Visual Short-Term Memory Activation Patterns in Adult Survivors of Childhood Acute Lymphoblastic Leukemia

Aubree Boulet-Craig, MSc 1,2; Philippe Robaey, MD, PhD 1,2,3,4,5; Fanny Barlaam, PhD 1; Julie Laniel, BSc 1,2; Victor Oswald, MSc 1; Karim Jerbi, PhD 2; Serge Sultan, PhD 1,2; Laurence Affret-Bertout, MSc 1; Simon Drouin, PhD 1; Maja Krajinovic, PhD 1,6; Caroline Laverdière, MD, PhD 1,6; Daniel Sinnett, PhD 1,6; Pierre Jolicoeur, PhD 2; and Sarah Lippé, PhD 1,2

BACKGROUND: Acute lymphoblastic leukemia (ALL) is the most common childhood cancer. Treatments against ALL might lead to later cognitive effects and alterations in brain structure in survivors but to the authors’ knowledge the observed variability in the severity of neurocognitive deficits is not fully understood. The objective of the current study was to investigate abnormalities in visual short-term memory (VSTM) brain activation in survivors of childhood ALL using magnetoencephalography. METHODOLOGY: A VSTM task was completed by 40 survivors of ALL and 26 controls. VSTM capacity (Cowan K) and brain activation were assessed during the retention period of the task (400-1400 milliseconds) using a standard minimum norm source localization method. RESULTS: Performance (Cowan K) was found to be similar between survivors of ALL and controls. Atypical brain activation was found in survivors of ALL during the task, including overactivation of regions usually involved in VSTM (lateral occipital, precentral gyrus, and postcentral gyrus), recruitment of regions that typically are not involved in VSTM (superior/middle temporal gyrus and supramarginal gyrus), and lower activation of frontal brain regions (inferior frontal gyrus). These patterns of activation were modulated by the age at the time of cancer onset (P = .01) because activity was found to be reduced in participants who were younger at diagnosis. CONCLUSIONS: The results of the current study suggest a pattern of neural inefficiency and compensatory activity during VSTM in survivors of ALL. Cancer 2019;0:1-10. © 2019 American Cancer Society.

KEYWORDS: acute lymphoblastic leukemia, cognitive function, short-term memory, survivors of childhood cancer.

INTRODUCTION
Acute lymphoblastic leukemia (ALL) is the most common pediatric cancer, accounting for approximately 27% of cases in Western countries.1 Because of improvements in treatment protocols over the last several decades, the survival rate today exceeds 80%.2 Treatment of childhood ALL is central nervous system–directed and tracks malignant cells behind the blood-brain barrier. However, treatment can cause health outcomes in survivors, which persist throughout adulthood.3

Neurocognitive sequelae are common among survivors of childhood ALL. Approximately one-half of survivors of childhood cancer experience at least one clinically significant cognitive deficit after treatment,3 most commonly attentional and executive functioning deficits, including lowered attentional capacities,4 working memory,5 processing speed,6 flexibility, verbal fluency, and inhibition.8 Neurocognitive sequelae in survivors of ALL persist decades after treatment. Time elapsed increases the risk of presenting with impairment.8,9

The treatment of ALL can lead to smaller volumes of gray and white matter and alterations in white matter microstructure in cortical and subcortical structures,10-14 often associated with decreased cognitive performance.10,11,15

Even when considering the impact of risk factors such as sex, age at diagnosis, treatment dosage, and cranial radiotherapy (CRT) on cognition and brain structure, the causes of and variability in neurocognitive deficits among survivors of ALL are not fully appreciated.2,8,10 Growing evidence has suggested that investigating the neural basis of neurocognitive impairment could lead to a better understanding of deficits and help to create cognitive remediation interventions.17

Recent studies have demonstrated brain processes particularities in survivors of ALL, such as stronger activation than...
matched controls in frontal regions and cingulate cortex in working memory tasks.\textsuperscript{17,18} Stronger activations were associated with higher doses of methotrexate (MTX), a chemotherapeutic agent known for its neurotoxic properties.\textsuperscript{7,17} To our knowledge, these studies have investigated children and adolescents only.\textsuperscript{7,17,18} Investigating adult survivors may demonstrate the evolution of neuronal functioning across lifespan in that population.

Visual short-term memory (VSTM), an active cognitive system allowing for the storage and temporary maintenance of a limited amount of visual information, frequently is lowered in survivors of ALL. It activates a network involving the intraparietal and intracerebral sulci for maintenance of information.\textsuperscript{19-22} In fact, occipito-parietal activity increases in amplitude with the amount of information retained in VSTM and reaches a plateau when the capacity of VSTM is at its maximum. VSTM also activates the precuneus, posterior parietal regions, lateral occipital regions, prefrontal cortex, and superior and middle frontal gyri.\textsuperscript{19,20,23,24} Investigating VSTM and its neuronal support in survivors of ALL will increase our understanding of the consequences of ALL treatment.

The objective of the current study was to investigate abnormalities in neural correlates of VSTM in a cohort of adult survivors of childhood ALL using magnetoencephalography (MEG). We verified the impact of the most common neurocognitive impairment risk factors on cerebral activity and compared different subgroups of survivors as a function of their performance in a battery assessing cognitive domains sensitive to cancer treatments.\textsuperscript{25} We hypothesized that survivors of ALL would present a lower VSTM capacity (Cowan’s K)\textsuperscript{26} and stronger activation patterns compared with controls during the retention period (400-1400 milliseconds) of the task, mainly in parieto-occipital and frontal regions.

MATERIALS AND METHODS

Participants

As part of the PETALE study,\textsuperscript{27} we recruited 50 adults who were survivors of childhood ALL; were at least 5 years after diagnosis; were treated with 1985 to 2005 Dana-Farber Cancer Institute ALL protocols (DFCI-ALL 87-01 to 2005-01); had no history of refractory ALL, disease recurrence, or hematopoietic stem cell transplantation; and were diagnosed between ages birth to 17 years (see Supporting Table 1). We also recruited 29 age-matched and sex-matched healthy subjects with no reported history of cancer or neurological or psychiatric disorders. Ten survivors and 3 controls were removed from the analysis (Fig. 1).\textsuperscript{27,28} Participants had normal color vision. The study was approved by the institutional review board at the Sainte-Justine University Health Center. All participants provided written informed consent. Including neuropsychological assessment (see Supporting Table 2), the testing lasted approximately 5 hours and was completed in 1 day. Survivors were classified in 3 distinct performance groups—low performance (LP), average performance (AP), and high performance (HP)—based on their results on the digit span verbal fluency grooved pegboard trail making test (DIVERGT) screening procedure, a valid index of neurocognitive impairment in ALL survivors (see Boulet-Craig et al for comprehensive information regarding the classification process\textsuperscript{28}).

MEG Procedure: VSTM Task

Brain activity during the task was recorded using a CTF-VSM whole-head, 275-sensor MEG system in a magnetically shielded room. The sampling rate was 1200 Hz. No filter was applied during recordings. We recorded 29 reference channels to compute a third-order gradiometer noise reduction. Electrocardiography was recorded to monitor pulse (electrodes were placed on each shoulder). Horizontal (electrodes at the right and left canthi) and vertical (electrodes above and below the left eye) electrooculography also was recorded to monitor eye movements and eye blinks. Stimuli (colored disks) were back projected onto a translucent screen located approximately 75 cm from and facing the participants.

Each trial started with a fixation cross that was presented for 400 to 500 milliseconds (random jitter) and centered within the display (Fig. 2). Two arrowheads then appeared above and below the fixation cross (800 milliseconds), pointing left or right to indicate the visual hemifield relevant for that trial, and were followed by a blank interval (200 milliseconds). One or three easily distinguishable colored disks (blue, green, orange, pink, or brown) then appeared (200 milliseconds) on each side of the screen. Load 1 and load 3 were chosen to ensure an adequate performance in both participants. Colored disks randomly were positioned on an invisible 3×3 grid within each hemifield. The luminance of the colors was calibrated to be approximately equiluminant (Minolta CS100 chroma meter), and no color was shown more than once in the same hemifield. After the retention period (1800 milliseconds), one colored disk in each hemifield within the same location as one of the previously presented disks was presented (1500 milliseconds). In approximately one-half of the trials, the color of the test disk matched the color in the memory array (same trial). Otherwise, one of the
other colors was shown (different trial). Participants had to decide whether the test disk was identical or different from the disk in the memory array at that location (hemifield indicated by arrows). Subjects pressed a button with their index finger to indicate a “same” response and a button with their middle finger to indicate a “different” response. Participants answered on a response pad with their dominant hand (56 with their right hand and 10
with their left hand). Feedback was presented visually at the center of the screen after the responses and indicated whether the response was correct, incorrect, or not produced in the allotted time (1500 milliseconds).

Complete settings of the task (ie, blank interval before arrows, memory load, hemifield, same and different trials) were distributed equally, individually by participant, and were presented in a different random order. The experiment had 4 blocks of 80 trials. Before the recordings, the subjects performed 3 practice blocks (20 trials each) outside the magnetically shielded room to ensure they understood the task and performed above chance.

**MEG Analysis**

Data analyses were performed using Brainstorm software. Raw data first was notch filtered (60 Hz, 120 Hz, and 180 Hz). Then, Signal-Space Projection (SSP) was used to correct the MEG signals contaminated by ocular, cardiac, and respiratory artifacts by identifying and removing the components that best captured these artifacts. Data then were segmented into trials from -200 milliseconds to 2500 milliseconds relative to the onset of the memory array for each condition, and were baseline corrected using the mean prestimulus onset activity. Supplementary artifact rejection through visual inspection was performed to remove trials containing large artefactual deflections (an average of 4.5% of trials were removed in controls and 5.1% in survivors). Disregarded trials were found to be distributed evenly across conditions. Correct and incorrect response trials were included in the analysis and low-pass filtered at 40 Hz. Data for each of the 4 main conditions (encode from left or right, 1 or 3 disks) were averaged for each participant in a 400-millisecond to 1400-millisecond window to produce event-related magnetic fields. This specific window was chosen based on previous work regarding VSTM retention activity.

To compute source localization analyses using a standard minimum-norm method, the T1-weighted brain volumes were acquired previously from each participant (GE Discovery MR-750, 3-Tesla) and were used to create a cortical surface model. Using the overlapping spheres forward model, we computed the minimum-norm estimate using a constrained dipolar orientation model (15,000 dipoles), with a signal-to-noise ratio of 3 and a depth weighting of 0.5. The noise covariance matrix for each participant was estimated from a 2-minute empty-room recording performed earlier on the same day. Source images were created based on event-related magnetic fields. Resulting images were interpolated back on the Brainstorm default anatomy and spatially smoothed (6 mm). Conditions (right and left hemifield) then were averaged at the source level in absolute values to produce mean activations by memory load (load 1 and load 3). Bilateral activation was used in analyses because previous work demonstrated that VSTM activates a large bilateral load-related response and is likely more reliable than statistics based on contralateral minus ipsilateral differences. Source images for the load 1 condition then were subtracted from the load 3 condition. This operation was performed to minimize brain activity related to perceptual processes (eg, the sudden onset of the test disk) and to isolate memory retention processes, which we specifically wanted to investigate.

**Statistical Analysis**

A Monte Carlo permutation test using 2000 randomizations was computed to assess differences between survivors and controls regarding the load 3 minus load 1 difference (statistical threshold of $P < .05$). To concentrate our analyses on the most prominent brain regions, cortical areas of a minimum of 20 vertices were considered in the results. An exploratory multivariate analysis of variance was used to assess differences in brain activation between performance groups (LP, AP, HP, and control groups). A multivariate analysis of covariance was performed to verify any relation between risk factors (ie, sex, CRT, age at diagnosis, cumulative doses of corticosteroids, doses of intrathecal MTX, and doses of intravenous MTX) and brain activation during the retention period.
Cowan K
The average number of disks maintained in VSTM by each subject was estimated using the Cowan K formula, which is an index of VSTM capacity:

\[ K = (p(H) - p(FA)) \times N, \]

in which \( p(H) \) is the percentage of hits (ie, answering the same when the color was the same), \( p(FA) \) is the percentage of false alarms (ie, answering the same when the color was different), and \( N \) is the number of items to be remembered. The rejected trials were disregarded from calculation of the Cowan K. Differences between controls and survivors were assessed using the Mann-Whitney \( U \) test for independent samples. A Kruskal-Wallis test for independent samples then was conducted to assess differences between performance groups. Spearman correlations and Mann-Whitney \( U \) tests for independent samples were used to verify the relation between performance (Cowan K) and risk factors. Nonparametric tests were chosen because the variable “K” did not follow a normal distribution.

RESULTS
Behavior
Analysis using the Mann-Whitney \( U \) test demonstrated that performance (see Supporting Table 3) did not differ significantly between survivors and controls at load 1 (controls, Median of 0.89 and survivors, Median of 0.88 \([U = 463; P = .454]\)) nor at load 3 (controls, Median of 1.54 and survivors, Median of 1.56 \([U = 460.5; P = .435]\)). No differences in VSTM performance were detected between performance groups (LP, AP, HP, and control groups) and no relation was found between K and any of the risk factors.

Brain Activation
Figure 3 presents the mean activation during the retention period for each group. Both groups demonstrated a distributed pattern of neuronal activity, including parietal regions such as the intraparietal sulcus and occipital and prefrontal regions.

Brain Activation Differences Between Groups
The Monte Carlo permutation test demonstrated differences in brain activation related to VSTM load effects during retention between survivors and controls (Fig. 4). Survivors presented with greater activation in multiple regions (left lateral occipital, left precentral, postcentral gyrus, central sulcus, left supramarginal gyrus, left superior temporal gyrus, left transverse temporal gyrus, right central sulcus, and right middle temporal gyrus \([P < .05]\)). Controls only exhibited greater activation in 1 brain region, the left inferior frontal region \((P < .05)\) (Table 1).

Exploratory multivariate analysis of variance demonstrated performance group differences with regard to brain activation \((F(10,53) = 1.7; P = .016; \eta^2_p = 0.24)\). Post hoc analysis using the Tukey procedure demonstrated that
controls had smaller amplitudes compared with HP survivors in the left precentral/central ($P = .022$), left postcentral ($P = .013$), left supramarginal ($P < .0001$), and left superior temporal ($P < .0001$) regions and smaller amplitudes compared with AP survivors in the left superior temporal region ($P = .015$), right middle temporal region ($P = .016$), and right central sulcus ($P = .016$). LP survivors had higher amplitudes compared with controls in the inferior frontal region ($P = .027$).

Multivariate analysis of covariance demonstrated that smaller amplitudes in multiple regions were related to younger age at diagnosis ($F(10.22) = 3.2; P = .01; \eta^2_p = 0.60$), including left precentral ($F(1.31) = 6.1; P = .02; \eta^2_p = 0.16$), left postcentral ($F(1.31) = 5.9; P = .021; \eta^2_p = 0.16$), left supramarginal ($F(1.31) = 6.7; P = .015; \eta^2_p = 0.18$) (Fig. 5), and left superior temporal ($F(1.31) = 4.6; P = .04; \eta^2_p = 0.13$) activation.

**DISCUSSION**

We examined the capacity and neural substrates of VSTM in a cohort of adult survivors of childhood ALL. We found that survivors presented with VSTM load–related brain activation differences in several regions during the task, despite a lack of difference in capacity as estimated by the Cowan K. Specifically, we found a widespread increase in activation during retention in the left lateral occipital cortex.

**TABLE 1. Activation Differences Between Groups by Brain Region**

<table>
<thead>
<tr>
<th>Brain Region</th>
<th>No. of Vertices</th>
<th>X</th>
<th>Y</th>
<th>t Value</th>
<th>Activation Difference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Left lateral occipital</td>
<td>33</td>
<td>−101.2</td>
<td>−4.7</td>
<td>3.6</td>
<td>S&gt;C</td>
</tr>
<tr>
<td>Left precentral/central sulcus</td>
<td>43</td>
<td>−10.2</td>
<td>44.1</td>
<td>3.0</td>
<td>S&gt;C</td>
</tr>
<tr>
<td>Left postcentral</td>
<td>29</td>
<td>−21.1</td>
<td>40.8</td>
<td>2.3</td>
<td>HP&gt;C</td>
</tr>
<tr>
<td>Left supramarginal</td>
<td>75</td>
<td>−21.4</td>
<td>14.8</td>
<td>3.7</td>
<td>S&gt;C</td>
</tr>
<tr>
<td>Left superior temporal</td>
<td>22</td>
<td>−23.6</td>
<td>−2.0</td>
<td>4.1</td>
<td>S&gt;C</td>
</tr>
<tr>
<td>Left transverse temporal</td>
<td>45</td>
<td>−17.9</td>
<td>8.9</td>
<td>2.6</td>
<td>S&gt;C</td>
</tr>
<tr>
<td>Left inferior frontal</td>
<td>24</td>
<td>36.1</td>
<td>−16.6</td>
<td>2.4</td>
<td>C&gt;S; C&gt;LP</td>
</tr>
<tr>
<td>Right central sulcus</td>
<td>26</td>
<td>−7.7</td>
<td>32.4</td>
<td>2.4</td>
<td>S&gt;C; AP&gt;C</td>
</tr>
<tr>
<td>Right middle temporal</td>
<td>32</td>
<td>−40.8</td>
<td>7.6</td>
<td>2.1</td>
<td>S&gt;C; AP&gt;C</td>
</tr>
</tbody>
</table>

Abbreviations: AP, average performance group; C, controls; HP, high performance group; LP, low performance group; MNI, Montreal Neurological Institute and Hospital; S, survivors.
The t-value is the Monte-Carlo Permutation T-Test result.
occipital, left precentral/central, postcentral, supramarginal, left superior and transverse temporal, right central, and middle temporal regions. We also found decreased activity in the left inferior frontal region. These results can be understood in light of the Compensation-Related Utilization of Neural Circuits Hypothesis (CRUNCH). This model proposes that more cortical regions are activated as task load increases. If brain processing is less efficient in survivors, it may be necessary to recruit more regions of the brain for the same load increase compared with controls. Neural efficiency is the amount of performance-relevant computations per unit of synaptic activity that we operationalized as MEG signal load–related change. We compared the performance-relevant computational power by using subgroups based on the DIVERGT battery, which is a sensitive index of performance decline in survivors of ALL. We found one region in the inferior frontal cortex for which survivors of ALL demonstrated a smaller load–related activation compared with controls. Exploratory analyses revealed that only the survivors who performed poorly on the DIVERGT were less activated than the controls. Using a visual n-back task with letters, survivors demonstrated significantly greater activation in the dorsolateral and ventrolateral prefrontal cortex compared with controls. Persistent activity in the prefrontal cortex (especially dorsolateral) in working memory is well established. The decrease in brain activity we observed in LP survivors suggests a reduction in neural efficiency.

We found increased activity in the left lateral occipital region. In VSTM, the sustained MEG activity observed during retention in healthy controls is generated by a network of cortical sources that includes bilateral parietal loci, most likely the intraparietal/intraoccipital cortex, and contralateral parietal sources. Activity in visual brain areas specifically was found to encode remembered visual content across delays. This increase in left lateral occipital activity observed in survivors does not appear to be as beneficial for any performance subgroups. Therefore, increasing the activity in this region could reflect a response to a loss of specialization (dedifferentiation).

We found an increase in activity in adjacent brain regions. Exploratory analyses demonstrated that these increased activations were repeatedly significant for HP survivors: increased activity was found in the left precentral, central, and postcentral regions. These regions also were found to be activated in a study using a spatial working memory task. The activity in the superior parietal gyrus extended anteriorly toward the postcentral gyrus and the frontal activation extended from the superior and middle frontal gyri to the precentral gyrus. Both postcentral and precentral activities were modulated by the memory load, which may be a sign of motor representation for future actions.

The increase in activity in the left supramarginal regions also was significant for HP survivors. Neural activity in the posterior region of the right supramarginal gyrus, lying along the intraparietal sulcus, is critical for...
mediating both spatial working memory and shifts in spatial attention. In normal controls, delays of >1 second in a visual working memory task favor the verbal/abstract recoding of visual material. Accordingly, when visual working memory is insufficient to support performance, damage to medial temporal lobe structures impairs performance. One intriguing possibility would be that subjects would recode the visual information using a long-term verbal memory process (eg, by verbally rehearsing the colors during the delay). In survivors of ALL, the maintenance of visual information may be impaired (as suggested by increased activity in occipital regions without performance benefits), and compensatory mechanisms could lead to the additional use of linguistic encoding. This hypothesis of the verbal recoding of visual information is compatible with the observed increased activity in the left superior temporal regions in AP and HP survivors.

We also found increased activity in the right hemisphere. The increased activities in the right central and right middle temporal regions were significant for AP survivors. The activation in the right hemisphere is more in keeping with the visuospatial nature of the task, whereas the previous activity differences mainly were left lateralized. This lack of lateralization may reflect an hemispheric asymmetry reduction that is found in older age: older adults activate homologous brain regions in the opposite hemisphere whereas the activity remains lateralized in younger adults (the HAROLD effect). The HAROLD effect can be considered to be a special manifestation of age-related compensatory processes in the more general CRUNCH model. The fact that homologous brain regions are activated in both hemispheres in survivors without deficits in DIVERGT also may indicate an asymmetrical reduction in survivors.

Alternatively, it is possible that engaging additional regions represents unselective recruitment or dedifferentiation of function. However, the increase in activity was observed among survivors with average to high DIVERGT performance. Dedifferentiation is expected to be associated with a decrease in performance. Moreover, the increased load-related activity at the level of the left supramarginal and left superior temporal regions was correlated with younger age at diagnosis, which is a well-known risk factor for cognitive deficits. Figure 5 shows that survivors diagnosed after the age of 5 years recruited these regions, whereas those diagnosed at age <5 years recruited them inconsistently. In another study using a visual continuous performance test (CPT) task, younger age at diagnosis was similarly associated with lower brain activation in the bilateral superior temporal and parietal cortices.

The theoretical models we used were proposed in studies of the aging brain. The similarities observed between brain functions and structures after ALL treatment and in aging are striking. For certain delayed memory domains, survivors of ALL function at a cognitive age of 2 and 3 decades beyond their chronological age. Healthy aging is accompanied by a decrease in white matter integrity that, in turn, contributes to the different forms of cognitive decline. Overrecruitment among older adults may compensate for white matter decline. Adult survivors of ALL also demonstrated differences in fractional anisotropy, especially after combined treatment with irradiation and chemotherapy.

**Study Limitations**

In the current study, the simplicity of the task could be considered as a limitation. We decided to use load 1 and load 3 in a time-efficient perspective and to avoid overloading the survivors because high loads tend to be difficult, even in healthy subjects. Nevertheless, higher loads may have provided further understanding of between-group neural modulation differences. In addition, a large percentage of the survivors were treated with CRT (75%), which has a strong impact on brain development. The current study findings may not be readily applicable in survivors treated without CRT. Last, the average intelligence quotient (IQ) in the control group in the current study was slightly above average (106.5) and, to a certain extent, may have impacted group differences.

**Conclusions**

The results of the current study suggest a pattern of neural inefficiency and compensatory activity for impaired functioning in survivors of ALL, similar to what is observed in older adults. These results may guide cognitive remediation programs promoting more efficient brain activation.

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**AUTHOR CONTRIBUTIONS**

Aubree Boulet-Craig: Conceptualization, data curation, formal analysis, investigation, methodology, project administration, writing–original draft,
and writing–review and editing. Philippe Robaey: Conceptualization, methodology, project administration, and writing–review and editing.

Fanny Barlaam: Formal analysis, validation, and writing–review and editing. Julie Laniel: Investigation and data curation.

Laurence Affret-Bertout: Conceptualization, supervision, and project administration. Simon Drouin: Investigation and project administration. Majka Krajnovic: Conceptualization, funding acquisition, supervision, and project administration.

Caroline Laverdier: Conceptualization, funding acquisition, supervision, and project administration. Daniel Sinnett: Conceptualization, funding acquisition, supervision, and project administration.

Victor Oswald: Conceptualization, supervision, and project administration. Laurence Affret-Bertout: Conceptualization, supervision, and project administration.

Sarah Lippé: Conceptualization, methodology, supervision, project administration, and writing–review and editing.

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