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Does a high intake of fruit in a dietary intervention study help counteract DNA hypomethylation changes brought on by cigarette smoking?

Submission date
12/12/2022

Recruitment status
No longer recruiting

Registration date
15/12/2022

Overall study status
Completed

Last Edited
14/12/2022

Condition category
Other

[X] Prospectively registered
[_] Protocol
[_] Statistical analysis plan
[_] Results
[_] Individual participant data
[_] Record updated in last year

Plain English summary of protocol

Background and study aims
Despite advances made in genome sequencing and the knowledge of genetic variants in DNA, these events alone are not enough to predict the risk of disease because of other dynamic regulatory elements, called epigenomes, that can regulate the expression of DNA sequences. Among the different epigenetic mechanisms, regulation by methylation is one of the most studied in humans. The epigenetic modification of DNA is produced by the enzymatic addition of a methyl group to carbon 5 of cytosine. Advances in omics technologies have made it possible to incorporate them into epidemiological research so that we can obtain more specific knowledge of the effects of exposure to different factors on the epigenome and provide useful knowledge for precision medicine or precision nutrition. It is known that tobacco is one of the main cardiovascular and cancer risk factors, as well as other chronic diseases. There is also consistency in the studies that have analyzed the effect of tobacco consumption on DNA methylation, which indicates that it can produce the hypomethylation of CpG sites in several genes. This hypomethylation is dynamic because, in ex-smokers, demethylated CpG sites associated with tobacco consumption have been shown to be methylated again and can reach similar methylation levels to non-smokers after decades of quitting smoking. Although multiple international studies have confirmed these results in smokers and ex-smokers, there are hardly any studies that have analyzed the influence of diet on modulating the demethylation and remethylation of said CpG sites in smokers and ex-smokers. For this reason, the objective of this study is to analyze changes in the methylation pattern with frequent consumption of fruit among the general smoking and non-smoking populations.

Who can participate?
Healthy men and women (white European) from the general Mediterranean Spanish population aged 18-75 years old

What does this study involve?
The study design will be a randomized controlled crossover clinical trial. Each person will have

the intervention with fruit (1 kg of peeled mandarins per day for a week) and the control group (no fruit will be administered). The sequence of interventions is randomized for each participant. We will start with 30 people, 15 of whom will be assigned first to the intervention group and 15 to the control group. Between the control group and the intervention group, there will be a washout period of two weeks.

What are the possible benefits and risks of participating?
There are no direct medical benefits to participating in the study. Participants will be informed that there are no benefits and risks expected.

Where is the study run from?
University of Valencia (Spain)

When is the study starting and how long is it expected to run for?
October 2022 to June 2024

Who is funding the study?
University of Valencia (Spain)

Who is the main contact?
Prof Carolina Ortega-Azorín Carolina, Ortega@uv.es (Spain)

Contact information

Type(s)
Principal Investigator

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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
Influence of frequent fruit consumption on changes in the methylation of hypomethylated CpG sites by tobacco exposure: A randomized controlled clinical trial

Acronym
TOBAM

Study objectives
The current study tests the hypothesis that frequent daily fruit eating for a week can modify the DNA methylation profile in CpG sites associated with tobacco consumption (AHRR, PRSS23, F2RL3, RARA, and GRP15 genes) in current and former smokers compared to the non-smoking population.

Ethics approval required
Old ethics approval format

Ethics approval(s)
Approved 06/12/2022, Human Research Ethics Committee University of Valencia (Avda. Blasco Ibañez, 13, 46010, Valencia, Spain; +34 (0) 963864109; vicerec.investigacio@uv.es), ref: 2448740

Study design
Interventional randomized controlled crossover trial

Primary study design
Interventional

Secondary study design
Randomised cross over trial

Study setting(s)
Community

Study type(s)
Prevention

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

Health condition(s) or problem(s) studied
General population (smokers and non-smokers)

Interventions
In this randomized and controlled cross-over trial, the same person will receive both interventions (randomly allocated using the appropriate online tool to receive fruit or the control group). The intervention with fruit will be carried out with mandarin oranges (Clemenules) that the research group will provide with the same origin and composition. A total of 1000 g of peeled oranges will be taken daily with a frequency of 4-5 times a day for a week. The rest of the diet in the intervention group will be usual. In the control group, no fruit will be provided and they will consume their usual diet (recommending fruit restriction) for a week.

Comparative methylation profiles will be measured in leukocytes (using DNA isolated from blood) to identify methylome signatures (detection of differential DNA methylation sites) of a habitual diet with frequent consumption of fruit versus habitual diet consumption after both interventions in
healthy subjects from a Mediterranean population. Differential CpG methylation sites will be statistically analyzed and the DNA methylation sites will be combined by computational analyses to build specific signatures for each intervention.

Baseline and samples from at the end of each intervention will be obtained to store them deep-frozen and with future funding to analyze the influence of the microbiome on methylation changes.

Intervention Type
Other

Primary outcome measure
Comparative methylation profiles (% of DNA-methylated cytosines) in DNA isolated from blood leukocytes measured using arrays and for selected CpG sites by Massarray, according to the standard protocols, at baseline and after one week of the corresponding intervention (frequent consumption of provided fruit or control group without the supplied fruit). DNA will be isolated by standard procedures and quality control measures will be carried out.

Secondary outcome measures
1. Plasma glucose and biochemical parameters (lipids) measured using standard procedures at baseline and after one-week interventions in both groups
2. Height, weight, waist circumference and body composition by bioimpedance measured using validated methods at baseline and after one-week interventions
3. Blood pressure and heart rate measured using standard protocols at baseline and after one-week interventions in both groups
4. Analysis of differential methylation pathways in the whole sample and by sex measured using computer tools
5. Food intake measured using food frequency questionnaires and adherence to the Mediterranean diet measured using a validated scale at baseline
6. Sleep duration, sleep quality and chronotype measured using the Horne and Östberg questionnaire at baseline
7. Additional analysis (depending on the budget) of plasma metabolites (lipids, amino acids, inflammatory markers measured using a standard high-throughput nuclear magnetic resonance metabolomics platform) at baseline and after the one week of the interventions
8. The habit of tobacco consumption will be self-reported by the participants themselves, at baseline and one week for each intervention

Overall study start date

01/10/2022

Completion date
30/06/2024

Eligibility

Key inclusion criteria
1. White European subjects (men and women) recruited from the Mediterranean population
2. Age ranging from 18 to 75 years old
3. BMI <40 Kg/m2

Participant type(s)
Healthy volunteer

Age group
Adult

Lower age limit
18 Years

Upper age limit
75 Years

Sex
Both

Target number of participants
30

Key exclusion criteria
1. Diabetic subjects
2. Other chronic diseases (cardiovascular, cancer, respiratory diseases, liver diseases, kidney diseases, etc)
3. Subjects with food allergies or food intolerances
4. Alcohol abuse or addiction
5. Immunodeficiency, HIV-positive status, COVID-19 positive status or other acute infections
6. Serious psychiatric disorders: schizophrenia, bipolar disease, eating disorders, depression, etc
7. Any severe co-morbid condition
8. History of major organ transplantation
9. Concurrent therapy with immunosuppressive drugs or cytotoxic agents
10. Current treatment with systemic corticosteroids
11. Current use of weight loss medication
12. Any other condition that may interfere with the completion of the study protocol

Date of first enrolment
21/12/2022

Date of final enrolment
21/01/2024

Locations

Countries of recruitment
Spain

Study participating centre
University of Valencia
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46010

Study participating centre
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University/education

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Funder(s)

Funder type
University/education

Funder Name
Universitat de València

Alternative Name(s)
University of Valencia, 85|86

Funding Body Type
Private sector organisation

Funding Body Subtype
Universities (academic only)

Location
Spain

Results and Publications

Publication and dissemination plan
Planned publications in an international peer-reviewed journal

Intention to publish date
01/04/2025

Individual participant data (IPD) sharing plan
The datasets generated and/or analyzed during the current study are not expected to be made available due to the limitations expressed in the informed consent

IPD sharing plan summary
Not expected to be made available