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Multicentric assessment of safety and efficacy of combinatorial adjuvant brain metastasis treatment by intraoperative radiotherapy and immunotherapy

Safety and efficacy of IORT plus immunotherapy

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Conflict of Interest Statement for All Authors

J.P.L reports stocks and travel expenses from TME Pharma AG; travel expenses from Carl Zeiss Meditec AG; stocks and honoraria from Siemens Healthineers and stocks from Bayer AG and BioNTech AG. S.B. reports travel expenses and honoraria from Carl Zeiss Meditec AG. C.D.D. reports travel expenses from Carl Zeiss Meditec AG. M.H. reports travel expenses from Carl Zeiss Meditec AG. H.V. reports travel expenses from Carl Zeiss Meditec AG. M.S. reports travel expenses from Carl Zeiss Meditec AG. S.E.C. reports travel expenses and honorarium from Carl Zeiss Meditech AG. L.C.S. reports travel expenses from Carl Zeiss Meditec AG. C.P.C reports travel expenses and speaking honorarium from Carl Zeiss Meditech AG. F.A.G. reports research grants and

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Data availability

The data presented in this study are available in this article. Further datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

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Abstract

Purpose

Following surgical resection of brain metastases (BMs), intraoperative radiation therapy (IORT) provides a promising alternative to adjuvant external beam radiation therapy (EBRT) by enabling superior organ at risk preservation, reduction of in-hospital times and timely admission to subsequent

systemic treatments, which increasingly comprise novel targeted immunotherapeutic approaches. We sought to assess safety and efficacy of IORT in combination with immune checkpoint inhibitors (ICIs) and other targeted therapies (TTs).

Methods

In a multicentric approach incorporating individual patient data from six international IORT centers, all patients with BMs undergoing IORT were retrospectively assessed for combinatorial treatment with ICIs/TTs and evaluated for toxicity and cumulative rates, including wound dehiscence, radiation necrosis (RN), leptomeningeal spread (LMS), local control (LC), distant brain progression (DBP) and estimated overall survival (OS).

Results

A total of 103 lesions with a median diameter of 34 mm receiving IORT combined with immunomodulatory systemic treatment or other TTs were included. The median follow up was 13.2 (1.2-102.4) months and the median IORT dose was 25 (18-30) Gy prescribed to the applicator surface. There was one grade 3 adverse event related to IORT recorded (2.2%). A 4.9% cumulative RN rate was observed. The 1-year LCR was 98.0% and the 1-year DBP-free rate 60.0%. Median time to DBP was 5.5 (1.0-18.5) months in the subgroup of patients experiencing DBP and the cumulative LMS rate was 4.9%. The median estimated OS was 26 (1.2-not reached) months with a 1-year survival rate of 74.0%. Early initiation of IT/TT was associated with a non-significant trend towards improved DBP rate and OS.

Conclusion

The combination of ICIs/TT with IORT for resected BMs does not seem to increase toxicity, while yielding encouraging local control outcomes in the difficult-to-treat subgroup of larger BMs. Time gaps between surgery and systemic treatment could be shortened or avoided. The definitive role of IORT in local control after BM resection will be defined in a prospective trial.

Introduction

The rise of novel immunotherapeutic agents has redrawn the treatment patterns for many tumor entities in recent years (1, 2). As a consequence of improved local control and prolonged survival, the diagnostic incidence of brain metastases (BMs) has increased significantly (3, 4) with nearly every second patient developing BM over the course of the disease (5, 6). This is also attributed to the fact that many of the novel immunotherapeutic drugs cannot penetrate the blood-brain-barrier (BBB) sufficiently to induce stable tumor control within the brain (7, 8). While overall survival (OS) is largely dictated by extracranial disease progression (9), BMs usually require medical intervention to prevent or stabilize neurological deterioration and impairment of quality of life (QOL) (10, 11). Local treatment options include surgery, radiosurgery, fractionated stereotactic radiotherapy and surgery followed by adjuvant radiotherapy of the resection cavity. Surgery and adjuvant radiotherapy are usually indicated for larger BMs to improve local control rates, as smaller volume BM do not need surgery (12–14). While the most common form of RT application is stereotactic external-beam RT (EBRT) with one to seven fractions (12–15), intraoperative RT (IORT) provides an excellent alternative yielding equal clinical outcome (16–19) at superior organ at risk (OAR) preservation (20) and a favorable toxicity profile (21, 22). However, data are very limited regarding potential desirable and undesirable effects (23, 24) of concomitant or sequential treatment with increasingly available immunostimulating systemic therapy (25). We thus sought to assess safety and efficacy of combination treatment with IORT to BM and immunotherapy (IT) in this multicentric retrospective series.

Methods

Patients

In a multicentric approach, patient databases of four XXXX, one XXXX and one XXXX university hospitals were retrospectively screened for patients with BMs receiving IORT with concomitant or sequential IT or targeted therapy (TT) between 2014 and 2023. IT was defined as authority-approved administration of an ICI, i.e., anti-PD-L1, anti-PD-1, anti-CTLA4 antibodies. TT was defined as authority-approved administration of a drug using a tumor-specific, either non-immunogenic or

immunogenic target other than immune checkpoint blockade, i.e., BRAF/MEK inhibition, (multi-) tyrosine kinase inhibition or antibodies against essential tumor signaling pathways. For inclusion, at least one available imaging follow-up and information on received systemic treatment was mandatory. All patients underwent surgical resection and IORT following interdisciplinary evaluation in a neuro-oncological tumor board. BMs were pathologically confirmed in all cases. The criteria for surgical resection were presence or severe risk of acute neurological impairment, clinically significant mass effects as abnormal intracranial pressure or hemispheric shift and histopathological confirmation of diagnosis in case of cancer of unknown primary. Only the clinically relevant lesion receiving IORT was considered for surgical removal in case of multiple BMs. Requirements for IORT were gross total resection, intraoperative confirmation of BM on frozen tumor sections and fulfillment of dose constraints. The data collected from eligible patients included sociodemographic characteristics, functional status with Karnofsky performance score (KPS), tumor location, histology, baseline and follow-up (FU) radiological features of the lesion and systemic therapy status. Diagnostic-Specific Graded Prognostic Assessment (DS-GPA) (26) scores were calculated by standard procedures.

Intraoperative radiotherapy

3D image guidance for both surgery and IORT was provided by preoperative contrast-enhanced T1-weighted magnetic resonance imaging (MRI). Optic nerves, chiasm and brain stem were identified preoperatively and intraoperatively as OARs for IORT and delivered doses were defined based on dose-depth template profiles corresponding to each applicator diameter. Following macroscopic complete resection of the lesion, a frozen section was assessed intraoperatively by a board-certified neuropathologist confirming the presence of malignant cells with an extracranial solid tumor origin. Neurosurgical MRI-navigation was used to intraoperatively assess the minimum distance of the resection cavity to organs at risk and cavity extends, followed by selection of the optimal fitting for spherical applicators ranging from 1.5 to 5.0 cm diameter. The selected applicator was placed in the resection cavity without applying pressure to the adjacent healthy brain tissue, but with the aim of ubiquitous direct tissue contact avoiding air entrapment for optimal dose distribution. The IORT was only performed when a safe and orderly execution was ensured. The INTRABEAM® 600 (Carl Zeiss

Meditec AG, Germany) was used to deliver IORT by application of nominal 50-kV photons at a standard dose of 20 to 30 Gy prescribed to the applicator surface. The dose profile in depth was obtained prior to each procedure according to pre-performed Monte Carlo calculations with Radiance (GMV, Spain). Decreasing the prescribed dose down to 16 Gy was acceptable in case of OAR doses exceeding the constraints of 8 Gy to the optical system or the brain stem following QUANTEC (Quantitative Analyses of Normal Tissue Effects in the Clinic) recommendations (27) with consideration of the specific (1.3-1.5 times higher) RBE of low energy photons. In individual cases, anatomical positioning of the applicator required consideration of further, not regularly assessed OAR, e.g., cochlea or thalamus, with equal consideration of the QUANTEC recommendations. The irradiation time ranged from seven to 49 minutes, depending on the applicator size and the prescribed dose. Following removal of the applicator, the surgery was continued as per standard procedures with wound sealing.

Follow-up

All patients had regular FU visits including physical examination and MRI as per guideline recommendations. Adverse events (AEs) were assessed and graded by clinicians according to the National Cancer Institute Common Terminology Criteria for Adverse Events (CTCAE), Version 5.0. Acute toxicities were considered AEs occurring within the first eight weeks of FU, whereas late toxicities were defined as all AEs recorded at a later time point. MRI assessments were performed according to the RANO criteria (28) by board-certified radiologists. In case of uncertain clinical/radiographic response, the interdisciplinary neuro-oncological tumor board was consulted for shared decision-making. The following conditions qualified for diagnosis of RN: (1) after initial suspected progressive disease (PD), a minimum of two FU MRIs showed no sign of ongoing PD; (2) advanced MRI incorporating dynamic susceptibility contrast (DSC) perfusion imaging or diffusion-weighted imaging (DWI) was concordantly suggestive of RN; (3) positron emission tomography (PET) imaging such as ^{18}F-fluoroethyl-tyrosine PET with findings consistent with RN; (4) RN was confirmed histopathologically following resection.

Study endpoints

The primary endpoints were toxicity, namely cumulative RN rates, and 1-year LCR. The secondary endpoints were cumulative distant brain progression (DBP) rates, leptomeningeal spread (LMS) rates, 1-year OS rates and estimated OS. For toxicity assessment, simultaneous IORT and IT/TT was defined as an initiation of treatment within the first 2 months after date of surgery. Local control was defined as the absence of MRI-radiographic PD as per RANO-BM criteria (28) within 1 cm surrounding the previously irradiated BM resection cavity and absence of clinical deterioration attributable to the treated lesion. Local control was calculated from the day of surgery until the local PD date. Patients lost to FU or deceased prior to radiographic progression were censored at the last FU time point. DBP was defined as an MRI-radiographic emergence/progression of intracranial lesions as per RANO-BM criteria in at least 1 cm distance to the resection cavity receiving IORT or clinical deterioration not attributed to the IORT, but a distant brain lesion. DBP rates were calculated from the day of surgery until the PD date. Patients lost to FU or deceased prior to the event were censored at the last FU time point. Leptomeningeal spread was defined as either MRI-radiographic suspicion or cytologic confirmation of pachymeningeal or leptomeningeal tumor cell spread. OS was defined as time interval between the date of surgery and the date of either last FU (censored) or death.

Ethics

The study was conducted in accordance with the principles of the Declaration of Helsinki and approved by the Ethics Committee XXXX (XXXX).

Statistics

The software package used for the data analyses was GraphPad Prism (version 9, GraphPad Software, USA). Figures and Graphs were created using GraphPad Prism and Adobe Illustrator 2023 (Adobe Inc., USA). Descriptive statistics incorporated calculation of percentages and median values with minimum to maximum range. For survival analysis, the Kaplan-Meier method was employed and curves with 95% confidence intervals were generated. Hazard ratios and their 95% confidence intervals were calculated using the Mantel-Haenszel method. Fisher's exact test was used to analyze

categorical variables. The Mann-Whitney test was used to compare continuous variables, as the data were not normally distributed. Statistical significance was defined as a p-value < 0.05. The particular statistical methods applied are specified in the corresponding figures.

Results

Patient and tumor characteristics

A total sample size (n) of 114 consecutive patients with BMs receiving IORT to the resection cavity combined with immune checkpoint inhibitors (ICIs) or other TTs were screened. Of these, sufficient FU information (at least one imaging follow-up and systemic therapy information) was available for 99 patients with 105 treated lesions. Two cases were removed from the outcome analyses since the IORT lesion received additional immediate SBRT leaving a total of 103 lesions analyzed. The median patient age was 63 (range 35-85; n=99) years and the median KPS was 80 (40-100). The median DS-GPA score was 2 (0-4; n=99). The most frequent BM localization was the frontal lobe (35.0%), while most histopathology results corresponded to lung cancer (54.4%). With a range of 1 to 16 intracranial lesions, 48 cases (46.6%) suffered from multiple BMs at the time of surgery. Further details on patient characteristics are provided in Table 1.

Treatment

The median FU was 13.2 (1.2-102.4; n=99) months. The brainstem and the optic tracts (optic nerves and chiasma) were regularly assessed as OARs and no dose constraints were exceeded. All patients completed treatment. The median IORT prescription dose was 25 (16-30; n=103) Gy to the surface, which corresponds to a dose delivery of approximately 60% in 3 mm, 45% in 5 mm and 22% in 10 mm tissue depth, slightly varying depending on applicator diameter. The median applicator size was 2 (1.5-4.0; n=103) cm. While 90 patients (87.4%) received IORT plus ICIs, another 25 patients (24.3%) received other TTs. Of note, some patients received both ICI and TT in parallel or combinations of either substance group. The median time to ICI initiation after IORT was 1.1 (-22.3-34; n=90)

months. TT was initiated after a median time of 1.2 (-38.9-22.9; n=25) months. Table 2 depicts further treatment characteristics and lists the specific administered substances.

Toxicity

Under combinatory treatment, mild and anticipated toxicity was reported. A summary of the observed AEs is provided in Table 3. No grade 4 or 5 events were deemed related to IORT. Fig. 1a and 1b show the maximum toxicity observed for individual patients. A cumulative RN rate of 4.9% (n=5) was observed with a median time to RN of 12.8 (7.8-18.9) months (Fig. 1c). Of these RN events, four were grade 1 and one a grade 3 event. The latter occurred in a patient with RCC receiving IORT with 30 Gy to a frontal 34 mm BM after 7.8 months. This patient had received systemic treatment with cabozantinib initiated 5 weeks after surgery for a total of 7 months, before it was terminated due to an unfavorable overall toxicity profile. The RN was treated successfully with bevacizumab after previous failure of dexamethasone treatment. No wound dehiscences of any grade were noted. There were significantly more severe AEs (p=0.049; Fig. 1d) in total, but also treatment-related (p=0.025; Fig. 1e; RN, autoimmune infection) recorded for patients that commenced systemic treatment in parallel to resection and IORT, defined as initiation of treatment within the first 2 months following surgery. The full list of acute and long-term AEs is provided in Suppl. Table 1.

Outcome

The overall 1-year and 2-year LCRs were 98.0% (Fig. 2a) and 93.7%, respectively. With an overall DBP rate of 36.9%, the median DBP-free rate (DBPR) was not reached, while the 1-year DBPR was 60.0% (Fig. 2b). The median time to DBP was 5.5 (range 1.0-18.5; n=38) months in the subgroup of patients experiencing distant intracranial progression. The cumulative LMS rate was 4.9% with a median time to LMS of 6.2 (4.2-18.2) months (Fig. 2c). The median OS after IORT was 26 (1.2-not reached) months and the 1-year OS rate 74.0% (Fig. 2d). The initiation of IT/TT within two months following IORT was associated with a non-significant trend towards prolongation of both distant brain control and overall survival (Fig. 2e). There were no variables significantly associated with local recurrence or RN in uni- or multivariate analysis, while DS-GPA provided the best prognostic

separation (HR 0.05, p=0.173) for local recurrence. However, median-based classification of the dose prescription (≤24 Gy vs. ≥25 Gy) showed a trend for increased RN risk (p=0.158; Suppl. Fig. 1), but not local recurrence (p>0.999), BDP-FS (p=0.782) or OS (p=0.318). Age (p=0.022) and DS-GPA (p=0.049) were significantly associated with OS in multivariate analysis.

Discussion

In contrast to the preceding era of uniform chemotherapy, ITs have reshaped the landscape of oncology dramatically within very few years towards precision-tailored treatments. This success is due to promising efficacy in a growing number of tumor entities and good patient tolerability with a relatively favorable toxicity profile, also in combination with other local or systemic therapies. We here provide first proof that IORT is an overall well-tolerated combination partner for ICI and other novel TTs.

Particularly in highly immunogenic entities such as melanoma there are several reports of synergistic systemic effects of combined focal RT and systemic IT, often referred to as "abscopal effect" (29, 30). However, the brain was long considered a privileged organ where the underlying mechanisms do not apply due to the filtering properties of the BBB, thus preventing sufficient penetration of the tumor tissue and limiting bioavailability of the drugs (8) in an *a priori* immune-cold, secluded microenvironment (31). Nevertheless, there are numerous clinical case reports of abscopal systemic tumor response following high-dose RT of BMs, particularly with concomitant IT (32, 33). Recent advances in research have shed more light on the characteristics of the immunologic tumor microenvironment of BMs claiming a very distinct, yet non-negligible role of the immune system for the brain compartments (34–36). RT generates neo-antigens (37), activates non-redundant immune pathways in the tumor (38) and increases permeability of the BBB, thus improving brain penetration of Its/TTs (39). These mechanisms make RT a specifically interesting combination partner for targeted approaches in entities and individual patients considered non-responsive to treatment (40).

Independent of prognostic factors, BM resection necessitates additional RT to improve local tumor control. Nonetheless, depending on individual tumor features and clinical context, it remains controversial which RT sequencing and technique achieves best long-term outcomes at lowest toxicity levels. Our observed 1-year LCR of 97.1% is in line with previous reports on IORT (16, 19) and furthermore strengthens the notion that this RT technique might be superior to both definitive and adjuvant EBRT regimens where LCRs of 85 to 90% can be expected at most (12, 14, 15, 41–43). Yet, prospective trials are required to confirm this hypothesis. A large pooled analysis with 179 patients assessed very recently outcomes for the combination of SBRT and IT (44) reporting a LCR of 94.2% and a cumulative ≥grade 2 RN rate of 6.9% after a median FU of 14.8 months. Notably, the median diameter of the investigated lesions was only 7 mm. With a median lesion diameter of 34 mm, we provide with IORT plus IT/TT a treatment rationale with particularly good outcome and tolerability for large lesions. Of note, the tumor lesions reported here are measured presurgically for obvious technical reasons, but adjuvant EBRT faces the dilemma of about 30% target volume increase (45). This additionally strengthens the data provided here for these already presurgically large lesions with a median volume of 22.9 cm^3. Furthermore, larger lesion size was not associated with inferior outcome in this collective. Besides good local tumor control, we also demonstrated convincing intracranial control with a 1-year DBPS of 61.1% and a cumulative LMS rate of only 4.8%. Even though the exact underlying mechanisms remain unknown and require further scientific attention, a positive effect of the instant dose application thus preventing intracranial or leptomeningeal spread of tumor cells from around the resection cavity appears reasonable. Besides this timely eradication of remaining tumor cells, IORT may synergistically prevent the re-establishment of a protumorigenic tumor microenvironment. IT and TT may benefit from the high-dose local RT effects facilitating antigen presentation and subsequent immune-stimulatory properties, thereby enabling more effective killing of distantly circulating tumor cells (24). Proteomic profiles of wound fluids from breast cancer patients exhibited an abrogation of pathways promoting migration and invasiveness following IORT, which may particularly explain the LCRs and DB-PFS observed (46). The kV photon energy of IORT furthermore encompasses a compared to MV energy 1.3 to 1.5 times higher relative biologic effectiveness (RBE) (47), possibly overcoming typical limitations of common RT dosing like tumor

hypoxia, repair and reduced radiosensitivity of surviving tumor cells (48). On the contrary, the particularly local dose distribution of IORT (20) prevents tumoricidal effects of in principal undesired distant dose exposure which may occur in EBRT and which represent the primary rationale for whole brain RT (15) where not visible tumor burden but the complete potentially tumor cell-baring compartment is targeted to prevent intracranial spread. Yet, the healthy brain-sparing properties of IORT both prevent neurological and cognitive impairment of the patients and allow for targeted reirradiation in case of distant recurrence.

The "one-stop-shop" characteristic of IORT enables timely admission to subsequent systemic treatments while reducing in-hospital times (49) and might furthermore allow for earlier reduction of often necessary systemic corticosteroids compared to EBRT, which is a known risk factor for TT efficacy predominantly in the early initiation phase (50). Although the OS reported in this series needs to be interpreted cautiously due to its retrospective nature and potential selection bias, we additionally provide first evidence of encouraging survival outcomes following combinatorial treatment, at least non-inferior to previous reports on IORT (18) but also a matched retrospective comparison of IORT and EBRT cases (19). As mentioned, this is hypothesis-generating and should be evaluated within a prospective clinical trial.

Overall, our data indicate good tolerability and a favorable safety profile of this combinatorial approach. While predominantly confirming a lack of sufficient data for most drugs, a systematic meta-analysis previously reported generally acceptable toxicity of cranial stereotactic EBRT with IT (51). Yet, TTs and particularly BRAF inhibitors were associated with a high risk of severe toxicity (51) which we cannot confirm for our IORT cohort. The toxicity reported here is rather mild and in line with previous reports on IORT, that did not specifically address IT/TT cases (21, 22). Of note, only a minority of IORT patients of previous series received concomitant systemic therapy at all. Patel et al. observed a non-significant trend towards higher RN incidence for RT and ipilimumab versus RT only (52). Regardless of this, the reported RN rate of 30% exceeds significantly the cumulative RN rate of 5.7% presented here, despite the numerous patients in this collective receiving

duplet immune checkpoint blockade, which is associated with increased toxicity (2), let alone a less favorable toxicity profile in combination with SRT (53). Similar to previous retrospective single-center reports (16–19), IORT patients seem to have a very low RN risk which is not altered by concomitant IT/TT.

Timing of IT matters, but the optimal sequence of and time intervals between RT and IT remain controversial. Patient- and tumor-centered factors cannot be excluded to additionally influence this question. The PACIFIC trial showed strong evidence for sequential durvalumab treatment in locally advanced lung cancer with a time gap of at least one day, but up to six weeks (1). In the RTOG 3505 trial, IT with nivolumab was initiated four to twelve weeks after RT (54). However, a large retrospective analysis noted improved clinical outcome when ICI were started at least one month prior to RT (55). This divergence prompted us to assess the IORT+IT effects in a wide time range of treatment initiation and to investigate possible timing effects. Within the low toxicity collective reported here, we notably observed increased toxicity for the subgroup of patients commencing their systemic treatment not before, but in the first two months following resection and IORT. Additionally, we noted a trend towards improved clinical outcome in both of these groups compared to even later initiation of IT. While requiring confirmation in prospective data, this would contradict the common concept of preventing increased perioperative risks by decidedly long post-surgery treatment gaps but suggest a benefit both in regard to outcome and tolerability for even earlier, pre-interventional initiation of the systemic treatment.

Wound dehiscence is a common complication following BM resection (56, 57) with reported increased incidence for synchronous IT in head and neck cancer (58). In this series, we observed not a single case of wound dehiscence rendering IORT safe for patients with BMs receiving IT/TT. Notably, there is a well-known risk for wound infections with concomitant bevacizumab (59), which was underrepresented in our collective with just two patients receiving this VEGF pathway-TT. Our data are thus in accordance with previous reports that claim fewer toxicity for cranial RT with bevacizumab than for extracranial RT (51). It is worth highlighting, that two patients were *a priori*

removed from the analyses due to receiving an additional sequential SRT boost after IORT. One of these patients with a RCC receiving the VEGF-targeting multikinase inhibitor axitinib and avelumab later suffered from both a wound dehiscence requiring surgical intervention and a RN grade 3. Our observations raise suspicion over the safety of this treatment combination, while also the RT prescription requires reconsideration, as IORT with a sequential SRT boost was previously reported to be related to higher toxicity (18). Although IORT to BMs without sequential SRT boosting appears safe independent of the dose prescription, the results of the multivariate analyses suggest to limit the dose to 25 Gy to the surface. This limits the IORT duration and thus the window of risk for anesthesia side effects, but may also be protective for RN while non-inferior in regard to clinical outcome. Again, this will have to be confirmed in larger prospective trials.

Our study carries several limitations. The retrospective nature of the assessment may cause an incomplete portrait of toxicity in comparison to controlled prospective clinical trials, as well as patient selection bias. This is particularly important since the multicenter aspect additionally attributes to heterogeneity in this regard. Notably, most of our patients presented with lung primary histology. Other histologies, such as breast cancer, were underrepresented. Furthermore, the IORT dose prescription and FU protocols of the contributing centers were not derived from a single trial and not homogenized, which may impact the generalizability of the findings. Given the current small number of IORT patients in this setting, randomized prospective data are yet required. Our efforts thus mark a first step towards a multicentric, prospective study of IORT cases in the world-wide centers to ease the interpretation of its therapeutic value. This is the largest investigation on an IORT patient cohort thus far, incorporating patient data from over a hundred BM treatments in six international, tertiary-referral centers and it is the first assessment of IORT as a potential combination partner for IT and TT approaches, paving the way to a more patient-centered, fast and safe individual care for patients with BMs.

Conclusions

The combination of IT/TT with IORT for resected BMs does not seem to increase toxicity, while yielding encouraging local control and leptomeningeal spread rates, particularly for large brain metastases. Times between surgery and systemic treatment should be shortened with this approach, as timely admission to systemic therapy was associated with a trend towards improved clinical outcome. A prospective clinical trial will elucidate the actual role of IORT in this setting.

References

1. Antonia SJ, Villegas A, Daniel D, *et al.* Overall Survival with Durvalumab after Chemoradiotherapy in Stage III NSCLC. *N Engl J Med*. 2018;379:2342–2350.
2. Larkin J, Chiarion-Sileni V, Gonzalez R, *et al.* Combined Nivolumab and Ipilimumab or Monotherapy in Untreated Melanoma. *N Engl J Med*. 2015;373:23–34.
3. Davis FG, Dolecek TA, McCarthy BJ, *et al.* Toward determining the lifetime occurrence of metastatic brain tumors estimated from 2007 United States cancer incidence data. *Neuro-Oncology*. 2012;14:1171–1177.
4. Lamba N, Wen PY, Aizer AA. Epidemiology of brain metastases and leptomeningeal disease. *Neuro Oncol*. 2021;23:1447–1456.
5. Cagney DN, Martin AM, Catalano PJ, *et al.* Incidence and prognosis of patients with brain metastases at diagnosis of systemic malignancy: a population-based study. *Neuro Oncol*. 2017;19:1511–1521.
6. Chamberlain MC, Baik CS, Gadi VK, *et al.* Systemic therapy of brain metastases: non-small cell lung cancer, breast cancer, and melanoma. *Neuro Oncol*. 2017;19:i1–i24.
7. Ishiyama H, Teh BS, Ren H, *et al.* Spontaneous Regression of Thoracic Metastases While Progression of Brain Metastases After Stereotactic Radiosurgery and Stereotactic Body Radiotherapy for Metastatic Renal Cell Carcinoma: Abscopal Effect Prevented by the Blood-Brain Barrier? *Clinical Genitourinary Cancer*. 2012;10:196–198.
8. Di Giacomo AM, Valente M, Cerase A, *et al.* Immunotherapy of brain metastases: breaking a "dogma." *J Exp Clin Cancer Res*. 2019;38:419.
9. Yamamoto M, Sato Y, Serizawa T, *et al.* Subclassification of Recursive Partitioning Analysis Class II Patients With Brain Metastases Treated Radiosurgically. *International Journal of Radiation Oncology*Biology*Physics*. 2012;83:1399–1405.
10. Schödel P, Schebesch K-M, Brawanski A, *et al.* Surgical Resection of Brain Metastases—Impact on Neurological Outcome. *IJMS*. 2013;14:8708–8718.
11. Verhaak E, Gehring K, Hanssens PEJ, *et al.* Health-related quality of life of patients with brain metastases selected for stereotactic radiosurgery. *J Neurooncol*. 2019;143:537–546.
12. Mahajan A, Ahmed S, McAleer MF, *et al.* Post-operative stereotactic radiosurgery versus observation for completely resected brain metastases: a single-centre, randomised, controlled, phase 3 trial. *The Lancet Oncology*. 2017;18:1040–1048.
13. Lehrer EJ, Peterson JL, Zaorsky NG, *et al.* Single versus Multifraction Stereotactic Radiosurgery for Large Brain Metastases: An International Meta-analysis of 24 Trials. *International Journal of*

*Radiation Oncology*Biology*Physics*. 2019;103:618–630.
14. Eitz KA, Lo SS, Soliman H, *et al.* Multi-institutional Analysis of Prognostic Factors and Outcomes After Hypofractionated Stereotactic Radiotherapy to the Resection Cavity in Patients With Brain Metastases. *JAMA Oncol*. 2020;6:1901.
15. Brown PD, Ballman KV, Cerhan JH, *et al.* Postoperative stereotactic radiosurgery compared with whole brain radiotherapy for resected metastatic brain disease (NCCTG N107C/CEC·3): a multicentre, randomised, controlled, phase 3 trial. *Lancet Oncol*. 2017;18:1049–1060.
16. Cifarelli CP, Brehmer S, Vargo JA, *et al.* Intraoperative radiotherapy (IORT) for surgically resected brain metastases: outcome analysis of an international cooperative study. *J Neurooncol*. 2019;145:391–397.
17. Kahl K-H, Balagiannis N, Höck M, *et al.* Intraoperative radiotherapy with low-energy x-rays after neurosurgical resection of brain metastases-an Augsburg University Medical Center experience. *Strahlenther Onkol*. 2021;197:1124–1130.
18. XXXX
19. Layer JP, Hamed M, Potthoff A-L, *et al.* Outcome assessment of intraoperative radiotherapy for brain metastases: results of a prospective observational study with comparative matched-pair analysis. *J Neurooncol*. 2023.
20. Sarria GR, Smalec Z, Muedder T, *et al.* Dosimetric Comparison of Upfront Boosting With Stereotactic Radiosurgery Versus Intraoperative Radiotherapy for Glioblastoma. *Front Oncol*. 2021;11:759873.
21. Hamed M, Potthoff A-L, Layer JP, *et al.* Benchmarking Safety Indicators of Surgical Treatment of Brain Metastases Combined with Intraoperative Radiotherapy: Results of Prospective Observational Study with Comparative Matched-Pair Analysis. *Cancers*. 2022;14:1515.
22. Krauss P, Steininger K, Motov S, *et al.* Resection of supratentorial brain metastases with intraoperative radiotherapy. Is it safe? Analysis and experiences of a single center cohort. *Front Surg*. 2022;9:1071804.
23. Demaria S, Bhardwaj N, McBride WH, *et al.* Combining radiotherapy and immunotherapy: a revived partnership. *Int J Radiat Oncol Biol Phys*. 2005;63:655–666.
24. Weichselbaum RR, Liang H, Deng L, *et al.* Radiotherapy and immunotherapy: a beneficial liaison? *Nat Rev Clin Oncol*. 2017;14:365–379.
25. Haslam A, Prasad V. Estimation of the Percentage of US Patients With Cancer Who Are Eligible for and Respond to Checkpoint Inhibitor Immunotherapy Drugs. *JAMA Netw Open*. 2019;2:e192535.
26. Sperduto PW, Kased N, Roberge D, *et al.* Summary Report on the Graded Prognostic Assessment: An Accurate and Facile Diagnosis-Specific Tool to Estimate Survival for Patients With Brain Metastases. *JCO*. 2012;30:419–425.
27. Marks LB, Yorke ED, Jackson A, *et al.* Use of Normal Tissue Complication Probability Models in the Clinic. *International Journal of Radiation Oncology*Biology*Physics*. 2010;76:S10–S19.
28. Lin NU, Lee EQ, Aoyama H, *et al.* Response assessment criteria for brain metastases: proposal from the RANO group. *The Lancet Oncology*. 2015;16:e270–e278.
29. Grimaldi AM, Simeone E, Giannarelli D, *et al.* Abscopal effects of radiotherapy on advanced melanoma patients who progressed after ipilimumab immunotherapy. *Oncoimmunology*. 2014;3:e28780.
30. Koller KM, Mackley HB, Liu J, *et al.* Improved survival and complete response rates in patients with advanced melanoma treated with concurrent ipilimumab and radiotherapy versus ipilimumab alone. *Cancer Biol Ther*. 2017;18:36–42.
31. Engelhardt B, Vajkoczy P, Weller RO. The movers and shapers in immune privilege of the CNS. *Nat Immunol*. 2017;18:123–131.
32. Lin X, Lu T, Xie Z, *et al.* Extracranial abscopal effect induced by combining immunotherapy with

brain radiotherapy in a patient with lung adenocarcinoma: A case report and literature review. *Thorac Cancer*. 2019;10:1272–1275.
33. Suwinski R. Combination of immunotherapy and radiotherapy in the treatment of brain metastases from non-small cell lung cancer. *J Thorac Dis*. 2021;13:3315–3322.
34. Louveau A, Smirnov I, Keyes TJ, *et al.* Structural and functional features of central nervous system lymphatic vessels. *Nature*. 2015;523:337–341.
35. Quail DF, Joyce JA. The Microenvironmental Landscape of Brain Tumors. *Cancer Cell*. 2017;31:326–341.
36. Strickland MR, Alvarez-Breckenridge C, Gainor JF, *et al.* Tumor Immune Microenvironment of Brain Metastases: Toward Unlocking Antitumor Immunity. *Cancer Discov*. 2022;12:1199–1216.
37. Lhuillier C, Rudqvist N-P, Yamazaki T, *et al.* Radiotherapy-exposed CD8+ and CD4+ neoantigens enhance tumor control. *J Clin Invest*. 2021;131:e138740, 138740.
38. Twyman-Saint Victor C, Rech AJ, Maity A, *et al.* Radiation and dual checkpoint blockade activate non-redundant immune mechanisms in cancer. *Nature*. 2015;520:373–377.
39. Sprowls SA, Arsiwala TA, Bumgarner JR, *et al.* Improving CNS Delivery to Brain Metastases by Blood-Tumor Barrier Disruption. *Trends Cancer*. 2019;5:495–505.
40. Lussier DM, Alspach E, Ward JP, *et al.* Radiation-induced neoantigens broaden the immunotherapeutic window of cancers with low mutational loads. *Proc Natl Acad Sci U S A*. 2021;118:e2102611118.
41. Minniti G, Scaringi C, Paolini S, *et al.* Single-Fraction Versus Multifraction (3 × 9 Gy) Stereotactic Radiosurgery for Large (>2 cm) Brain Metastases: A Comparative Analysis of Local Control and Risk of Radiation-Induced Brain Necrosis. *Int J Radiat Oncol Biol Phys*. 2016;95:1142–1148.
42. Lehrer EJ, Ahluwalia MS, Gurewitz J, *et al.* Imaging-defined necrosis after treatment with single-fraction stereotactic radiosurgery and immune checkpoint inhibitors and its potential association with improved outcomes in patients with brain metastases: an international multicenter study of 697 patients. *Journal of Neurosurgery*. 2022:1–10.
43. XXXX
44. Hall J, Lui K, Tan X, *et al.* Factors associated with radiation necrosis and intracranial control in patients treated with immune checkpoint inhibitors and stereotactic radiotherapy. *Radiotherapy and Oncology*. 2023;189:109920.
45. Jarvis LA, Simmons NE, Bellerive M, *et al.* Tumor bed dynamics after surgical resection of brain metastases: implications for postoperative radiosurgery. *Int J Radiat Oncol Biol Phys*. 2012;84:943–948.
46. Belletti B, Vaidya JS, D'Andrea S, *et al.* Targeted intraoperative radiotherapy impairs the stimulation of breast cancer cell proliferation and invasion caused by surgical wounding. *Clin Cancer Res*. 2008;14:1325–1332.
47. Liu Q, Schneider F, Ma L, *et al.* Relative Biologic Effectiveness (RBE) of 50 kV X-rays measured in a phantom for intraoperative tumor-bed irradiation. *Int J Radiat Oncol Biol Phys*. 2013;85:1127–1133.
48. Herskind C, Ma L, Liu Q, *et al.* Biology of high single doses of IORT: RBE, 5 R's, and other biological aspects. *Radiat Oncol*. 2017;12:24.
49. Dejonckheere CS, Layer JP, Hamed M, *et al.* Intraoperative or postoperative stereotactic radiotherapy for brain metastases: time to systemic treatment onset and other patient-relevant outcomes. *J Neurooncol*. 2023.
50. Maslov DV, Tawagi K, Kc M, *et al.* Timing of steroid initiation and response rates to immune checkpoint inhibitors in metastatic cancer. *J Immunother Cancer*. 2021;9:e002261.
51. Kroeze SGC, Fritz C, Hoyer M, *et al.* Toxicity of concurrent stereotactic radiotherapy and

targeted therapy or immunotherapy: A systematic review. *Cancer Treatment Reviews*. 2017;53:25–37.
52. Patel KR, Shoukat S, Oliver DE, *et al.* Ipilimumab and Stereotactic Radiosurgery Versus Stereotactic Radiosurgery Alone for Newly Diagnosed Melanoma Brain Metastases. *American Journal of Clinical Oncology*. 2017;40:444–450.
53. Bodensohn R, Werner S, Reis J, *et al.* Stereotactic radiosurgery and combined immune checkpoint therapy with ipilimumab and nivolumab in patients with melanoma brain metastases: A retrospective monocentric toxicity analysis. *Clinical and Translational Radiation Oncology*. 2023;39:100573.
54. Gerber DE, Urbanic JJ, Langer C, *et al.* Treatment Design and Rationale for a Randomized Trial of Cisplatin and Etoposide Plus Thoracic Radiotherapy Followed by Nivolumab or Placebo for Locally Advanced Non–Small-Cell Lung Cancer (RTOG 3505). *Clinical Lung Cancer*. 2017;18:333–339.
55. Samstein R, Rimner A, Barker CA, *et al.* Combined Immune Checkpoint Blockade and Radiation Therapy: Timing and Dose Fractionation Associated with Greatest Survival Duration Among Over 750 Treated Patients. *International Journal of Radiation Oncology*Biology*Physics*. 2017;99:S129–S130.
56. Barami K, Fernandes R. Incidence, risk factors and management of delayed wound dehiscence after craniotomy for tumor resection. *Journal of Clinical Neuroscience*. 2012;19:854–857.
57. Gupta S, Dawood H, Giantini Larsen A, *et al.* Surgical and Peri-Operative Considerations for Brain Metastases. *Front Oncol*. 2021;11:662943.
58. Mays AC, Yarlagadda B, Achim V, *et al.* Examining the relationship of immunotherapy and wound complications following flap reconstruction in patients with head and neck cancer. *Head Neck*. 2021;43:1509–1520.
59. Sharma K, Marcus JR. Bevacizumab and wound-healing complications: mechanisms of action, clinical evidence, and management recommendations for the plastic surgeon. *Ann Plast Surg*. 2013;71:434–440.

Table captions

Table 1: Patient characteristics for the evaluated BMs (n=103).

BM: brain metastasis; DS-GPA: Diagnostic-Specific Graded Prognostic Assessment; KPS: Karnofsky performance score; NSCLC: non-small cell lung cancer; RCC: renal cell carcinoma; RT: radiotherapy; SCLC: small cell lung cancer.

Variable	**n (%)**	**Median (range)**
Gender		
Male	56 (54.4)	
Female	47 (45.6)	
Age (years)		63 (35-85)
Tumor entity		

NSCLC	53 (51.5)	
Melanoma	25 (24.3)	
RCC	13 (12.6)	
Breast	4 (3.9)	
SCLC	3 (2.9)	
Others	5 (4.9)	
Localization		
Frontal lobe	36 (35.0)	
Parietal lobe	28 (27.2)	
Occipital lobe	18 (17.5)	
Temporal lobe	13 (12.6)	
Cerebellum	8 (7.8)	
Max. pre-surgical diameter (mm)		34 (8-70)
Pre-surgical tumor volume (ccm)		22.9 (1.2-701.7)
Multiple BMs	48 (46.6)	
Number of BMs		1 (1-16)
RT to other BMs	50 (48.5)	
Relevant overlap (≥10% isodose)	14 (13.6)	
Extracranial metastases	66 (64.1)	
KPS		80 (40-100)
DS-GPA		2 (0-4)

Table 2: Treatment characteristics (n=103).

* Some patients received both IT and TT in parallel or combinations of either substance group. IT: immunotherapy; IORT: intraoperative radiotherapy; MKI: multikinase inhibitor; SRT: stereotactic radiotherapy; TKI: tyrosine kinase inhibitor; TT: targeted therapy.

Variable	**n (%)**	**Median (range)**
IORT dose (Gy)		25 (16-30)
18	5 (4.9)	
20	40 (38.8)	
24	4 (3.9)	
25	3 (2.9)	
26	1 (1.0)	
30	50 (48.5)	
Applicator diameter (mm)		20 (15-40)
Time from first diagnosis to IORT (months)		1 (0-297)
Immune checkpoint inhibitor	90 (87.4)*	
Pembrolizumab	36 (40.0)	
Ipilimumab + nivolumab	20 (22.2)	
Atezolizumab	16 (15.5)	
Nivolumab	13 (14.4)	
Durvalumab	4 (4.4)	
Ipilimumab	1 (1.1)	
Time from IORT to IT (months)		1.1 (-22.3-34)
Number of IT cycles		6 (1-93)
TT drug	25 (24.3)*	
BRAF/MEK inhibitor	6 (24.0)	
TKI	5 (20.0)	
MKI	6 (24.0)	
VEGF targeting*	6 (24.0)	

Androgen deprivation	3 (12.0)	
Anti-Her2neu	3 (12.0)	
Anti-TNFa	1 (4.0)	
Time from IORT to TT (months)		1.2 (-38.9-22.9)
Duration of TT treatment (months)		7 (2-68)

Table 3: Summary of adverse events (n=147).

* Fulminant auto-immune hepatitis unrelated to IORT but likely related to pembrolizumab.

** One patient suffered from an ICI-related auto-immune vasculitis of grade 4 under pembrolizumab and another patient experienced an unrelated cardiac infarction causing pulmonary vein congestion and ultimately an atypical pneumonia.

*** An 81-year-old patient with lung cancer experienced reactivation of a pre-existing chronic lymphocytic leukemia ultimately causing his demise following septicemia. Another patient deceased due to a distant brain progression-related intracranial bleeding four months after IORT.

	Acute events	**Late events**	**All events**
Grade	**n (%)**	**n (%)**	**n (%)**
Grade 1	22 (44.9)	43 (43.9)	65 (44.2)
Grade 2	20 (40.8)	18 (18.4)	38 (25.9)
Grade 3	6 (12.2)	33 (33.7)	39 (26.5)
Grade 4	1 (2.0)*	2 (2.0)**	3 (2.0)
Grade 5	0 (0.0)	2 (2.0)***	2 (1.4)
Any grade	49 (33.3)	98 (66.7)	147 (100.0)

* Fulminant auto-immune hepatitis unrelated to IORT but likely related to pembrolizumab.
** One patient suffered from an ICI-related auto-immune vasculitis of grade 4 under pembrolizumab and another patient experienced an unrelated cardiac infarction causing pulmonary vein congestion and ultimately an atypical pneumonia.

*** An 81-year-old patient with lung cancer experienced reactivation of a pre-existing chronic lymphocytic leukemia ultimately causing his demise following septicemia. Another patient deceased due to a distant brain progression-related intracranial bleeding four months after IORT.

Supp. Table 1: Full list of adverse events.

Figure captions

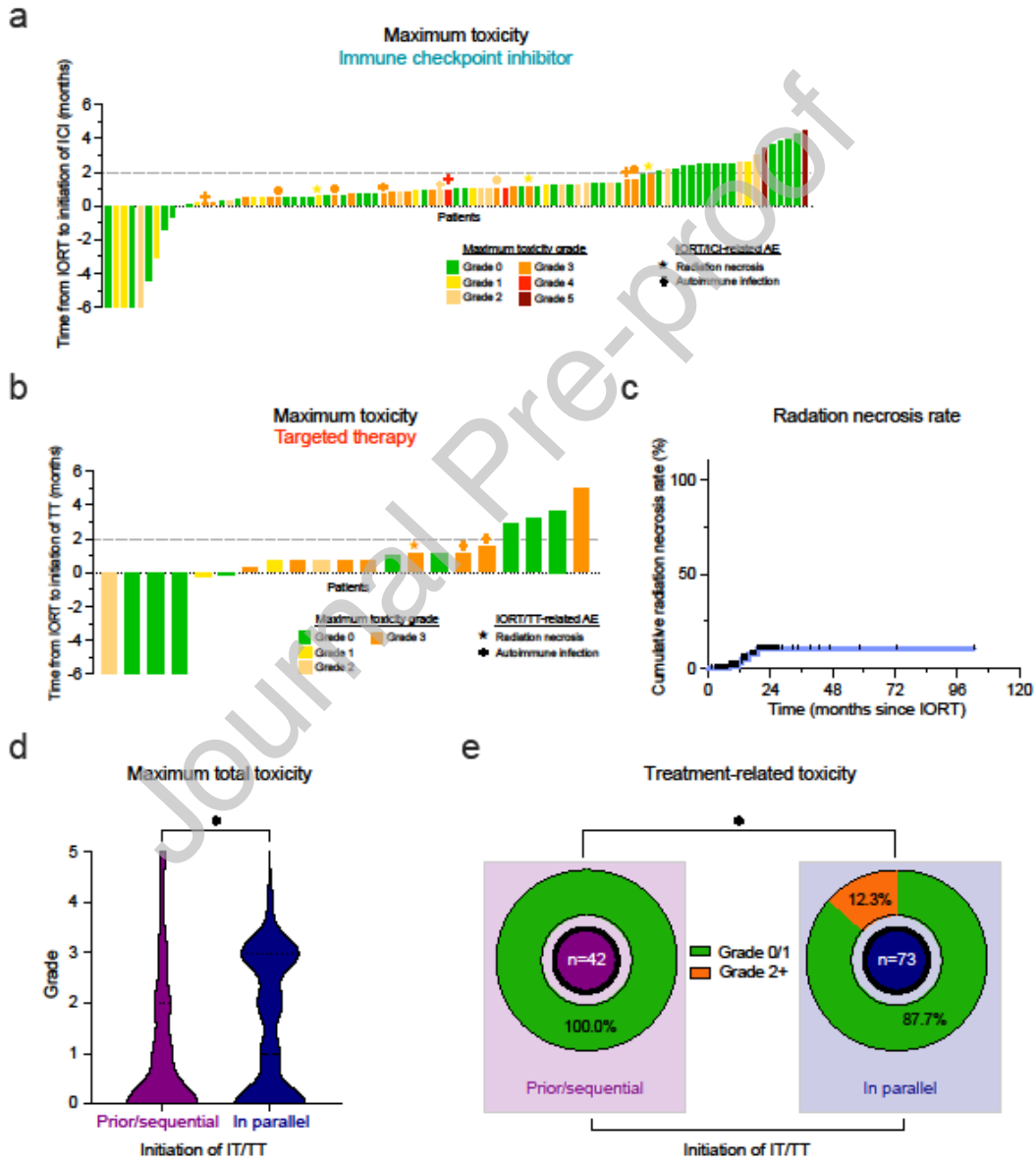

Fig. 1: Toxicity profile of IORT with immune checkpoint inhibition or other targeted therapies. **a, b** Waterfall plot illustrating time from IORT to initiation of ICI therapy (a) and TT (b) for each individual patient with treatment initiation between 6 months prior to and after IORT. Color labeling represents the maximum overall toxicity observed as per CTCAE grading (independent of relation to treatment) and icons symbolize occurrence of therapy-associated AEs (IORT/ IT/TT-related AEs). **c** Cumulative radiation necrosis rate (%) over time in months since IORT. **d** Violin plots demonstrating the distribution of the maximum reported toxicity as per CTCAE grading for patients receiving IT/TT prior or sequential (later than 2 months following surgery) to IORT compared to treatment initiation in parallel with IORT (within 2 months following surgery). Dashed lines indicate the median. * $p<0.05$, Mann-Whitney test. **e** Donut chart depicting maximum grade IORT/IT/TT-related toxicity (≤ grade 1 vs. ≥ grade 2) depending on time point of IT/TT initiation as defined in (d). * $p>0.05$, Fisher's exact test. AE: adverse event; CTCAE: Common Terminology Criteria for Adverse Events; ICI: immune checkpoint inhibitor: IORT: intraoperative radiotherapy; IT: immunotherapy; TT: targeted therapy.

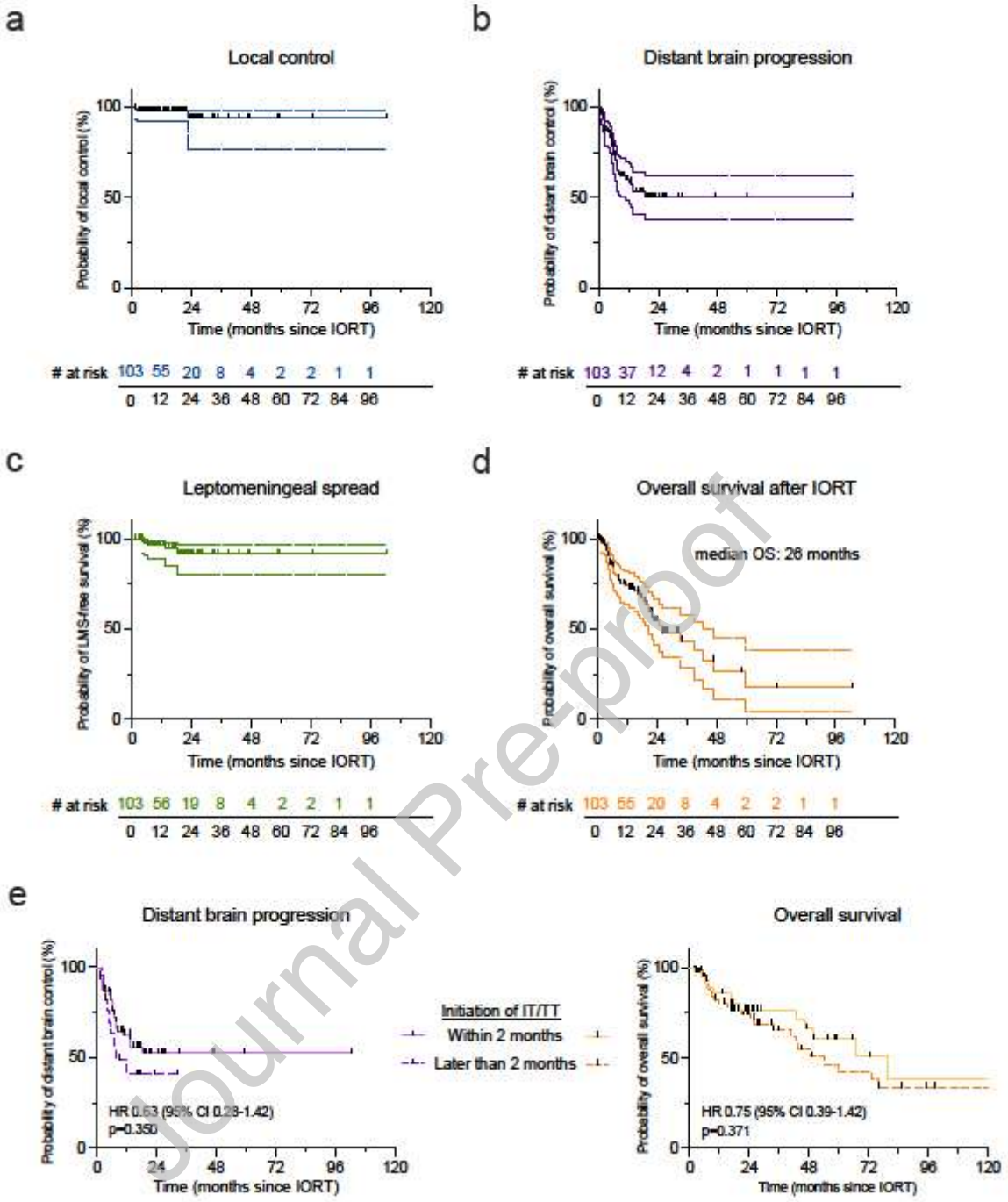

Fig. 2: Outcome parameters of IORT with immune checkpoint inhibition and other targeted therapies. **a,b,c,d** Kaplan-Meier plots depicting percentual local control rate (a), distant brain progression rate (b), leptomeningeal spread rate (c) and overall survival (d) over time in months since IORT. Dashed lines indicate 95% confidence intervals. **e,f** Kaplan-Meier plots for distant brain progression (e) and overall survival (f) dependent on timepoint of IT/TT initiation (within vs. later than 2 months after IORT); HR with CI and p indicated in the lower left corner and calculated for initiation of IT/TT

within 2 months vs. later than 2 months. Log-rank test. CI: confidence interval; HR: Hazard ratio; IT: immunotherapy; LMS: leptomeningeal spread; OS: overall survival; TT: targeted therapy.

Suppl. Fig. 1: Dose dependency of radiation necrosis rate. Kaplan-Meier plot for cumulative radiation necrosis rate dependent on IORT prescription dose (≤24 Gy vs. ≥25 Gy).); HR with CI and p indicated in the upper left corner and calculated for dose prescription of 24 Gy or less vs. 25 Gy or more. Log-rank test. CI: confidence interval; HR: Hazard ratio.