Systematic analysis of nutrigenomic effects of polyphenols related to cardiometabolic health in humans – Evidence from untargeted mRNA and miRNA studies

Tatjana Ruskovska, Irena Budić-Leto, Karla Fabiola Corral-Jara, Vladimir Ajdžanović, Anna Arola-Arnal, Francisca Isabel Bravo, Georgia-Eirini Deligiannidou, Jaroslav Havlik, Milkica Janeva, Elena Kistanova, et al.

Systematic analysis of nutrigenomic effects of polyphenols related to cardiometabolic health in humans - evidence from untargeted mRNA and miRNA studies

Tatjana Ruskovska, Irena Budić-Leto, Karla Fabiola Corral-Jara, Vladimir Ajdžanović, Anna Arola-Arnal, Francisca Isabel Bravo, Georgia-Eirini Deligiannidou, Jaroslav Havlik, Milkica Janeva, Elena Kistanova, Christos Kontogiorgis, Irena Krga, Marika Massaro, Marko Miler, Hicham Harnafi, Verica Milosevic, Christine Morand, Egeria Scoditti, Manuel Suárez, David Vauzour, Dragan Milenkovic

PII: S1568-1637(22)00091-5

DOI: https://doi.org/10.1016/j.arr.2022.101649

Reference: ARR101649

To appear in: *Ageing Research Reviews*

Received date: 22 July 2021
Revised date: 15 April 2022
Accepted date: 13 May 2022

Please cite this article as: Tatjana Ruskovska, Irena Budić-Leto, Karla Fabiola Corral-Jara, Vladimir Ajdžanović, Anna Arola-Arnal, Francisca Isabel Bravo, Georgia-Eirini Deligiannidou, Jaroslav Havlik, Milkica Janeva, Elena Kistanova, Christos Kontogiorgis, Irena Krga, Marika Massaro, Marko Miler, Hicham Harnafi, Verica Milosevic, Christine Morand, Egeria Scoditti, Manuel Suárez, David Vauzour and Dragan Milenkovic, Systematic analysis of nutrigenomic effects of polyphenols related to cardiometabolic health in humans - evidence from untargeted mRNA and miRNA studies, *Ageing Research Reviews,* (2021) doi:https://doi.org/10.1016/j.arr.2022.101649

Systematic analysis of nutrigenomic effects of polyphenols related to cardiometabolic health in humans - evidence from untargeted mRNA and miRNA studies

Tatjana Ruskovska[1], Irena Budić-Leto[2], Karla Fabiola Corral-Jara[3], Vladimir Ajdžanović[4], Anna Arola-Arnal[5], Francisca Isabel Bravo[5], Georgia-Eirini Deligiannidou[6], Jaroslav Havlik[7], Milkica Janeva[1], Elena Kistanova[8], Christos Kontogiorgis[6], Irena Krga[3,9], Marika Massaro[10], Marko Miler[4], Hicham Harnafi[11], Verica Milosevic[4], Christine Morand[3], Egeria Scoditti[10], Manuel Suárez[5], David Vauzour[12,#] and Dragan Milenkovic[3,13,#,*]

1. Faculty of Medical Sciences, Goce Delcev University, 2000 Stip, North Macedonia

2. Institute for Adriatic Crops and Karst Reclamation, 21000 Split, Croatia

3. Université Clermont Auvergne, INRAE, UNH, F-63000 Clermont-Ferrand, France

4. Institute for Biological Research "Siniša Stanković"- National Institute of Republic of Serbia, University of Belgrade, 11060 Belgrade, Serbia

5. Universitat Rovira i Virgili, Departament de Bioquímica i Biotecnologia, Nutrigenomics Research Group, 43007 Tarragona, Spain

6. Department of Medicine, Democritus University of Thrace, 68100 Alexandroupolis, Dragana, Greece

7. Department of Food Science, Czech University of Life Sciences, 16500 Prague, Czech Republic

8. Institute of Biology and Immunology of Reproduction, Bulgarian Academy of Sciences, 1113 Sofia, Bulgaria

9. Centre of Research Excellence in Nutrition and Metabolism, Institute for Medical Research, National Institute of Republic of Serbia, University of Belgrade, 11000 Belgrade, Serbia

10. National Research Council (CNR) Institute of Clinical Physiology (IFC), 73100 Lecce, Italy

11. Department of Biology, Faculty of Sciences, University Mohammed I, 60000 Oujda, Morocco

12. Norwich Medical School, University of East Anglia, Norwich NR4 7TJ, UK

13. Department of Nutrition, University of California Davis, Davis, CA, 95616, United States of America

#: These authors share senior authorship

*Corresponding author: Dr Dragan Milenkovic, PhD

University of California Davis,

Department of Nutrition,

College of Agricultural and Environmental Sciences

Davis, CA, 95616

United States of America

Email: dmilenkovic@ucdavis.edu

Highlights

- Dietary polyphenols modulate expression of a large number of genes in humans.
- Modulated genes are involved in the regulation of cell adhesion and mobility, immune system, metabolism, or cell signaling.
- Polyphenols can regulate both protein coding and protein non-coding RNAs in humans.
- Gene expression profile is inversely correlated with cardiometabolic diseases and correlated with gene expression following intake of drugs against cardiometabolic disorders.

Abstract

Cardiovascular and metabolic disorders present major causes of mortality in the ageing population. Polyphenols present in human diets possess cardiometabolic protective properties, however their underlying molecular mechanisms in humans are still not well identified. Even though preclinical and *in vitro* studies advocate that these bioactives can modulate gene expression, most studies were performed using targeted approaches. With the objective to decipher the molecular mechanisms underlying polyphenols cardiometabolic preventive properties in humans, we performed integrative multi-omic bioinformatic analyses of published studies which reported improvements of cardiometabolic risk factors following polyphenol intake, together with genomic analyses performed using untargeted approach. We identified 5 studies within our criteria and nearly 5,000 differentially expressed genes, both mRNAs and miRNAs, in peripheral blood cells. Integrative bioinformatic analyses (e.g. pathway and gene network analyses, identification of transcription factors, correlation of gene expression profiles with those associated with diseases and drug intake) revealed that these genes are involved in the processes such as cell adhesion and mobility, immune system, metabolism, or cell signaling. We also identified 27 miRNAs known to regulate processes such as cell cytoskeleton, chemotaxis, cell signaling, or cell metabolism. Gene expression profiles negatively correlated with expression profiles of cardiovascular disease patients, while a positive correlation was observed with gene expression profiles following intake of drugs against cardiometabolic disorders. These analyses further advocate for health protective effects of these bioactives against age-associated diseases. In conclusion, polyphenols can exert multi-genomic modifications in humans and use of untargeted methods coupled with bioinformatic analyses represent the best approach to decipher molecular mechanisms underlying healthy-ageing effects of these bioactives.

Keywords: polyphenols, genomics, bioinformatics, cardiometabolic health, integrated multi-omics

1. Introduction

Worldwide, cardiometabolic diseases (CMD), including heart disease, stroke, and type 2 diabetes mellitus (T2DM) are in constant rise, presenting important societal and economic burdens [WHO, 2018; https://www.who.int/nmh/publications/ncd-profiles-2018/en/; accessed on 27 May 2021]. Dietary factors are important contributors to the incidence of these chronic diseases, and poor dietary habits are recognized as a major determinant of CMD risks (Mozaffarian et al., 2016). It is therefore important to develop nutritional strategies targeting risk factors and consequently to help preventing CMD. Epidemiological and clinical data provide evidence for a consensus on the beneficial effects of a diet rich in plant-based foods for the prevention of obesity, diabetes, and cardiovascular disease (CVD) (Mullins and Arjmandi, 2021) (Satija et al., 2016) (Johnson et al., 2020). In addition to providing low energy and essential micronutrients, plant-based foods are important sources of biologically active phytochemicals with known health-promoting benefits. These bioactive compounds include polyphenols (i.e., flavonoids, phenolic acids, ellagitannins) known to display a wide range of biological activities linked to the prevention of a broad range of chronic diseases (Del Rio et al., 2013) (Vetrani et al., 2018). Human intervention studies have shown that consumption of polyphenols can exert physiological effects with implications for cardiometabolic health, such as improvements in endothelial function, platelet function, blood pressure, arterial stiffness, blood lipid profile and insulin sensitivity (Schroeter et al., 2006) (Rodriguez-Mateos et al., 2018) (Morand et al., 2011) (Tomé-Carneiro et al., 2013a). The mechanisms of action underlying their health effects are related to their ability to modulate oxidative and inflammation processes but also through the regulation of

nitric oxide bioavailability, insulin resistance, glucose and lipid metabolism, amongst others (Kerimi and Williamson, 2016).

Recent studies reported that most of the physiological effects of polyphenols are being mediated by their capacity to modulate the expression of genes and proteins (Krga et al., 2016). For example, it has been described that resveratrol inhibits the expression of the proteins ICAM and GM-CSF in TNF-activated human umbilical vein endothelial cells (HUVECs), as well as cytokines (TNF-α, IL-1, IL-6) and chemokines (CCL2/MCP-1, CCL4/MIP-1, CCL5/RANTES, CXCL10/IP-10) in macrophages (Schwager et al., 2017). A recent systematic analysis of nutrigenomic data from 37 studies identified 54 differentially expressed genes in *in vitro* models of CMD exposed to flavanols and their metabolites (Ruskovska et al., 2020). Global bioinformatic analysis revealed that these genes are predominantly involved in the regulation of inflammation, leukocyte adhesion and transendothelial migration, and lipid metabolism. This systematic analysis also reported that most nutrigenomic studies focused on the analysis of expression of few specific genes and using non-physiologically relevant conditions, which are high concentrations of non-circulating forms of polyphenols for a long period of time, conditions that do not consider the current knowledge on the bioavailability and metabolism of ingested polyphenols. Several *in vivo* and *in vitro* studies using a non-targeted genomic approach revealed, however, that polyphenols can exert complex genomic effects by simultaneously modulating the expression of a large number of genes. Supplementation of diets with anthocyanin-rich extract (Mauray et al., 2012), curcumin (Coban et al., 2012), catechin (Auclair et al., 2009) or quercetin (de Boer et al., 2006) has been shown to modulate expression of several genes spanning from hundred to over a thousand. Similarly, several studies have also revealed that polyphenol circulating metabolites at physiologically relevant concentrations can modulate

large numbers of genes in specific cells in *in vitro* studies. For example, epicatechin metabolites modulated the expression of 751 genes in brain microvascular endothelial cells (Corral-Jara et al., 2021) or over 1,200 in HUVECs (Claude et al., 2014) (Milenkovic et al., 2018). In HUVECs exposed to the mixture of oligomeric procyanidins there were above 1,000 genes for which the expression levels were significantly altered (García-Conesa et al., 2009) or resveratrol that changed the expression of 1,318 genes in human fibrosarcoma cells (Harati et al., 2015). Similarly, a few studies have also identified the genomic impact of consumption of these bioactive compounds in humans and using peripheral blood mononuclear cells (PBMCs) which are easily accessible in clinical trials. Moreover, PBMC transcriptome may reflect systemic health given the fact that they are cells circulating throughout the body and are exposed to different tissues (Afman et al., 2014). Gene expression profiling of PBMCs could reflect different physio-pathological conditions, such as coronary artery stenosis, chronic heart failure, atherosclerosis or inflammation and therefore provide insight in relation to how nutrients can modulate cellular and physiological processes within the context of human nutrition and health (Afman et al., 2014). Moreover, it has been shown that the use of transcriptomics in cardiovascular research can lead to an increased understanding of underlying molecular mechanisms and allows linking mRNA changes to the physiological state of diseases (Witt et al., 2019). Compilation of genomic data from different studies is useful in the identification of novel genes and their potential function for a better understanding of gene networks during the development of diseases.

In this context, the aim of the current study was to perform a systematic analysis of genomic modifications induced by polyphenols or polyphenol-rich foods/extracts consumption, using untargeted approaches, from human clinical studies and to perform a global bioinformatic

analysis of these data with the aim to obtain a more precise picture of the molecular mechanisms underlying the effects of polyphenols in cardiometabolic health.

2. Methods

2.1 Strategy for literature search and data extraction

All literature relevant to the effect of polyphenols on gene expression and cardiometabolic endpoints in human trials was searched and obtained using the Preferred Reporting Items for Systematic Reviews statement guidelines with a predetermined search strategy (Moher et al., 2009). A comprehensive search on PubMed and Web of Science, using Medical Subject Headings (MeSH) and Boolean operators was conducted in July 2018, with an update in November 2019 (Ruskovska et al., 2021) and further in January 2021 only for studies that used untargeted approach for evaluation of gene expression. The search included keywords referring to bioactives and bioactive-rich foods (polyphenols, flavonoids, flavanols, flavanones, epicatechin, catechin, procyanidin, anthocyanins, resveratrol, hydroxytyrosol, extracts, fruits, juice, grapes, citrus, pomegranate, apple, tea, coffee, cocoa, olive oil, chocolate, berries, isoflavones, daidzein, equol, hesperetin), type of studies (human, clinical trials, randomized, patients, volunteers, males, females), nutrigenomic analysis (nutrigenomic, genomic, genome, gene, gene expression, transcription, mRNA, messenger RNA, RT-PCR, PCR-arrays, microarrays, macroarrays, epigenome, miRNA, ncRNA) and target tissues (peripheral blood mononuclear cells, T-cells, B-cells, lymphocytes, monocytes, blood, blood cells, platelets, adipose tissue, adipocytes, liver, plasma, serum). Following the identification of the publications using the aforementioned terms, the search results were narrowed down by only selecting studies that used array technology for analysis of gene expression published in English language.

Inclusion criteria for data extraction were as follows: human intervention studies of CMD or cardiometabolic risk factors where polyphenols were used for intervention, and modulation of gene expression was studied at mRNA and/or miRNA level, which was analyzed with macro- or microarray methods. Furthermore, in order to be included in our analysis, the publications should have reported at least one beneficial cardiometabolic outcome, such as: improved plasma lipid status (total cholesterol, triglycerides, HDL-cholesterol, LDL-cholesterol, apoA1, apoB or oxLDL, etc.), improved oral glucose tolerance test, decreased fasting glucose, glycated hemoglobin, insulin resistance, blood pressure, body weight, waist circumference, systemic inflammation (circulating CRP, TNF-α, interleukins, etc.), or decreased oxidative stress. Studies reporting an unfavorable cardiometabolic outcome, for example, increased LDL-cholesterol, were excluded from our analysis. This criterion was chosen with the aim to only identify changes in the expression of genes that will result in health promoting properties of bioactives. In addition, studies that did not include a control of polyphenol intervention, or studies of co-interventions (for example, co-interventions of polyphenols with vitamins or exercise), were also excluded.

A template for data extraction was designed specifically for this study. The template was pilot tested to verify the accuracy of the data extraction. The final version of the template was distributed among the co-authors. Extracted data included: details about the publication (PMID, authors, year of publication, title), information about the study population (gender, age, number of participants, health status), information about the intervention (bioactive/s, dose and duration of intervention), study design, information about the cardiometabolic outcomes (including both studied and significantly modulated), and information about gene expression (mRNA and/or miRNA, analytical method, tissue/cells analyzed for gene expression). Only genes that were

significantly modulated (positive or negative regulations) by the intervention ($p<0.05$) were extracted from the eligible publications, and subsequently subjected to bioinformatic analyses. Extracted data were cross-checked by two co-authors. In case of doubts or disagreement, a third co-author was consulted.

2.2 Bioinformatic analyses

Pathway enrichment analyses were conducted using the bioinformatic tool GeneTrail2 (https://genetrail2.bioinf.uni-sb.de/) (Stockel et al., 2016), as a platform to access KEGG database. InteractiVenn (http://www.interactivenn.net/) (Heberle et al., 2015) was used as a tool to retrieve elements that are in common for different datasets. Networks of pathways were visualized using the Cytoscape software (https://cytoscape.org) (Shannon et al., 2003), version 3.7.2. Official gene names and symbols were identified using GeneCards (https://www.genecards.org/) (Stelzer et al., 2016).

Potential transcription factors which activity could be modulated by polyphenols were identified with bioinformatic tool Enrichr (https://amp.pharm.mssm.edu/Enrichr/) (Chen et al., 2013) (Kuleshov et al., 2016), as a platform to interrogate two transcription factor databases, TRRUST (Han et al., 2018) and TRANSFAC (Matys et al., 2003).

MiRNA targets were identified, and a network of miRNAs and their target genes was constructed using the online tool MIENTURNET (http://userver.bio.uniroma1.it/apps/mienturnet/) (Licursi et al., 2019). The MIENTURNET tool was also used for functional enrichment analysis, to identify cellular pathways significantly over-represented for individual miRNAs in the 3 studies.

The association of polyphenol modulated genes with human diseases was analyzed using the Comparative Toxicogenomics Database (https://ctdbase.org/) (Davis et al., 2021) together with GWAS (https://www.ebi.ac.uk/gwas/) (Buniello et al., 2019) and DisGeNET (https://www.disgenet.org/) (Piñero et al., 2020). The last two databases were interrogated using bioinformatic tool Enrichr (https://amp.pharm.mssm.edu/Enrichr/) (Chen et al., 2013) (Kuleshov et al., 2016).

To explore correlations between polyphenol modulated genes and genes associated with CMD, we first searched the GEO (https://www.ncbi.nlm.nih.gov/gds) for suitable datasets to obtain gene expression profiles of CMD and drugs commonly used for the treatment of insulin resistance, dyslipidemia, or hypertension. The available datasets were further analyzed with GEO2R. Pearson's correlation coefficients between these gene expression profiles and polyphenol modulated genes were calculated using the R program (https://www.r-project.org/) (Dessau and Pipper, 2008).

3. Results

3.1 Studies and bioactives

A comprehensive search on PubMed and Web of Science, conducted in July 2018, with an update in 2020, resulted in 8,678 documents. The update of literature search in January 2021 did not retrieve any additional eligible publications. After removal of duplicates and using “Human” and “RCT” and “Clinical study” and “Clinical trial” and “Controlled Clinical Trial” and “Multicenter Study” filters, this number dropped to 465 manuscripts, which were further screened and assessed for eligibility. Of these, 12 publications were identified as having adopted holistic approaches (Ruskovska et al., 2021). In one of these 12 publications, only proteomic

biomarkers were analyzed (Fuchs et al., 2007), and as such, it was ineligible for inclusion in our study. One publication reported modulation in global DNA methylation (Crescenti et al., 2013), two publications reported modulation of gene expression in abdominal subcutaneous white adipose tissue (Konings et al., 2014) (Most et al., 2018), three in skeletal muscle (Most et al., 2016) (Timmers et al., 2011) (Pollack et al., 2017), and five in blood samples. In our study, we included only the publications that reported significant changes in gene expression in blood samples, analyzed with macro- or microarray methods, which were accompanied by at least one favorable cardiometabolic outcome in controlled human intervention studies with polyphenols (Figure 1). Further in the text, these publications are denoted as Publication 1 (De Groote et al., 2012), Publication 2 (Milenkovic et al., 2011), Publication 3 (Tomé-Carneiro et al., 2013b), Publication 4 (Milenkovic et al., 2014) and Publication 5 (Rodriguez-Mateos et al., 2019). For three of these publications, the information about the cardiometabolic outcomes was extracted from previously published papers (Morand et al., 2011) (Tomé-Carneiro et al., 2013a) (Weseler et al., 2011) (Table 1).

The study population comprises almost exclusively men, adults up to 80 years old. Only one of the studies included women (Publication 1). The health status of the participants is heterogeneous and includes obese men and women with BMI between 30 and 40 kg/m^2; overweight men; men with type 2 diabetes, hypertension, and coronary artery disease; non-obese healthy smokers (men); and healthy men. The five selected studies were chronic interventions. Four of them are similar in duration – three lasted for four weeks/one month and one lasted two months. Only one of the studies is much longer and has been conducted in duration of one year (Publication 3). Bioactives used for intervention include: trans-resveratrol phosphate, catechin-rich grape seed extract, hesperidin, grape extract plus resveratrol, monomeric and oligomeric

flavanols from grape seeds, and wild blueberry anthocyanins (Table 1). Total RNA for global gene expression analysis was isolated either from whole blood (Publication 1 and 4), peripheral blood mononuclear cells (Publication 3 and 5) or total leukocytes (Publication 2).

3.2 Functional pathways analysis of genomic data

The number of differentially expressed genes at mRNA level extracted from each of the selected publications varied from less than 50 (Publication 1) to more than 2,000 (Publication 3). The lists of identified differentially expressed genes and their fold-changes (FC) are presented in the Supplemental table S1. The total number of differentially expressed genes extracted from all selected publications reached N=5,465. After the removal of duplicate genes, this number dropped to N=4,990 (Supplemental table S2a). Comparisons of genes identified as differentially expressed among these 5 studies showed that 456 genes are in common in at least 2 studies (Supplemental table S2b). Comparison of FC of the common genes showed that for over 65% of them, the direction of change in the expression is the same among the studies. Differencies in the direction of change in the expression between the studies can be due to different study populations with potential influence of age or health status, but also due to the use of differerent bioactives, as well as the different types of arrays.

For each of the selected publications, differentially expressed genes at the mRNA level were subjected to pathways enrichment analysis to identify pathways that are the most significantly associated with each specific gene set (Supplemental table S3). For this purpose, we used the platform GeneTrail2 to access the KEGG database. Next, we analyzed which of these pathways are in common for at least three of the selected publications. The pathways related to various diseases and addictions were excluded, while the pathways related to cellular processes that are

in common for at least three of the selected publications are presented in Figure 2. These pathways are involved in cell signaling, immune system, cell motility and interaction, endocrine system, nervous system, metabolism, and other cellular processes. Pathways that are in common for four or five publications included: PI3K-Akt signaling pathway, MAPK signaling pathway, Ras signaling pathway (cell signaling), chemokine signaling pathway, natural killer cell mediated cytotoxicity, B cell receptor signaling pathway, T cell receptor signaling pathway (immune system), focal adhesion, cell adhesion molecules (cell motility and interaction), estrogen signaling pathway, thyroid hormone signaling pathway (endocrine system), dopaminergic synapse, cholinergic synapse, serotonergic synapse, neurotrophin signaling pathway (nervous system), glutathione metabolism (metabolism), osteoclast differentiation, phagosome, protein processing in endoplasmic reticulum, ribosome, RNA transport, endocytosis (other cellular processes) (Figure 2). Networks of these pathways, both global and networks related to specific cellular processes are presented in Figure 3A and 3B.

Next, the differentially expressed genes that were in common in at least two of the selected publications (N=456, Supplemental table S2b) were further subjected to pathways enrichment analysis, using the platform GeneTrail2 to access the KEGG database. Excluding pathways that are related to various diseases and addictions, this pathways enrichment analysis revealed that the genes were associated with pathways that are involved in cell signaling (Ras signaling pathway, PI3K-Akt signaling pathway, mTOR signaling pathway, NF-kappa B signaling pathway, HIF-1 signaling pathway), immune response (chemokine signaling pathway, NOD-like receptor signaling pathway, cytosolic DNA-sensing pathway, B cell receptor signaling pathway, T cell receptor signaling pathway), the endocrine and circulatory system (thyroid hormone signaling pathway, adrenergic signaling in cardiomyocytes, vascular smooth muscle contraction,

adipocytokine signaling pathway, progesterone-mediated oocyte maturation), the nervous system (dopaminergic synapse, retrograde endocannabinoid signaling, serotonergic synapse, neurotrophin signaling pathway, cholinergic synapse), metabolism (pyruvate metabolism, carbon metabolism, pyrimidine metabolism, citrate cycle, propanoate metabolism), and other processes (RNA transport, ribosome, mineral absorption, circadian entrainment, proteasome) (Supplemental table S2c). Within our analysis we further extracted the genes that were in common in at least three of the selected publications. These include: *FSTL4 (down-regulated), AHCTF1 (up-regulated), TMEM201 (up-regulated), POLR2G (up-regulated), MRPS18C (down-regulated),* and *SUCLA2 (up-regulated)* (Table 2).

3.3 Transcription factors regulating the expression of polyphenol modulated genes

Following the extraction of differentially expressed genes, our next step was to use the extracted genomic data and, using the available databases and bioinformatic tools, to search for potential transcription factors of which activity could be modulated by these food bioactives. For this analysis, the Enrichr tool was used to interrogate two transcription factor databases, TRRUST and TRANSFAC. Using this approach, we identified between 9 and 37 potential transcription factors depending on the study with significance values $p<0.05$. Among these transcription factors, several were identified in common in two or more studies. For example, following analysis of genomic data, SP1, SREBF2 and CRX were found in both Publication 1 and 5; STAT3 was common in Publication 1, 2 and 5; Nf-kB1 and ATF1 were found in both Publication 1 and 4. Other transcription factors seem to be specific to each study and potentially to the polyphenol under investigation. For example, PPARG, ATF3 or NFIA were identified as specific in Publication 1; SMAD4 or PAX6 in Publication 2; ATF6, KLF4 or ATF4 in

Publication 3; ZFP36 or NR2F1 in Publication 4; SP3, FOXF2 or GATA1 in Publication 5 (Figure 4).

3.4 Polyphenols modulate miRNAs expression in PBMCs in human volunteers

Among the identified studies, 3 publications also reported changes in the expression of miRNAs in human PBMCs following consumption of polyphenols. In the genomic analysis reported in Publication 2, 13 miRNAs were identified as differentially expressed following consumption of hesperidin, but also following regular intake of a grape extract enriched with resveratrol as reported in Publication 3. Three miRNAs were also identified as differentially expressed in PBMCs in volunteers following consumption of anthocyanins (Publication 5). These observations suggest that different families of polyphenols may have the capacity to modulate the expression of these small protein non-coding RNAs. Comparisons of miRNAs identified miR-211 and miR-25, in both Publication 2 and Publication 3. No other miRNAs were identified in common between these 2 studies. Publication 5 only identified 3 differentially expressed miRNAs (Figure 5).

Our next step was to identify target genes of these differentially expressed miRNAs. Using MIENTURNET, an online tool for identification of miRNA targets, networks of miRNAs and their target genes have been constructed (Figure 6A, 6B, 6C). We identified 2,473 target genes for Publication 2, 3,208 for Publication 3 and 705 for Publication 5. Functional enrichment was also performed using the MIENTURNET tool, and identified cellular pathways significantly over-represented for individual miRNAs in the 3 studies. For example, miR-135a-5p's targets were involved in pathways regulating amino acid metabolism or cell signaling pathways; miR-193-3p was involved in pathways regulating cancer development, cell signaling or cell cycle.

Other miRNAs were also found in pathways regulating inflammation, lipid metabolism or cardiomyopathy (Figure 7A). For miRNAs identified as differentially expressed in Publication 3, functional enrichment analysis identified pathways involved in cell signaling, lipolysis, apoptosis, or cytoskeleton organization; while only few pathways were identified for target miRNAs identified in Publication 5, including inflammation, MAPK signaling or cancer (Figure 7B and 7C).

Together with functional enrichment using MIENTURNET, significantly over-represented pathways for miRNA targets were also searched using the GeneTrail2 tool. Consequently, we identified 51, 78 and 49 pathways for Publication 2, Publication 3 and Publication 5 respectively. Comparisons of these pathways revealed 9 common pathways for the 3 studies: adrenergic signaling in cardiomyocytes, PI3K-Akt signaling pathway, pancreatic secretion, cholinergic synapse, purine metabolism, glutamatergic synapse, arrhythmogenic right ventricular cardiomyopathy, Ras signaling pathway and dilated cardiomyopathy (Figure 8). Ten pathways were identified in common between Publication 3 and Publication 5; 3 between Publication 2 and Publication 5 and 31 between Publication 3 and Publication 2 (Figure 8). These major pathways are involved in the regulation of cellular processes such as cell signaling, inflammation, chemotaxis, lipid metabolism or cell-cell adhesion and permeability.

3.5 Integrated analysis of protein-coding and non-coding gene expression

Because miRNAs are small RNAs involved in the post-transcriptional regulation, our next goal was to perform an integrated data analysis of genomic modifications of protein-coding RNAs with non-coding RNAs, miRNAs. Comparison of differentially expressed genes with the target genes of miRNAs showed that there were 8.8% of differentially expressed genes in common

with miRNA target genes, with the percentage ranging from 3.1 to 15.1 (Figure 9A). This suggests that about 9% of changes in genes expression could be related to changes in the expression of miRNAs.

Following this step, we compared pathways identified using GeneTrail2 for differentially expressed genes and targets of miRNAs. As presented in Figure 9B, we identified 35 common pathways, including those related to cell adhesion molecules, chemokine signaling pathway, focal adhesion, insulin secretion, leukocyte transendothelial migration, MAPK signaling pathway, Ras signaling pathway or regulation of actin cytoskeleton. This observation suggests that these pathways could be affected by polyphenols through the expression of protein-coding genes and miRNAs. Examples of 2 common pathways, chemokine signaling pathway and leukocyte transendothelial migration are presented in Figure 9C, which show that modulation of protein-coding and non-coding genes can affect a large number of genes in the pathways and consequently the functionality of the pathways identified.

3.6 Disease bioinformatic analyses

We next analyzed the association of polyphenol modulated genes with human diseases. For this purpose, we used the Comparative Toxicogenomics Database, together with GWAS and DisGeNET that were interrogated using Enrichr. This analysis showed an association with 20, 77, 50 and 56 human diseases for publications 2-5, respectively. Metabolic, cardiovascular and heart diseases are in common for publications 2-5, while vascular diseases and myocardial ischemia are in common for publications 3 and 4 (Figure 10). A comparison of all polyphenol-modulated genes (N=4,990) with genes associated with disease development from the DisGeNET database showed that 367 polyphenol-modulated genes were associated with CVD,

224 with metabolic syndrome, and 126 with both CVD and metabolic syndrome (Figure S1). We also searched for GWAS and meta-analyses of GWAS that identify genes associated with various CMD and risk factors. For each of these studies we analyzed the overlapping genes with polyphenol-modulated genes from our study. We found that 19.6, 19.0, 22.8, 21.5, and 17.6% of genes associated with body fat distribution (Shungin et al., 2015), insulin resistance (Lotta et al., 2017), blood pressure (Liu et al., 2016a), metabolic syndrome (Lind, 2019), and coronary artery disease (LeBlanc et al., 2016) respectively overlap with polyphenol-modulated genes. Altogether, the results of these analyses highlight the involvement of polyphenol-modulated genes in CMD and risk factors.

To get further insight into the genomic effects of dietary polyphenols, we analyzed the presence of correlations between polyphenol modulated genes and genes associated with CMD. To this aim, available GEO datasets were analyzed with GEO2R and differentially expressed mRNAs were correlated with those extracted from the analyzed publications using the R program. These analyses showed that gene expression profile of metabolic syndrome (GSE98895) (D'Amore et al., 2018) is significantly and inversely correlated with gene expression profiles after consumption of wild blueberry anthocyanins (Publication 5), grape extract plus resveratrol (Publication 3) and monomeric and oligomeric flavanols from grape seeds (Publication 4) (Figure 11 A-C). Also, the gene expression profile after consumption of grape extract plus resveratrol (Publication 3) showed an inverse and highly significant correlation with the gene expression profile of T2DM (GSE23561) (Grayson et al., 2011), and a weak inverse correlation with the gene expression profile of arterial stiffness and hypertension (GSE6599) (Figure 11 D-E).

Using the same approach, we also tested correlations between polyphenol modulated genes and gene expression profiles of common drugs used for the treatment of cardiometabolic diseases. For example, the metformin gene expression profile (GSE153318) (Seneviratne et al., 2021) showed a high and statistically significant positive correlation with grape extract plus resveratrol (Publication 3) (Figure 12A). Similarly, grape extract plus resveratrol gene expression profile exerted a significant positive correlation with the gene expression profile of statins, as reported in GSE11393 (Llaverias et al., 2008) and GSE71220 (Obeidat et al., 2015) (Figure 12B and 12D). Also, the gene expression profile of wild blueberry anthocyanins (Publication 5) showed a trend towards significant positive correlation with the gene expression profile of statins GSE71220 (Obeidat et al., 2015) (Figure 12C). We also tested correlations between genes extracted from the analyzed publications and gene expression profiles of some of the common antihypertensive drugs. We found a trend towards the positive correlation of gene expression profile of grape extract plus resveratrol (Publication 3) with gene expression profile of the treatment with telmisartan (Figure 12H), along with a high and statistically significant positive correlation with a combination of telmisartan and amlodipine (GSE42808) (Siragusa and Sessa, 2013) (Figure 12G). Potential correlation with gene expression profile of anti-hypertensive drugs was also observed for blueberry and hesperidin intake, even though a statistical significance was not reached (Figure 12E, 12F and 12I). Altogether, these results suggest that polyphenols can have a positive effect on cardiometabolic risk factors.

4. Discussion

Cardiovascular and metabolic diseases are major causes of mortality in the ageing population, which is rapidly increasing worldwide. Therefore, there is a need and interest to find efficient

solutions which could contribute to the prevention of development of these diseases. Epidemiological and human intervention studies have shown that polyphenols exert protective effects on human cardiometabolic health (Hertog et al., 1993) (Grassi et al., 2008). However, molecular mechanisms underlying health properties of polyphenols are not entirely identified. A number of *in vitro*, preclinical and human intervention studies have shown that polyphenols and their metabolites can modulate the expression of different genes. Systematic bioinformatic analyses showed that by modulating the gene expression, polyphenols exert multilayer and multitarget modes of actions (Ruskovska et al., 2020) (Monfoulet et al., 2021) (Ruskovska et al., 2021). However, most of the studies have been performed using targeted approach, which only provides incomplete pictures of molecular mechanisms involved. Few *in vitro* studies and animal models used untargeted approach, such as microarrays, and revealed the capacity of polyphenols to modulate simultaneously several hundreds of genes (Corral-Jara et al., 2021) (Milenkovic et al., 2021) (Coban et al., 2012). In humans, the mode of action of polyphenols is still largely unidentified. With the goal to elucidate the molecular mechanisms underlying the beneficial effects of polyphenols on cardiometabolic health in humans, we conducted a systematic literature search of studies that used an untargeted approach followed by comprehensive bioinformatic analyses. Even though the number of available studies so far remains small, available data allowed us to integrate both mRNAs and miRNAs data, to perform functional analysis and to predict transcription factors of which activity could be regulated by polyphenols and may play an important role in molecular mechanisms of action of these bioactives.

Integrated bioinformatic analyses of the genes that have been identified as differentially expressed in humans showed that they were involved in different cellular processes such as inflammation, cell signalling, cell mobility and adhesion, or cell metabolism. One of the

pathways that we looked at in our bioinformatics study was the PI3K-Akt signaling pathway. Recent research suggests that cardiomyocyte-secreted vascular endothelial growth factor (VEGF) plays a key role in cardiac hypertrophy and myocardial infarction. Akt and mTOR have been shown to promote angiogenesis by raising the expression of VEGF and angiopoietin (Ang)-2 (Shiojima et al., 2005) (Shiojima and Walsh, 2006). Furthermore, atherosclerotic plaque formation is a typical feature of atherosclerosis. Activation of PI3K/Akt signaling can induce monocyte chemotaxis, macrophage migration, increased intracellular lipid accumulation, neovascularization, smooth muscle cells proliferation and dysfunction in lesions, all of which are involved in plaque formation (Zhao et al., 2021). In cardiovascular disorders, cell adhesion molecules (CAMs) are particularly involved in atherogenesis and atherosclerotic plaque progression. Obese adults have significantly greater sICAM-1, E-selectin, and P-selectin serum concentrations than healthy ones, demonstrating that the increased cardiovascular risk associated with obesity may be reflected and possibly mediated by significantly increased CAMs (Mulhem et al., 2021).

Furthermore, in our analysis, we discovered a link between the neurological and cardiovascular systems. For instance, brain-derived neurotrophic factor (BDNF) is critical throughout the development of the cardiovascular system because it activates the TrkB receptor, which leads to endothelial cell survival and the construction of the heart vasculature (Emanueli et al., 2014). BDNF can enhance vascular flow and can regulate revascularization of ischemic tissues. The metabolic system, on the other hand, is intimately linked to cardiovascular diseases. Increased amounts of free radicals are hypothesized to cause impaired "redox homeostasis", a balance between oxidative and reductive stress. Glutathione is the most abundant antioxidant in the heart,

and a low level of red-cell glutathione peroxidase 1 activity was linked to an increased risk of cardiovascular events in people with coronary artery disease (Blankenberg et al., 2003).

Our bioinformatic analysis of genomic data also allowed us to identify potential transcription factors of which activity could be modulated by polyphenols and underlie changes in the expression of genes observed in genomic studies. Among the transcription factors identified and common for 2 studies, was the Activating Transcription Factor 1 (ATF1). It has been shown that metformin, the most widely used anti-diabetic drug with anti-atherogenic properties, independently of its effect on glycaemia (Luo et al., 2019) acts through modification of phosphorylation of ATF1 transcription factor and increase the expression of atheroprotective genes (Salt et al., 2021). It has also been described that omega-3 polyunsaturated fatty acids, which have several beneficial effects on CVD risk factors, can modulate the DNA methylation of genes in obese subjects including ATF1. Change in epigenetic profile of these genes has been correlated with changes in plasma triglyceride and glucose levels as well as with changes in the ratio of total cholesterol/HDL-cholesterol following the supplementation therefore providing new potential insights on the mechanisms of action (Tremblay et al., 2017). Another transcription factor identified from our bioinformatic analysis was the signal transducer and activator of transcription 3 (STAT3), which has been described as involved in the development of cardiovascular and cerebrovascular diseases and obesity (Chen et al., 2019). Indeed, aberrant STAT3 activation was shown to contribute to endothelial cell dysfunction, macrophage polarization, inflammation, and immunity. Different studies described that several STAT3 inhibitors could be used as therapeutic drugs to treat diseases (Chen et al., 2019). The capacity of bioactive molecules to act as STAT3 inhibitors has been reported in several studies. For

example, it was observed that curcumin could inhibit STAT3 *in vitro* (Alexandrow et al., 2012) and that resveratrol could contribute to the treatment of thrombosis and atherosclerosis via STAT3 inhibition (Sun et al., 2018).

Our bioinformatic analysis also identified the transcription factor NFκB as being involved in the genomic modifications induced by polyphenols. This transcription factor is well known to be involved in the regulation of inflammation and atherosclerotic processes but also in the pathogenesis of cardiac remodeling and heart failure (Fiordelisi et al., 2019). Atherosclerosis is characterized by the deposition of lipids in arterial walls and involves leukocyte recruitment, adhesion to the endothelium and their transendothelial migration into the subendothelial layer of blood vessels. These processes require the expression of adhesion molecules, interleukins, chemokines, which are regulated by NFκB in response to stress (Panday et al., 2016). Consequently, anti-inflammatory treatments were developed in recent years and NFκB has been identified as one of the major candidates. Several studies have shown that inhibition of this transcription factor results in endothelial and cardiovascular protection. It has been furthermore observed that inhibition of NFκB could prevent immune response and adhesion of monocytes to the endothelium, *in vitro* and *ex vivo* (Ward et al., 2020). Phosphodiesterase inhibitors, used for the treatment of heart failure, are able to reduce the production of cytokines; for example pimobendan was shown to lower the production of IL-1β, TNF-α and nitric oxide by inhibiting the activation of NFκB in mouse models of heart failure (Iwasaki et al., 1999) (Matsumori et al., 2000). Interestingly and in agreement with our observation, polyphenols, such as flavonoids have been observed to exert anti-inflammatory effects by acting on the NFκB pathway (Hussain et al., 2020) (Chu, 2014). Our analysis also identified few other transcription factors, probably to be involved in the regulation of genomic response following consumption of these bioactives.

Among those is PPARγ, implicated in cardiovascular diseases and T2DM (Han and Qu, 2020) along with RUNX2 (Runt-related transcription factor 2) involved in atherosclerosis development (Chen et al., 2021) or KLF5 identified as playing a role in CVDs (Dong and Chen, 2009). Taken together, our integrated omic analysis has identified highly probable transcription factors involved in the genomic modification following consumption of these polyphenols, presenting important molecular mechanisms underlying their health properties.

Expression of genes, and consequently proteins, can also be regulated post-transcriptionally by non-coding RNAs, such as miRNAs. MiRNAs are small non-coding RNAs whose expression can be regulated by different factors, and can bind to different mRNAs and induce either their degradation or prevent protein synthesis (Friedman et al., 2009) (Bartel, 2004). There is accumulating data showing that changes in the expression of miRNAs play an important role in the development of cardiovascular and metabolic diseases (Bielska et al., 2021) (Ramzan et al., 2021). Studies have shown that polyphenols can regulate the expression of miRNAs, *in vivo* and *in vitro*, and present one of the molecular mechanisms underlying their health properties (Milenkovic et al., 2018) (Krga and Milenkovic, 2019) (Krga et al., 2018). Among the miRNAs identified are miR-211 and miR-25, differentially expressed by hesperidin and grape seed extract rich in resveratrol. MiR-211 has been identified in obese patients with non-alcoholic fatty liver disease (Mehta et al., 2016). MiR-25 was observed to be correlated with coronary stenosis, possibly by regulating processes such as "angiogenesis" and "leukocyte cell-cell adhesion" (Nariman-Saleh-Fam et al., 2019), as well as with increased heart failure risk in coronary heart disease patients (Yao et al., 2018). Interestingly, it has been reported the polyphenol taxifolin can regulate the expression of miR-211 (Dostal et al., 2021) and epigallocatechin gallate or resveratrol can regulate the expression of miR-25 (Zan et al., 2019) (de Queiroz et al., 2018).

Several other miRNAs have been identified as having expression modulated by polyphenols. For example, miR-181b has been identified as differentially expressed by grape-seed rich in resveratrol consumption, a miRNA that has been described to regulate for example, arterial stiffness related to ageing (Hori et al., 2017) or vascular inflammation in T2DM (Witkowski et al., 2020). MiRNA-126, which has been reported as a predictor of long-term all-cause mortality in patients with T2DM (Pordzik et al., 2021), was identified to be regulated by anthocyanin-rich berries. In addition, miRNA-7, differentially expressed in volunteers that consumed grape-seed rich in resveratrol, has been identified as differentially expressed in hypertensive patients with left ventricular hypertrophy (Kaneto et al., 2017). Comparisons of the identified differentially expressed miRNAs with miRNAs from the Human microRNA Disease Database (HMDD) are in agreement with our analysis. This analysis showed that some of the miRNAs identified as having expression modulated by polyphenols have been described to be causally involved in the development of atherosclerosis (hsa-mir-135a), cardiomyopathy (hsa-mir-27a, hsa-mir-218), diabetes (hsa-mir-27a, hsa-mir-424), hypertension (hsa-mir-27a, hsa-mir-424), inflammation (hsa-mir-135a, hsa-mir-25, hsa-mir-424) or obesity (hsa-mir-27a) but also coronary artery diseases (hsa-mir-197, hsa-mir-25) and vascular diseases (hsa-mir-27a). This observation shows the importance of miRNAs as molecular targets of flavonoids underlying the health benefits of consumption of these food bioactives.

Our bioinformatic analysis also included identification of potential target genes of these miRNAs as well as pathways in which they are involved in. Using this functional analysis, we identified that miRNAs, differentially expressed following consumption of polyphenols by volunteers, are involved in pathways related to the regulation of different processes such as inflammation, cell adhesion, chemotaxis, or apoptosis. These processes have been described as involved in the

development of atherosclerosis and consequently the development of vascular dysfunction, including arterial stiffness and cardiovascular diseases (VanderBurgh et al., 2018). Interestingly, comparisons of pathways identified from protein-coding genes and miRNAs have shown a number of common pathways. Among these pathways are those regulating inflammation, cell adhesion, cytoskeleton, cellular metabolism, again the pathways playing key roles in endothelial permeability leading to atherosclerosis development and consequently cardiovascular diseases but also metabolic disorders (van Steen et al., 2020) (van Buul and Hordijk, 2004) (Li et al., 2021). These results suggest that polyphenols can prevent the development of cardiometabolic diseases by modulating both the expression of protein-coding but also non-coding genes. Therefore, to gain a better understanding of the molecular mechanisms involved in the health benefits of these bioactives it is important to take into account the analysis of both types of RNAs in future studies.

Genomic modulations that are reported in the publications included in our study are associated with beneficial effects on key determinants of cardiometabolic health, such as blood pressure and vascular function, lipid status, blood glucose and oxidative stress. Hence, our bioinformatic analysis shows that more than 9% of polyphenol modulated genes are in common with the genes associated with metabolic syndrome and cardiovascular disease. Metabolic syndrome, a cluster of several cardiometabolic risk factors that include abdominal obesity, insulin resistance, dyslipidemia, and hypertension (Alberti et al., 2009), leads to an increased cardiovascular morbidity and mortality (Saklayen, 2018) (Gluvic et al., 2017). The protective role of polyphenols on the incidence of metabolic syndrome and some of its components has been recently reported in a large-scale European epidemiological study (Grosso et al., 2017). These findings are further corroborated with our bioinformatic analyses showing a highly significant

negative correlation between the gene expression profile of metabolic syndrome and dietary polyphenols such as wild blueberry anthocyanins, grape extract plus resveratrol or flavanols from grape seeds.

Our bioinformatic analyses also revealed a negative correlation between the gene expression profile of grape extract plus resveratrol and vascular dysfunction (arterial stiffness and hypertension), as well as a highly significant positive correlation between the gene expression profile of the same mixture of bioactives and the treatment with a combination of two antihypertensive drugs (telmisartan and amlodipine). These findings highlight the genomic mechanisms underlying the antihypertensive mode of action of polyphenols in cardioprotection. The effects of polyphenols on hypertension and vascular dysfunction as key components of metabolic syndrome have been widely studied. Bioactives under investigation include hesperidin (Valls et al., 2021), anthocyanins (Igwe et al., 2019), grape seed polyphenols (Schön et al., 2021), or resveratrol (Breuss et al., 2019) (Ferreira et al., 2020), for which global genomic effects have been reported in publications included in our study. Meta-analyses of human intervention studies showed modest effects of various polyphenols or polyphenol-rich foods on blood pressure when compared with antihypertensive drugs (Zhang et al., 2016) (Sahebkar et al., 2017). Nevertheless, in addition to medical treatment, even modest improvements in blood pressure are shown to be clinically significant for the risk reduction of stroke and coronary heart disease (Cook et al., 1995) (Whelton et al., 2002).

Atherogenic dyslipidemia is yet another hallmark of metabolic syndrome. The first line of defense in its management are lifestyle changes, which also include plant-based dietary regimens (Trautwein and McKay, 2020). Studies have demonstrated improvements in serum lipid status following interventions with anthocyanins (Liu et al., 2016b) (Li et al., 2015), grape seed extract

(Asbaghi et al., 2020) (Yousefi et al., 2021) or resveratrol (Simental-Mendía and Guerrero-Romero, 2019) (Hoseini et al., 2019). Although these effects are mild, on a genomic level they are comparable to the effects of statins, as shown in our bioinformatic analyses, and as such can contribute to the improvement of cardiometabolic health.

While metabolic syndrome and T2DM often coexist, non-diabetic individuals with metabolic syndrome are at a much greater risk of developing T2DM (Punthakee et al., 2018). Medical nutrition therapy, together with adequate pharmacological treatment, is an important cornerstone in the management of T2DM, aiming to improve glycemic control, but also dyslipidemia and hypertension, for preventing or delaying diabetic complications. However, there is mounting evidence that Mediterranean dietary pattern might be more effective at improving glycemic control than the traditional carbohydrate-centered approach for management of T2DM (Papamichou et al., 2019), which to some extent can be attributed to the high amount of plant food bioactives, including polyphenols consummed (Guasch-Ferré et al., 2017). Studies have shown that polyphenols (anthocyanins, polyphenols from grape seed, or resveratrol, among others) exhibit antidiabetic properties by various mechanisms that include reduction of insulin resistance or improvement of glycemic control (Cao et al., 2019). Concordantly, our bioinformatic analyses showed a highly significant negative correlation between the gene expression profile of grape extract enriched with resveratrol and T2DM, as well as a highly significant positive correlation between the same bioactives and the antidiabetic drug metformin.

This systematic analysis presents however few limitations. Publications included in our analysis presented high heterogenicity regarding the study populations, duration, doses used for intervention, mode of extractions of total RNAs and analytical platforms employed. These differences may impact on the identification of differentially expressed genes following

bioactive intake and may explain why for some common genes, the expression profiles varied from one study to another. For these reasons, pathway analyses were performed without considering the directionality of the genomic effects. Also, this analysis pointed out that cellular pathways, functions and cell signalling regulations were affected by bioactives in humans and would need to be validated using different cellular and molecular methods.

In conclusion, the application of untargeted approaches in nutrigenomic studies allows for a complete coverage of the genomic effects of food ingredients under study, which also includes plant food bioactives such as polyphenols. The subsequent processing of obtained data using the available bioinformatic tools enables conceptualization and visualization of the genomic effects, thus generating information about the biological effects of the observed genomic modulations. In this study we applied for the first time an integrative multi-omics approach, for integration of available data from existing untargeted nutrigenomic studies evaluating cardiometabolic health benefits of polyphenols in humans, but also for comparison with data on the genomic effects of cardiometabolic diseases and common drugs used for treatment of cardiometabolic diseases and risk factors that are available from publicly available databases. This innovative approach allowed us to elucidate at least to some extent some of the key molecular mechanisms underlying the beneficial effects of polyphenols on cardiometabolic health in humans. Moreover, the results from the correlation analyses with genomic effects of cardiometabolic diseases and common drugs used for treatment of cardiometabolic diseases and risk factors are yet another confirmation of the beneficial effects of polyphenols on human cardiometabolic health, but on a deeper, molecular level. We believe that the results of previous human intervention studies, along with the results of our integrative bioinformatic analyses of available data, will encourage further research on nutrigenomic effects of polyphenols in diverse study population that will

include participants of both genders, various age groups, but also healthy people as well as patients with diverse pathologies. In that way, the collected data will allow to study also the interindividual variability in response to consumption of specific polyphenolic compounds and will provide a solid background for future nutrigenetic studies.

Acknowledgements

This article is based upon work from the COST Action FA1403 POSITIVe (Interindividual variation in response to consumption of plant food bioactives and determinants involved) supported by COST (European Cooperation in Science and Technology; www.cost.eu). Authors thank Zan Mitrev Klinik and Dr. Rodney Rosalia for their support. A. A.-A. and F. I. B. are Serra Húnter Fellows and thank the Serra Húnter Programme (Generalitat de Catalunya) for the academic positions with reference numbers URV-AG-587 and URV-LE-621, respectively. I.K. is supported by Ministry of Education, Science and Technological Development of the Republic of Serbia (451-03-9/2021-14/200015).

Funding

COST Action FA1403-European Cooperation in Science and Technology (www.cost.eu).

Author contribution

All authors contributed to the design and the editing of this review.

Conflict of interest

The authors have declared no conflict of interest.

Table 1

No.	Participants				Intervention			Study design	Significantly altered biomarkers of cardiometabolic health	Gene expression (mRNA or miRNA)	Method	Ref.
	Gender	Age (years)	N	Health status	Plant food/ extract/ bioactive	Dose	Duration of intervention					
1	M and F	38.0 ± 5.8	12	Obese (BMI between 30 and 40 kg/m^2)	Transresveratrol phosphate	300 mg transresveratrol phosphate (0.66 mmol) /day	28 days	Sequential design: 28 days placebo followed by 28 days transresveratrol phosphate	Decreased lipid peroxides Increased plasma total antioxidant power Increased glutathione peroxidase	mRNA	Macroarray Custom-made low-density array with 200 oligonucleotide probes	(De Groote et al., 2012)
	M and F	34.2 ±7.7	10	Obese (BMI between 30 and 40 kg/m^2)	Catechin-rich grape seed extract	400 mg (0.66 mmol) catechin-rich grape seed extract/day	28 days	Sequential design: 28 days placebo followed by 28 days catechin-rich grape seed extract	Increased glutathione peroxidase	mRNA	Macroarray Custom-made low-density array with 200 oligonucleotide probes	
2	M	50-65	10	Overweight	Hesperidin	500 ml control drink plus hesperidin (292 mg/day) *vs.* 500 ml contro	4 weeks	Randomized, double-blind, placebo-controlled, crossover study	Decreased diastolic blood pressure (Morand et al., 2011)	mRNA and miRNA	Microarray Commercial Operon array	(Milenkovic et al., 2011)

						l drink plus placebo						
3	M	Adults, up to 80 years old	18	Type 2 diabetes, hypertension, and coronary artery disease	Grape extract (GE) or grape extract plus resveratrol (GE-Res)	One capsule/day of GE, GE-Res, or placebo in the morning for the first 6 months, and 2 capsules/day for the following 6 months. The phenolic content of the GE and the GE-Res was very similar (151± 17 mg and 139±18 mg phenolics per capsule, respec	1 year	Randomized, triple-blind, placebo-controlled, dose-response, 1-year follow-up study with three parallel arms designated as placebo (maltodextrin), GE (conventional grape extract) and GE-Res (grape extract containing resveratrol)	Increased adiponectin in GE-Res group *vs.* placebo group Decreased plasminogen activator inhibitor type 1 (PAI1), total cholesterol, glucose, and hemoglobin A1c in GE-Res group *vs.* placebo group (Tomé-Carneiro et al., 2013a)	mRNA and miRNA	Microarray Commercial Affymetrix arrays	(Tomé-Carneiro et al., 2013b)

						tively) but GE-Res also contained 8.1±0.5 mg of resveratrol per capsule.						
4	M	30-60	7	Non-obese healthy smokers	Monomeric and oligomeric flavanols from grape seeds	200 mg/day	8 weeks	Randomized, double-blind, placebo-controlled, parallel study	Improvement of vascular health index Decreased TNF release (ex vivo, LPS induced) Decreased rate of collagen-induced aggregation of platelets (ex vivo) (Weseler et al., 2011)	mRNA	Microarray Commercial Agilent arrays	(Milenkovic et al., 2014)
5	M	33 ± 6 years, for the total study population (N=40)	10	Healthy	Wild blueberry anthocyanins	11 g wild blueberry powder, equivalent to 100 g fresh wild blueberries, containing 150 mg of anthocyanins	1 month	Two-arm, parallel, double-blind randomized controlled trial	Increased FMD Decreased 24-hour systolic blood pressure	mRNA and miRNA	Microarray Commercial Agilent arrays	(Rodriguez-Mateos et al., 2019)

						, dissolved in 500 ml water, bi-daily more than 28 days *vs.* matched control drink (11 g powder, bi-daily)						

Table 2

Genes in common for at least three of the analyzed publications			
No.	**Gene symbol**	**Gene name**	**Publications**
1	*SOX3*	SRY-Box Transcription Factor 3	[Publication 2] and [Publication 3] and [Publication 4] and [Publication 5]
2	*ZNF770*	Zinc Finger Protein 770	
3	*PRDX1*	Peroxiredoxin 1	[Publication 1] and [Publication 4] and [Publication 5]
4	*AJAP1*	Adherens Junctions Associated Protein 1	[Publication 2] and [Publication 3] and [Publication 4]
5	*FSTL4*	Follistatin Like 4	
6	*PSMB5*	Proteasome 20S Subunit Beta 5	[Publication 2] and [Publication 3] and [Publication 5]
7	*NDUFS2*	NADH:Ubiquinone Oxidoreductase Core Subunit S2	
8	*AHCTF1*	AT-Hook Containing Transcription Factor 1	
9	*SSR1*	Signal Sequence Receptor Subunit 1	
10	*ATP7A*	ATPase Copper Transporting Alpha	
11	*TMEM201*	Transmembrane Protein 201	[Publication 2] and [Publication 4] and [Publication 5]
12	*POLR2G*	RNA Polymerase II Subunit G	
13	*HRASLS5*	Phospholipase A And Acyltransferase 5	
14	*MRPS18C*	Mitochondrial Ribosomal Protein S18C	
15	*GJC3*	Gap Junction Protein Gamma 3	
16	*KRTAP12-2*	Keratin Associated Protein 12-2	[Publication 3] and [Publication 4] and [Publication 5]
17	*SUCLA2*	Succinate-CoA Ligase ADP-Forming Subunit Beta	

Table legends

Table 1. Intervention studies on nutrigenomic effects of polyphenols accompanied with favorable cardiometabolic outcomes, assessed in blood samples with macro- or microarray methods.

Table 2. Genes in common for at least three of the analyzed publications.

Publication 1 (De Groote et al., 2012); bioactives - transresveratrol phosphate and catechin-rich grape seed extract. Publication 2 (Milenkovic et al., 2011); bioactive - hesperidin. Publication 3 (Tomé-Carneiro et al., 2013b); bioactives - grape extract plus resveratrol. Publication 4 (Milenkovic et al., 2014); bioactives - monomeric and oligomeric flavanols from grape seeds. Publication 5 (Rodriguez-Mateos et al., 2019); bioactives - wild blueberry anthocyanins.

Figure legends

Figure 1. Flowchart of the literature search and data extraction.

Figure 2. Pathways related to cellular processes that are in common for at least three of the analyzed publications.

Publication 1 (De Groote et al., 2012); bioactives - transresveratrol phosphate and catechin-rich grape seed extract. Publication 2 (Milenkovic et al., 2011); bioactive - hesperidin. Publication 3 (Tomé-Carneiro et al., 2013b); bioactives - grape extract plus resveratrol. Publication 4 (Milenkovic et al., 2014); bioactives - monomeric and oligomeric flavanols from grape seeds. Publication 5 (Rodriguez-Mateos et al., 2019); bioactives - wild blueberry anthocyanins.

Green circle: identified from the publication genomic data; red circle: not identified from the publication genomic data.

Figure 3. Networks of pathways related to cellular processes that are in common for at least three of the analyzed publications.

A) Networks of pathways related to specific cellular processes. B) Global network of pathways.

Figure 4. Potential transcription factors which activity could be modulated by polyphenols.

Set A - Publication 1 (De Groote et al., 2012); bioactives - transresveratrol phosphate and catechin-rich grape seed extract. Set B - Publication 2 (Milenkovic et al., 2011); bioactive - hesperidin. Set C - Publication 3 (Tomé-Carneiro et al., 2013b); bioactives - grape extract plus resveratrol. Set D - Publication 4 (Milenkovic et al., 2014); bioactives - monomeric and

oligomeric flavanols from grape seeds. Set E - Publication 5 (Rodriguez-Mateos et al., 2019); bioactives - wild blueberry anthocyanins.

Figure 5. miRNAs that have been identified as differentially expressed following consumption of polyphenols.

Publication 2 (Milenkovic et al., 2011); bioactive - hesperidin. Publication 3 (Tomé-Carneiro et al., 2013b); bioactives - grape extract plus resveratrol. Publication 5 (Rodriguez-Mateos et al., 2019); bioactives - wild blueberry anthocyanins.

Figure 6. miRNAs and their targets networks. Blue circles present miRNAs and yellow circles target genes of miRNAs.

A) Publication 2 (Milenkovic et al., 2011); bioactive - hesperidin. B) Publication 3 (Tomé-Carneiro et al., 2013b); bioactives - grape extract plus resveratrol. C) Publication 5 (Rodriguez-Mateos et al., 2019); bioactives - wild blueberry anthocyanins.

Figure 7. Functional enrichment analysis of differentially expressed miRNAs' targets.

Dot plot of functional enrichment analysis for target genes of selected miRNAs resulting from the enrichment analysis. The Y-axis reports the annotation categories (e.g., KEGG pathways) and the X-axis reports the selected miRNAs. The color of the dots represents the adjusted p-values, and the size of the dots represents gene ratio.

A) Publication 2 (Milenkovic et al., 2011); bioactive - hesperidin. B) Publication 3 (Tomé-Carneiro et al., 2013b); bioactives - grape extract plus resveratrol. C) Publication 5 (Rodriguez-Mateos et al., 2019); bioactives - wild blueberry anthocyanins.

Figure 8. Comparison of significantly over-represented pathways for miRNAs' targets.

Publication 2 (Milenkovic et al., 2011); bioactive - hesperidin. Publication 3 (Tomé-Carneiro et al., 2013b); bioactives - grape extract plus resveratrol. Publication 5 (Rodriguez-Mateos et al., 2019); bioactives - wild blueberry anthocyanins.

Figure 9. Integrated analysis of protein coding and non-coding gene expression.

A) Comparison of differentially expressed genes with the target genes of miRNAs identified as differentially expressed from the same studies. Publication 2 (Milenkovic et al., 2011); bioactive - hesperidin. Publication 3 (Tomé-Carneiro et al., 2013b); bioactives - grape extract plus resveratrol. Publication 5 (Rodriguez-Mateos et al., 2019); bioactives - wild blueberry anthocyanins. B) Comparison of pathways identified for differentially expressed genes and targets of miRNAs. C) Pathways are affected by polyphenols by modulating the expression of protein coding genes and miRNAs.

Figure 10. Association of polyphenol modulated genes with human diseases.

Publication 2 (Milenkovic et al., 2011); bioactive - hesperidin. Publication 3 (Tomé-Carneiro et al., 2013b); bioactives - grape extract plus resveratrol. Publication 4 (Milenkovic et al., 2014); bioactives - monomeric and oligomeric flavanols from grape seeds. Publication 5 (Rodriguez-Mateos et al., 2019); bioactives - wild blueberry anthocyanins.

Figure 11. Correlations between polyphenol modulated genes and gene expression profiles of cardiometabolic diseases.

Blueberry - Publication 5 (Rodriguez-Mateos et al., 2019); Grape seed extract - Publication 4 (Milenkovic et al., 2014); Resveratrol - Publication 3 (Tomé-Carneiro et al., 2013b). Gene expression profiles associated with cardiometabolic diseases were obtained from Gene Expression Omnibus database: Metabolic syndrome - GSE98895; T2DM - GSE23561; Arterial stiffness and hypertension - GSE6599.

Figure 12. Correlations between polyphenol modulated genes and drug effects gene expression profiles.

Blueberry - Publication 5 (Rodriguez-Mateos et al., 2019); Hesperidin - Publication 2 (Milenkovic et al., 2011); Resveratrol - Publication 3 (Tomé-Carneiro et al., 2013b). Gene expression profiles associated with drug intake were obtained from Gene Expression Omnibus database: Statin study 1 - GSE11393; Statin study 2 - GSE71220; Metformin - GSE153318; Losartan - GSE37824; Telmisartan + Amlodipine - GES42808; Telmisartan - GES42808.

References

Afman, L., Milenkovic, D., Roche, H.M., 2014. Nutritional aspects of metabolic inflammation in relation to health--insights from transcriptomic biomarkers in PBMC of fatty acids and polyphenols. Mol Nutr Food Res 58, 1708-1720.

Alberti, K.G., Eckel, R.H., Grundy, S.M., Zimmet, P.Z., Cleeman, J.I., Donato, K.A., Fruchart, J.C., James, W.P., Loria, C.M., Smith, S.C., Jr., 2009. Harmonizing the metabolic syndrome: a joint interim statement of the International Diabetes Federation Task Force on Epidemiology and Prevention; National Heart, Lung, and Blood Institute; American Heart Association; World Heart Federation; International Atherosclerosis Society; and International Association for the Study of Obesity. Circulation 120, 1640-1645.

Alexandrow, M.G., Song, L.J., Altiok, S., Gray, J., Haura, E.B., Kumar, N.B., 2012. Curcumin: a novel Stat3 pathway inhibitor for chemoprevention of lung cancer. Eur J Cancer Prev 21, 407-412.

Asbaghi, O., Nazarian, B., Reiner, Ž., Amirani, E., Kolahdooz, F., Chamani, M., Asemi, Z., 2020. The effects of grape seed extract on glycemic control, serum lipoproteins, inflammation, and body weight: A systematic review and meta-analysis of randomized controlled trials. Phytother Res 34, 239-253.

Auclair, S., Milenkovic, D., Besson, C., Chauvet, S., Gueux, E., Morand, C., Mazur, A., Scalbert, A., 2009. Catechin reduces atherosclerotic lesion development in apo E-deficient mice: a transcriptomic study. Atherosclerosis 204, e21-27.

Bartel, D.P., 2004. MicroRNAs: genomics, biogenesis, mechanism, and function. Cell 116, 281-297.

Bielska, A., Niemira, M., Kretowski, A., 2021. Recent Highlights of Research on miRNAs as Early Potential Biomarkers for Cardiovascular Complications of Type 2 Diabetes Mellitus. Int J Mol Sci 22.

Blankenberg, S., Rupprecht, H.J., Bickel, C., Torzewski, M., Hafner, G., Tiret, L., Smieja, M., Cambien, F., Meyer, J., Lackner, K.J., 2003. Glutathione peroxidase 1 activity and cardiovascular events in patients with coronary artery disease. N Engl J Med 349, 1605-1613.

Breuss, J.M., Atanasov, A.G., Uhrin, P., 2019. Resveratrol and Its Effects on the Vascular System. Int J Mol Sci 20.

Buniello, A., MacArthur, J.A.L., Cerezo, M., Harris, L.W., Hayhurst, J., Malangone, C., McMahon, A., Morales, J., Mountjoy, E., Sollis, E., Suveges, D., Vrousgou, O., Whetzel, P.L., Amode, R., Guillen, J.A., Riat, H.S., Trevanion, S.J., Hall, P., Junkins, H., Flicek, P., Burdett, T., Hindorff, L.A., Cunningham, F., Parkinson, H., 2019. The NHGRI-EBI GWAS Catalog of published genome-wide association studies, targeted arrays and summary statistics 2019. Nucleic Acids Res 47, D1005-d1012.

Cao, H., Ou, J., Chen, L., Zhang, Y., Szkudelski, T., Delmas, D., Daglia, M., Xiao, J., 2019. Dietary polyphenols and type 2 diabetes: Human Study and Clinical Trial. Crit Rev Food Sci Nutr 59, 3371-3379.

Chen, E.Y., Tan, C.M., Kou, Y., Duan, Q., Wang, Z., Meirelles, G.V., Clark, N.R., Ma'ayan, A., 2013. Enrichr: interactive and collaborative HTML5 gene list enrichment analysis tool. BMC Bioinformatics 14, 128.

Chen, Q., Lv, J., Yang, W., Xu, B., Wang, Z., Yu, Z., Wu, J., Yang, Y., Han, Y., 2019. Targeted inhibition of STAT3 as a potential treatment strategy for atherosclerosis. Theranostics 9, 6424-6442.

Chen, Y., Zhao, X., Wu, H., 2021. Transcriptional Programming in Arteriosclerotic Disease: A Multifaceted Function of the Runx2 (Runt-Related Transcription Factor 2). Arterioscler Thromb Vasc Biol 41, 20-34.

Chu, A.J., 2014. Antagonism by bioactive polyphenols against inflammation: a systematic view. Inflamm Allergy Drug Targets 13, 34-64.

Claude, S., Boby, C., Rodriguez-Mateos, A., Spencer, J.P., Gerard, N., Morand, C., Milenkovic, D., 2014. Flavanol metabolites reduce monocyte adhesion to endothelial cells through modulation of expression of genes via p38-MAPK and p65-Nf-kB pathways. Mol Nutr Food Res 58, 1016-1027.

Coban, D., Milenkovic, D., Chanet, A., Khallou-Laschet, J., Sabbe, L., Palagani, A., Vanden Berghe, W., Mazur, A., Morand, C., 2012. Dietary curcumin inhibits atherosclerosis by affecting the expression of genes involved in leukocyte adhesion and transendothelial migration. Mol Nutr Food Res 56, 1270-1281.

Cook, N.R., Cohen, J., Hebert, P.R., Taylor, J.O., Hennekens, C.H., 1995. Implications of small reductions in diastolic blood pressure for primary prevention. Arch Intern Med 155, 701-709.

Corral-Jara, K.F., Nuthikattu, S., Rutledge, J., Villablanca, A., Morand, C., Schroeter, H., Milenkovic, D., 2021. Integrated Multi-Omic Analyses of the Genomic Modifications by Gut Microbiome-Derived Metabolites of Epicatechin, 5-(4'-Hydroxyphenyl)-γ-Valerolactone, in TNFalpha-Stimulated Primary Human Brain Microvascular Endothelial Cells. Front Neurosci 15, 622640.

Crescenti, A., Solà, R., Valls, R.M., Caimari, A., Del Bas, J.M., Anguera, A., Anglés, N., Arola, L., 2013. Cocoa Consumption Alters the Global DNA Methylation of Peripheral Leukocytes in Humans with Cardiovascular Disease Risk Factors: A Randomized Controlled Trial. PLoS One 8, e65744.

D'Amore, S., Härdfeldt, J., Cariello, M., Graziano, G., Copetti, M., Di Tullio, G., Piglionica, M., Scialpi, N., Sabbà, C., Palasciano, G., Vacca, M., Moschetta, A., 2018. Identification of miR-9-5p as direct regulator of ABCA1 and HDL-driven reverse cholesterol transport in circulating CD14+ cells of patients with metabolic syndrome. Cardiovasc Res 114, 1154-1164.

Davis, A.P., Grondin, C.J., Johnson, R.J., Sciaky, D., Wiegers, J., Wiegers, T.C., Mattingly, C.J., 2021. Comparative Toxicogenomics Database (CTD): update 2021. Nucleic Acids Res 49, D1138-d1143.

de Boer, V.C., van Schothorst, E.M., Dihal, A.A., van der Woude, H., Arts, I.C., Rietjens, I.M., Hollman, P.C., Keijer, J., 2006. Chronic quercetin exposure affects fatty acid catabolism in rat lung. Cell Mol Life Sci 63, 2847-2858.

De Groote, D., Van Belleghem, K., Devière, J., Van Brussel, W., Mukaneza, A., Amininejad, L., 2012. Effect of the intake of resveratrol, resveratrol phosphate, and catechin-rich grape

seed extract on markers of oxidative stress and gene expression in adult obese subjects. Ann Nutr Metab 61, 15-24.
de Queiroz, K.B., Dos Santos Fontes Pereira, T., Araújo, M.S.S., Gomez, R.S., Coimbra, R.S., 2018. Resveratrol Acts Anti-Inflammatory and Neuroprotective in an Infant Rat Model of Pneumococcal Meningitis by Modulating the Hippocampal miRNome. Mol Neurobiol 55, 8869-8884.
Del Rio, D., Rodriguez-Mateos, A., Spencer, J.P., Tognolini, M., Borges, G., Crozier, A., 2013. Dietary (poly)phenolics in human health: structures, bioavailability, and evidence of protective effects against chronic diseases. Antioxid Redox Signal 18, 1818-1892.
Dessau, R.B., Pipper, C.B., 2008. ["R"--project for statistical computing]. Ugeskr Laeger 170, 328-330.
Dong, J.T., Chen, C., 2009. Essential role of KLF5 transcription factor in cell proliferation and differentiation and its implications for human diseases. Cell Mol Life Sci 66, 2691-2706.
Dostal, Z., Sebera, M., Srovnal, J., Staffova, K., Modriansky, M., 2021. Dual Effect of Taxifolin on ZEB2 Cancer Signaling in HepG2 Cells. Molecules 26.
Emanueli, C., Meloni, M., Hasan, W., Habecker, B.A., 2014. The biology of neurotrophins: cardiovascular function. Handb Exp Pharmacol 220, 309-328.
Ferreira, A.C., Serejo, J.S., Durans, R., Pereira Costa, J.M., Maciel, A.W.S., Vieira, A.S.M., Dias-Filho, C.A.A., Dias, C.J., Bomfim, M.R.Q., Mostarda, C.T., Brito-Monzani, J.O., 2020. Dose-related Effects of Resveratrol in Different Models of Pulmonary Arterial Hypertension: A Systematic Review. Curr Cardiol Rev 16, 231-240.
Fiordelisi, A., Iaccarino, G., Morisco, C., Coscioni, E., Sorriento, D., 2019. NFkappaB is a Key Player in the Crosstalk between Inflammation and Cardiovascular Diseases. Int J Mol Sci 20.
Friedman, R.C., Farh, K.K., Burge, C.B., Bartel, D.P., 2009. Most mammalian mRNAs are conserved targets of microRNAs. Genome Res 19, 92-105.
Fuchs, D., Vafeiadou, K., Hall, W.L., Daniel, H., Williams, C.M., Schroot, J.H., Wenzel, U., 2007. Proteomic biomarkers of peripheral blood mononuclear cells obtained from postmenopausal women undergoing an intervention with soy isoflavones. Am J Clin Nutr 86, 1369-1375.
García-Conesa, M.T., Tribolo, S., Guyot, S., Tomás-Barberán, F.A., Kroon, P.A., 2009. Oligomeric procyanidins inhibit cell migration and modulate the expression of migration and proliferation associated genes in human umbilical vascular endothelial cells. Mol Nutr Food Res 53, 266-276.
Gluvic, Z., Zaric, B., Resanovic, I., Obradovic, M., Mitrovic, A., Radak, D., Isenovic, E.R., 2017. Link between Metabolic Syndrome and Insulin Resistance. Curr Vasc Pharmacol 15, 30-39.
Grassi, D., Desideri, G., Necozione, S., Lippi, C., Casale, R., Properzi, G., Blumberg, J.B., Ferri, C., 2008. Blood pressure is reduced and insulin sensitivity increased in glucose-intolerant, hypertensive subjects after 15 days of consuming high-polyphenol dark chocolate. J Nutr 138, 1671-1676.
Grayson, B.L., Wang, L., Aune, T.M., 2011. Peripheral blood gene expression profiles in metabolic syndrome, coronary artery disease and type 2 diabetes. Genes Immun 12, 341-351.

Grosso, G., Stepaniak, U., Micek, A., Stefler, D., Bobak, M., Pająk, A., 2017. Dietary polyphenols are inversely associated with metabolic syndrome in Polish adults of the HAPIEE study. Eur J Nutr 56, 1409-1420.

Guasch-Ferré, M., Merino, J., Sun, Q., Fitó, M., Salas-Salvadó, J., 2017. Dietary Polyphenols, Mediterranean Diet, Prediabetes, and Type 2 Diabetes: A Narrative Review of the Evidence. Oxid Med Cell Longev 2017, 6723931.

Han, C.L., Qu, C.Z., 2020. Cardiovascular Risk and Safety Evaluation of a Dual Peroxisome Proliferator-Activated Receptor-Alpha/Gamma Agonist, Aleglitazar, in Patients With Type 2 Diabetes: A Meta-analysis. J Cardiovasc Pharmacol 75, 351-357.

Han, H., Cho, J.W., Lee, S., Yun, A., Kim, H., Bae, D., Yang, S., Kim, C.Y., Lee, M., Kim, E., Lee, S., Kang, B., Jeong, D., Kim, Y., Jeon, H.N., Jung, H., Nam, S., Chung, M., Kim, J.H., Lee, I., 2018. TRRUST v2: an expanded reference database of human and mouse transcriptional regulatory interactions. Nucleic Acids Res 46, D380-D386.

Harati, K., Slodnik, P., Chromik, A.M., Goertz, O., Hirsch, T., Kapalschinski, N., Klein-Hitpass, L., Kolbenschlag, J., Uhl, W., Lehnhardt, M., Daigeler, A., 2015. Resveratrol induces apoptosis and alters gene expression in human fibrosarcoma cells. Anticancer Res 35, 767-774.

Heberle, H., Meirelles, G.V., da Silva, F.R., Telles, G.P., Minghim, R., 2015. InteractiVenn: a web-based tool for the analysis of sets through Venn diagrams. BMC Bioinformatics 16, 169.

Hertog, M.G., Feskens, E.J., Hollman, P.C., Katan, M.B., Kromhout, D., 1993. Dietary antioxidant flavonoids and risk of coronary heart disease: the Zutphen Elderly Study. Lancet 342, 1007-1011.

Hori, D., Dunkerly-Eyring, B., Nomura, Y., Biswas, D., Steppan, J., Henao-Mejia, J., Adachi, H., Santhanam, L., Berkowitz, D.E., Steenbergen, C., Flavell, R.A., Das, S., 2017. miR-181b regulates vascular stiffness age dependently in part by regulating TGF-β signaling. PLoS One 12, e0174108.

Hoseini, A., Namazi, G., Farrokhian, A., Reiner, Ž., Aghadavod, E., Bahmani, F., Asemi, Z., 2019. The effects of resveratrol on metabolic status in patients with type 2 diabetes mellitus and coronary heart disease. Food Funct 10, 6042-6051.

Hussain, T., Murtaza, G., Yang, H., Kalhoro, M.S., Kalhoro, D.H., 2020. Exploiting Anti-Inflammation Effects of Flavonoids in Chronic Inflammatory Diseases. Curr Pharm Des 26, 2610-2619.

Igwe, E.O., Charlton, K.E., Probst, Y.C., 2019. Usual dietary anthocyanin intake, sources and their association with blood pressure in a representative sample of Australian adults. J Hum Nutr Diet 32, 578-590.

Iwasaki, A., Matsumori, A., Yamada, T., Shioi, T., Wang, W., Ono, K., Nishio, R., Okada, M., Sasayama, S., 1999. Pimobendan inhibits the production of proinflammatory cytokines and gene expression of inducible nitric oxide synthase in a murine model of viral myocarditis. J Am Coll Cardiol 33, 1400-1407.

Johnson, S.A., Navaei, N., Pourafshar, S., Jaime, S.J., Akhavan, N.S., Alvarez-Alvarado, S., Proaño, G.V., Litwin, N.S., Clark, E.A., Foley, E.M., George, K.S., Elam, M.L., Payton, M.E., Arjmandi, B.H., Figueroa, A., 2020. Effects of Montmorency Tart Cherry Juice Consumption on Cardiometabolic Biomarkers in Adults with Metabolic Syndrome: A Randomized Controlled Pilot Trial. J Med Food 23, 1238-1247.

Kaneto, C.M., Nascimento, J.S., Moreira, M.C.R., Ludovico, N.D., Santana, A.P., Silva, R.A.A., Silva-Jardim, I., Santos, J.L., Sousa, S.M.B., Lima, P.S.P., 2017. MicroRNA profiling identifies miR-7-5p and miR-26b-5p as differentially expressed in hypertensive patients with left ventricular hypertrophy. Braz J Med Biol Res 50, e6211.

Kerimi, A., Williamson, G., 2016. At the interface of antioxidant signalling and cellular function: Key polyphenol effects. Mol Nutr Food Res 60, 1770-1788.

Konings, E., Timmers, S., Boekschoten, M.V., Goossens, G.H., Jocken, J.W., Afman, L.A., Müller, M., Schrauwen, P., Mariman, E.C., Blaak, E.E., 2014. The effects of 30 days resveratrol supplementation on adipose tissue morphology and gene expression patterns in obese men. Int J Obes (Lond) 38, 470-473.

Krga, I., Milenkovic, D., 2019. Anthocyanins: From Sources and Bioavailability to Cardiovascular-Health Benefits and Molecular Mechanisms of Action. J Agric Food Chem 67, 1771-1783.

Krga, I., Milenkovic, D., Morand, C., Monfoulet, L.E., 2016. An update on the role of nutrigenomic modulations in mediating the cardiovascular protective effect of fruit polyphenols. Food Funct 7, 3656-3676.

Krga, I., Tamaian, R., Mercier, S., Boby, C., Monfoulet, L.E., Glibetic, M., Morand, C., Milenkovic, D., 2018. Anthocyanins and their gut metabolites attenuate monocyte adhesion and transendothelial migration through nutrigenomic mechanisms regulating endothelial cell permeability. Free Radic Biol Med 124, 364-379.

Kuleshov, M.V., Jones, M.R., Rouillard, A.D., Fernandez, N.F., Duan, Q., Wang, Z., Koplev, S., Jenkins, S.L., Jagodnik, K.M., Lachmann, A., McDermott, M.G., Monteiro, C.D., Gundersen, G.W., Ma'ayan, A., 2016. Enrichr: a comprehensive gene set enrichment analysis web server 2016 update. Nucleic Acids Res 44, W90-97.

LeBlanc, M., Zuber, V., Andreassen, B.K., Witoelar, A., Zeng, L., Bettella, F., Wang, Y., McEvoy, L.K., Thompson, W.K., Schork, A.J., Reppe, S., Barrett-Connor, E., Ligthart, S., Dehghan, A., Gautvik, K.M., Nelson, C.P., Schunkert, H., Samani, N.J., Ridker, P.M., Chasman, D.I., Aukrust, P., Djurovic, S., Frigessi, A., Desikan, R.S., Dale, A.M., Andreassen, O.A., 2016. Identifying Novel Gene Variants in Coronary Artery Disease and Shared Genes With Several Cardiovascular Risk Factors. Circ Res 118, 83-94.

Li, D., Zhang, Y., Liu, Y., Sun, R., Xia, M., 2015. Purified anthocyanin supplementation reduces dyslipidemia, enhances antioxidant capacity, and prevents insulin resistance in diabetic patients. J Nutr 145, 742-748.

Li, Z., Zhao, H., Wang, J., 2021. Metabolism and Chronic Inflammation: The Links Between Chronic Heart Failure and Comorbidities. Front Cardiovasc Med 8, 650278.

Licursi, V., Conte, F., Fiscon, G., Paci, P., 2019. MIENTURNET: an interactive web tool for microRNA-target enrichment and network-based analysis. BMC Bioinformatics 20, 545.

Lind, L., 2019. Genome-Wide Association Study of the Metabolic Syndrome in UK Biobank. Metab Syndr Relat Disord 17, 505-511.

Liu, C., Kraja, A.T., Smith, J.A., Brody, J.A., Franceschini, N., Bis, J.C., Rice, K., Morrison, A.C., Lu, Y., Weiss, S., Guo, X., Palmas, W., Martin, L.W., Chen, Y.D., Surendran, P., Drenos, F., Cook, J.P., Auer, P.L., Chu, A.Y., Giri, A., Zhao, W., Jakobsdottir, J., Lin, L.A., Stafford, J.M., Amin, N., Mei, H., Yao, J., Voorman, A., Larson, M.G., Grove, M.L., Smith, A.V., Hwang, S.J., Chen, H., Huan, T., Kosova, G., Stitziel, N.O., Kathiresan, S., Samani, N., Schunkert, H., Deloukas, P., Li, M., Fuchsberger, C., Pattaro, C., Gorski, M., Kooperberg, C.,

Papanicolaou, G.J., Rossouw, J.E., Faul, J.D., Kardia, S.L., Bouchard, C., Raffel, L.J., Uitterlinden, A.G., Franco, O.H., Vasan, R.S., O'Donnell, C.J., Taylor, K.D., Liu, K., Bottinger, E.P., Gottesman, O., Daw, E.W., Giulianini, F., Ganesh, S., Salfati, E., Harris, T.B., Launer, L.J., Dörr, M., Felix, S.B., Rettig, R., Völzke, H., Kim, E., Lee, W.J., Lee, I.T., Sheu, W.H., Tsosie, K.S., Edwards, D.R., Liu, Y., Correa, A., Weir, D.R., Völker, U., Ridker, P.M., Boerwinkle, E., Gudnason, V., Reiner, A.P., van Duijn, C.M., Borecki, I.B., Edwards, T.L., Chakravarti, A., Rotter, J.I., Psaty, B.M., Loos, R.J., Fornage, M., Ehret, G.B., Newton-Cheh, C., Levy, D., Chasman, D.I., 2016a. Meta-analysis identifies common and rare variants influencing blood pressure and overlapping with metabolic trait loci. Nat Genet 48, 1162-1170.

Liu, C., Sun, J., Lu, Y., Bo, Y., 2016b. Effects of Anthocyanin on Serum Lipids in Dyslipidemia Patients: A Systematic Review and Meta-Analysis. PLoS One 11, e0162089.

Llaverias, G., Pou, J., Ros, E., Zambón, D., Cofán, M., Sánchez, A., Vázquez-Carrera, M., Sánchez, R.M., Laguna, J.C., Alegret, M., 2008. Monocyte gene-expression profile in men with familial combined hyperlipidemia and its modification by atorvastatin treatment. Pharmacogenomics 9, 1035-1054.

Lotta, L.A., Gulati, P., Day, F.R., Payne, F., Ongen, H., van de Bunt, M., Gaulton, K.J., Eicher, J.D., Sharp, S.J., Luan, J., De Lucia Rolfe, E., Stewart, I.D., Wheeler, E., Willems, S.M., Adams, C., Yaghootkar, H., Forouhi, N.G., Khaw, K.T., Johnson, A.D., Semple, R.K., Frayling, T., Perry, J.R., Dermitzakis, E., McCarthy, M.I., Barroso, I., Wareham, N.J., Savage, D.B., Langenberg, C., O'Rahilly, S., Scott, R.A., 2017. Integrative genomic analysis implicates limited peripheral adipose storage capacity in the pathogenesis of human insulin resistance. Nat Genet 49, 17-26.

Luo, F., Das, A., Chen, J., Wu, P., Li, X., Fang, Z., 2019. Metformin in patients with and without diabetes: a paradigm shift in cardiovascular disease management. Cardiovasc Diabetol 18, 54.

Matsumori, A., Nunokawa, Y., Sasayama, S., 2000. Pimobendan inhibits the activation of transcription factor NF-kappaB: a mechanism which explains its inhibition of cytokine production and inducible nitric oxide synthase. Life Sci 67, 2513-2519.

Matys, V., Fricke, E., Geffers, R., Gössling, E., Haubrock, M., Hehl, R., Hornischer, K., Karas, D., Kel, A.E., Kel-Margoulis, O.V., Kloos, D.U., Land, S., Lewicki-Potapov, B., Michael, H., Münch, R., Reuter, I., Rotert, S., Saxel, H., Scheer, M., Thiele, S., Wingender, E., 2003. TRANSFAC: transcriptional regulation, from patterns to profiles. Nucleic Acids Res 31, 374-378.

Mauray, A., Felgines, C., Morand, C., Mazur, A., Scalbert, A., Milenkovic, D., 2012. Bilberry anthocyanin-rich extract alters expression of genes related to atherosclerosis development in aorta of apo E-deficient mice. Nutr Metab Cardiovasc Dis 22, 72-80.

Mehta, R., Otgonsuren, M., Younoszai, Z., Allawi, H., Raybuck, B., Younossi, Z., 2016. Circulating miRNA in patients with non-alcoholic fatty liver disease and coronary artery disease. BMJ Open Gastroenterol 3, e000096.

Milenkovic, D., Berghe, W.V., Morand, C., Claude, S., van de Sandt, A., Gorressen, S., Monfoulet, L.E., Chirumamilla, C.S., Declerck, K., Szic, K.S.V., Lahtela-Kakkonen, M., Gerhauser, C., Merx, M.W., Kelm, M., 2018. A systems biology network analysis of nutri(epi)genomic changes in endothelial cells exposed to epicatechin metabolites. Sci Rep 8, 15487.

Milenkovic, D., Deval, C., Dubray, C., Mazur, A., Morand, C., 2011. Hesperidin displays relevant role in the nutrigenomic effect of orange juice on blood leukocytes in human volunteers: a randomized controlled cross-over study. PLoS One 6, e26669.

Milenkovic, D., Krga, I., Dinel, A.-L., Morand, C., Laye, S., Castanon, N., 2021. Nutrigenomic modification induced by anthocyanin-rich bilberry extract in the hippocampus of ApoE-/- mice. Journal of Functional Foods 85, 104609.

Milenkovic, D., Vanden Berghe, W., Boby, C., Leroux, C., Declerck, K., Szarc vel Szic, K., Heyninck, K., Laukens, K., Bizet, M., Defrance, M., Dedeurwaerder, S., Calonne, E., Fuks, F., Haegeman, G., Haenen, G.R., Bast, A., Weseler, A.R., 2014. Dietary flavanols modulate the transcription of genes associated with cardiovascular pathology without changes in their DNA methylation state. PLoS One 9, e95527.

Moher, D., Liberati, A., Tetzlaff, J., Altman, D.G., 2009. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. Ann Intern Med 151, 264-269, w264.

Monfoulet, L.E., Ruskovska, T., Ajdžanović, V., Havlik, J., Vauzour, D., Bayram, B., Krga, I., Corral-Jara, K.F., Kistanova, E., Abadjieva, D., Massaro, M., Scoditti, E., Deligiannidou, E., Kontogiorgis, C., Arola-Arnal, A., van Schothorst, E.M., Morand, C., Milenkovic, D., 2021. Molecular Determinants of the Cardiometabolic Improvements of Dietary Flavanols Identified by an Integrative Analysis of Nutrigenomic Data from a Systematic Review of Animal Studies. Mol Nutr Food Res, e2100227.

Morand, C., Dubray, C., Milenkovic, D., Lioger, D., Martin, J.F., Scalbert, A., Mazur, A., 2011. Hesperidin contributes to the vascular protective effects of orange juice: a randomized crossover study in healthy volunteers. Am J Clin Nutr 93, 73-80.

Most, J., Timmers, S., Warnke, I., Jocken, J.W., van Boekschoten, M., de Groot, P., Bendik, I., Schrauwen, P., Goossens, G.H., Blaak, E.E., 2016. Combined epigallocatechin-3-gallate and resveratrol supplementation for 12 wk increases mitochondrial capacity and fat oxidation, but not insulin sensitivity, in obese humans: a randomized controlled trial. Am J Clin Nutr 104, 215-227.

Most, J., Warnke, I., Boekschoten, M.V., Jocken, J.W.E., de Groot, P., Friedel, A., Bendik, I., Goossens, G.H., Blaak, E.E., 2018. The effects of polyphenol supplementation on adipose tissue morphology and gene expression in overweight and obese humans. Adipocyte 7, 190-196.

Mozaffarian, D., Benjamin, E.J., Go, A.S., Arnett, D.K., Blaha, M.J., Cushman, M., Das, S.R., de Ferranti, S., Després, J.P., Fullerton, H.J., Howard, V.J., Huffman, M.D., Isasi, C.R., Jiménez, M.C., Judd, S.E., Kissela, B.M., Lichtman, J.H., Lisabeth, L.D., Liu, S., Mackey, R.H., Magid, D.J., McGuire, D.K., Mohler, E.R., 3rd, Moy, C.S., Muntner, P., Mussolino, M.E., Nasir, K., Neumar, R.W., Nichol, G., Palaniappan, L., Pandey, D.K., Reeves, M.J., Rodriguez, C.J., Rosamond, W., Sorlie, P.D., Stein, J., Towfighi, A., Turan, T.N., Virani, S.S., Woo, D., Yeh, R.W., Turner, M.B., 2016. Heart Disease and Stroke Statistics-2016 Update: A Report From the American Heart Association. Circulation 133, e38-360.

Mulhem, A., Moulla, Y., Klöting, N., Ebert, T., Tönjes, A., Fasshauer, M., Dietrich, A., Schön, M.R., Stumvoll, M., Richter, V., Blüher, M., 2021. Circulating cell adhesion molecules in metabolically healthy obesity. Int J Obes (Lond) 45, 331-336.

Mullins, A.P., Arjmandi, B.H., 2021. Health Benefits of Plant-Based Nutrition: Focus on Beans in Cardiometabolic Diseases. Nutrients 13, 519.

Nariman-Saleh-Fam, Z., Vahed, S.Z., Aghaee-Bakhtiari, S.H., Daraei, A., Saadatian, Z., Kafil, H.S., Yousefi, B., Eyvazi, S., Khaheshi, I., Parsa, S.A., Moravej, A., Mousavi, N., Bastami, M., Mansoori, Y., 2019. Expression pattern of miR-21, miR-25 and PTEN in peripheral blood mononuclear cells of patients with significant or insignificant coronary stenosis. Gene 698, 170-178.

Obeidat, M., Fishbane, N., Nie, Y., Chen, V., Hollander, Z., Tebbutt, S.J., Bossé, Y., Ng, R.T., Miller, B.E., McManus, B., Rennard, S., Paré, P.D., Sin, D.D., 2015. The Effect of Statins on Blood Gene Expression in COPD. PLoS One 10, e0140022.

Panday, A., Inda, M.E., Bagam, P., Sahoo, M.K., Osorio, D., Batra, S., 2016. Transcription Factor NF-κB: An Update on Intervention Strategies. Arch Immunol Ther Exp (Warsz) 64, 463-483.

Papamichou, D., Panagiotakos, D.B., Itsiopoulos, C., 2019. Dietary patterns and management of type 2 diabetes: A systematic review of randomised clinical trials. Nutr Metab Cardiovasc Dis 29, 531-543.

Piñero, J., Ramírez-Anguita, J.M., Saüch-Pitarch, J., Ronzano, F., Centeno, E., Sanz, F., Furlong, L.I., 2020. The DisGeNET knowledge platform for disease genomics: 2019 update. Nucleic Acids Res 48, D845-d855.

Pollack, R.M., Barzilai, N., Anghel, V., Kulkarni, A.S., Golden, A., O'Broin, P., Sinclair, D.A., Bonkowski, M.S., Coleville, A.J., Powell, D., Kim, S., Moaddel, R., Stein, D., Zhang, K., Hawkins, M., Crandall, J.P., 2017. Resveratrol Improves Vascular Function and Mitochondrial Number but Not Glucose Metabolism in Older Adults. J Gerontol A Biol Sci Med Sci 72, 1703-1709.

Pordzik, J., Eyileten-Postuła, C., Jakubik, D., Czajka, P., Nowak, A., De Rosa, S., Gąsecka, A., Cieślicka-Kapłon, A., Sulikowski, P., Filipiak, K.J., Mirowska-Guzel, D., Siller-Matula, J.M., Postuła, M., 2021. MiR-126 Is an Independent Predictor of Long-Term All-Cause Mortality in Patients with Type 2 Diabetes Mellitus. J Clin Med 10.

Punthakee, Z., Goldenberg, R., Katz, P., 2018. Definition, Classification and Diagnosis of Diabetes, Prediabetes and Metabolic Syndrome. Can J Diabetes 42 Suppl 1, S10-s15.

Ramzan, F., Vickers, M.H., Mithen, R.F., 2021. Epigenetics, microRNA and Metabolic Syndrome: A Comprehensive Review. Int J Mol Sci 22.

Rodriguez-Mateos, A., Istas, G., Boschek, L., Feliciano, R.P., Mills, C.E., Boby, C., Gomez-Alonso, S., Milenkovic, D., Heiss, C., 2019. Circulating Anthocyanin Metabolites Mediate Vascular Benefits of Blueberries: Insights From Randomized Controlled Trials, Metabolomics, and Nutrigenomics. J Gerontol A Biol Sci Med Sci 74, 967-976.

Rodriguez-Mateos, A., Weber, T., Skene, S.S., Ottaviani, J.I., Crozier, A., Kelm, M., Schroeter, H., Heiss, C., 2018. Assessing the respective contributions of dietary flavanol monomers and procyanidins in mediating cardiovascular effects in humans: randomized, controlled, double-masked intervention trial. Am J Clin Nutr 108, 1229-1237.

Ruskovska, T., Budić-Leto, I., Corral-Jara, K.F., Ajdžanović, V., Arola-Arnal, A., Bravo, F.I., Deligiannidou, G.-E., Havlik, J., Janeva, M., Kistanova, E., Kontogiorgis, C., Krga, I., Massaro, M., Miler, M., Milosevic, V., Morand, C., Scoditti, E., Suárez, M., Vauzour, D., Milenkovic, D., 2021. Systematic Bioinformatic Analyses of Nutrigenomic Modifications

by Polyphenols Associated with Cardiometabolic Health in Humans—Evidence from Targeted Nutrigenomic Studies. Nutrients 13, 2326.
Ruskovska, T., Massaro, M., Carluccio, M.A., Arola-Arnal, A., Muguerza, B., Vanden Berghe, W., Declerck, K., Bravo, F.I., Calabriso, N., Combet, E., Gibney, E.R., Gomes, A., Gonthier, M.P., Kistanova, E., Krga, I., Mena, P., Morand, C., Nunes Dos Santos, C., de Pascual-Teresa, S., Rodriguez-Mateos, A., Scoditti, E., Suárez, M., Milenkovic, D., 2020. Systematic bioinformatic analysis of nutrigenomic data of flavanols in cell models of cardiometabolic disease. Food Funct 11, 5040-5064.
Sahebkar, A., Ferri, C., Giorgini, P., Bo, S., Nachtigal, P., Grassi, D., 2017. Effects of pomegranate juice on blood pressure: A systematic review and meta-analysis of randomized controlled trials. Pharmacol Res 115, 149-161.
Saklayen, M.G., 2018. The Global Epidemic of the Metabolic Syndrome. Curr Hypertens Rep 20, 12.
Salt, I.P., Nunes, J.R.C., Fullerton, M.D., 2021. Metformin again? Atheroprotection mediated by macrophage AMPK and ATF1. Cardiovasc Res 117, 1233-1234.
Satija, A., Bhupathiraju, S.N., Rimm, E.B., Spiegelman, D., Chiuve, S.E., Borgi, L., Willett, W.C., Manson, J.E., Sun, Q., Hu, F.B., 2016. Plant-Based Dietary Patterns and Incidence of Type 2 Diabetes in US Men and Women: Results from Three Prospective Cohort Studies. PLoS Med 13, e1002039.
Schön, C., Allegrini, P., Engelhart-Jentzsch, K., Riva, A., Petrangolini, G., 2021. Grape Seed Extract Positively Modulates Blood Pressure and Perceived Stress: A Randomized, Double-Blind, Placebo-Controlled Study in Healthy Volunteers. Nutrients 13.
Schroeter, H., Heiss, C., Balzer, J., Kleinbongard, P., Keen, C.L., Hollenberg, N.K., Sies, H., Kwik-Uribe, C., Schmitz, H.H., Kelm, M., 2006. (-)-Epicatechin mediates beneficial effects of flavanol-rich cocoa on vascular function in humans. Proc Natl Acad Sci U S A 103, 1024-1029.
Schwager, J., Richard, N., Widmer, F., Raederstorff, D., 2017. Resveratrol distinctively modulates the inflammatory profiles of immune and endothelial cells. BMC Complement Altern Med 17, 309.
Seneviratne, A., Cave, L., Hyde, G., Moestrup, S.K., Carling, D., Mason, J.C., Haskard, D.O., Boyle, J.J., 2021. Metformin directly suppresses atherosclerosis in normoglycaemic mice via haematopoietic adenosine monophosphate-activated protein kinase. Cardiovasc Res 117, 1295-1308.
Shannon, P., Markiel, A., Ozier, O., Baliga, N.S., Wang, J.T., Ramage, D., Amin, N., Schwikowski, B., Ideker, T., 2003. Cytoscape: a software environment for integrated models of biomolecular interaction networks. Genome Res 13, 2498-2504.
Shiojima, I., Sato, K., Izumiya, Y., Schiekofer, S., Ito, M., Liao, R., Colucci, W.S., Walsh, K., 2005. Disruption of coordinated cardiac hypertrophy and angiogenesis contributes to the transition to heart failure. J Clin Invest 115, 2108-2118.
Shiojima, I., Walsh, K., 2006. Regulation of cardiac growth and coronary angiogenesis by the Akt/PKB signaling pathway. Genes Dev 20, 3347-3365.
Shungin, D., Winkler, T.W., Croteau-Chonka, D.C., Ferreira, T., Locke, A.E., Mägi, R., Strawbridge, R.J., Pers, T.H., Fischer, K., Justice, A.E., Workalemahu, T., Wu, J.M.W., Buchkovich, M.L., Heard-Costa, N.L., Roman, T.S., Drong, A.W., Song, C., Gustafsson, S., Day, F.R., Esko, T.,

Fall, T., Kutalik, Z., Luan, J., Randall, J.C., Scherag, A., Vedantam, S., Wood, A.R., Chen, J., Fehrmann, R., Karjalainen, J., Kahali, B., Liu, C.T., Schmidt, E.M., Absher, D., Amin, N., Anderson, D., Beekman, M., Bragg-Gresham, J.L., Buyske, S., Demirkan, A., Ehret, G.B., Feitosa, M.F., Goel, A., Jackson, A.U., Johnson, T., Kleber, M.E., Kristiansson, K., Mangino, M., Leach, I.M., Medina-Gomez, C., Palmer, C.D., Pasko, D., Pechlivanis, S., Peters, M.J., Prokopenko, I., Stančáková, A., Sung, Y.J., Tanaka, T., Teumer, A., Van Vliet-Ostaptchouk, J.V., Yengo, L., Zhang, W., Albrecht, E., Ärnlöv, J., Arscott, G.M., Bandinelli, S., Barrett, A., Bellis, C., Bennett, A.J., Berne, C., Blüher, M., Böhringer, S., Bonnet, F., Böttcher, Y., Bruinenberg, M., Carba, D.B., Caspersen, I.H., Clarke, R., Daw, E.W., Deelen, J., Deelman, E., Delgado, G., Doney, A.S., Eklund, N., Erdos, M.R., Estrada, K., Eury, E., Friedrich, N., Garcia, M.E., Giedraitis, V., Gigante, B., Go, A.S., Golay, A., Grallert, H., Grammer, T.B., Gräßler, J., Grewal, J., Groves, C.J., Haller, T., Hallmans, G., Hartman, C.A., Hassinen, M., Hayward, C., Heikkilä, K., Herzig, K.H., Helmer, Q., Hillege, H.L., Holmen, O., Hunt, S.C., Isaacs, A., Ittermann, T., James, A.L., Johansson, I., Juliusdottir, T., Kalafati, I.P., Kinnunen, L., Koenig, W., Kooner, I.K., Kratzer, W., Lamina, C., Leander, K., Lee, N.R., Lichtner, P., Lind, L., Lindström, J., Lobbens, S., Lorentzon, M., Mach, F., Magnusson, P.K., Mahajan, A., McArdle, W.L., Menni, C., Merger, S., Mihailov, E., Milani, L., Mills, R., Moayyeri, A., Monda, K.L., Mooijaart, S.P., Mühleisen, T.W., Mulas, A., Müller, G., Müller-Nurasyid, M., Nagaraja, R., Nalls, M.A., Narisu, N., Glorioso, N., Nolte, I.M., Olden, M., Rayner, N.W., Renstrom, F., Ried, J.S., Robertson, N.R., Rose, L.M., Sanna, S., Scharnagl, H., Scholtens, S., Sennblad, B., Seufferlein, T., Sitlani, C.M., Smith, A.V., Stirrups, K., Stringham, H.M., Sundström, J., Swertz, M.A., Swift, A.J., Syvänen, A.C., Tayo, B.O., Thorand, B., Thorleifsson, G., Tomaschitz, A., Troffa, C., van Oort, F.V., Verweij, N., Vonk, J.M., Waite, L.L., Wennauer, R., Wilsgaard, T., Wojczynski, M.K., Wong, A., Zhang, Q., Zhao, J.H., Brennan, E.P., Choi, M., Eriksson, P., Folkersen, L., Franco-Cereceda, A., Gharavi, A.G., Hedman Å, K., Hivert, M.F., Huang, J., Kanoni, S., Karpe, F., Keildson, S., Kiryluk, K., Liang, L., Lifton, R.P., Ma, B., McKnight, A.J., McPherson, R., Metspalu, A., Min, J.L., Moffatt, M.F., Montgomery, G.W., Murabito, J.M., Nicholson, G., Nyholt, D.R., Olsson, C., Perry, J.R., Reinmaa, E., Salem, R.M., Sandholm, N., Schadt, E.E., Scott, R.A., Stolk, L., Vallejo, E.E., Westra, H.J., Zondervan, K.T., Amouyel, P., Arveiler, D., Bakker, S.J., Beilby, J., Bergman, R.N., Blangero, J., Brown, M.J., Burnier, M., Campbell, H., Chakravarti, A., Chines, P.S., Claudi-Boehm, S., Collins, F.S., Crawford, D.C., Danesh, J., de Faire, U., de Geus, E.J., Dörr, M., Erbel, R., Eriksson, J.G., Farrall, M., Ferrannini, E., Ferrières, J., Forouhi, N.G., Forrester, T., Franco, O.H., Gansevoort, R.T., Gieger, C., Gudnason, V., Haiman, C.A., Harris, T.B., Hattersley, A.T., Heliövaara, M., Hicks, A.A., Hingorani, A.D., Hoffmann, W., Hofman, A., Homuth, G., Humphries, S.E., Hyppönen, E., Illig, T., Jarvelin, M.R., Johansen, B., Jousilahti, P., Jula, A.M., Kaprio, J., Kee, F., Keinanen-Kiukaanniemi, S.M., Kooner, J.S., Kooperberg, C., Kovacs, P., Kraja, A.T., Kumari, M., Kuulasmaa, K., Kuusisto, J., Lakka, T.A., Langenberg, C., Le Marchand, L., Lehtimäki, T., Lyssenko, V., Männistö, S., Marette, A., Matise, T.C., McKenzie, C.A., McKnight, B., Musk, A.W., Möhlenkamp, S., Morris, A.D., Nelis, M., Ohlsson, C., Oldehinkel, A.J., Ong, K.K., Palmer, L.J., Penninx, B.W., Peters, A., Pramstaller, P.P., Raitakari, O.T., Rankinen, T., Rao, D.C., Rice, T.K., Ridker, P.M., Ritchie, M.D., Rudan, I., Salomaa, V., Samani, N.J., Saramies, J., Sarzynski, M.A., Schwarz, P.E.,

Shuldiner, A.R., Staessen, J.A., Steinthorsdottir, V., Stolk, R.P., Strauch, K., Tönjes, A., Tremblay, A., Tremoli, E., Vohl, M.C., Völker, U., Vollenweider, P., Wilson, J.F., Witteman, J.C., Adair, L.S., Bochud, M., Boehm, B.O., Bornstein, S.R., Bouchard, C., Cauchi, S., Caulfield, M.J., Chambers, J.C., Chasman, D.I., Cooper, R.S., Dedoussis, G., Ferrucci, L., Froguel, P., Grabe, H.J., Hamsten, A., Hui, J., Hveem, K., Jöckel, K.H., Kivimaki, M., Kuh, D., Laakso, M., Liu, Y., März, W., Munroe, P.B., Njølstad, I., Oostra, B.A., Palmer, C.N., Pedersen, N.L., Perola, M., Pérusse, L., Peters, U., Power, C., Quertermous, T., Rauramaa, R., Rivadeneira, F., Saaristo, T.E., Saleheen, D., Sinisalo, J., Slagboom, P.E., Snieder, H., Spector, T.D., Stefansson, K., Stumvoll, M., Tuomilehto, J., Uitterlinden, A.G., Uusitupa, M., van der Harst, P., Veronesi, G., Walker, M., Wareham, N.J., Watkins, H., Wichmann, H.E., Abecasis, G.R., Assimes, T.L., Berndt, S.I., Boehnke, M., Borecki, I.B., Deloukas, P., Franke, L., Frayling, T.M., Groop, L.C., Hunter, D.J., Kaplan, R.C., O'Connell, J.R., Qi, L., Schlessinger, D., Strachan, D.P., Thorsteinsdottir, U., van Duijn, C.M., Willer, C.J., Visscher, P.M., Yang, J., Hirschhorn, J.N., Zillikens, M.C., McCarthy, M.I., Speliotes, E.K., North, K.E., Fox, C.S., Barroso, I., Franks, P.W., Ingelsson, E., Heid, I.M., Loos, R.J., Cupples, L.A., Morris, A.P., Lindgren, C.M., Mohlke, K.L., 2015. New genetic loci link adipose and insulin biology to body fat distribution. Nature 518, 187-196.

Simental-Mendía, L.E., Guerrero-Romero, F., 2019. Effect of resveratrol supplementation on lipid profile in subjects with dyslipidemia: A randomized double-blind, placebo-controlled trial. Nutrition 58, 7-10.

Siragusa, M., Sessa, W.C., 2013. Telmisartan exerts pleiotropic effects in endothelial cells and promotes endothelial cell quiescence and survival. Arterioscler Thromb Vasc Biol 33, 1852-1860.

Stelzer, G., Rosen, N., Plaschkes, I., Zimmerman, S., Twik, M., Fishilevich, S., Stein, T.I., Nudel, R., Lieder, I., Mazor, Y., Kaplan, S., Dahary, D., Warshawsky, D., Guan-Golan, Y., Kohn, A., Rappaport, N., Safran, M., Lancet, D., 2016. The GeneCards Suite: From Gene Data Mining to Disease Genome Sequence Analyses. Curr Protoc Bioinformatics 54, 1.30.31-31.30.33.

Stockel, D., Kehl, T., Trampert, P., Schneider, L., Backes, C., Ludwig, N., Gerasch, A., Kaufmann, M., Gessler, M., Graf, N., Meese, E., Keller, A., Lenhof, H.P., 2016. Multi-omics enrichment analysis using the GeneTrail2 web service. Bioinformatics 32, 1502-1508.

Sun, J., Zhang, M., Chen, K., Chen, B., Zhao, Y., Gong, H., Zhao, X., Qi, R., 2018. Suppression of TLR4 activation by resveratrol is associated with STAT3 and Akt inhibition in oxidized low-density lipoprotein-activated platelets. Eur J Pharmacol 836, 1-10.

Timmers, S., Konings, E., Bilet, L., Houtkooper, R.H., van de Weijer, T., Goossens, G.H., Hoeks, J., van der Krieken, S., Ryu, D., Kersten, S., Moonen-Kornips, E., Hesselink, M.K.C., Kunz, I., Schrauwen-Hinderling, V.B., Blaak, E., Auwerx, J., Schrauwen, P., 2011. Calorie restriction-like effects of 30 days of resveratrol supplementation on energy metabolism and metabolic profile in obese humans. Cell Metab 14, 612-622.

Tomé-Carneiro, J., Gonzálvez, M., Larrosa, M., Yáñez-Gascón, M.J., García-Almagro, F.J., Ruiz-Ros, J.A., Tomás-Barberán, F.A., García-Conesa, M.T., Espín, J.C., 2013a. Grape resveratrol increases serum adiponectin and downregulates inflammatory genes in

peripheral blood mononuclear cells: a triple-blind, placebo-controlled, one-year clinical trial in patients with stable coronary artery disease. Cardiovasc Drugs Ther 27, 37-48.
Tomé-Carneiro, J., Larrosa, M., Yáñez-Gascón, M.J., Dávalos, A., Gil-Zamorano, J., Gonzálvez, M., García-Almagro, F.J., Ruiz Ros, J.A., Tomás-Barberán, F.A., Espín, J.C., García-Conesa, M.T., 2013b. One-year supplementation with a grape extract containing resveratrol modulates inflammatory-related microRNAs and cytokines expression in peripheral blood mononuclear cells of type 2 diabetes and hypertensive patients with coronary artery disease. Pharmacol Res 72, 69-82.
Trautwein, E.A., McKay, S., 2020. The Role of Specific Components of a Plant-Based Diet in Management of Dyslipidemia and the Impact on Cardiovascular Risk. Nutrients 12.
Tremblay, B.L., Guénard, F., Rudkowska, I., Lemieux, S., Couture, P., Vohl, M.C., 2017. Epigenetic changes in blood leukocytes following an omega-3 fatty acid supplementation. Clin Epigenetics 9, 43.
Valls, R.M., Pedret, A., Calderón-Pérez, L., Llauradó, E., Pla-Pagà, L., Companys, J., Moragas, A., Martín-Luján, F., Ortega, Y., Giralt, M., Romeu, M., Rubió, L., Mayneris-Perxachs, J., Canela, N., Puiggrós, F., Caimari, A., Del Bas, J.M., Arola, L., Solà, R., 2021. Effects of hesperidin in orange juice on blood and pulse pressures in mildly hypertensive individuals: a randomized controlled trial (Citrus study). Eur J Nutr 60, 1277-1288.
van Buul, J.D., Hordijk, P.L., 2004. Signaling in leukocyte transendothelial migration. Arterioscler Thromb Vasc Biol 24, 824-833.
van Steen, A.C.I., van der Meer, W.J., Hoefer, I.E., van Buul, J.D., 2020. Actin remodelling of the endothelium during transendothelial migration of leukocytes. Atherosclerosis 315, 102-110.
VanderBurgh, J.A., Hotchkiss, H., Potharazu, A., Taufalele, P.V., Reinhart-King, C.A., 2018. Substrate stiffness heterogeneities disrupt endothelial barrier integrity in a micropillar model of heterogeneous vascular stiffening. Integr Biol (Camb) 10, 734-746.
Vetrani, C., Vitale, M., Bozzetto, L., Della Pepa, G., Cocozza, S., Costabile, G., Mangione, A., Cipriano, P., Annuzzi, G., Rivellese, A.A., 2018. Association between different dietary polyphenol subclasses and the improvement in cardiometabolic risk factors: evidence from a randomized controlled clinical trial. Acta Diabetol 55, 149-153.
Ward, A.O., Angelini, G.D., Caputo, M., Evans, P.C., Johnson, J.L., Suleiman, M.S., Tulloh, R.M., George, S.J., Zakkar, M., 2020. NF-κB inhibition prevents acute shear stress-induced inflammation in the saphenous vein graft endothelium. Sci Rep 10, 15133.
Weseler, A.R., Ruijters, E.J., Drittij-Reijnders, M.J., Reesink, K.D., Haenen, G.R., Bast, A., 2011. Pleiotropic benefit of monomeric and oligomeric flavanols on vascular health--a randomized controlled clinical pilot study. PLoS One 6, e28460.
Whelton, P.K., He, J., Appel, L.J., Cutler, J.A., Havas, S., Kotchen, T.A., Roccella, E.J., Stout, R., Vallbona, C., Winston, M.C., Karimbakas, J., 2002. Primary prevention of hypertension: clinical and public health advisory from The National High Blood Pressure Education Program. Jama 288, 1882-1888.
Witkowski, M., Witkowski, M., Saffarzadeh, M., Friebel, J., Tabaraie, T., Ta Bao, L., Chakraborty, A., Dörner, A., Stratmann, B., Tschoepe, D., Winter, S.J., Krueger, A., Ruf, W., Landmesser, U., Rauch, U., 2020. Vascular miR-181b controls tissue factor-dependent thrombogenicity and inflammation in type 2 diabetes. Cardiovasc Diabetol 19, 20.

Witt, E., Hammer, E., Dörr, M., Weitmann, K., Beug, D., Lehnert, K., Nauck, M., Völker, U., Felix, S.B., Ameling, S., 2019. Correlation of gene expression and clinical parameters identifies a set of genes reflecting LV systolic dysfunction and morphological alterations. Physiol Genomics 51, 356-367.

Yao, Y., Song, T., Xiong, G., Wu, Z., Li, Q., Xia, H., Jiang, X., 2018. Combination of peripheral blood mononuclear cell miR-19b-5p, miR- 221, miR-25-5p, and hypertension correlates with an increased heart failure risk in coronary heart disease patients. Anatol J Cardiol 20, 100-109.

Yousefi, R., Parandoosh, M., Khorsandi, H., Hosseinzadeh, N., Madani Tonekaboni, M., Saidpour, A., Babaei, H., Ghorbani, A., 2021. Grape seed extract supplementation along with a restricted-calorie diet improves cardiovascular risk factors in obese or overweight adult individuals: A randomized, placebo-controlled trial. Phytother Res 35, 987-995.

Zan, L., Chen, Q., Zhang, L., Li, X., 2019. Epigallocatechin gallate (EGCG) suppresses growth and tumorigenicity in breast cancer cells by downregulation of miR-25. Bioengineered 10, 374-382.

Zhang, H., Liu, S., Li, L., Liu, S., Liu, S., Mi, J., Tian, G., 2016. The impact of grape seed extract treatment on blood pressure changes: A meta-analysis of 16 randomized controlled trials. Medicine (Baltimore) 95, e4247.

Zhao, Y., Qian, Y., Sun, Z., Shen, X., Cai, Y., Li, L., Wang, Z., 2021. Role of PI3K in the Progression and Regression of Atherosclerosis. Front Pharmacol 12, 632378.

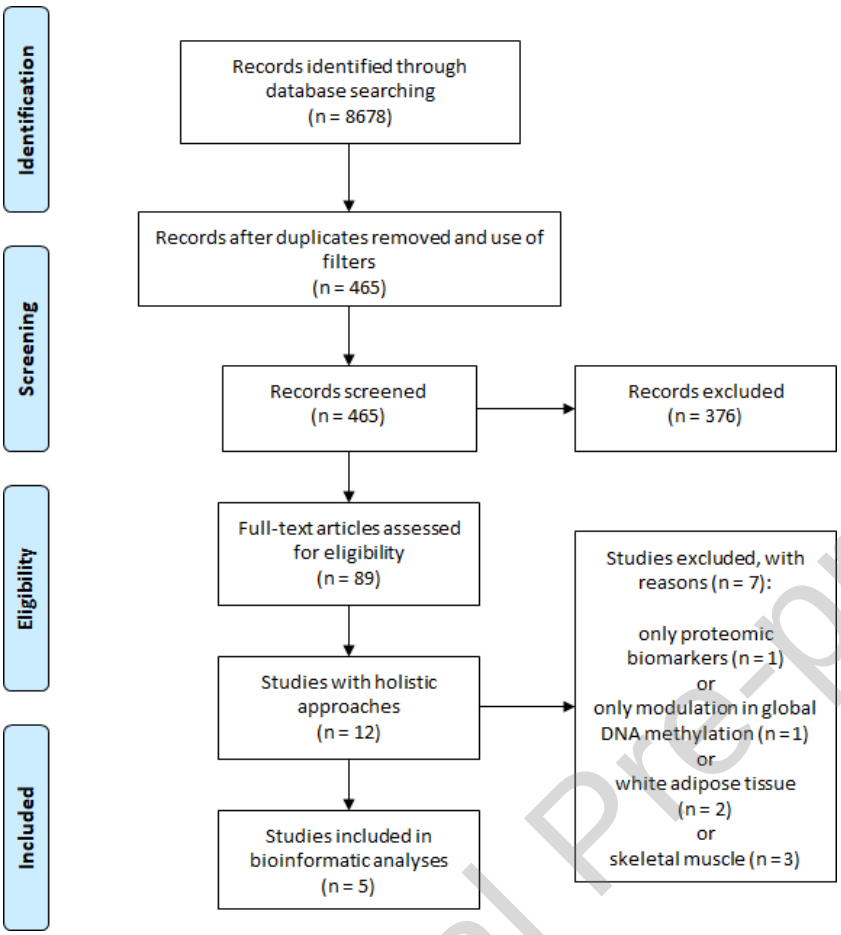
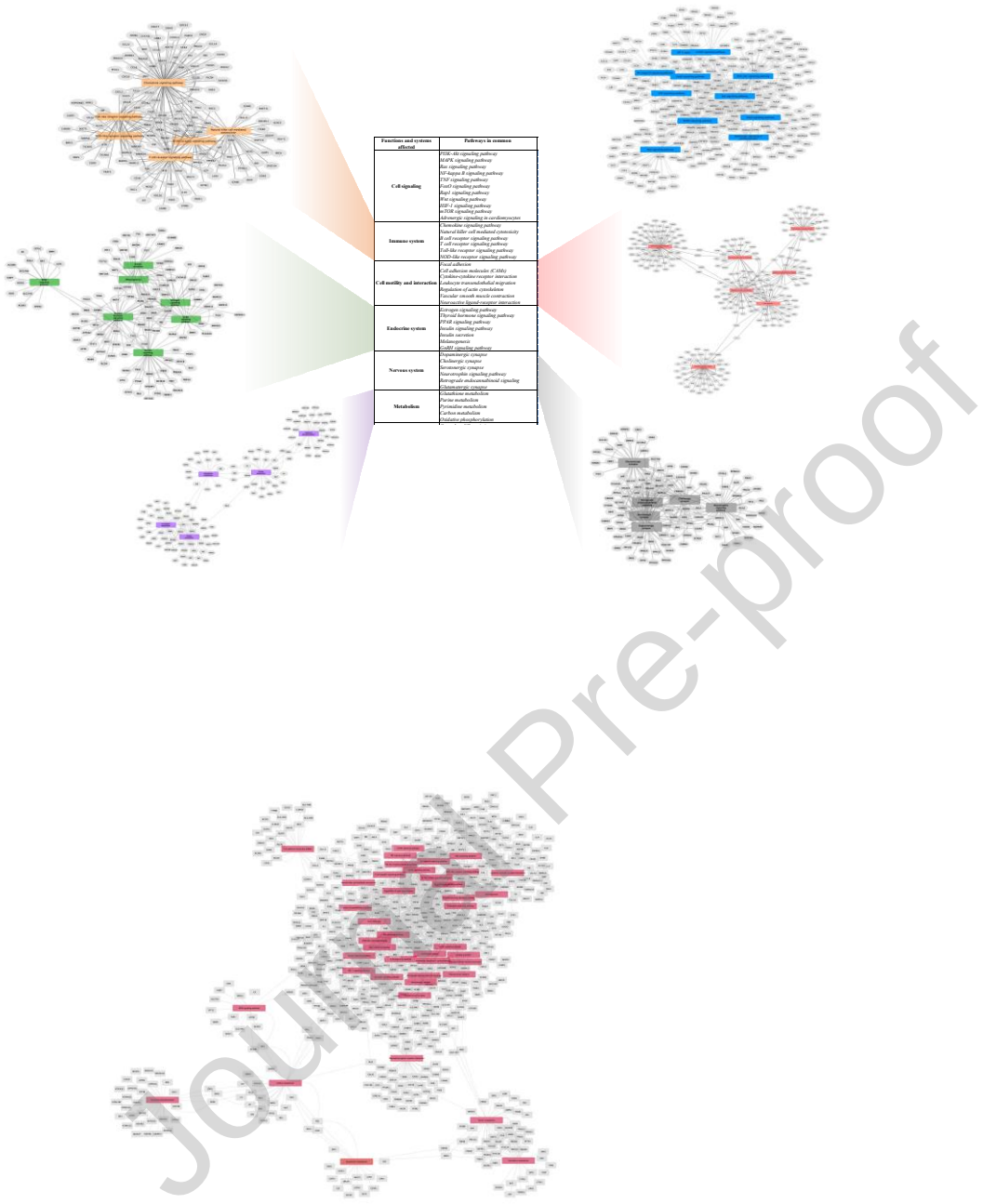


Fig 1

Functions and systems affected	Pathways in common	Number of publications	Bioactives				
			1	2	3	4	5
Cell signaling	*PI3K-Akt signaling pathway*	5	●	●	●	●	●
	MAPK signaling pathway	4	●	●	●	●	●
	Ras signaling pathway	4	●	●	●	●	●
	NF-kappa B signaling pathway	3	●	●	●	●	●
	TNF signaling pathway	3	●	●	●	●	●
	FoxO signaling pathway	3	●	●	●	●	●
	Rap1 signaling pathway	3	●	●	●	●	●
	Wnt signaling pathway	3	●	●	●	●	●
	HIF-1 signaling pathway	3	●	●	●	●	●
	mTOR signaling pathway	3	●	●	●	●	●
	Adrenergic signaling in cardiomyocytes	3	●	●	●	●	●
Immune system	*Chemokine signaling pathway*	4	●	●	●	●	●
	Natural killer cell mediated cytotoxicity	4	●	●	●	●	●
	B cell receptor signaling pathway	4	●	●	●	●	●
	T cell receptor signaling pathway	4	●	●	●	●	●
	Toll-like receptor signaling pathway	3	●	●	●	●	●
	NOD-like receptor signaling pathway	3	●	●	●	●	●
Cell motility and interaction	*Focal adhesion*	4	●	●	●	●	●
	Cell adhesion molecules (CAMs)	4	●	●	●	●	●
	Cytokine-cytokine receptor interaction	3	●	●	●	●	●
	Leukocyte transendothelial migration	3	●	●	●	●	●
	Regulation of actin cytoskeleton	3	●	●	●	●	●
	Vascular smooth muscle contraction	3	●	●	●	●	●
	Neuroactive ligand-receptor interaction	3	●	●	●	●	●
Endocrine system	*Estrogen signaling pathway*	4	●	●	●	●	●
	Thyroid hormone signaling pathway	4	●	●	●	●	●
	PPAR signaling pathway	3	●	●	●	●	●
	Insulin signaling pathway	3	●	●	●	●	●
	Insulin secretion	3	●	●	●	●	●
	Melanogenesis	3	●	●	●	●	●
	GnRH signaling pathway	3	●	●	●	●	●
Nervous system	*Dopaminergic synapse*	4	●	●	●	●	●
	Cholinergic synapse	4	●	●	●	●	●
	Serotonergic synapse	4	●	●	●	●	●
	Neurotrophin signaling pathway	4	●	●	●	●	●
	Retrograde endocannabinoid signaling	3	●	●	●	●	●
	Glutamatergic synapse	3	●	●	●	●	●
Metabolism	*Glutathione metabolism*	4	●	●	●	●	●
	Purine metabolism	3	●	●	●	●	●
	Pyrimidine metabolism	3	●	●	●	●	●
	Carbon metabolism	3	●	●	●	●	●
	Oxidative phosphorylation	3	●	●	●	●	●
Other	*Osteoclast differentiation*	4	●	●	●	●	●
	Phagosome	4	●	●	●	●	●
	Protein processing in endoplasmic reticulum	4	●	●	●	●	●
	Ribosome	4	●	●	●	●	●
	RNA transport	4	●	●	●	●	●
	Endocytosis	4	●	●	●	●	●
	mRNA surveillance pathway	3	●	●	●	●	●
	Circadian entrainment	3	●	●	●	●	●
	Apoptosis	3	●	●	●	●	●
	Oocyte meiosis	3	●	●	●	●	●
	Mineral absorption	3	●	●	●	●	●
	RNA degradation	3	●	●	●	●	●
	Proteasome	3	●	●	●	●	●
	Spliceosome	3	●	●	●	●	●
	Ribosome biogenesis in eukaryotes	3	●	●	●	●	●
	Lysosome	3	●	●	●	●	●
	Olfactory transduction	3	●	●	●	●	●

Fig 2



Functions and systems affected	Pathways in common
Cell signaling	*PI3K-Akt signaling pathway* *MAPK signaling pathway* *Ras signaling pathway* *NF-kappa B signaling pathway* *TNF signaling pathway* *FoxO signaling pathway* *Rap1 signaling pathway* *Wnt signaling pathway* *HIF-1 signaling pathway* *mTOR signaling pathway* *Adrenergic signaling in cardiomyocytes*
Immune system	*Chemokine signaling pathway* *Natural killer cell mediated cytotoxicity* *B cell receptor signaling pathway* *T cell receptor signaling pathway* *Toll-like receptor signaling pathway* *NOD-like receptor signaling pathway*
Cell motility and interaction	*Focal adhesion* *Cell adhesion molecules (CAMs)* *Cytokine-cytokine receptor interaction* *Leukocyte transendothelial migration* *Regulation of actin cytoskeleton* *Vascular smooth muscle contraction* *Neuroactive ligand-receptor interaction*
Endocrine system	*Estrogen signaling pathway* *Thyroid hormone signaling pathway* *PPAR signaling pathway* *Insulin signaling pathway* *Insulin secretion* *Melanogenesis* *GnRH signaling pathway*
Nervous system	*Dopaminergic synapse* *Cholinergic synapse* *Serotonergic synapse* *Neurotrophin signaling pathway* *Retrograde endocannabinoid signaling* *Glutamatergic synapse*
Metabolism	*Glutathione metabolism* *Purine metabolism* *Pyrimidine metabolism* *Carbon metabolism* *Oxidative phosphorylation*

Fig 3

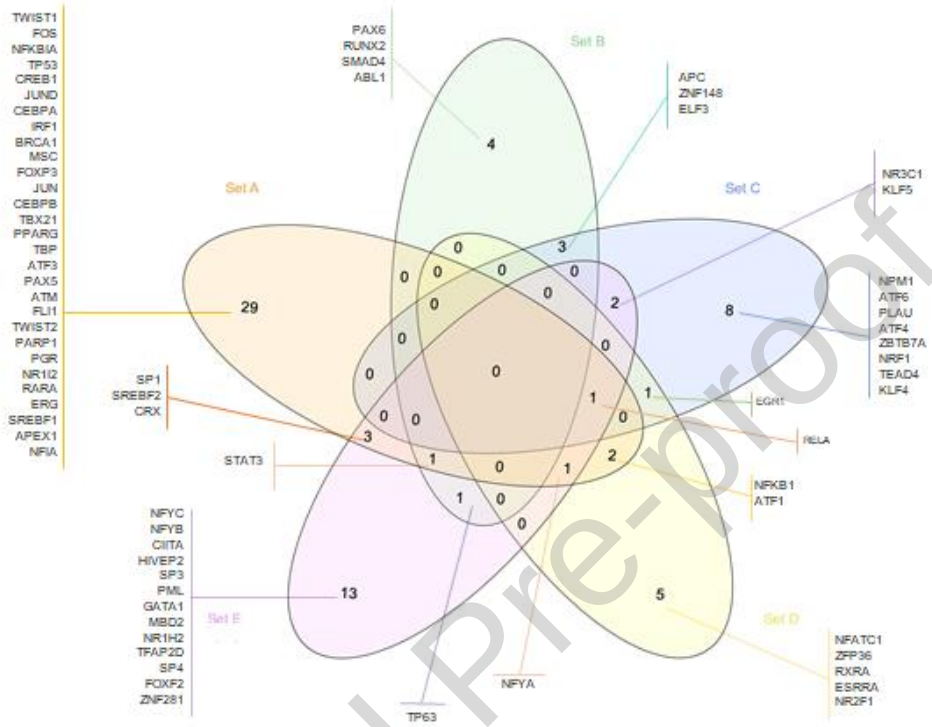


Fig 4

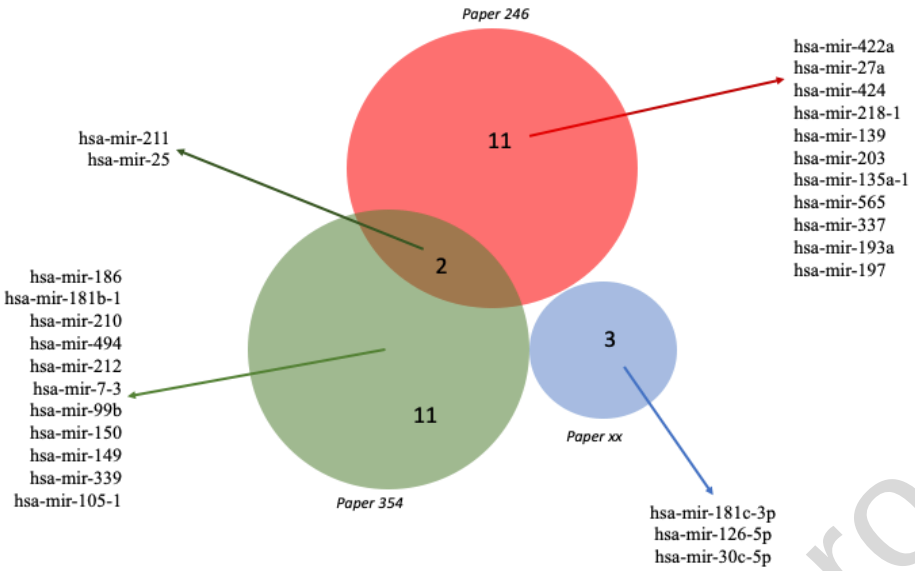


Fig 5

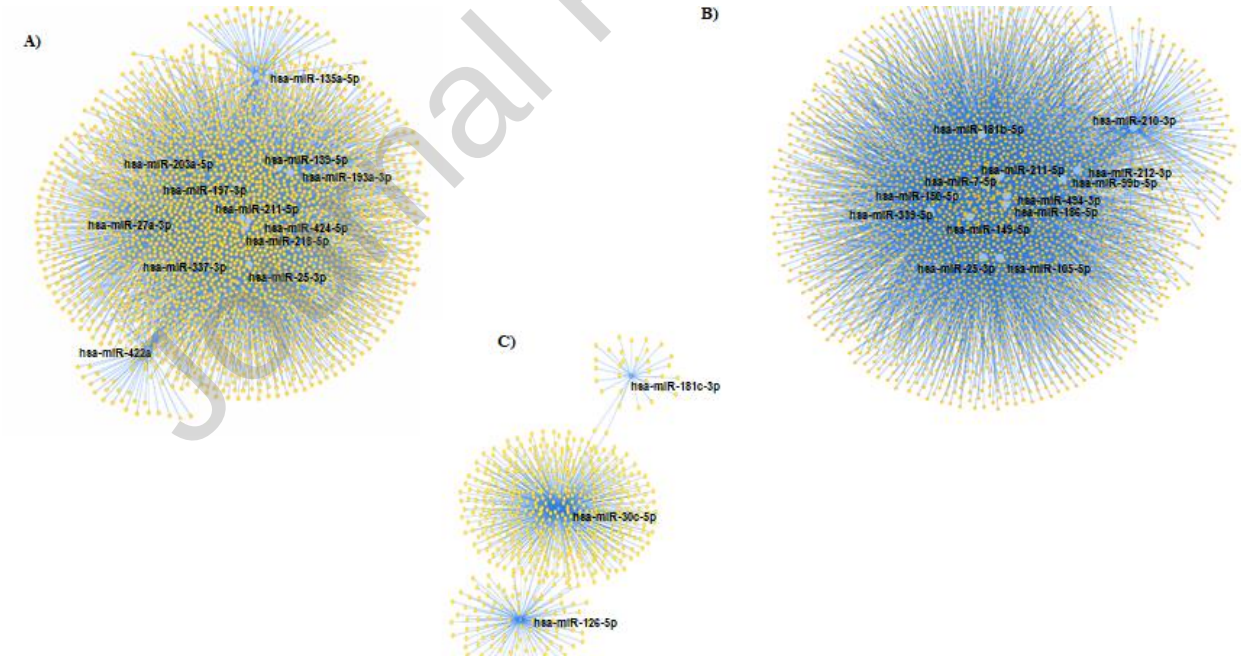
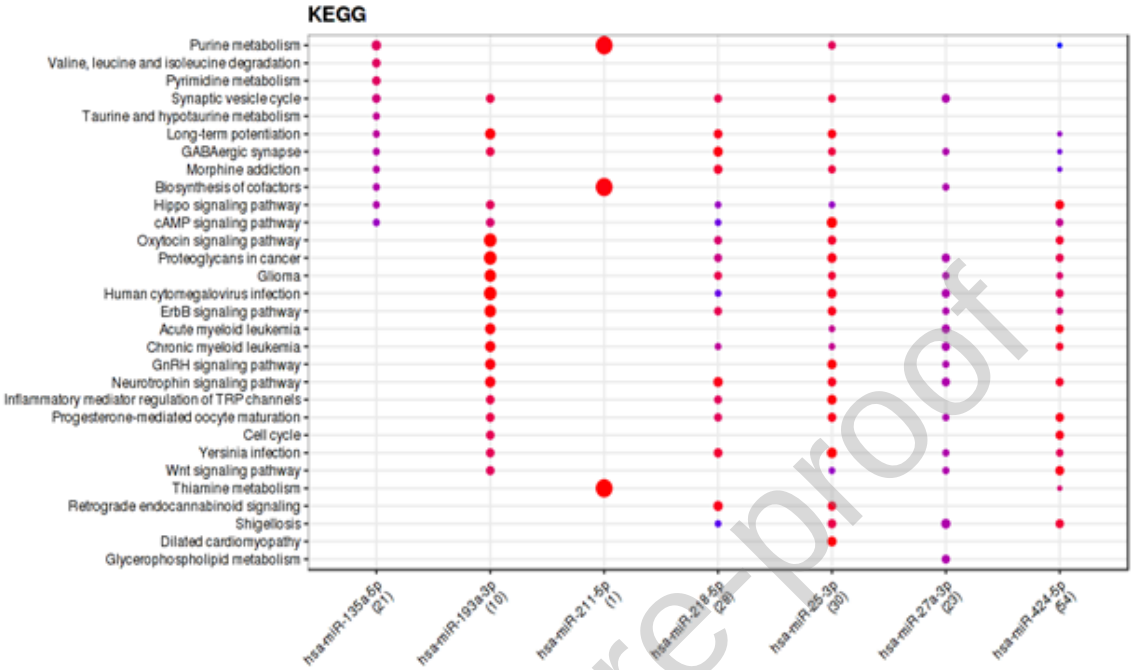
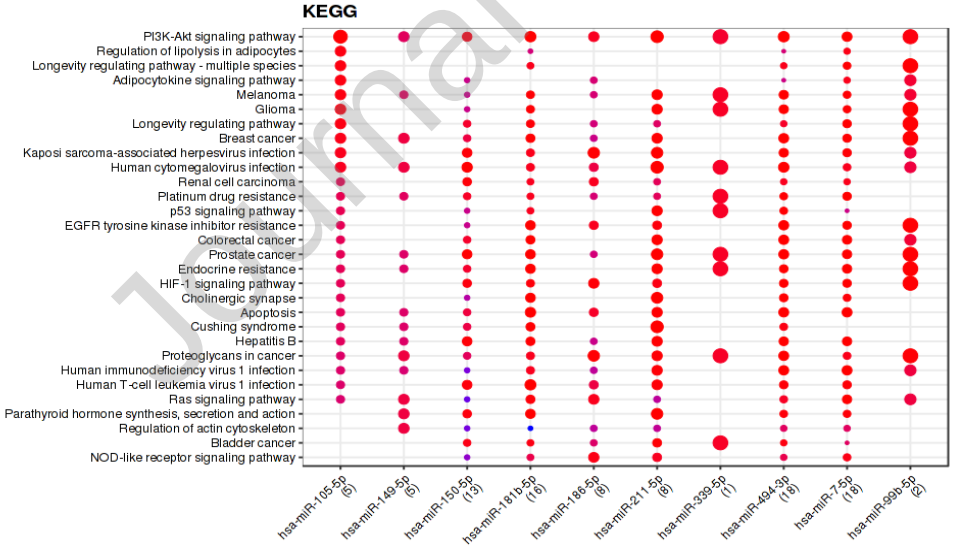


Fig 6





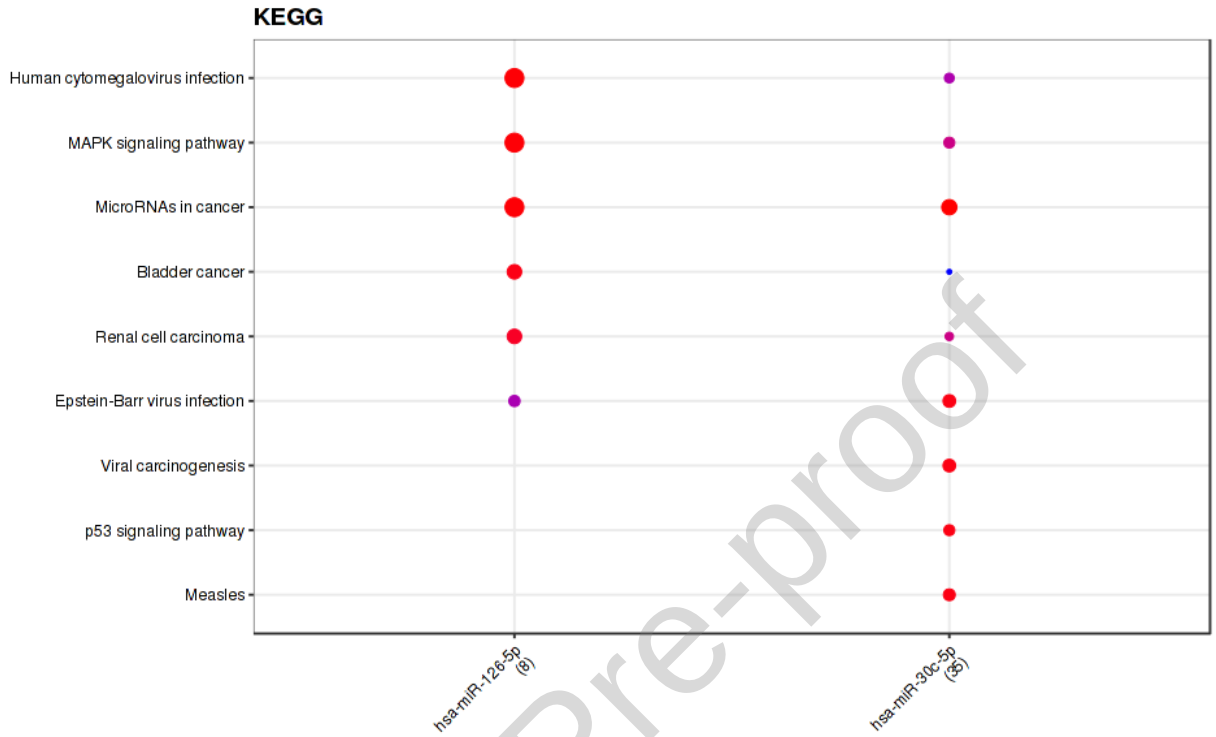


Fig 7

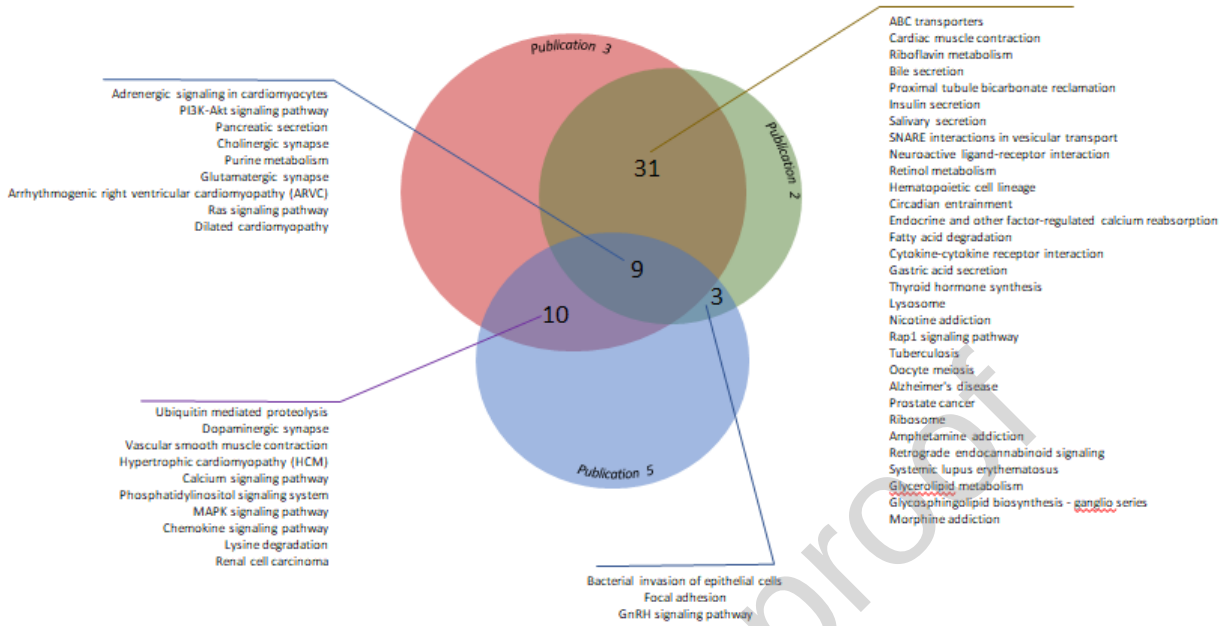
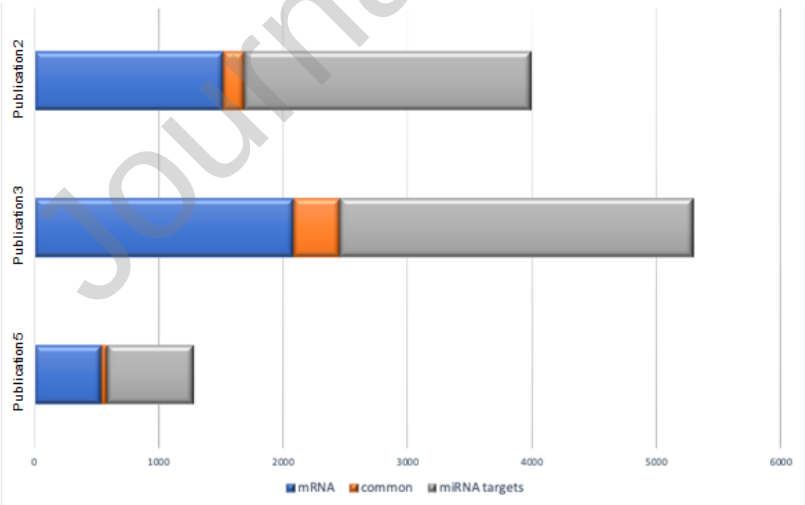


Fig 8



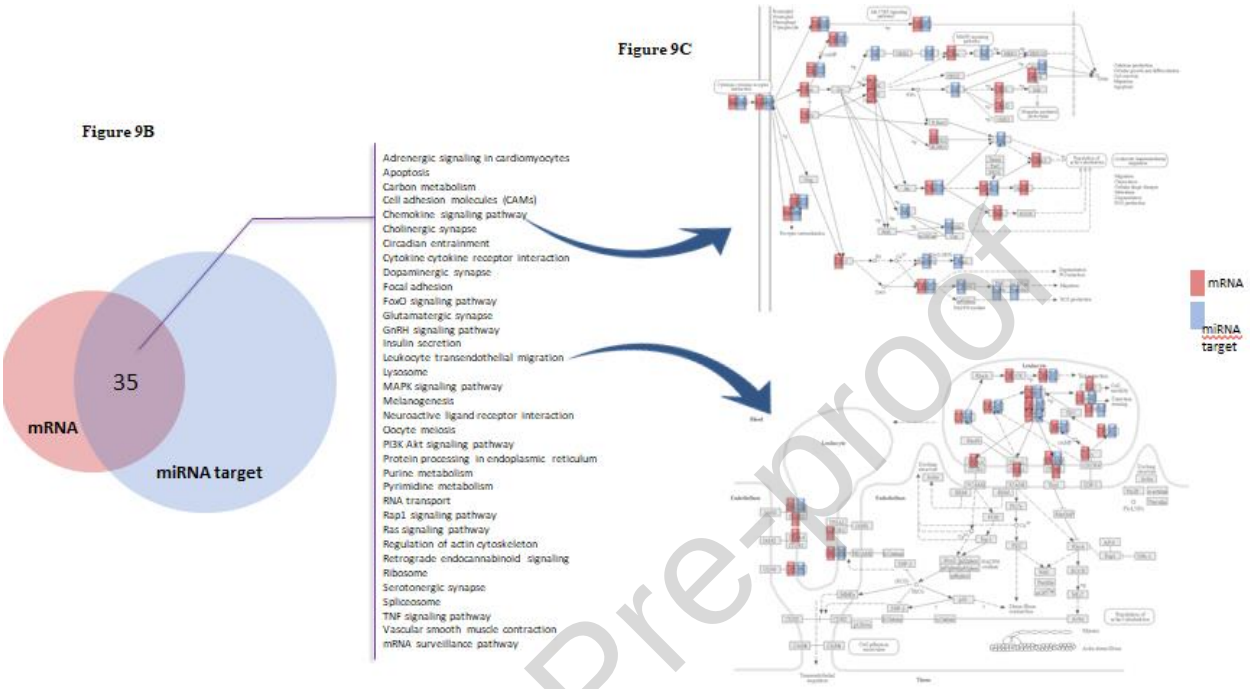


Fig 9

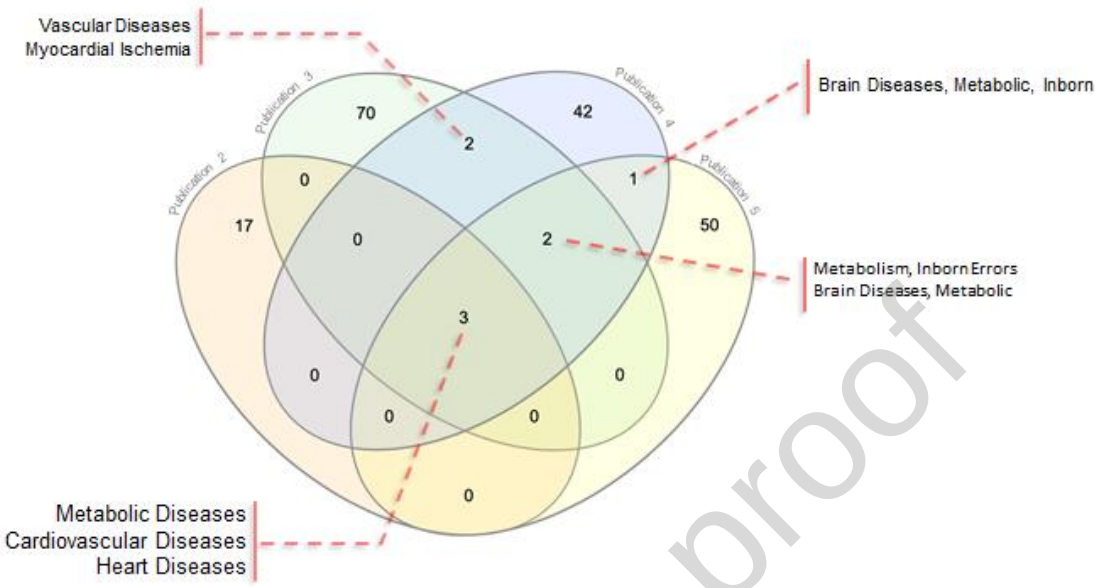


Fig 10

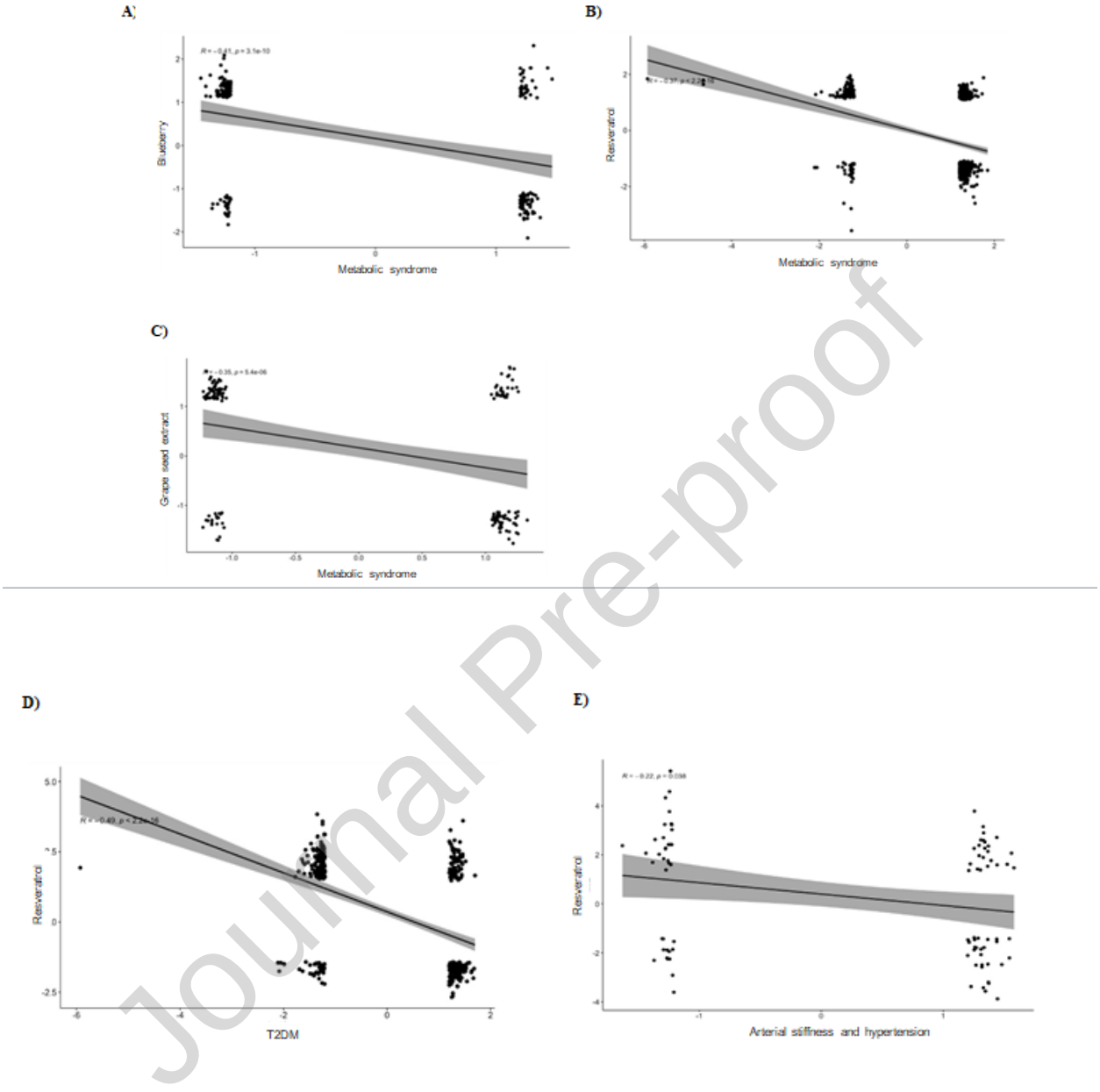
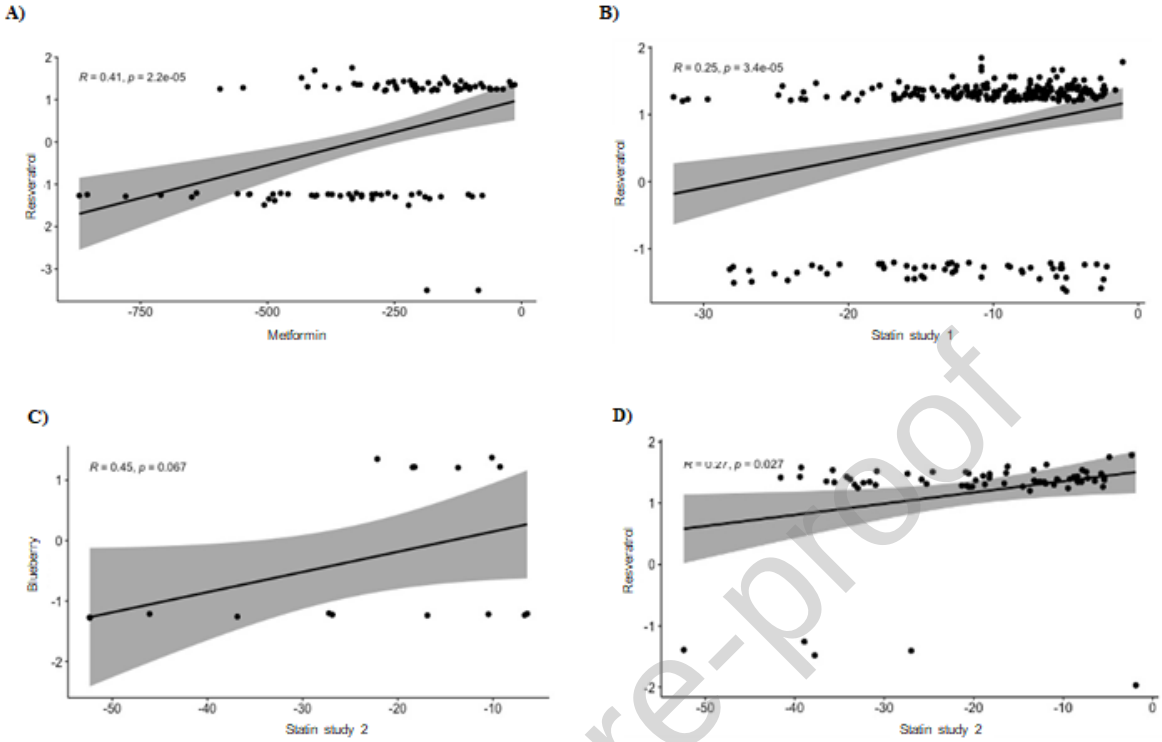


Fig 11



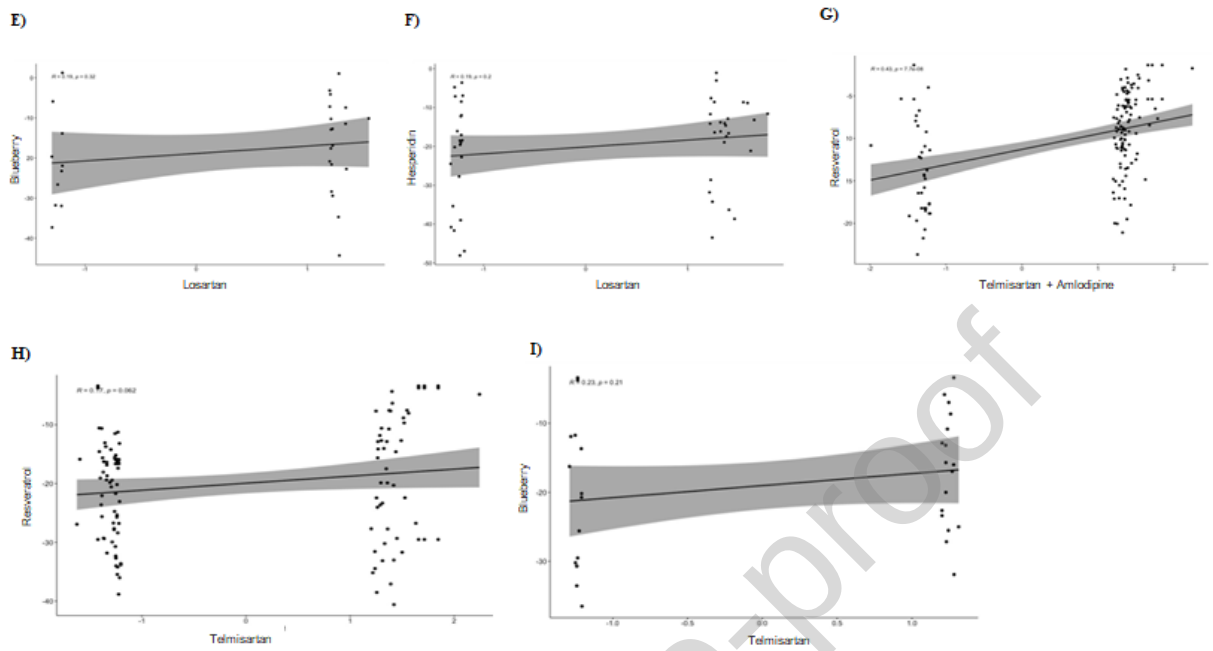
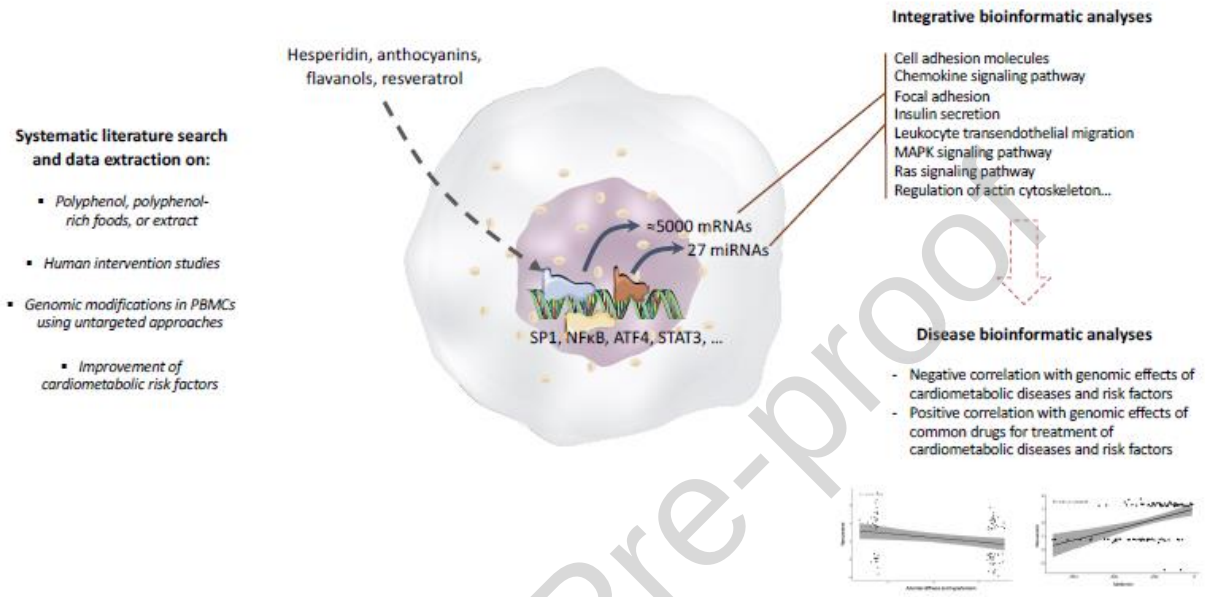


Fig 12



Graphical abstract

Highlights

- Dietary polyphenols module expression of large number of genes in humans.
- Modulated genes are involved in the regulation of cell adhesion and mobility, immune system, metabolism, or cell signaling
- Polyphenols can regulate both protein coding and protein non-coding RNAs in humans
- Gene expression profile is inversely correlated with cardiometabolic diseases and correlated with gene expression following intake of drugs against cardiometabolic disorders.