Conflicts Hurt: Social Stress Predicts Elevated Pain and Sadness Following Mild Inflammatory Increases

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Abstract

Individuals respond differently to inflammation. Pain, sadness, and fatigue are common correlates of inflammation among breast cancer survivors. Stress may predict response intensity. The current study tested whether breast cancer survivors with greater exposure to acute or chronic social or non-social stress had larger increases in pain, sadness, and fatigue during an acute inflammatory response. In total, 156 postmenopausal breast cancer survivors (ages 36-78, stage I-IIIA, 1-9 years post-treatment) were randomized to either a typhoid vaccine/saline placebo or the placebo/vaccine sequence, which they received at two separate visits at least one month apart. Survivors had their blood drawn every 90 minutes for the next 8 hours post-injection to assess levels of interleukin-6 (IL-6) and interleukin-1 receptor antagonist (IL-1Ra). Shortly after each blood draw, they rated their current levels of pain, sadness, and fatigue. Women also completed the Test of Negative Social Exchange to assess chronic social stress and the Trier Inventory of Chronic Stressors screen to index chronic general stress. At each visit, a trained experimenter administered the Daily Inventory of Stressful Events to assess social and non-social stress exposure within the past 24 hours. After statistical adjustment for relevant demographic and behavioral covariates, the most consistent results were that survivors who reported more chronic social stress reported more pain and sadness in response to IL-1Ra increases. Frequent and ongoing social stress may sensitize the nervous system to the effects of inflammation, with potential implications for chronic pain and depression risk among breast cancer survivors.

1. Introduction

Individuals differ in their psychological responsivity to inflammation [42,45]. In response to an acute inflammatory response, some individuals report more pain, sadness, and fatigue than others. In the presence of chronic systemic inflammation, this symptom constellation may result in diagnosable pain, depressive, and fatigue-related disorders. Differences in psychological responsivity to inflammation help to explain why people can have clinically elevated inflammation but not chronic pain, fatigue, or clinical depression. Over one-third of middle-aged U.S. adults have chronic inflammation, but a subset of individuals have clinically elevated inflammation yet do not have a chronic pain or fatigue condition nor any history of clinical depression [13,18,51,64]. Conversely, even subthreshold inflammation may drive pain, fatigue, and depressive symptoms among particularly sensitive individuals [59]. Identifying risk factors for heightened psychological responses to acute inflammatory challenges may provide insight into risk for inflammation-driven chronic pain, fatigue, and depression.

Prior work has identified several psychosocial factors that predict inflammation-related symptom intensity. For example, expectations can play a role, as individuals who expected to be less sick than they actually felt reported more anxiety, negative affect, and fatigue after a strong inflammatory stimulus [45]. Heightened perceived stress, sensitivity to social disconnection, and anxiety and depressive symptoms all tracked with a lower mood following endotoxin administration, compared to placebo [30]. Other risk factors for experiencing more severe symptoms include higher levels of negative affect [41], neuroticism [14], sleep disturbances [12], and low socioeconomic status [14]. A combination of physiological processes (e.g., cytokine concentrations) and psychosocial processes, such as environment, experience, and expectations, shape psychological responses to inflammation.

Another important factor is sex. Women mount stronger inflammatory responses to immune challenges than men and are more sensitive to inflammation’s psychological effect [44,76]. This adverse combination may help to explain women’s higher prevalence of chronic

pain, fatigue, and depression [17,50]. Therefore, it is important to identify risk factors for heightened psychological responsivity to inflammation among women [44]. The current study did so among breast cancer survivors – a population at particularly high risk for depression, fatigue, and chronic pain [1,39,75].

1.1 The Current Study

In these secondary analyses of a parent trial (ClinicalTrials.gov Identifier: NCT02415387) [36], we examined whether acute or chronic social and non-social stress sensitized women to an inflammatory challenge (i.e., a typhoid vaccine), resulting in elevated pain, sadness, and fatigue. It is well-known that stress activates proinflammatory pathways [68], but here we investigate whether stress may also track with heightened psychological responsivity to inflammation. Our prior publication showed that women's inflammatory responses varied following typhoid vaccination, even though none of the women had received a prior typhoid vaccine [36]. Other work has also found variability in the typhoid vaccine inflammatory response [7,27,73]. Therefore, we predicted that stress would interact with inflammatory trajectories, rather than injection type (vaccine, placebo), to predict increases in pain, sadness, or fatigue. Also, building on our prior work [52], we hypothesized that higher levels of chronic social stress, but not other types of stress, would strengthen the relationship between inflammation and symptom increases.

2. Methods

2.1 Procedure

Participants were breast cancer survivors (*N* = 156, stage I-IIIA, ages 36-78) who were one to nine years post-completion of all primary cancer treatment except for longer-term hormonal therapies (tamoxifen, aromatase inhibitors). Most of the survivors were recruited via direct referrals from the James Cancer Hospital outpatient clinic at XXX University Medical Center. Exclusions included any other current or past cancer diagnosis except basal or squamous cell skin cancers, strokes, diabetes, anemia, current heart disease or uncontrolled

hypertension, liver disease, autoimmune or other inflammatory diseases, a prior typhoid vaccination, any other vaccination within the past month, alcohol or drug abuse, smoking, and medical conditions that would limit participation (e.g., dementia). Blood tests confirmed that women were not diabetic or anemic. Women were also excluded if they took steroids, statins, or other medications with anti-inflammatory actions. Additional sample information is available in the initial publication from this trial [36].

The study was approved by The XXX University Institutional Review Board, and all survivors provided written informed consent. Women completed a screening visit during which time they completed questionnaires and a graded cycle ergometry exercise test as previously described [58], and were assessed for eligibility. If they met criteria, they returned for the first of two study visits and were randomized to sequence (vaccine/placebo or placebo/vaccine) for this within-subjects crossover design. At the beginning of each 9.5-hour visit (approximately 07:30), nurses inserted an intravenous catheter and drew a baseline blood sample. The nurse then injected saline (the placebo) or Typhoid capsular polysaccharide vaccine (Typhim-Vi, Sanofi Pasteur) into the non-dominant deltoid muscle. Women ate standardized meals. Approximately 20 minutes after receiving the injection, a trained experimenter conducted the Daily Inventory of Stressful Events (DISE) interview. Prior to the injections and then every 90 minutes for 8 hours after, they had their blood drawn to assess the inflammatory response trajectory. Shortly after each blood draw, they rated their sadness, pain, and fatigue.

Data collection occurred between August 2013 and May 2021. Although the goal was one month between visits, the two ~ 9.5-hour sessions occurred 26–420 days apart (M = 48.2, SD = 49.4) because the COVID-19 pandemic delayed some sessions. Overall, 139 participants completed both visits prior to the pandemic (M: 43.1 days between visits, SD=32.4), seven had Visit 1 before the pandemic and never completed Visit 2, two had Visit 1 before the pandemic and Visit 2 when data collection resumed during the COVID-19 pandemic (M=309.0, SD=157.0), and eight had both visits when data collection resumed (M=72.1, SD=82.4). Among

those who completed at least one visit during the pandemic, one reported having a positive COVID-19 test, and another suspected that they had been infected with COVID-19; actual or suspected infections occurred four to six months before the visit.

2.2 Measures

2.2.1 Chronic Stress

The 12-item Trier Inventory of Chronic Stressors (TICS) screening measure asked women how frequently in the past three months they experienced various stressors, such as “I try in vain to get recognition for my good work,” “I experience having too much to do” (Cronbach's α=.90) [63]. Women responded on a five-point Likert scale ranging from 0 ‘never’ to 4 ‘very often.’ Although some items of the TICS chronic stress screen tap into social elements (e.g., relationship dynamics with co-workers, caregiving), it is not primarily, predominately, or exclusively a measure of social stress. Items are summed such that a higher total score indicates more chronic stress.

Survivors also completed the 21-item the Test of Negative Social Exchange (TENSE), which asks how frequently in the past month they experienced various stressful social interactions with important people in their lives (e.g., “someone wouldn’t let me finish talking,” “someone was rude to me”) [69]. The three subscales are interfering, insensitive, and angry interactions, which contained 6, 11, and 4 items, respectively (.88 < αs < .96). Women answered on 10-point Likert scales ranging from 0 ‘not at all’ to 9 ‘frequently.’ Item-level scores are summed, such that higher total scores indicate more frequent tense interactions. Thus, the maximum scores for the interfering, insensitive, and angry subscales were 54, 99, and 36, respectively. After the first 13 participants, TENSE and TICS screen measure were moved from the screening visit to Visit 2 due to a lack of time at the screening visit and the fact that these measures index long-standing stress exposure. There were no differences in scores of those who completed the measures at the screen visit versus those who completed them at Visit 2 (*p*s>.24).

2.2.2 Acute Stress

The well-validated Daily Inventory of Stressful Events assessed the occurrence of stressors in the 24 hours prior to each visit [2]. This flexible instrument captures individuals' idiosyncratic stressors. In consensus meetings, trained research assistants coded transcribed interview responses using the Daily Inventory of Stressful Event's extensive electronic 'dictionary' [2]. Social stress was defined as reporting of a stressor that involved the participant as well as at least one other person. Variables of interest were the total number of social stressors and total number of non-social stressors women experienced in the past 24 hours.

2.2.3 Symptom Ratings

To measure sadness, women completed the widely-used Self-Assessment Manikin (SAM), which pictorially represents a continuum of affect from 1 "happy, satisfied, hopeful, and glad" to 10 "sad, unsatisfied, hopeless, depressed" [6]. To ensure that symptom ratings were on a similarly structured scale for ease of comparison, we created separate 10-point fatigue (weariness, tiredness) and overall pain Likert scales ranging from "no pain" or "no fatigue" to "pain as bad as you can imagine" or fatigue as bad as you can imagine." Importantly, for all three symptoms, women were asked about how they felt *right now.* Our fatigue and pain scales are comparable to other numerical rating scales with demonstrated psychometric soundness and clinical relevance [19,55]

2.2.4 Cytokines

We measured interleukin-6 (IL-6) and interleukin-1 receptor antagonist (IL-1Ra) based on evidence that they increase in response to the typhoid vaccine [29,33]. Although IL-1Ra is anti-inflammatory, it rises in tandem with the proinflammatory cytokine interleukin-1 beta (IL-1β) [22]. Compared to IL-1β, IL-1Ra is more detectable in the bloodstream [10,25], and it is a stronger correlate of disease severity across many chronic conditions [20,25,62]. Serum IL-6 was assayed with the Quantikine HS ELISA kit (R & D Systems, Minneapolis, MN), and serum IL-1Ra was assayed using an electrochemilluminescence method with Meso Scale Discovery

kits. Sensitivity for IL-6 was 0.03 pg/mL, the intra-assay coefficient of variation was 4.1%, and the inter-assay coefficient of variation was 6.5%; corresponding values for IL-1Ra were 6.3 pg/mL, 4.1% and 8.6%. Each woman's stored serum samples were assayed for each inflammatory marker in one run, thus using the same controls for all time points.

2.2.5 Covariates

At the screening visit, the ergometry test assessed peak oxygen consumption (VO_2max), the gold standard cardiorespiratory fitness measure [31]. Also at this visit, the valid and reliable Charlson Index, which was first tested among breast cancer survivors, provided data on comorbidities [11,16]. At the screening visit and second study visit, the Pittsburgh Sleep Quality Index assessed sleep quality in the past month (.62< αs < .68) [9]. At the first study visit, dual x-ray absorptiometry (DXA) assessed central adiposity (model DPX-NT/software version 5.60, GE Lunar, Madison, WI) – a better measure than BMI, which misclassifies adiposity status in one-third of women [32]. Lastly, participants reported their depressive symptoms in the past week on the Center for Epidemiological Studies Depression Scale at both study visits (Cronbach's α=.86 at both visits) [5,66].

2.3 Analytic Method

Area under the curve with respect to increase (AUCi) was used to summarize changes in pain, fatigue, and sadness across the day (six timepoints, one following each blood draw). IL-6 and IL-1Ra values were highly right-skewed, so they were log-transformed to reduce the impact of large values. With these transformed values, we calculated AUCi to summarize the magnitude of IL-6 change across the day (six timepoints: pre-injection baseline, 1.5, 3, 5, 6.5, 8 hours post-injection). IL-1Ra does not change as quickly as IL-6, so it was not assessed as frequently and an AUCi calculation was not possible. To capture change in IL-1Ra, we subtracted logIL-1Ra values at baseline from values at 6 hours post-injection.

For all analyses, we used general estimating equation (GEE) models with robust standard error and working correlation structure [3,77]. These models capture the within-subject

correlation arising from the repeated measurements on a subject both within a visit and between visits without requiring distributional assumptions. Preliminary models tested whether each stress measure (predictor of interest, each tested in a separate model) modulated the inflammatory response to the vaccine. Then we tested each inflammatory change score/AUCi as a predictor of AUCi for sadness, pain, and fatigue, in separate models. Primary models mirrored these analyses, except they added stress measures (1. Acute social, 2. Acute non-social, 3. Chronic social, 4. Chronic non-social) as moderators, in separate models. All models controlled for the same covariates included in our initial paper, which are known correlates of the vaccine-induced inflammatory response: age, sleep quality, trunk fat, depressive symptoms, VO_2max, time from their primary cancer treatment end date to Visit 1, visit order, stage (I versus II/III), cancer treatment type (surgery versus all, surgery and chemotherapy versus all, surgery, radiation versus all), comorbidities (0 versus 1 or more). Note that we additionally controlled for the baseline values of the inflammatory marker to account for regression to the mean. We also adjusted for injection type because our initial publication from this trial showed that the vaccine predicted heightened pain and inflammation [36], compared to the placebo, and we wanted to test whether those who had larger inflammatory responses to the vaccine also had more depression-relevant symptoms. Significant interactions were probed at the 25^{th} and 75^{th} percentiles of the moderator (i.e., stress) to obtain estimates for the simple slope of the inflammatory marker on the symptom outcome at high and low levels of stress.

3. Results

3.1 Demographic Information

Breast cancer survivors were middle-aged (*M*=56.53, *SD*=8.36), primary White (92.31%), highly educated (71.16% graduated from college), and most were overweight or obese according to BMI classifications (64.10%). In terms of stress exposure, participants experienced between zero and three social stressors (25^{th} percentile: 0, 50^{th} percentile: 0, 75^{th} percentile: 1) and between zero and four non-social stressors (25^{th}: 0, 50^{th}: 1, 75^{th}: 1) in the prior

24 hours. Scores on the TICS general life stress screen ranged from zero to 33 (25^{th}: 9, 50^{th}: 15, 75^{th}: 19). For chronic social stress, scores ranged from zero to 36 for angry interactions (25^{th}: 1, 50^{th}: 4, 75^{th}: 10), from zero to 54 for interfering interactions (25^{th}: 1, 50^{th}: 5, 75^{th}: 13), and from zero to 99 for insensitive interactions (25^{th}: 3, 50^{th}: 9, 75^{th}: 22). See Table 1 for demographic information.

3.2 Manipulation Check: Injection Type and Inflammatory Response

As reported in our initial publication, both IL-6 and IL-1Ra increased in response to the typhoid vaccine [37]. Both inflammatory responses were more variable following the vaccine than the placebo, as depicted by the wider boxes and longer whiskers in Figure 1A-B. None of the stress measures influenced the post-injection inflammatory response (*p*s>.07).

3.3 Typhoid Vaccine, Inflammatory Response, and Symptom Ratings

As we have reported elsewhere, survivors reported more pain but not sadness or fatigue following the typhoid vaccine [37,53]. Symptom trajectories are depicted in Supplemental Figure 1. Here we additionally report that IL-6 increases predicted greater pain increases ($\chi^2 = 4.3$, p=.038), but inflammatory responsivity was unrelated to any other symptom trajectory (*p*s>.07) (Table 2).

3.4 Primary Models

3.4.1 Acute Social Stress

Results from all primary models are shown in Table 3. Acute social stress occurrence did not interact with IL-1Ra or IL-6 to predict changes in pain (*p*s>.65), sadness (*p*s>.39), or fatigue (*p*s>.90) ratings. Thus, women who reported social stressors in the 24 hours prior to vaccination were not more psychologically sensitive to increases in IL-1Ra or IL-6.

Chronic Social Stress

None of the chronic social stress subscales interacted with IL-6 trajectories to predict changes in pain (*p*s>.16), sadness (*p*s>.29), or fatigue (*p*s>.29). In models with IL-1Ra trajectories, frequency of angry ($\chi^2 = 6.16$, p=.013) and interfering ($\chi^2 = 7.74$, p=.005) but not

insensitive (p=.07) interactions modulated IL-1Ra increase's relationship with pain. That is, among women who had more angry and interfering interactions, IL-1Ra increases predicted more pain. The relationship between IL-1Ra and pain increases was significant for those who scored at least 11 on the angry interactions subscale or at least 13 on the interfering interactions subscale. A similar pattern of results emerged when modeling sadness ratings: Frequency of angry (χ^2 = 4.99, p=.026), insensitive (χ^2 = 8.32, p=.004), and interfering (χ^2 = 4.00, p=.046) exchanges modulated IL-1Ra's relationship with sadness. Specifically, IL-1Ra increases predicted sadness increases among women who scored at least 21 on the angry interactions subscale, or at least 37 on the insensitive interactions subscale, or at least 27 on the interfering interactions subscale. Frequency of social stress in the prior month did not modulate IL-1Ra's relationship with fatigue (ps>.53). Significant models are depicted in Figure 2A-B.

3.4.2 Acute Non-Social Stress

Experiences of non-social stress in the past 24 hours did not render women more susceptible to the effect of IL-6 or IL-1Ra increases on changes in pain (ps>.20) or fatigue (ps>.26). However, acute stress interacted with IL-6 increases (χ^2 = 4.77, p=.029), but not IL-1Ra increases (p=.38) to predict changes in sadness. Even so, when probing the significant interaction, there were no values of the moderator (acute non-social stress) at which IL-6 increases predicted sadness.

3.4.3. Chronic General Stress

Frequency of general stress over the past three months did not modulate the relationship between IL-1Ra increases and pain (p=.90) or sadness (p=.44), yet it interacted with IL-1Ra increases to predict fatigue (χ^2 = 8.80, p=.003). Specifically, IL-1Ra increases predicted changes in sadness only among people who scored 16 or lower on the TICS stress screen (Figure 3). Frequency of general life stress over the past three months did not interact with IL-6 trajectories to predict changes in pain (p=.86), sadness (p=.27), or fatigue (p=.14).

4. Discussion

In this large, randomized, placebo-controlled trial among middle-aged breast cancer survivors, the most consistent finding was that chronic conflict-related social stress interacted with IL-1Ra increases to predict changes in pain and sadness—in line with hypotheses. Put another way, survivors who reported more frequent social stress in the past month were more sensitive to the pain and sadness-related effects of IL-1Ra increases. Notably, standardized slopes in the significance regions were small to moderate (Supplemental Figures 2-3). We also observed a direct, positive relationship between IL-6 and pain increases. Contrary to hypotheses, our variables of interest did not predict fatigue. Our results suggest that breast cancer survivors who experience more conflict-related social stress may experience more pain and sadness in response to inflammation.

4.1 Psychological Responsivity to Inflammation

Notably, none of the stress measures predicted greater inflammatory responsivity to the vaccine; rather, chronic social stress predicted greater psychological responsivity to inflammation. This distinction is subtle but critical, showing that breast cancer survivors who experience frequent social stress may respond with more sadness and pain following even modest inflammatory responses. This finding adds to nascent translational research showing, for instance, that inflammatory responses following influenza vaccination translated to more cognitive difficulties and depressed mood among those who experienced early life stress, as compared to their peers [40]. Also, our data align with observational findings that stress predicts greater pain in various chronic inflammatory diseases, such as rheumatic diseases [28] and irritable bowel syndrome [23].

4.2 The Biological Relevance of Chronic or Repetitive Social Stress

Our results mirror prior findings from our lab that demonstrated the unique and powerful effects of conflict-related social stress, compared to work-related stress [52]. In one healthy sample, those with heightened and prolonged inflammatory reactivity to a 20-minute marital

conflict, as well as frequent social stress over the past month, reported heightened depressive symptoms one month later. In another sample of breast cancer survivors, those with exaggerated inflammatory reactivity to a laboratory speech stressor and greater loneliness (or less social support) experienced steeper depressive symptom rises over a four- and eight-month follow-up period. These findings were specific to social stress and did not extend to general perceived stress. Our prior and current findings tell a coherent story: Those who have greater inflammatory responses to acute environmental stressors – either pathogenic or psychosocial – and who experience frequent, chronic social stress may experience more pain and sadness in the short term, which may lead to depressive symptom worsening over time.

In the current study, no consistent story emerged for other types of stress. For example, chronic general stress interacted with IL-1Ra responses to predict fatigue. Heightened IL-1Ra responses predicted greater fatigue ratings only among women with lower chronic general stress. Although we would expect that inflammation increases would be most strongly associated with fatigue at higher stress levels, this finding nonetheless aligns with extant literature, which generally shows a positive, causal relationship between inflammation and fatigue [43].

4.3 Possible Mechanisms

There are several mechanisms by which psychosocial factors can increase emotional and pain responsivity to fluctuations in inflammation. Most of this literature has yet to be translated from animals to humans. Firstly, chronic stress can dysregulate HPA axis function, leading to flatter diurnal cortisol slopes, higher basal cortisol levels, and hippocampal function – contributing to persistent pain [26,74]. Chronic and repetitive activation of another stress-sensitive system – the sympatho-adrenal-medullary axis – can also mediate stress-induced hyperalgesia [34]. Specifically, stress hormones released via these axes (e.g., cortisol, epinephrine) can sensitize neurons [35], and proinflammatory cytokines can directly activate these sensory neurons [78], contributing to lower pain thresholds. Another possibility is that

chronic social stress may weaken the blood-brain barrier [46], affording peripheral inflammatory mediators greater access to the brain. Indeed, peripheral IL-1Ra and IL-6 can cross the blood-brain barrier [4]. Social stress can also prime microglia – a key brain immune cell – to overreact, promoting neuroinflammation [70,71]. Thus, chronic social stress may promote neuroinflammation following a peripheral inflammatory challenge, perhaps contributing to more intense sickness symptoms. In short, several stress-responsive neuroendocrine and immune pathways can help to explain why those who experience more frequent social stress may be more psychologically sensitive to inflammation.

4.4 IL-1 and Psychological Symptoms

Although we observed a positive and direct relationship between IL-6 and pain ratings, our most consistent finding was the multiplicative impact of IL-1Ra and chronic social stress on sadness and pain trajectories. Our results parallel work in animal models that demonstrates IL-1's centrality in psychological symptoms. In animals, IL-1β administration alone promotes psychological symptoms, whereas IL-6 potentiates IL-1's effects [15,47]. Specifically, there is strong evidence that IL-1 in the brain mediates pain [54] and depressive-like behaviors [24,38]. In a small human sample, those who were hospitalized for unmedicated depression had higher cerebrospinal fluid concentrations of IL-1β – but not IL-6 or TNF-alpha – than healthy controls, and levels of IL-1β correlated with depression severity [48]. IL-1β is implicated in human pain disorders, and IL-1β antagonism is a promising therapeutic strategy [67]. These findings add to the translational literature showing IL-1's centrality to pain and low mood in humans.

We were not able to reliably predict fatigue trajectories. Meta-analytic evidence among patients with chronic fatigue syndrome found that other cytokines (tumor necrosis factor – alpha, interleukin-2, interleukin-4, C-reactive protein, transforming growth factor) may track more closely with fatigue [60,72]. Moreover, there is evidence of biological heterogeneity in fatigue; certain cytokines (e.g., IL-6) only tracked with fatigue in healthy controls, yet tumor-necrosis factor-alpha was a more universal predictor across subsamples [61].

4.5 Clinical Implications

Meta-analytic evidence suggests that 45% of breast cancer survivors struggle with lingering pain after curative treatment, and almost half of these cases are moderate to severe [75]. Nearly half of people with chronic pain have a mood disorder, and over 60% of people with depression report a chronic pain condition, such as fibromyalgia [56,65]. The high comorbidity rate is particularly troubling given that these disorders are more treatment-resistant when they present together [21]. Results from our study point to IL-1 and chronic social stress as two potentially modifiable etiological factors and treatment targets to explore among breast cancer survivors with chronic pain and depression.

4.6 Strengths and Limitations

The paradigm has many strengths. Firstly, we used a mild inflammatory stimulus (i.e., a typhoid vaccine), in which the inflammatory response magnitude is on par with the inflammation observed in depression; stronger inflammatory stimuli like endotoxin may be a better model for infectious illness [49]. Further, we repeatedly drew blood to assess the inflammatory response following both the saline placebo injection and typhoid vaccine. There was substantial variability in inflammatory responses, which is why we used inflammatory response magnitude rather than injection type in our models; yet we controlled for injection type so that we could determine whether those with heightened inflammatory responses to the typhoid vaccine and chronic social stress had higher symptom ratings.

In terms of limitations, an acute inflammatory stimulus as a model of chronic pain and depression is imperfect due to its transient nature. Furthermore, we measured only two cytokines, and one (IL-1Ra) is an anti-inflammatory proxy for proinflammatory IL-1β. We also acknowledge that larger inflammatory responses and more sickness symptoms following vaccination are not necessarily a correlate or harbinger of poor health; in fact, there is some evidence that a stronger innate immune response and more intense sickness symptoms predict a better adaptive immune response to vaccination [8,57]. Indeed, the problem may be

heightened psychological responsivity to even modest inflammatory rises, rather than high raw values of sickness symptoms or inflammatory responses by themselves. Another consideration is that most women completed the chronic stress measures at the second visit; although these measures assess exposure to long-lasting stressors, additional research should assess whether these observations are robust regardless of chronic stress measurement timing. In terms of our pain measure, because we did not use the 11-point Numeric Rating Scale [19], it is difficult to interpret the clinical significance of pain changes throughout the day. Also, primary hypotheses necessitated 36 statistical tests (18 for each inflammatory marker), and we did not correct for multiple tests. Thus, results should be replicated before being clinically applied. Even so, we only expected the tests involving chronic social stress to be significant; in that vein, models in which chronic social stress moderated the relationships between IL-1Ra and pain or fatigue were notable because the significance rate was much higher than would be expected if results were due to chance. Lastly, the current study included female breast cancer survivors who were primarily White and highly educated, and it is unclear if results generalize to males or more diverse or cancer-free populations.

4.7 Conclusions

This randomized, placebo-controlled trial demonstrated that chronic conflict-related social stress was associated with greater increases in pain and sadness among those with steeper IL-1Ra response trajectories. These results parallel findings over a longer timescale [52]. Chronic social stress, especially among those with a robust inflammatory response, may increase risk for pain and low mood. These findings may help to explain chronic pain and depression comorbidity in breast cancer survivorship.

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Figure Captions

Figure 1A-B. Variability in the typhoid vaccine-induced inflammatory response. As depicted by the wider box and longer whiskers for the interleukin-6 (IL-6; Figure 1A) and interleukin-1 receptor antagonist (IL-1Ra; Figure 1B) responses, the typhoid vaccine triggered a more variable inflammatory response than the saline placebo injection.

Figure 2A-B. Chronic social stress predicts greater sadness and pain responsivity to IL-1Ra response. These Johnson-Neyman Plots show the regions of significance (i.e., values of the social stress moderator) in which IL-1Ra increases predict higher sadness (Figure 2A) and pain (Figure 2B) ratings. In the shaded regions that do not include a 0 effect (the dotted, horizontal line), IL-1Ra increases predict greater pain and sadness. That is, women who reported more frequent angry, insensitive, and interfering exchanges were more psychologically sensitive to IL-1Ra increases.

Figure 3. Chronic general stress moderates the relationship between IL-1Ra increases and fatigue ratings. This Johnson-Neyman Plot depicts the regions of significance (i.e., values of chronic general stress) in which IL-1Ra increases predict fatigue rating increases. In the shaded regions that do not include a 0 effect (the dotted, horizontal line), IL-1Ra increases predict greater fatigue increases. That is, among women with lower levels of chronic general stress, IL-1Ra increases predicted greater fatigue ratings.

Tables

Table 1. Demographic information at visit 1.

Variable	N	Mean (SD) or N (%)	Range
Age	156	56.53 (8.36)	36.00-78.00
Race			
White		144 (92.31%)	
Black		10 (6.41%)	
Mixed		2 (1.28%)	
BMI, kg/m^2	156	27.84 (5.89)	18.70-45.50
Trunk fat, kg	156	15.65 (6.71)	3.14-34.26
VO_2max, mL/kg/min	156	22.04 (5.35)	10.20-33.60
Education	149		
High school or less		19 (12.18%)	
Some college		26 (16.67%)	
College graduate		55 (35.26%)	
Graduate school/professional training		56 (35.90%)	
Months since treatment	156	43.10 (27.86)	9.00-119.00
Chemotherapy treatment	156	106 (67.95%)	
Radiation treatment	156	93 (59.62%)	
Current hormone therapy	156	126 (80.77%)	
Cancer stage	156		
Stage I		74 (47.44%)	
Stage II		75 (48.08%)	
Stage III		7 (4.49%)	
Any comorbidities	156	20 (12.82%)	
CES-D score	156	7.43 (6.67)	0-37.00
Fasting IL-6, pg/mL	155	2.85 (6.42)	0.42-78.37
Fasting IL-1Ra, pg/mL	155	547.83 (388.12)	121.04-1991.10
Baseline fatigue rating	156	1.71 (1.64)	0-7
Baseline pain rating	156	0.54 (1.02)	0-7
Baseline sadness rating	156	2.68 (1.44)	1-8
TENSE angry interactions	150	6.57 (8.06)	0-36
TENSE insensitive interactions	150	17.20 (20.19)	0-99
TENSE interfering interactions	150	8.79 (10.42)	0-54
TICS chronic stress scale	150	14.60 (7.64)	0-33
DISE number of personal stressors	156	0.64 (0.71)	0-3
DISE number of non-personal stressors	156	0.98 (0.91)	0-4

CES-D = Center for Epidemiological Studies Depression Scale; TENSE = Test of Negative Social Exchange; TICS = Trier Inventory of Chronic Stressors; DISE = Daily Inventory of Stressful Events; IL-6 = interleukin-6, IL-1Ra = interleukin-1 receptor antagonist

Table 2. P-values for main effect of inflammation.

		P-value	
Outcome	**N**	**Vaccine***	**Predictor**
Predictor = IL-6 increase			
Pain increases	156	0.13	**0.038**
Sadness increases	156	0.68	0.69
Fatigue increases	156	0.97	0.91
Predictor = IL-1Ra increase			
Pain increases	156	0.0022	0.22
Sadness increases	156	0.82	0.68
Fatigue increases	156	0.66	0.071

IL-6 increase = area under the curve with respect to increase (AUCi) of log-transformed interleukin-6 values; IL-1Ra increase = AUCi of log-transformed interleukin-1 receptor antagonist values; *As stated in the methods section, we controlled for injection type (vaccine, placebo) in models testing inflammation's relationship with symptoms; here we report the p-values for the vaccine as well as the inflammation predictor of interest (included in the same model).

Table 3. P-values for inflammation by stress interactions.

	Inflammation = IL-6 increase				Inflammation = IL-1Ra increase			
Outcome	**N**	**Stress**	**IL-6 increase**	**Stress*IL-6 increase**	**N**	**Stress**	**IL-1Ra increase**	**Stress*IL-1Ra increase**
Stress = Chronic social stress (TENSE angry interactions)								
Pain increases	150	0.17	0.50	0.17	150	0.11	0.20	**0.013**
Sadness increases	150	0.43	0.99	0.30	150	0.84	0.20	**0.026**
Fatigue increases	150	0.11	0.79	0.66	150	0.14	0.40	0.54
Stress = Chronic social stress (TENSE insensitive interactions)								
Pain increases	150	0.20	0.42	0.39	150	0.07	0.42	0.07
Sadness increases	150	0.46	0.64	0.91	150	0.90	0.13	**0.004**
Fatigue increases	150	**0.005**	0.49	0.30	150	**0.004**	0.23	0.82
Stress = Chronic social stress (TENSE interfering interactions)								
Pain increases	150	0.24	0.54	0.23	150	0.11	0.14	**0.005**
Sadness increases	150	0.86	0.86	0.59	150	0.82	0.24	**0.046**
Fatigue increases	150	**0.040**	0.77	0.73	150	**0.010**	0.23	0.97
Stress = Chronic general stress (TICS screen)								
Pain increases	150	0.95	0.25	0.86	150	0.76	0.32	0.90
Sadness increases	150	0.12	0.20	0.27	150	0.21	0.64	0.44
Fatigue increases	150	0.05	0.25	0.14	150	**0.036**	**0.001**	**0.003**
Stress = Acute social stress (DISE)								
Pain increases	156	0.80	0.10	0.98	156	0.94	0.35	0.66
Sadness increases	156	0.95	0.99	0.40	156	0.58	0.95	0.47
Fatigue increases	156	0.51	0.98	0.99	156	0.30	0.18	0.91
Stress = Acute non-social stress (DISE)								
Pain increases	156	0.13	**0.013**	0.21	156	0.36	0.17	0.84
Sadness increases	156	**0.010**	0.06	**0.029**	156	0.19	0.19	0.38
Fatigue increases	156	0.10	0.62	0.27	156	0.11	0.11	0.94

TENSE = Test of Negative Social Exchange; TICS = Trier Inventory of Chronic Stressors; DISE = Daily Inventory of Stressful Events; IL-6 increase = area under the curve with respect to increase (AUCi) of log-transformed interleukin-6 values; IL-1Ra increase = AUCi of log-transformed interleukin-1 receptor antagonist values; **p<.05**