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Dear Editor:

Thank you for the thorough review of our paper now entitled “Stress-related disorders and risk of cardiovascular disease: a population-based sibling-controlled cohort study” and for the opportunity to revise and resubmit an improved version for publication in BMJ. We appreciate the insightful comments from the editors and reviewers; please find our point-by-point responses to these comments below. We have now performed some additional analyses and made revisions to the manuscript in accordance with the important issues raised by the editors and reviewers. We believe that the quality of the manuscript has, as a result, been significantly improved. While we are certainly willing to make further revisions on your request, we hope that you will find the present version of our manuscript suitable for publication in BMJ.

Yours sincerely,

Huan Song
On behalf of all co-authors

Center of Public Health Sciences
Faculty of Medicine
University of Iceland
Sturlugata 8, 101 Reykjavík,
Iceland

Responses to the comments of the editorial committee and referees*:

*All page and line marks correspond to the line numbers of the '**Revised Manuscript_clean**' version.

Comments by the committee:

- Please note that the Patient and Public Involvement (PPI) declaration is missing and there was no acknowledgment to patients for their data. Some of the limitations could have been mitigated by PPI and it is alarming that a study with psychiatric issues linked to morbidity and mortality would be without PPI

Authors' responses: We have now added a statement of 'Patient and Public Involvement' in the revised manuscript. This study leverages Swedish administrative health utilization data for a historical analysis addressing the primary hypothesis. Such register-based studies are, in accordance with Swedish law, conducted without an informed consent, or consultation with patients, after an ethical review by a Regional Ethics Committee (in this case in Stockholm). We regret that patients were not invited to comment on the study design or interpretation of the result during the research process, and therefore highly appreciate the comments from reviewer 1 providing us with unique insights into this point (we thank for his contribution in the 'Acknowledgement' part).

In the revised manuscript (Page 10, Line 216-223):

*'**Patient and Public Involvement***

As this study leverages nationwide register data, no patients were involved in setting the research question or the outcome measures, nor were they involved in developing plans for design or implementation of the study. A patient was invited to contribute to the review process of the study and we are grateful for his input on readability and accuracy of this document. There are no plans to directly disseminate the results of the research to study participants or the relevant patient community. The dissemination to the Swedish population (which constitutes the study population) will be achieved through a media outreach (e.g. press release and communication) upon publication of this study.'

- Our statistician made the following comments:

The analyses seem appropriate and I have only minor points to add to the reviewer comments:

1. Follow up time is skew and so should be summarised non-parametrically (table 1). The same may also be true for patient age.

Authors' responses: Requested changes have been made. We now provide median and interquartile range for age at index date and follow-up time.

2. Need to remove causal inference in what this study adds.

Authors' responses: Requested changes have been made. In the revised manuscript (Page 4, Line 68-76):

'*What this study adds*

After careful control of familial background (through a sibling design), multiple risk factors, and comorbidities, our results indicate that severe stress reactions, indicated by a clinical diagnosis of PTSD, acute stress reaction, or adjustment disorder and other stress reactions, are associated with considerably higher risk of multiple types of CVDs (in seven major categories and 16 individual diagnoses of CVDs), especially during the first year after the diagnosis of a stress-related disorder (i.e., 64% higher risk among patients with stress-related disorders compared to their full siblings). The association between stress-related disorders and CVD seems particularly strong in cases of early-onset CVDs (occurring before the age of 50)'

- One editor would like you to clarify what does it mean that all the individual elements of the composite outcome are raised considering they arise from different conditions? He also wondered if you could include "control conditions" as other outcomes, as well as mortality. For example, some common cancers (e.g. skin and GIT), chest infection, joint surgery, back pain, etc. The results for these would add whether they were raised across the board (stress as a universal risk factor for instance) or only raised for some conditions (stress affecting specific disease mechanisms).

Authors' responses: Thanks for your comment and good suggestions. We certainly agree that different subtypes of CVDs have various pathological processes, which may be the reason why different magnitude of HRs was observed for different CVDs. However, some common general pathways may have a role for all CVDs under the condition of a severe stress response. For instance, behavior changes, such as smoking which is a risk factor for all CVDs, are common after developing stress-related disorders. Also, increased inflammatory response has been demonstrated in the traumatized population, probably due to the alterations in the glucocorticoid (GC) receptor itself and reduced GC signaling (Cohen S, et al. PNAS 2012). Given that the inflammatory process has been linked to the development of multiple CVDs, e.g. atherosclerosis, plaque formation, hypertension, and/or ischemic diseases (Dinarello CA, et al. Nat Rev Drug Discov. 2012; Wong BW, et al. Can J Cardiol. 2012), it is possible that stress-related disorders have a general impact on CVD development. Therefore, although we agree that a composite category of CVDs might at first glance appear vague, the above rationale of a general behavioral- or inflammatory mechanism potentially mediating associations to all CVD supports our current approach/presentation.

In the revised manuscript, we highlighted these statements in the Discussion,' Meaning of the study' part (Page 15, Line 346-354):

'*Although the crucial underlying mechanisms remain unclear, numerous potential mechanisms have been proposed to explain the association between stress-related disorders, particularly PTSD, and CVD. Physiologic impact of an acute stressor can directly work on cardiovascular system (e.g. increased arterial blood pressure), which consequently forms favorable conditions for the onset of acute CVD events[40 41], as well as the further development of hypertension, endothelial dysfunction, and arteriosclerosis[42]. In addition, the long-lasting impact of severe stress reactions on CVD risk is also plausible through prolonged biological disturbances (e.g., inflammation, autonomic dysfunction, dysregulation of hypothalamic-pituitary-adrenal axis, and abnormal neurochemistry)[43-45] and behavior-related changes (e.g., smoking, sleeping disturbance)[32].*'

We appreciate the editor's idea of presenting 'control conditions' for identifying potential disease-specific pathophysiologic alteration after stress-related disorders. However, after careful consideration, we believe that we, and others, have previously reported varyingly strong associations between stress-related disorders and different disease phenotypes, including several null associations, which may be informative for this point:

- ✓ First, we recently reported associations between stress-related disorders with 18 out of 41 tested autoimmune diseases (Song et al., JAMA 2018). That is, stress-related disorders were modestly associated with several autoimmune diseases (i.e., rheumatoid arthritis, sarcoidosis, and ulcerative colitis with HR ranging from 1.08 and 1.17, generally weaker than reported now for

CVD) whereas more than half of the tested autoimmune disease were not significantly associated with past exposure to stress-related disorders.

- ✓ Second, a Danish group previously reported null associations between posttraumatic stress disorder and most cancers (Gradus JL et al. Eur J Epidemiol. 2015) while approximately doubled rate of overall mortality after all stress-related disorders (Gradus JL et al. Am J Epidemiol 2015).
- ✓ Third, although of great interest, some of the conditions suggested by the editor have established associations with severe stress (e.g. pain) (Vachon-Presseau E et al. *The stress model of chronic pain: evidence from basal cortisol and hippocampal structure and function in humans.* Brain 2015) or represent an indication of an immune-system impairment (infections in respiratory tract, Cohen S, et al. *Psychological stress and susceptibility to the common cold*. N Engl J Med. 1991) that has previously been associated with severe stress.
- ✓ Forth, our dataset does not have complete information for surgery (joint surgery), so the inclusion of joint surgery as a control condition is not feasible.

Taken together, we believe that the abovementioned data suggest that stress-related disorders are not associated with all disease outcomes (including many autoimmune diseases and cancers) pointing against a general pathophysiologic mechanism for all diseases or, alternatively, general surveillance bias. On the other hand, we believe that our current study convincingly demonstrates a particularly strong association with CVD and that this paper should be focused on precisely these associations, potentially mediated through altered immune function, chronic/systemic inflammation, and trauma-related behavior changes.

We have now added these points to in the Introduction section (Page 5, Line 84-87):
Accumulating evidence suggests that such adversities might lead to an increased risk of several major diseases (including cardiovascular morbidity[3], injury[4], infection[5], certain autoimmune diseases[6], but not cancer[7,8]) and mortality[9,10], with the largest risk elevations usually noted among individuals who develop psychiatric disorders as a result of their trauma[11-13].
With this said, we are certainly willing to perform additional analyses if the editor does not agree with our rationale.

- Another editor wondered how come the 'Family history of cardiovascular disease' is different for exposed and their siblings?

Authors' responses: Thank you for your comment. The 'family history of cardiovascular disease' is the same for exposed patients and their siblings. The small difference shown in Table 1 is due to the different number of siblings for each exposed patient (i.e., some exposed patients have only

one, while others have several siblings without stress-related disorders). Within each stratum (by family identifier), the family history of cardiovascular disease is always the same.

In the revised manuscript, we added one footnote in Table 1 (Page 23):

£ The marginal difference between exposed and unexposed sibling group is due to the different number of siblings for exposed patients. Within each family, the family history of cardiovascular disease is always the same.

- Another editor suggested that you make it clearer that this looks at the more severely affected people with stress as this is a cohort who were referred to psychiatric services.

Authors' responses: Thank you for the important comment. To have a clearer description about our target population, in the revised manuscript, we modified the Summary box and Discussion, pointing out that we targeted on patients with a clinically diagnosed stress-related disorder, and hence the findings here may not be directly applicable to individuals with less severe reactions or daily stress.

Summary box:

(Page 4, Line 68-74):

'*What this study adds*

After careful control of familial background (through a sibling design), multiple risk factors, and comorbidities, our results indicate that severe stress reactions, indicated by a clinical diagnosis of PTSD, acute stress reaction, or adjustment disorder and other stress reactions, are associated with considerably higher risk of multiple types of CVDs (in seven major categories and 16 individual diagnoses of CVDs), especially during the first year after the diagnosis of a stress-related disorder (i.e., 64% higher risk among patients with stress-related disorders compared to their full siblings).'

In the 'Strengths and weaknesses of this study' part of Discussion, we added

(Page 13, Line 301-304):

'Limitations include, first, the absence of information from primary care as well as late inclusion of outpatient specialist care records in the NPR, which might have led to an underestimation of the number of patients with milder forms of stress-related disorders or less severe CVD events.'

(Page 14, Line 319-321):

'Sixth, since this study focus on patients who received a clinical diagnosis of stress-related disorders through hospital visit, its findings might not be directly applicable to individuals with less severe stress reaction or daily stress'

In the 'Conclusions' part of Discussion, we added

(Page 16, Line 379-381):

'This population-based sibling-controlled analysis showed a clear association between clinically confirmed stress-related disorders and a higher subsequent risk of CVD, particularly during the months following a diagnosis of stress-related disorder, in the Swedish population.'

In your response please provide, point by point, your replies to the comments made by the reviewers and the editors, explaining how you have dealt with them in the paper.

Comments from Reviewers

Reviewer: 1

Recommendation:

Comments:
As a previous sufferer of stress-related illness I found this paper interesting and reassuring. My illness was some years ago, I have not developed CVD, and this paper suggests my past stress is unlikely to give me an increased risk level. I received very little in the way of care or treatment for my stress illness, other than removing me from the cause and occasional blood pressure checks. I would hope that anyone involved in treating or caring for a victim of severe stress would be alerted to the increased risk of CVD and be vigilant in monitoring the signs. I think this paper would be of interest to a large number of people.
I found the article comprehensive and informative and I have been unable to identify any areas where I wished for more information.
Patients with stress disorders might find it difficult to distinguish between imagined and real symptoms, so increased vigilance from healthcare professionals could be required. For example, I often had feelings of palpitations or felt my heart rate was raised. I also imagined my blood pressure must have been raised. However, none of these were observed on the occasions I had health checks, leading me to doubt if I would have been able to differentiate these from real physical symptoms if I had experienced any.
The outcomes described in the paper are relevant to patients and clinicians and could form the basis of discussions between them to highlight the increased risk of CVD. This would have to be done sympathetically in order to avoid raising the patients stress levels further.
There is no evidence of patient involvement in this study, which has been produced from large volumes of patient data. Perhaps the authors could have recruited some patients and clinicians to discuss the findings and assess their impact on stressed patients and how clinicians were able to reassure them or put a monitoring plan in place.

Authors' responses: Our sincere thanks for sharing your experience and thoughts on this paper. Your comments are extremely important to us since no other patients were invited to comment on

the study design nor implications of these findings during the research process. However, we did work closely with clinicians for data analyses and interpretation of results. We recognize how difficult it must be for patients with stress-related disorders to disentangle their psycho-physiologic symptoms from symptoms of CVD, and the importance of gently conveying the message of such risks to patients in order not to further add to their stress levels. Importantly, we share your opinion and recommendation that patients with stress-related disorders need to be monitored closely for any signs of symptoms of emerging CVDs.

We have now put more emphasis on this implication of effective monitoring plan in the revised version of our paper.

'Discussion' section, under the 'Meaning of the study' (Page 16, Line 375-377):

'*These sudden CVD events carry a high risk of a fatal outcome[44], thus increased clinical awareness of these risks among patients with recently diagnosed stress-related disorders deserves further attention*'

'Discussion' section, under the 'Conclusions' (Page 16, Line 379-386):

'This population-based sibling-controlled analysis showed a clear association between clinically confirmed stress-related disorders and a higher subsequent risk of CVD, particularly during the months following a stress-related disorder diagnosis, in the Swedish population. This association applies equally to both men and women and is independent of familial factors, history of somatic/psychiatric diseases, and psychiatric comorbidities. These findings call for enhanced clinical awareness and, if verified, monitoring or early intervention among patients with recently diagnosed stress-related disorders.'

Reviewer: 2

Recommendation:

Comments:

This study is a well-conducted large register-based cohort study using two different approaches in relation to selecting the comparison groups, as both a cohort of siblings and a cohort of birth-year- and sex-matched controls were examined in relation to the exposed group with stress. The study is well argued, designed, analysed and written.

The exposure was based on hospital discharge diagnosis within posttraumatic stress disorders, acute stress reaction or adjustment disorder and the cohorts were followed for incident cases of cardiovascular diseases, likewise based on hospital discharge or death certificates. Adjustment and analyses of interactions were conducted based on register-based information.

The study shows a higher hazard rate ratio for CVD among those exposed to stress-related

disorders, higher within the first 6-12 months and with stronger associations with CVD before the age of 50.

Major comments:

Both in the summary box, the introduction and in the supplementary files, the focus is on posttraumatic stress disorder, which is clinically different from acute stress and adjustment disorders. This is to some extent explained, as PTSD has been subject to most research, but it would, in my opinion, be relevant to discuss this further or to change the focus of the text and handle all three types of stress equally. This is also relating to the introduction, stating that most individuals will be exposed to psychological trauma or life events, and clinically this may not necessarily be in the form of stress examined in the study. A more thorough discussion on the type of stress (leading to hospitalization) in comparison to ”stressful life events” could be included.

Authors' responses: Thank you for the encouraging remarks on our study and this important comment. In the revised manuscript, we have now added descriptions about the other two types of stress-related disorders.

Summary box (Page 4, Line 69-74):

'*After careful control of familial background (through sibling design), multiple risk factors, and comorbidities, our results indicate that severe stress-related reactions, indicated by a clinical diagnosis of PTSD, acute stress reaction, or adjustment disorder and other stress reactions, are associated with considerably higher risk of multiple types of CVDs (in seven major categories and 16 individual diagnoses of CVDs), especially during the first year after the diagnosis of a stress-related disorder (i.e., 64% higher risk among patients with stress-related disorders compared to their full siblings).*'

'Introduction' section (Page 5, Line 88-96), we now include a description of the type of traumatic events that relevant to different diagnoses of stress-related disorder:

'*Stress-related disorders are a group of psychiatric disorders where one of the diagnostic criteria is the presence of a preceding stressful life event. Depending on the type of stressor, the reported symptoms, and their duration, such disorders are mainly categorized as acute stress reaction, post-traumatic stress disorder (PTSD), and adjustment disorder[14]. The presence of a life-threatening traumatic event is a prerequisite for the former two disorders while adjustment disorder generally refers to physical or psychological distress ('adjustment syndromes'[15]) triggered by an identifiable life change. PTSD is the most severe and widely studied form of stress-related disorders, characterized by re-experiencing, avoidance, negative cognitions and mood, and hyperarousal following the traumatic event[16]'*

Also, we modified the statements in the Discussion (Page 15, Line 348-360):

'*Although the crucial underlying mechanisms remain unclear, numerous potential mechanisms have been proposed to explain the association between stress-related disorders, particularly PTSD, and CVD. Physiologic impact of an acute stressor can directly affect the cardiovascular system (e.g. increased arterial blood pressure), which consequently forms favorable conditions for the onset of acute CVD events[40 41], as well*

as the further development of hypertension, endothelial dysfunction, and arteriosclerosis[42]. In addition, the long-lasting impact of severe stress reaction on CVD risk is also plausible through prolonged biological disturbances (e.g., inflammation, autonomic dysfunction, dysregulation of hypothalamic-pituitary-adrenal axis, and abnormal neurochemistry)[43-45] and behavior-related changes (e.g., smoking, sleeping disturbance)[32]. In particular, as our analyses revealed that patients with stress-related disorders other than PTSD were also at a considerably excess risk of adverse cardiovascular outcomes, further studies with a broader research scope are warranted to explore the underlying mechanisms between severe stress reactions and the onset of CVD.'

As the clinical entities have changed over the years it is somewhat surprising that stratification on calendar time yields the same results. This could be discussed further, as it may imply potential bias in the study. ?

Authors' responses: We are aware of some changes of diagnostic criteria, primarily for adjustment disorder, over the study period. Yet, it is not clear that such historical trends are differential with respect to the studied outcome, i.e., CVD. Hence, although the incidence of diagnosed stress-related disorders changes over time, it does not necessarily have to result in varying associations with CVDs over time. Nevertheless, it is indeed possible that misclassification between diagnostic categories results in lower observed differences in associations with CVD across categories of stress-related disorders. In the present analyses, we observed similar results when stratifying by calendar period. In our view, this suggests a minor impact of such changes of diagnostic criteria of stress-related disorders on the studied associations. In the 'Strengths and weaknesses of this study' part of the Discussion, we have added (Page 13, Line 304-307) the following:

'*Second, changes in the diagnostic criteria of stress-related disorders over the 27 year study period may have influenced the observed associations, although our stratified analyses across different calendar periods suggest a minor impact of such diagnostic instability on the reported association.*'

As is rightly discussed, most patients suffering from stress are not hospitalized and will thus not be entering the study, but is treated in general practice. The conditions leading to hospitalization could be the more grave ones, and this could be discussed further, especially as the implications of the study would lead to suggestions to focus on the prevention of CVD. This may be only the more severe ones.

Authors' responses: Thanks for the important comment. We agree that this study mainly captured individuals with severe stress responses (severe enough to make a hospital visit). However, since outpatient specialist care records are also added to the National Patient Register since 2001, the identified exposed patients here were not necessarily hospitalized. To have a clearer description about our target population, in the revised manuscript, we modified in the Summary box and the Discussion, pointing out that we targeted on patients with ***clinically confirmed stress-related***

disorders, and hence the findings here may not be directly applicable to individuals with less severe reactions or daily stress.

Summary box:

(Page 4, Line 68-74):

'*What this study adds*
After careful control of familial background (through a sibling design), multiple risk factors, and comorbidities, our results indicate that severe stress reactions, indicated by a clinical diagnosis of PTSD, acute stress reaction, or adjustment disorder and other stress reactions, are associated with considerably higher risk of multiple types of CVDs (in seven major categories and 16 individual diagnoses of CVDs), especially during the first year after the diagnosis of a stress-related disorder (i.e., 64% higher risk among patients with stress-related disorders compared to their full siblings).'

In the 'Strengths and weaknesses of this study' part of Discussion, we added

(Page 13, Line 301-304):

'Limitations include, first, the absence of information from primary care as well as late inclusion of outpatient specialist care records in the NPR, which might have led to an underestimation of the number of patients with milder forms of stress-related disorders or less severe CVD events.'

(Page 14, Line 319-321):

'Sixth, since this study focus on patients who received a clinical diagnosis of stress-related disorders through hospital visit, its findings might not be directly applicable to individuals with less severe stress reaction or daily stress'

In the 'Conclusions' part of Discussion, we added

(Page 16, Line 379-381):

'This population-based sibling-controlled analysis showed a clear association between clinically confirmed stress-related disorders and a higher subsequent risk of CVD, particularly during the months following a diagnosis of stress-related disorder, in the Swedish population.'

The study finds a long-term (up to 25 years) elevated risk of CVD with an HR of 1.2-1.4. This is somewhat surprising, I think, as stress should wear off if the biological cause is related to an increased workload for the heart (p. 14), and the consistent risk could be the result of residual confounding or unaccounted bias; this could also be discussed in more details.

Authors' responses: We agree with the reviewer that the most relevant risk period for CVD is the first year after a stress-related disorder diagnosis, which might attribute to direct physiologic impact of stress on the cardiovascular system. We further agree that the risk of unmeasured or residual confounding influencing our results cannot be fully excluded– although, in our opinion, we have made every possible effort to exclude such risks (e.g. by using a sibling design and carefully adjusting for a range of relevant covariates). Alternatively, the long-term impact of stress-related disorders on CVD risk is indeed plausible due to prolonged dysregulation of inflammatory responses

or lasting behavior changes (e.g. increased smoking) after a stress-related disorder. We have now highlighted this in the revised version of our paper:

In the Discussion (Page 15, Line 346-354):

'*Although the crucial underlying mechanisms remain unclear, numerous potential mechanisms have been proposed to explain the association between stress-related disorders, particularly PTSD, and CVD. Physiologic impact of an acute stressor can directly work on cardiovascular system (e.g. increased arterial blood pressure), which consequently forms favorable conditions for the onset of acute CVD events[40 41], as well as the further development of hypertension, endothelial dysfunction, and arteriosclerosis[42]. In addition, the long-lasting impact of severe stress reactions on CVD risk is also plausible through prolonged biological disturbances (e.g., inflammation, autonomic dysfunction, dysregulation of hypothalamic-pituitary-adrenal axis, and abnormal neurochemistry)[43-45] and behavior-related changes (e.g., smoking, sleeping disturbance)[32].*'

Minor comments:

The abstract (p. 3-4) gives results from the sibling-controlled study as is also implicated on p. 4, l. 5. The abstract might be reformulated in order to balance the results

Authors' responses: Thanks for the comment. We now have added more information about the results of population-based analyses in the abstract (Page 2, Line 51-53):

'*Analyses within the population-matched cohort yielded similar results (HR=1.71 [95% CI 1.59 to 1.83] for any CVD during the first year of follow-up, and 1.36 [95% CI 1.33 to 1.39] for thereafter)*.'

The term "16 individual CVDs" is used in the summary box (p. 5) and elsewhere in the manuscript. It covers diagnoses and thus not necessarily different conditions, and I would suggest rephrasing the term.

Authors' responses: Thanks for the comment. We agree with the reviewer. To further clarify this, in the revised manuscript, we call it '16 individual **diagnoses** of CVDs'.

table 1: the total number of participants (p. 22, l. 11) in both columns below population-matched cohort seems to be ten times too high.

Authors' responses: We are sorry for this typo. The numbers have been corrected in the revised manuscript.

Several entries should have the "n (%)" explanation given, eventually just in the heading as a general description and then "…unless otherwise stated".

Authors' responses: Requested changes have been made.

Supplementary files, figure on p. 39. In the legend on p. 40, no explanation of the red line at HR=1 is given. Is the red line necessary?

Authors' responses: Thank you for this comment. We agree and have removed the red line. Please refer to the new Supplementary Figure 1.

Reviewer: 3

Comments:
The current manuscript includes details of a population-based cohort study in Sweden exploring the link between PTSD and other similar psychiatric disorders and the development of CVD. Using novel sibling-controlled and population-matched designs the authors were able to provide information on a large number of participants (ca. 130,000 patients with the disorder, 170,000 full siblings, and 1.4 million unexposed members of the general population). The authors report finding that there was a consistent, increased association between having a disorder and a future CVD event, which was generally robust to all sensitivity analyses. Furthermore, there was a signal suggesting that this association was stronger in the 1st year post diagnosis compared to after 1 year post diagnosis.

Overall, this is a well-structured study which extends the current literature exploring PTSD (mostly) and CVD. The study design and samples size are considerable strengths. That being said there are several areas where some further reflection by the authors may enhance the manuscript.

It would seem that the authors are trying to explore the relationship between PTSD and related disorders and CVD. However, they are using the term psychiatric reactions to severe stress, which is rather nebulous. For example, one could argue that certain forms of depressive disorders may appear in response to severe stress, which would also fall under the realm of 'psychiatric reactions to severe stress' (of note, there is an abundance of literature linking depression to CVD). As such, I would suggest that the authors rework their title and be consistent in their wording throughout the manuscript (may be PTSD and similar stress-related disorders?).

Authors' responses: Thanks for the nice comments on our study and for this important comment. We agree and have now reconsidered the title according to the reviewer's comment. Now the title

has been changed to '*Stress-related disorders and risk of cardiovascular disease: a population-based sibling-controlled cohort study'*.

Non-effected siblings were matched at the point of diagnosis for the effected sibling. If the non-effected sibling demonstrated a PTSD or similar diagnosis during follow-up how was this handled analytically? Was some kind of censoring included? This may have a notable effect on the course of associations. Of note, due to the 1:10 matching for the general population I don't think this is as much of an issue in those analyses, where any effects are likely to be washed out by the magnitude of the general population.

Authors' responses: Thanks for highlighting this issue. As mentioned in the 'Method' (Page 7, Line 140-142: '*The follow-up time for the unaffected full siblings or matched unexposed individuals was additionally censored at the time of their first diagnosis of stress-related disorder, if any, during the follow-up.*'), the unaffected siblings/matched unexposed individuals that got a stress-related disorder diagnosis during follow-up were censored at the time of their diagnosis. We agree with the reviewer that it is possible that delayed diagnosis of a stress-related disorder in the not yet diagnosed sibling controls (for instance cases of same index event causing disorders in both siblings), might result in somewhat conservative point estimates, since these 'unaffected' siblings contributed person-time to the unexposed group when in fact exposed. In the revised manuscript, we comment on this:

In the 'Strengths and weaknesses of this study' part of Discussion (Page 14, Line 307-309): *'Third, as traumatic life experiences may be shared within families it is possible that a proportion of the reference-sibling population also suffered from milder or undiagnosed stress-related disorders yielding conservative estimates in the sibling analysis.'*

Table 1 only reports psychiatric comorbidity (i.e., the development of a non-PTSD or related disorder) in the effected cohort only. I am assuming that the siblings and the general population both developed these disorders as well. It would be good to report these in the table. I am also assuming that if the other 2 cohorts had one of these disorders then this was included in the analyses, though I am now not 100% sure that this is correct. As a side-bar this should probably be 'other psychiatric disorders' rather than comorbidity to cover all 3 populations.

Authors' responses: Thank you for this comment. In Table 1 and the analyses for psychiatric comorbidity shown in Supplementary Figure 2, the 'psychiatric comorbidity' always refers to the concurring incidence of other psychiatric disorders, diagnosed 3 months before to 1 year after the stress-related disorder, among the exposed individual. We assume other psychiatric disorders

occurring around the time of the stress-related disorder diagnosis may be relevant to the severity of the stress-related disorder. Of course, individuals in the unexposed cohorts might also get diagnosis of other psychiatric disorder during this pre-defined period. But since the percentage is very small (only 0.71% among unaffected siblings), further adjustment for the 'other psychiatric disorder' made no difference for the estimates. Nevertheless, in table 1 we do report history of other psychiatric disorders (occurring before the index date) and control for this factor in our analyses.

In the second paragraph on page 14 the authors speculate that the association between a stressful event (and not disorder) and CVD events (which has been shown in a number of previous studies) might be mediated by severe emotional reactions. However, the data they have make it hard to make this assertion. To be able to study this they would have needed to have 2 groups of individuals who experienced stressful events and then compared those who developed a PTSD or similar disorder vs. those that didn't. I understand where the authors are going with this, but there is some tempering of this assumption which is needed in this paragraph.

Authors' responses: Thanks for the helpful comment. We agree and have revised this sentence. In the revised manuscript ('Discussion', Page 15, Line 342-346)

'Clinical observations suggest that experiencing severe emotional or physical stress is linked to increased workload for the heart, which consequently may trigger immediate cardiovascular consequences, such as heart attack, sudden cardiac arrest, and heart failure, even in apparently healthy individuals. Our results consolidate the reported association by showing that a clinically confirmed stress-related disorder (requiring a preceding occurrence of a trauma or significant life stressor) is associated with these acute CVD events.'

At the end of the 2nd paragraph on page 15 the authors argue that because their analyses show similar patterns with acute CVD events as well as non-acute events that this 'diminishes the possible impact of surveillance bias or reverse causality.' I'm not sure why this would be the case, especially for surveillance bias. Furthermore, the fact that heart failure had one of the largest HRs within the first year would strongly suggest reverse causation (HF is unlikely to be caused in a 6 month window) but this is not offset by the findings from the acute events (NB. the HRs for the acute events of MI, hemorrhagic stroke, and Takotsubo cardiomyopathy were all lower than would be expected compared other events). I think most people will appreciate the limitation, without diminishing the importance of the findings, if that sentence was removed.

Authors' responses: We understand that this sentence may easily be misunderstood. We only meant to convey that we believed that the results of these additional analyses (on acute events) pointed against the notion that the increased HR were solely due to surveillance bias and/or reverse causality, since acute and severe events reasonably always present in the health care system and

thus these diagnoses might be less affected by these factors. To avoid misunderstanding, we have edited this sentence in the revised manuscript ('Discussion', Page 16, Line 372-375):
'*However, the additional analyses on acute and severe CVD events (e.g., acute coronary events) that typically resulting in prompt hospital visit and medical care provide further evidence supporting an immediate impact of stress-related disorder on CVD, since such analyses are less likely to be affected by surveillance bias or reverse causality.*'

Minor comments.

The authors use the term gender to describe their population. However, gender is a complex psychosocial construct made up of a number different facets. If this is truly what the authors studied then they should provide details of their conceptualisation of gender and the measures they used to assess it. Alternatively, if they used sex, which is an anatomical/physiological construct usually represented as a binary man/woman variable, then they should adjust their terminology within the manuscript.

Authors' responses: Thank you for the comment. We have changed 'gender' to 'sex'.

There are some inconsistencies in the tables with the use of decimal places which should be corrected.

Authors' responses: Thank you. We have corrected the typo in Table 1, and closely checked all numbers throughout the manuscript.

Figure 2, the forest plots, should be on the same scale so has to help comparisons.

Authors' responses: Thank you. We have now re-plotted this figure using the same scale for both <1 and ≥1 year period.

Reviewer: 4

Recommendation:

Comments:
Song et al. have produced an excellent manuscript regarding stress disorders and risk of CVD. Although the overall finding of increased risk is not particularly surprising, the novelty here is the use

of a sibling-matched analysis to address potential confounding. The manuscript is well-written and analyses conducted thoroughly. Some potential areas for improvement are discussed below.

Introduction

A fundamental argument of this manuscript is that stress increases CVD risk and that persons who have developed a stress disorder are at greater risk. However, this point is not properly supported by the authors. Reference 5 from the Nurses Health Study is used to support this point, but in that study, trauma with no PTSD symptoms vs. trauma with 4+ PTSD symptoms were generally equivalent in increased risk of incident CVD across various model adjustments. That study would support that trauma, regardless of psychiatric disorders or not, is the important determinant of CVD risk. The authors should find evidence that better supports their point (while acknowledging incompatible evidence) …or consider reframing their argument. It should also be noted that the Nurses Health Study participants are all women, and stress disorders and CVD both exhibit sex differences that may make this reference even more unhelpful to the present study.

Authors' responses: Thank you for the encouraging remarks on our study and the excellent comment. We acknowledge that in our study, only individuals experienced trauma/stressful events that eventually developed a stress-related disorder were included in the analyses. We assume that, in our comparison population there are also individuals who have been exposed to stressful events with or without psychiatric disorders that have not been diagnosed. This misclassification inevitably results in attenuation of our estimates. However, we agree that disentangling the influence of the event itself from resulting psychiatric sequel is complex and the findings from Summer et al. (the original reference 5) in the Nurses Health Study do not provide a clear answer to that question. We have therefore replaced this reference with a study focusing on 9-11 survivors (Jordan HT, Stellman SD, Morabia A, et al. Cardiovascular Disease Hospitalizations in Relation to Exposure to the September 11, 2001 World Trade Center Disaster and Posttraumatic Stress Disorder. J Am Heart Assoc 2013;2(5)), where they suggested an association between the presence of PTSD and elevated risk of heart disease hospitalization.

Methods:

Sibling cohort – this section is presented in a confusing manner. The first paragraph seems to be more relevant to the overall study rather than the specific sibling cohort – a simple fix would be to move the header to the actual paragraph describing the sibling cohort. It also would be helpful to the reader to introduce the rationale for a sibling cohort design here.

Authors' responses: Thanks for the comment. We have moved the header 'Sibling cohort' to the next paragraph where the construction of the sibling cohort was described. We have further added a rationale (including a reference, D'Onofrio BM et al. Critical need for family-based, quasi-experimental designs in integrating genetic and social science research. Am J Public Health. 2013) for why a sibling comparison is needed (Page 6, Line 125-128):

'To control for the familial components[22], we identified 106,180 clusters of full siblings discordant for stress-related disorders, with a total of 171,314 unaffected full siblings who were alive and free of stress-related disorders and CVD at the diagnosis date of the affected sibling, through the Multi-Generation Register.'

The sample selection may be problematic because of potential immortal time bias. The age at index date is 36 +/- 14 years SD, which suggests that there are very young participants included who are very unlikely to be diagnosed with stress disorders (due to lower detection in pediatric populations) and CVD even with long follow-up (due to lower inherent risk). Implementing a minimum age eligibility requirement as well as an assessment of the distributions of the ages of diagnosis of the stress disorders would help mitigate this potential bias.

Authors' responses: Thank you for this important comment. In our previous study (Song et al. JAMA 2018) we found that exposure to stress-related disorders at a younger age was more strongly associated with subsequent risk of autoimmune diseases, compared to exposure during later life. Thus we believe it's of great interest to study CVD risk among individuals exposed to stress-related disorders at young ages. Therefore, we provide a stratified analysis by tertiles of the age at exposure to a stress-related disorder. However, as the reviewer points out, due to the relatively limited follow-up time, we are only able to assess the risk of early-onset CVD in this relatively young population. Nevertheless, the same applies for the population controls and siblings of the exposed – as their age at start of follow-up is matched to (or adjusted for in the sibling analysis) to that of the young population exposed to a stress-related disorder. Hence, our position is that our study design is not subjected to immortal time bias (time from study start until exposure) although the baseline incidence of CVD is indeed quite low in the younger population (Suissa S, Immortal time bias in pharmaco-epidemiology. Am J Epidemiol. 2008).

Nevertheless, we agree with the reviewer that, to assure the diagnosis accuracy, we should have a minimum age requirement for the diagnosis of stress-related disorders, and have now reran all analyses **excluding exposed patients with diagnosis age younger than 5 years** (NICE Clinical Guidelines, Post-Traumatic Stress Disorder: The Management of PTSD in Adults and Children in Primary and Secondary Care. National Collaborating Centre for Mental Health (UK). Leicester (UK):

Gaskell; 2005). ***Based on the new analyses, the number of participants and risk estimates have been updated (and closely checked) in all figures, tables, and relevant text***.

From a causal inference perspective, the sibling matched analysis still has some problems since shared genetic or environmental confounding is still a possibility. It may make sense to use a case-crossover analysis to better control for this, so that each person is his/her own control. Having this 3rd matching analysis triangulate with the sibling and population-matched analyses would be powerful support of the authors' results.

Authors' responses: Thank you for this excellent suggestion. We indeed considered initially to include a case-crossover analysis but then decided not to since the pattern in CVD incidence after exposure to a stress-related disorder is **not completely transient**, which is a formal requirement for the case-crossover design (Turner J.R. Case-Crossover Studies. In: Gellman M.D., Turner J.R. (eds) Encyclopedia of Behavioral Medicine. Springer, New York, NY 2013). Nevertheless, below we show the results from the case-crossover design, which indeed support our original findings and suggest limited, if any, influence of unmeasured confounding.

As mentioned above, our data do not fit perfectly for a case-crossover design as the CVD risk never returns to normal after a diagnosis of stress-related disorder. Therefore, the results from the case-crossover analysis (OR 1.48, see below) is not directly comparable with the results from the cohort analysis (HR 1.64).

We now describe the analysis and its results here:

Method: *We performed a nested, self-matched case-crossover analysis among all individuals with a CVD event subsequent to a diagnosis of stress-related disorder, to allay the concern of unmeasured confounders (e.g., smoking). Conditional logistic regression was used to estimate the odds ratios (ORs) of having a diagnosis of any stress-related disorder in the case period (i.e., 1 year prior to the CVD, as 'at risk' period), compared to the control periods (i.e., 2-4 years prior to the CVD, as periods providing an estimate of the expected frequency of exposure for each case).*

Results: *In the case-crossover analysis where we compared the incidence of stress-related disorders 1 year ('case' period) to the 2-4 years window ('control' periods) prior to CVD event, the OR for any stress-related disorder was 1.46 (95% CI 1.38-1.53), suggesting some but minor impact of unmeasured confounding.*

Due to the abovementioned reasons, we believe this analysis should not be included in the manuscript, but we are open to reconsider our position if the editor or reviewer feel strongly it should.

The authors mention the possibility of surveillance bias in that patients with stress disorders have more healthcare visits and greater likelihood of receiving CVD diagnoses. Sensitivity analyses would be useful to defend against this, e.g.: adjusting/stratifying on number of health care contacts or days in hospital.

Authors' responses: Thank you for this important suggestion. We agree with the reviewer that surveillance bias may influence our estimation of the association between stress-related disorders and less severe, lingering outcomes where timing of onset is less clear (e.g. hypertension). However, this should not be the case with more severe, potentially fatal, outcomes, such as most of the CVD outcomes in this study (e.g. MI, stroke, embolism, etc.) that usually will end up in the health care system, irrespective of potential differential surveillance of the exposed population. Indeed, the findings from our additional analyses focusing on severe acute CVD outcomes, relieve our concern that surveillance bias explains, other than to a very limited extent, our findings.

In the revised manuscript, we wrote:

('Discussion', Page 16, Line 372-375):

'*However, the additional analyses on acute and severe CVD events (e.g., acute coronary events) that typically resulting in prompt hospital visit and medical care provide further evidence supporting an immediate impact of stress-related disorder on CVD, since such analyses are less likely to be affected by surveillance bias or reverse causality.*'

Nevertheless, in order to assess the possibility of considerable influence of surveillance bias on our assessment after stress-related disorders, we performed an additional analysis on risk of ***all CVD,*** *and* ***hypertension*** *specifically,* ***beyond one year*** after stress-related disorders by further adjusting for the number of hospital visits due to any existing psychiatric disorder or severe somatic diseases (i.e.,a group of severe somatic diseases as we defined in the method part) ***during the first year of follow-up***.

Our results show, the HRs slightly decreased but remained largely the same after such additional adjustment.

	HRs for ≥1 year, from main analysis	HRs for ≥1 year, additionally adjusted for the number of hospital visit due to *any existing psychiatric and severe somatic diseases* during the first year of follow-up
All CVD		
All stress-related disorder	1.29 (1.24-1.34)	1.25 (1.20-1.30)

PTSD	1.44 (1.23-1.68)	1.39 (1.19-1.64)
Acute stress reactions	1.31 (1.24-1.38)	1.28 (1.21-1.35)
Adjustment disorder and other stress reactions	1.23 (1.17-1.31)	1.19 (1.12-1.26)
Hypertension		
All stress-related disorder	1.16 (1.08-1.26)	1.13 (1.04-1.23)
PTSD	1.58 (1.14-2.18)	1.48 (1.06-2.09)
Acute stress reactions	1.15 (1.03-1.29)	1.14 (1.01-1.28)
Adjustment disorder and other stress reactions	1.13 (1.00-1.27)	1.08 (0.96-1.23)

However, it has to be considered that the number of hospital visits are also an indicator of the severity of the exposure, i.e. the stress-related disorder. Therefore, this slight attenuation of point estimates might not be an indication of enhanced medical surveillance but control for an indication of the severity of the exposure. Also, since such sensitivity analysis can only be done for HRs ≥1 year, **but not HRs <1 year (a focus of our paper)**, we feel it provides only limited information on the impact of surveillance bias. We therefore did not add this result to the revised manuscript but we are certainly willing to reconsider our position on the editor's request.

Were pain disorders considered as covariates? Pain and pain control are very relevant to stress-disorders.

Authors' responses: We appreciate the idea of using 'pain disorder' or 'pain control' as a covariate, although at the same time, we fear that by adjusting for such comorbidities, we might be controlling for factors in the causal mechanistic chain between stress-related disorders and CVD (e.g. chronic inflammation). Moreover, the diagnostic criteria for these conditions have been modified in each of the recent editions of the DSM and clinical applicability is highly debated (Mark DS, et al. International Review of Psychiatry. 2009). Also, as many types of pain control medications can be purchased over the counter, using the Drug Prescription Registers (2006-) would only capture a small proportion of subjects with the pain-related problems.

Table 1: Numbers in the columns do not sum to the listed number of exposed "patients". Also: some standardization of terminology would be helpful, since unexposed and unaffected are seemingly used interchangeably, as well as patients and individuals. Could just use exposed vs. unexposed.

Authors' responses: Thank you for these comments. We have corrected the typos in Table 1, and closely checked all numbers throughout the manuscript.

Regarding terminology, with the aim to keep consistency with the full text and to distinguish the sibling ‘control’ from the population ‘control’, we believe it’s best to always name them differently ‘unaffected siblings’ and ‘matched unexposed individuals’. Similarly, the use of ‘patients’ for ‘exposed’ and ‘individuals’ for ‘unexposed’, is for emphasizing the difference in the exposure status. We are certainly willing to reconsider the terminology if the reviewer or editors have further suggestions for improved clarity.