LABORATORY INVESTIGATION - HUMAN/ANIMAL TISSUE

Pharmacokinetic analysis of etoposide distribution after administration directly into the fourth ventricle in a piglet model

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Abstract We hypothesize that infusion of chemotherapeutic agents directly into the fourth ventricle potentially may play a role in treating malignant posterior fossa brain tumors. Accordingly, we used a piglet model developed in our laboratory to test the safety of etoposide infusions into the fourth ventricle and to study the pharmacokinetics associated with these infusions. In 5 piglets, closed-tip silicone catheters were inserted into the fourth ventricle and lumbar cistern. Five consecutive daily infusions of etoposide (0.5 mg) were administered via the fourth ventricle catheter. Serum and CSF from both catheters were sampled for measurement of etoposide level by reversed-phase high performance liquid chromatography (HPLC). For CSF samples, area under the concentration-time curve (AUC) was calculated. Piglets underwent daily neurological examinations, a 4.7 Tesla MRI scan, and then were sacrificed for post-mortem brain examination. No neurological deficits or signs of meningitis were caused by intraventricular chemotherapy infusions. MRI scans showed catheter placement within the fourth ventricle but no signal changes in the brain stem or cerebellum. In all piglets, the mean fourth ventricular CSF peak etoposide level exceeded the mean peak lumbar etoposide levels by greater than 10-fold. Statistically significant differences between fourth ventricle and lumbar AUC were noted at peaks (ΔAUC = 3384196 ng h/ml with 95%CI: 1758625, 5009767, P = 0.0044) and at all collection time points (ΔAUC = 1422 977 ng h/ml with 95%CI: 732188, 2113766, P = 0.0046) but not at troughs (ΔAUC = −29546 ng h/ml (95%CI: −147526, 88434.2, P = 0.5251). Serum etoposide was absent at two and four hours after intraventricular infusions in all animals. Pathological analysis demonstrated meningitis, choroid plexitis, and ependymitis in the fourth and occasionally lateral ventricles. Etoposide can be infused directly into the fourth ventricle without clinical or radiographic evidence of damage. Autopsy examination revealed ventriculitis and meningitis which did not have a clinical correlate. Etoposide does not distribute evenly throughout CSF spaces after administration into the fourth

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ventricle, and higher peak CSF levels are observed in the fourth ventricle than in the lumbar cistern.

Introduction

Our laboratory developed a piglet model for direct infusion of chemotherapeutic agents into the fourth ventricle based upon the hypothesis that such infusions can potentially play a role in treating patients with malignant posterior fossa brain tumors [1]. This novel treatment approach may offer several advantages over current regimens used to treat medulloblastoma, the most common malignant brain tumor of childhood [2], as well as other tumors occurring in this location. Medulloblastoma typically arises from the cerebellar vermis and fills the fourth ventricle. Initial surgical resection is often incomplete due to adherence of tumor to the floor of the fourth ventricle [3]. Postoperative adjuvant treatment typically includes radiation therapy and systemic chemotherapy, both of which can be associated with significant morbidity, especially in young children. When medulloblastoma recurs, local recurrence is often accompanied by leptomeningeal spread via cerebrospinal fluid (CSF) pathways, emphasizing the importance of treatments which address both local disease and central nervous system (CNS) spread [4–6].

Previous human studies for various malignancies have utilized intrathecal or intraventricular chemotherapy in order to increase drug concentrations within the CSF while minimizing systemic exposure [7–10]. Treatment has been administered either by repeated lumbar punctures or via a ventricular access device (Ommaya reservoir) connected to a catheter which is inserted into the lateral ventricle of the brain [11]. There are several potential advantages of catheter placement into the fourth ventricle over lumbar or lateral ventricle infusions. Repeated lumbar punctures are painful, sometimes technically challenging, and often require sedation in children. Catheter placement into the lateral ventricle requires a separate surgical procedure, can be technically challenging in patients with small ventricles, and can be associated with multiple complications. Catheter malposition is relatively common [11–14], intraparenchymal hemorrhage can occur with passage of catheters through normal brain en route to the ventricle [11, 13], and symptomatic leukoencephalopathy can occur following drug administration when all catheter holes are not within the ventricle [11, 15–18]. Direct catheter placement into the fourth ventricle could be performed at the time of surgery for primary or recurrent tumor resection without requiring an additional operation. Because the catheter would be placed under direct vision without passage through any brain parenchyma, the risk of intraparenchymal hemorrhage would be eliminated. Moreover, placing the catheter under direct vision would ensure that all catheter holes are within the ventricle, thus reducing the risk of treatment-related leukoencephalopathy.

Previous work from our laboratory established the feasibility of this approach and demonstrated preliminary safety data for infusions of etoposide into the fourth ventricle in piglets [1]. The objective of the current study was to investigate the pharmacokinetics associated infusions into the fourth ventricle by measuring subsequent etoposide levels in CSF samples from the fourth ventricle and lumbar cistern.

Materials and methods

Surgical procedures

Experiments were conducted on five Yorkshire piglets, each weighing between 16 and 18 kg, with the approval of the University of Miami Miller School of Medicine Institutional Animal Care and Use Committee. Anesthesia was induced using intramuscular ketamine (40 mg/kg), xylazine (4 mg/kg), and acepromazine (0.4 mg/kg) followed by endotracheal intubation and mechanical ventilation. Central venous and arterial lines were placed for infusion of medications, blood pressure monitoring, and blood gas sampling. Anesthesia and paralysis were maintained with intravenous (IV) fentanyl (50 mcg/kg bolus then 10 mcg/kg/h continuous infusion), propofol (50–100 mcg/kg/min), and pancuronium (0.3 mg/kg every 30–60 min as needed). After completion of the surgical procedure, pancuronium was reversed with neostigmine (0.1 mg/kg IV) and glycopyrolate (0.02 mg/kg IV), and piglets were extubated.

Piglets were positioned prone with the head in a flexed position. The skin was infiltrated with 1% bupivicaine and an incision was made to expose the subocciput and the posterior elements of C1 and C2. Posterior cervical muscles and fascia were incised and retracted, and then a suboccipital craniectomy, C1 laminectomy, and partial C2 laminectomy were performed. The dura mater was opened and the inferior cerebellum was gently elevated to identify the obex. A closed-tip silicone lumbar drain catheter (Medtronic, product reference number 46419) that was precut to a length of 23 cm was placed into the fourth ventricle under direct vision to ensure that all catheter holes were within the fourth ventricle. The dura was closed in a water-tight fashion by first approximating the edges with sutures and then sealing a pericranial graft to the dura with a small amount of Instant Krazy Glue (Elmer's Products,

Inc.). Normal saline was infused into the catheter to ensure that the closure was water-tight. The catheter was tunneled through the skin and secured with sutures, and a luerlock connector was placed to allow subsequent access. The muscle, fascia, and skin were then sutured closed.

A separate incision in the lumbar region was made after infiltration of the skin with 1% bupivicaine. After a limited laminectomy and small dural opening, a closed-tip silicone lumbar drain catheter (Medtronic, product reference number 46419) that was pre-cut to a length of 23 cm was inserted rostrally into the subdural space. The dura was sutured closed around the catheter and a fascial graft was sealed to the dura with a small amount of Instant Krazy Glue (Elmer's Products, Inc.). Normal saline was infused into the catheter to ensure that the closure was water-tight. The catheter was tunneled through the skin and secured with sutures, and a luerlock connector was placed to allow subsequent access. The muscle, fascia, and skin were then sutured closed.

After closure of both wounds, the piglet was allowed to emerge from anesthesia and extubated. Femoral arterial lines were removed, and central venous catheters maintained for subsequent access.

Clinical assessment

Neurological examinations were performed at least once per day until sacrifice. Level of alertness was observed, and pigs were monitored for neck stiffness, lethargy, or other signs of infection. Gait was observed with particular attention paid to symmetric movement of each forelimb and hindlimb. Only a limited sensory examination, consisting of assessing limb withdrawal to touch, was possible. Examination of cranial nerve function included responses to visual and auditory threat, checking the corneal reflex (by dripping water into each eye), and observation of mouth movement and feeding. Eating and drinking patterns were noted.

Chemotherapy infusions and CSF and serum collection

Etoposide solution (Sicor Pharmaceuticals; Irvine, California) was diluted in sterile preservative-free normal saline so that each infusion contained 0.5 mg of etoposide in 0.25 ml of total volume. Immediately after each infusion, 0.25 ml of normal saline was infused to ensure that all drug was flushed out of the tubing and into the fourth ventricle. This volume was chosen based upon the measured dead space of approximately 0.15 ml in the pre-cut catheter.

Etoposide was infused into the fourth ventricle once daily for five consecutive days beginning 2 days after surgery. Piglets were sedated with IV propofol during chemotherapy infusions and collection of serum and CSF samples. CSF samples were obtained simultaneously from the fourth ventricle and lumbar catheters at 15 min and then 1, 2, 4, 8, 12, and 24 h time intervals after the first etoposide infusion. CSF samples were then obtained just prior to and 15 min after each subsequent etoposide infusion to monitor trough and peak levels. Each time CSF was sampled, 0.2 ml was first aspirated and discarded to ensure that the fluid sampled was from within the ventricle. 0.5 ml of CSF was then aspirated, and 0.7 ml of sterile, preservative-free normal saline was then infused to flush the catheter and replace the aspirated volume.

Serum samples (3 ml) were obtained 2 and 4 h after intraventricular etoposide infusion. CSF and serum samples were placed into heparinized tubes, centrifuged at 2,000 rpm for 2 min, and then stored at a temperature below −20 °C until pharmacokinetic analysis was performed.

CSF samples were also collected for gram stain and culture at the conclusion of the experiment (after administration of all chemotherapy doses). If adequate CSF could be obtained, a cell count was also performed.

Pharmacokinetic analysis of CSF and serum samples

Etoposide levels in CSF and serum samples were determined using reversed-phase high performance liquid chromatography (HPLC) as previously described by Reif et al. and in our previous publication [1, 19]. Investigators performing this analysis were blinded regarding the time of sample collection as well as whether each sample was from the fourth ventricle or lumbar catheter.

For CSF samples from the fourth ventricle and lumbar catheters, area under the concentration-time curves (AUC) by trapezoidal rule were calculated for each piglet for peak, for trough, and for all time points of etoposide measurement, respectively. The differences in mean AUCs were calculated by comparing mean AUCs between lumbar and fourth ventricle samples. Statistical significances of the differences in the mean AUCs were tested by paired t-tests at 5% significance level.

Magnetic resonance imaging (MRI) scans

MRI (magnetic resonance imaging) scans were performed after the completion of the five daily intraventricular etoposide infusions prior to sacrifice. A 4.7-Tesla (200 MHz) 40-cm bore magnet with a Bruker Avance™ console using an actively shielded gradient set was used. A homemade quadrature saddle shaped transmit-receive surface RF (radio frequency) coil was used for imaging the brain in order to increase sensitivity and reduce field-of-view sizes. For the MRI scans, piglets were sedated with IV propofol and then intubated. A new femoral arterial line was placed

for monitoring of arterial blood gases. Pancuronium (0.3 mg/kg IV every 30–60 min) was administered, and piglets were placed in an MRI-compatible cradle which was inserted into the MRI scanner. Sagittal and coronal T2-RARE (Rapid Acquisition Relaxation Enhanced) sequences and coronal T1-weighted Fluid Attenuated Inversion Recovery (FLAIR) sequences were obtained to assess catheter position and detect any signal changes in the brainstem or cerebellum.

Tissue preparation and histological analysis

At the conclusion of each experiment, piglets were fully anesthetized with IV propofol (10 mg/kg) and then killed with IV potassium chloride. Transcardiac perfusion/fixation of the brain was performed in situ using 10% buffered formalin as previously described [1]. Brains were then removed and placed in fixative for at least 1 week prior to cutting. Brain specimens were sectioned and then tranverse sections of medulla and pons with cerebellum and the dorsal hippocampi were embedded in paraffin, cut, and stained with hematoxylin and eosin. Sections were analyzed for evidence of inflammation, necrosis, and disruption of cytoarchitecture. Inflammation was characterized as "mild" if it consisted of relatively few inflammatory cells involving the leptomeninges or choroid plexus, "moderate" if focal infiltrates of many inflammatory cells were observed in the leptomeninges, choroid plexus, or ventricular surface, and "severe" if dense inflammatory infiltrates also involved subependymal brain parenchyma.

Results

Clinical findings

There were no neurological deficits attributed to chemotherapy administration into the fourth ventricle. All piglets were noted to have a normal level of alertness, and none exhibited neck stiffness, lethargy, or other signs of meningitis. One piglet (Piglet 3) was noted postoperatively (prior to chemotherapy infusion) to avoid bending the right hindlimb when walking, but this improved each day until the pig was walking and running normally by postoperative day 6. The right hindlimb had been the site of arterial line placement, and it is possible that the animal was experiencing pain from the procedure. Normal gait and symmetrical forelimb and hindlimb movements both spontaneously and in response to touch were observed in all other piglets throughout the experiment and in this piglet by the conclusion of the experiment. All piglets responded to auditory and visual threats by moving rapidly away from the stimuli. All piglets were noted to have normal corneal reflexes, spontaneous mouth movement, and appropriate feeding and drinking patterns. One piglet (Piglet 4) was noted to have stridor on postoperative day 4 attributed to likely airway obstruction, possibly from a mucous plug, and was briefly re-intubated. The animal was extubated several hours later and had no further difficulties for the remainder of the experiment.

Imaging findings

T2-RARE MRI scans demonstrated accurate catheter placement within the fourth ventricle in all five piglets (Fig. 1a, b). T1-weighted FLAIR sequences performed in all piglets did not demonstrate any signal changes in the brain stem or cerebellum (Fig. 1c). There was no evidence of ventriculomegaly in any piglets.

Histological analysis

Pathological analysis of post-mortem brain sections demonstrated an inflammatory response involving the meninges, choroid plexus, and ependyma of the fourth ventricle and, to a lesser extent, the lateral ventricles (Fig. 2a–c). This response was characterized as severe in 2 piglets (Piglets 1 and 2), moderate in 2 piglets (Piglets 3 and 4), and mild in one piglet (Piglet 5). In three piglets (piglets 1, 2, and 4), focal areas of necrosis very close to the ependymal surface were noted (Fig. 2d). Beyond the area just adjacent to the ependymal surface, none of the piglets demonstrated any histological evidence of necrosis, edema, or disruption of normal cytoarchitecture in the brain stem or cerebellum.

CSF Cultures were negative in piglets 1 and 2 and positive in piglets 3, 4, and 5 (for Staphlycoccus aureus, Pseudomonas fluroescens/putida, and Acinetobacter baumannii/haemolyticus, respectively). CSF cell counts were analyzed in 3 of 5 piglets (piglets 1, 2, and 5), and an elevated white blood cell count was noted in each. In the remaining 2 piglets, adequate CSF could not be obtained for cell count. Results of CSF analysis and correlation with autopsy findings of inflammation are listed in Table 1.

Pharmacokinetic analysis

Etoposide was not detectable in serum samples at 2 and 4 h after intraventricular etoposide infusion in all five piglets. Over the first 24 h, mean etoposide levels in fourth ventricular CSF peaked immediately and then steadily decreased. Mean lumbar etoposide levels started lower and then gradually increased until the four hour time point, at which time etoposide levels in the fourth ventricle and lumbar cistern were similar. This data is graphically displayed in Fig. 3.

Fig. 1 MRI scans obtained after 5 consecutive days of etoposide infusions into the fourth ventricle. **a** Sagittal T2-RARE MRI scan demonstrating catheter position (marked by *arrow*) within the fourth ventricle. **b** Coronal T2-RARE MRI scan demonstrating catheter position (marked by *arrow*) within the fourth ventricle. **c** Coronal T1-weighted FLAIR image through cerebellum and brainstem. No signal changes are observed
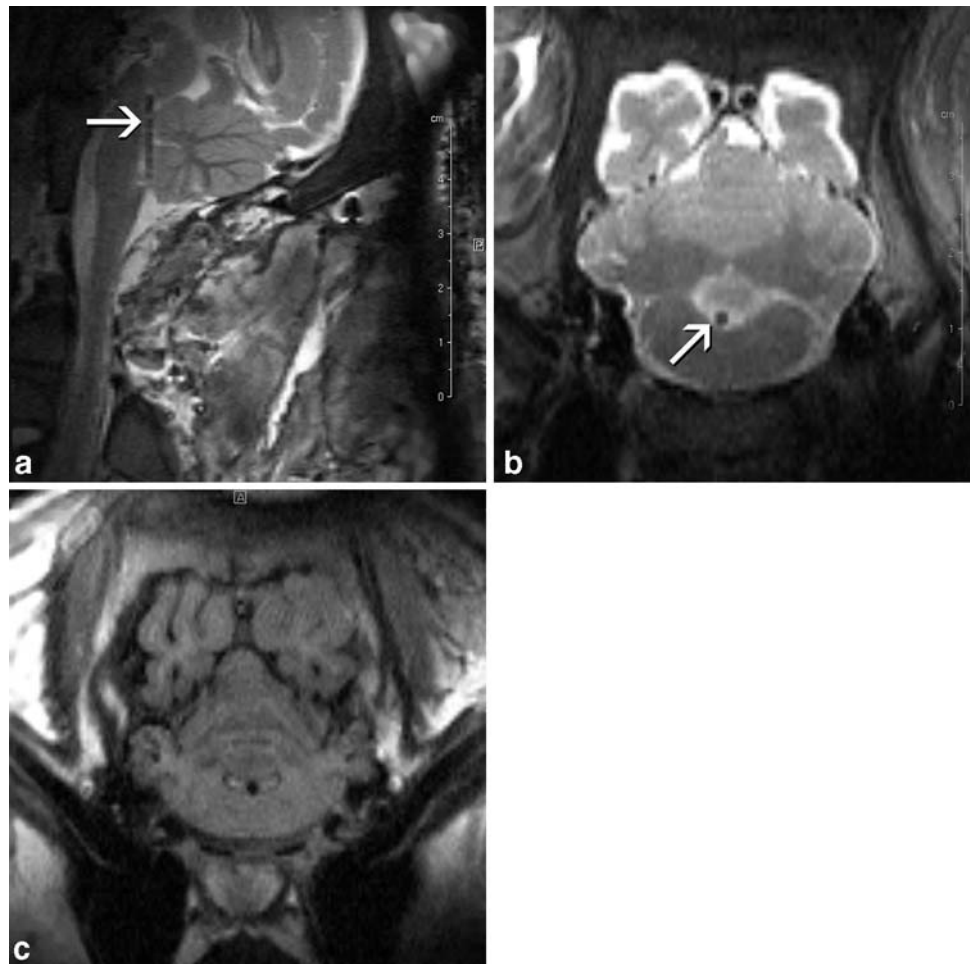

Fig. 2 Photomicrograhs of histological specimens obtained from piglets. **a** Base of brain stem section from Piglet 1 demonstrating severe meningitis (*arrows*). **b** Section through choroid plexus from Piglet 2 demonstrating profound inflammatory response of the choroid plexus (*arrow*). **c** Section through fourth ventricle in Piglet 3. Moderate inflammation is observed in the choroid plexus and subependymal zone (*arrow*) with preservation of normal cytoarchitecture in the brain stem and cerebellum beyond the area just adjacent to the ependymal surface. **d** Section through fourth ventricle in Piglet 2 showing foci of subependymal necrosis (*arrow*)
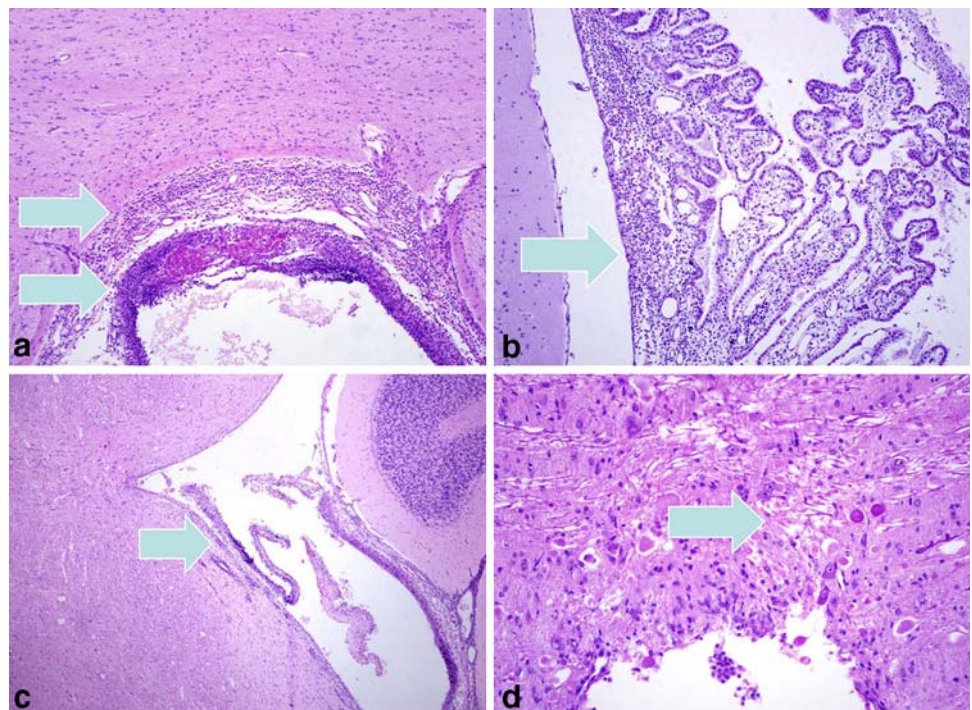
Table 1 CSF culture and cell count results after completion of 5 daily doses of intraventricular etoposide and correlation with autopsy findings of inflammation

	Culture	Cell count	Inflammation
1	Negative	1212 RBC	Severe
		308 WBC (47% P, 35% M, 18% L)	
2	Negative	259 RBC	Severe
		621 WBC (4% P, 50% M, 46% L)	
3	Staphlycoccus aureus	Not measured—inadequate specimen	Moderate
4	Pseudomonas fluroescens/putida	Not measured—inadequate specimen	Moderate
5	Acinetobacter baumannii/ haemolyticus	12312 RBC	Mild
		408 WBC (40% P, 7% M, 53% L)	

RBC red blood cells, *WBC* white blood cells, *P* polymorphonuclear cells, *M* monocytes, *L* Lymphocytes
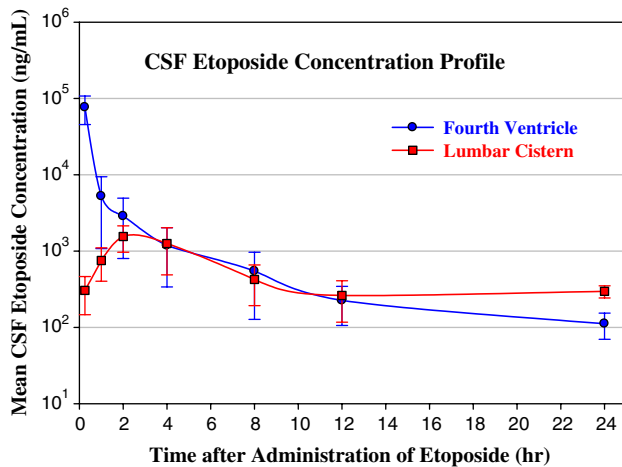

Fig. 3 Mean ± Standard Error (SE) CSF etoposide levels in the fourth ventricle and lumbar cistern over the first 24 h after etoposide infusion into the fourth ventricle. CSF Etoposide Concentration Profile first 24 h (Mean + Standard Error)
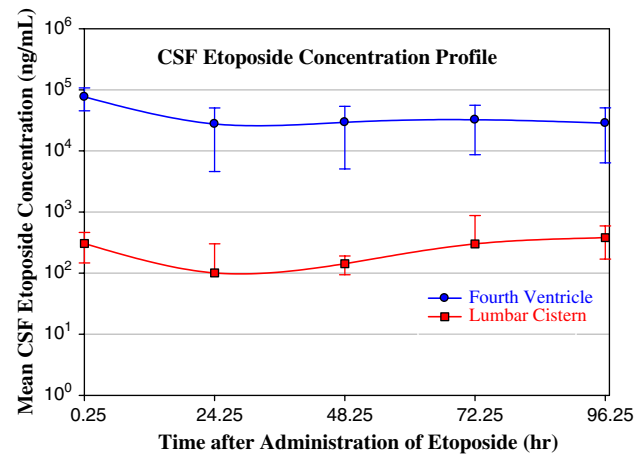

Fig. 4 Mean ± SE peak CSF etoposide levels in the fourth ventricle and lumbar cistern after five consecutive daily infusions into the fourth ventricle. CSF Etoposide Concentration Profile at Peaks (Mean + Standard Error)
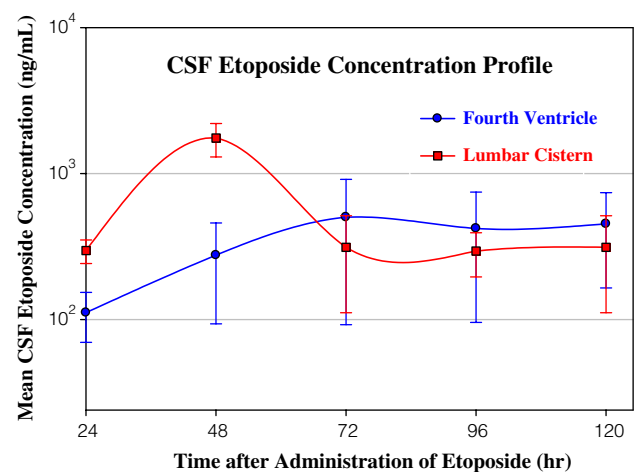
In all piglets, the mean fourth ventricular peak CSF etoposide level exceeded the mean peak lumbar etoposide level by at least 10-fold. Mean peak CSF etoposide levels from the fourth ventricle and lumbar cistern are displayed in Fig. 4. Mean trough CSF etoposide levels from the fourth ventricle and lumbar cistern were similar, as displayed in Fig. 5.

AUC analysis of CSF etoposide levels is listed in Table 2 for each piglet individually, and Table 3 displays overall statistical results when comparing fourth ventricle and lumbar CSF at peaks, troughs, and all time points. When peak etoposide levels were assessed by AUC analysis, statistically significant differences between fourth ventricle and lumbar AUC were noted (ΔAUC = 3384196 ng h/ml with 95%CI: 1758625, 5009767, $P = 0.0044$). Statistically significant differences were also noted when all time collection time points were assessed (ΔAUC = 1422977 ng h/ml with 95%CI: 732188, 2113766, $P = 0.0046$). At troughs, there was no statistically significant difference between fourth ventricle and lumbar CSF etoposide levels (ΔAUC = −29546 ng h/ml (95%CI: −147526, 88434.2, $P = 0.5251$).

Fig. 5 Mean ± SE trough CSF etoposide levels in the fourth ventricle and lumbar cistern after five consecutive daily infusions into the fourth ventricle. CSF Etoposide Concentration Profile at Troughs (Mean + Standard Error)

Table 2 AUC values for each piglet (ng h/ml), calculated by linear trapezoidal method

Piglet ID	Peaks		Troughs		All	
	Fourth ventricle	Lumbar cistern	Fourth ventricle	Lumbar cistern	Fourth ventricle	Lumbar cistern
1	3760019.52	50146.08	53644.20	63661.44	1846702.42	73828.58
2	1379789.52	25778.28	37730.16	30983.52	736163.07	38551.63
3	3035536.80	0.00	33952.44	7974.24	1589740.56	10015.57
4	4874408.52	17052.36	15291.87	212611.44	1161420.54	149174.08
5	3977599.92	13398.36	37254.24	10372.20	2064688.67	12260.11

Table 3 Statistical test results assessing differences between fourth ventricle and lumbar etoposide levels by AUC analysis

Time	Difference (SD)[a]	95% CI[b]	P-value*
Peaks	3384196 (1309187)	(1758625, 5009767)	0.0044
Troughs	−29546 (95018)	(−147526, 88434)	0.5251
All	1422977 (556341)	(732188, 2113766)	0.0046

[a] Difference is defined as mean difference in *AUC* mean AUCventricular − mean AUClumbar; all values are in ng h/ml; *SD* Standard Deviation of the difference

[b] 95% Confidence Interval for the mean difference in AUC

* Based on two-sided paired *t*-test at 5% significance level

Discussion

Although survival for patients with medulloblastoma has improved dramatically over time, current adjuvant therapy regimens are associated with significant morbidity, and survival rates for patients with recurrence are still poor. Our laboratory has developed a piglet model to investigate local chemotherapy infusions directly into the fourth ventricle. Such infusions would potentially enable high local drug concentrations while minimizing systemic exposure and associated side effects. This treatment approach would also potentially provide regional therapy throughout CSF spaces if administered prior to the presence of bulky leptomeningeal disease which obstructs CSF flow.

Results of preliminary experiments using the piglet model developed in our laboratory were recently published [1]. These experiments demonstrated that etoposide can be infused into the fourth ventricle without causing recognizable neurological deficits or imaging changes on MRI scans. An inflammatory response consisting predominantly of T-lymphocytes was demonstrated, but this did not correspond to any recognizable clinical findings. These results were replicated in the current experiments. In all 5 piglets, the catheter was noted on MRI scans to be within the fourth ventricle, and there were no neurological deficits or signal changes in the brainstem or cerebellum noted on MRI scan. The main pathological finding was a significant

inflammatory response of the meninges, choroid plexus, and ependyma, but this did not correspond to recognized clinical findings in any piglets. Of note, CSF analysis showed positive bacterial cultures in 3 of 5 piglets and leukocytosis was noted in all 3 piglets in which adequate CSF was available for cell count. None of the piglets demonstrated any clinical signs of meningitis, and we attribute the positive cultures to the multiple time points at which CSF was accessed from externalized ports despite attempts to maintain sterile technique. Based upon previous published studies from our laboratory in which control piglets not receiving etoposide had a minimal inflammatory response, the authors hypothesize that the inflammation observed on histological sections and CSF leukocytosis were likely a direct response to the etoposide or its constituents rather than to infection [1]. This hypothesis is supported by the fact that meningitis and ventriculitis noted on autopsy was most severe in the 2 patients with negative CSF cultures.

The primary objective of the current study was to assess pharmacokinetics associated with etoposide infusions into the fourth ventricle. Etoposide was utilized for these studies based upon its significant cytotoxic activity against medulloblastoma in vitro and the fact that it has been well-tolerated in previous clinical trials when administered into the lateral ventricle [8, 10, 20, 21]. In these trials, peak CSF etoposide levels over one hundred times higher than with systemic administration were achieved [8, 10].

In the current experiments, CSF samples were obtained from the fourth ventricle and lumbar cistern to study both local and regional pharmacokinetics. Ideally, CSF would have been obtained from the lateral ventricle as well to evaluate distribution throughout the ventricles. Unfortunately, the small size of the lateral ventricle in piglets renders collection of adequate CSF for analysis not possible with this model.

We found that a single infusion of etoposide into the fourth ventricle at a dose of 0.5 mg achieved cytotoxic etoposide levels, defined as (>0.1 µg/ml) by previous studies [22], at all time points measured for the first 24 h in both the fourth ventricle and lumbar cistern. Following

daily etoposide infusion over 5 days, mean peak and trough etoposide levels in both the fourth ventricle and lumbar cistern continued to exceed cytotoxic levels. CSF etoposide concentrations in the fourth ventricle during the first 24 h and for peaks and troughs after daily infusion for 5 days were similar to those reported in previous human studies involving infusions into the lateral ventricles [10, 21].

As expected, etoposide levels in CSF from the fourth ventricle peaked immediately after infusion into the fourth ventricle and then gradually declined, while lumbar CSF levels started lower and then gradually increased. By four hours after infusion, etoposide levels in the fourth ventricle and lumbar cistern were similar. These findings suggest that this treatment approach, if utilized prior to the presence of bulky leptomeningeal disease which obstructs CSF flow, may provide both local and regional therapy. Moreover, the fact that serum etoposide levels were not measurable suggests that systemic side effects will be unlikely with intraventricular infusions.

While cytotoxic drug levels were achieved and maintained in both the fourth ventricle and lumbar cistern, the mean peak etoposide level achieved in the fourth ventricle exceeded the mean peak etoposide level in the lumbar cistern by greater than 10-fold. AUC analysis confirmed significantly higher drug concentrations at peak measurements in the fourth ventricle. While the benefit of providing a substantially higher drug concentration than needed for killing tumor cells is unknown, the authors propose that aggressive treatment directly at the site of residual tumor immediately after surgery may offer a theoretical advantage as long as there is no associated toxicity. This hypothesis needs to be tested by future experiments.

In conclusion, etoposide can be infused directly into the fourth ventricle in piglets without causing recognized clinical toxicity or MRI evidence of damage. An inflammatory response is observed without associated clinical correlate, and future studies will determine if this response can be minimized with simultaneous administration of systemic corticosteroids or by administering chemotherapeutic agents other than etoposide. Cytotoxic etoposide levels can be achieved in the fourth ventricle and lumbar cistern with significantly higher peak levels in the fourth ventricle. Etoposide is not detected in serum after these infusions. These preliminary findings suggest that direct infusion of chemotherapeutic agents into the fourth ventricle will enable cytotoxic concentrations at the site of tumor origin without causing significant systemic toxicity.

References

1. Sandberg DI et al (2008) Chemotherapy administration directly into the fourth ventricle in a new piglet model. J Neurosurg Pediatrics 1(5):373–380
2. Reddy AT, Packer RJ (1999) Medulloblastoma. Curr Opin Neurol 12(6):681–685
3. Rutka JT (1997) Medulloblastoma. Clin Neurosurg 44:571–585
4. Choux M et al (1982) Medulloblastoma. Neurochirurgie 28(Suppl 1): 1–229
5. Silverman CL, Simpson JR (1982) Cerebellar medulloblastoma: the importance of posterior fossa dose to survival and patterns of failure. Int J Radiat Oncol Biol Phys 8(11):1869–1876
6. Suit HD, Westgate SJ (1986) Impact of improved local control on survival. Int J Radiat Oncol Biol Phys 12(4):453–458
7. Rutkowski S et al (2005) Treatment of early childhood medulloblastoma by postoperative chemotherapy alone. N Engl J Med 352(10):978–986
8. Slavc I et al (2003) Feasibility of long-term intraventricular therapy with mafosfamide ($n = 26$) and etoposide ($n = 11$): experience in 26 children with disseminated malignant brain tumors. J Neurooncol 64(3):239–247
9. Slavc I et al (1998) Intrathecal mafosfamide therapy for pediatric brain tumors with meningeal dissemination. J Neurooncol 38(2–3):213–218
10. Fleischhack G et al (2001) Feasibility of intraventricular administration of etoposide in patients with metastatic brain tumours. Br J Cancer 84(11):1453–1459
11. Sandberg DI et al (2000) Ommaya reservoirs for the treatment of leptomeningeal metastases. Neurosurgery 47(1):49–54; discussion 54-5
12. Jacobs A, Clifford P, Kay HE (1981) The Ommaya reservoir in chemotherapy for malignant disease in the CNS. Clin Oncol 7(2):123–129
13. Obbens EA et al (1985) Ommaya reservoirs in 387 cancer patients: a 15-year experience. Neurology 35(9):1274–1278
14. Shapiro WR et al (1977) Treatment of meningeal neoplasms. Cancer Treat Rep 61(4):733–743
15. Rubinstein LJ et al (1975) Disseminated necrotizing leukoencephalopathy: a complication of treated central nervous system leukemia and lymphoma. Cancer 35(2):291–305
16. Macdonald DR (1991) Neurologic complications of chemotherapy. Neurol Clin 9(4):955–967
17. Chamberlain MC, Kormanik PA, Barba D (1997) Complications associated with intraventricular chemotherapy in patients with leptomeningeal metastases. J Neurosurg 87(5):694–699
18. Bleyer WA et al (1978) The Ommaya reservoir: newly recognized complications and recommendations for insertion and use. Cancer 41(6):2431–2437
19. Reif S et al (2001) Bioequivalence investigation of high-dose etoposide and etoposide phosphate in lymphoma patients. Cancer Chemother Pharmacol 48(2):134–140
20. Gajjar A et al (1999) Chemotherapy of medulloblastoma. Childs Nerv Syst 15(10):554–562
21. Sirisangtragul C et al (2003) Cerebrospinal fluid pharmacokinetics after different dosage regimens of intraventricular etoposide. Int J Clin Pharmacol Ther 41(12):606–607
22. Henwood JM, Brogden RN (1990) Etoposide. A review of its pharmacodynamic and pharmacokinetic properties, and therapeutic potential in combination chemotherapy of cancer. Drugs 39(3):438–490