Accelerated Aging, Decreased White Matter Integrity, and Associated Neuropsychological Dysfunction 25 Years After Pediatric Lymphoid Malignancies

Ilse Schuitema, Sabine Deprez, Wim Van Hecke, Marita Daams, Anne Uyttebroeck, Stefan Sinaert, Frederik Barkhof, Elina van Dulmen-den Broeder, Helena J. van der Pal, Cor van den Bos, Anjo J.P. Veerman, and Leo M.J. de Sonneveld

See accompanying editorial doi: 10.1200/JCO.2013.50.8879

ABSTRACT

Purpose
CNS-directed chemotherapy (CT) and cranial radiotherapy (CRT) for childhood acute lymphoblastic leukemia or lymphoma have various neurotoxic properties. This study aimed to assess their impact on the maturing brain 20 to 30 years after diagnosis, providing a much stronger perspective on long-term quality of life than previous studies.

Patients and Methods
Ninety-three patients treated between 1978 and 1990 at various intensities, with and without CRT, and 49 healthy controls were assessed with magnetic resonance diffusion tensor imaging (DTI) and neuropsychological tests. Differences in fractional anisotropy (FA)—a DTI measure describing white matter (WM) microstructure—were analyzed by using whole brain voxel-based analysis.

Results
CRT-treated survivors demonstrated significantly decreased FA compared with controls in frontal, parietal, and temporal WM tracts. Trends for lower FA were seen in the CT-treated survivors. Decreases in FA correlated well with neuropsychological dysfunction. In contrast to the CT group and controls, the CRT group showed a steep decline of FA with age at assessment. Younger age at cranial irradiation and higher dosage were associated with worse outcome of WM integrity.

Conclusion
CRT-treated survivors show decreased WM integrity reflected by significantly decreased FA and associated neuropsychological dysfunction 25 years after treatment, although effects of CT alone seem mild. Accelerated aging of the brain and increased risk of early onset dementia are suspected after CRT, but not after CT.

J Clin Oncol 31. © 2013 by American Society of Clinical Oncology

INTRODUCTION

Prophylaxis to prevent meningeal relapse after childhood acute lymphoblastic leukemia (ALL) or lymphoma used to consist of both intrathecal (IT) chemotherapy (CT) and cranial radiotherapy (CRT). The neurotoxic adverse effects of prophylactic CRT became apparent during the 1980s and are well documented.1 Around that time, CRT was mostly abolished and replaced with more intensive IT CT.2-4 Knowledge of late effects is mostly restricted to the first decade after treatment,5-8 but these effects may have an impact on quality of life for many decades because both long-term survival rates and general life expectancy keep increasing. This study provides information about the long-term impact of CT and CRT on the maturing brain 20 to 30 years after diagnosis by using magnetic resonance (MR) diffusion tensor imaging (DTI) and neuropsychological assessment. The outcomes will help to better understand these survivors’ current needs and could aid in anticipating late effects of CT and of CRT that is currently applied for brain tumors and is still used for ALL in some countries.

There are many reports on late neurocognitive deficits after CT alone, particularly in the domain of executive functions, although these are relatively subtle compared with the effects of CRT.9-12 Sequelae seem more severe after high-risk treatment.13 Advanced neuroimaging techniques allow us to investigate possible neural substrates of these cognitive sequelae. An MR voxel-based morphometry study by Carey et al12 linked reduction of white matter (WM) volumes within the right frontal lobe...
to decreased cognitive functioning 10 years after treatment with CT only. The effects of CT beyond 10 years after treatment are sparsely studied. Porto et al13 studied 10 female ALL survivors, on average 15 years post-treatment with CT only, and found distributed reductions of gray matter and WM concentrations and a trend for decreased WM integrity. Research by Koppelmans et al indicates that cognitive effects of adjuvant CT for breast cancer can persist beyond 20 years after treatment, but it remains unclear how this translates to IT CT in childhood.

Late neurotoxic effects of CRT include an increased risk of neurologic complications such as vascular malformations, secondary neoplasms, and focal necrosis.16–18 In the cognitive domain, deficits in memory, information processing speed, and attention have been reported.19–24 WM volume reduction is more apparent after CRT than after CT and correlates significantly with cognitive impairment.6

On a microstructural level, DTI is able to quantify WM organization by assessing the restriction of randomly moving water molecules. The degree of directional preference of diffusion is quantified by the DTI parameter fractional anisotropy (FA). Damage to WM microstructures will result in lower FA because of relatively more diffusion of water perpendicular to the fiber orientation.25 Significant differences in FA between cancer survivors (up to 10 years after treatment) and controls have been reported and linked to cognitive impairment.26–30 Dellani et al demonstrated WM alterations in 13 CRT-treated ALL survivors 16 to 28 years after treatment. Porto et al demonstrated reduced FA after CRT in 11 males 15 years after treatment for childhood ALL. Associations with cognition remained unaddressed by Dellani and Porto, and they were both unable to demonstrate a relation between FA and age at diagnosis.

Younger age at treatment with CRT has been associated with more severe WM lesions and worse neurocognitive outcome.32–35 Progression of radiation-induced WM changes has also been reported.36,37 Prophylactic CRT (20-30 Gy) for small-cell lung cancer in adults has been associated with early-onset dementia.38–41 In general, theories are emerging that cancer and cancer treatment may cause accelerated aging of the brain and of cells in general, even after CT without CRT.32,42 This study investigated WM changes and associated neuropsychological dysfunction 25 years after treatment. The hypothesis of accelerated WM decay will be addressed by comparing the correlations of FA and age between survivors treated with and without CRT and controls. In addition, the relation between age at diagnosis and vulnerability to neurotoxicity of treatment will be explored.

**PATIENTS AND METHODS**

**Patients**

We identified 285 survivors of ALL or lymphoma from patient records of the Vrije Universiteit University Medical Center, the Academic Medical Center Amsterdam in the Netherlands, and the University Hospitals Leuven in Belgium. Dates of diagnoses after 1978 were included, and time since diagnosis had to be at least 18 years. After exclusion of 42 survivors (Appendix Fig A1; online only), 243 eligible survivors were contacted. Ninety-six survivors were willing to participate and were asked to recruit a control (sibling, partner, or friend; n = 49). Assessments took place between 2007 and 2011.

Survivors were treated according to Berlin-Frankfurt-Munster (BFM)–based protocols with a duration of approximately 2 years. Between 1979 and 1983, standard-risk CRT patients were treated according to the Dutch Childhood Leukemia Study Group (DCLSG) protocol ALL-5 or the Richem protocol, both characterized by CRT (15 to 25 Gy) in addition to five to seven IT injections of 12 to 12.5 mg methotrexate (MTX). This cohort included 24 survivors treated according to a standard-risk CRT protocol. We included seven high-risk (HR) patients from this period, treated with additional customized high-dose MTX intravenously (IV). Around 1983, CRT was abolished and standard-risk patients were treated according to DCLSG protocol ALL-6 (13 × 12 mg MTX IT and 6 g/m² MTX IV) or European Organisation for Research and Treatment of Cancer (EORTC) Trial 58831 (6 × 12 mg MTX IT and 2 g/m² MTX IV), of which 29 patients were included. HR patients were treated with customized protocols based on either EORTC Trial 58832 (8 × 12 mg MTX IT and 10 g/m² MTX IV), the BACOP [bleomycin, doxorubicin, cyclophosphamide, vincristine, and prednisone] protocol (customized dose of MTX IV), or ALL-6 (customized additional MTX IV) without CRT (HR CT, n = 20).43 Administration of MTX IV was always followed by leucovorin (12 to 15 mg/m² every 6 hours until serum levels of MTX had dropped below 10–7 mol/L). Thirteen patients were treated for relapse after standard-risk or HR treatment, seven of whom were irradiated only during relapse and six of whom were irradiated at both initial occurrence and relapse. Group means of dosages were calculated excluding missing data. Dosages of CRT were available for all patients, but dosages of MTX IV were missing for three survivors in the CRT group and dosages of MTX IT were missing for one survivor in the CT group. Three irradiated survivors were excluded because meningiomas were discovered during assessment. Survivors were grouped into irradiated (n = 44) and nonirradiated (n = 49), and dose-dependent effects of CRT, MTX IV, and MTX IT were studied. The ethical principles of the Helsinki Declaration were followed and approval was obtained from the local ethical committees.

**Acquisition Details for MR Imaging**

Patients were scanned on a 1.5T Sonata system (Siemens, Erlangen, Germany), including a T1-weighted 3D gradient sequence (TR, 2,700 ms; TE, 5.17 ms; flip angle, 8 degrees; 160 coronal slices; voxel size, 1 × 1 × 1.5 μL). DTI was measured by using a 9-minute echo planar imaging sequence (TR, 5,800 ms; TE, 86 ms; voxel size, 2 mm isotropic; 59 slices; acquisition matrix, 128 × 128 mm; FOV, 256 mm; 60 directions [b value, 700 s/mm²]; 10 b0 images).46

**DTI Processing and Statistical Analysis**

DTI preprocessing was done by using ExploreDTI consisting of motion and distortion correction with reorientation of the b-matrix and an iterative weighted nonlinear tensor estimation process to generate FA maps.47 Individual DTI data sets were nonrigidly registered to a population-based DTI atlas generated from DTI images from all patients.48–50 Finally, the resulting images were smoothed with a 3D-Gaussian kernel of full width at half maximum of 6 mm.

Statistical Parametric Mapping 8 (SPM8) whole-brain voxel-based analysis of variance was performed to assess differences in FA between the different groups.51 Age at assessment (AaA) was used as covariate. A WM mask was applied to limit the analysis to WM voxels only. The resulting statistical parametric maps were thresholded at the voxel-level P < .001. Only clusters significant at the family-wise error P < .05 level corrected for multiple comparisons were retained.

**Neuropsychological Assessment and Statistical Analysis**

The Amsterdam Neuropsychological Tasks (ANT) program was used to assess executive functions.52 The computerized ANT provides standardized assessments and automated recordings of speed and accuracy of information processing, attention processes, and working memory (see Appendix, online only). Intelligence quotient was estimated by using a four subset short-form of the Wechsler Adult Intelligence Scale Revised (WAIS-R III).53 Differences between groups were tested by using analysis of variance and simple contrasts with controls as the reference group. AaA was used as a covariate. Task parameters that discriminated between survivors and controls were selected for correlation analyses with FA.

**Correlations Between FA and Discriminative Neuropsychological Parameters**

To study general associations between FA and cognition, a whole-brain voxel-based correlation analysis was conducted that included all survivors and controls, with FA as dependent variable, the selected neuropsychological test...
### Table 1. Characteristics and Neuropsychological Task Performance of the Included Patients

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Controls (n = 49)</th>
<th>CRT (n = 44)</th>
<th>CT (n = 49)</th>
<th>ANOVA (between-patients effects)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>% M SD</td>
<td>% M SD M SD</td>
<td>% M SD M SD</td>
<td>% M SD M SD M SD M SD M SD M SD</td>
</tr>
<tr>
<td>Males</td>
<td>42.9 52.3</td>
<td>57.1</td>
<td>26.7 5.1</td>
<td>24.5 2.3</td>
</tr>
<tr>
<td>Age at assessment (years)</td>
<td>26.5 5.9</td>
<td>31.2 4.8</td>
<td>31.9 4.3</td>
<td>32.6 4.0</td>
</tr>
<tr>
<td>Age at diagnosis (years)</td>
<td>N/A</td>
<td>5.7 3.7</td>
<td>5.4 3.4</td>
<td>7.1 4.6</td>
</tr>
<tr>
<td>Time since diagnosis (years)</td>
<td>N/A</td>
<td>25.4 3.2</td>
<td>26.4 2.7</td>
<td>25.4 3.2</td>
</tr>
<tr>
<td>CRT (Gy)</td>
<td>N/A 22.591</td>
<td>6.991 20.83</td>
<td>6.991 20.83</td>
<td>24.43 3.51</td>
</tr>
<tr>
<td>Cumulative dose</td>
<td>N/A</td>
<td>24.43 3.51</td>
<td>24.43 3.51</td>
<td>24.85 11.52</td>
</tr>
<tr>
<td>Cumulative dose</td>
<td>N/A</td>
<td>22.591 6.991</td>
<td>22.591 6.991</td>
<td>24.43 3.51</td>
</tr>
<tr>
<td>MTX IV (mg/m²)</td>
<td>N/A 15,853.7</td>
<td>30,498.0</td>
<td>15,853.7</td>
<td>30,498.0</td>
</tr>
<tr>
<td>Cumulative dose</td>
<td>N/A</td>
<td>15,853.7</td>
<td>30,498.0</td>
<td>15,853.7</td>
</tr>
<tr>
<td>MTX IT (mg)</td>
<td>N/A 111.9</td>
<td>95.7 20.2</td>
<td>111.9</td>
<td>95.7 20.2</td>
</tr>
<tr>
<td>Estimated IQ</td>
<td>107.4 19.4</td>
<td>96.9 20.2</td>
<td>107.4</td>
<td>19.4 20.2</td>
</tr>
<tr>
<td>Visuomotor accuracy</td>
<td>3.3 0.6</td>
<td>3.9 0.9</td>
<td>3.3 0.6</td>
<td>3.9 0.9</td>
</tr>
<tr>
<td>Visuomotor stability</td>
<td>2.1 0.7</td>
<td>2.6 0.9</td>
<td>2.1 0.5</td>
<td>2.6 0.9</td>
</tr>
<tr>
<td>Sustained attention</td>
<td>8.2 1.4</td>
<td>9.5 2.3</td>
<td>8.2 1.4</td>
<td>9.5 2.3</td>
</tr>
<tr>
<td>Visuospatial sequencing</td>
<td>3.2 3.2</td>
<td>8.2 5.7</td>
<td>3.2 3.2</td>
<td>8.2 5.7</td>
</tr>
</tbody>
</table>

**NOTE.** Larger values for visuomotor accuracy, visuomotor stability, sustained attention work pace, and visuospatial sequencing denote worse performance.

**Abbreviations:** AaA, age at assessment; AaD, age at diagnosis; ANOVA, analysis of variance; CRT, cranial radiotherapy; CT, chemotherapy; IQ, intelligence quotient; IT, intrathecally; IV, intravenously; M, mean; MTX, methotrexate; N/A, not applicable. Significant contrast result (simple contrast v controls).
scores as regressor, and age as covariate. Pearson’s r was calculated within clusters showing significant correlations.

**Age at Diagnosis and Time Since Diagnosis**

The separate effects of AaA and age at diagnosis (AaD) on WM organization were studied with a whole-brain voxel-based correlation analysis, controlling the correlation between FA and AaA for AaD, and between FA and AaD for AaA. Only participants between age 20 and 40 years were included, because within a healthy population, FA values are minimally affected by normal maturation and aging in this age range (CRT, n = 30; CT, n = 45; controls, n = 44).63 Relapse patients needed to be excluded because of their double AaD. Subsequently, the relationship between FA and AaA and AaD in CRT-treated survivors was studied with linear regression models within significant clusters also relevant to cognition.64

**Dosage Correlations**

The potential influence of therapy dosage on FA was explored with a voxel-based correlation analysis between FA and cumulative doses of CRT and doses of MTX IV and/or MTX IT within the CRT and CT groups, respectively. AaA and AaD were included as covariates.

**RESULTS**

**Patient Demographics and Neuropsychological Function**

Table 1 provides a summary of participants’ characteristics and cognitive performance. As expected, the groups differed significantly in AaA and time since diagnosis because the protocols with and without CRT were applied consecutively. However, the age of controls (range, 17.3 to 43.4 years) covers the full age range of survivors (range, 18.9 to 43.7 years). Estimated intelligence quotient was significantly decreased in the CRT group.

Visuomotor accuracy, visuomotor stability, work pace during sustained attention, and visuospatial sequential working memory showed a significant overall group difference. Contrasting each survivor group with controls showed that irradiated survivors performed worse than controls. These neuropsychological tasks were selected for correlation with FA maps.

**Assessment of Differences in FA**

CRT-treated survivors demonstrated significantly decreased FA (family-wise error corrected P < .05) compared with controls in orbitofrontal WM, genu, anterior body, and forceps minor of corpus callosum (CC), cingulum (frontal and parietal), and inferior fronto-occipital fasciculus (IFOF), superior longitudinal fasciculus (SLF) and uncinate fasciculi (Fig 1). After CT, trends for lower FA were seen in frontal WM tracts (Table 2).

**Correlation Analysis of Neuropsychological Performance With FA Values**

Voxel-based correlation analysis between FA maps of all patients and task performance revealed significant correlations in frontal, parietal, and temporal WM tracts with measures of visuomotor control, visuospatial sequencing, and sustained attention work pace (family-wise error corrected P < .05; Table 3 and Fig 2A). Figure 2D illustrates the correlation between FA in frontal WM and visuomotor accuracy.

**Fig 1.** (A) Sagittal and (B) axial slices of regions showing significantly decreased fractional anisotropy in cranial radiotherapy–treated survivors when compared with healthy controls (thresholded T maps [P < .001]). Color indicates significance.
Table 2. FA Voxel-Wise Analysis for Brain Regions Showing Significantly Reduced FA* in Survivors Compared With Healthy Controls

<table>
<thead>
<tr>
<th>Contrast</th>
<th>Region</th>
<th>Cluster Family-Wise Error Corrected P</th>
<th>Cluster Size (in number of voxels)</th>
<th>Anatomic Extent of Cluster</th>
<th>T</th>
<th>Mean FA Controls</th>
<th>Mean FA Survivors</th>
</tr>
</thead>
<tbody>
<tr>
<td>Controls v CRT-treated survivors</td>
<td>R + L frontal</td>
<td>&lt; .001</td>
<td>2,514</td>
<td>Cluster covering orbitofrontal WM, genu, anterior body, and forceps minor of CC, cingulum, IFOF, uncinate fasciculus, ALIC, SLF</td>
<td>7.58</td>
<td>0.374</td>
<td>0.332</td>
</tr>
<tr>
<td></td>
<td>R parietal</td>
<td>.02</td>
<td>154</td>
<td>Cluster covering part of cingulum and CC</td>
<td>4.12</td>
<td>0.415</td>
<td>0.375</td>
</tr>
<tr>
<td>Controls v CT-treated survivors</td>
<td>L frontal</td>
<td>.031</td>
<td>474</td>
<td>Cluster covering forceps minor of CC, cingulum, corona radiata</td>
<td>4.73</td>
<td>0.388</td>
<td>0.375</td>
</tr>
</tbody>
</table>

NOTE. Threshold set at P < .001 (clusters significant at family-wise error corrected P < .05 were retained for the CRT group). For the CT group, threshold was set at P < .01 (clusters significant at family-wise error corrected P < .05 were retained). Mean fractional anisotropy (FA) values are reported for the identified clusters.

Abbreviations: ALIC, anterior limb of internal capsule; CC, corpus callosum; CRT, cranial radiation therapy; CT, chemotherapy; FA, fractional anisotropy; IFOF, inferior fronto-occipital fasciculus; L, left; R, right; SLF, superior longitudinal fasciculus; WM, white matter.

*Trends for significantly reduced FA are provided for CT-treated survivors v controls.
| TABLE 3. | Brain Regions Showing Significantly Negative Correlations (Family-Wise Error Corrected $P < .05$) Between FA and Cognitive Scores and FA and CRT Dosage |
|----------------|-------------------------------------------------|----------------|---------------------------------|
| **ANT Task**   | **Side** | **Region** | **Cluster Family-Wise Error Corrected $P$** | **Cluster Size** | **Anatomical Extent of Cluster** | **$T$** | **Pearson $r$’** |
| **Visuomotor accuracy** |         |            |                                      |                  |                                          |        |                 |
| R + L          | Frontal + parietal | .045 | 118 | Cluster including WM under the anterior superior frontal gyrus | 4.63 | -0.374 |
| R              | Frontal | .002  | 263 | Cluster including subcortical WM, inferior longitudinal fasciculus, and inferior fronto-occipital fasciculus | 4.41 | -0.338 |
| R              | Temporal | .042  | 122 | Forceps major CC | 3.99 | -0.323 |
| **Visuomotor stability** |         |            |                                      |                  |                                          |        |                 |
| R + L          | Frontal | .002  | 226 | Cluster covering anterior part of SLF | 4.82 | -0.354 |
| R              | Parietal | .001  | 308 | Cingulum | 4.99 | -0.379 |
| R              | Parietal | .001  | 973 | Cluster covering left and right cingulum, right genu of CC | 4.96 | -0.433 |
| R              | Parietal | .007  | 200 | Cluster including SLF | 4.55 | -0.356 |
| L              | Frontal | .002  | 247 | Cluster including anterior part of SLF | 4.52 | -0.410 |
| **ANT sustained attention** |         |            |                                      |                  |                                          |        |                 |
| Work pace      | Frontal | .026  | 142 | Cluster covering body of CC and cingulum | 4.28 | -0.340 |
| **CRT dosage** |         |            |                                      |                  |                                          |        |                 |
| L              | Frontal | .05   | 102 | Cluster including part of IFOF and uncinate fasciculus | 6.08 | -0.673 |
| R              | Frontal | .001  | 272 | Cluster including part of cingulum and forceps major CC | 5.26 | -0.660 |
| R              | Frontal | .022  | 135 | Cluster including part of IFOF and uncinate fasciculus | 4.84 | -0.631 |

Abbreviations: ANT, Amsterdam Neuropsychological Tasks; CC, corpus callosum; FA, fractional anisotropy; IFOF, inferior fronto-occipital fasciculus; L, left; PLIC, posterior limb of internal capsule; R, right; SLF, superior longitudinal fasciculus; WM, white matter.

Pearson’s correlation coefficient calculated between ANT variable and average FA of sphere of 3 mm around peak voxel of cluster.
Correlations With AaD and AaA

Within the CRT group, significant positive correlations (family-wise error $P < .05$) between FA and AaD (corrected for AaA) were found in frontal and parietal WM, including parts of the forceps major, forceps minor, and body of CC; PLIC, anterior limb of internal capsula (PLIC), SLF, and orbitofrontal WM (Fig 2B). Significant negative correlations (family-wise error corrected $P < .05$) between FA and AaA, controlled for AaD, were found in frontal, parietal, and temporal WM, including parts of the forceps minor, forceps major, and body of CC; PLIC and anterior limb of internal capsula (PLIC), SLF, thalamic radiation, corona radiata, IFOF, and the uncinate fasciculus (Fig 2B). Within controls, correlations with AaA were not significant. In the CRT group, FA declined...
with age, as demonstrated by interacting regressions of FA on AaA between survivors and controls. This significant interaction indicates accelerated aging of WM (Fig 2E). This effect was shown to be larger when the effect of AaD was taken into account (Table 4). Note that a scatterplot of FA in cluster splenium CC from Table 4 is displayed in Fig 2B and a scatterplot of FA in cluster IFOF, uncinate fasciculus in Fig 2C. These effects are also visible on the neuropsychological variables (Table 4). No significant correlations with AaA or AaD were found within the CT group. Correlations between AaA and the neuropsychological variables are described in Table 4.

**Dosage Correlations**

Significantly negative correlations between FA and cumulative doses of CRT were found in clusters covering the CC, corona radiata, IFOF, and uncinate fasciculus (Fig 2C; Table 3). Interactions or correlations with doses of MTX IT or IV could not be established. Within the CT group, no correlations with doses of MTX IT or IV were found. No effects of doses of CRT or MTX IT or IV on neuropsychological outcome were observed.

**DISCUSSION**

This study demonstrated decreased WM integrity, as determined by FA, in a large group of leukemia and lymphoma survivors 25 years after treatment. Younger age at cranial irradiation and higher dosage were associated with lower FA. Accelerated aging of the irradiated brain was suggested. In addition, decreased FA was significantly associated with neuropsychological dysfunction.

For irradiated survivors, both FA and neuropsychological function were significantly below average. The dependence of these long-term outcomes on AaA and dosage is consistent with literature describing short-term outcomes of CRT.65 The steep decline of FA with AaA compared with controls, most importantly within the frontal and parietal WM, is a strong indication of accelerated aging. In general, the risk of developing dementia increases with age. There are also anatomic similarities between our survivors and patients with Alzheimer’s disease. Parente et al.66 reported decreased FA in the CC, cingulum, and SLF in patients with Alzheimer’s disease and those with mild cognitive impairment, similar to what we found. Furthermore, our own magnetoencephalography findings from the same cohort displayed an oscillatory activity pattern resembling the pattern found in patients with Alzheimer’s disease.67 Together, these findings suggest that the irradiated survivors could be at increased risk of developing early-onset dementia.

In general, FA values correlate well with cognition, in particular with tests of executive functions.68,69 In this cohort, neuropsychological dysfunction correlated significantly with lower FA in the CC, cingulum, and SLF in patients with Alzheimer’s disease and those with mild cognitive impairment, similar to what we found. Furthermore, our own magnetoencephalography findings from the same cohort displayed an oscillatory activity pattern resembling the pattern found in patients with Alzheimer’s disease. Together, these findings suggest that the irradiated survivors could be at increased risk of developing early-onset dementia.

The observed decreases in FA might be related to decreased myelin and/or axonal injury. There is evidence that suggests that both CT and CRT can cause early apoptosis of oligodendrocytes—essential for the myelination of axons—and vasculopathy leading to ischemia.6,14,70,73 Both CT and CRT can limit neural repair by damaging periventricular progenitor cells that would otherwise maintain WM integrity and stimulate hippocampal neurogenesis.74,77 Evidence is accumulating that these processes are fundamental to understanding late cognitive effects and WM decay and are likely to provide targets for future therapeutic interventions.74,77,78

### Table 4. Linear Regression Models

<table>
<thead>
<tr>
<th>Independent Variables</th>
<th>Step 1</th>
<th>Step 2</th>
<th>Step 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>AaA</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>AaD</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dosage CRT</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>FA splenium CC</td>
<td>-0.409</td>
<td>-0.766</td>
<td>-0.765</td>
</tr>
<tr>
<td>FA cingulum</td>
<td>-0.462</td>
<td>-0.592</td>
<td>-0.581</td>
</tr>
<tr>
<td>FA subcortical orbitofrontal WM</td>
<td>-0.579</td>
<td>-0.641</td>
<td>-0.624</td>
</tr>
<tr>
<td>FA genu CC</td>
<td>-0.507</td>
<td>-0.631</td>
<td>-0.614</td>
</tr>
<tr>
<td>FA body CC</td>
<td>-0.474</td>
<td>-0.633</td>
<td>-0.616</td>
</tr>
<tr>
<td>TIQ (normalized for age)</td>
<td>0.056</td>
<td>-0.233</td>
<td>-0.153</td>
</tr>
<tr>
<td>Visuomotor accuracy*</td>
<td>0.270</td>
<td>0.425</td>
<td>0.399</td>
</tr>
<tr>
<td>Visuomotor stability†</td>
<td>0.287</td>
<td>0.410</td>
<td>0.371</td>
</tr>
<tr>
<td>Sustained attention work pace†</td>
<td>0.265</td>
<td>0.410</td>
<td>0.371</td>
</tr>
<tr>
<td>Visuospatial sequencing†</td>
<td>0.316</td>
<td>0.322</td>
<td>0.287</td>
</tr>
<tr>
<td>FA IFOF, uncinate fasciculus</td>
<td>-0.228</td>
<td>-0.362</td>
<td>-0.318</td>
</tr>
</tbody>
</table>

**NOTE.** Bold correlation coefficients are indicative of accelerated aging. Linear regression models assessing the effects of age at assessment (AaA), age at diagnosis (AaD), and dosage of cranial radiotherapy (CRT) on fractional anisotropy (FA) in cognitively relevant regions and on the neuropsychological variables. The analysis was performed for the CRT group (n = 30), with relapse patients being excluded because of double AaD. The first five dependent variables were selected on the basis of both a significant correlation (family-wise error corrected P < .05) between FA and a neuropsychological deficiency and a significant correlation between FA and AaA. The correlations with AaA are supposedly zero in the normal population, and therefore, the large negative correlations in these regions indicate accelerated aging. The correlation between FA (or cognitive variables) and AaD is suppressed by the correlation with AaA, as shown in step 2 of the linear regression models. Without controlling for AaA, the accelerated aging effect is underestimated.

**Abbreviations:** CC, corpus callosum; IFOF, inferior fronto-occipital fasciculus; TIQ, total IQ (estimated based on four subtests); WM, white matter.

†Higher scores mean worse performance.
Furthermore, younger age at treatment with CRT was associated with lower FA, mostly within frontal and parietal WM tracts. These tracts are known to myelinate at a later age than the rest of the brain.²⁹ This could suggest that CRT affects the cells that create myelin more than it damages existing myelin. This might leave survivors with a lower peak level of WM density in young adulthood. Concurrently, treatment could injure the axons unprotected by myelin.

For nonirradiated survivors, both FA values and neuropsychological performance were lower on average, but not more than one standard deviation below the mean of controls. Trends for decreased FA can be shown, confirming findings by Porto et al.,¹⁴ but impact on cognition seems to be limited. The CT group displays no signs of accelerated aging. This suggests that, although acute leukoencephalopathy is frequently reported, long-term effects after 20 years are mild.¹⁰,¹¹ No dose-effect relationship could be established for MTX. Although outcome of nonirradiated survivors was on average within the normal range, there is a small subgroup with below-average FA and related neuropsychological deficiencies. This group should be acknowledged by clinicians. Future research should focus on the identification of this subgroup, identification of the risk factors, and development of preventive measures.

With older AaD, the risk of relapse and therefore treatment intensity increase. However, evidence indicates that CRT is more detrimental at younger AaD. This interaction is difficult to quantify but important to acknowledge.

In line with previous studies, we observed similar impairments in cognitive functioning nineteen–twenty-four and WM integrity in frontal, parietal, and temporal WM tracts. Porto et al.¹⁴ reported that WM around the frontal horns of the lateral ventricles and subcortical frontal WM were the areas most affected. Dellani et al.¹¹ reported decreased FA in the temporal lobes, the hippocampi, and thalami, in which we found decreased FA in WM surrounding these structures. We also found decreased FA in the cingulum, CC, and SLF. Our analysis might have been more sensitive because we used a population-based atlas, which allows for more accurate image registration, and a region of interest–independent analysis. A major strength of this study is the stronger perspective on long-term quality of life than previous studies. We were able to demonstrate age dependence by disentangling the effects of AaD and AaA, a method not applied by either Porto or Dellani. Thirty-one We also used larger patient samples and more homogeneous patient groups, and we excluded patients with pre-existing CNS disorders. The established association between decreased FA and neuropsychological dysfunction in this population is a major contribution to the existing literature. Evidently, longitudinal research with even larger groups is necessary to confirm our accelerated aging hypothesis, which is now based only on cross-sectional data. Alternatively, data from older controls, acquired in the same experimental setting, could be used to investigate similarity of FA levels and cognitive status between older adults and cancer survivors to further support the accelerated aging hypothesis. More research is needed to elucidate the clinical relevance of the observed trends of decreased FA after CT only.

The variability in treatment regimens, especially in terms of chemotherapy agents, might be a limitation of this study. However, this is inevitable when aiming for larger samples. Dose-effect relationships of agents other than MTX should be studied.

In conclusion, this study stresses the importance of following cohorts many decades after neurotoxic treatment in childhood, preferably throughout life. The growing support for the concept of accelerated aging after CRT implicates screening for early-onset dementia. Recommending lifestyle modifications that are implicated in slowing the progression of dementia, such as not smoking and getting regular physical exercise, could be considered.³² Although detrimental effects of CT on WM and neuropsychological function are not completely absent, they are mild compared with CRT, although CT is equally effective in terms of survival and recurrence rates after ALL.³ This warrants a recommendation to use CRT only as a last resort.

**AUTHORS’ DISCLOSURES OF POTENTIAL CONFLICTS OF INTEREST**

The author(s) indicated no potential conflicts of interest.

**AUTHOR CONTRIBUTIONS**

Conception and design: Ilse Schuitema, Frederik Barkhof, Eline van Dulmen-den Broeder, Cor van den Bos, Anjo J.P. Veerman, Leo M.J. de Sonneville

Administrative support: Ilse Schuitema, Marita Daams, Anne Uyttebroeck, Frederik Barkhof

Provision of study materials or patients: Anne Uyttebroeck, Frederik Barkhof, Cor van den Bos, Anjo J.P. Veerman, Leo M.J. de Sonneville

Collection and assembly of data: Ilse Schuitema, Marita Daams, Anne Uyttebroeck, Frederik Barkhof, Helena J. van der Pal, Cor van den Bos

Data analysis and interpretation: Ilse Schuitema, Sabine Deprez, Wim Van Hecke, Marita Daams, Stefan Sunaert, Frederik Barkhof, Leo M.J. de Sonneville

Manuscript writing: All authors

Final approval of manuscript: All authors

**REFERENCES**


**OF INTEREST**

The author(s) indicated no potential conflicts of interest.


42. Gunnther T, Herpertz-Dahlmann B, Konrad K: [Reliability of attention and verbal memory tests with normal children and adolescents: Clinical implications]. [In German]. Z Kinder Jugendpsychiatr Psychother 33:189-197, 2005


48. Schultema et al

© 2013 by American Society of Clinical Oncology. JOURNAL OF CLINICAL ONCOLOGY
71. Smith B: Brain damage after intrathecal methotrexate. J Neurol Neurosurg Psychiatr 38:810-815, 1975
The computerized Amsterdam Neuropsychological Tasks (ANT) provides standardized assessments and automated recordings of speed and accuracy of information processing, attention processes, and working memory. The program has proved to be helpful in defining neuropsychological deficit profiles in various clinical domains associated with generally diffuse impact on the brain and particularly in middle-late effects of childhood acute lymphoblastic leukemia (ALL). On the basis of these studies, tasks evaluating baseline response speed, pattern recognition, sustained attention (work pace and attentional fluctuations), cognitive flexibility (set shifting and inhibition), visuomotor skills, and visuospatial sequential working memory were selected for assessment of this study's population. The reliability and validity of these tasks are excellent. The following are task descriptions.

Baseline speed. A simple reaction time task in which cognitive demands are restricted to the mere detection of a stimulus. Provides a reference level for response speed. Parameters: T_bs (mean response time of left and right hand), S_bs (standard deviation of response times).

Feature identification. To test speed and accuracy of processing complex abstract visuospatial patterns. Task demands include maintenance and manipulation of working memory representations. Parameters: Ts_fi (mean response time in similar condition, in which target is surrounded by similar patterns and distinction is based on detailed information processing), Td_fi (mean response time in dissimilar condition, in which target is surrounded by dissimilar patterns and distinction is based on more global and simple information processing), PES_fi (percentage of errors in similar condition), PED_fi (percentage of errors in dissimilar condition).

Memory search objects. Patients have to detect a predefined target set in a signal of four two-dimensional symbols (red, green, blue, yellow, circle, triangle, cross, square). Memory load is increased across two task parts. Part 2 requires continuous monitoring and updating of the contents of the working memory. Parameters: T1_2d = (th1_2d + tc1_2d)/2 (mean of response times hits and correct rejections in part 1), T2_2d = (th2_2d + tc2_2d)/2 (mean of response times hits and correct rejections in part 2), Ne1_2d (mean number of errors part 1), Ne2_2d (mean number of errors part 2).

Sustained attention. To evaluate changes and fluctuations in speed and accuracy of processing over time. The paradigm induces a response bias providing indices for response inhibition and behavioral adaptation to feedback. Parameters: TSa (mean response time), SSa (standard deviation of response time, a measure for fluctuations of attention), PMSa (percentage of misses; answer is no although it should be yes), PF Sa (percentage of false alarms; answer is yes although it should be no). Note: Tsa is referred to in the main text as sustained attention work pace.

Shifting attentional set. To evaluate inhibition of prepotent responses and attentional flexibility. In the signal—a horizontal bar—a colored square may jump from left to right or vice versa. Depending on the color of the square, the patient should execute a compatible response (part 1), that is, press right (left) key when square jumped to the right (left), or is required to execute an incompatible response (press opposite keys, part 2). In part 3, trials of part 1 and 2 are randomly mixed, which requires a switch between the two types of response sets. Parameters: Tinhib (mean response time inhibition: difference between condition 2 and 1), Tflex (mean response time flexibility: difference between condition 3 and 1), P_inhib (percentage of errors on inhibition), P_flex (percentage of errors on flexibility).

Pursuit. This task evaluates the quality of visuomotor control. The patient has to track a small star, which continuously moves across the screen in random directions. The task requires concurrent planning and execution of unpredictable movements. Parameters: Dpu (mean distance to target), Spu (standard deviation of Dpu). Note: Dpu is referred to in the main text as visuomotor accuracy; Spu is referred to as visuomotor stability.

Tracking. This task serves the same purpose as the pursuit task, but now the patient is asked to execute planned, more automated movements. The patient has to draw a circle by moving the mouse cursor in between two large concentric circles on the screen. This task requires less controlled processing than the pursuit task. Parameters: Da_tr (mean absolute distance to target), St_tr (standard deviation of Da_tr). Diff_Dpu_tr (difference of Dpu and Da_tr) and Diff_Spu_tr (difference of Spu and S_tr) are measures of working memory.

Visuospatial sequencing. This task evaluates memory of visuospatial temporal patterns. In each trial, several circles are pointed out in an array of nine circles, arranged in a 3x3 matrix on the computer screen. The patient has to point out the same circles in the same order by moving the mouse cursor and must press a button when the cursor is positioned at the right location(s). The test consists of 24 trials in which the number of target circles varies from 3 to 7 and in which the spatial sequential patterns increase gradually in complexity. Parameters: Nitvs (number of correctly identified circles), Nitco_vs (number of correctly identified circles in the correct order). Diff_vs (Nit_vs – Nitco_vs) is a measure of the sequential working memory component. Note: where visuospatial sequencing is mentioned in the main text, it refers to the parameter Diff_vs.
Fig A1. Flow diagram of survivor selection. CRT, cranial radiotherapy; neuropsy., neuropsychological; NOS, not otherwise specified.