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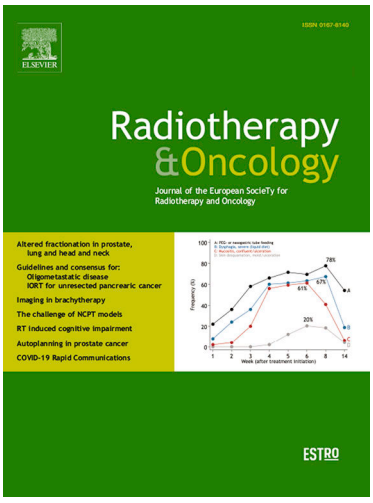

Original Article

Dose dependent cerebellar atrophy in glioma patients after radio(chemo)therapy

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Dose dependent cerebellar atrophy in glioma patients after radio(chemo)therapy

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Keywords: cerebellum, radiotherapy, atrophy, volume, proton therapy,

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Keywords: cerebellum, radiotherapy, atrophy, volume, proton therapy,

Abstract

Background and purpose:

Radiotherapy is a standard treatment option for high-grade gliomas. Brain atrophy has previously been associated with radiotherapy. The goal of this study was to investigate dose dependent cerebellar atrophy using prospective, longitudinal MR data from adult glioma patients who received radiotherapy.

Materials and methods:

Cerebellar volumes were measured using T1-weighted MR images from 91 glioma patients before radiotherapy (N = 91) and from longitudinal follow-ups acquired in three monthly intervals (N = 349). Relative cerebellar volumes were calculated as ratios to the corresponding baseline values. Univariate mixed effects models were used to determine factors that were significantly associated with relative cerebellar volumes. These factors were subsequently included as fixed effects in a final multivariate linear mixed effects model.

Results:

In multivariate analysis, cerebellar volume decreased significantly as a function of time ($p < 0.001$), time × dose ($p < 0.001$) and patient age ($p=0.007$). Considering a 55 year patient receiving a mean cerebellar dose of 0 Gy (10 Gy), the linear mixed effects model predicts a relative cerebellar volume loss of 0.4 % (2.0 %) after 1 year and 0.7 % (3.6 %) after 2 years. Compared to patients treated with photons, the cerebellar dose was significantly lower in patients treated with proton therapy ($p < 0.001$, $r = 0.62$).

Conclusion:

Cerebellar volume decreased significantly and irreversibly after radiotherapy as function of time and mean cerebellar dose. Further work is now needed to correlate these results with cognitive function and motor performance.

Abbreviations

CT	-	computed tomography
CET1w	-	post-contrast T1w images
CSF	-	cerebrospinal fluid
CTV	-	clinical target volume
GM	-	grey matter
GTV	-	gross tumour volume
MWU	-	Mann-Whitney *U* test
PTV	-	planning target volume
T1w	-	T1-weighted images
TBV	-	tumour bed volume
TFE	-	Turbo Field Echo
WM	-	white matter

Introduction

Radiotherapy is part of the standard treatment of high-grade gliomas following primary resection [1,2]. In particular, expanded clinical target volumes, but also technical limitations in dose delivery, such as positioning uncertainties and physical properties of the used irradiation technique, inevitably lead to irradiation of surrounding normal appearing brain. This can cause radiation induced brain injury, which is commonly categorised into acute, early delayed and late side effects, whereas late side effects occur several months after irradiation and are typically irreversible and progressive [3]. Late side effects include histopathological changes such as vascular alterations, demyelination and gliosis but also cognitive impairment, even when no obvious anatomical abnormalities are visible [3,4]. Such radiation-induced side effects can severely affect quality of life, particularly in long-term survivors [5-7].

We have previously shown that irradiation of normal appearing brain leads to cerebral atrophy and reduced perfusion [8,9] as well as changes in MR diffusion [10]. Both the anatomical and functional changes were significantly correlated with the regional radiation dose and progressed over time. The dose correlation also meant that these changes were significantly lower across the whole brain in patients treated with proton therapy, because they received a lower whole brain dose compared to patients treated with photon therapy [9,10].

Dose-dependent atrophy has been observed after irradiation across the whole brain [7,9,11] and substructures such as the hippocampus [12-15] and amygdala [16]. Brain atrophy has been linked with cognitive decline [6,7], however it is hypothesized that substructures, e.g. the hippocampus, are more radiosensitive [14] and therefore, together with their known function in memory and cognition [17], might be the driving force of cognitive decline after radiotherapy [15,18].

Recently, the question was raised if the cerebellum should be considered as an organ at risk due to increasing evidence of its contribution to cognition besides motor function [19]. A recent study by Dutz et al. [20] has shown that the volume receiving more than 30 or 40 Gy in the anterior cerebellum of adult patients treated for brain tumours correlated with a decrease in cognitive performance. Previous studies have found cognitive performance to decline with increasing cerebellar dose in children treated for infratentorial ependymoma [21], and linked cerebellar atrophy with cognitive performance in adult survivors of childhood brain cancer [21,22]. A preclinical study by Zhou et al. [23] demonstrated drastic atrophy and reduced perfusion in the cerebellum following a moderate radiation dose in juvenile rats. Given these findings, we wanted to analyse cerebellar volume changes after radiotherapy in a cohort of adult glioma patients using longitudinal structural

MR imaging. We related those changes to the mean cerebellar radiation dose, time after radiotherapy and patient age.

Methods

Patient cohorts

Data from two different prospective studies was combined for this analysis. The first patient cohort comprised data from an ongoing, longitudinal study (study A) of grade I-IV glioma patients, approved by the local ethics committee (NCT02824731, EK22012016). The second patient cohort included grade IV glioblastoma patients (study B). These data were acquired as part of a prospective, longitudinal study investigating the effect of 11C-methionine PET/MR for tailoring the treatment of patients with glioblastoma, approved by the local ethics committee (NCT01873469, EK41022013, BO-EK-167052020).

Gross tumour resection was performed in most patients prior to radio(chemo-)therapy. Baseline MR images were acquired after surgery and typically two weeks before the start of radio(chemo-)therapy. Follow-up MRIs were first acquired approximately three months after the end of radiotherapy and then every three months thereafter. However, patients occasionally skipped follow-up MRs or received clinical follow-up MRs at other centres, which were not used for analysis in this study. The scans were generally performed until either patient status worsened or patients required further clinical intervention due to clinical progression.

The following exclusion criteria were used: No baseline MRI, no follow-up MRI, additional treatment, previous irradiation, lesion in the cerebellum, severe motion artefacts, aspergillus infection (one patient), external head trauma (one patient).

Data acquisition

All MRI data were acquired on a 3 T Philips Ingenuity PET/MR scanner (Philips, Eindhoven, The Netherlands) using an eight channel head coil.

Pre- and post-contrast T1-weighted (T1w) MR images were used for cerebellar segmentation. In study A, pre-contrast T1w images were acquired using a 3D gradient spoiled echo sequence acquired in sagittal orientation with 1 mm isotropic resolution. In study B, pre-contrast T1w MR images were acquired using a 3D Turbo Field Echo (TFE) sequence acquired in sagittal orientation at 1 mm isotropic resolution. This 3D TFE sequence was used in both studies to acquire post-contrast T1w images (CET1w) following the injection of intravenous contrast agent.

Radiation treatment planning

Computed tomography (CT) scans for radiation treatment planning were performed prior to radio(chemo)therapy with the patient positioned supine with an individual head support and mask. For radiation treatment planning, the CTs were co-registered with the post-surgery MRI scans (T1w, T2w, CET1w) to define the tumour bed and potential residual tumour (tumour bed volume; TBV, or gross tumour volume; GTV, respectively). Depending on the tumour histology, the TBV (including the GTV) was expanded by a 1-2 cm isotropic margin, corrected for anatomical boundaries, to derive the clinical target volume (CTV). For proton therapy, dose was prescribed to the CTV taking into account inherent proton range uncertainties, whereas for photon beam irradiation, a planning target volume (PTV) was created by expanding the CTV using an isotropic margin of 0.5 cm. For this analysis, the planning CTs and corresponding dose maps were retrieved from the planning workstation. Prescribed total dose was typically either 54 Gy or 60 Gy depending on tumour histology and delivered in 27-30 fractions.

Data processing

Cerebellar volumes were calculated using a three-step process as illustrated in Figure 1. In step one, the Montreal Neurological Institute (MNI) 152 brain atlas [24] provided by the FMRIB Software Library (FSL) [25] was transformed to individual brain extracted T1w images using deformable coregistration implemented in Advanced Normalization Tools (ANTs) [26,27]. A cerebellum mask created in MNI space was then transformed to the individual T1w images to cut out the cerebellum. Step two cleaned up the resulting cerebellar volumes by removing any residual parts of the transverse sinuses. This was achieved by rigidly coregistering the CET1w images to the T1w images, segmenting the contrast enhancement in the CET1w images using Atropos [28] and subsequently removing these voxels from the T1w cerebellum images. In step three, Atropos segmentation [28] was used to segment the cerebellum into grey matter (GM), white matter (WM) and cerebrospinal fluid (CSF) using the T1w images and prior probability maps from the publicly available SUIT atlas [29]. The volume of each cerebellum was then calculated as the sum of all GM and WM probabilities. The segmentation of the cerebellum was visually inspected for all patients and time points. Manual correction was performed in cases where the automated pipeline still included parts of the transverse sinuses or parts of the cerebrum.

The mean dose delivered to the cerebellum was calculated by rigidly coregistering the planning CT and corresponding dose maps to the baseline T1w images.

Statistical analysis

Relative cerebellar volumes V_{rel} were calculated for each patient as ratios to their corresponding pre-radiotherapy baseline values.

Analysis was carried out in R [30] using a linear mixed effects model with the relative cerebellar volume as the response variable and patient ID as grouping variable. To establish which fixed effects should be considered in this model, we first carried out multiple univariate analyses on all potential fixed effects (see supplementary material): time after radiotherapy, mean cerebellar dose, patient age, gender, low grade (grade I-II) or high grade (grade III-IV) tumour, radiotherapy type (proton/photon), chemotherapy (yes/no). Significant predictors were then used as fixed effects in the linear mixed effects model.

The difference in mean cerebellar radiation doses between proton and photon radiotherapy was compared using a non-parametric Mann-Whitney *U* test performed in MATLAB (The MathWorks, Inc., Natick, Massachusetts, USA). A corresponding effect size was calculated as $r = Z/\sqrt{N}$.

Results

In total, 91 patients with baseline MRIs and 349 follow-up MRIs were eligible for this study. Follow-up MRIs were available at 3 (N = 87), 6 (N = 58), 9 (N = 38), 12 (N = 30), 15 (N = 25), 18 (N = 25), 21 (N = 18), 24 (N = 16), 27 (N = 15) and > 27 months (N = 37) after the end of radio(chemo)therapy. Patients treated with protons (N = 38) received a significantly lower mean cerebellar dose compared to patients treated with photons (N = 52) (median [range]: 0.4 Gy [0 Gy – 6.8 Gy] vs. 4.9 Gy [0.3 Gy – 16.9 Gy] ; MWU: $p < 0.001$, $r = 0.62$). Full patient details are summarised in Table 1. Plotting the relative cerebellar volumes over time for all patients illustrates a general trend for cerebellar volume reduction (Figure 2).

Univariate analysis using linear mixed effects modelling revealed that neither gender, tumour grade, radiotherapy type nor chemotherapy were able to predict relative cerebellar volumes (see supplementary material). On the other hand, time after radiotherapy, mean cerebellar dose and patient age were found to significantly predict the relative cerebellar volume. Consequently, we used the following multivariate linear mixed effects model:

$$V_{\mathrm{rel}} = a \times dose + b \times time + c \times time \times dose + d \times age + e,$$

R syntax: lmeModel = lme(V_{rel} ~ dose*time + age, data, random = ~ 1|id).

The model coefficients are given in Table 2. All included parameters remained significantly related to the relative cerebellar volume, except for the cerebellar dose that showed a statistical trend. The model revealed that cerebellar atrophy increased with higher mean radiation dose and with increasing time after radiotherapy. Additionally, older patients were predicted to develop greater cerebellar atrophy over the same time span than younger patients (Figure 3). One year after receiving a mean cerebellar dose of 10 Gy, patients aged 30, 55 and 80 years were expected to lose approximately 1.6 %, 2.0 % and 2.4 % cerebellar volume, respectively (Figure 3A). After two years, these values increased to approximately 3.2 %, 3.6 % and 4.0 %, respectively (Figure 3B).

Discussion

To the best of our knowledge, this is the first longitudinal study reporting on dose dependant cerebellar volume loss after radiotherapy in adults. The cerebellar volume loss showed no signs of recovery within our observational period. This highlights the potential impact particularly for long-term survivors following radiotherapy.

We found that time after radiotherapy, the product of time after radiotherapy and mean cerebellar dose as well as patient age were significant predictors for cerebellar volume loss. From the linear mixed effects model, we can estimate an expected cerebellar volume loss of around 2 % for a 55 year old patient receiving a mean cerebellar dose of 10 Gy (Figure 3A). While we found no previous studies investigating dose dependent cerebellar volume changes, Ailion et al. [22] found radiotherapy as a predictor of cerebellar atrophy in a cohort of 25 adult survivors of cerebellar childhood tumours. Several studies reported atrophy of cerebral subvolumes after radio(chemo)therapy. Karunamuni et al. [11] reported significant dose dependent cortical thinning 1 year after radiotherapy in a cohort of 15 high grade glioma patients. Petr et al. [9] investigated volume loss of the cerebrum 3 and 6 months after the end of radiotherapy using a subset of patients used in this study and found significant grey matter atrophy of approximately 0.9 % per 10 Gy at 3 months after the end of radio(chemo)therapy. Gommlich et al. [31] found a trend for decreasing white matter in adult glioma patients after irradiation, although the study was hampered by heterogeneous MR data acquired during routine follow-up, i.e. with different protocols, field strengths and resolutions. In a longitudinal study, Prust et al. [32] found progressive whole brain volume loss during and after radiotherapy with concurrent chemotherapy in a cohort of eight glioblastoma patients. Volume loss was about 1 % at approximately 3 months after the end of radiotherapy. In a cohort of primary brain tumour patients analysed one year after radiotherapy, Seibert et al. [12,16,33] found regional and dose dependant cortical thinning, a 6 % decrease in volume of the hippocampus receiving a radiation dose greater than 40 Gy and a decrease of the amygdala volume at a rate of 1.7 % per 10 Gy. Takeshita et al. [14] assessed hippocampal volumes of 20 metastases patients receiving whole brain irradiation with a total dose of 30 Gy and found volume reductions of 1.8 %, 5.8 % and 9.2 % after 0-3, 4-7 and 8-11 months, respectively. The volume changes reported in these studies are thus comparable to the volume changes that we found in the cerebellum, despite focusing on different brain subvolumes, treatment regimens and radiotherapy techniques.

For this study, radiation treatment planning was performed without specifically considering the dose to the cerebellum. Nevertheless, in our cross-section of patients we could show that the cerebellar

mean dose was significantly lower in patients treated with proton therapy compared to patients treated with photon therapy.

The benefit of dose sparing using proton therapy has already been shown in children treated for brain cancer [34-36]. Previous studies have linked cognitive decline with radiation induced atrophy of the whole brain [7] and the hippocampus [15]. In children with infratentorial ependymoma, Merchant et al. [21] have shown that cerebellar dose correlated with poor cognitive performance assessed over a follow-up period of five years. Cerebellar atrophy in adult survivors of cerebellar childhood tumours was linked to a decrease in processing speed [22]. Dutz et al. [20] found that cognitive decline was linked to the volume receiving more than 30 or 40 Gy in the anterior cerebellum of adult patients treated for brain tumours. Cerebellar atrophy could explain this link between cerebellar dose and decreasing cognitive function, although the underlying mechanisms might be more complex.

There are several limitations of this study. (1) Although neurocognitive testing was available for some patients, the data could not be included here since these are primary endpoints of ongoing studies. We can therefore not assess the connection of cerebellar atrophy to cognition and at what level cerebellar atrophy becomes clinically relevant. (2) Segmentation accuracy of the cerebellar volume is limited by the MRI resolution and the ability to separate its outlines from surrounding tissues. Small errors in the cerebellar volume estimation in the baseline MRIs will impact all relative volumes determined in the follow-up MRIs. (3) The number of patients not receiving adjuvant chemotherapy was low (N = 12), of which only one patient received a cerebellar mean dose greater than 1 Gy. Consequently, we cannot assess the effect of radiotherapy alone on the cerebellar volume.

Future work is now needed to analyse the potential connection between motor function, cognitive decline and cerebellar atrophy. Such work would be vital to establish at which point cerebellar atrophy becomes symptomatic and clinically relevant. However, we also recommend to simultaneously measure the volumes of the hippocampus, amygdala and the whole cerebrum to rule out that cerebellar atrophy is simply an indirect marker of atrophy of another brain region that is more relevant to cognitive function. Longer observational times of the patients are also needed to establish if cerebellar atrophy continues to progress beyond two years.

In conclusion, cerebellar volume decreases significantly and irreversibly after radio(chemo)therapy as function of follow-up time and radiation dose. The magnitude of atrophy is comparable to previously published results for the cerebrum, hippocampus and amygdala. Further work is required to correlate cerebellar volume loss with cognitive function and motor performance.

Acknowledgements

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Conflict of Interest

In the past 5 years, Dr. Michael Baumann attended an advisory board meeting of MERCK KGaA (Darmstadt), for which the University of Dresden received a travel grant. He further received funding for his research projects and for educational grants to the University of Dresden by Teutopharma GmbH (2011-2015), IBA (2016), Bayer AG (2016-2018), Merck KGaA (2014-open), Medipan GmbH (2014-2018). He is on the supervisory board of HI-STEM gGmbH (Heidelberg) for the German Cancer Research Center (DKFZ, Heidelberg) and also member of the supervisory body of the Charité University Hospital, Berlin. As former chair of OncoRay (Dresden) and present CEO and Scientific Chair of the German Cancer Research Center (DKFZ, Heidelberg), he has been or is still responsible for collaborations with a multitude of companies and institutions, worldwide. In this capacity, he discussed potential projects with and has signed/signs contracts for his institute(s) and for the staff for research funding and/or collaborations with industry and academia, worldwide, including but not limited to pharmaceutical corporations like Bayer, Boehringer Ingelheim, Bosch, Roche and other corporations like Siemens, IBA, Varian, Elekta, Bruker and others. In this role, he was/is further responsible for commercial technology transfer activities of his institute(s), including the DKFZ-PSMA617 related patent portfolio [WO2015055318 (A1), ANTIGEN (PSMA)] and similar IP portfolios.

Within the past years, Dr. Krause received funding for her research projects by IBA (2016), Merck KGaA (2014-2018 for preclinical study; 2018-2020 for clinical study), Medipan GmbH (2014-2018), Attomol GmbH (2019-2021), GA Generic Assays GmbH (2019-2021), BTU Cottbus-Senftenberg (2019-2021), Gesellschaft für medizinische und wissenschaftliche genetische Analysen (2019-2021), Lipotype GmbH (2019-2021), PolyAn GmbH (2019-2021).

Dr. Baumann and Dr Krause confirm that to the best of his knowledge none of the above funding sources were involved in the preparation of this paper.

The other authors have nothing to disclose.

Figures

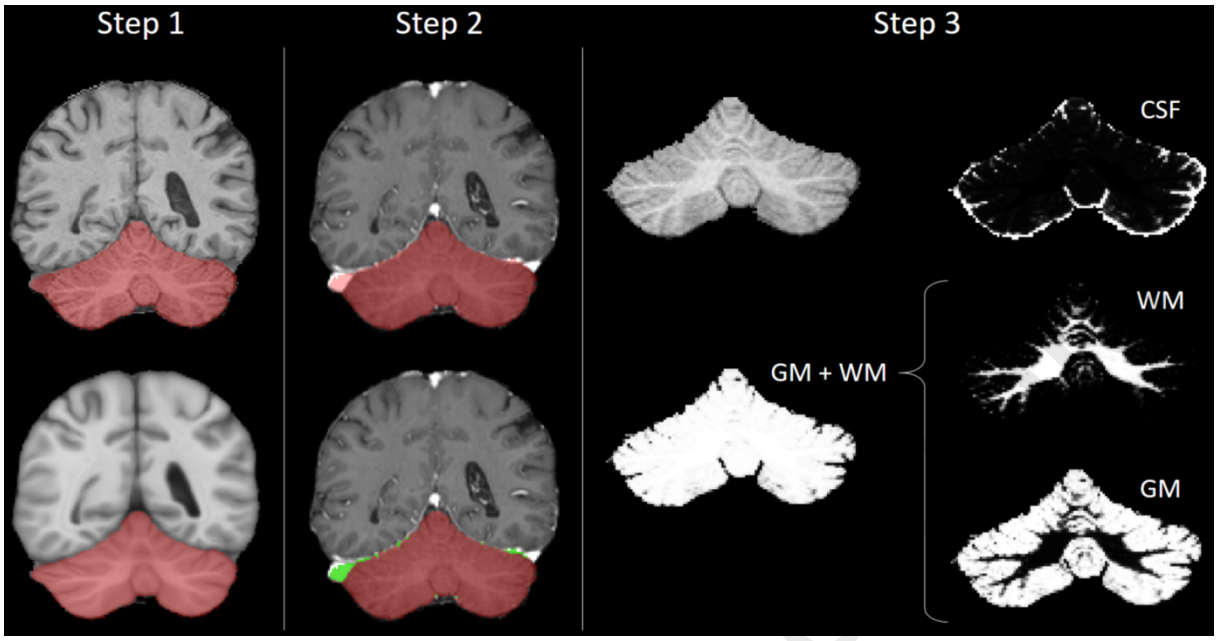

Figure 1: Segmentation and volume measurement of the cerebellum illustrated for a patient. Step 1: Non-linear coregistration of the MNI152 brain atlas (bottom) and corresponding cerebellum mask (red) to the brain extracted T1w image of a patient (top). Step 2: Rigid coregistration of the CET1w image to the T1w image and segmentation of contrast enhancement (green) within the cerebellum mask (red). Contrast enhancing voxels were removed from the final cerebellum mask. Step 3: Segmenting the extracted cerebellum into CSF, WM and GM. The sum of GM and WM probabilities was used to calculate the cerebellar volume.

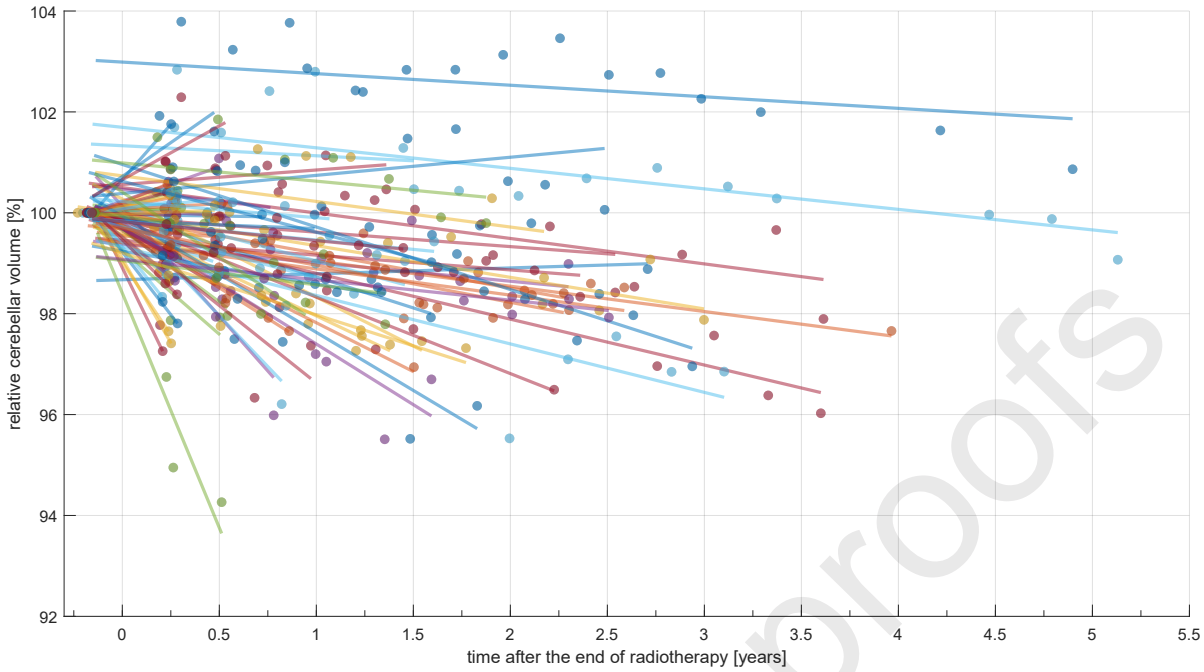

Figure 2: Trend lines for all patients, created across all available time points to illustrate the general trend of the relative cerebellar volumes reduction over time in our patient cohort.

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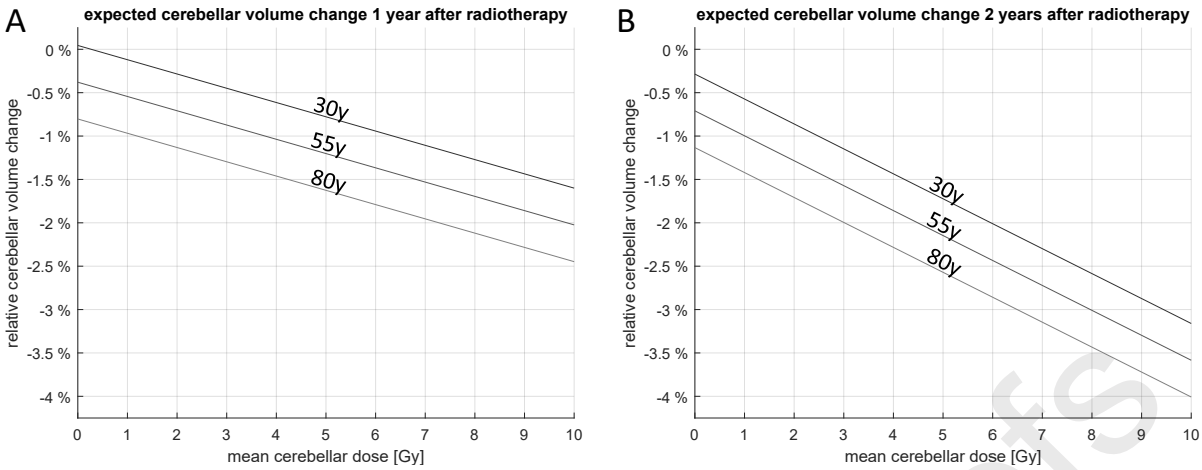

Figure 3: Illustration of the expected cerebellar volume loss 1 year (A) and 2 years (B) after radiotherapy for patients 30, 55 and 80 years of age. These plots were generated using the coefficients of the final linear mixed effects model given in Table 2.

Tables

Table 1: Patient information. For this analysis data was combined from two separate studies, study A (NCT02824731, EK22012016) and study B (NCT01873469, EK41022013). TMZ – temozolomide, PCV – procarbazine, lomustine (CCNU) and vincristine.

	study A	study B	study A + B
eligible Patients [N]			
total/male/female	24/11/13	67/38/29	91/49/42
age at baseline [years]			
mean ± std	45.6 ± 14.5	54.7 ± 13.9	52.3 ± 14.5
range [min - max]	[20.1 - 76.7]	[23.2 - 81.8]	[20.1 - 81.8]
glioma grade [N]			
grade I/II/III/IV	1/3/13/7	0/0/0/67	1/3/13/74
cerebellum volume at baseline [ml]			
mean ± std	120.3 ± 12.1	117.4 ± 11.4	118.2 ± 11.9
range [min - max]	[99.6 - 144.9]	[91.2 - 143.1]	[91.2 - 144.9]
mean cerebellum dose [Gy]			
mean ± std	1.5 ± 2.3	4.8 ± 4.4	3.9 ± 4.2
range [min - max]	[0 - 7.8]	[0 - 16.9]	[0 - 16.9]
RTx treatment [N]			
Ph/H+/mix	4/19/1	48/19/0	52/38/1
number of MR scans [N]			
total/manual correction	130/12	310/43	440/55
follow-ups			
mean number of follow-ups [N]	4.4 ± 3.0	3.6 ± 3.2	3.8 ± 3.2
mean follow-up period [days]	444.8 ± 311.4	422.2 ± 438.5	428.1 ± 407.3
range follow-up period [days]	[85 - 1073]	[63 - 1874]	[63 - 1874]
chemotherapy [N]			
TMZ/PCV/none	7/5/12	67/0/0	74/5/12

Table 2: Results of the multivariate linear mixed effects model $V_{\mathrm{rel}} = a \times dose + b \times time + c \times time \times dose + d \times age + e$.

	Coefficient	Std. Error	p value
cerebellar dose [Gy]	a = -4.1×10^{-4}	2.3×10^{-4}	0.072
time [days]	b = -9.0×10^{-6}	1.6×10^{-6}	<0.001
time × dose [years x Gy]	c = -3.4×10^{-6}	-0.4×10^{-6}	<0.001
age [years]	d = -1.7×10^{-4}	-0.6×10^{-4}	0.007
intercept	e = 1.009	0.0032	<0.001

5

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- Radiotherapy causes cerebellar atrophy in adult glioma patients
- Cerebellar atrophy is dose dependant and progressive over time

- Cerebellar dose is lower in the proton group compared to the photon group