FRIGID ZONE **MEDICINE**
Original Article • DOI: 10.1515/fzm-2025-0006

Cold environments and health: proteomic analysis of health impacts

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Abstract

Background: Cold temperatures cause blood vessels to constrict, shallow breathing, and slight thickening of the blood. Working in extremely cold environments can have negative effects on health, yet there are currently no effective biomarkers to monitor these health conditions. Proteins are important intermediate phenotypes that can provide a theoretical basis for understanding disease pathophysiology. Proteins in the circulatory system reflect the physiological status of individuals, and plasma proteins have significant potential as biomarkers for various health conditions. **Methods:** In this study, we employed the Mendelian randomization (MR) method to analyze the effects of freezing temperatures on over 2900 plasma proteins. Subsequently, the selected plasma proteins were subjected to causal analysis in relation to 55 diseases, including respiratory disorders, cardiovascular diseases, various cancers, and oral diseases. The aim was to identify proteins that could serve as biomarkers for health status. **Results:** Our results indicate that cold environments may affect the concentrations of 78 plasma proteins. Further MR analysis revealed that nine of these plasma proteins are associated with the risk of respiratory disorders, cardiovascular diseases, various cancers, and oral diseases. **Conclusion:** These proteins show promise as biomarkers for monitoring the hazards and risks faced by individuals working in cold environments. These findings provide valuable insights into the biological mechanisms underlying occupational hazards.

Keywords

plasma protein; biomarker; cold environment; Mendelian randomization

Received 17 December 2023 , accepted 11 November 2024

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1 Introduction

Over the past century, scientists have observed substantial increases in greenhouse gas concentrations, and numerous indicators confirm that our climate is changing worldwide[1]. These changes in the climatic system have resulted in more extreme weather events. A major challenge facing human society is the threat of climate change, which is closely linked to food security, the protection of water resources, and public health[2]. Climate change is highly likely to have both direct and indirect effects on human health. These effects range from deaths due to extremely low temperatures to shifts in disease patterns. As a result, public health systems may experience greater strain[3].

Climate change, by causing abnormal ambient temperatures, is increasing the risk of a wide range of diseases, according to epidemiological evidence. In addition to cardiovascular and respiratory diseases, cancer is among the most common diseases. Numerous studies have demonstrated that climatic conditions affect the clinical manifestations of cardiopulmonary disorders. For example, acute coronary syndromes, myocardial infarctions, and their morbidity and mortality rates vary seasonally, with acute events peaking in winter and declining in summer[4-5]. Despite these findings, no biomarkers have yet been identified that can accurately assess human health before the development of diseases with obvious symptoms.

In understanding disease pathophysiology, proteins provide both theoretical foundations and potential solutions. The proteins in the circulatory system can offer insight into an individual's physiology. Protein levels in plasma reflect genetic differences, and inflammatory status, and metabolic conditions. Dysregulation of plasma proteins has been frequently observed in a variety of diseases, suggesting that these dysregulated proteins play a role in disease pathogenesis. The U.S. Food and Drug Administration has cleared

or approved hundreds of plasma proteins as biomarkers in clinical trials, with many more currently being developed[6-8]. Rapid developments in proteomic analyses of human blood have made it possible to identify plasma biomarkers that can monitor a person's health in extreme environments.

Recently, Mendelian Randomization (MR) has emerged as a new epidemiological tool. The primary objective of MR is to estimate the causal effect of an exposure on an outcome using Single Nucleotide Polymorphisms (SNPs) from Genome-Wide Association Studies (GWAS). Because genetic variants are randomly allocated during conception, they are largely independent of confounders, thus reducing the risk of confounders[9]. Two-sample MR was used in the present study to screen and evaluate the relationship between working in extremely cold environments and plasma concentrations of 2992 proteins in European populations[10]. A GWAS dataset of 55 disease-related plasma proteins was analyzed as part of this study to determine if workplace environments affect plasma proteins associated with disease risk. It is necessary to estimate whether variations in these plasma protein levels affect disease risks.

2 Methods

An analysis of genetic associations with workplace environments was conducted by the UK Biobank (UKB) cohort, which collected genetic and health information on over half a million individual participants. We retrieved GWAS data from subjects working in extremely cold environments (self-reported: often, N = 7 881; $N_{control}$ = 82,307) from the second round of GWAS results released by the UK Biobank in August 2018 (http://www.nealelab.is/uk-biobank). MRBASE (http://app.mrbase.org/) was used to summarize the data for this study[11-12]. Detailed GWAS IDs and descriptions of these data are available in Supplementary Data 1.

2.1 Genetic associations with plasma protein concentrations: data sources

The INTERVAL study, which included 3301 Europeans, provided genetic instruments to assess plasma protein levels[10]. National Research Ethics Service approval was granted for the INTERVAL study, which required participants to complete trial consent. As a result, this study used the GWAS summaries of 2992 non-redundant plasma proteins collected by MRBASE (Supplementary Data 1).

2.2 Data sources of genetic associations with diseases

Numerous studies have recognized the impact of climatic conditions on the clinical presentations of certain diseases. Acute events consistently peak in winter and experience troughs in summer[4-5]. These studies have indicated seasonal variation in the incidence of cardiovascular diseases, respiratory disorders, oral diseases, and cancers. The disease summary GWAS data in this study were derived from the EU Open GWAS and the FinnGen consortium to examine the role of plasma proteins identified in the preliminary analysis. A comprehensive description of these GWAS datasets is provided in Supplementary Data 1.

2.3 Selection of genetic instruments and statistical analysis

MR uses genetic variations as Instrumental Variables (IVs) to determine whether a particular exposure causally affects a particular outcome. A valid IV must meet three core criteria: First, it should be strongly correlated with the exposure; second, the SNP must not be associated with any traits that could confound the relationship between the exposure and the outcome; lastly, the variant must not be linked to the outcome through alternative pathways other than the exposure. A SNP is considered to exhibit horizontal pleiotropy when it violates the last two assumptions[13-14]. The procedural flow of this analysis is depicted in Fig. 1.

With default parameters, aside from an alternative significance threshold, two-sample multiple regression was used to identify SNPs that strongly and independently predicted genome-wide significant exposures ($P < 5 \times 10^{-6}$). The inverse variance weighted (IVW) approach was employed to estimate the preliminary relationships between exposures and outcomes. MR-Egger and weighted median methods were used for sensitivity analyses of significant IVW results. For the second round of MR analysis, we applied the IVW method to evaluate the relationships between plasma protein concentrations and diseases related to the working environment, with plasma protein concentration as the exposure and disease as the outcome[15]. Fig. 2 shows the workflow of this analysis.

The TwoSampleMR package was used to conduct all statistical analyses in R version 4.0.3. Ethical approval was not required for the use of the summary data, as all data are publicly available.

3 Results

3.1 Investigation of plasma proteins affected by cold environments

Seventy-eight plasma proteins were significantly associated with cold environments based on a preliminary analysis of 2992 plasma protein levels using the IVW method ($P < 0.05$) (Supplementary Data 2). To enhance the sensitivity of the MR analysis, the statistical significance threshold was set to $P < 0.01$ (highly significant). Ten plasma proteins passed this higher significance threshold ($P < 0.01$). The top 10 proteins identified were Allergin-1, Protein TMEPAI, Mucin-1, Protein FAM19A5, DnaJ

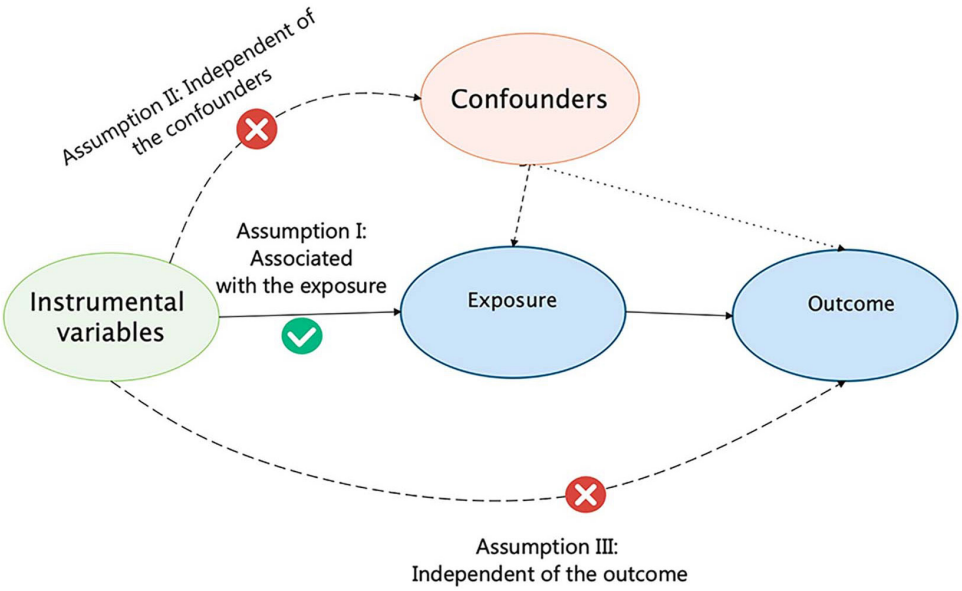

Fig. 1 The basic principles of Mendelian randomization

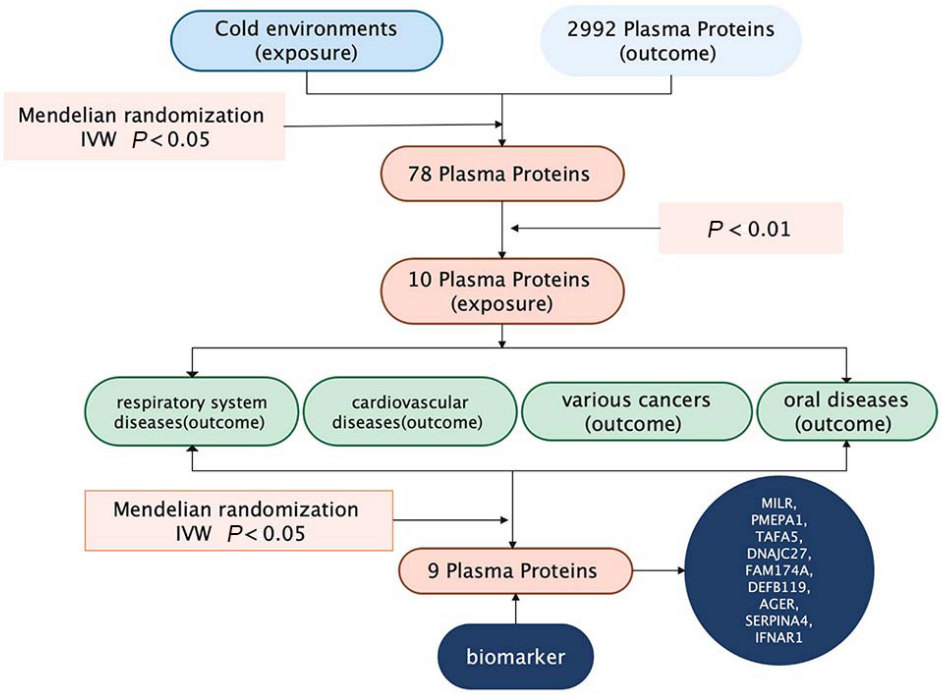

Fig. 2 Flowchart of the study design

Homolog Subfamily C Member 27, Membrane Protein FAM174A, Beta-Defensin 119, Advanced Glycosylation End Product-Specific Receptor, Kallistatin, and Interferon Alpha/Beta Receptor 1.

Extreme cold environments were found to increase the levels of Allergin-1 (MILR1), Protein TMEPAI (PMEPA1), Mucin-1 (MUC1), Protein FAM19A5 (TAFA5), DnaJ Homolog Subfamily C Member 27 (DNAJC27), Membrane Protein FAM174A (FAM174A), Advanced Glycosylation End Product-Specific Receptor (AGER), Kallistatin (SERPINA4), and Interferon Alpha/Beta Receptor 1 (IFNAR1), while decreasing the levels of Beta-Defensin 119 (DEFB119). It can be found in Fig. 3.

These plasma proteins may serve as potential biomarkers for monitoring workers' health during extreme cold weather exposure.

3.2 A biomarker for health monitoring based on plasma proteins

An MR analysis was conducted to assess the association between the concentrations of 10 plasma proteins and 55 diseases, evaluating their potential link to disease risk. In contrast, although MUC1 was influenced by cold environments, it was not found to be associated with any of the diseases tested (Fig. 4).

DEFB119, which is reduced in cold environments, may provide protection against hypertension, unstable angina pectoris, ischemic heart disease, and malignant neoplasms of the colon and ovary.

In contrast, FAM174A, TAFA5, IFNAR1, MILR1, MUC1, PMEPA1, SERPINA4, AGER, and DNAJC27, which are elevated in cold environments, have been identified risk factors for the following diseases: Acute bronchitis (FAM174ATAFA5, PMEPA1), Adult respiratory distress syndrome (TAFA5, DNAJC27), COPD (MILR1,SERPINA4), Viral pneumonia (FAM174A), pneumoniae (TAFA5, PMEPA1), Pneumothorax (SERPINA4), Heart failure (IFNAR1, PMEPA1), ischemic heart disease (MILR1), Non-ischemic cardiomyopathy (PMEPA1), Paroxysmal tachycardia (MILR1, DNAJC27), Right bundle-branch block (SERPINA4), Malignant melanoma (FAM174A, AGER), Malignant neoplasm of intrahepatic ducts, biliary tract and gallbladder (MILR1, DNAJC27), Malignant neoplasm of prostate (SERPINA4), Malignant neoplasm of rectum (SERPINA4), Head and neck cancer (ieu-b-4912) (AGER), Oral and oropharyngeal cancer (ieu-b-4962) (MILR1, AGER), Oropharyngeal cancer (ieu-b-4968) (MILR1), Bruxism (TAFA5), Oral leukoplakia (MILR1, SERPINA4, AGER), Mouth ulcers (TAFA5, PMEPA1), Erythema multiforme (DNAJC27) and Sicca syndrome (FAM174A, PMEPA1).

Nine plasma proteins have been identified as potential markers of cold environments and associated diseases. These candidate biomarkers may prove valuable for monitoring the health of individuals living in cold environments.

4 Discussion

As a result of climate change, extreme temperatures have become more prevalent, and mortality rates have increased. The Global Burden of Disease Study has revealed that nonoptimal temperatures are now among the top 10 risk factors for mortality worldwide. To mobilize healthcare resources and inform public health recommendations, extreme temperature analysis can be used to identify vulnerable subgroups and disease outcomes[16-18]. Several plasma proteins were found to be affected by cold environments in this study, indicating that cold environments affect specific plasma proteins.

The thermoregulatory mechanisms in humans have evolved over millions of years, making them resilient and dependable. While it is well-documented that cold temperatures can elevate the incidence and fatality rates of cardiovascular diseases, the overall impact of cold exposure remains ambiguous[19-20].

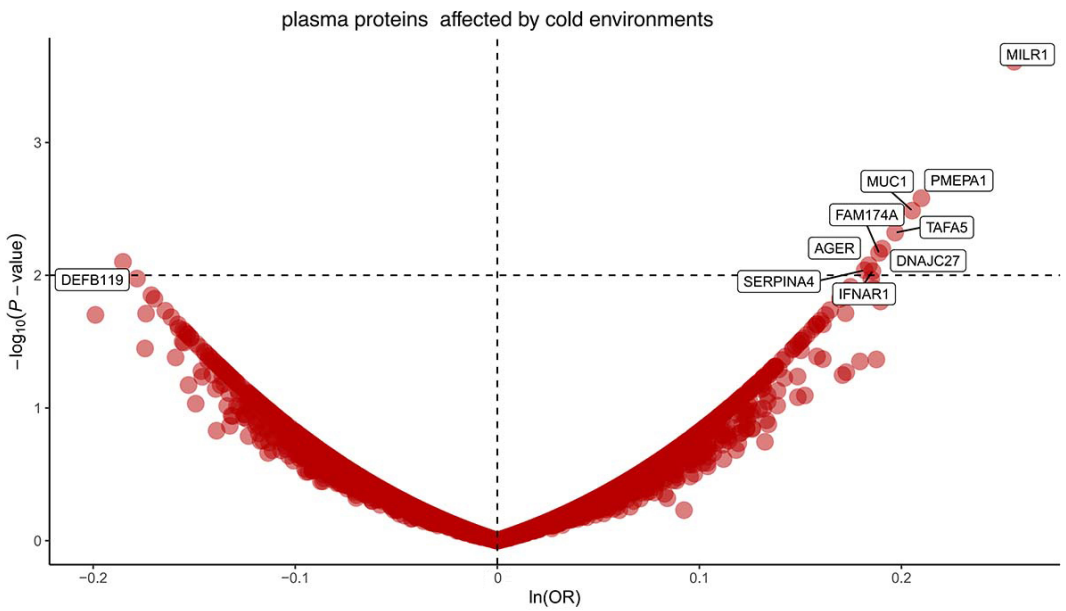

Fig. 3 Plasma proteins affected by cold environments

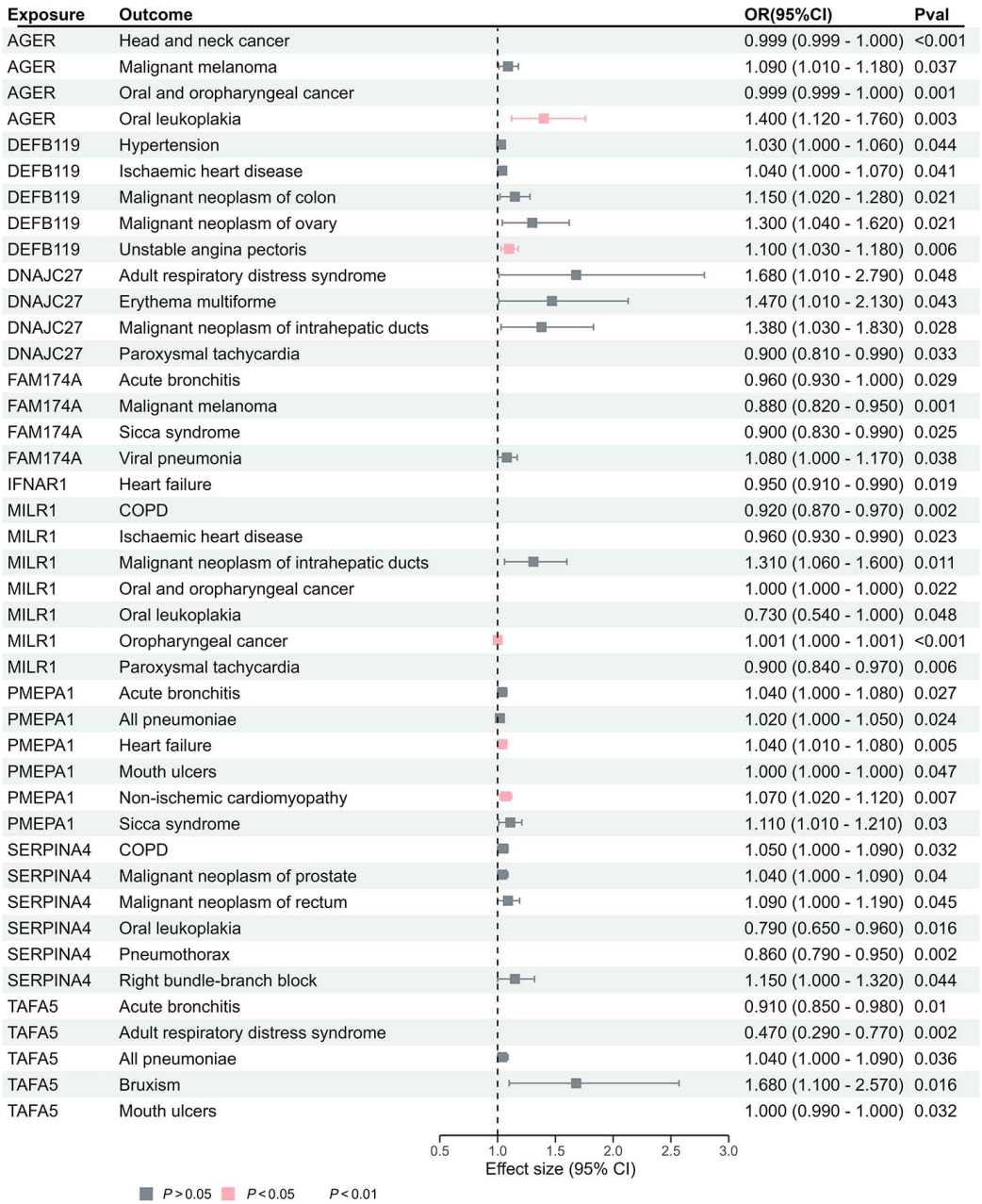

Fig. 4 Estimated causal effects of plasma proteins on selected diseases using MR

The current study reveals that exposure to cold environments has discernible effects on the concentrations of plasma proteins. Moreover, this study demonstrated that occupational cold exposure leads to elevated plasma levels of SERPINA4, which is a known risk factor for prostate cancer, rectal cancer, and oral leukoplakia. A recent study also found that SERPINA4 plays a significant inhibitory role in malignant tumor progression, suggesting its potential as a prognostic marker and therapeutic target for colorectal cancer[21]. These findings align with our results, reinforcing the consistency of these observations.

Additionally, the study showed that cold exposure leads to elevated plasma levels of MILR1, which is a risk factor for COPD, paroxysmal tachycardia, ischemic heart disease, oral and oropharyngeal cancer, oral leukoplakia, and malignant neoplasm of intrahepatic ducts, biliary tract, and gallbladder. A recent investigation also revealed that Allergin-1, an immunoglobulin-like receptor,

functions to inhibit immediate hypersensitivity reactions mediated by immunoglobulin E. Allergin-1 may be involved in the onset of diseases through an immune-related mechanism[22]. MR analyses in the present study further revealed that cold environments resulted in elevated levels of FAM174A, PMEPA1, and TAFA5 in the plasma, contributing to the development of various respiratory system diseases associated with these proteins. Additionally, this study uncovered correlations between AGER-encoded proteins influenced by the environment and oral pathologies.

Based on the findings of this study, it is plausible to infer that DEFB119 exhibits reduced expression under cold working conditions. This gene encodes a constituent of the beta subfamily of defensins, which are antimicrobial peptides that protect tissues and organs from a wide range of microorganisms, thereby preventing infection[23]. In this study, DEFB119 may serve as a protective factor against conditions such as hypertension, unstable angina pectoris, ischemic heart disease, malignant neoplasm of the colon, and malignant neoplasm of the ovary. Collectively, these findings suggest that environmental factors affect plasma protein expression levels, which, in turn, influence the development of various diseases.

The results of this study highlight the impact of cold environments on the levels of multiple plasma proteins. The observed changes in plasma protein concentrations present a promising opportunity for using these proteins as biomarkers to predict risks and hazards. Nine distinct plasma proteins are proposed as potential biomarkers for monitoring the health status of individuals exposed to cold environments. While the MR analysis employed in this study is considered reliable and robust, further research is needed to validate these biomarkers. Additional epidemiological investigations are warranted to evaluate the sensitivity, specificity, and overall accuracy of these prospective biomarkers.

Acknowledgments

We thank all the investigators and participants for their contribution to the GWAS and making the summary data publicly available.

Research ethics

This study was based on publicly available datasets. Ethical review and approval was not required for the study, in accordance with the local legislation and institutional requirements.

Informed consent

Informed consent was obtained from all subjects involved in this study.

Author contributions

Conceptualization: Yan J Q, Jiao X H; Methodology: Song H Q, Zhang R; Software: Song H Q, Validation: Lu Z X; Formal analysis: Lu Z X, Zhang R; Investigation: Jiao X H; Resources: Song H Q; Data Curation: Song H Q, Zhang R; Writing, original draft preparation: Song H Q, Zhang R, Lu Z X; Writing, review and editing: Song H Q, Yan J Q, Jiao X H; Visualization: Yan J Q, Jiao X H; Supervision: Song H Q; Project administration: Yan J Q, Jiao X H; Funding acquisition: Yan J Q, Jiao X H.

Use of Large Language Models, AI and Machine Learning Tools

In this study, we utilized the R programming language and its associated packages for data analysis and processing.

Conflict of interest

The authors declare no conflict of interest.

Research funding

This study was funded by the Health Commission of Heilongjiang Province (Project Number: 20230808010517).

Data availability

We declare that the data presented in this study are available.

References

[1] Agache I, Sampath V, Aguilera J, *et al.* Climate change and global health: A call to more research and more action. Allergy, 2022; 77(5): 1389-1407.
[2] Ebi K L, Vanos J, Baldwin J W, *et al.* Extreme weather and climate change: population health and health system implications. Annu Rev Public Health, 2021; 42: 293-315.
[3] Ashrafuzzaman M, Furini G L. Climate change and human health linkages in the context of globalization: An overview from global to southwestern coastal region of Bangladesh. Environ Int, 2019; 127: 402-411.
[4] Chen R, Yin P, Wang L, *et al.* Association between ambient

temperature and mortality risk and burden: time series study in 272 main Chinese cities. BMJ, 2018; 363: k4306.
[5] González Hernández E, Cabadés O'Callaghan A, Cebrián Doménech J, *et al.* Variaciones estacionales en los ingresos por infarto agudo de micardio. El estudio PRIMVAC [Seasonal variations in admissions for acute myocardial infarction. The PRIMVAC study]. Rev Esp Cardiol, 2004; 57(1): 12-19.
[6] Geyer P E, Kulak N A, Pichler G, *et al.* Plasma proteome profiling to assess human health and disease. Cell Syst, 2016; 2(3): 185-195.
[7] Geyer P E, Holdt L M, Teupser D, *et al.* Revisiting biomarker discovery by plasma proteomics. Mol Syst Biol, 2017; 13(9): 942.
[8] Anderson N L. The clinical plasma proteome: a survey of clinical assays for proteins in plasma and serum. Clin Chem, 2010; 56(2): 177-185.
[9] Davey Smith G, Hemani G. Mendelian randomization: genetic anchors for causal inference in epidemiological studies. Hum Mol Genet, 2014; 23(R1): R89-R98.
[10] Sun B B, Maranville J C, Peters J E, *et al.* Genomic atlas of the human plasma proteome. Nature,2018; (2018) 558: 73-79.
[11] Hemani G, Zheng J, Elsworth B, *et al.* The MR-Base platform supports systematic causal inference across the human phenome. Elife, 2018; 7: e34408.
[12] Sudlow C, Gallacher J, Allen N, *et al.* UK biobank: an open access resource for identifying the causes of a wide range of complex diseases of middle and old age. PLoS Med, 2015; 12(3): e1001779. .
[13] He Y, Zhang H, Wang T, *et al.* Impact of serum calcium levels on Alzheimer's disease: A mendelian randomization study. J Alzheimers Dis, 2020; 76(2): 713-724.
[14] Zhang H, Wang T, Han Z, *et al.* Mendelian randomization study to evaluate the effects of interleukin-6 signaling on four neurodegenerative diseases. Neurol Sci, 2020; 41(10): 2875-2882.
[15] Li A, Liao W, Xie J, *et al.* Plasma proteins as occupational hazard risk monitors for populations working in harsh environments: A mendelian randomization study. Front Public Health, 2022; 10: 852572.
[16] Vicedo-Cabrera A M, Scovronick N, Sera F, *et al.* The burden of heat-related mortality attributable to recent human-induced climate change. Nat Clim Chang, 2021; 11(6): 492-500.
[17] GBD 2019 Risk Factors Collaborators. Global burden of 87 risk factors in 204 countries and territories, 1990-2019: a systematic analysis for the Global Burden of Disease Study 2019. Lancet, 2020; 396(10258): 1223-1249.
[18] Zhao Q, Guo Y, Ye T, *et al.* Global, regional, and national burden of mortality associated with non-optimal ambient temperatures from 2000 to 2019: a three-stage modelling study. Lancet Planet Health, 2021; 5(7): e415-e425.
[19] Moholdt T, Afoakwah C, Scuffham P, *et al.* Excess mortality at Christmas due to cardiovascular disease in the HUNT study prospective population-based cohort in Norway. BMC Public Health, 2021; 21(1): 549.
[20] Li Y, Zhou Z, Chen N, *et al.* Seasonal variation in the occurrence of ischemic stroke: A meta-analysis. Environ Geochem Health, 2019; 41(5): 2113-2130.
[21] Sun H M, Mi Y S, Yu F D, *et al.* SERPINA4 is a novel independent prognostic indicator and a potential therapeutic target for colorectal cancer. Am J Cancer Res, 2016; 6(8): 1636-1649.
[22] Hitomi K, Tahara-Hanaoka S, Someya S, *et al.* An immunoglobulin-like receptor, Allergin-1, inhibits immunoglobulin E-mediated immediate hypersensitivity reactions. Nat Immunol, 2010; 11(7): 601-607.
[23] Pazgier M, Hoover D M, Yang D, *et al.* Human beta-defensins. Cell Mol Life Sci, 2006; 63(11): 1294-1313.