OBSERVATIONS OF THE PHENOMENON OF EXPERIMENTAL CEREBRAL CONCUSSION IN CONSCIOUS RATS

BY MICHAEL WEST, M.D.

A THESIS SUBMITTED TO THE FACULTY OF GRADUATE STUDIES
IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE
DEGREE OF

DOCTOR OF PHILOSOPHY

DEPARTMENT OF PHYSIOLOGY
WINNIPEG, MANITOBA
JUNE, 1980

OBSERVATIONS OF THE PHENOMENON OF EXPERIMENTAL CEREBRAL CONCUSSION IN CONSCIOUS RATS

BY

MICHAEL WEST

A thesis submitted to the Faculty of Graduate Studies of the University of Manitoba in partial fulfillment of the requirements of the degree of

DOCTOR OF PHILOSOPHY

ACKNOWLEDGEMENTS

With sincere gratitude, I acknowledge the enormous contributions made by the following individuals to the completion of this work. A few lines in print cannot completely express my feelings of indebtedness for their constant encouragement and constructive criticism.

Dr. Viktor Havlicek
Dr. Dwight Parkinson
Dr. Henry Friesen
Dr. Frank LaBella
Dr. Nabumaso Kato
Dr. John Taylor
Dr. John Hyde

Invaluable instruction in specialized laboratory techniques and assistance with various technical aspects of this research were provided by:

Ms. Valdine Sundmark
Dr. Allan Wright
Mrs. Sona Pacin
Mr. Doug Stein
Mr. Gary Queen

Very special thanks is due to Miss Emi Okamoto, who typed this manuscript.

I am grateful to the Department of Physiology, for allowing me the privilege of continuing my studies in this stimulating academic environment and to the Medical Research Council of Canada for their support during my Fellowship year (1979-80).

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF FIGURES Cont'd

LIST OF TABLES

OBSERVATIONS OF THE PHENOMENON OF EXPERIMENTAL CONCUSSION IN CONSCIOUS RATS

The syndrome of cerebral concussion has been defined as a transient disturbance of neural function due to the sudden application of mechanical forces to the brain. The most common manifestation is traumatic unconsciousness, due to sudden acceleration or deceleration of the head. Previous authors have studied the experimental phenomenon in anesthetized animal preparations. These studies have been unable to correlate the parameters examined, i.e. neuronal damage, changes in neurotransmitters, intracranial pressure, and cerebral metabolism, with the level of consciousness, due to the use of anesthesia. This investigation began by studying concussion in the clinical situation (i.e. film-strips of boxing ring knock-outs) and dividing concussion into four stages: Stage 1--Impairment of memory, Stage 2--Impairment of memory and mobility; Stage 3--Impairment of mobility and irregular respirations; Stage 4--absent mobility and transient cessation of respirations. Experimental concussion was then produced in conscious rats. The apparatus employed was a spring-loaded pistol which shot a blunt-tipped dart of known mass and velocity. The momentum and kinetic energy of this system was calculated and determined to be constant between experiments. With these materials, concussion could be repeatably produced in Sprague-Dawley rats (males; 200-250 grams) and graded in a scale similar to the human situation.

PART I

Rats were trained in a multiple-choice maze before and after concussion. Post-concussion amnesia was suggested by the facts that rats demonstrated no fear on repeated handling in the experimental environment and that there was a short-lived period during which performance in the previously learned maze was markedly impaired. Compared to its baseline performance, each rat now required a much longer period of time in which to negotiate the maze, and made more errors in doing so. Performance usually returned to baseline within one hour of concussion.

PART II

The electroencephalographic response to concussion was then studied in 32 non-anesthetized Sprague-Dawley male rats, previously implanted with chronic epidural electrodes for bipolar recording of the electroencephalogram (EEG). EEG records were taken continuously preceding, during and following concussion. Spectral analysis of the EEG was performed by means of a PDP8 computer, using fast Fourier transform. In Stages 1-2 of concussion, the power spectra of alpha, beta, and theta frequency bands were decreased by 25 percent, 37 percent, and 10 percent respectively. The delta spectrum alone was increased by 15 percent. Stages 3-4 of concussion were characterized by more profound depression of the power spectra of alpha, beta, theta, and delta frequency bands (56 percent, 49 percent, 34 percent, and 31 percent respectively). EEG depression occurred immediately, reached a peak at five minutes and recovered at an average of two hours in the concussed rats. In five rats, which did not recover completely, post-mortem examination revealed cerebral contusion or intracranial hematomas. These rats were not included in the analysis of "concussion". The EEG changes suggest the involvement of the cerebral cortex in the traumatic unconsciousness of concussion, and are similar to those of spreading depression, as noted by previous investigators. The rapid recovery of the EEG which was observed suggests that the changes which produce this syndrome are functional, rather than structural.

PART III

It is generally accepted that pressure gradients are produced within the brain substance at the moment of impact, due to inertial loading of the brain. These pressure gradients, and the well-documented rotation of the brain in response to acceleration trauma, produce shear strains within the cerebral cortex and craniospinal junction. Based upon the hypothesis that such an abrupt imposition of pressure gradients may produce transient perturbations of neuronal

membranes, and as a consequence, of neurotransmission, the effect of concussion on receptor binding was studied. Sprague-Dawley rats (males; 200-250 grams) were decapitated within two seconds of the concussing blow. The brains were immediately removed and, following dissection on ice, were tested for receptor binding using one of the following radioreceptor assays; opiate, cholinergic, spiroperidol, and ouabain. The results suggested significantly decreased binding of endogenous opiates and cholinergic transmitters to their receptor sites at the time of concussion in the hypothalamus specifically. Five to ten seconds following concussion, once the rats had regained consciousness, binding had returned to normal. Spiroperidol and ouabain binding were unchanged in any brain region.

The brain content of immunoreactive β-endorphin was also studied. This potent inhibitory peptide was decreased significantly in the hypothalamus alone, within five seconds of concussion. This decrease was detectable for 45 minutes following concussion. The change in content most probably reflects increased release of β-endorphin from hypothalamic neurons.

These results suggest that the interruption of specific neuronal circuits in the hypothalamus may contribute to the production of traumatic unconsciousness. Following the acute impairment of receptor binding, secretion of endorphin may continue at higher than normal values, accounting for continued suppression of neural function. The observed alteration of β-endorphin is much more dramatic than that previously recorded following acute stress, but the possibility remains that this change may be a response specific to stress rather than to concussion.

Overall, our experiments suggest that cerebral concussion relates to a transient functional, rather than a persistent structural change in the nervous system. No single subunit of this system can be singled out as being responsible for the entire complex of clinical signs.

OBSERVATIONS OF THE PHENOMENON OF EXPERIMENTAL CEREBRAL CONCUSSION IN CONSCIOUS RATS

INTRODUCTION

Trauma to the central nervous system has become the most common form of neurological disease (Caveness (1977). Loss of consciousness secondary to head injury is a frequent feature of such trauma. Many cases of permanent post-traumatic unconsciousness may be explained by the post-mortem observations of generalized neuronal damage in the cortex and/or brain stem. More frequently, however, the injury is less severe and produces unconsciousness followed by apparent complete recovery. This latter syndrome of transient loss of consciousness has been termed "concussion", and has been the subject of considerable investigation.

The word "concussion" is derived from the Latin "concutere" which means "to shake violently". Ambrose Paré (1510-1590) and his contemporaries used this term to describe the "ébranlement du cerveau" (shaking of the brain) which they believed was responsible for the sudden neuronal paralysis which followed a concussive head injury. These early investigators believed that no structural defect in the brain need occur to account for the clinical picture. Littré (1705) substantiated this opinion when he performed a post-mortem examination on a patient who ran head first into a wall, was instantly rendered unconscious, and died minutes later. There was no macroscopic evidence of damage to the brain. Similarly, Polis (1894) described patients who had suffered a concussion and died, in whom he could demonstrate no pathological lesions.

Before discussing the phenomenon of concussion, it is advisable to define the normal state of consciousness. This concept is best outlined in Ommaya's definition as:

> "that state of awareness in the organism which is characterized by maximum capacity to utilize its sensory input and motor output potential in order to achieve accurate storage and retrieval of events related to contemporary time and space."

The classic definition of concussion appears in the authoritative study of Denny-Brown and Russell (1941); "an essentially reversible syndrome (of neural dysfunction) without detectable pathology". The current generally accepted definition is that of the Committee to Study Head Injury Nomenclature (1966):

> "a clinical syndrome characterized as an immediate and transient impairment of neural function, such as alteration of consciousness, equilibrium, etc., due to mechanical forces".

It has been emphasized that, although usually reversible, a concussion may be fatal. For instance, if the respiratory and vasomotor centers of the medulla are transiently paralyzed, the affected individual may expire before these centers have had time to recover (Fulton, 1942; Symonds, 1974; Gurdjian, 1975).

For the reader who has not witnessed the instantaneous loss of neural function produced by a concussing blow, the eloquent description of Sir Charles Sherrington (1906) is instructive:

> "the knock-out blow...reduces in a moment a vigorous athlete to an unstrung bulk of flesh, whose weight alone determines its attitude, if indeed a reactionless mass can be described as having attitude at all".

Loss of consciousness and postural reflexes are fundamental signs of concussion in Sherrington's description of boxers. Maintenance of consciousness depends upon continuing interplay between the cerebral cortex and the brain stem reticular formation, while maintenance of posture depends upon a large number of complex reflexes encompassing almost the entire brain stem and spinal cord, with modulation by the cerebral cortex (Govons, 1968).

Walker (1973) also described the clinical picture:

> "the classic 'concussion' is featured by 'a fall like a log' with unresponsiveness for varying periods of time. However, other types may occur--a stiffening of the body in extension, a gradual slumping with glassy apparently unseeing eyes, and even retention of posture and some automatic movements with no response to the environment".

In man, concomitants of loss of consciousness are an impairment of higher neurological functions, especially memory (attested to by the frequent questions "where am I?" and "what happened?"); also, transient apnea, bradycardia, hypotension, and impairment of brain stem reflexes.

Many anecdotal reports have been derived from observations made in sports such as boxing, football, soccer and polo, where head injuries are not uncommon. The impairment of memory described by Walker is common to such reports. Amnesia may occur for minutes or hours prior to the injury, in which case, it is known as retrograde amnesia. Amnesia occurring for different time periods following the injury is known as post-traumatic, or antegrade amnesia. It is known that post-traumatic amnesia may follow trauma that does not produce unconsciousness (Fisher, 1966, Symonds, 1966). This period of post-traumatic amnesia has been likened to the "epileptic twilight state" (petit mal epilepsy) (Winterstein, 1937) in that the behavior of the individual appears to be "automatic". For example, after being concussed, the boxer or football player may continue, successfully, at his match and when questioned later, have no recollection of the concussing blow, nor of the events following it (Martland, 1928; Blonstein and Clarke, 1957; Yarnell and Lynch, 1970, 1973).

Following concussion, return of awareness to stimuli usually precedes sensory and motor recovery, which in turn, recover before complete restoration of memory and other cognitive functions (Ommaya and Gennarelli, 1975). This sequence reflects the dictum that the higher levels of the nervous system are more susceptible to injury and require longer periods for recovery from such an injury.

Although less frequently recognized than loss of consciousness, concussion may also produce transient focal neurological deficits, such as paresis in an arm or leg, without clouding of the sensorium.

Discussion to this point has emphasized the transient nature of the disturbance of neural function. Several reports have claimed to show neuronal chromatolysis and diminished neuronal cell counts in the brain stem reticular formation and related areas following concussion in anesthetized animals (Windle, et al. 1944; Groat, et al. 1945 A, B; Groat and Simmons, 1950; Chason, et al. 1957; Brown, et al, 1972). As a result of such studies, several scientists and clinicians have accepted the conclusion that concussion always produces irreversible brain damage. The quoted studies were poorly controlled and used relatively primitive techniques, but are still widely quoted. More recent, well-controlled studies, using both light and electron microscopy have demonstrated temporary mitochondrial swelling and abnormal microvascular permeability, but have not confirmed the presence of permanent structural damage (see below).

The controversy regarding the presence of permanent structural alterations following concussion is far from resolved. However, when head injury is viewed as a spectrum, ranging from mild concussion to severe cerebral laceration, it is obvious that there will be an overlap between the different gradations of severity. Hence, concussion occurs as a clinical syndrome that may be associated with, but unrelated to a variety of pathological states (Kaplan and Browder, 1954). In these complicated cases, one would be mistaken to attempt to relate the dramatic, generalized, transient loss of neural function to the permanent loss of a small circumscribed population of neurons. In an attempt to avoid the controversy that surrounds the term concussion, Walker (1973) has introduced the term "traumatic unconsciousness". Traumatic unconsciousness is common to a wide variety of head injuries--it is the basic response of the nervous system to trauma. It may be seen alone--in concussion--or may be complicated by irreversible brain damage in more serious injuries. Less common than cerebral concussion and also of unknown etiology are concussion of the spinal cord or of a peripheral nerve, which may

produce a transient loss of neural function. The latter entities are not within the scope of this paper and will not be discussed further.

Since the syndrome of concussion is one of the minor forms of injury to the nervous system, appears to be reversible, and requires no special treatment, why then has it attracted so much continuing attention? One reason is that the pathophysiology of the syndrome remains unknown in spite of numerous investigations. Another is that the cellular substrate of concussion may contribute in some way to the prolonged traumatic unconsciousness frequently observed in humans, which may last several weeks, to be followed eventually by a good recovery (Becker, 1978). Becker speculated that neurons may have been rendered "dysfunctional" during this period, although morphologically intact and therefore, capable of recovery. His views are in agreement with those of Govons (1968), who stated that "within the limits of our present knowledge, no definite time limit can be set for maximum duration of coma following a concussive blow".

Understanding the pathophysiology of concussion may conceivably lead to improved treatment of central nervous system trauma.

MECHANISMS OF HEAD INJURY

The brain of all species is well-protected from low velocity blunt injuries by a series of protective layers. The scalp although thin and vascular, is important in that it is free to slide, which helps to absorb applied tangential forces (Gurdjian, 1975); it also absorbs significant energy when it is disrupted. In both examples, the scalp aids in absorbing forces that otherwise would be transmitted to the brain. Similarly the skull, consisting of two layers of cortical bone, separated by a delicate lattice of marrow, is analogous to a corrugated cardboard box. Significant forces must be applied to this structure before it is penetrated, and hence, it is another shock-absorber. From this point onwards, the protective devices are rather flimsy. The dura, arachnoid and their processes aid in the support of the brain, as does the cerebrospinal fluid in which the brain "floats".

No combination of the above-mentioned protective layers will protect the brain from blunt injuries which apply acceleration or deceleration forces to the head. The importance of acceleration-deceleration forces in the production of concussion was elucidated by Denny-Brown and Russell (1941) and Denny-Brown (1945) who were the first investigators to appreciate the fact that damage to the brain is not proportional to the energy of the striking blow alone. They found that it was difficult, if not impossible, to produce experimental concussion in cats when the head was fixed, but relatively easy when the head was free to move. Holbourn (1943, 1945) further underscored the importance of acceleration, in particular rotational acceleration. Employing gelatin models of the brain, he surmised that shear strains would be more severe with rotational (angular) than with translational (linear) acceleration. This fact has become generally accepted because of the large body of clinical and experimental evidence suporting it. (Pudenz and Shelden, 1947; Rowbotham, 1964; Sellier and Unterharnscheidt, 1966; Ommaya, et al.

1968; Yarnell and Ommaya, 1969; Joseph and Crisp, 1971; Adams and Graham, 1972; Ripperger, 1975). Translational (linear) acceleration is such that all parts of the impacted body are accelerated uniformly. Rotational (angular) acceleration is more complex in that there occurs asymmetrical acceleration of the impacted body, in relation to its center of gravity (Fig. 1). During rotational acceleration, the brain tends to remain essentially stationary as the skull rotates. This can be compared to water in a bucket which remains at rest while the bucket is twirled around its geometric axis. Such rotation produces shear stresses, which are maximal at the brain surface, and decrease at increased distance from the surface (Joseph and Crisp, 1971; Ripperger, 1975; Ommaya and Gennarelli, 1975). Nature also supports this theory. May, et al. (1979) studied a natural model in which the effects of acceleration may be observed. The woodpecker may pound its head incessantly without the development of concussion. The reason is probably that, as high-speed cinematography has shown, the acceleration of the woodpecker's head in linear, rather than rotational. This movement requires more complex neuromuscular innervation than if the acceleration were rotational. The authors suggested that linear acceleration had been "selected" because it is safer.

Ommaya and Gennarelli (1975) have shown that while translational acceleration is most likely to produce focal lesions (such as temporal lobe contusions and hemorrhages), such focal lesions do not often result in concussion.

Rotational acceleration, on the other hand, is more effective in producing diffuse brain injury and hence, resulting in cerebral concussion. More recently, Joseph and Crisp (1971) have documented elegant mathematical analyses of the mechanics of brain movement within the skull at the time of injury and confirmed Holbourn's theories. The most convincing clinical correlate is the efficient use of an "upper-cut" blow in boxing. This type of a blow--striking the chin sideways and upwards--is very effective in producing a concussion, secondary to rotational

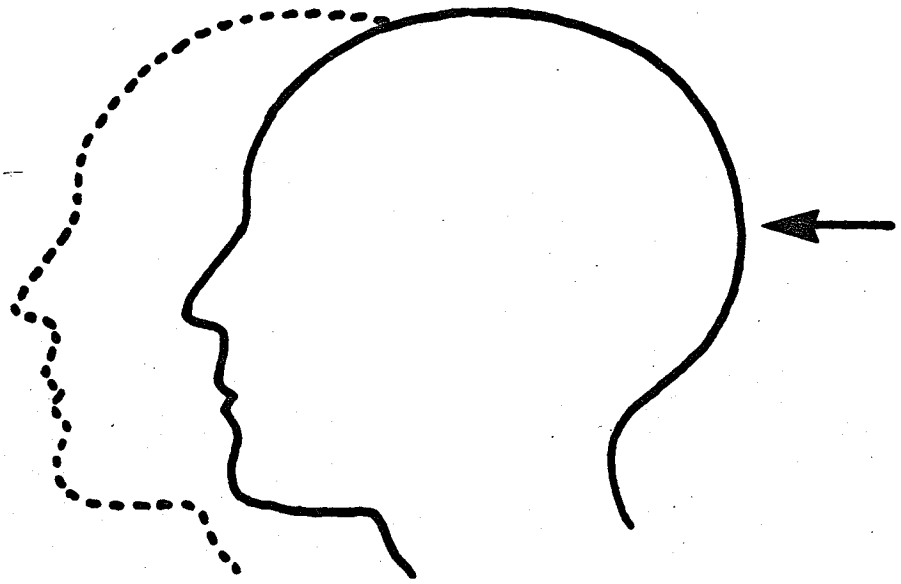
Figure 1

Ommaya (1976) has confirmed the more dramatic effects of rotational acceleration (the head is accelerated asymmetrically in relation to its center of gravity) compared to translational acceleration (all parts of the head are accelerated uniformly).

The solid lines indicate head position pre-acceleration. The interrupted lines indicate head position post-impact.

The accelerating force is from right to left (arrow).

TRANSLATION (LINEAR)



ROTATION (ANGULAR)

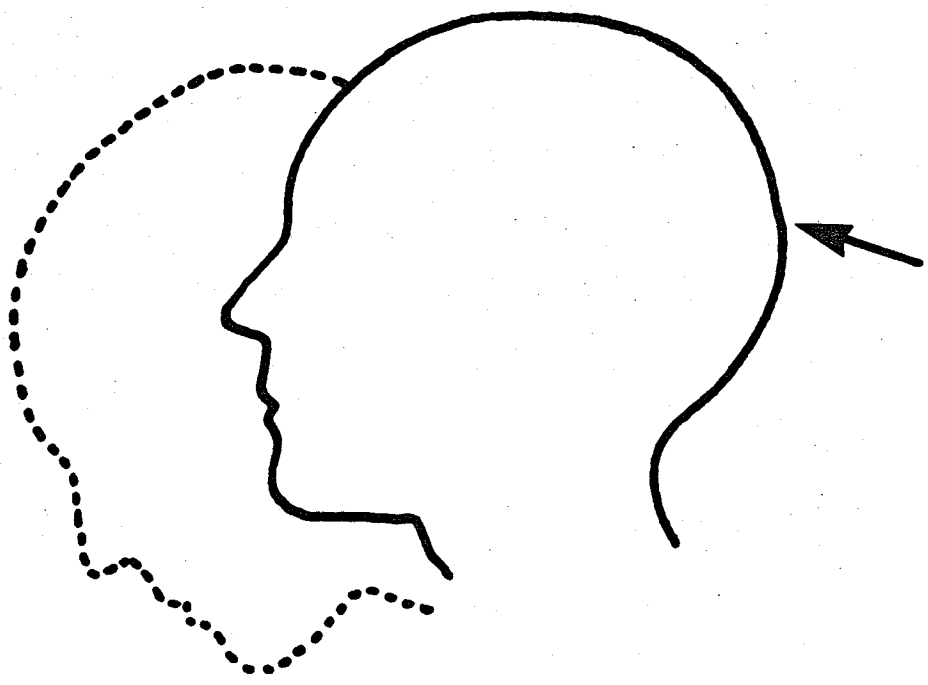


FIGURE 1
(modified from Ommaya, 1976)

acceleration to the head.

Several authors have recorded movement of the brain within the skull at the time of experimental concussion (Shelden and Pudenz, 1947; Gurdjian, et al. 1968; Gosch, et al. 1970; Ommaya, et al. 1974; and Shatsky, et al. 1974). The original experiments of Shelden and Pudenz were ingenious. These authors replaced the calvaria of monkeys with form-fitting lucite "calvaria", through which the brain could be observed. When an impact producing rotational acceleration was applied to the skull, the brain could be observed, by high-speed cinemaphotography, to "swirl" around within the calvarium.

Having witnessed this swirling movement of the brain within the skull at the time of concussion, succeeding experiments focussed upon the question as to whether or not this sudden, violent movement of the brain was accompanied by any variation in intracranial pressure (ICP).

Scott (1940) measured intracranial pressure in anesthetized dogs subjected to a head injury by a weight dropped on a fixed, immobile head. This study reported results from a model which would now be described as a compression-type injury. However, Scott did record an instantaneous, but transient elevation of intracranial pressure, which averaged 300 mm Hg. and lasted .2-.6 seconds.

Walker, et al. (1944) also demonstrated elevation of intracranial pressure in an experimental model of concussion. These authors believed that it was not the absolute height of the ICP, but rather oscillations in the ICP that were important. They believed that, through the production of pressure-waves, damaging shearing forces would be produced within the brain substance.

Gurdjian, et al. (1954) demonstrated that, although acceleration was important in the production of concussion, unless the impact acceleration produced a