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# Chemokines Are Associated With Delirium After Cardiac Surgery

James L. Rudolph,[1,10] Basel Ramlawi,[2,6] George A. Kuchel,[4] Janet E. McElhaney,[3,4,5] Dongxu Xie,[3,4] Frank W. Sellke,[6] Kamal Khabbaz,[6] Sue E. Levkoff,[9] and Edward R. Marcantonio[7,8]

[1]Geriatric Research, Education, and Clinical Center, VA Boston Healthcare System, Massachusetts.
[2]Division of Cardiac Surgery, University of Western Ontario, London, Ontario, Canada.
[3]Center for Immunotherapy of Cancer and Infectious Diseases, University of Connecticut School of Medicine, Farmington.
[4]University of Connecticut Center on Aging, University of Connecticut Health Center, Farmington.
[5]Geriatric Medicine, University of British Columbia, Vancouver, Canada.
Divisions of [6]Cardiac Surgery, [7]General Medicine and Primary Care, and [8]Gerontology,
Beth Israel Deaconess Medical Center, Boston, Massachusetts.
[9]Department of Psychiatry and [10]Division of Aging, Brigham and Women's Hospital, Boston, Massachusetts.

***Background.*** Delirium has been hypothesized to be a central nervous system response to systemic inflammation during a state of blood–brain barrier compromise. The purpose of this study was to compare postoperative changes in groups of inflammatory markers in persons who developed delirium following cardiac surgery and matched controls without delirium.

***Methods.*** Serum samples were drawn from 42 patients undergoing cardiac surgery preoperatively and postoperatively at 6 hours and postoperative day 4. The serum concentrations of 28 inflammatory markers were determined with a microsphere flow cytometer. A priori, inflammatory markers were assigned to five classes of cytokines. A class *z* score was calculated by averaging the standardized, normalized levels of the markers in each class. Beginning on postoperative day 2, patients underwent a daily delirium assessment.

***Results.*** Twelve patients with delirium were matched by surgical duration, age, and baseline cognition to 12 patients without delirium. At the 6-hour time point, patients who went on to develop delirium had higher increases of chemokines compared to matched controls (class *z* score $0.3 \pm 1.0$, $p < .05$). Among the five classes of cytokines, there were no other significant differences between patients with or without delirium at either the 6 hour or postoperative day 4 assessments.

***Conclusion.*** After cardiac surgery, chemokine levels were elevated in patients who developed delirium in the early postoperative period. Because chemokines are capable of disrupting blood–brain barrier integrity in vitro, future studies are needed to define the relationship of these inflammatory mediators to delirium pathogenesis.

***Key Words*:** Inflammation—Delirium—Cardiac surgery—Chemokines—Cytokines.

DELIRIUM, a multifactorial geriatrics syndrome (1), is a common complication of cardiac surgery, occurring in 32%–72% of older patients (2). Postoperative delirium has been associated with increased mortality (3), postoperative complications (3), functional decline (4), and increased cost (5). Despite the high prevalence and negative sequelae of postoperative delirium, its pathophysiology remains unknown (6,7). Nevertheless, it has been proposed that systemic inflammation may actually contribute to delirium pathogenesis by compromising blood–brain barrier (BBB) integrity (8–10).

The magnitude of the operative inflammatory response has been implicated as a risk factor of neurocognitive decline, including delirium, after surgery (11–13). Normally, the BBB inhibits cytokines and many medications from passing across capillaries into the brain parenchyma (14). Thus, the brain is relatively protected from the effects of systemic inflammation. Delirium is felt to be a central nervous system (CNS) manifestation of a systemic disease state that may indeed cross the BBB. In many of the situations in which delirium is likely to occur (e.g., infections, postoperative states), BBB integrity may be compromised. Chemokines are locally acting cytokines that enhance migration of inflammatory cells into the brain by compromising the BBB (15,16). When BBB integrity is compromised, the brain becomes more susceptible to the effects of systemic inflammation (10,15).

The inflammatory response to surgery involves a complex coordination of cytokines, chemokines, and adaptive physiological responses required to maintain homeostasis. A transient postoperative increase in levels of circulating inflammatory markers (10–100 times more than baseline) has been hypothesized to result from tissue damage, adrenal stress response, cardiopulmonary bypass, and/or anesthesia (17–19). Inflammatory marker levels peak 6–24 hours postoperatively and return to baseline levels over 2–4 days (18,19). Inflammatory cytokines are produced by activated immune cells and are involved in the early amplification of the inflammatory response. Chemokines rapidly mediate leukocyte movement to sites of inflammation, including across the BBB. Cytokines that promote a T-helper 1 (TH-1) leukocyte and cytotoxic T-lymphocyte (CTL) response increase cell proliferation and CD8 expression. Cytokines that promote a T-helper 2 (TH-2) response are responsible

for B-cell proliferation and humoral immune maturation. As a result of their respective roles, inflammatory cytokines and chemokines should be elevated early, with cytokines promoting TH-1/CTL and TH-2 responses increased during later stages of the postoperative inflammatory response.

The purpose of this study was to determine if a difference exists in the postoperative pattern of change in *a priori* determined classes of inflammatory markers in matched patients with and without delirium after cardiac surgery. We hypothesized that, as compared to their baseline levels, patients with delirium would have (i) increased inflammatory cytokines and chemokines 6 hours postoperatively and (ii) increased cytokines that promote TH-1/CTL and TH-2 responses 4 days postoperatively.

## Methods

### *Patient Enrollment*

We prospectively enrolled 42 patients undergoing elective or urgent cardiac surgery at an academic medical center. Eligible procedures included coronary artery bypass graft (CABG), valve replacement, and combined CABG–valve surgery. Patients with preoperative delirium, active substance abuse, psychiatric disease, and/or aortic procedures were excluded. Patients provided their written informed consent, and the study was approved by the institutional review board.

### *Anesthetic and Surgical Methods*

Operative procedures were completed by three surgeons using the same conventional approach, including induction of general anesthesia, invasive monitoring, midline sternotomy, and systemic heparinization. Mild hypothermic cardiopulmonary bypass (CPB) with cold-blood hyperkalemic cardioplegia was used. All patients received antibiotics preoperatively and up to 48 hours postoperatively.

### *Measurement of Inflammation*

Prior to surgery and 6 hours after surgery in the intensive care unit (ICU), blood samples were collected from the central venous line. Postoperative day 4 samples were collected peripherally. Blood samples were processed, and serum samples were frozen at −80°C until the time of assay.

Samples were analyzed on a Luminex 100 dual-laser, microsphere flow cytometer (Luminex, Austin, TX) using combined BioSource human cytokine 25-plex and death receptor 3-plex bead kits (Invitrogen, Carlsbad, CA). Samples were incubated with the beads for 2 hours, washed, incubated with biotinylated detector antibodies for 1 hour, washed, incubated for 30 minutes with a conjugated fluorescent protein, and again washed. For each inflammatory marker measured, a standard curve was developed using four known concentration standards. The fluorescence of each inflammatory marker was converted to a concentration using the standard curve. In accordance with standard practice, samples with undetectable cytokine levels were entered at half of the minimum detection level derived from the standard curve.

*A priori* and based on the consensus of three experts in the study of inflammation, inflammatory markers were assigned to one of five classes: (i) inflammatory cytokines; (ii) cytokines that promote TH-1/CTL responses; (iii) cytokines that promote TH-2 responses; (iv) chemokines, and (v) lymphatic chemokines. Interleukin 17 (IL-17) and death receptor 5 were not assigned to any class and were analyzed independently.

### *Delirium*

A brief delirium assessment (<15 minute) was performed preoperatively and daily postoperatively, beginning on day 2. Patients were not assessed on postoperative days 0 or 1 because of the intensive medical care required after CABG surgery. Delirium was assessed using the diagnostic algorithm of the Confusion Assessment Method (CAM) (20). Prior to its completion, a standardized mental status interview was conducted, including the Mini-Mental State Examination (MMSE) (21), digit span, the Delirium Symptom Interview (DSI) (22), and the Memorial Delirium Assessment Scale (MDAS) (23). The MMSE is a screening assessment of mental status. The digit span asks patients to repeat a series of random digits forward and backward and is an assessment of working memory and attention. The DSI is an interview for eliciting eight key symptoms of delirium. The MDAS is a severity scale for delirium. This combined assessment for delirium has been shown to be highly reliable ($\kappa = 0.95$) (24) when administered by trained, nonclinician interviewers.

### *Matching*

An analyst unaware of study aims and inflammatory marker results matched patients with delirium to patients who did not develop delirium on the basis of surgery duration (±90 minutes), age (±5 years) and baseline MMSE (±3 points), respectively. Because of the small and diverse sample, the matching process was appropriate to allow comparisons of baseline characteristics that might influence the inflammatory response. We used a Student *t* test to compare the baseline characteristics of the matched controls to those of the patients with delirium and to those of the unmatched group.

### *Statistics*

As the distribution of circulating inflammatory markers is generally nonnormal, we log normalized the inflammatory marker concentrations. To calculate the postoperative inflammatory response, we subtracted the baseline log-normalized concentration from the postoperative log-normalized concentration (log[Postoperative] − log[Baseline]). The concentrations among the inflammatory markers were standardized to the mean and standard deviation of the matched nondelirious control group (marker *z* score). We created a class *z* score by averaging the marker *z* scores of the inflammatory markers within each class. We compared the mean *z* score among the assigned classes using a Student *t* test. All statistical calculations were performed using SPSS version 11.5.0 (SPSS Inc., Chicago, IL).

## Results

Among the 42 patients enrolled, 12 (29%) developed delirium. Table 1 describes the baseline characteristics of

the matched control and delirium individuals, as well as the unmatched patients. There were no significant differences between matched controls and patients with delirium in the parameters used for matching (age, MMSE score, and surgery time). Interestingly, no patients younger 60 years developed delirium. Matched controls were more likely to have a diagnosis of hypertension than were the patients with delirium, but all patients were taking preoperative medications that lower blood pressure. The unmatched patients were significantly younger (59.9 ± 5.5 years) and had significantly higher preoperative MMSE scores (28.6 ± 1.5) than the matched controls. There was no significant difference in preoperative comorbidity, medication usage, or surgical time among the groups.

Preoperative levels of inflammatory markers are compared in Table 2. Class *z* scores in patients with delirium did not differ significantly from those of matched controls. Thus, the patients were well matched for baseline inflammatory levels. Also, based on these data, it does not appear that greater levels of baseline inflammation are a risk factor for delirium.

Table 3 depicts the postoperative change in circulating inflammatory markers. At 6 hours postoperatively, circulating levels of chemokines were significantly higher (Marker *z* score >0) in patients who went on to develop delirium than in matched controls without delirium ($p < .05$). However, the inflammatory cytokine group was not elevated. On the 4th postoperative day, the levels of cytokines that promote TH-1/CTL and TH-2 responses tended to be lower (Class *z* scores <0) in patients who developed delirium, yet this difference was not statistically significant.

Table 1. Baseline Characteristics of the Matching Procedure

| Patient Characteristic | No Delirium ($N = 12$) | Delirium ($N = 12$) | Unmatched ($N = 18$) |
|---|---|---|---|
| Age, y | 73.9 (8.4) | 74.7 (7.0) | 59.9 (5.5)* |
| Male gender, *n* (%) | 9 (75%) | 11 (92%) | 17 (94%) |
| Charlson Comorbidity Index | 2.1 (2.0) | 2.9 (1.9) | 1.7 (1.6) |
| Number of medications | 7.9 (2.4) | 7.2 (2.7) | 6.2 (2.9)* |
| Body mass index, kg/m$^2$ | 30.1 (6.1) | 26.7 (4.1) | 31.3 (5.7) |
| MMSE | 25.3 (3.6) | 26.1 (2.9) | 28.6 (1.5)* |
| GDS | 2.5 (2.2) | 2.5 (1.3) | 2.4 (2.1) |
| History of *n* (%): | | | |
| Hypertension | 10 (83%) | 5 (42%)* | 10 (62%) |
| Diabetes | 5 (42%) | 4 (33%) | 7 (44%) |
| Hyperlipidemia | 8 (67%) | 5 (42%) | 7 (44%) |
| WBC (per mL) | 8.3 (2.8) | 7.4 (2.3) | 7.4 (1.6) |
| Hematocrit, % | 35.9 (5.3) | 35.1 (4.4) | 36.0 (5.4) |
| Creatinine, mg/dL | 1.9 (2.6) | 1.2 (0.4) | 1.1 (0.5) |
| Medications, *n* (%) | | | |
| Aspirin | 12 (100%) | 10 (83%) | 16 (89%) |
| NSAIDs | 0 (0%) | 2 (17%) | 2 (11%) |
| Steroids | 0 (0%) | 1 (8%) | 1 (6%) |
| Beta-blockers | 10 (83%) | 7 (58%) | 14 (78%) |
| ACE-I/ARB | 7 (58%) | 6 (50%) | 12 (67%) |
| CCB | 3 (25%) | 4 (33%) | 4 (22%) |
| Nitrate | 3 (25%) | 2 (17%) | 2 (11%) |
| Diuretic | 4 (33%) | 7 (58%) | 6 (33%) |
| Surgery time, min | 210.3 (53.9) | 202.8 (57.3) | 221.1 (54.9) |
| CPB time, min | 90.4 (38.6) | 91.4 (50.4) | 94.6 (36.3) |

*Notes*: Values are shown as mean ± standard deviation. There were no differences between patients with delirium ($n = 12$) and the matched controls without delirium ($n = 12$). The unmatched group ($n = 18$) was significantly younger and had increased Mini-Mental State Examination (MMSE) scores.

*$p < .05$ compared to No Delirium group.

ACE-I/ARB = angiotensin-converting enzyme inhibitor/angiotensin receptor blocker; CCB = calcium channel blocker; CPB = cardiopulmonary bypass; GDS = Geriatric Depression Scale (15 point scale); NSAIDs = nonsteroidal antiinflammatory drugs; WBC = white blood cell count.

## Discussion

Delirium after surgery has been hypothesized to occur as a result of the inflammatory response (9). However, the basic pathophysiology is not understood. This study used new technology to examine peripheral inflammatory marker responses to cardiac surgery in patients with delirium and in matched controls without delirium. Based on the temporal nature of typical inflammatory responses, we hypothesized that inflammatory cytokines and chemokines would be initially elevated because of their role in the control of subsequent inflammatory responses and that cytokines known to promote TH-1/CTL and TH-2 responses would be elevated on postoperative day 4. We postulated that at both times delirium would be associated with a greater increase in circulating inflammatory marker response compared to that of patients without delirium. Unexpectedly, our findings indicate a much more restricted inflammatory response. In the immediate postoperative periods, patients who later developed delirium had increased chemokine levels, but levels of classical inflammatory cytokines were similar between those who developed delirium and matched controls. At 4 days after surgery, patients with delirium tended to demonstrate smaller increases of cytokines that promote the TH-1/CTL and TH-2 response.

Chemokines have not been traditionally included in delirium research, but recent studies have added biological plausibility to a hypothesis implicating these inflammatory mediators in the earliest events contributing to delirium. Although our study was not designed to detect changes in levels of individual chemokines, elevations in inflammatory cytokines (particularly IL-6) have been linked to postoperative delirium in other studies (25,26). Chemokines are known to promote leukocyte migration into the CNS (27) and, in the case of chemokine C-C motif ligand 2 (CCL2), mediate BBB disruption in the context of ischemic injury (15). Moreover, at least three important risk factors for delirium—aging (28,29), brain injury(29), and anticholinergic medications (30)—have been associated with greater basal and induced CCL-2 activity. Our observed elevations in systemic chemokine levels are quite transient, with no detectable differences between individuals with and without delirium by the 4th postoperative day.

The cause of the nonsignificant trend toward lower TH-1/CTL and TH-2 responses in individuals with delirium at postoperative day 4 is unclear. We propose three hypotheses for this finding. First, there may be a lack of reserve capacity to mount an inflammatory response. Second, most of these cytokine elevations are very modest (exceptions: IL-4, IL-10), and these substances exerted predominantly local autocrine effects and were being consumed locally (9). Third, stress mediators, such as glucocorticoids, are likely to be elevated in the setting of delirium (31) and tend to suppress TH-1 immunity, while promoting TH-2 immunity (32). Thus, as we learn more about the nature of the

Table 2. Preoperative Inflammatory Markers

| Class | Cytokine | Raw (pg/μL) | | Normalized | | Marker *z* Scores | | Class *z* Scores | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | No Delirium | Delirium | No Delirium | Delirium | No Delirium | Delirium | No Delirium | Delirium | *p* Value |
| Group 1: Inflammatory | IL-1β | 678 (1019.5) | 368.9 (1027.5) | 4.6 (2.4) | 3.4 (2.0) | 0.0 (1.0) | −0.5 (0.9) | 0.0 (1.0) | −0.1 (1.1) | .45 |
| | IL-1Rα | 327 (518) | 247.8 (495.3) | 4.3 (2.0) | 4.1 (1.8) | 0.0 (1.0) | −0.1 (0.9) | | | |
| | IL-6 | 295.8 (422.7) | 144.1 (179.3) | 5.0 (1.2) | 4.2 (1.4) | 0.0 (1.0) | −0.7 (1.2) | | | |
| | IFN-α | 19.6 (29.1) | 45.9 (74.2) | 2.4 (1.0) | 2.7 (1.4) | 0.0 (1.0) | 0.4 (1.5) | | | |
| | TNF-α | 18.3 (21.3) | 30 (35.5) | 2.4 (0.9) | 2.7 (1.2) | 0.0 (1.0) | 0.3 (1.3) | | | |
| | TNF-R1 | 1024.6 (906) | 831.3 (895.4) | 6.5 (1.0) | 6.3 (1.0) | 0.0 (1.0) | −0.2 (1.0) | | | |
| | TNF-R2 | 7470.2 (3056.6) | 7441.9 (2125.8) | 8.8 (0.4) | 8.9 (0.3) | 0.0 (1.0) | 0.1 (0.7) | | | |
| Group 2: T-Helper 1 | IL-2 | 532.3 (838.8) | 272.4 (715.9) | 4.0 (2.6) | 3.1 (2.1) | 0.0 (1.0) | −0.3 (0.8) | 0.0 (1.0) | −0.1 (1.8) | .79 |
| | IL-2R | 87.5 (91.1) | 81 (68.2) | 4.1 (0.9) | 4.0 (0.9) | 0.0 (1.0) | 0.0 (1.0) | | | |
| | IL-7 | 65.1 (112.9) | 52.2 (83.6) | 3.0 (1.5) | 2.9 (1.4) | 0.0 (1.0) | −0.1 (1.0) | | | |
| | IL-12p40_p70 | 72 (58.5) | 94.4 (91.9) | 4.0 (0.8) | 4.1 (1.0) | 0.0 (1.0) | 0.1 (1.3) | | | |
| | IL-15 | 164.8 (256.9) | 179 (255.2) | 4.2 (1.4) | 4.1 (1.6) | 0.0 (1.0) | −0.1 (1.1) | | | |
| | IFN-γ | 26.5 (45.6) | 78.4 (141.2) | 2.6 (1.0) | 3.1 (1.6) | 0.0 (1.0) | 0.5 (1.6) | | | |
| | IP-10 | 1011.4 (598.9) | 1315.3 (1152.7) | 6.8 (0.5) | 6.5 (2.0) | 0.0 (1.0) | −0.6 (3.8) | | | |
| Group 3: T-Helper 2 | IL-4 | 25.9 (39.4) | 54.9 (102.4) | 2.6 (1.0) | 2.8 (1.4) | 0.0 (1.0) | 0.2 (1.4) | 0.0 (1.0) | 0.1 (1.3) | .59 |
| | IL-5 | 60.7 (76) | 73.1 (127.8) | 3.3 (1.4) | 3.1 (1.5) | 0.0 (1.0) | −0.1 (1.1) | | | |
| | IL-10 | 109 (107.2) | 130.8 (147) | 4.3 (1.0) | 4.4 (1.0) | 0.0 (1.0) | 0.1 (1.0) | | | |
| | IL-13 | 25.7 (21.3) | 49.9 (84.6) | 3.0 (0.6) | 3.2 (1.0) | 0.0 (1.0) | 0.3 (1.8) | | | |
| Group 4: Chemokines | MIP-1α | 121.5 (197.1) | 113.4 (196.5) | 3.6 (1.6) | 3.4 (1.7) | 0.0 (1.0) | −0.1 (1.0) | 0.0 (1.0) | −0.2 (1.2) | .28 |
| | MIP-1β | 53.3 (68.8) | 94.8 (115) | 3.4 (1.1) | 3.6 (1.5) | 0.0 (1.0) | 0.2 (1.4) | | | |
| | MIG | 42 (48.1) | 81.9 (114.8) | 3.1 (1.2) | 3.3 (1.8) | 0.0 (1.0) | 0.2 (1.4) | | | |
| | Eotaxin | 593.4 (548) | 421.2 (222.1) | 6.1 (0.7) | 5.9 (0.7) | 0.0 (1.0) | −0.4 (1.0) | | | |
| | RANTES | 7241.2 (1521.1) | 6445.7 (1427.6) | 8.9 (0.2) | 8.7 (0.2) | 0.0 (1.0) | −0.6 (1.2) | | | |
| | CCL-2 | 558 (419.7) | 424.7 (257.2) | 6.1 (0.6) | 5.8 (0.8) | 0.0 (1.0) | −0.5 (1.2) | | | |
| Group 5: Lymphatic Chemokines | IL-8 | 674.9 (1667.5) | 146.7 (133.6) | 4.7 (1.9) | 4.2 (1.6) | 0.0 (1.0) | −0.3 (0.9) | 0.0 (1.0) | −0.2 (0.8) | .52 |
| | GM–CSF | 60.4 (116.5) | 35.8 (50.6) | 2.9 (1.4) | 2.8 (1.2) | 0.0 (1.0) | −0.1 (0.8) | | | |
| Miscellaneous | IL-17 | 22.7 (29.7) | 38.5 (51.3) | 2.7 (0.9) | 3 (1.1) | 0.0 (1.0) | 0.4 (1.3) | | | |
| | DR5 | 87.7 (76.3) | 88.7 (62.4) | 3.9 (1.2) | 4.1 (1.0) | 0.0 (1.0) | 0.2 (0.8) | | | |

*Notes*: This table describes the process leading to inflammatory marker comparisons. Raw scores were log normalized because of data skew. Normalized scores were standardized to a mean of 0 and standard deviation (*SD*) of 1 (*z* scores) in the population without delirium (marker *z* scores). Thus, the marker *z* zcores for the ''No Delirium'' group are 0 with an *SD* of 1. This step allows the comparison of the individual inflammatory markers and the effect of inflammatory class (class *z* score). The class *z* score is the average of the marker *z* scores in the designated class. At baseline, there were no significant class *z* score differences between patients with delirium and matched controls.

IL = interleukin; IFN = interferon; TNF = tumor necrosis factor; TNF-R = tumor necrosis factor receptor; MIP = macrophage inflammatory protein; MIG = monokine induced by IFN-γ; RANTES = regulated upon activation, normal T-cell expressed, and secreted; CCL = C-C motif ligand; GM–CSF = granulocyte macrophage colony-stimulating factor; DR5 = death receptor 5; IP = interferon inducible protein.

relationships among different cytokines, we can better understand the cognitive effects of cytokines.

The inflammatory response is a highly complex and dynamic inter-related process during which any one cytokine can modulate a proinflammatory and/or antiinflammatory response depending on multiple clinical, physiologic, and immune considerations. Traditionally, studies of inflammation and geriatric syndromes measured individual inflammatory markers using enzyme linked immunosorbent assays (ELISAs) to measure a single protein in a blood sample (33,34). Although reliable, ELISAs are time-, labor-, and sample-consuming, resulting in increased cost and inefficient use of available blood samples. The dual-channel microsphere flow cytometer is a recent advance that allows assessment of multiple cytokines simultaneously by using a blood sample (100 microliters) comparable to that required for an individual ELISA. As with all new technologies, some cautionary notes have been raised. First, a reliable uniplex assay cannot be merely added into a reliable multiplex array without additional validation (35). Studies have demonstrated good correlations, but often poor concurrence of quantitative values between multiplex kits made by different manufacturers (36), as well as between multiplex kits and ELISA measurements (35). Because inflammatory markers are likely to be interdependent, grouping markers into classes for analysis increases statistical power and reduces the likelihood of finding a spurious association as a result of multiple testing. Nevertheless, carefully performed multiplex assays offer investigators opportunities for evaluating varied elements of inflammatory responses in the same individual over time.

Our study's approach to examine peripheral cytokine panels after cardiac surgery has both strengths and limitations. Its major advantage is the utilization of the microsphere flow cytometer, which allowed the measurement of 28 cytokines simultaneously. Thus, we were able to acquire a broad picture of changes in many cytokines and group them according to their roles within the overall inflammation response. However, the number of inflammatory markers measured increases the number of individuals needed to definitively draw conclusions about the relationship between delirium and inflammation. In this study, we focused on class effects because the large number of inflammatory markers limited the power to make definitive statements regarding

Table 3. Postoperative Change in Inflammatory Markers at 6 Hours (Intensive Care Unit [ICU]) and 4 Days (Postoperative Day [POD] 4)

| | ICU | | | | | POD 4 | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Marker *z* Scores | | Class *z* Scores | | | Marker *z* Scores | | Class *z* Scores | | |
| Class | No Delirium | Delirium | No Delirium | Delirium | *p* Value | No Delirium | Delirium | No Delirium | Delirium | *p* Value |
| Group 1: Inflammatory | | | | | | | | | | |
| IL-1β | 0.0 (1.0) | −0.5 (0.8) | 0.0 (1.0) | −0.1 (1.0) | .57 | 0.0 (1.0) | 0.1 (0.7) | 0.0 (1.0) | 0.0 (0.9) | .75 |
| IL-1Rα | 0.0 (1.0) | −0.1 (0.9) | | | | 0.0 (1.0) | −0.2 (0.8) | | | |
| IL-6 | 0.0 (1.0) | 0.5 (0.9) | | | | 0.0 (1.0) | 0.2 (1.0) | | | |
| IFN-α | 0.0 (1.0) | −0.4 (1.4) | | | | 0.0 (1.0) | −0.5 (1.0) | | | |
| TNF-α | 0.0 (1.0) | −0.5 (1.0) | | | | 0.0 (1.0) | −0.5 (0.8) | | | |
| TNF-R1 | 0.0 (1.0) | 0.3 (0.9) | | | | 0.0 (1.0) | 0.2 (1.1) | | | |
| TNF-R2 | 0.0 (1.0) | 0.1 (0.7) | | | | 0.0 (1.0) | 0.4 (0.8) | | | |
| Group 2: T-Helper 1 | | | | | | | | | | |
| IL-2 | 0.0 (1.0) | −0.2 (0.6) | 0.0 (1.0) | −0.2 (1.2) | .34 | 0.0 (1.0) | 0.0 (0.8) | 0.0 (1.0) | −0.3 (1.2) | .10 |
| IL-2R | 0.0 (1.0) | −0.7 (0.8) | | | | 0.0 (1.0) | −0.2 (0.8) | | | |
| IL-7 | 0.0 (1.0) | −0.2 (0.8) | | | | 0.0 (1.0) | −0.1 (0.8) | | | |
| IL-12p40_p70 | 0.0 (1.0) | −0.3 (0.9) | | | | 0.0 (1.0) | −0.6 (0.9) | | | |
| IL-15 | 0.0 (1.0) | −0.0 (1.0) | | | | 0.0 (1.0) | −0.3 (0.7) | | | |
| IFN-γ | 0.0 (1.0) | −0.4 (1.6) | | | | 0.0 (1.0) | −0.4 (1.1) | | | |
| IP-10 | 0.0 (1.0) | 0.8 (2.0) | | | | 0.0 (1.0) | −0.3 (2.4) | | | |
| Group 3: T-Helper 2 | | | | | | | | | | |
| IL-4 | 0.0 (1.0) | −0.4 (1.6) | 0.0 (1.0) | −0.1 (1.1) | .80 | 0.0 (1.0) | −0.4 (1.1) | 0.0 (1.0) | −0.3 (0.9) | .08 |
| IL-5 | 0.0 (1.0) | 0.1 (0.9) | | | | 0.0 (1.0) | −0.1 (0.8) | | | |
| IL-10 | 0.0 (1.0) | 0.4 (0.8) | | | | 0.0 (1.0) | −0.3 (0.8) | | | |
| IL-13 | 0.0 (1.0) | −0.3 (0.7) | | | | 0.0 (1.0) | −0.5 (0.9) | | | |
| Group 4: Chemokines | | | | | | | | | | |
| MIP-1α | 0.0 (1.0) | −0.1 (1.0) | 0.0 (1.0) | 0.3 (1.0)* | .04 | 0.0 (1.0) | −0.3 (0.9) | 0.0 (1.0) | 0.0 (1.1) | .68 |
| MIP-1β | 0.0 (1.0) | −0.1 (0.9) | | | | 0.0 (1.0) | −0.5 (1.0) | | | |
| MIG | 0.0 (1.0) | 0.5 (1.5) | | | | 0.0 (1.0) | −0.4 (1.1) | | | |
| Eotaxin | 0.0 (1.0) | 0.7 (0.6)* | | | | 0.0 (1.0) | 0.8 (0.8)* | | | |
| RANTES | 0.0 (1.0) | 0.3 (0.6) | | | | 0.0 (1.0) | 0.0 (1.2) | | | |
| CCL-2 | 0.0 (1.0) | 0.8 (0.8)* | | | | 0.0 (1.0) | 0.5 (0.9) | | | |
| Group 5: Lymphatic Chemokines | | | | | | | | | | |
| IL-8 | 0.0 (1.0) | 0.4 (0.8) | 0.0 (1.0) | 0.2 (0.8) | .44 | 0.0 (1.0) | 0.1 (1.3) | 0.0 (1.0) | 0.1 (1.1) | .93 |
| GM–CSF | 0.0 (1.0) | 0.0 (0.8) | | | | 0.0 (1.0) | 0.1 (0.9) | | | |
| Miscellaneous | | | | | | | | | | |
| IL-17 | 0.0 (1.0) | −0.6 (1.1) | | | | 0.0 (1.0) | −0.5 (0.8) | | | |
| DR5 | 0.0 (1.0) | −0.2 (0.7) | | | | 0.0 (1.0) | −0.4 (0.8) | | | |

*Notes*: The postoperative inflammatory marker concentration (ICU and POD 4) was divided by the preoperative concentration (fold change) and log normalized. To obtain the change *z* score, the population was standardized with a mean of 0 and standard deviation of 1 relative to the control group without delirium. We present the change *z* score for each inflammatory marker and class change *z* score, which represents the mean of the inflammatory marker class. Overall, circulating cytokine increased after surgery at both times. Among patients with delirium, there was a greater increase in chemokine class levels in the ICU and a trend toward decreased T-helper 1 and T-helper 2 class responses at POD 4.

*$p < .05$.

IL = interleukin; IFN = interferon; TNF = tumor necrosis factor; TNF-R = tumor necrosis factor receptor; MIP = macrophage inflammatory protein; MIG = monokine induced by IFN-γ; RANTES = regulated upon activation, normal T-cell expressed, and secreted; CCL = C-C motif ligand; GM–CSF = granulocyte macrophage colony stimulating factor; DR5 = death receptor 5; IP = interferon inducible protein.

any individual cytokine. Although our study would clearly benefit from more patients with and without delirium, this type of analysis is commonly used in the gene microarray literature, where the expression of thousands of different genes may be measured in a single individual to identify specific areas of interest for future research.

Our study is limited by the number of patients. The matched analysis provided a baseline level of risk adjustment. Inflammation and delirium can be affected by cognitive performance, age, and the surgical procedure. By matching for these factors, we were able to control some of the baseline risk. In future studies with more patients, it would be helpful to match by gender, comorbidity, and atherosclerosis burden, which can affect inflammation (9).

Because delirium is a disorder of the CNS, our assumption that serum levels of inflammatory markers parallel those in the CNS inflammatory response cannot be definitively proven at this time. However, we did find associations with the inflammatory response and delirium, suggesting that they may indeed be related. Moreover, because delirium may be associated with breakdown of the BBB, serum levels may indeed be correlated with CNS levels, although this hypothesis requires further evaluation.

## *Conclusion*

By using new technology to examine a large panel of cytokines, we found that early postoperative increased elevations of chemokines were associated with the development

of delirium after cardiac surgery. Although more research is needed to definitively establish a causative link, our evidence suggests that delirium may be at least in part mediated via chemokines, as these potent immune mediators attract inflammatory cells to the site of brain injury, may cause breakdown of the BBB, and may ultimately suppress T-cell–mediated immunity in patients who develop delirium in the postoperative period. These results represent a potential pathophysiologic mechanism of delirium after surgery. Moreover, after being replicated in a larger sample that will allow analysis of individual chemokines or cytokines, our findings may also offer a first potential target for mechanism-driven interventions for this common and morbid syndrome.

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CORRESPONDENCE

Address correspondence to James L. Rudolph, MD, SM, VABHS Geriatric Research, Education, and Clinical Center, 150 South Huntington Ave (JP-182), Boston, MA 02130. E-mail: jrudolph@partners.org

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