Intravascular coagulation: a common phenomenon in minor experimental head injury

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Fibrin microthrombi were demonstrated by an immunoenzymehistochemical method in the small blood vessels of the lung and, to a lesser extent, in the brain in rats after minor experimental head injury. It was concluded that intravascular coagulation is a common phenomenon in head injury.

Key Words • intravascular coagulation • fibrinolysis • pulmonary microthrombi • head injury

Disseminated intravascular coagulation (DIC) is a well known complication of many disorders. Its occurrence in both penetrating and blunt head injury has been recognized more recently. In a large prospective study, laboratory evidence of DIC was found in at least 8% of patients with blunt head injury, and less specific coagulation changes were found in another 32%. However, postmortem examination using conventional fibrin staining methods has revealed some intravascular microthrombi in only a few cases. This discrepancy between laboratory results and morphological findings may be explained by 1) insufficiency of the conventional histological and histochemical fibrin staining techniques, and 2) the occurrence of fibrinolysis during the interval between intravascular clotting and death.

The present experimental study was undertaken to evaluate morphologically the occurrence of intravascular coagulation after blunt head injury using immunoenzymehistochemical fibrin staining methods.

Materials and Methods

Male Wistar rats weighing 200–250 gm were subjected to head injury by an iron pendulum, as described by Bakay, et al. Although the eventual results were not affected by the method of inducing anesthesia, ether anesthesia was preferred to anesthesia induced by pentobarbital (6 mg/100 mg), as the former lasts only a few minutes and thus allows a better evaluation of the clinical consequences of head injury.

Fibrinolysis was inhibited by administration of epsilon-aminocaproic acid (EACA), 200 mg intravenously, 5 minutes before head injury. The head of the animal was then shaved to reduce the loss of traumatic impact and to allow a better positioning of the blow. The animal, still anesthetized, was fixed with sticking plaster in an upright position on a vertical movable wooden board, as shown in Fig. 1, and the pendulum (weight 1370 gm) was released to hit the top of the skull, just in front of the ears and behind the eyes, from a position of 30° to the vertical line of gravity. The wooden board toppled over, thus ensuring a certain acceleration-deceleration effect. Immediately after head injury the sticking plaster was removed.

Control animals underwent an identical procedure except for the blow on the head. Ether anesthesia was induced for a second time at intervals varying in different animals from 5 to 45 minutes after head injury. In the control animals, the second anesthesia was induced 45 minutes after removal of the sticking plaster. Perfusion for 2 minutes (10 ml/min) was performed via the portal vein with phosphate-buffered saline (pH 7.4). Brain and pieces of lung, liver, and kidney were fixed in 4% p-formaldehyde or ethanol 96%, and embedded in Paraplast. Sections were stained for the detection of fibrin microthrombi by an indirect immunoenzymehistochemical method using rabbit antisera against rat fibrin monomer in the first
Clinical Features

When the interval between head injury and second anesthesia was long enough to allow observation, unconsciousness lasted about 10 minutes usually followed by a period of unsteadiness and sluggishness until the moment of induction of the second anesthesia. Some blood loss from the nose after head injury was observed in seven animals, four of which showed a slight dyspnea. Except for a temporary nystagmus occurring in three animals, no neurological deficit was observed. Respiratory arrest occurred in two animals within 1 minute after head injury, making perfusion impossible in one of these.

Macroscopic Examination

On gross examination, the brain and skull of all animals appeared normal, with the exception of one animal that had contusion and hemorrhage of both frontal lobes and a skull fracture. Small petechial subpleural hemorrhages were found in the lungs of seven animals from the group with head injury, and in one control animal. Liver and kidney appeared normal in all cases.

Microscopic Examination

On microscopic examination, small contusional hemorrhages were found in four animals with head injury, in addition to the above-mentioned animal with macroscopic evidence of cerebral contusion. Inter-alveolar edema was found in the lungs of 15 animals, 14 of which had been subjected to head injury. Microscopic evidence of pulmonary hemorrhage was found in 10 animals, eight of which belonged to the group with head injury. Of the latter animals, only three had shown pulmonary hemorrhages macroscopically. In the above-mentioned control animal with macroscopic pulmonary hemorrhages, signs of "shock" lung were observed.

Results

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Intravascular coagulation

Fig. 2. Photomicrographs of various vessels. Indirect immunoperoxidase staining method. A: White blood cell attached to a network of fine fibrin fibers in a pulmonary capillary. × 650. B: Fibrin microthrombus obstructing a pulmonary capillary. × 650. C: Fibrin microthrombus in the middle of a small pulmonary vessel. On the right side, many red cells are seen trapped behind the clot. On the left side are two crescent-shaped schistocytes and a white blood cell. × 650. D: Multiple cerebral capillaries filled with fibrin. × 165. E and F: Brain capillaries filled with fibrin. × 650.

later) were observed, consisting of interstitial and intra-alveolar edema, interstitial and intra-alveolar hemorrhage, and patchy atelectasis. Another control animal showed a single microscopic pulmonary hemorrhage. Liver and kidney appeared normal in all cases.

Immunoenzymehistochemical Staining

Staining with antiserum against fibrin monomer showed fibrillar fibrin(-ogen)-related material (FRM) in the pulmonary capillaries, and sometimes also in small venules and arterioles in 17 of the 19 animals with head injury (Table 1, Fig. 2A–C). Because of its
resistance to p-formaldehyde fixation, this material can be presumed to represent fibrin microthrombi formed in vivo. Occasionally, fragmented red cells or schistocytes were found, which are considered to be characteristic of intravascular coagulation (Fig. 2C). No FRM was found in the lungs of seven of the 10 control animals nor in one of the 19 animals with head injury. Results were equivocal in two of the control animals and in one animal with head injury. There was FRM present in the lung of the control rat with shock lung.

In the brain, fibrillar FRM was found in the capillaries of six of the 19 animals with head injury (Fig. 2D-F). Five of these six animals had con
tusion hemorrhages. Interestingly, FRM was not found in the hemorrhages themselves, but in the surrounding vessels. Otherwise no preferential localization of FRM-positive vessels in the cerebral hemispheres could be determined. Virtually no FRM was detected in the cerebellum. No FRM was found in the brains of control animals.

**Discussion**

This study demonstrates the presence of pulmonary microthrombi in 17 of 19 rats with minor experimental head injury. In six of these 17 cases, microthrombi were also found in the small blood vessels of the brain, which seems to justify the diagnosis of DIC. The occasional observation of schistocytes or fragmented red cells (Fig. 2C) confirms this conclusion, as fragmentation is thought to occur when rapidly moving red cells encounter thin fibrin strands and are subjected to buffeting from other cells in the onrushing blood. The slight degree of the head injury is reflected by an uneventful recovery and the absence of macroscopic and microscopic brain damage in the majority of cases. In a control group of 10 rats, pulmonary microthrombi were present in only one case, which in addition revealed evidence of "shock lung." This syndrome, which is also referred to as "congestive atelectasis" or "pulmonary insufficiency," may be observed not only in trauma, but also in various other conditions, such as hypoxia. Whether the abnormalities suggesting "shock lung" in this case were due to an insufficient perfusion or some other cause is uncertain.

A striking feature is the predominant occurrence of microthrombi in the lung, whereas no fibrin could be demonstrated in liver and kidney. Pulmonary insufficiency after general trauma is a well known complication and may be due to various causes. The occurrence of DIC in trauma has been discussed by McKay and Hardaway, and more recently by Saldeen, who stressed the prevalence of microthrombi in the lungs in contrast to other organs and postulated the theory of the so-called "early" and "delayed microembolism syndrome" after trauma. In the early microembolism syndrome, thromboplastic material is released from the damaged area and enters the venous circulation, subsequently giving rise to the formation of thrombin and fibrin, which is filtered out in the microcirculation of the lungs. In the delayed microembolism syndrome, elimination of microemboli from the pulmonary circulation is impaired on account of inhibition of the fibrinolytic system.

Pulmonary complications following head injury were recognized a long time ago. During World War I, Moutier reported that in several patients with head injury death was due to pulmonary edema occurring within 24 hours after injury. Whether these clinical signs were related to the presence of pulmonary microthrombi, neurogenic pulmonary edema, or to both, is unknown. Pulmonary microthrombi were demonstrated histologically in 24% of patients with fatal head injury with little or no injury elsewhere. Interestingly in this respect are the experiments of Dupuy and de Blainville nearly 150 years ago: they injected an extract of cerebral tissue intravenously in animals and reported clotted blood in heart and blood vessels in addition to petechial hemorrhages in the lungs at postmortem examination.

The hypothesis that tissue thromboplastin is liberated into the circulation is indirectly supported by the demonstration of increased concentrations of myelin basic protein, a nervous system-specific protein, in the serum of patients with severe head injury. Occasionally, even brain tissue emboli have been found in pulmonary vessels after head injury. Recently, posttraumatic DIC has been associated with the finding of brain tissue within the lumen of the sagittal sinus and within the pulmonary arteries.

Another argument in favor of this hypothesis is the presence of more pronounced coagulation abnormalities in the cerebral venous blood than in the systemic venous and arterial blood of patients with head injury. Regardless of the question whether the underlying mechanism should be called "disseminated intravascular coagulation" or "microembolism syndrome," the findings in the present study suggest that, even in cases with minor head injury, intravascular coagulation seems to be the rule rather than the exception. This is in accordance with a previous clinical study that revealed laboratory evidence of DIC much more frequently than expected. To what extent this phenomenon affects the clinical picture and course is still unknown, and this problem needs further investigation.

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