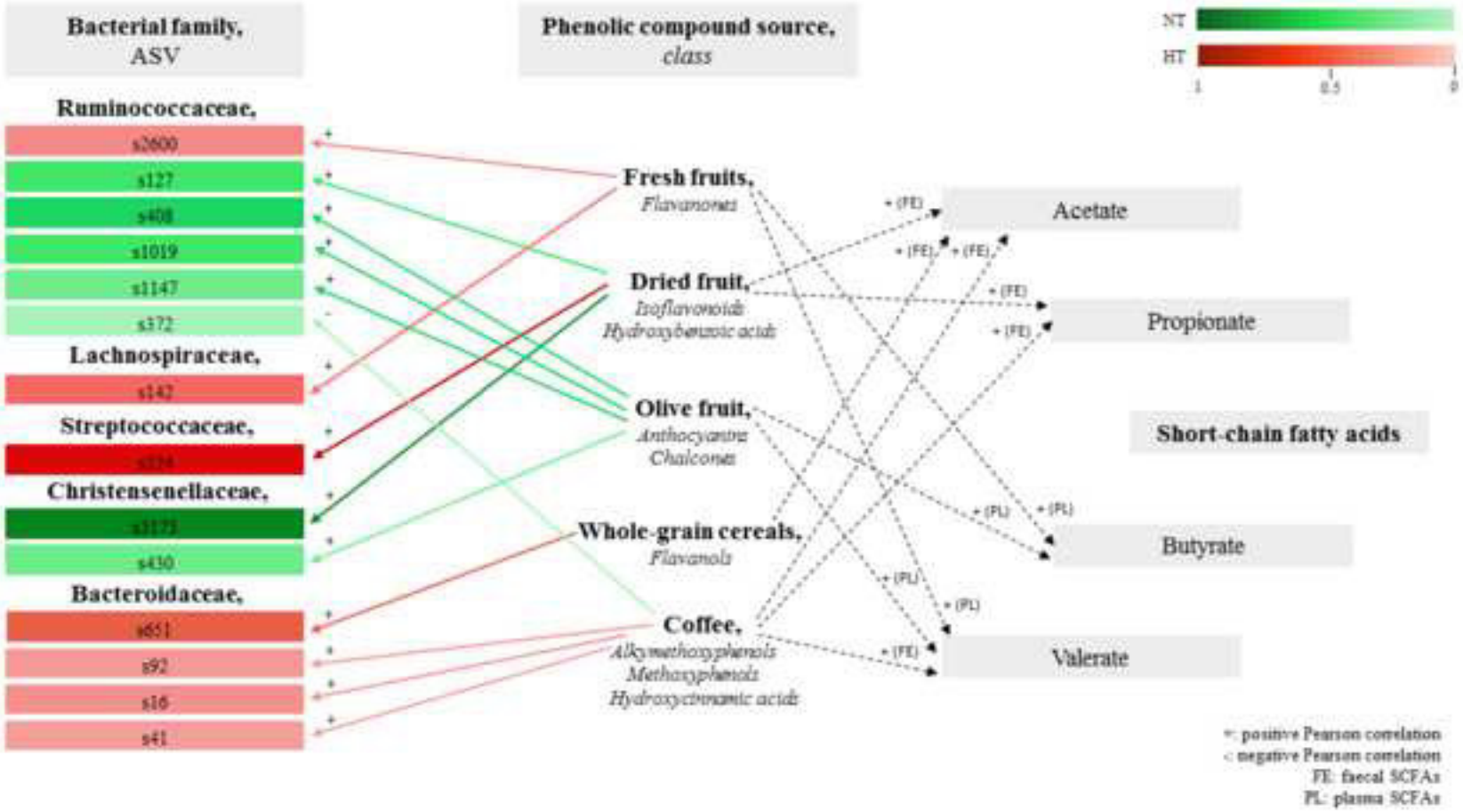

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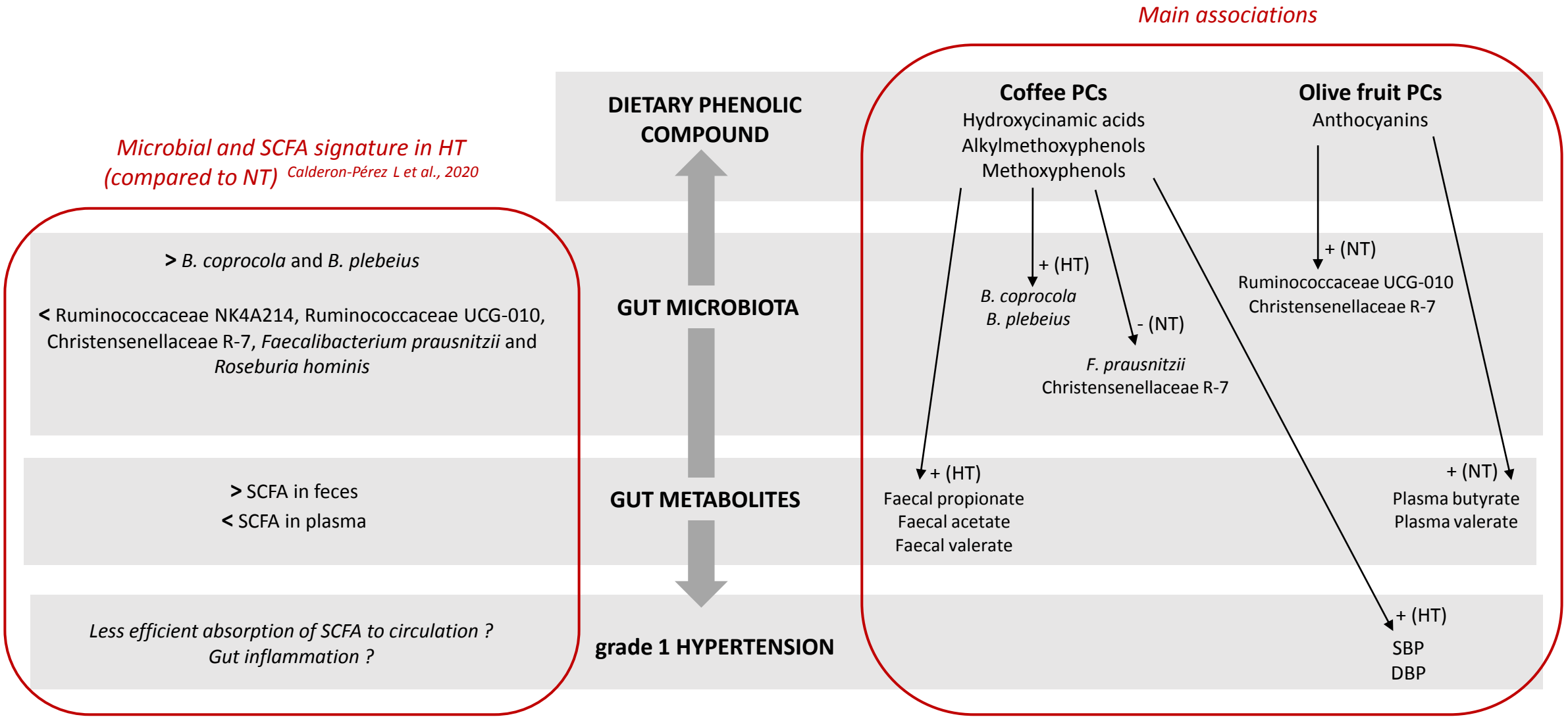


Supplementary Table 1 | Mean daily intake according to food groups.

Food group intake *(g/day)*	Hypertensive (n=27)	Normotensive (n=32)	P-value
Dairy products	240.17 ± 150.38	329.26 ± 295.26	0.513
Whole dairy	65.62 ± 59.35	103.40 ± 154.12	0.610
Semi-skimmed dairy	86.28 ± 112.30	105.46 ± 151.72	0.766
Skimmed dairy	76.28 ± 115.24	85.78 ± 112.70	0.926
Eggs	25.25 ± 12.80	27.11 ± 28.71	0.537
Meats	193.07 ± 122.15	148.55 ± 106.27	0.140
Red meat	64.95 ± 44.70	53.30 ± 50.40	0.150
White meat	82.09 ± 81.11	65.71 ± 78.23	0.125
Processed meat	46.03 ± 28.52	29.54 ± 25.13	0.016
Fish and seafood	103.98 ± 57.60	107.83 ± 71.10	0.976
Lean fish	37.40 ± 26.50	36.46 ± 27.75	0.691
Fatty fish	42.86 ± 23.09	48.13 ± 43.72	0.517
Shellfish	23.72 ± 18.93	23.20 ± 16.14	0.770
Vegetables	467.05 ± 261.35	433.03 ± 217.74	0.533
Tubers	66.22 ± 38.16	40.37 ± 32.58	0.005
Fresh fruits	399.99 ± 239.93	403.71 ± 338.54	0.420
Dried fruit	10.81 ± 22.90	11.07 ± 24.17	0.713
Nuts	12.18 ± 17.51	6.61 ± 7.35	0.132
Olive fruit	12.57 ± 13.19	24.77 ± 27.83	0.107
Legumes	27.72 ± 8.78	25.45 ± 11.92	0.417
Cereals	213.50 ± 176.76	238.93 ± 188.35	0.553
Refined cereals	121.42 ± 86.69	104.10 ± 85.13	0.312
Whole-grain cereals	26.51 ± 42.88	50.63 ± 68.66	0.034
Oils and fats	23.13 ± 14.96	27.70 ± 21.99	0.503
Olive oil	20.50 ± 14.61	25.30 ± 22.18	0.374
Sunflower oil	0.36 ± 0.89	1.17 ± 2.64	0.511
Butter	2.03 ± 4.58	1.21 ± 1.73	0.407
Pastries	14.52 ± 12.60	20.39 ± 19.76	0.583
Chocolate	7.03 ± 7.95	6.36 ± 8.40	0.590

Pre-cooked food	53.70 ± 40.00	39.53 ± 23.82	0.191
Non-alcoholic beverages	235.82 ± 172.11	144.79 ± 136.20	0.004
Sugary carbonated drinks	28.85 ± 45.34	9.46 ± 16.31	0.190
Soft drinks	12.66 ± 34.15	22.70 ± 56.08	0.713
Commercial juices	30.63 ± 96.65	18.83 ± 50.27	0.574
Natural juices	85.32 ± 81.47	27.88 ± 45.24	0.008
Coffee	60.11 ± 45.30	44.70 ± 57.45	0.033
Tea	18.23 ± 33.01	21.18 ± 36.97	0.906
Alcoholic beverages	160.80 ± 145.21	131.70 ± 162.22	0.108
Wine	57.60 ± 81.08	41.35 ± 61.76	0.278
Beer	101.90 ± 102.93	88.83 ± 112.07	0.213
Distilled spirits	1.28 ± 2.56	1.52 ± 4.05	0.756

Data expressed as mean ± standard deviation. Daily mean intake by food groups was estimated from adapted Food Frequency Questionnaires performed in 59 subjects. P-value estimated by Student's t-test and Mann-Whitney U test. The level of statistical significance was set at $p<0.05$.

Supplementary Table 2 | Correlations of total phenolic intake, total polyphenol excretion in urine and phenolic compounds from food sources and phenolic classes with systolic and diastolic blood pressure in all subjects

	Systolic Blood Pressure (mmHg)		Diastolic Blood Pressure (mmHg)	
	r	**p**	**r**	**p**
TPI, *mg/day*	-0.046	0.365	-0.058	0.331
TPE, *GAE/g creatinine*	0.183	0.083	0.166	0.103
Age, *y*	0.567	**0.000**	0.528	**0.000**
Dietary fiber, *g/day*	-0.070	0.298	-0.110	0.202
Energy intake, *kcal/day*	-0.044	0.369	-0.094	0.237
Phenolic compound dietary source, m*g/day*				
Tubers	0.162	0.108	0.143	0.138
Cereals	-0.070	0.296	-0.028	0.417
Whole-grain cereals	-0.119	0.189	-0.138	0.153
Vegetables	-0.008	0.476	-0.148	0.130
Legumes	-0.066	0.308	-0.008	0.476
Fresh fruits	-0.099	0.227	-0.155	0.119
Berries	0.056	0.336	0.029	0.414
Dried fruit	-0.018	0.445	0.016	0.453
Nuts	-0.011	0.467	-0.030	0.411
Olive fruit	-0.104	0.216	-0.080	0.273
Oils	0.005	0.486	-0.148	0.129
Olive oil	0.004	0.487	-0.147	0.131
Chocolate	-0.198	0.064	-0.175	0.091
Non-alcoholic beverages	0.300	**0.010**	0.344	**0.004**
Natural fruit juices	0.227	**0.040**	0.125	0.171
Coffee	0.278	**0.016**	0.295	**0.011**
Tea	0.005	0.486	0.048	0.359
Alcoholic beverages	0.236	**0.035**	0.259	**0.023**
Wine	0.231	**0.038**	0.218	**0.047**
Beer	0.135	0.152	0.295	**0.011**
Seasonings	-0.044	0.369	0.005	0.484
Phenolic compound class and subclass, *mg/day*				
Flavonoids	0.144	0.136	0.123	0.174
Anthocyanins	0.203	0.060	0.198	0.065
Chalcones	0.202	0.061	0.343	**0.004**
Dihydrochalcones	-0.107	0.208	-0.218	**0.047**
Dihydroflavanols	0.231	**0.038**	0.218	**0.047**
Flavanols	0.040	0.380	0.031	0.407
Flavanones	0.226	**0.042**	0.208	0.055
Flavones	-0.119	0.182	-0.207	0.056
Flavonols	0.050	0.351	-0.020	0.441
Isoflavonoids	-0.056	0.335	0.029	0.413
Phenolic acids	0.284	**0.014**	0.258	**0.023**
Hydroxybenzoic acid	0.030	0.409	0.021	0.436
Hydroxycinnamic acids	0.310	**0.008**	0.283	**0.014**
Stilbenes	0.234	**0.036**	0.224	**0.043**
Lignans	0.148	0.129	-0.023	0.432
Other polyphenols	0.028	0.416	0.038	0.385
Alkymethoxyphenols	0.290	**0.012**	0.348	**0.003**
Alkylphenols	-0.019	0.443	0.006	0.483
Curcuminoids	-0.023	0.432	-0.029	0.413
Hydroxybenzaldehydes	0.226	**0.041**	0.217	**0.048**
Hydroxybenzoketones	-0.033	0.402	0.097	0.231
Hydroxycoumarins	0.080	0.272	0.225	**0.042**
Methoxyphenols	0.285	**0.014**	0.299	**0.010**
Tyrosols	0.056	0.336	0.018	0.447

SBP. systolic blood pressure; DBP. diastolic blood pressure; TPI. total polyphenol intake; TPE. total polyphenol excretion. Significant correlations were set at $p<0.05$ (depicted in **bold**). Hypertensive n=28; Normotensive n=32; All subjects n=60. r: Pearson correlation coefficient; p: 1-tailed test of significance for Pearson correlations.

Supplementary Table 3 | Correlations of phenolic compounds from food sources and phenolic classes with faecal short-chain fatty acid concentrations in hypertensive and normotensive subjects

	Propionate *(mg/g feces)*				**Butyrate** *(mg/g feces)*				**Acetate** *(mg/g feces)*				**Valerate** *(mg/g feces)*			
	Hypertensive		**Normotensive**		**Hypertensive**		**Normotensive**		**Hypertensive**		**Normotensive**		**Hypertensive**		**Normotensive**	
	r	**p**	**r**	**p**	**r**	**p**	**r**	**p**	**r**	**p**	**r**	**p**	**r**	**p**	**r**	**p**
TPI. *mg/day*	-0.256	0.188	0.292	0.105	-0.267	0.170	0.134	0.464	-0.196	0.319	0.223	0.221	-0.226	0.248	0.239	0.188
TPE. *GAE/g creatinine*	0.157	0.415	0.237	0.199	0.135	0.485	0.290	0.113	0.290	0.127	0.244	0.185	-0.193	0.316	0.274	0.136
Phenolic compound dietary source, *g/day*																
Tubers	-0.119	0.545	0.044	0.810	0.023	0.908	0.177	0.332	0.011	0.955	0.079	0.668	-0.182	0.353	0.078	0.673
Whole-grain cereals	0.192	0.174	0.262	0.077	0.201	0.162	0.189	0.154	0.321	**0.055**	0.119	0.261	-0.054	0.396	0.040	0.416
Vegetables	-0.115	0.561	0.009	0.962	-0.110	0.578	0.175	0.339	0.028	0.887	0.007	0.969	-0.152	0.439	0.048	0.794
Legumes	-0.315	0.103	0.143	0.433	-0.362	0.059	-0.027	0.882	-0.334	0.082	0.108	0.558	-0.103	0.603	0.032	0.862
Fresh-fruits	0.116	0.557	0.349	0.051	0.205	0.296	0.243	0.180	0.147	0.456	0.292	0.104	0.054	0.784	0.311	0.083
Berries	0.266	0.172	0.231	0.203	0.045	0.821	0.192	0.292	0.128	0.516	0.141	0.441	-0.092	0.643	0.102	0.579
Dried fruits	0.375	**0.049**	0.142	0.440	0.246	0.207	0.158	0.388	0.385	**0.043**	0.105	0.569	0.196	0.318	0.212	0.243
Nuts	-0.219	0.263	-0.060	0.745	-0.216	0.269	0.177	0.332	-0.216	0.270	-0.056	0.759	-0.158	0.422	-0.024	0.894
Olive fruits	-0.117	0.553	-0.001	0.997	-0.171	0.383	0.156	0.395	-0.025	0.899	-0.064	0.729	-0.291	0.133	0.102	0.580
Olive oil	-0.036	0.857	0.280	0.120	-0.016	0.935	0.103	0.575	0.056	0.779	0.202	0.267	-0.064	0.745	0.240	0.185
Chocolate	0.112	0.572	0.239	0.187	0.062	0.755	-0.124	0.499	0.240	0.219	0.129	0.483	-0.285	0.142	0.075	0.683
Natural fruit juices	0.045	0.821	0.166	0.365	-0.074	0.708	0.284	0.115	0.105	0.595	0.092	0.618	-0.170	0.388	0.201	0.270
Coffee	0.475	**0.006**	0.270	0.165	0.013	0.949	0.079	0.667	0.401	**0.023**	0.191	0.329	0.506	**0.003**	0.271	0.263
Tea	-0.023	0.908	-0.040	0.828	-0.118	0.551	0.017	0.928	0.039	0.844	-0.046	0.803	-0.140	0.476	0.052	0.779
Wine	-0.229	0.241	0.029	0.875	-0.173	0.379	-0.101	0.583	-0.309	0.109	0.143	0.436	-0.049	0.806	0.138	0.452
Beer	-0.040	0.841	0.248	0.170	0.165	0.402	0.147	0.422	0.057	0.774	0.264	0.144	-0.058	0.768	0.508	**0.003**
Seasonings	-0.017	0.931	0.031	0.867	-0.078	0.692	0.149	0.416	0.013	0.947	0.070	0.703	-0.204	0.297	-0.095	0.606
Phenolic compound class, *mg/day*																
Anthocyanins	0.048	0.807	0.152	0.405	-0.022	0.913	0.213	0.241	-0.111	0.573	0.095	0.606	-0.070	0.724	0.099	0.589
Chalcones	0.019	0.924	0.242	0.183	-0.033	0.870	0.139	0.446	0.024	0.902	0.261	0.148	-0.046	0.816	0.505	**0.003**
Dihydrochalcones	-0.031	0.875	0.316	0.078	-0.112	0.570	0.137	0.456	-0.060	0.761	0.368	**0.038**	-0.215	0.271	0.367	**0.039**
Dihydroflavanols	-0.228	0.242	0.028	0.877	-0.173	0.378	-0.101	0.584	-0.308	0.111	0.144	0.431	-0.049	0.803	0.137	0.456

Flavanols	-0.060	0.763	0.273	0.130	-0.085	0.666	0.034	0.853	-0.053	0.791	0.279	0.122	-0.153	0.437	0.246	0.174
Flavanones	0.121	0.539	0.201	0.271	0.212	0.278	-0.008	0.963	0.122	0.537	0.159	0.384	0.141	0.474	0.217	0.234
Flavones	0.180	0.360	-0.312	0.082	-0.042	0.831	-0.316	0.078	0.074	0.709	-0.246	0.174	-0.233	0.233	-0.173	0.343
Flavonols	-0.241	0.217	0.041	0.825	-0.282	0.146	0.003	0.986	-0.141	0.474	0.018	0.921	-0.231	0.237	0.060	0.743
Isoflavonoids	-0.298	0.124	-0.146	0.425	-0.218	0.265	-0.041	0.826	-0.161	0.413	-0.126	0.491	-0.149	0.448	-0.009	0.960
Hydroxybenzoic acid	0.130	0.510	0.132	0.473	0.167	0.395	0.284	0.115	0.119	0.547	0.081	0.658	0.120	0.541	0.150	0.411
Hydroxycinnamic acids	0.454	**0.009**	0.193	0.325	-0.096	0.625	0.129	0.481	0.376	**0.034**	0.122	0.537	0.583	**0.000**	0.060	0.762
Stilbenes	-0.232	0.235	0.034	0.855	-0.176	0.370	-0.100	0.585	-0.316	0.102	0.143	0.434	-0.050	0.799	0.143	0.435
Lignans	-0.240	0.218	-0.059	0.748	-0.261	0.180	-0.011	0.954	-0.149	0.450	-0.080	0.663	-0.236	0.226	-0.079	0.669
Alkymethoxyphenols	0.450	**0.010**	0.234	0.230	0.076	0.701	0.076	0.680	0.411	**0.019**	0.213	0.277	0.566	**0.001**	0.236	0.227
Alkylphenols	-0.138	0.483	0.037	0.839	0.031	0.876	0.009	0.962	-0.125	0.526	0.071	0.700	-0.071	0.719	-0.069	0.706
Curcuminoids	-0.038	0.846	-0.137	0.456	-0.192	0.328	-0.147	0.422	0.064	0.746	-0.052	0.779	-0.087	0.662	0.076	0.680
Hydroxybenzaldehydes	-0.222	0.257	0.042	0.820	-0.160	0.415	-0.092	0.617	-0.295	0.128	0.159	0.384	-0.049	0.806	0.156	0.395
Hydroxybenzoketones	-0.044	0.825	0.015	0.936	0.170	0.386	-0.021	0.908	0.050	0.799	0.084	0.649	-0.057	0.773	0.161	0.379
Hydroxycoumarins	-0.044	0.822	0.244	0.178	0.180	0.361	0.138	0.451	0.061	0.759	0.263	0.146	-0.039	0.844	0.506	**0.003**
Methoxyphenols	0.434	**0.013**	0.262	0.179	0.010	0.959	0.045	0.806	0.383	**0.031**	0.197	0.314	0.482	**0.005**	0.262	0.179
Tyrosols	-0.141	0.475	0.139	0.449	-0.214	0.275	0.191	0.295	-0.101	0.608	0.078	0.672	-0.245	0.208	0.250	0.168

SBP, systolic blood pressure; DBP, diastolic blood pressure. Significant correlations were set at $p<0.05$ (depicted in **bold**). Hypertensive n=28; Normotensive n=32. r: Pearson correlation coefficient; p: 2-tailed test of significance for Pearson correlations.

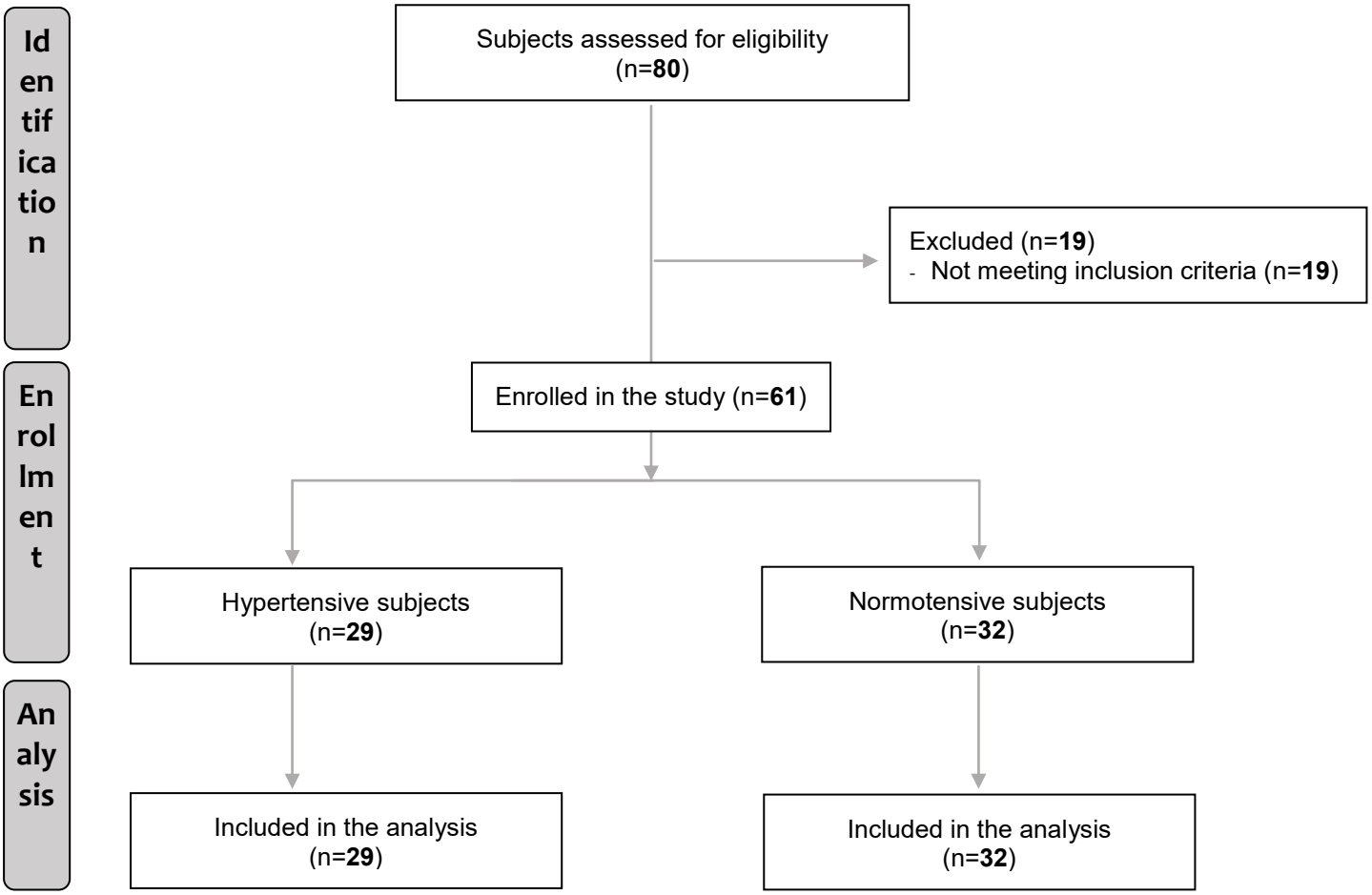
Supplementary Table 4| Correlations of phenolic compounds from food sources and phenolic classes with plasma short-chain fatty acid concentrations in hypertensive and normotensive subjects

	Propionate *(mg/mL plasma)*				**Butyrate** *(mg/mL plasma)*				**Acetate** *(mg/mL plasma)*				**Valerate** *(mg/mL plasma)*			
	Hypertensive		**Normotensive**		**Hypertensive**		**Normotensive**		**Hypertensive**		**Normotensive**		**Hypertensive**		**Normotensive**	
	r	**p**	**r**	**p**	**r**	**p**	**r**	**p**	**r**	**p**	**r**	**p**	**r**	**p**	**r**	**p**
Phenolic compound dietary source, *g/day*																
Tubers	0.173	0.378	0.199	0.282	0.158	0.423	-0.106	0.569	0.159	0.418	0.060	0.750	0.119	0.546	0.054	0.774
Whole-grain cereals	0.036	0.857	-0.207	0.263	-0.108	0.583	-0.177	0.341	-0.188	0.338	-0.140	0.454	-0.123	0.533	-0.069	0.711
Vegetables	-0.026	0.894	0.109	0.559	0.052	0.793	-0.207	0.265	0.244	0.210	0.330	0.070	-0.102	0.606	-0.303	0.097
Legumes	0.186	0.344	-0.172	0.354	-0.167	0.395	0.080	0.671	0.185	0.345	-0.053	0.776	-0.119	0.547	0.168	0.367
Fresh-fruits	0.060	0.760	0.197	0.288	0.053	0.790	**0.368**	**0.042**	0.172	0.380	0.142	0.446	0.185	0.345	**0.576**	**0.001**
Berries	-0.028	0.889	-0.073	0.696	-0.136	0.489	0.250	0.175	-0.109	0.582	0.056	0.765	-0.009	0.963	0.326	0.073
Dried fruits	-0.066	0.737	-0.001	0.997	0.155	0.432	0.197	0.289	-0.164	0.405	0.054	0.771	0.144	0.465	0.291	0.112
Nuts	0.140	0.476	0.026	0.889	-0.179	0.363	0.287	0.117	0.204	0.298	0.148	0.426	-0.258	0.184	0.311	0.089
Olive fruits	-0.139	0.479	0.011	0.952	-0.267	0.170	**0.356**	**0.049**	-0.025	0.900	0.192	0.301	-0.264	0.174	**0.415**	**0.020**
Olive oil	0.078	0.693	-0.011	0.953	-0.098	0.619	-0.065	0.727	0.027	0.891	0.116	0.534	-0.121	0.539	0.087	0.642
Chocolate	0.026	0.894	-0.107	0.567	0.028	0.888	-0.058	0.755	-0.112	0.571	-0.172	0.356	0.054	0.785	0.074	0.691
Natural fruit juices	0.047	0.811	0.011	0.954	0.063	0.752	0.189	0.309	-0.041	0.834	0.272	0.139	0.284	0.143	0.002	0.992
Coffee	0.107	0.588	0.034	0.857	-0.152	0.439	-0.189	0.309	0.309	0.110	0.049	0.792	-0.238	0.222	-0.052	0.780
Tea	0.353	0.065	-0.028	0.880	0.138	0.483	-0.226	0.221	**0.403**	**0.034**	-0.176	0.344	0.011	0.955	-0.181	0.331
Wine	-0.191	0.330	0.060	0.748	-0.201	0.304	-0.152	0.415	0.104	0.597	0.204	0.271	-0.228	0.244	-0.243	0.188
Beer	0.251	0.198	0.037	0.842	-0.101	0.610	-0.168	0.367	0.026	0.895	0.269	0.144	-0.110	0.576	-0.149	0.423
Seasonings	0.209	0.286	0.046	0.805	0.141	0.473	-0.160	0.391	0.183	0.351	-0.066	0.725	0.169	0.391	-0.252	0.171
Phenolic compound class, *mg/day*																
Anthocyanins	-0.185	0.345	-0.048	0.796	-0.208	0.289	0.347	0.056	-0.201	0.304	0.146	0.435	-0.070	0.723	**0.360**	**0.047**
Chalcones	0.326	0.091	0.044	0.815	0.005	0.979	-0.165	0.375	**0.415**	**0.028**	0.275	0.134	0.037	0.852	-0.151	0.417
Dihydrochalcones	0.123	0.535	0.235	0.204	-0.037	0.852	0.047	0.802	-0.008	0.968	0.216	0.243	0.005	0.979	0.194	0.295
Dihydroflavanols	-0.194	0.323	0.060	0.747	-0.204	0.298	-0.152	0.415	0.104	0.598	0.201	0.277	-0.231	0.237	-0.240	0.194
Flavanols	0.168	0.394	0.086	0.645	0.026	0.896	-0.220	0.234	0.215	0.271	-0.014	0.940	-0.033	0.866	-0.038	0.838
Flavanones	-0.061	0.759	0.108	0.564	0.002	0.991	0.041	0.826	0.152	0.439	0.047	0.804	0.116	0.556	-0.030	0.872

Flavones	0.004	0.983	0.014	0.942	-0.138	0.484	-0.186	0.317	-0.037	0.851	-0.096	0.607	-0.002	0.991	-0.129	0.490
Flavonols	0.218	0.265	0.196	0.292	0.094	0.633	-0.110	0.555	**0.446**	**0.017**	0.230	0.213	-0.045	0.822	-0.071	0.704
Isoflavonoids	0.305	0.115	0.130	0.487	0.280	0.148	**0.485**	**0.006**	**0.477**	**0.010**	0.289	0.115	0.291	0.133	0.340	0.061
Hydroxybenzoic acid	0.156	0.427	-0.063	0.736	-0.129	0.512	0.207	0.263	0.259	0.184	0.133	0.475	-0.254	0.193	0.222	0.229
Hydroxycinnamic acids	0.065	0.744	0.226	0.221	-0.241	0.217	-0.140	0.452	0.229	0.241	0.188	0.311	-0.303	0.117	0.126	0.501
Stilbenes	-0.191	0.331	0.060	0.748	-0.208	0.287	-0.150	0.422	0.099	0.617	0.211	0.255	-0.227	0.245	-0.246	0.182
Lignans	0.020	0.919	0.033	0.862	-0.054	0.784	0.083	0.658	0.245	0.208	0.173	0.353	-0.127	0.518	0.052	0.782
Alkymethoxyphenols	0.154	0.434	0.046	0.806	-0.177	0.367	-0.236	0.202	0.256	0.189	0.104	0.576	-0.259	0.183	-0.127	0.496
Alkylphenols	0.125	0.527	-0.029	0.879	0.001	0.996	-0.042	0.822	-0.010	0.958	-0.041	0.829	0.078	0.692	-0.114	0.541
Curcuminoids	**0.422**	**0.025**	-0.177	0.341	0.121	0.539	-0.216	0.244	**0.520**	**0.005**	-0.014	0.940	0.023	0.906	-0.257	0.163
Hydroxybenzaldehydes	-0.182	0.355	0.059	0.754	-0.203	0.301	-0.157	0.400	0.106	0.592	0.202	0.276	-0.241	0.217	-0.230	0.212
Hydroxybenzoketones	0.187	0.341	0.075	0.689	-0.123	0.533	-0.036	0.849	-0.061	0.757	0.211	0.255	-0.139	0.480	-0.054	0.771
Hydroxycoumarins	0.223	0.255	0.043	0.820	-0.060	0.762	-0.167	0.369	-0.013	0.946	0.273	0.137	-0.073	0.714	-0.151	0.417
Methoxyphenols	0.075	0.704	0.037	0.842	-0.160	0.415	-0.210	0.256	0.272	0.162	0.032	0.863	-0.241	0.216	-0.092	0.624
Tyrosols	-0.301	0.120	0.196	0.291	-0.326	0.090	0.098	0.599	-0.103	0.604	0.254	0.167	-0.374	0.050	0.294	0.108

SBP, systolic blood pressure; DBP, diastolic blood pressure. Significant correlations were set at $p<0.05$ (depicted in **bold**). Hypertensive n=28; Normotensive n=32. r: Pearson correlation coefficient; p: 2-tailed test of significance for Pearson correlations.

Supplementary Figure 1 | Flowchart based on the STROBE (Strengthening the Reporting of Observational Studies in Epidemiology) statement



***Declaration of Interest Statement**

Declaration of interests

☒ The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

☐The authors declare the following financial interests/personal relationships which may be considered as potential competing interests:

CRediT Author contributions statement

RMV and RS: Project administration, Conceptualization. AP, LR and RMV: Supervision. LC-P: Formal analysis, Writing - Original draft and preparation, Visualization. LC-P, AP, LR, RMV, RS, EL, JC, LP-P and SY: Investigation, Writing - Reviewing and Editing. MJG and LR: Data curation, Validation, Formal analysis. All authors read and approved the final manuscript.