Istituti Clinici Scientifici Maugeri

IDEA Protocol N. 41379/24
Pavia, 07/26/2024

Finanziato dall'Unione europea NextGenerationEU | Ministero dell'Università e della Ricerca | Italiadomani Piano Nazionale di Ripresa e Resilienza | National Biodiversity Future Center

Project Title: **Impact of the Urban Environment on Biological Age: Effects of Air Quality and Occupational Stress on Inflammatory Cytokines and DNA Methylation Profiles in Workers in Pavia (IDEA)**

NBFC Project - CUP B83C22002930006 – Identification code CN00000033

Document date: the uploaded document was most recently updated and approved by a human subjects protection review board on 03/25/2025.

NCT number not assigned yet.

SYNOPSIS and INFORMED CONSENT FORM

Note:

The attached documents were submitted to the competent Territorial Ethics Committee and therefore were originally prepared in Italian, as required for the ethics review process. For the purpose of ClinicalTrials.gov registration, English translations of the original Italian documents are provided below. In the event of any discrepancy between the translated versions and the original documents, the Italian versions shall prevail.

IDEA Protocol N. 41379/24
Pavia, 07/26/2024

SYNOPSIS (version n. 1, 05/23/2024)

Project title	IDEA – Impact of the Urban Environment on Biological Age: effect of Air Quality and Occupational Stress on inflammatory cytokines and DNA methylation profiles in workers in Pavia.
Sponsor	ICS Maugeri
Coordinator	Prof. XXX, ICS Maugeri
Study type	Low-intervention experimental study
Number of Italian Centers involved	Single-center
Number of Participants	200 subjects, equally divided between outdoor workers exposed to traffic during their work activities and a control group carrying out work indoors in an environment without additional exposures. Subjects exposed to traffic will be recruited from the staff of the Pavia Municipal Police (approximately 60 individuals), the Pavia State Police (approximately 500 individuals), and public transport drivers (approximately 120 individuals), with an estimated participation rate of 35-40%. Preliminary contacts have already been made with the Municipal Police and the State Police, while public transport drivers in Pavia will be involved as part of the health surveillance carried out at the Occupational Medicine Unit of ICS Maugeri, Pavia Institute
Background and rationale	The University of Pavia is part of the National Biodiversity Future Centre (NBFC), funded by PNRR funds, and coordinates Spoke 6, which is aimed at studying air quality in urbanized areas and the related health impacts. Within the NBFC national center, several key activities have been identified that require external support to be selected through a call for proposals. In particular, this project was prepared in response to the call requesting 'analysis of the effect of the internal and external exposome on human health'. A reference population was therefore identified for the study, namely outdoor workers, that is, workers who operate outdoors and are exposed to occupational risks related to environmental and pollution conditions in an urban context. The urban environment is responsible for approximately 70% of air pollutant emissions which, together with other exposure factors, influence the individual's biological responses (epigenetic, oxidative, endocrinological), thereby composing the exposome, namely the set of exposures to which an individual is subjected over the course of life. Outdoor workers represent a significant target for assessing the potential health effects of air pollution, given their continuous and prolonged exposure. ICS Maugeri proposes, according to its areas of expertise, a series of activities examining this particular occupational category in order to characterize: i) external exposure, and in particular air quality (Environmental Research Centre); ii) working conditions and general health status (Occupational Medicine Unit); iii) nutritional status and lifestyle (CUO - Clinical Nutrition); iv) possible epigenetic effects,

	effects on inflammatory status, and biological age (Epigenetics Laboratory + Endocrinology Laboratory).
Study design	Low-intervention, cross-sectional experimental study. The project involves the recruitment of 200 workers, equally divided between outdoor workers and a group with negligible exposure to the urban outdoor environment. The general external exposome will be measured by carrying out a sampling campaign of personal exposure among workers who operate outdoors and among the group of workers who operate mainly indoors, with negligible direct exposure to urban air. Exposure assessment will be expressed as the mean value of personal exposure collected during the five-day monitoring campaign. The internal exposome will be determined by analyzing biological samples (blood, saliva, urine) to assess levels of inflammatory chemokines and oxidative stress markers, as well as to study epigenetic changes, such as DNA methylation.
Objectives	The IDEA study has four specific objectives: • Specific Objective 1 (SO1): To establish the concentration of the main atmospheric pollutants correlated with inflammatory processes and oxidative stress in the air breathed by the subjects involved in the project, through direct measurements of inhalation exposure in situations representative of the external exposome specific to the urban environment of the city of Pavia. The data collected (as mean values of exposure measurements over five working days) will be integrated with data from urban air-quality monitoring stations in the city of Pavia. • Specific Objective 2 (SO2): To assess the specific external exposome, both in terms of lifestyle and in terms of reconstruction of occupational exposures. Both these factors influence the characteristics of the internal exposome, contributing to or counteracting potential developments toward clinically evident and diagnosable diseases. • Specific Objective 3 (SO3): To measure the levels of key inflammatory cytokines (IL-1, IL-6, IL-8) and 8-oxoguanosine (a marker of oxidative stress) in individuals exposed to urban environmental factors. • Specific Objective 4 (SO4): To analyze DNA methylation in individuals exposed to urban environmental factors, in order to assess the impact of exposure on this epigenetic process.
Endpoints	The IDEA study investigates the impact of the urban exposome on health by assessing biological effects in a population of outdoor workers and comparing them with a group of workers minimally exposed to the urban environment of the city of Pavia, while also collecting information on participants' nutrition and lifestyle. In particular, the identified endpoints relate to five different categories of indicators: • DNA methylation

	• Assessment of the biological clock • Levels of inflammatory chemokines (CCL2, CXCL8, CXCL10) • Oxidative stress markers (hydrogen peroxide H2O2, hydroxyl radicals OH-, peroxyl radicals ROO-) • Thyroid function parameters and anti-thyroid antibodies. These endpoints will be related to levels of exposure to the different atmospheric pollutants in order to evaluate possible correlation profiles in exposed subjects.
Population	Inclusion criteria: the exposed group includes adult subjects of both sexes who carry out their activity predominantly outdoors and in an urban environment, presenting exposure to air pollution. In particular, preliminary contacts have been made with the Municipal Police and State Police forces of the city of Pavia, who have expressed their interest in participating in the study. The non-exposed group includes adult subjects of both sexes recruited from the Occupational Medicine Service, ICS Maugeri of Pavia, and will be identified among workers performing office duties and without particular exposures (e.g., workers of the University of Pavia). Exclusion criteria: the detailed definition of the exclusion criteria is the subject of an activity included in the project itself. As a preliminary approach, subjects with active oncological disease and those currently receiving immunosuppressive therapy are expected to be excluded.
Sample size	Number of subjects in the study: 200. Given the exploratory nature of the study, which is configured as a pilot investigation, no formal sample-size calculations were performed. The number of subjects to be included was defined on the basis of scientific rationale and the resources made available by the project call. Furthermore, given the time constraints imposed by the funding provided through the PNRR, the number of subjects was defined in such a way as to ensure the sustainability of recruitment.
Study duration	15 months
Study phases	The project activities are organized into five work packages, the first of which is managerial and the other four of which describe the operational phases of the study: 1. Overall project organization (M1-M15): aimed at coordinating activities in scientific and economic-financial management terms, as well as communication and scientific dissemination actions. 2. Recruitment of the population and assessment of occupational history (M1-M15): in this phase, operating protocols will be defined and authorization will be requested from the ethics committee. In addition, the study population will be involved; their occupational history will be reconstructed and biological samples will be collected for subsequent analyses. 3. Measurement of exposure to urban atmospheric pollutants (M4-M14), which involves personal characterization of the level of pollution to which

	exposed and non-exposed subjects are subjected (assessment of the general external exposome) and the creation of an exposure database for the city. 4. Assessment of nutritional status and lifestyle (M3-M14), which deals with the measurement of variables related to the 'specific' external exposome in terms of eating habits and lifestyle through the administration of questionnaires and interviews concerning smoking habits, stress level, eating habits (Food Frequencies Questionnaire and MEDI-LITe), level of physical activity, and sleep quality. 5. Assessment of the DNA methylation profile, determination of biological age, levels of inflammatory cytokines, and oxidative stress (M3-M14). This is aimed at determining the identified endpoints as a function of the monitored exposure levels. The results of the four operational phases will be integrated for an overall assessment of the health and exposure conditions of the subjects involved in the study, in order to identify their impact on the selected endpoints in relation to work activity.
Statistical methods	For the description of the main variables under study, standard significance tests will be used, taking into account the normal distribution (Student's t-test, chi-square test) or non-normal distribution of the variables themselves. With regard to inferential statistical analyses, linear or logistic regression models will be used, respectively, for continuous quantitative variables and for discrete or qualitative variables. These analyses will be conducted both for the group of non-exposed subjects (reference category) and for exposed subjects, in order to compare the biological indicators under study in relation to exposure. In addition, the models will be appropriately adjusted to correct for the main confounding variables.

Istituti Clinici Scientifici Maugeri

IDEA Protocol N. 41379/24
Pavia, 07/26/2024

INFORMATION SHEET AND INFORMED CONSENT FORM

for adult patients capable of personally providing consent

PRINCIPAL INVESTIGATOR: Prof. XXX
Occupational Medicine Unit
Telephone: +39 0382 592740

INFORMATION SHEET

Dear Sir/Madam,

The ICS Maugeri IRCCS Scientific Clinical Institutes – Pavia Institute have promoted an experimental study entitled: “Impact of the Urban Environment on Biological Age: Effect of Air Quality and Occupational Stress in Pavia on Inflammatory Cytokines and DNA Methylation Profile.”

This is a national, single-center research study.
To carry out this research, we require the collaboration and availability of individuals who, like you, meet the scientific eligibility criteria for the assessments to be performed.

Based on the predefined inclusion and exclusion criteria, you appear to be a suitable candidate for this study.

Before deciding whether or not to provide your consent to participate, please read these pages carefully, taking all the time you need, and ask for clarification if anything is unclear or if you require further information. If you wish, before making your decision you may also seek advice from your family members or your trusted physician. Please note that signing the consent form is voluntary, but it is required in order to participate in the study.

PURPOSE OF THE STUDY

IDEA is a clinical study that observes and analyzes how the urban environment affects human health. It focuses on workers who perform outdoor activities in urban settings and compares them with a group of individuals whose exposure to the urban environment is limited to daily non-work-related activities, for a total of 200 participants equally distributed between the two groups.

The study uses a multidisciplinary approach and includes a comprehensive assessment of exposure to environmental factors such as pollution and lifestyle, as well as an evaluation of the body's biological responses, including exposure biomarkers and biological age. This is a low-intervention experimental study involving the collection of biological material (blood sampling).

WHAT PARTICIPATION IN THE STUDY INVOLVES

If you agree to participate in this study, you will undergo an assessment visit to verify that your condition meets the required eligibility criteria. This visit will take place at the Hospital Occupational Medicine Unit of ICS Maugeri, Pavia Institute. During this visit, the following activities will be carried out:

- Reconstruction of current and past occupational exposure history.
- Provision of a personal passive sampler (Radiello) and instructions for its use during the sampling period (5 days, from the Monday following the visit until Friday).
- Blood sampling for the evaluation of biochemical and exposure-related parameters.

In addition, a second visit is planned, during which nutritional status will be assessed, including body composition analysis (bioelectrical impedance analysis) and assessment of liver fibrosis/steatosis status.

The data collected during the first and subsequent visits will be used for the purposes of this study.

The study will last 15 months and a total of 200 participants are expected to be enrolled.

STUDY RESULTS AND CONFIDENTIALITY OF COLLECTED INFORMATION

All your data will be coded and recorded electronically. You will be assigned a personal numerical or alphanumeric code that will not allow your identification outside the research center (pseudonymization[1]). For information regarding the processing of personal data, please refer to the separate privacy notice that will be provided to you.

RISKS ASSOCIATED WITH PARTICIPATION IN THE STUDY

As this is a low-intervention experimental study involving the collection of biological material (blood sampling), participation does not require examinations or treatments that differ from those performed in routine clinical practice. Therefore, participation in the study does not involve risks beyond those associated with normal clinical practice, and no specific insurance coverage for the study is provided.

[1] Pseudonymization means the processing of personal data in such a manner that the data can no longer be attributed to a specific individual without the use of additional information, provided that such information is kept separately and protected through appropriate technical and organizational measures, in accordance with Article 4(5) of the GDPR.

POTENTIAL BENEFITS OF PARTICIPATION

By participating in this study, you will receive the results of your blood tests (including thyroid function assessment) and a report from the nutritional counseling session. We believe these results may help you better understand your current health status and support any preventive or health-improvement measures you may wish to undertake.

WITHDRAWAL FROM THE STUDY

Your participation in this study is entirely voluntary, and you may withdraw at any time without providing any explanation.

By signing this consent form, you explicitly agree that if you decide to withdraw from the study, the medical data collected before your withdrawal may still be processed, in pseudonymized form, together with the other data collected as part of the study.

The Sponsor may terminate the study prematurely at any time, provided that the reasons are communicated to the Ethics Committee.

PRIVACY NOTICE
Pursuant to Article 13 of Italian Legislative Decree 196/03
(Personal Data Protection Code)

Data controllers and purposes of processing:

ICS Maugeri IRCCS Scientific Clinical Institutes – Pavia Institute (headquartered on Via Maugeri) and the National Biodiversity Future Center (NBFC) are the Data Controllers of your personal data and, in accordance with Good Clinical Practice requirements (Legislative Decree 211/2003), will process your personal data—particularly health-related data and only to the extent necessary for achieving the objectives of the Study—exclusively for the conduct of the Study.

Prof. XXX has been appointed Data Processing Manager on behalf of ICS Maugeri.

The processing of your personal data is essential for participation in the Study. Refusal to provide such data will prevent your participation.

Nature of the data:

The responsible physician will collect your data together with the code assigned to you as a participant so that only this code will appear on the data collection form, rather than your surname, first name, or initials. Furthermore, only the month and year of your birth will be recorded on the form (and the day only if indispensable for proper study conduct). The physician will maintain the correspondence file linking personal identification data and participant codes

on a password-protected computer located in a restricted-access area separate from the location where other study documents are stored. Only the physician and authorized personnel will be able to link the code to your identity.

Methods of processing and disclosure to third parties

The above-mentioned data will be collected, managed, and stored, both in paper and electronic form, by ICS Maugeri IRCCS Scientific Clinical Institutes – Pavia Institute.
Your data will not be disclosed except in aggregated form during scientific conferences or through scientific or statistical publications.
The data will not be transferred outside the territory of the European Union.

Exercise of your rights

Pursuant to Article 7 of Legislative Decree 196/03, by contacting ICS Maugeri IRCCS directly, you have the right to obtain:

a) confirmation as to whether personal data concerning you exist and communication of such data in an intelligible form;
b) updating, rectification, or integration of the data where appropriate;
c) deletion, anonymization, or blocking of data processed unlawfully, including data whose retention is no longer necessary;
d) certification that the operations referred to in points b) and c) have been notified to those to whom the data were communicated, except where this proves impossible or requires disproportionate effort.

You have the right to object, in whole or in part, on legitimate grounds, to the processing of your personal data.
You may discontinue your participation in the Study at any time and without providing any justification. In that case, no further data concerning you will be collected, while data already collected may continue to be used to preserve the scientific validity of the study results.

For further information during the study, you may contact: project.idea@icsmaugeri.it

ADDITIONAL INFORMATION

You may freely access the documentation relating to the study (clinical-scientific and therapeutic documentation) and the opinion expressed by the Ethics Committee by requesting them directly from the Ethics Committee. The study protocol has been prepared in accordance with the European Union Good Clinical Practice Guidelines and the current revision of the Declaration of Helsinki and has been approved by the Ethics Committee of this institution. You may report any matter you consider relevant concerning the research to: Territorial Ethics Committee Lombardia 6 c/o Fondazione I.R.C.C.S. Policlinico San Matteo, Viale Golgi 19, 27100 Pavia, Italy.

Istituti Clinici Scientifici Maugeri

IDEA Protocol N. 41379/24
Pavia, 07/26/2024

WHO ORGANIZES AND FUNDS THIS STUDY?

The study is promoted and funded by the National Biodiversity Future Center (NBFC – PNRR) and the ICS Maugeri Scientific Clinical Institutes, Pavia Institute.

INFORMED CONSENT DECLARATION

This declaration must be personally signed and dated by both the patient and the physician who conducted the informed consent discussion ONLY IF THE PATIENT HAS DECIDED TO PARTICIPATE IN THE STUDY.

I, the undersigned _____

declare that I have received comprehensive explanations from Dr. _____ regarding the request to participate in the above-mentioned study, according to the information sheet attached hereto, a copy of which was provided to me sufficiently in advance.

I further declare that I have had the opportunity to discuss the information provided, to ask all questions I considered necessary, and to receive satisfactory answers. I have also had the opportunity to consult a person of my choice regarding the details of the study.

I therefore freely agree to participate in the study, having understood the purpose of the request and the risks and benefits involved. I understand my right to discontinue participation at any time.

I have also been informed of my right to access the documentation relating to the study and the opinion expressed by the Ethics Committee.

I am furthermore aware that, during the study, a representative of the Sponsor or its delegate, the Ethics Committee, or national or international Regulatory Authorities may review the conduct of the study and verify the accuracy of the data reported in the medical records and case report forms.

Date _____ Patient Signature _____

Date _____ Signature of Physician Providing Information _____

(If the patient is unable to read or sign, an independent witness who is not affiliated with the Investigator or the Sponsor must be present throughout the informed consent discussion. The witness must personally sign and date the informed consent form after the form and all written information have been read and explained to the participant and after the participant has verbally consented to participate in the study.)

IDEA Protocol N. 41379/24
Pavia, 07/26/2024

In this case: I, the undersigned _____, hereby attest that Dr. _____ fully explained to Mr./Ms. _____ the characteristics of the above-mentioned study according to the attached information sheet and that the participant, after having had the opportunity to ask all necessary questions, freely agreed to participate in the study.

Date _____ Signature of Independent Witness _____

Date _____ Signature of Physician Providing Information _____