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The effect of blueberries on blood vessel function, inflammation and oxidative stress in older people

Submission date	Recruitment status	[X] Prospectively registered
30/04/2021	No longer recruiting	[X] Protocol
Registration date	**Overall study status**	[_] Statistical analysis plan
07/05/2021	Completed	[_] Results
Last Edited	**Condition category**	[_] Individual participant data
02/12/2022	Circulatory System	[_] Record updated in last year

Plain English summary of protocol

Background and study aims
Blueberries contain large amounts of polyphenols and in particular anthocyanins, which are the compounds that give to the skin of these berries their distinguishing blue coloration. An increasing number of studies support the health benefits of such compounds in the reduction of high blood pressure, platelet aggregation and blood vessel (vascular) function. Endothelial dysfunction (the loss of functionality of the internal layer of the arteries) represents a risk factor for the development of heart disease. In previous studies blueberries were found to improve vascular response, reduce blood pressure, and increase protection against oxidative stress in healthy or at-risk people (e.g. smokers or with heart disease risk factors). However, there is no evidence related to the effects of blueberry polyphenols in older people, even though they are more prone to develop heart diseases. The aim of this study is to find out whether eating blueberry products can improve vascular function, oxidative stress and inflammatory markers in older people.

Who can participate?
Adults aged 60 and over who are free from major diseases.

What does the study involve?
Participants are randomly allocated to one of two groups to consume a blueberry product or a control product after 2 days of avoiding consumption of polyphenol-rich foods, with a 2-week period (wash-out) of consuming their usual diet in between. The treatment product consists of a 250 g portion of a mousse of blueberry rich in polyphenols, while the control product consists of 250 ml of water containing the same amount and type of sugars provided by blueberry (i.e. fructose and glucose). The test will be performed two times. In the first test, blood and urine samples will be collected to evaluate the absorption of polyphenols and salicylates, and to analyse their effect on oxidative stress, vascular and inflammatory-related markers. In the second test, participants will undergo a non-invasive test to assess different markers of vascular function using a device.

What are the possible benefits and risks of participating?
Consuming polyphenol-rich products such as blueberry may help to improve vascular function and related markers. There are no notable risks involved with participating.

Where is the study run from?
University of Milan (Italy)

When is the study starting and how long is it expected to run for?
November 2020 to June 2022

Who is funding the study?
Regione Lombardia (Italy)

Who is the main contact?
Prof. Patrizia Riso
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Contact information

Type(s)
Scientific

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Additional identifiers

EudraCT/CTIS number
Nil known

IRAS number

ClinicalTrials.gov number
Nil known

Secondary identifying numbers
Nil known

Study information

Scientific Title
Evaluation of blueberry polyphenols absorption and their role in modulation of vascular function, inflammation and oxidative stress in a group of older subjects

Study objectives
The consumption of polyphenol-rich blueberries can acutely ameliorate markers of vascular function, oxidative stress and inflammatory markers, after a single ingestion, in older subjects.

Ethics approval required
Old ethics approval format

Ethics approval(s)
Approved 14/12/2020, Ethics Committee of the University of Milan (Università degli Studi di Milano, Via Festa del Perdono 7, Milan, 20122, Italy; + 39 (0)2 503 12667; comitato.etico@unimi.it), ref: 121/20_Verbale_All-11

Study design
Randomized controlled cross over study

Primary study design
Interventional

Secondary study design
Randomised cross over trial

Study setting(s)
Other

Study type(s)
Other

Participant information sheet
Not available in web format, please use the contact details to request a participant information shee

Health condition(s) or problem(s) studied
Vascular function in older people

Interventions
Based on a computer randomization plan, participants are randomized to receive the two study products in a random order. The trial is divided into two phases.

For the first phase of the trial, participants will be asked to consume a portion of blueberry products obtained from a specific cultivar of blueberry, rich in polyphenols, or a control product. Before the ingestion and after 1 h, 1,5 h, 2 h and 4 h, blood samples will be collected by all participants to evaluate the kinetics of absorption of main blueberry polyphenols, salicylates, glycaemic and insulinemic response, markers of inflammation (e.g. IL-6, IL-8, TNF-alpha), oxidative stress (e.g. cell resistance against H_2O_2-induced DNA damage) and vascular function (e.

g. nitric oxide, endotelin-1). Following a 2-week wash out period the participants will be asked to undergo the intervention again, since the trial has a cross over design.

For the second phase of the trial, participants will be asked to consume the same blueberry product or control product of the first phase. Then, they will undergo the measurement of peripheral arterial function markers (i.e. Reactive Hyperaemia Index (RHI) and Framingham RHI (FRHI)), arterial stiffness markers (dAix and dAix@75), systolic blood pressure and heart rate before the ingestion and after 2 h from the consumption of blueberry or control product, which correspond to the peak of main blueberry polyphenols absorption. Participants will also provide urine samples 24 and 48 h after the consumption of blueberry or control product.

In both phases participants will be informed about dietary habits that they should follow. They will also receive a list of food that they should avoid before and after the interventions (e.g. polyphenols rich food such as chocolate, red fruits, tea and coffee).

The polyphenol-rich blueberry product consists of a mousse obtained from about 250 g of frozen blueberry obtained from organic farming and characterized for its polyphenol content.

The control product consists of about 250 ml of water containing the same amount and type of sugars provided by the blueberry product.

Intervention Type
Other

Primary outcome measure
Phase I: polyphenols and salicylates measured through high-performance liquid chromatography (HPLC) and liquid chromatography–mass spectrometry (LC-MS) analysis before and after intake at 1 h, 1,5 h, 2 h and 4 h
Phase II: reactive hyperaemia markers (i.e. Reactive Hyperaemia Index (RHI) and Framingham RHI) and arterial stiffness markers (Augmentation Index (AI) and AI@75) measured through non-invasive EndoPAT 2000 device before and after intake at 2 h

Secondary outcome measures
1. Systolic and diastolic blood pressure measured through a sphygmomanometer following standard procedure before and after intake at 2 h
2. Plasma levels of glucose and insulin evaluated through routine laboratory clinical assessment before and after intake at 1 h, 1,5 h, 2 h and 4 h
3. DNA damage evaluated through the comet assay before and after intake at 1 h, 1,5 h, 2 h and 4 h
4. Vascular function markers (e.g. nitric oxide, endothelin-1, VEGF, VCAM-1, ICAM-1) and inflammation markers (e.g. IL-6, IL-8, TNF-alpha) evaluated using an enzyme-linked immunosorbent assay (ELISA) kit before and after intake at 1 h, 1,5 h, 2 h and 4 h

Overall study start date
03/11/2020

Completion date
30/06/2022

Eligibility

Key inclusion criteria
1. Age ≥60 years old
2. Absence of major diseases

Participant type(s)
Healthy volunteer

Age group
Senior

Sex
Both

Target number of participants
20 older subjects completing the whole protocol

Key exclusion criteria
1. Subjects with allergies or other adverse reaction to the ingestion of blueberry
2. Presence of major diseases, with pharmacological treatments

Date of first enrolment
10/05/2021

Date of final enrolment
30/04/2022

Locations

Countries of recruitment
Italy

Study participating centre
ICANS - centro internazionale per lo studio della composizione corporea
Via Sandro Botticelli, 21
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Sponsor information

Organisation
Regione Lombardia

Sponsor details

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Sponsor type
Government

Website
https://www.regione.lombardia.it/wps/portal/istituzionale/HP

Funder(s)

Funder type
Government

Funder Name
Regione Lombardia

Alternative Name(s)
Lombardy Region, Region of Lombardy

Funding Body Type
Government organisation

Funding Body Subtype
Local government

Location
Italy

Results and Publications

Publication and dissemination plan
Planned publication in a high-impact peer-reviewed journal. Additional documents (e.g. study protocol) will be available as soon as possible. The researchers are willing to publish the study protocol separately and are preparing the manuscript.

Intention to publish date
03/12/2022

Individual participant data (IPD) sharing plan

The datasets generated during and/or analysed during the current study are/will be available upon request from Prof. Patrizia Riso (patrizia.riso@unimi.it). Upon request, the corresponding author will provide access to individual de-identified participant datasets, the study protocol or informed consent form. Data may be requested from the corresponding author beginning 3 months and ending 5 years following article publication. Data are anonymous and consent from participants was obtained.

IPD sharing plan summary

Available on request

Study outputs

Output type	Details	Date created	Date added	Peer reviewed?	Patient-facing?
Protocol article		01/12/2022	02/12/2022	Yes	No