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# Title: Depressive Symptoms and Levels of C-reactive Protein: A Population Based Study

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Abstract: Background: Depression and depressive symptoms have been repeatedly linked to elevated levels of C-reactive protein (CRP) but questions remain as to the statistical robustness of the association and particularly whether the association between depression and CRP reflects the presence of a chronic disease.
Methods: A random sample of 6,126 men and women aged 45-69 years was examined in a cross-sectional study in 7 towns in the Czech Republic. Depressive symptoms were assessed by the CESD scale.
Results: CESD score was significantly related to increased levels of CRP in a linear fashion. After controlling for a range of potential confounders, subjects with depressive symptoms (CESD score ≥16)
had CRP concentrations 0.43 mg/l (95% CI 0.16 to 0.72) higher than those without symptoms. The association remained significant when study sample was restricted to healthy subjects; among individuals who did not report any chronic disease, the difference between those with and without

depressive symptoms was 0.44 mg/l (95% CI 0.14 to 0.74), and among persons who did not visit a doctor in the last 12 months the difference was 1.20 mg/l (95% CI 0.52 to 1.87).
Conclusions: These results confirm that there is a statistically robust association between depressive symptoms and increased levels of CRP. We did not find evidence that the association is due presence of a chronic condition.

Response to Reviewers: Our response is in a separate attachement.

**Social Psychiatry and Psychiatric Epidemiology**

**Pikhart et al: Depressive Symptoms and Levels of C-reactive Protein: A Population Based Study**

**Response to reviewer's comments**

We would like to thank the reviewer for thoughtful and constructive comments. Below, we list all reviewer's comments and our responses. We also attach the manuscript with changes described below.

Reviewer #1:

*1. In the methods section on the subjects I find the numbers to be unclear. Does the response rate of 55% on line 57 refer to 55% from the whole original sample and this is the 8856? They then say that the proportion of subjects with full data is 82% but 82% of 8856 is not the 6126 mentioned as the final sample. A flow chart or extended description here is needed.*

We agree with reviewer that the description is unclear. We have changed the sample description into following:
The subjects were first visited at home, to complete a structured questionnaire (N=8856, response rate 55%), and then invited to a clinic for a short examination. For this reason, not all subjects have data on both questionnaire and examination; the proportion of subjects with full data is 82% (N=7264). Only subjects with complete data on the outcome, valid CRP measurements, and all other covariates were used in presented analyses (N=6126).

*2. On the next page on lines 37-40 they talk about re-calculating the CESD scores but do not explain how this was done and they should do so.*

We have added explanation how this re-calculating was done (page 5):
The depression score was calculated if at least 16 out of 20 questions were answered; if fewer than 20 questions were answered, the score was recalculated to have values between 0 and 60 (average scores were calculated if a minimum of 16 out of 20 questions on depressive symptoms contained valid answers, and then they were multiplied by 20 to give scores between 0 and 60).

*3. In their discussion they appropriately make the point that they were not assessing clinical depression but depressive symptoms. They might add that the way in which they have obtained data, that is by first visiting with a questionnaire and then inviting people to clinic, makes it likely that those with more severe clinical depression were not sampled and that perhaps their findings would be slightly stronger if such subjects had been included.*

We thank referee for this useful comment. Prompted by the reviewer, we have compared subjects included in the analysis with those who completed the questionnaire but did not come to examination at the clinic. Although we found some differences in smoking prevalence and self rated health, we have not seen important differences in CESD scores and other health measures, such as diabetes or hypertension. This suggests that excluding subjects with missing data did not introduce a major selection bias.

We have added a brief section regarding the non-respondents and clinic non-attendees to the discussion (page 10).

# Depressive Symptoms and Levels of C-reactive Protein: A Population Based Study

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**Abstract**

**Background:** Depression and depressive symptoms have been repeatedly linked to elevated levels of C-reactive protein (CRP) but questions remain as to the statistical robustness of the association and particularly whether the association between depression and CRP reflects the presence of a chronic disease.

**Methods:** A random sample of 6,126 men and women aged 45-69 years was examined in a cross-sectional study in 7 towns in the Czech Republic. Depressive symptoms were assessed by the CESD scale.

**Results:** CESD score was significantly related to increased levels of CRP in a linear fashion. After controlling for a range of potential confounders, subjects with depressive symptoms (CESD score ≥16) had CRP concentrations 0.43 mg/l (95% CI 0.16 to 0.72) higher than those without symptoms. The association remained significant when study sample was restricted to healthy subjects; among individuals who did not report any chronic disease, the difference between those with and without depressive symptoms was 0.44 mg/l (95% CI 0.14 to 0.74), and among persons who did not visit a doctor in the last 12 months the difference was 1.20 mg/l (95% CI 0.52 to 1.87).

**Conclusions:** These results confirm that there is a statistically robust association between depressive symptoms and increased levels of CRP. We did not find evidence that the association is due presence of a chronic condition.

**Key words:**

Depressive symptoms, CRP, inflammation

## Introduction

Depression and depressive symptoms are common in many different populations [2;18;37], and it has been estimated that depression will be the second major source of disability worldwide by 2030 and the main source of disability in high-income countries [23]. In addition to the burden of depression per se, there is also an extensive literature suggesting that depression and depressive symptoms are associated with cardiovascular morbidity and mortality [14;15;17;25]. An important issue in assessing the causality of this association is identification of the underlying biological mechanisms.

In recent years, several authors hypothesised that psychological stress may lead to chronic activation of inflammatory processes [5;19]. Indeed, over the last decade, several studies reported depression and depressive symptoms to be associated with elevated levels of CRP [10;12;20;35;36]. It has been proposed that such chronic inflammation may lead to atherosclerosis and cardiovascular disease (CVD) [6;7;31]. In such a way inflammatory markers, such as C-reactive protein (CRP), could provide the link between psychological stress, depression and atherosclerosis or CVD. Although a recent meta-analysis of prospective studies [9] and a Mendelian randomisation study [8] suggest that the effect of serum CRP on the risk of coronary heart disease is weaker than previously thought, the possible association between depression and CRP remains of major scientific interest, because of the potential effect of psychosocial and psychological factors on immunological functions. [33;34;40]

There are, however, several problems with assessing whether the association between CRP and depression is genuine and causal. First, most studies addressing this issue were relatively small. Second, virtually all studies were conducted in western countries; in establishing whether an association is genuine, it is important to examine its consistency across populations. Since depression shows a social gradient [21], and since the social patterning of different confounders (for example

smoking , body-mass index and waist-hip ratio) differs between eastern and western Europe, a study in a non-western population would help to break the socioeconomic confounding. Third, the independence of the relationship is not clear; it is possible the higher levels of CRP reflect presence of existing chronic condition, such as heart disease, which could also lead to depression (i.e. confounding by an existing disease)[1].

In this report, we analysed cross-sectional data from the Czech Republic, with the objective to test the hypothesis that CRP levels are positively associated with depressive symptoms. The size, scope and location of the study should allow us to address, at least in part, some of the problems described above, namely (1) the statistical stability and precision of the association, (2) the socioeconomic confounding, (3) whether the association is replicated in non-western study populations, and (4) the effect of existing chronic disease.

## Methods

### *Subjects*

The data come from the Czech part of the HAPIEE (Health, Alcohol and Psychosocial factors In Eastern Europe) study, a baseline survey for a longitudinal study of chronic diseases, conducted in six towns in the Czech Republic (Havirov/Karvina, Hradec Kralove, Jihlava, Kromeriz, Liberec and Usti nad Labem) in 2002-2005. The study has been described in detail elsewhere [27]. Briefly, men and women aged 45-69 years, stratified by gender and 5 year age groups and randomly selected from population registers, were invited to participate. The subjects were first visited at home, to complete a structured questionnaire (N=8856, response rate 55%), and then invited to a clinic for a short examination. For this reason, not all subjects have data on both questionnaire and examination; the proportion of subjects with full data is 82% (N=7264). Only subjects with complete data on the

outcome, valid CRP measurements, and all other covariates were used in presented analyses (N=6126).

The study received ethical approval from the UCL/UCLH joint research ethics committee and from the ethical committee in the Czech National Institute of Public Health. All participants gave informed consent.

*Measurements*

The questionnaire covered health (self-rated health status, medical history, health behaviours, life style, socioeconomic circumstances, psychosocial factors and depressive symptoms). All questions were translated from English into Czech and back translated into English to check for accuracy.

Depressive symptoms were measured using the Center for Epidemiologic Studies Depression (CESD) scale [28]; the scale has previously been used and evaluated in the Czech Republic[26]. This instrument consists of 20 self-reported items (presence of symptoms in the past week) and scores range between 0 and 60. The depression score was calculated if at least 16 out of 20 questions were answered; if fewer than 20 questions were answered, the score was recalculated to have values between 0 and 60 (average scores were calculated if a minimum of 16 out of 20 questions on depressive symptoms contained valid answers, and then they were multiplied by 20 to give scores between 0 and 60). The subjects with a score of 16 and above (shown to be predictive of major depressive disorder in a range of populations [3;22;32]) have been classified as having depressive symptoms. Subsequently, the full scale was used in additional analyses to confirm results based on the binary outcome.

Education was classified into 3 categories: primary or less, secondary or vocational, and university. Material deprivation was evaluated by three separate questions asking whether it ever happened that participants did not have enough money for food, for clothes or ever had problems paying household

bills. Each question had five response levels: all the time, often, sometimes, rarely or never. A cumulative score was constructed using these responses. The following covariates were also included: smoking (categorized as never smoker, past smoker, current smoker), physical activity (number of hours per week), alcohol consumption (mean dose of alcohol per drinking occasion and annual intake of alcohol, both estimated from the graduated frequency questionnaire [16;29;30]), and serum concentration of total and HDL-cholesterol, and triglycerides. As indicators of obesity, we used measures of body-mass index (BMI) and waist-hip ratio (WHR). Finally we used measurement of systolic blood pressure (SBP) and diastolic blood pressure (DBP) (blood pressure was measured three times in sitting position, with a two minute interval between measurements, using an Omron M5-I digital blood pressure monitor; and we used arithmetic mean of $2^{nd}$ and $3^{rd}$ measurement).

The examination included a fasting venous blood sample. Venous blood samples were collected in EDTA-treated tubes, centrifuged within 2 hours of venepuncture, and plasma samples were stored at -80 ºC. The CRP assay used in the study was designed to measure high sensitive CRP - ultrasensitive CRP was determined by immunoturbidimetry (Orion Diagnostica, Espoo, Finland, kit no. 68025), using a Cobas Mira autoanalyzer (Hoffman-LaRoche, USA). The lower detection limit of the assay was 0.25 mg/l. The intra-assay coefficient of variation of the CRP levels was 2.88% at 2 mg/l, while the inter-assay coefficient of variation was 4.84% at 2 mg/l. All samples were analysed at lipid laboratory of Institute for Clinical and Experimental Medicine, Prague, Czech Republic.

*Statistical analysis*

Depressive symptoms were initially analysed both as continuous (the CESD score; both original and logarithmically transformed values) and dichotomous variables (CESD scores of 16 and above). Because both analyses produced essentially identical results, the results on the dichotomised outcomes are reported here.

The analytical strategy was as follows. First, summary measures of all variables were cross tabulated by gender. Second, since there was no statistically significant interaction between depression and gender (p=0.88 for likelihood ratio test comparing models with and without the interaction terms), we pooled the data for men and women, and we estimated the age-sex-adjusted difference in CRP concentrations between persons with and without depressive symptoms, using linear regression. Third, the difference in CRP in persons with and without depressive symptoms was further adjusted for socioeconomic factors (education and deprivation), obesity, health behaviours, serum lipids and blood pressure, in order to take into account potential confounding. Finally, we compared the fully adjusted results in the full sample with results in subsets of people who (1) reported no previous history of MI, angina, stroke or cancer; reported history of MI, stroke or angina; and those who reported history of cancer; (2) reported no long-term health problems for which medical treatment has been sought over last 12 months, and those who reported such problems; (3) reported no visit to a doctor in last 12 months, reported 1-3 visits and those who reported 4 or more visits. All analyses were performed using STATA version 9 for Windows.

**Results**

The descriptive characteristics of the study sample are presented in table 1. There were 6126 individuals (2829 men and 3297 women) with valid CRP measurements, CESD scores and all other covariates. Mean levels of CRP were 2.39 mg/l for men and 2.60 mg/l for women. The prevalence of depressive symptoms (CESD score 16+) was 13.0% in men and 22.8% in women. Men had higher education than women (18.8% of men and 10.7% of women reported university education) and more than 70% of population were in the secondary or vocational education category. 29% of men and 23% of women were current smokers. More than half of the women had never smoked compared to one

third of men. Men and women reported similar physical activity, and the mean body mass index was high (above 28 kg/m$^2$ in both genders). Men reported higher systolic blood pressure than women (144 mmHg vs. 134 mmHg), lower total cholesterol (5.61 mmol/l vs. 5.82 mmol/l), lower HDL and higher triglycerides. Both systolic blood pressure and total cholesterol levels were high in this population.

There was a strong association between depressive symptoms and CRP in both genders: the age-adjusted difference between those reporting depressive symptoms and those who did not was 0.57 mg/l (0.10-1.04) for men and 0.61 (0.27-0.96) mg/l for women. The effect of depressive symptoms was not statistically significantly modified by gender. Data from men and women were therefore pooled (table 2). The unadjusted mean CRP concentrations were 0.60 mg/l (95% CI 0.32 to 0.87) higher in subjects with depressive symptoms than in those without symptoms. This association persisted at all levels of adjustment. The major reduction in the effect was observed after controlling for education, material deprivation and body mass index but even then the relationship remained statistically significant. Further adjustment for smoking, alcohol consumption, physical activity, serum lipid levels and systolic blood pressure did not reduce the association. In the fully adjusted model, subjects with depressive symptoms had CRP concentrations 0.44 mg/l (95% CI 0.16 to 0.72) higher than those without symptoms.

Since presence of an existing disease may affect both CRP levels and depression, we compared the results in the full sample, reported above, with subsets of subjects stratified by health conditions reported at the baseline (table 3). Both the prevalence of depressive symptoms and mean CRP were higher in persons who reported history of myocardial infarction, angina, stroke or cancer, who had a long-term illness, and who visited a doctor 4 or more times in the last year. However, the difference in CRP between persons with and without depressive symptoms was also present in apparently healthy

participants. If anything, the association between depressive symptoms and CRP tended to be smaller in persons with long-term health problem and in those who frequently visited a doctor.

As mentioned in Methods, we analysed CESD score as both dichotomised and continuous variables. Both analyses showed virtually identical pattern. We found an approximately linear association between CRP concentration and the CESD scale; an increase in CESD score by 1 SD was associated with an increase in CRP of 0.23 mg/l (95% CI 0.12 to 0.34) in age-sex adjusted analysis and 0.14 mg/l (95% CI 0.03 to 0.25) in the fully adjusted model. Figure 1 shows mean values of CRP (with 95% CI) by CESD score categories. Because CESD score was not normally distributed we also used logarithmically transformed CESD score as a continuous variable. We have found that an increase in log(CESD score) by 1 SD was associated with an increase in CRP of 0.19 mg/l (95% CI 0.09 to 0.30) in age-sex adjusted analysis and 0.12 mg/l (95% CI 0.01 to 0.22) in the fully adjusted model.

**Discussion**

To our knowledge, this is one of the largest population-based studies examining the association between depression and CRP so far, and one of the first CRP studies conducted in Central and Eastern Europe. We found a strong and statistically robust positive association between presence of depressive symptoms and CRP. The association was not explained by extensive adjustment for socioeconomic and behavioural covariates, and it was not attenuated by restricting the sample to subjects free of existing disease.

When interpreting the results of this study, several limitations need to be considered. Firstly, though CESD is an internationally recognized, extensively used and validated instrument [13], it is not a measure of clinical depression. Participants who scored above the threshold of 16 points included

persons with minor distress states and anxiety disorders. The scale may also detect some personality characteristics, for example high negative affectivity [39]. The CESD measure is therefore not unambiguously specific to depression. In addition, it is possible that non-respondents had more severe clinical depression than respondents, and the results may have been slightly stronger if non-respondents had been included. On the other hand, persons who completed questionnaire during a home visit but who did not attend examination in a clinic had CESD scores similar to those with full data; this suggest that non-attendance to a clinic did not introduce a major selection bias.

Secondly, it is not possible to assess temporality using a cross-sectional design. While we assessed the role of existing conditions on the relationship between depression and CRP indirectly, by restricting the sample, we could not investigate this relationship prospectively over time. We therefore cannot make definitive statements about the causality of the association between presence of depressive symptoms and elevated CRP levels.

Finally, we did not have information on recent infections. It has been previously shown that infections (which involve immune activation and cytokine release) were associated with psychological distress [12;24]. However, when we restricted the analyses to only those who had not visited doctor in the last 12 months, the association between depressive symptoms and CRP was even stronger than in the whole dataset. Assuming that most persons with acute infections would visit a doctor, these results do not support a major role of acute infections in the observed association between CRP and depression.

The present findings are consistent with previous studies reporting the association between depression or depressive symptoms and CRP levels[10;12] in confirming the presence of the relationship. But our results differ in the extent to which other factors partly confound/mediate it. In previous studies, overweight/obesity either explained most of the association between CRP and depression [11] or the

association was substantially attenuated by adjustment for obesity [12]. In our study, body mass index and waist hip ratio did reduce the effect of presence of depressive symptoms on CRP levels but the association remained strong and statistically significant in both genders. We also examined whether the effect of depressive symptoms on CRP differed by the level of obesity. However, we found no evidence for an interaction between depressive symptoms and body-mass index. In addition, our data do not suggest the existence of any threshold. When we used score of depressive symptoms as a continuous variable, we found a statistically significant linear association between CESD-score and CRP, indicating a relationship existing even in those not classified as depressed. This dose-response effect of the depressive symptoms score on CRP is consistent with a genuine relationship, rather than an artefact. The association between depression and CRP is biologically plausible, as depression may lead to activation of inflammation system via autonomic nervous system[4;38].

The fact that the study was conducted in Eastern Europe is important. In the West, both depression and CRP show a socioeconomic gradient, and the relationship between them may therefore be due to social confounding. As the social patterning of disease and risk factors in Eastern Europe is generally weaker than in the West, and indicators of material circumstances are only weakly related to health outcomes in Eastern Europe, our finding of a robust association between depression and CRP minimises the risk of residual confounding by socioeconomic status.

A crucial question about the relationship between depression and CRP is the possibility that both depression and CRP are the result of a chronic disease. The size of this study was sufficient to allow us to stratify the study sample by several markers of a presence of chronic health conditions. As expected, both depression and CRP were positively associated with such markers. The relationship between depressive symptoms and CRP, however, was not attenuated in the sample restricted to the apparently healthy subjects. While the cross-sectional nature of the study does not allow causal inference, these

results do not support the common cause argument and suggest that the observed association between depression and raised CRP is not due to poor physical health.

Reference List


1. Almeida OP, Norman P, Hankey GJ, Jamrozik K, Flicker L. The association between C-reactive protein concentration and depression in later life is due to poor physical health: results from the Health in Men Study (HIMS). Psychol Med 2007; 37:1775-86.

2. Ayuso-Mateos JL, Vazquez-Barquero JL, Dowrick C et al. Depressive disorders in Europe: prevalence figures from the ODIN study. The British Journal of Psychiatry 2001; 179:308-16.

3. Beekman AT, Deeg DJ, Van Limbeek J, Braam AW, De Vries MZ, Van Tilburg W. Criterion validity of the Center for Epidemiologic Studies Depression scale (CES-D): results from a community-based sample of older subjects in The Netherlands. Psychol Med 1997; 27:231-5.

4. Bierhaus A, Wolf J, Andrassy M et al. A mechanism converting psychosocial stress into mononuclear cell activation. Proceedings of the National Academy of Sciences 2003; 100:1920-5.

5. Black PH, Garbutt LD. Stress, inflammation and cardiovascular disease. J Psychosom Res 2002; 52:1-23.

6. Blake GJ, Ridker P. Inflammatory biomarkers and cardiovascular risk prediction. J Intern Med 2002; 252:283-94.

7. Boekholdt S, Hack C, Sandhu M et al. C-reactive protein levels and coronary artery disease incidence and mortality in apparently healthy men and women: The EPIC-Norfolk prospective population study 1993-2003. Atherosclerosis 2006; 187:415-22.

8. Casas JP, Shah T, Cooper J et al. Insight into the nature of the CRP–coronary event association using Mendelian randomization. Int J Epidemiol 2006; 35:922-31.

9. Danesh J, Wheeler JG, Hirschfield GM et al. C-reactive protein and other circulating markers of inflammation in the prediction of coronary heart disease. N Engl J Med 2004; 350:1387-97.

10. Danner M, Kasl SV, Abramson JL, Vaccarino V. Association between depression and elevated C-reactive protein. Psychosom Med 2003; 65:347-56.

11. Douglas KM, Taylor AJ, O'Malley PG. Relationship Between Depression and C-Reactive Protein in a Screening Population. Psychosom Med 2004; 66:679-83.

12. Elovainio M, Keltikangas-Jarvinen L, Pulkki-Raback L et al. Depressive symptoms and C-reactive protein: the Cardiovascular Risk in Young Finns Study. Psychol Med 2006; 36:797-805.

13. Fountoulakis K, Iacovides A, Kleanthous S et al. Reliability, validity and psychometric properties of the Greek translation of the Center for Epidemiological Studies-Depression (CES-D) Scale. BMC Psychiatry 2001; 1:3.

14. Gump BB, Matthews KA, Eberly LE, Chang YF. Depressive symptoms and mortality in men. Results from the Multiple Risk Factors Intervention Trial. Stroke 2005; 36:98-102.

15. Hemingway H, Marmot M. Evidence based cardiology: psychosocial factors in the aetiology and prognosis of coronary heart disease. Systematic review of prospective cohort studies. BMJ 1999; 318:1460-7.

16. Hilton ME. A comparison of a prospective diary and two summary recall techniques for recording alcohol consumption. Br J Addict 1989; 84:1085-92.

17. Kamphuis MH, Kalmijn S, Tijhuis MAR et al. Depressive symptoms as risk factor of cardiovascular mortality in older European men: the Finland, Italy and Netherlands Elderly

(FINE) study. European Journal of Cardiovascular Prevention and Rehabilitation 2006; 13:199-206.

18. Kessler RC, McGonagle KA, Zhao S et al. Lifetime and 12-month prevalence of DSM-II-R psychiatric disorders in the United States. Results from the National Comorbidity Survey. Archives of General Psychiatry 1994; 51:8-19.

19. Kop WJ. The integration of cardiovascular behavioral medicine and psychoneuroimmunology: New developments based on converging research fields. Brain,Behavior and Immunity 2003; 17:233-7.

20. Liukkonen T, Silvennoinen-Kassinen S, Jokelainen J et al. The Association Between C-Reactive Protein Levels and Depression: Results from the Northern Finland 1966 Birth Cohort Study. Biol Psychiatry 2006; 60:825-30.

21. Lorant V, Deliege D, Eaton W, Robert A, Philippot P, Ansseau M. Socioeconomic inequalities in depression: a meta-analysis. Am J Epidemiol 2003; 157:98-112.

22. Lyness JM, Noel TK, Cox C, King DA, Conwell Y, Caine ED. Screening for depression in elderly primary care patients. Arch Intern Med 1997; 157:449-54.

23. Mathers CD, Loncar D. Projections of Global Mortality and Burden of Disease from 2002 to 2030. PLoS Med 2006; 3:e442.

24. Meijer A, Zakay-Rones Z, Morag A. Post-influenzal psychiatric disorder in adolescents. Acta Psychiatrica Scandinavica 1988; 78:176-81.

25. Nicholson A, Kuper H, Hemingway H. Depression as an aetiologic and prognostic factor in coronary heart disease: a meta-analysis of 6362 events among 146 538 participants in 54 observational studies. Eur Heart J 2006; 27:2763-74.

26. Osecká L, Škála deprese CESD - psychometrická analýza. Brno : Akademie ved Ceské republiky - Psychologický ústav, 1999.

27. Peasey A, Bobak M, Kubinova R et al. Determinants of cardiovascular disease and other non-communicable diseases in Central and Eastern Europe: rationale and design of the HAPIEE study. BMC Public Health 2006; 6:255.

28. Radloff LS. The CES-D scale: a self-report depression scale for research in the general population. Applied Psychological Measurement 1977; 1:385-401.

29. Rehm J. Measuring quantity, frequency and volume drinking. Alcoholism: Clinical and Experimental Research 1998; 22:4S-14S.

30. Rehm J, Greenfield TK, Walsh G, Xie X, Robson L, Single E. Assessment methods for alcohol consumption, prevalence of high risk drinking and harm: a sensitivity analysis. Int J Epidemiol 1999; 28:219-24.

31. Ridker P, Hennekens CH, Buring JE, Rifai N. C-reactive protein and other markers of inflammation in the prediction of cardiovascular disease in women. N Engl J Med 2000; 342:836-43.

32. Roberts RE, Vernon SW. The Center for Epidemiologic Studies Depression Scale: its use in a community sample. Am J Psychiatry 1983; 140:41-6.

33. Segerstrom S, Miller G. Psychological Stress and the Human Immune System: A Meta-Analytic Study of 30 Years of Inquiry. Psychological Bulletin 2004; 130:601-30.

34. Sher L. Psychological Factors, Immunity, and Heart Disease Psychosomatics. Psychosomatics 2000; 41:372-3.

35. Suarez EC. C-reactive protein is associated with psychological risk factors of cardiovascular disease in apparently healthy adults. Psychosom Med 2004; 66:684-91.

36. Toker S, Shirom A, Shapira I, Berliner S, Melamed S. The association between burnout, depression, anxiety, and inflammation biomarkers: C-reactive protein and fibrinogen in men and women. J Occup Health Psychol 2005; 10:344-62.

37. Ustun TB, Ayuso-Mateos JL, Chatterji S, Mathers C, Murray CJ. Global burden of depressive disorders in the year 2000. The British Journal of Psychiatry 2004; 184:386-92.

38. Vaccarino V, Johnson BD, Sheps DS et al. Depression, Inflammation, and Incident Cardiovascular Disease in Women With Suspected Coronary Ischemia: The National Heart, Lung, and Blood Institute-Sponsored WISE Study. Journal of the American College of Cardiology 2007; 50:2044-50.

39. Watson D, Clark LA. Negative affectivity; the disposition to experience aversive emotional states. Psychological Bulletin 1984; 96:465-90.

40. Zorrilla EP, Luborsky L, McKay JR et al. The Relationship of Depression and Stressors to Immunological Assays: A Meta-Analytic Review. Brain, Behavior, and Immunity 2001; 15:199-226.

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**Authors contributions:**

HP, AN, AP, RK and NC participated in the design and coordination of the study, and acquisition of the data. RP and JAH coordinated laboratory analysis. HP and MB conducted statistical analyses and drafted the manuscript. All the authors helped to draft and revise the manuscript. All authors read and approved the final manuscript.

**Competing interests:** None

**Ethical approval:** the study was approved by the UCL/UCLH joint research ethics committee and at the ethical committee in the Czech National Institute of Public Health.

**Table 1. Descriptive characteristics of the study sample**

| Variable | Category | Men | Women | p-value* |
|---|---|---|---|---|
| | | N=2829 | N=3297 | |
| | | | | |
| CRP | mg/l - mean (SD) | 2.39 (4.30) | 2.60 (4.26) | 0.08 |
| | | | | |
| CESD | 16 and more | 13.0 % | 22.8 % | <0.001 |
| | Mean (SD) | 8.8 (7.3) | 11.1 (9.1) | <0.001 |
| | | | | |
| | | | | |
| Age | Years - mean (SD) | 58.5 (7.2) | 57.7 (7.1) | <0.001 |
| | | | | |
| Education | Primary or less | 5.3 % | 17.3 % | <0.001 |
| | Secondary/vocational | 75.9 % | 72.1 % | |
| | University | 18.8 % | 10.7 % | |
| | | | | |
| Deprivation | Score (0=low depr. 12=high depr.) – mean (SD) | 1.4 (2.2) | 1.8 (2.4) | <0.001 |
| | | | | |
| Smoking | Never | 32.2 % | 54.4 % | <0.001 |
| | Past | 38.5 % | 22.6 % | |
| | Currently | 29.3 % | 23.0 % | |
| | | | | |
| Alcohol – mean dose per occasion | ml – mean (SD) | 35.9 (36.4) | 20.3 (22.8) | <0.001 |
| Alcohol – annual intake | Litres, mean (SD) | 6.5 (11.3) | 1.6 (5.2) | <0.001 |
| | | | | |
| Physical activity | Hours per week - mean (SD) | 4.5 (5.5) | 4.4 (5.3) | 0.87 |
| | | | | |
| Body-mass index (BMI) | $kg/m^2$ – mean (SD) | 28.2 (3.9) | 28.1 (5.0) | 0.11 |
| Waist-hip ratio (WHR) | Mean (SD) | 0.94 (0.06) | 0.83 (0.07) | <0.001 |
| Total cholesterol | mmol/l – mean (SD) | 5.61 (1.02) | 5.82 (1.04) | <0.001 |
| HDL | mmol/l – mean (SD) | 1.25 (0.34) | 1.52 (0.39) | <0.001 |
| Triglycerides | mmol/l – mean (SD) | 2.08 (1.39) | 1.70 (0.99) | <0.001 |
| SBP | mmHg – mean (SD) | 144.0 (18.5) | 134.4 (19.6) | <0.001 |
| DBP | mmHg – mean (SD) | 90.8 (10.5) | 86.9 (10.8) | <0.001 |

*p-value for gender difference; unpaired t-test used for continuous variables, chi-square test for categorical variables

**Table 2. The difference between those reporting depressive symptoms* and those not reporting depressive symptoms (coefficient and 95% CI) on CRP (mg/l) in different levels of adjustment (N=6126)**

| Level of adjustment | Difference (95% CI) | p-value |
|---|---|---|
| 1. Unadjusted | 0.60 (0.32-0.87) | <0.001 |
| 2. Age adjusted | 0.63 (0.35-0.90) | <0.001 |
| 3. Age + sex adjusted | 0.60 (0.32-0.87) | <0.001 |
| 4. (3) + education | 0.53 (0.25-0.81) | <0.001 |
| 5. (4) + material deprivation | 0.47 (0.18-0.75) | 0.001 |
| 6. (5) + BMI+WHR | 0.42 (0.14-0.70) | 0.003 |
| 7. (6) + smoking | 0.43 (0.15-0.71) | 0.002 |
| 8. (7) + alcohol consumption | 0.43 (0.15-0.71) | 0.002 |
| 9. (8) + physical activity | 0.43 (0.15-0.71) | 0.002 |
| 10. (9) + HDL + total cholesterol + triglycerides | 0.44 (0.16-0.72) | 0.002 |
| 11. (10) + SBP + DBP | 0.44 (0.16-0.72) | 0.002 |

* CESD score ≥ 16

**Table 3. Difference in CRP (mg/l) between persons with and without depressive symptoms in different subsets of subjects, fully adjusted*.**

| Subset | N | Depressive symptoms (%) | Mean CRP | B (95% CI) | p-value |
|---|---|---|---|---|---|
| Full sample | 6126 | 18.9 | 2.52 | 0.43 (0.16-0.72) | 0.002 |
| | | | | | |
| Medical history | | | | | |
| No history of MI, angina, stroke or cancer | 4946 | 17.3 | 2.41 | 0.44 (0.14-0.74) | 0.004 |
| History of MI, angina or stroke | 723 | 22.9 | 3.08 | 0.43 (-0.57, 1.43) | 0.40 |
| History of cancer | 364 | 28.1 | 2.63 | 0.83 (-0.18,1.85) | 0.11 |
| | | | | | |
| Long-term health problems for which medical treatment has been sought over last 12 months | | | | | |
| No | 2448 | 12.2 | 2.15 | 0.67 (0.25,1.10) | 0.002 |
| Yes | 3632 | 23.3 | 2.74 | 0.27 (-0.10,0.64) | 0.15 |
| | | | | | |
| Number of visits to a doctor in the last 12 months | | | | | |
| None | 1131 | 12.2 | 2.30 | 1.20 (0.52,1.87) | 0.001 |
| 1-3 | 2254 | 15.7 | 2.22 | 0.45 (0.03,0.87) | 0.03 |
| 4 and more | 2557 | 24.7 | 2.84 | 0.20 (-0.26,0.66) | 0.40 |

* = adjusted for age, sex, education, material deprivation, BMI, WHR, smoking, alcohol consumption, physical activity, HDL, total cholesterol, triglycerides, SBP and DBP

**Figure 1. Mean CRP levels and 95% CI by CESD score (age-sex adjusted)**