Rhesus Macaque as an Animal Model for Posterior Fossa Syndrome following Tumor Resection

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Abstract

Background/Aims: Posterior fossa tumors are the most common brain tumors in children. Surgeons usually remove these tumors via a midline incision through the posterior vermis of the cerebellum. Though often effective, this surgery causes hypotonia, ataxia, oculomotor deficits, transient mutism, difficulty in swallowing and nausea. To date, there is no animal model that mimics these complications. We found that the rhesus macaque is a good model for the consequences of this surgery. Methods: We made a midline incision through the cerebellar vermis of one monkey to mimic the posterior fossa surgery. Then, we closely monitored the monkey for deficits following the surgery. Results: In the first few days, the monkey exhibited nausea, hypotonia, ataxia, difficulty in swallowing and an absence of vocalization. At 28 days, we recorded eye movements and found severe deficits in the accuracy of rapid eye movements and smooth pursuit of a target. Additionally, the animal had trouble fixating and a rightward-beating nystagmus. Oculomotor signs persisted until we sacrificed the animal 99 days after surgery, but the other effects resolved by 37 days. Conclusion: Our surgery in a monkey caused the same postsurgical signs observed in humans. We expect to use this model to improve the posterior fossa surgery methods.

Introduction

Pediatric brain tumors account for >20% of all childhood cancers and are second only to leukemia in the number of deaths they cause. Nearly half of the brain tumors in children 0–14 years old are in the posterior fossa, involving the cerebellum and/or brainstem in the area around the IVth ventricle [1–4].

These tumors usually require aggressive surgical resection to maximize the likelihood of survival. To remove posterior fossa tumors, surgeons commonly make an incision through the midline of the posterior lobe of the cerebellum. Though effective for removing tumors and for improving survival [3, 5], this method often leaves patients with short- and long-term neurological deficits, some of which are severe [1, 6, 7]. Up to 25% of the patients...
with medulloblastoma (the most common pediatric posterior fossa tumor) suffer from a posterior fossa syndrome characterized by trunk and limb ataxia, hypotonia, and oculomotor abnormalities as well as mutism, i.e. diminished or absent vocalization [8, 9]. Though previously considered transient, this syndrome was recently shown to have persistent features and poor long-term outcomes [1, 3].

An accurate animal model would help develop techniques to improve these outcomes. Our results show that the rhesus monkey is an animal model that mimics the major deficits observed in human patients after a midline incision through the posterior cerebellum, particularly following extensive damage to midline cerebellar structures. Here, we emphasize the assessment of eye movements because they are easily quantifiable and similar data could be obtained from human patients.

Materials and Methods

One male juvenile rhesus monkey was used for this study. We implanted a scleral search coil in the animal’s left eye to record eye movements and trained the monkey to track a small moving spot of light with rapid eye movements (saccades) and smooth pursuit movements. After recording normal eye movements we anesthetized the monkey and, in a sterile procedure, made an incision through the posterior cerebellar vermis that mimicked the surgery to remove tumors near the IVth ventricle. After surgery we monitored the monkey's behavior for 99 days. We began recording eye movements 28 days after surgery, the earliest that the monkey would make trained eye movements. Veterinary records documented the animal’s recovery for the previous 27 days, as well as after we had started recording eye movements. To record eye movements, we digitized horizontal and vertical eye and target positions at 1 kHz using a Power 1401 digitizer (Cambridge Electronic Design, Cambridge, UK) and stored them on a hard drive. Custom MATLAB (MathWorks, Inc., Natick, Mass., USA) programs measured the properties of saccade and smooth pursuit eye movements. After we had completed the data collection, we sacrificed the monkey with an overdose of barbiturate and perfused it through the left ventricle with formalin and sucrose formalin. We cut 50-μm frozen parasagittal sections and stained them with cresyl violet.

All experiments and procedures were approved by the Animal Care and Use Committee at the University of Washington.

Results

Histology showed evidence of a midline cerebellar hemorrhage, presumably at the time of surgery. There was bilateral damage to the posterior vermis. It extended to lobules VI, VII, VIII, IX and X (see fig. 1). On the right, lobules VI and VII were damaged, and on the left damage extended from lobule VII to X. The dentate nucleus was intact, bilaterally, though outflow fibers from the dentate nucleus to the superior cerebellar peduncle may have been involved (tracer confirmation is not available) due to the extent of this incision.

Like many humans who have undergone posterior fossa tumor resection, this monkey exhibited postoperative ataxia, hypotonia and oculomotor abnormalities, as well as transient mutism, difficulty in swallowing and nausea.

Nonoculomotor Deficits

Ataxia and Hypotonia

Immediately after recovery from surgical anesthesia the animal exhibited severe hypotonia. It lay on the floor of its cage unable to sit up. It also exhibited severe ataxia of both its trunk and limbs. The arms oscillated widely around targets and, when we assisted the monkey in sitting up, its trunk movements oscillated strongly.

Three days after the surgery the monkey was able to grasp the juice syringe despite limb oscillations. At 5 days it was able to sit up and drink water. Eleven days after surgery it could climb slowly onto its perch. At day 17 its movements were more effective but still very ataxic and exhibited large oscillations. By day 37 the ataxic oscillations had subsided substantially and the animal’s movements had largely recovered but were clearly slower than normal.

Transient Mutism

Transient mutism occurs in about 25% of humans following posterior fossa surgery for midline tumors [3, 10]. Though monkeys do not speak, they vocalize, especially when presented with food. Before surgery our monkey consistently vocalized several times per minute. After surgery our monkey consistently vocalized several times per minute. In contrast, after surgery it was completely mute for 35 days. On postsurgery day 36 it vocalized for the first time as we presented food. Its initial vocalizations were unusually soft, abnormally short and high pitched. The animal vocalized with increasing frequency and within a few days the frequency and sound of vocalizations were normal.

Difficulty in Swallowing

The monkey had difficulty in swallowing after surgery. For 8 days after the operation the animal choked every time it tried to drink water or juice. On day 9 it was...
able to swallow small volumes but still often choked until day 12. After that it was able to swallow liquids and solids apparently without trouble.

**Nausea**

Though common in human patients immediately after surgery, nausea appeared in our animal at 4 days after surgery and persisted for 2 days.

**Oculomotor Deficits**

**Saccades**

This surgery severely impaired saccades. Figure 2a shows vertical target and eye positions during saccades before surgery and at 1 and 3.5 months after surgery. Before surgery the monkey’s saccades were accurate or occasionally slightly undershooting (e.g. fig. 2a, left panel, 6.5 s). After surgery both upward and downward saccades were larger than normal so that they often oscillated around the target (fig. 2a, middle panel). Presumably, these oscillations occurred because saccades could not stop directly on the target and each saccade toward the target was too big and elicited another overshooting saccade in the opposite direction. Horizontal saccades were also disturbed but not as severely. Saccade size accuracy did not recover; they were too large at both 1 and 3.5 months after surgery.

In addition to being hypermetric, postsurgery saccades exhibited 2 other abnormalities. They were unusually variable and their deceleration was impaired (fig. 3 and 4, respectively). Figure 3 shows that 1 month after surgery the standard deviation of vertical saccade size is ~11 times its presurgery size when tracking identical 5° target movements. Though this variability decreased with increasing time after surgery, it was still ~6 times larger than normal at 3.5 months.

Deceleration of postsurgery horizontal and vertical saccades lasted longer than normal (fig. 4a), while saccade acceleration was not strongly changed after surgery. Figure 4 plots the deceleration duration at 1 and 3.5 months after surgery. The average deceleration duration was roughly 140% of normal for all sizes and directions of saccades. This may be why saccades did not stop accurately on the target.
Smooth Pursuit
The left panel of figure 2b shows records of eye and target position during tracking of a target oscillating left and right sinusoidally ±10° at 0.2 Hz before surgery. The eye closely tracked the moving target. After surgery horizontal tracking was much less accurate. The middle and right panels of figure 2a show records during smooth pursuit 1 and 3.5 months after surgery, respectively. The monkey made many small saccades to compensate for poor smooth pursuit tracking. On average, the gain (eye velocity divided by target velocity) of horizontal smooth pursuit was only ~50% of normal after surgery for 0.2 and 0.5 Hz (see fig. 5). We did not test vertical tracking after the surgery.

Fixation Instability
After surgery the monkey was unable to look steadily at a stationary target. Figure 2c shows vertical eye and target positions during fixation before (left panel) and 1 (middle panel) and 3.5 (right panel) months after surgery.
**Fig. 3.** Comparison of saccade size and standard deviation before, as well as 1 and 3.5 months after surgery of 13–14 sequential saccades to leftward 5° target movements. The solid line (blue in the online version) marks the mean saccade size and the gray area above and below the mean marks ±1 SD. *p < 0.05.

**Fig. 4.** Deceleration duration. a Eye movement trajectories and velocity profiles to 10° target steps before (thick lines) and 3.5 months after the cut (thin lines). After the cut saccades are more variable and the deceleration phase is prolonged and kinked. b Deceleration duration for 10° rightward saccades. Data normalized to percent of control and collected prior to, as well as 1 and 3.5 months after surgery. Error bars are ± SD. *p < 0.05; **p < 0.001 (t test).
Before surgery the monkey’s eye stayed on or very near the target. After surgery apparent saccades, both large and small, deflected the eye abruptly away from the target. The deflections were both larger and more frequent after surgery. There was no predominant direction for these deflections.

**Nystagmus**

Immediately after surgery the monkey exhibited a rightward-beating nystagmus. We did not measure the velocity of this nystagmus, but it was clearly seen on visual inspection. The nystagmus decreased significantly within 3 days and we were no longer able to detect it visually, but it was still evident in eye movement recordings 1 month after surgery. These recordings showed an intermittent ~3°/s rightward-beating nystagmus with an occasional small downward component. The nystagmus was very rare or absent by 3.5 months after surgery.

**Persistence of Signs**

The monkey’s ataxia, swallowing difficulty and mutism all resolved by 37 days after the posterior fossa surgery. In contrast, the hypermetria of vertical saccades never improved significantly. For example, 1 month after the lesion the monkey’s saccades tracking 10° vertical target movements were 30% larger than normal. At 3.5 months after surgery they were even more hypermetric, 59% larger than normal. Oscillations around the target persisted throughout our tests (right panel, fig. 2a).

**Discussion**

This work shows that a surgery in a rhesus monkey that mimics the procedure for removing a posterior fossa tumor in humans causes the same postsurgical signs. This animal sustained more extensive damage to the vermis than we had initially planned with the midline incision. Nevertheless, this lesion is very similar to that in humans undergoing removal of a posterior fossa tumor because the damage remained within a few millimeters of the midline, within the cerebellar vermis, with no direct damage to the cerebellar nuclei or the brainstem.

Like human patients after surgery, this monkey showed persistent disturbances of saccades, smooth pursuit and fixation. The oculomotor deficits of humans after posterior fossa tumor resection are not described in detail yet but, like our monkey, these patients have persistent difficulty in aiming their foveae consistently at a target whether or not the target is moving [11]. It is not surprising that damage to the posterior vermis in both humans and monkeys impairs eye movements, since the posterior vermis is strongly implicated in both saccades [12, 13] and pursuit [14].

Also like human patients, our monkey showed severe but transient hypotonia, ataxia, difficulty in swallowing and mutism. The duration of this animal’s mutism is similar to that evident in posterior fossa syndrome [1, 3, 6–10]. There are currently no other reports of vocalization function in the cerebellar vermis of primates, even though such deficits are increasingly recognized as significant morbidities following midline cerebellar surgery in humans. Current data do not indicate how vermis damage causes hypotonia, ataxia and mutism.

Increasingly, damage to the cerebellar outflow from the dentate to thalamus, through the superior cerebellar peduncle, is suspected in the etiology of posterior fossa syndrome [15–17] and this may prove to be the case in monkeys as well. It is possible that damage to equivalent structures, rather than to the vermis proper, resulted in the nonoculomotor deficits in this animal. Most importantly, this case offers the promise that future, targeted lesions in monkeys might resolve the question definitively, by correlating the surgical lesion in ways obviously impossible in clinical situations.
Conclusions

We mimicked the surgery most commonly used to remove posterior fossa tumors from humans in a rhesus monkey. Our surgery reproduced in a monkey the postoperative signs seen in human patients. Though we did this initial test in only 1 animal, because of the severity of postoperative effects, the strong similarity between the effects in monkeys and humans indicates that the rhesus macaque is a good animal model for this surgery. This animal model improves our ability to refine the surgical approach. In particular, we seek to develop an approach that avoids damaging eye movement-related structures like the oculomotor vermis. Damage to this structure might be the cause of an extremely high incidence of postoperative eye movement abnormalities and the consequent difficulty of seeing clearly [11].

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