A fluid percussion model of experimental brain injury in the rat

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Fluid percussion models produce brain injury by rapidly injecting fluid volumes into the cranial cavity. The authors have systematically examined the effects of varying magnitudes of fluid percussion injury in the rat on neurological, systemic physiological, and histopathological changes. Acute neurological experiments showed that fluid percussion injury in 53 rats produced either irreversible apnea and death or transient apnea (lasting 54 seconds or less) and reversible suppression of postural and nonpostural function (lasting 60 minutes or less). As the magnitude if injury increased, the mortality rate and the duration of suppression of somatomotor reflexes increased. Unlike other rat models in which concussive brain injury is produced by impact, convulsions were observed in only 13% of survivors. Transient apnea was probably not associated with a significant hypoxic insult to animals that survived. Ten rats that sustained a moderate magnitude of injury (2.9 atm) exhibited chronic locomotor deficits that persisted for 4 to 8 days. Systemic physiological experiments in 20 rats demonstrated that all levels of injury studied produced acute systemic hypertension, bradycardia, and increased plasma glucose levels. Hypertension with subsequent hypotension resulted from higher magnitudes of injury. The durations of hypertension and suppression of amplitude on electroencephalography were related to the magnitudes of injury. While low levels of injury produced no significant histopathological alterations, higher magnitudes produced subarachnoid and intraparenchymal hemorrhage and, with increasing survival, necrotic change and cavitation. These data demonstrate that fluid percussion injury in the rat reproduces many of the features of head injury observed in other models and species. Thus, this animal model could represent a useful experimental approach to studies of pathological changes similar to those seen in human head injury.

Key Words • head injury • fluid percussion model • concussion • rat

The consequences of mechanical brain injury range from a concussive syndrome characterized by brief loss of consciousness in the absence of significant histopathology to prolonged coma or death usually associated with extensive brain pathology. Several investigators have developed animal models of mechanical brain injury in an attempt to reproduce various aspects of the biomechanical responses, neurological syndromes, and pathology observed in human closed-head injury. These models use a variety of experimental techniques that have been developed to study these syndromes, including devices that accelerate or rotate the skull, impact injury to the intact and freely movable cranium, and rapid injection of fluid into a closed cranium (fluid percussion).

Fluid percussion devices reproduce a number of features of brain injury reported in humans. The technique produces pressure transients (approximately 20 msec) similar to those recorded in human cadaver skulls during sudden impact as well as neurological signs of behavioral suppression resembling signs of unconsciousness in humans. Fluid percussion injury also results in reduction or abolition of cerebrovascular responsiveness to changes in pCO2, loss in pressure autoregulation, immediate and late-developing increases in intracranial pressure, and edema, probably of vasogenic origin.

Studies of head injury have employed a variety of species, including rats, rabbits, cats, subhuman primates. Experimental brain injury in the rat has been produced by several whole-head impact methods, including the use of spring-loaded guns, gas-accelerated pistons, and lead-tipped darts. Recent reports have described the effects of a combined fluid...
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percussion-hypoxia insult to the rat on some neurological, electroencephalographic (EEG), histopathological, and metabolic variables. However, these studies did not attempt to characterize the rat’s response to fluid percussion injury as extensively as has the research presented here.

In contrast to mongrel species such as cats, the use of laboratory rats provides a number of advantages in studying experimental brain injury. These advantages include: 1) precise knowledge of age and genetic background of the animals; 2) existence of normative data for a wide range of physiological and behavioral variables; 3) compatibility with a number of neurochemical and neuropharmacological techniques not readily employed in larger species; 4) low purchase price and animal-care costs; 5) high resistance to infection; and 6) economical use of expensive radioisotopes and drugs due to low animal body weight.

The purpose of this study was to characterize in detail the neurological, physiological, and histopathological features of varying levels of mechanical brain injury produced in the rat by the fluid percussion technique. Our aim was to determine if such a model would produce graded levels of injury, and if fluid percussive injury to rats produces pathophysiological responses similar to those reported in other species. These data are necessary to determine if fluid percussion injury in the rat is a useful experimental model of brain injury. These data also provide important normative values for other researchers wishing to employ the rat model in experimental studies of brain injury.

Materials and Methods

Injury Device

The fluid percussion device used to produce experimental brain injury was identical to that which has been used in cats and described in greater detail elsewhere. Briefly, it consisted of a Plexiglas cylindrical reservoir, 60 cm long and 4.5 cm in diameter, bounded at one end by a Plexiglas cork-covered piston mounted on O-rings (Fig. 1). The opposite end of the reservoir was fitted with a 2-cm long metal housing on which a transducer was mounted. Fastened to the end of this was a 5-mm tube (2 mm inner diameter) that ended with a male Leur-Loc fitting. This was connected to a female Leur-Loc fitting that had been chronically implanted over the exposed dura overlying the parietal cortex (see Surgical Preparation). The entire system was filled with 37°C isotonic saline. The injury was induced by a metal pendulum which struck the piston of the injury device from a predetermined height. The resulting pressure pulse was measured extracranially by the transducer at the time of injury, recorded on a storage oscilloscope, and photographed with a Polaroid camera. The oscilloscope was triggered photoelectrically by the descent of the pendulum. The device injects varying volumes of saline into the closed cranial cavity, thereby producing brief displacement and deformation of neural tissue. Increased magnitudes of tissue deformation are associated with increased magnitudes of brain injury. The magnitude of the injury was regulated by varying the height of the pendulum. This resulted in corresponding variations of extracranial pressure pulses expressed in atmospheres (atm). Extracranial pressure pulses produced by fluid percussion injury have been found to be closely associated with intracranial pressure changes.

Surgical Preparation

All animals were surgically prepared under sodium pentobarbital anesthesia (54 mg/kg intraperitoneally) 24 hours prior to injury in order to reduce the possible confounding effects of the surgical trauma associated with the procedure and to minimize the effects of surgical anesthesia on postinjury neurological evaluations. Animals were placed in a stereotaxic frame and the scalp was sagittally incised. A hole 4.8 mm in diameter was trephined into the skull over the sagittal suture, midway between the bregma and lambda. Two stainless steel screws were placed 1 mm rostral to the bregma and 1 mm caudal to the lambda. A rigid plastic injury tube (modified Leur-Loc syringe hub, 2.6-mm inner diameter) was placed over the exposed dura and bonded with cyanoacrylate adhesive. Dental acrylic was then poured around the injury tube and around the stainless steel screws. After the acrylic had hardened, the injury tube was plugged with Gel foam sponge, the scalp was sutured closed, bacitracin was applied over the wound, and the animal was returned to its home cage.
Acute Neurological Evaluations

For the acute neurological evaluations, 53 male Sprague Dawley rats, each weighing 350 to 450 gm, were anesthetized with methoxyflurane and concussed at 2.1 atm (10 rats), 2.9 atm (nine rats), 3.4 atm (15 rats), 3.6 atm (13 rats), and 3.8 atm (six rats). Ten additional animals that were identically prepared but not injured served as a sham-treated control group. Within 15 seconds following injury, the animals were carefully removed from the injury device by disconnecting the implanted Leur-Loc and were placed on a table for neurological assessment.

A battery of tests was developed to characterize certain acute (60 minutes postinjury) neurological consequences of mechanical brain injury. This battery, which includes tests analogous to motor components of the Glasgow Coma Scale, allows quantification of various features of the suppression of animals' responsiveness to external stimuli and relationships between durations of behavioral suppression and magnitudes of injury. This battery is derived in part from neurological scales previously developed in our laboratories to evaluate the behavioral effects of brain injury on cats and rats.

The occurrence and duration of convulsive or apneic episodes were recorded throughout the acute assessment period, in addition to notations of whether such events were transient or irreversible (that is, associated with death). Assessments of simple nonpostural somatomotor functions were conducted by recording the duration of suppression of two responses to stimulation. The pinna reflex was assessed by touching the auditory meatus of the animal's ear to elicit a vigorous head shake. The corneal reflex was evaluated by lightly touching the cornea with a blunt instrument to elicit an eyeblink.

Assessments of simple postural somatomotor functions included measurements of the duration of suppression of flexion reflexes to stimulation. The assessment of the paw and tail flexion reflexes, a modification of an analgesia test developed by Collier, consisted of the gradual application of pressure on the hind paw or tail until paw or tail withdrawal was noted. Graded pressure was applied by means of specially designed needle-nosed pliers having a 4-sq mm jaw for grasping tissue. Normally, pinches with a calibrated force of 0.2 kg/sq mm consistently elicited the paw and tail flexion reflex. Paw and tail flexion reflexes were assessed at intervals of 1 minute until flexion occurred. In the event that no flexion reflex resulted, pressure greater than 2.0 kg/sq mm was not applied in order to prevent tissue damage. Another simple postural somatomotor function, the head-support reflex, was assessed by noting the duration of suppression of the animal's ability to support the weight of its head. Inability of the animal to support its head was reliably associated with a generalized loss of muscle tone.

A more complex nonpostural somatomotor function was assessed by recording the duration of suppression of the startle response. The startle response was determined by observing the presence of any motor response to a brief, loud noise resulting from snapping a clipboard's paper holder. In normal animals, this stimulus reliably elicits a profound twitch and stiffening of the whole body. More complex postural somatomotor functions were assessed by recording the duration of suppression of the following responses. The righting response was defined as the animal's ability to right itself three times consecutively after being placed on its back. After recovery of the righting response, spontaneous locomotion was evaluated by regularly placing the rat in a marked 25 x 25-cm area until the rat walked spontaneously out of the area. An escape response was assessed by briefly pinching the tail of the animal with the calibrated pinchers at a force of 1.0 kg/sq mm to elicit locomotive activity away from the noxious stimulus.

Chronic Neurological Evaluations

Two assessment tasks characterized more enduring motor deficits over a 10-day period following a 2.9-atm injury. This injury level produced intermediate levels of mortality and acute reflex suppression (see Results). For this evaluation, 10 male Sprague Dawley rats, each weighing 350 to 450 gm, were surgically prepared and injured using the procedure outlined in the acute evaluations. Body weights were recorded daily. The assessment of locomotor functioning was conducted by employing the following behavioral tasks. The beam-balancing task assessed motor and vestibular functioning by observing the animal's ability to balance on a beam. The beam-walking task allowed the assessment of more refined locomotor activity in the context of a learned avoidance task.

Beam-Balancing Task. The beam-balancing task consisted of placing the rat on a narrow wooden beam (1.5 cm wide) and noting the time (up to 60 seconds) during which the animal was able to maintain its balance. The balancing-beam durations were measured prior to injury and daily for 10 days after injury.

Beam-Walking Task. The beam-walking task was similar to that employed by Feeney and coworkers, in which rats traversed the top of a narrow wooden beam to avoid loud white noise and bright light. Rats were trained prior to injury using a negative-reinforcement paradigm in which termination of the adverse stimuli (noise and light) served as reinforcement (reward). During training and testing, the animals were placed on a start platform (15 x 17 cm) at one end of an elevated (1-m) wooden beam (120 x 2.5 cm) close to the source of the noise and light. The difficulty level of the task was increased by placing four equally spaced pegs (3 cm high) along the top of the beam, thus providing a more sensitive measure of motor performance. The noise was terminated after the animal had traversed the beam and entered a darkened goal box (30 x 15 x 18 cm). The length of time taken to traverse the beam was

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TABLE 1
Acute pathological responses following fluid percussion injury in the rat*

<table>
<thead>
<tr>
<th>Injury (atm)</th>
<th>No. of Rats</th>
<th>% Mortality</th>
<th>Pulmonary Edema</th>
<th>Convulsive Episodes</th>
<th>Transient Apnea</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Incidence (%)</td>
<td>Incidence (%)</td>
<td>Incidence (%)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Nonsurvivors</td>
<td>Survivors</td>
<td>Nonsurvivors</td>
</tr>
<tr>
<td>2.1</td>
<td>10</td>
<td>10</td>
<td>100</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>2.9</td>
<td>9</td>
<td>22</td>
<td>50</td>
<td>0</td>
<td>50</td>
</tr>
<tr>
<td>3.4</td>
<td>15</td>
<td>40</td>
<td>83</td>
<td>0</td>
<td>33</td>
</tr>
<tr>
<td>3.6</td>
<td>13</td>
<td>38</td>
<td>60</td>
<td>0</td>
<td>80</td>
</tr>
<tr>
<td>3.8</td>
<td>6</td>
<td>100</td>
<td>17</td>
<td>-</td>
<td>67</td>
</tr>
</tbody>
</table>

* - = not applicable.

measured. After baseline measurements the animals were injured and assessed daily for 10 days.

Systemic Physiological and EEG Evaluations

Twenty male Sprague Dawley rats, each weighing 350 to 500 gm, were initially anesthetized with methohexitol sodium (Brevital, 50 mg/kg intraperitoneally) and tracheostomized. During surgical preparation and throughout the experimental procedure, all wounds were infused with a topical anesthetic (2.0% lidocaine hydrochloride). After tracheostomy, the rats were paralyzed with intraperitoneal curare (0.3 ml) and artificially ventilated with a mixture of 70% nitrous oxide and 30% oxygen.* The femoral artery was cannulated to monitor heart rate and arterial blood pressure, as well as to sample arterial blood gases and plasma glucose levels. Arterial pressure changes were monitored by a strain-gauge transducer and recorded on a Beckman polygraph.† The cannula line was kept patent with buffered Ringer's solution. Prior to injury, the ventilation rate and volume were controlled to stabilize arterial blood gases within the normal range.‡ Blood gas assessments were made with a pH and blood gas analyzer.§ Aspiration and suspiration were routinely performed every 15 minutes. Body temperature was maintained at 37°C throughout the experimental procedure. Heart rate, arterial blood pressure, blood gases, plasma glucose level, hematocrit, and gross cortical EEG activity were recorded prior to and for 60 minutes after injury. Five animals were injured at 2.1 atm, five at 2.9 atm, and five at 3.8 atm. Five additional animals underwent identical procedures but were not injured, thus serving as a sham-treated control group.

In order to provide data on the magnitude of hypoxia associated with transient apnea following injury to animals used in the neurological studies, pO2 measurements were made in six additional animals. These animals were not injured but were surgically prepared and artificially ventilated similarly to the animals used in the physiological studies. After a blood gas sample was obtained, the animal's ventilation was stopped by turning off the respirator. At the end of a 15-, 30-, 45-, or 60-second interval (as determined by a Latin square design), another sample was taken and ventilation was then resumed. After pO2 levels returned to baseline, this procedure was repeated for each animal until pO2 measurements were made for all four durations.

Histopathological Evaluations

Fifteen experimental animals were prepared for gross inspection as well as for detailed light microscopy histological analyses following fluid percussion brain injuries of varying severity (2.1 to 3.8 atm). Corresponding to the acute and chronic phases of the neurological study, some animals were examined immediately following the traumatic event, while others were examined 4 to 7 days after injury. After the designated survival intervals, the animals were transcardially perfused with 10% buffered neutral formalin. Their brains were then removed, dehydrated in ascending grades of ethanols, cleared, and embedded in paraffin. Sections were cut 8 μ thick on a microtome and mounted, with alternative sections being stained with either hematoxylin and cosin or with the Bodian/Palmgren silver methods.

Results

Acute Neurological Evaluations

The percentage of animals that died generally increased with the severity of the injury. The largest increase in mortality occurred between the 3.6-atm and 3.8-atm levels of injury (Table 1). In many of the fatal injuries, the occurrence of pulmonary edema was suggested by the presence of pink fulminating exudate from the mouth and nostrils. The occurrence of pulmonary edema in nonsurvivors was not related to the magnitude of injury. It should be noted that animals injured at 3.8 atm generally died prior to an interval associated with the appearance of exudate, as was characteristic of the less severely injured groups. None of

* Ventilator, Model 680, manufactured by Harvard Apparatus Co., Inc., South Natick, Massachusetts.
† Beckman polygraph manufactured by Statham Instruments, Oxnard, California.
‡ Blood gas analyzer, Model 213, manufactured by Instrumentation Laboratory, Lexington, Massachusetts.

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FIG. 2. Effects of varying magnitudes of fluid percussion injury on acute neurological functioning. The data present mean durations of suppression ± standard error of the mean of: A) simple nonpostural reflexes; B) simple postural reflexes; C) complex nonpostural reflexes; D) complex postural responses; and E) complex locomotor responses.

the survivors showed pulmonary edema. Animals injured at 2.1 atm did not exhibit tonic-clonic hindleg convulsions (Table 1). At higher injury levels, the incidence of convulsions was much greater in nonsurvivors than in survivors. The incidence of convulsions at the higher injury levels was not related to the magnitude of injury.

The incidence of transient apneic episodes in which respiration resumed after cessation of at least 10 seconds was not related to the magnitudes of injury greater than 2.1 atm in nonsurvivors. However, in survivors the incidence increased from 10% for the 2.1-atm injury group to 37% for the 3.6-atm injury group (Table 1). All animals that subsequently died developed irreversible apnea even if they had a transient recovery from an initial apneic episode.

All acute somatomotor measurements exhibited a graded response to the magnitude of injury (Fig. 2). The duration of suppression of simple nonpostural responses correlated with the magnitude of injury: pinna, \( r = 0.30 \) (\( p < 0.05 \)); corneal reflex, \( r = 0.44 \) (\( p < 0.05 \)) (Fig. 2A). The duration of suppression of simple somatomotor postural responses was correlated with the magnitude of injury: paw flexion, \( r = 0.51 \) (\( p < 0.05 \)); tail flexion, \( r = 0.53 \) (\( p < 0.05 \)); and head support, \( r = 0.73 \) (\( p < 0.01 \)) (Fig. 2B). The more complex startle response was suppressed longer than all other somatomotor responses and was correlated with the magnitude of injury: \( r = 0.73 \) (\( p < 0.01 \)) (Fig. 2C). A more complex postural function, as assessed by the righting reflex, was suppressed longer than the simple postural responses and was correlated with the magnitude of injury: \( r = 0.47 \) (\( p < 0.05 \)) (Fig. 2D). The duration of disruption of locomotor responses was correlated with the magnitude of injury: escape, \( r = 0.53 \) (\( p < 0.05 \)); spontaneous locomotion, \( r = 0.55 \) (\( p < 0.05 \)) (Fig. 2E).

All sham-treated control animals were observed to be neurologically normal within 1 minute after removal from the injury restrainer.

Chronic Neurological Evaluations

Animals returned to preinjury latencies on the beam-balancing task within 4 days (\( p < 0.05 \), Dunnett's test, Fig. 3A). For the beam-walking task, the time taken to traverse the beam returned to preinjury values within 8 days (\( p < 0.05 \), Dunnett's test, Fig. 3B). Body weight (Fig. 3C), a crude measure of food and water intake, significantly decreased (\( p < 0.05 \)) from a preinjury mean of 373 ± 11 gm to 347 ± 11 gm at 24 hours after injury and remained significantly below preinjury values during the 10-day observation period.

Physiological Studies

The control values of physiological variables for all rats assessed prior to injury were within normal ranges (Table 2 and Fig. 4). Control plasma glucose levels were comparable to those in other reports in which rats were similarly prepared and maintained.

Systemic Cardiovascular Variables

All levels of injury produced increases (\( p < 0.05 \)) in mean arterial blood pressure (MABP) which peaked
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**TABLE 2**

<table>
<thead>
<tr>
<th>Variable</th>
<th>Experimental Group</th>
<th>Pretrauma</th>
<th>Posttrauma</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>5 Min</td>
<td>60 Min</td>
</tr>
<tr>
<td>pO2 (mm Hg)</td>
<td>control</td>
<td>118 ± 12.7</td>
<td>122 ± 9.9</td>
</tr>
<tr>
<td></td>
<td>2.1 atm</td>
<td>117 ± 6.6</td>
<td>104 ± 7.8</td>
</tr>
<tr>
<td></td>
<td>2.9 atm</td>
<td>123 ± 8.1</td>
<td>98 ± 15.4</td>
</tr>
<tr>
<td></td>
<td>3.8 atm</td>
<td>126 ± 4.5</td>
<td>81 ± 16.8†</td>
</tr>
<tr>
<td>pCO2 (mm Hg)</td>
<td>control</td>
<td>35.8 ± 1.1</td>
<td>35.4 ± 1.3</td>
</tr>
<tr>
<td></td>
<td>2.1 atm</td>
<td>36.4 ± 0.8</td>
<td>36.4 ± 1.7</td>
</tr>
<tr>
<td></td>
<td>2.9 atm</td>
<td>35.8 ± 1.6</td>
<td>37.0 ± 2.9</td>
</tr>
<tr>
<td></td>
<td>3.8 atm</td>
<td>35.4 ± 1.2</td>
<td>42.0 ± 3.3</td>
</tr>
<tr>
<td>pH</td>
<td>control</td>
<td>7.48 ± 0.03</td>
<td>7.49 ± 0.02</td>
</tr>
<tr>
<td></td>
<td>2.1 atm</td>
<td>7.49 ± 0.02</td>
<td>7.46 ± 0.03</td>
</tr>
<tr>
<td></td>
<td>2.9 atm</td>
<td>7.45 ± 0.03</td>
<td>7.42 ± 0.04</td>
</tr>
<tr>
<td></td>
<td>3.8 atm</td>
<td>7.46 ± 0.04</td>
<td>7.35 ± 0.05</td>
</tr>
<tr>
<td>glucose (mg/100 ml)</td>
<td>control</td>
<td>165.5 ± 7.1</td>
<td>176.7 ± 16.5</td>
</tr>
<tr>
<td></td>
<td>2.1 atm</td>
<td>174.4 ± 10.2</td>
<td>219.5 ± 27.3†</td>
</tr>
<tr>
<td></td>
<td>2.9 atm</td>
<td>170.0 ± 17.3</td>
<td>251.8 ± 26.2†</td>
</tr>
<tr>
<td></td>
<td>3.8 atm</td>
<td>166.2 ± 7.9</td>
<td>182.0 ± 20.3†</td>
</tr>
</tbody>
</table>

* Values represent means ± standard error of the means.
† Statistically significantly different from preinjury values at p < 0.05.

**Blood Gas and Plasma Glucose Responses**

Animals injured at either 2.1 atm or 2.9 atm demonstrated no significant changes in arterial blood gas levels during the course of the experiment (Table 2). However, animals injured at 3.8 atm demonstrated significant (p < 0.05) decreases in pO2 and increases in pCO2. No differences in hematocrit levels were observed either between or within the injury groups. Plasma glucose levels were significantly higher (p < 0.05) at 5 minutes postinjury for all injury groups and at 60 minutes for the 2.1- and 2.9-atm groups.

**Pulmonary edema, as suggested by the presence of pink fulminating exudate, was observed during routine aspiration in one animal in the 2.1-atm group, two in the 2.9-atm group, and all in the 3.8-atm group. All of these animals exhibited increased CO2 and decreased O2 values in blood.**

In a separate group of animals in which ventilation was systemically interrupted for 15-, 30-, 45-, and 60-second intervals, the mean pO2 values dropped to 70.8 ± 6.3, 50.8 ± 2.2, 44.2 ± 2.7, and 30.4 ± 2.4 mm Hg, respectively, from a baseline value of 132.0 ± 3.0 mm Hg. The pO2 decreased linearly between 15 and 60 seconds.

**Electroencephalographic Findings**

All injury levels produced an immediate reduction in EEG amplitude (Fig. 5) which persisted for 10 to 15 minutes in the 2.1-atm group, for 45 seconds for the 2.9-atm group, and for 5 seconds for the 3.8-atm group. At 60 minutes postinjury, animals injured at 2.9 and 3.8 atm showed a significant (p < 0.05) decrease in MABP, while in animals injured at 2.1 atm the MABP was not significantly different from control values (Fig. 4). A brief (5-to-10 second) period of bradycardia with heart rates dropping to approximately 50% of baseline was always observed immediately following trauma at all injury levels. No other bradycardic or tachycardic trends were observed for the duration of the experiment.

**Histopathological Studies**

Those animals sustaining minor brain injuries (2.1-atm group) were examined after a 4- to 7-day posttrau-
matic survival period. Such animals displayed no overt light microscopic changes. With the exception of subarachnoid blood overlying the traumatized convexities, their brains appeared unremarkable.

Animals subjected to injuries in the range of 2.9 to 3.6 atm demonstrated histological change in both the acute and chronic survival periods. Immediately following injury, subarachnoid blood was visualized under the injury site. Intraparenchymal hemorrhage was seen bilaterally in the frontoparietal and striate cortices at the gray/white matter interface and within the corpus callosum (Fig. 6 left). Additional intraparenchymal hemorrhage was also observed bilaterally in the hippocampus and fimbria hippocampi, with foci of petechial hemorrhage scattered throughout the midline in the pontomesencephalic and cervicomедullary junctions (Fig. 6 right). Occasional petechial hemorrhage was noted within the dorsal thalamus. With increasing post-traumatic survival (4 to 7 days), the frontoparietal and striate cortices showed necrotic change and cavitation, and silver staining revealed the presence of reactive axonal swellings (retraction balls) scattered throughout the brain stem, with their greatest concentration found at the cervicomedullary junction.

In the animals injured at 3.8 atm (100% mortality), changes comparable to those described above were noted, with the exception that the hemorrhage was now more extensive, particularly within the brain stem. Intraparenchymal hemorrhage was again recognized in the frontoparietal and striate cortices, corpus callosum, hippocampus, and fimbria hippocampi. Hemorrhage within the pontomesencephalic and cervicomедullary junctions was extensive and occupied the entire ventral midline portion of these regions (Fig. 7 left). In the presence of such extensive brain-stem hemorrhage, the overlying cerebellum displayed alteration, with hemorrhage occurring within the anterior and posterior lobes and deep cerebellar nuclei (Fig. 7 right). The material stained with silver failed to reveal reactive axonal swellings, as the animals' brief posttraumatic survival time was inadequate to allow for their complete genesis.

**Discussion**

This research has, for the first time, systematically characterized the neurological, physiological, and histopathological responses to graded levels of fluid percussion injury in the rat. In general, greater magnitudes of injury resulted in greater derangements. Lower magnitudes of injury produced a concussion-like injury characterized by brief neurological and systemic physiological alterations without remarkable structural damage. Higher magnitudes of injury resulted in greater neurological and histopathological alterations or death, with extensive structural damage.

Fluid percussion injury in the rat produces transient loss of muscle tone and suppression of many reflexes as well as more complexly organized behaviors. The transient flaccidity and reflex suppression observed following fluid percussion injury is similar to that seen in the cat fluid percussion model, as well as in other head-injury models and species, and closely resembles a reversible flaccid comatose state associated with moderate levels of human head injury.

The present data suggest that mortality in previous neurological studies was generally associated with hemorrhagic pulmonary edema and respiratory dysfunction similar to that observed in other species and head-injury models. It is important to consider the possible consequences of hypoxia associated with varying periods of apnea following injury to normally respiring...
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**Fig. 7.** Photomicrographs of brain sections cut sagittally showing typical alterations seen immediately after a 3.8-atm injury. Intraparenchymal hemorrhage within the cerebrum (left), although more extensive than in the animals receiving less severe injuries, is still confined to the corpus callosum, the cortical gray/white matter interface, and the fimbria hippocampi. The brain stem (right) reveals extensive intraparenchymal hemorrhage confined to the midline of the pontomesencephalic and cervicomedullary junctions, while the cerebellum also shows hemorrhagic focii. H & E. × 10.

animals. While pO₂ levels were not measured in the neurological study, the data from the animals in which ventilation was systematically interrupted suggest that the transient apneic episodes did not produce pO₂ levels of less than 30 mm Hg (50% hemoglobin saturation). Such arterial pO₂ levels in normal rats have previously been shown to result in only minor changes in energy metabolism which are readily reversible even after hypoxic periods of 15 to 30 minutes if perfusion pressure is maintained. Since the apneic period in our neurological studies (Table 1) coincided with the period of hypertension (Fig. 4), we think it unlikely that any major or persistent secondary insults resulted from the apnea seen in this study. Other reports have demonstrated that a rat fluid percussion model is useful in studying combined injury/hypoxia insults when hypoxia is artificially induced following injury.

Unlike other rat models of concussive brain injury, tonic-clonic seizure-like convulsions are not a prominent feature of fluid percussion brain injury. Until recently, experimental head injury in rats has been produced by impacts to the intact skull. Such impact injuries have generally resulted in seizure-like convulsions. Clinical studies have reported the incidence of posttraumatic epileptic seizures following non-military closed-head injury to be 2%. However, the occurrence of convulsions in humans immediately following closed-head injury has not been reliably demonstrated. The EEG tracings showed afterdischarges in our experiments reminiscent in pattern and delay of onset to reported afterdischarges observed following electroconvulsive shock-induced seizures originating in the hippocampus and amygdala. These patterns suggest the possibility of focal subcortical seizures in the present model.

Fluid percussion in the rat produces not only acute reflex suppression and flaccidity but also more enduring neurological deficits similar to deficits observed in other models. Interestingly, recovery of beam-walking ability took twice as long as recovery of beam-balancing ability. The beam-walking test may evaluate somatomotor function and also components of motivational and attentional function. These observations may parallel clinical reports in humans in which higher-order functioning takes longer to recover than does more simple somatomotor functioning.

Fluid percussion injury in rats produces changes in blood-pressure dynamics similar to the changes described in other reports of experimental head injury. A brief period of hypertension occurring immediately following injury has been observed in other models. Also, similar to other reports, the duration of hypertension and the incidence of hypotension were not related to injury magnitude. In contrast to the cat fluid percussion model, peak hypertension and incidence of bradycardia were not related to injury magnitude.

The histopathological changes resembled those observed in other models of head injury. Lower magnitudes of injury produced neurological and physiological signs of concussion in the absence of significant histopathological alterations, while higher injury magnitudes produced subarachnoid and intraparenchymal hemorrhages. Following fluid percussion injury, more reactive axonal swellings were observed in the cervicomedullary junction of the rat than of the cat, thereby suggesting greater spinal involvement. Similar spinal involvement has been reported in rats injured by impact acceleration methods and must be considered in interpreting neurological consequences of fluid percussion injury in the rat.
There are some disadvantages common to all fluid percussion models. Fluid percussion injury fails to replicate the prolonged or irreversible coma observed in many cases of human head injury. Moreover, the biomechanical features of fluid percussion injury have not been as well described as in some acceleration models, although in a recent study, Hayes, et al., considers this aspect in cats. At higher magnitudes of fluid percussion injury in other species, tissue deformation is maximized in the lower brain stem. The present histopathological data suggest that this phenomenon is accentuated in the rat. Thus, unlike acceleration models, fluid percussion injury in the rat is not usually associated with a predominance of supratentorial lesions.

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References

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