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LABORATORY INVESTIGATION

Safety, feasibility, and optimization of intra-arterial mitoxantrone delivery to gliomas

Jason A. Ellis[1] · Johann Cooke[3] · Rajinder P. Singh-Moon[2] · Mei Wang[3] · Jeffrey N. Bruce[1] · Charles W. Emala[3] · Irving J. Bigio[4] · Shailendra Joshi[3]

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Abstract Mitoxantrone is a highly cytotoxic antineoplastic drug, however, its poor penetration of the blood–brain barrier has limited its role in the treatment of brain cancers. We hypothesize that intra-arterial (IA) delivery of mitoxantrone may enhance its capacity for regional brain deposition thus expanding its potential as a brain tumor therapy agent. In this study we assessed the dose-response characteristics as well as the feasibility and safety of mitoxantrone delivery to the brain and specifically to gliomas in a rodent model. We show that delivery optimization utilizing the technique of intra-arterial transient cerebral hypoperfusion (IA-TCH) facilitates achieving the highest peak- and end- brain drug concentrations as compared to intravenous and IA delivery without hypoperfusion. Additionally, we observed significant tumor-specific uptake of mitoxantrone when delivered by the IA-TCH method. No untoward effects of IA-TCH delivery of mitoxantrone were observed. The IA-TCH method is shown to be a safely tolerated and feasible strategy for delivering mitoxantrone to tumors in the glioma model tested. Additional investigation is warranted to determine if IA-TCH delivery of mitoxantrone produces clinically relevant benefit.

✉ Jason A. Ellis
jae2109@cumc.columbia.edu

[1] Department of Neurological Surgery, Columbia University Medical Center, New York, NY, USA

[2] Department of Electrical Engineering, Columbia University, New York, NY, USA

[3] Department of Anesthesiology, Columbia University Medical Center, New York, NY, USA

[4] Departments of Electrical and Biomedical Engineering, Boston University, Boston, MA, USA

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Introduction

Mitoxantrone is semi-synthetic non-cell cycle specific antimitotic drug that has been used for the treatment of solid tumors [1–4]. It is poorly lipid soluble and thus has limited efficacy in the brain when delivered systemically. Loco-regional delivery of mitoxantrone has been shown to be effective in both experimental and human glioma studies [5–10]. Loco-regional delivery has several disadvantages, however, stemming from the need for direct tumor access. Intra-arterial (IA) delivery is an alternative, minimally invasive strategy that may be useful for targeting mitoxantrone to brain tumors.

In order to improve IA drug delivery for treating brain tumors we have suggested the use of transient cerebral hypoperfusion (TCH) during IA drug injections (IA-TCH). Using this strategy we have demonstrated improved efficacy in delivering lipid soluble agents such as carmustine, cationic liposomes, and lipid micelles to the brain [11–14]. Similar flow arrest techniques have been used clinically in human subjects to better target brain stem gliomas and cerebral vascular malformations as well as for breast and liver cancer treatment [15–17]. This technique is particularly attractive for the cationic mitoxantrone given its presumed affinity to the negative charge of the cerebral capillary endothelium.

While it has been previously reported that brain uptake of mitoxantrone after IA injections is enhanced by osmotic disruption of the blood brain barrier (BBB), the safety, feasibility, and optimization of physiological parameters for IA delivery to glioma-bearing animals has yet to be reported

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[18–20]. Thus, the goals of this study are threefold: (1) to demonstrate the dose-response characteristics of IA mitoxantrone delivery to the brain, (2) to demonstrate the feasibility of targeted mitoxantrone delivery to gliomas using the IA-TCH method, and (3) to demonstrate the short-term safety of IA mitoxantrone when delivered to glioma-bearing animals.

Materials and methods

Surgical preparation

All methods were approved by the Columbia University Institutional Review Board and the Animal Care and Use Committee. Sprague Dawley rats weighing 200–300 g were utilized. Anesthesia commenced with isoflurane induction, supplemental intramuscular ketamine, and placement of a tail vein cannula. Femoral arterial line placement and selective internal carotid artery (ICA) cannulation was then performed. The specific techniques for placement and obtaining laser Doppler blood flow measurements as well as for diffuse reflectance spectroscopy to assess tissue drug concentrations have been previously described [12, 18, 21]. During experiments, anesthesia was maintained by inhaled isoflurane. Vital signs were continuously recorded by an A-D instrument on a Mac Lab system. TCH was produced by intravenous (IV) administration of adenosine and esmolol (2 mg each) flushed with a bolus of cold saline (1.5 ml).

Dose response study and dose justification

Hemodynamic parameters (heart rate, mean arterial pressure, electrocardiographic activity, change in CBF) were recorded at baseline, at the peak of mitoxantrone injection (5 min) and at the end of the experiment (30 min after injection). To disrupt the blood–brain barrier, IA mannitol (0.25 mg/kg/s) was injected over 30 s. IA mitoxantrone was delivered through the ICA catheter using a Parker microinfusion pump controlled by a signal generator. The brain parenchymal concentrations of mitoxantrone were analyzed at peak and at the end.

A mitoxantrone dose of 12 mg/m^2 is typically utilized for clinical applications. Rarely, high dose therapy with 20–60 mg/m^2 can be used, however significant side effects have been reported [22, 23]. Given the body surface area of a 250 g rats is 0.04 m^2, we estimated that a dose of 0.48 mg corresponds to the standard 12 mg/m^2 dose of mitoxantrone used in clinical practice [24]. We therefore selected a dose range of 0.1–1 mg (1 ml) for these experiments. Four mitoxantrone concentrations of 0.1, 0.25, 0.5, and 1.0 mg/ml were used. Twelve total rats were injected (n = 4 IV, n = 4 IA, n = 4 IA-TCH) for this experiment.

Tumor implantation and harvest

Rats were stereotactically injected with 10^6 C-6 glioma cells. Injections were made with a 33 gauge Hamilton syringe after the animals were placed on a Knopf stereotactic frame. The coordinate used was 4 mm below the outer skull table and 3 mm behind and lateral to bregma. Tumors were allowed to grow for 10 days after cell implantation prior to starting drug delivery experiments. A total of 12 rats underwent tumor implantation followed by IA mitoxantrone delivery. Peak and end tissue concentrations were determined by sacrificing animals at 5 min (n = 6) and at 4 h (n = 6) respectively. Tumors were harvested by careful dissection along its margins and compared to the same volume of brain tissue in the corresponding contralateral hemisphere.

Histological analysis

Harvested brains were fixed, paraffin embedded, and cut into 1 mm thick cross sections. Slices were stained with hematoxylin and eosin (H&E) and/or with TUNEL immunoperoxidase to assess apoptosis. The tumor, surrounding peritumoral brain, and contralateral hemisphere were qualitatively assessed for evidence of toxicity including disruption of architecture, changes in neuron density and shape, and the presence of necrosis or hemorrhage.

Data analysis

Data were collected at specified time points as described. Statistical significance was set at $p < 0.05$. Stat View 5.2 (SAS Institute, Cary, North Carolina) was used for statistical calculations.

Results

IA-TCH improves mitoxantrone delivery to brain

Healthy animals without brain tumors (n = 12) demonstrated a dose-dependent increase in hemispheric mitoxantrone concentrations after IV, IA, and IA-TCH delivery (Fig. 1a, b). Peak and end concentrations were highest for the IA-TCH delivery method and negligible for IV delivery. By definition, IA-TCH was associated with a decrease in mean arterial pressure and heart rate during injection. In comparison, IA and IV delivery were not associated with changes in these physiological parameters (Table 1).

An additional ten healthy animals without brain tumors were studied to determine the effect of IA-TCH on mitoxantrone brain deposition. In these experiments, 0.5 mg of mitoxantrone were injected intra-arterially either with (IA-TCH, n = 5) or without (IA, n = 5) TCH. The peak concentrations

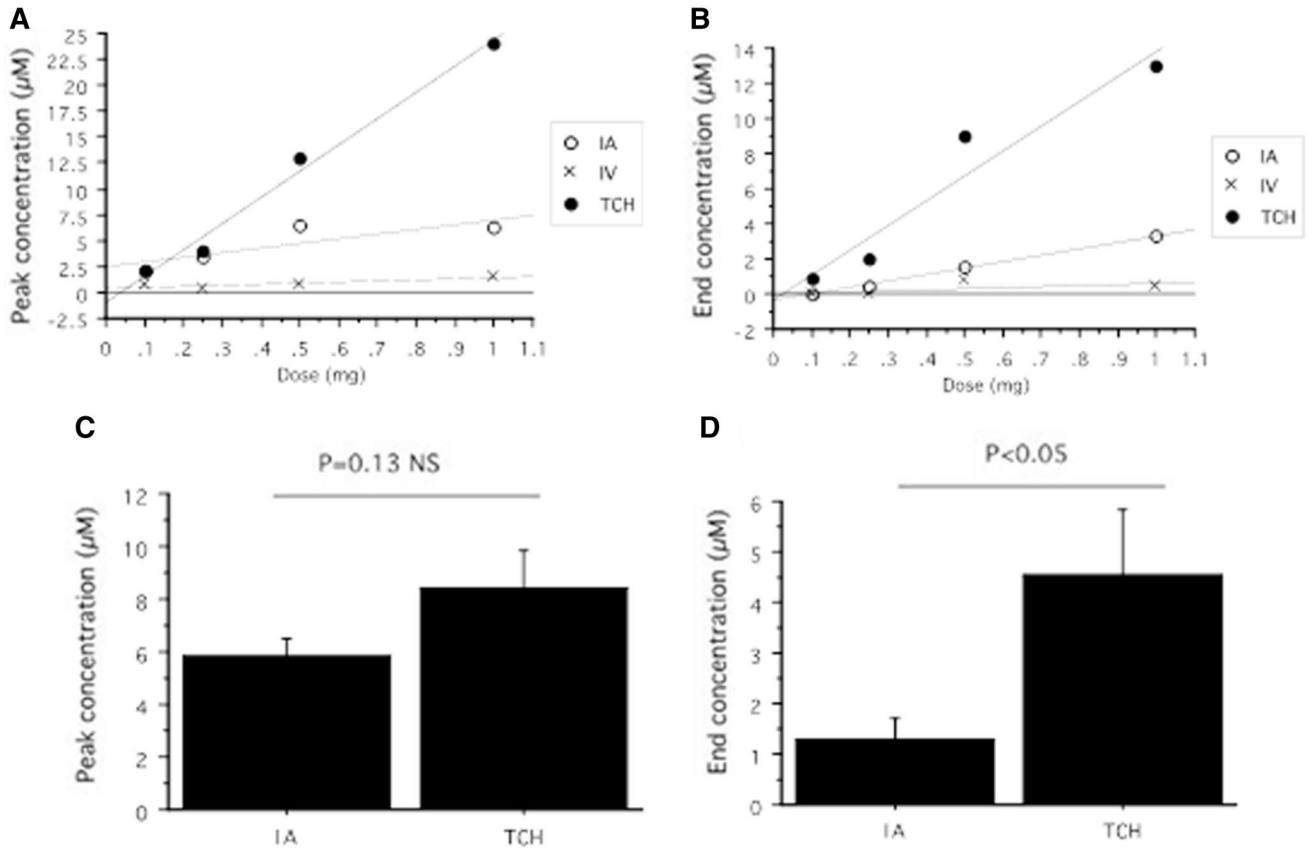

Fig. 1 Mitoxantrone levels in the brain. Graphs show the dose-response characteristics of IA, IA-TCH, and IV injection of 0.1, 0.25, 0.5, and 1 mg of mitoxantrone in each animal (**a, b**). Peak and end concentrations increase approximately linearly with increasing injection doses. IA-TCH injection show the highest efficiency of brain mitoxantrone delivery as compared to IA-only and IV injections. Transient cerebral hypoperfusion (TCH) significantly increases the end but not peak concentration of mitoxantrone delivered to the brain intra-arterially (**c, d**)

showed a trend toward higher mitoxantrone levels when IA-TCH was utilized, however the advantage did not meet statistical significance. On the other hand, IA-TCH delivery resulted in significantly increased ($p < 0.05$) end brain tissue concentrations (Fig. 1c, d). Thus, IA-TCH facilitates ipsilateral hemispheric brain deposition of mitoxantrone over IA delivery alone.

IA-TCH facilitates targeted delivery of mitoxantrone to C-6 gliomas

Mitoxantrone concentrations in harvested glioma tissue was considerably increased over the concentrations measured in the contralateral, non-tumor bearing brain tissue (Fig. 2). Peak and end concentrations of mitoxantrone in tumor tissue were more than tenfold greater than in the corresponding non-tumor tissue ($p < 0.05$). Importantly, therapeutic concentrations (12 μM) of mitoxantrone were achieved in the tumor tissue for at least 4 h during the course of these experiments. Conversely, the extremely low concentrations of mitoxantrone measured in non-tumor brain tissue supports the hypothesis that glioma-specific targeting occurs during IA-TCH. Fluorescence confocal microscopic imaging confirmed robust mitoxantrone deposition within the tumor (Fig. 3) and rare deposition elsewhere.

IA-TCH delivery of mitoxantrone to brain tumors can be safely tolerated

C-6 glioma-bearing rats injected with mitoxantrone (0.5 mg, $n = 5$) or saline ($n = 4$) via the IA-TCH method were studied to assess the short-term safety of the protocol utilized. All animals recovered from anesthesia without behavioral evidence of neurotoxicity prior to sacrifice. At the time of sacrifice 3 days after injection, tumor volumes were 44 and 45 mm^3 ($p > 0.05$) in the saline and mitoxantrone-treated groups, respectively. The degree of apoptosis was likewise no different between the two groups. No histopathological evidence of differential neural injury was observed between the two groups.

Table 1 Key hemodynamic changes during mitoxantrone delivery

	Group	Baseline	Injection	Peak	End
HR (bpm)	IV	269 ± 50	280 ± 26	280 ± 34	274 ± 46
% LD (Br)	IV	100 ± 0	103 ± 14	112 ± 22	77 ± 23
MAP (mmHg)	IV	55 ± 17	53 ± 13	71 ± 16	59 ± 9
Temp (R) (°C)	IV	37.7 ± 1.2	37.6 ± 1.1	37.6 ± 1.0	37.5 ± 0.5
HR (bpm)	IA	310 ± 13	245 ± 112	303 ± 19	302 ± 11
% LD (Br)	IA	100 ± 0	97 ± 19	132 ± 26	90 ± 42
MAP (mmHg)	IA	61 ± 13	49 ± 16	66 ± 19	64 ± 15
Temp (R) (°C)	IA	38.1 ± 0.6	37.8 ± 0.9	37.8 ± 0.9	36.8 ± 0.9[#]
HR (bpm)	IA-TCH	276 ± 57	77 ± 24[#]	252 ± 39	294 ± 46
% LD (Br)	IA-TCH	100 ± 0	11 ± 7[#]	125 ± 27	75 ± 24
MAP (mmHg)	IA-TCH	55 ± 7	20 ± 2[#]	48 ± 8	54 ± 14
Temp (R) (°C)	IA-TCH	37.1 ± 1.5	36.4 ± 1.7	36.4 ± 1.7	36.8 ± 1.2

IA Intra-arterial, *IA-TCH* intra-arterial with transient cerebral hypoperfusion, *HR* heart rate (beats/min), *%LD* (*Br*) percent change in laser Doppler blood flow from baseline in the brain, *MAP* (*mm Hg*) Mean arterial blood pressure, *Temp* (*R*) rectal temperature

[#]Significantly different from other stages of the experiment ($P < 0.0083$)

Discussion

The three major findings we herein report are that (1) TCH facilitates the IA delivery of mitoxantrone to the brain, (2) mitoxantrone can be specifically targeted at therapeutic levels to the site of glioma growth utilizing IA-TCH delivery, and (3) mitoxantrone is safely tolerated after IA-TCH delivery acutely and in the short-term. These new findings in concert with the known efficacy of loco-regional mitoxantrone against high grade glioma certainly set the stage for translation of IA-TCH delivery in a clinical setting. Although mitoxantrone was neither developed nor previously optimized for IA delivery to malignant gliomas our pre-clinical experiments suggests a workable role.

Intra-arterial drug delivery was initially proposed for the treatment of brain cancers more than a half century ago [25]. Although beneficial effects of this strategy have been noted for a number of systemic cancers, no significant benefits have been realized for the treatment of malignant gliomas. Recent glioma treatment clinical studies have focused on the IA delivery on safer agents such as carboplatin and bevacizumab, which may have limited direct cytotoxic effects [26–29]. However, agents that are highly tumoricidal but with increased systemic toxicity such as mitoxantrone may offer increased effectiveness. IA delivery may be a useful strategy for administering such agents specifically to the site of the tumor growth while limiting systemic side effects. Thus, we initiated preliminary pre-clinical experiments to study the safety, feasibility, and optimization techniques for IA mitoxantrone delivery to gliomas.

We hypothesized that cationic drugs such as mitoxantrone are particularly suited for the IA delivery techniques we employ. IA chemotherapy drugs that are rapidly taken up by the tumor during their first pass through the cerebral

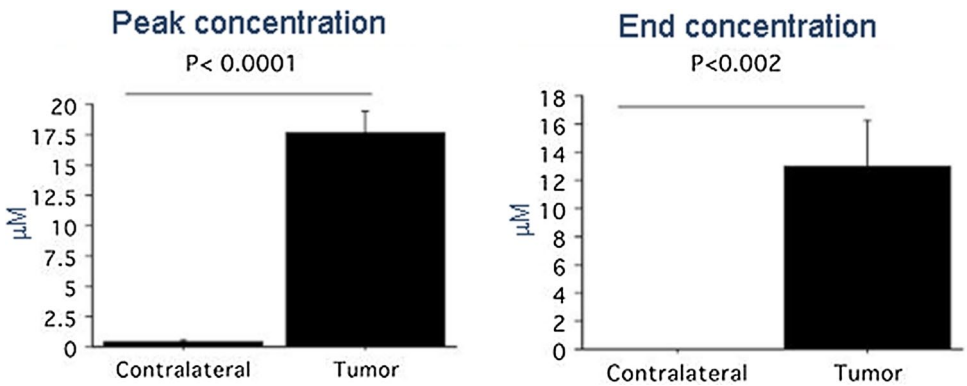

Fig. 2 Mitoxantrone delivery to gliomas. Significant retention of mitoxantrone was evident within tumor tissue ipsilateral to the side of IA delivery. The levels of mitoxantrone measured in the corresponding contralateral tissue was minimal both at peak (5 min) ad at the end of the experiment (4 h)

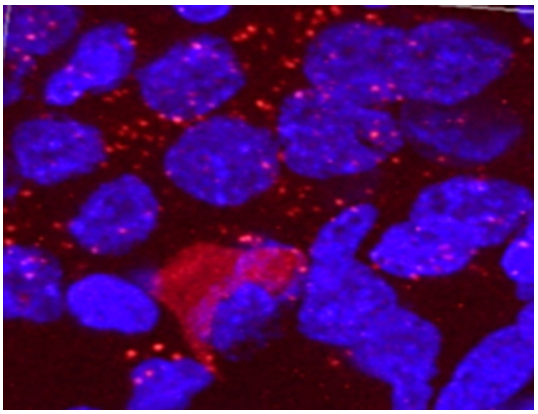

Fig. 3 Immunofluorescence of mitoxantrone in glioma tissue. Confocal microscopy indicates effective delivery of mitoxantrone molecules (*red*) to the site of glioma cells (nuclei stained *blue* with DAPI)

circulation are ideal. The attractive force between the positively charged drug molecules and the negatively charged endothelial cells likely facilitates at least a component of the delivery specificity we report. Furthermore, as anionic surface charges are overexpressed on tumor cell membranes, tumor specificity is also conferred. Although not specifically studied, we believe that an additional mediator of mitoxantrone's tumor specificity in this model is the inherent incompetence of the blood–tumor barrier. Of note, we did not pretreat tumor-bearing animals with IA mannitol so as to maintain the blood–brain barrier in areas of the brain that did not have infiltrated tumor.

The IA-TCH technique that we employed was an intentional strategy based on computational models of ideal IA drug delivery [30, 31]. These models show that IA drug delivery is most efficient when there is low cerebral blood flow, high regional extraction, and high systemic clearance. Although the brain has relatively high resting blood flow, this parameter is easily manipulated experimentally. High cerebral blood flow dilutes IA drugs, decreases their transit time through regional circulation, and leads to drug maldistribution due to streaming. As we have shown, a transient reduction in blood flow greatly improves drug delivery since it significantly increases the local arterial blood concentration and virtually delivers pure drug to the capillary endothelium. Drug binding with serum proteins is decreased and shear stress on the drug molecules as they interact with the endothelium is reduced as well.

While we did not intent for this study to investigate the anti-glioma response of mitoxantrone after IA-TCH, it is noteworthy that we did not observe one. There was no difference noted between mitoxantrone-treated animals and controls in terms of tumor size, apoptosis, or histopathological features up to 3 days after drug delivery. This may be for a number of reasons including that higher doses of mitoxantrone or a longer duration of exposure are needed to observe therapeutic effects in vivo. Future dose escalation, tumor response, and survival studies will be needed to address these issues.

Conclusion

Mitoxantrone can be safely and specifically targeted to gliomas in vivo by the technique of IA-TCH. Future studies are warranted to investigate the therapeutic efficacy and clinical translation of this strategy for the treatment of malignant gliomas.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval All applicable international, national, and/or institutional guidelines for the care and use of animals were followed. All procedures performed in this study were in accordance with the ethical standards of the Columbia University Institutional Review Board and the Animal Care and Use Committee.

References

1. Durr FE (1984) Biologic and biochemical effects of mitoxantrone. Semin Oncol 11(3 Suppl 1):3–10
2. Faulds D, Balfour JA, Chrisp P, Langtry HD (1991) Mitoxantrone. A review of its pharmacodynamic and pharmacokinetic properties, and therapeutic potential in the chemotherapy of cancer. Drugs 41(3):400–449
3. Jordan JP, Hand CM, Markowitz RS, Black P (1992) Test for chemotherapeutic sensitivity of cerebral gliomas: use of colorimetric MTT assay. J Neurooncol 14(1):19–35
4. White RJ, Durr FE (1985) Development of mitoxantrone. Invest New Drugs 3(2):85–93
5. Boiardi A, Bartolomei M, Silvani A, Eoli M, Salmaggi A, Lamperti E, Milanesi I, Botturi A, Rocca P, Bodei L, Broggi G, Paganelli G (2005) Intratumoral delivery of mitoxantrone in association with 90-Y radioimmunotherapy (RIT) in recurrent glioblastoma. J Neurooncol 72(2):125–131
6. Boiardi A, Eoli M, Pozzi A, Salmaggi A, Broggi G, Silvani A (1999) Locally delivered chemotherapy and repeated surgery can improve survival in glioblastoma patients. Ital J Neurol Sci 20(1):43–48
7. Boiardi A, Eoli M, Salmaggi A, Lamperti E, Botturi A, Broggi G, Bissola L, Finocchiaro G, Silvani A (2005) Systemic temozolomide combined with loco-regional mitoxantrone in treating recurrent glioblastoma. J Neurooncol 75(2):215–220
8. Ferroli P, Broggi M, Franzini A, Maccagnano E, Lamperti M, Boiardi A, Broggi G (2006) Surgifoam and mitoxantrone in the glioblastoma multiforme postresection cavity: the first step of locoregional chemotherapy through an ad hoc-placed catheter: technical note. Neurosurgery 59(2):E433–E434 (**discussion E433-434**)
9. Saini M, Roser F, Hussein S, Samii M, Bellinzona M (2004) Intralesional mitoxantrone biopolymer-mediated chemotherapy

prolongs survival in rats with experimental brain tumors. J Neurooncol 68(3):225–232
10. Yemisci M, Bozdag S, Cetin M, Soylemezoglu F, Capan Y, Dalkara T, Vural I (2006) Treatment of malignant gliomas with mitoxantrone-loaded poly (lactide-co-glycolide) microspheres. Neurosurgery 59(6):1296–1302 (**discussion 1302–1293**)
11. Joshi S, Wang M, Etu JJ, Suckow RF, Cooper TB, Feinmark SJ, Bruce JN, Fine RL (2007) Transient cerebral hypoperfusion enhances intraarterial carmustine deposition into brain tissue. J Neurooncol 86(2):123–132
12. Joshi S, Singh-Moon RP, Wang M, Chaudhuri DB, Holcomb M, Straubinger NL, Bruce JN, Bigio IJ, Straubinger RM (2014) Transient cerebral hypoperfusion assisted intraarterial cationic liposome delivery to brain tissue. J Neurooncol. doi:10.1007/s11060-014-1421-6
13. Joshi S, Singh-Moon R, Wang M, Chaudhuri DB, Ellis JA, Bruce JN, Bigio IJ, Straubinger RM (2014) Cationic surface charge enhances early regional deposition of liposomes after intracarotid injection. J Neurooncol 120(3):489–497. doi:10.1007/s11060-014-1584-1
14. Nguyen J, Cooke JR, Ellis JA, Deci M, Emala CW, Bruce JN, Bigio IJ, Straubinger RM, Joshi S (2016) Cationizable lipid micelles as vehicles for intraarterial glioma treatment. J Neurooncol. doi:10.1007/s11060-016-2088-y
15. Pile-Spellman J, Young WL, Joshi S, Duong DH, Vang MC, Hartmann A, Kahn RA, Rubin DA, Prestigiacomo CJ, Ostapkovich ND (1999) Adenosine-induced cardiac pause for endovascular embolization of cerebral arteriovenous malformations: technical case report. Neurosurgery 44(4):881–886 (**discussion 886–887**)
16. Riina HA, Knopman J, Greenfield JP, Fralin S, Gobin YP, Tsiouris AJ, Souweidane MM, Boockvar JA (2010) Balloon-assisted superselective intra-arterial cerebral infusion of bevacizumab for malignant brainstem glioma. A technical note. Interv Neuroradiol 16(1):71–76
17. Aigner KR (1998) Intra-arterial infusion: overview and novel approaches. Semin Surg Oncol 14(3):248–253
18. Reif R, Wang M, Joshi S, A'Amar O, Bigio IJ (2007) Optical method for real-time monitoring of drug concentrations facilitates the development of novel methods for drug delivery to brain tissue. J Biomed Opt 12(3):034036
19. Joshi S, Reif R, Wang M, Zhang J, Ergin A, Bruce JN, Housepian EM, Fine RL, Bigio IJ (2011) Intraarterial mitoxantrone delivery in rabbits: an optical pharmacokinetic study. Neurosurgery 69(3):706
20. Ergin A, Wang M, Zhang J, Bigio I, Joshi S (2012) Noninvasive in vivo optical assessment of blood brain barrier permeability and brain tissue drug deposition in rabbits. J Biomed Opt 17(5):057008
21. Joshi S, Singh-Moon RP, Ellis JA, Chaudhuri DB, Wang M, Reif R, Bruce JN, Bigio IJ, Straubinger RM (2014) Cerebral hypoperfusion-assisted intraarterial deposition of liposomes in normal and glioma-bearing rats. Neurosurgery NIHMS 624641 (in press)
22. Toledano A, Azria D, Garaud P, Fourquet A, Serin D, Bosset JF, Miny-Buffet J, Favre A, Le Floch O, Calais G (2007) Phase III trial of concurrent or sequential adjuvant chemoradiotherapy after conservative surgery for early-stage breast cancer: final results of the ARCOSEIN trial. J Clin Oncol 25(4):405–410. doi:10.1200/JCO.2006.07.8576
23. Damon LE, Wolf JL, Rugo HS, Gold E, Zander AR, Cassidy M, Cecchi G, Cohen N, Irwin D, Tracy M, Ries CA, Linker CA (2000) High-dose chemotherapy (CTM) for breast cancer. Bone Marrow Transplant 26(3):257–268. doi:10.1038/sj.bmt.1702481
24. Charest G, Sanche L, Fortin D, Mathieu D, Paquette B (2012) Glioblastoma treatment: bypassing the toxicity of platinum compounds by using liposomal formulation and increasing treatment efficiency with concomitant radiotherapy. Int J Radiat Oncol Biol Phys 84(1):244–249. doi:10.1016/j.ijrobp.2011.10.054
25. Klopp CT, Alford TC, Bateman J, Berry GN, Winship T (1950) Fractionated intra-arterial cancer; chemotherapy with methyl bis amine hydrochloride; a preliminary report. Ann Surg 132(4):811–832
26. Boockvar JA, Tsiouris AJ, Hofstetter CP, Kovanlikaya I, Fralin S, Kesavabhotla K, Seedial SM, Pannullo SC, Schwartz TH, Stieg P, Zimmerman RD, Knopman J, Scheff RJ, Christos P, Vallabhajosula S, Riina HA (2010) Safety and maximum tolerated dose of superselective intraarterial cerebral infusion of bevacizumab after osmotic blood–brain barrier disruption for recurrent malignant glioma. Clinical article. J Neurosurg 114(3):624–632
27. Riina HA, Burkhardt JK, Santillan A, Bassani L, Patsalides A, Boockvar JA (2012) Short-term clinico-radiographic response to super-selective intra-arterial cerebral infusion of bevacizumab for the treatment of vestibular schwannomas in neurofibromatosis type 2. Interv Neuroradiol 18 (2):127–132
28. Riina HA, Fraser JF, Fralin S, Knopman J, Scheff RJ, Boockvar JA (2009) Superselective intraarterial cerebral infusion of bevacizumab: a revival of interventional neuro-oncology for malignant glioma. J Exp Ther Oncol 8(2):145–150
29. Shin BJ, Burkhardt JK, Riina HA, Boockvar JA (2012) Superselective intra-arterial cerebral infusion of novel agents after blood–brain disruption for the treatment of recurrent glioblastoma multiforme: a technical case series. Neurosurg Clin of N Am 23(2):323–329, ix–x. doi:10.1016/j.nec.2012.01.008
30. Dedrick RL (1988) Arterial drug infusion: pharmacokinetic problems and pitfalls. J Natl Cancer Inst 80(2):84–89
31. Ellis JA, Banu M, Hossain SS, Singh-Moon R, Lavine SD, Bruce JN, Joshi S (2015) Reassessing the role of intra-arterial drug delivery for glioblastoma multiforme treatment. J Drug Deliv 2015:405735. doi:10.1155/2015/405735