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# Cerebrospinal fluid : with special reference to its formation, absorption and circulation

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# The Cerebrospinal Fluid with especial Reference to its Formation, Absorption and Circulation

by

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Outline and Table of Contents

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# I. Introduction

Comparing the amount of work done on the cerebrospinal fluid with what is actually known about the fluid I feel that it is quite a neglected subject. Although it has become important in the diagnosis of central nervous system disease and considerable work has been done on its composition and on its variation in composition in the various neurological diseases its mode of formation, absorption and relation to the central nervous system still are an enigma in many ways.

Unfortunately, the most vital problems concerning its formation and absorption are as yet unsolved. These problems are not problems unique to the study of the cerebrospinal fluid but are very far reaching, involving the question of capillary permeability in general, the osmotic relationships of fluids and the influence on cellular permeability of charged ions and certain substances in particular. However, I do not mean to imply that the riddles concerning the cerebrospinal fluid may be solved by studying another tissue in the body. In addition to possessing characteristics common to other tissues the brain presents peculiar anatomical features such as the ependymal lining through which the cerebrospinal fluid must pass and the absence of a true lymphatic system. The brain also possesses the unique feature

of being enclosed in a rigid box, the contents of that box being forced to occupy the same volume at all times. In the regulation of intracranial pressure the cerebrospinal fluid plays a vital part and it is for this function that its absorption and formation are undoubtedly so delicately controlled. It is this labile nature of the functions we will study that has made investigation of the cerebrospinal fluid so difficult. In no field is a careful control of the experiments being conducted more necessary. In no field is it more necessary to preserve physiological conditions during the experiments; its site of absorption and formation probably change with the conditions. Failure to observe these facts probably accounts for the contradictory results which have so often been obtained by different investigators.

I shall attempt to show how little we really know about the cerebrospinal fluid and yet how vitally important it is that we attempt to gain a correct knowledge of the mechanism of formation and absorption of this most interesting fluid.

## II. Historical Resume of the Knowledge of the Human Cerebrospinal Fluid

Some believe that the history of the cerebrospinal fluid begins with the Greeks.[67] This is due to the out-

standing eminence of Greece in medicine and surgery which has led many to believe that modern medicine was founded in Greece. This is held by others to be pure fallacy and they even go so far as to prove that both medicine and surgery were handed down in a high state of development, the Greeks receiving them through a long line of other civilized peoples from a remote antiquity. To prove their point, they say the first weapon in the hands of the first surgeon was a flint, and trepanning for the release of demons (early medicine being shrouded in mystery and magic) is the earliest surgical operation of which any evidence remains; and skulls bear witness to what was done "aeons before the dawn of civilization".[45] Thus they would prove that the history of the human cerebrospinal fluid goes back to the dawn of humanity as the fossil records disclose. We know that it was customary for the Greeks to tap the brain in certain disease states, such as hydrocepalus. They would not have done this had they not been aware of accumulations of fluid in the brain. For this reason some believe the Greeks to have been the first to note the presence of a cerebrospinal fluid.

## Historical Resume of the Knowledge of the Human Cerebrospinal Fluid

Even the semi-civilized peoples of the Americas performed craniotomy, probably for hydrocepalus. It is not certain whether the Greeks regarded the mucus which came from the posterior nasopharyngeal space as a fluid exuded from the brain. We do know, however, that they called it after the pituitary gland, thus they must have felt that it had its origin in that structure.[67]

The presence of a fluid both in the ventricles of the brain and the subarachnoid space was noted either in human beings or animals by Galen, Vesalius, Variolus, Valsalva, Sommering, Contugno and Haller.[48]

Claudius Galen with his immense knowledge dominated medical thought for fifteen centuries. His prolific writings with the doctrines he formulated, the compound medicines he either introduced or endorsed and the treatments he recommended comprised all that was known of medicine in his time. Someone has said "Galen was the last of the Greeks and when he spoke no more, the voice of the ancient world was hushed. Galen was the final star that shone in the twilight of antiquity, and when his effulgence was extinguished, there settled over Europe a darkness that was not lifted for many centuries." [45] It was Galen who described an excretion,

watery in character, emanating from the brain and collecting in the ventricles, whence it was forced into the nasal cavity through the ethmoid bone, by way of the infundibular process of the pituitary gland. It was he, who in the 16th century, knew that the ventricles as well as the chorioid plexus, contained a watery humor, yet he gave little or no weight to its importance or distribution. He was probably the first to recognize the fact that the normal content of the ventricles is fluid. Vesalius was the founder of modern anatomy. He is accorded the honor of having really performed the first dissection of a human body, and made observations unblinded by the veneration of Galen.

We have to go on to the end of the eighteenth century before we find investigators who recognized the presence of a fluid in the brain and chord even though they did not understand its nature or purpose.

The credit of having recognized the cerebrospinal fluid as a real, possibly circulating medium must be divided among Valsalva, Sommering, Contugno [9] and Baron Albertus Haller of Gottingen. These men were all investigators of the eighteenth century. Contugno is credited by some with the discovery for he found the

in like proportion absorbed again by the inhaling veins: or if any part abounds, that it decends through the bottom of the ventricles to the basis of the skull, and from thence into the loose cavity of the spinal medulla. That this is the case appears from the watery tumours in the lower part of the spinal medulla following in those who have an hydrocephalus." And again he says: "The cavity, of which these is one in each hemisphere of the brain, is called its anterior ventricle; and it is naturally filled with a vapour, which is frequently condensed into water or jelly." As for the cord: "Between the arachnoides and the dura water there exhales a vapour, which is frequently condensed into a reddish water and produces a true dropsy." His conclusions were that the "nervous fluid is the instrument of sense and motion and consists of elastic and electrical matter."

Experiments to show the protective character of the cerebrospinal fluid did not develop until another half century, and we are indebted to Francois Magendie for this advance. Magendie found that the space between the arachnoid and the medulla is much larger than is necessary to contain the organ, and that this space is filled up with a serous liquid, and that this liquid is under some pres-

sure, "for when the membrane is punctured it spurts out many inches in height." Magendie said that it was easy to see how efficacious the protection was from this liquid, and that it also surrounded the spinal marrow. Magendie was followed by Luschka and Naunyn with anatomical and physical discussions of the fluid . Investigators were beginning to speculate as to the origin, the distribution and circulation of the cerebrospinal fluid. What was it, an exudate, a transudate, or a secretion? It had been studied by obtaining it through a trephine opening in the skull, but, as this was a tedious and rather dangerous method, other means for obtaining it were being sought. Corning, in 1885, introduced a needle into the intravertebral cavity for the purpose of cocainizing the cord in certain spinal affections. His punctures were not for withdrawing any of the fluid. This is the first record we have of a spinal puncture. Unfortunately Corning left no record of his technique but we know that he entered at one of the lower dorsal levels.

Six years later we have three men, Quincke, Wynter and Morton, all with experiments with punctures for the removal of fluid. Wynter had drained the cerebrospinal

fluid in a case of tuberculous meningitis. Having made a skin incision at the level of the second lumbar vertebra, somewhat lateral to the spine, he introduced a trocar and cannula through the wound. By manipulation of the trocar downward and towards the median line, he finally entered the canal. As the fluid was drained continuously through a slender rubber cannula, he noted much improvement in the immediate symptoms of the disease. He followed this same procedure in several cases of tuberculous meningitis, giving temporary improvement in all, although the ultimate results of the disease were fatal.

Although the three men mentioned above all carried on their experiments simultaneously, most credit must go to Quincke.[55] It was he who first showed that the subarachnoid space could be punctured with a needle without incising the skin, that the fluid could be removed, and that diagnostic aid could be derived from its study. Quincke's technique for spinal puncture required no skin incision, needed no trocar and cannula; also he applied it at a much safer level for the integrity of the cord. He entered the spinal canal between the fourth and fifth lumbar vertebra, using a plain, strong needle. He was

able to secure much relief in the symptoms of several cases of hydrocepalus which he treated in this manner. From this, he concluded that spinal puncture for therapeutic purposes was indicated in cases of increased intracerebral pressure. Quincke measured the pressure of the fluid and examined it chemically and physically, although more or less crudely. Lichtheim, who followed Quincke, applied his technique with success to many meningitic conditions. As soon as the results of these investigators were published, many punctures were done not only in Europe but in America as well. The cytologic studies of Widal, Sicord, Ravant (1900 and 1901); the serologic studies of Plant (1909); the chemical studies of Mestrezat (1911-1912) and Lange (1912) (colloidal gold reaction); and the biologic studies of Kafka (1911-1930):- these were all advances in the understanding of the cerebrospinal fluid.

In 1912, with the publication of Mestrezat's [49] monograph, a new era began in knowledge concerning the composition of the normal cerebrospinal fluid and the changes it undergoes in disease. Mestrezat had the theory that the cerebrospinal fluid is a dialysate in equilibrium with the blood plasma. Following Deniges

and Sabroyes who suggested the idea, Mestrezat formulated cerebrospinal fluid syndromes characteristic for various diseases based on quantitative chemical analysis of the fluid.

In the early part of this century, Nonne emphasized the clinical importance of the examination of this fluid in syphilis of the nervous system. His "four reactions" in such cases were much written up in medical literature for over twenty years. The chemical, bacteriological and physical examinations have been more and more perfected: differences of pressures at varying levels, with their diagnostic import, have been scrutinized; the introduction of cisterna-magna puncture by Ayer in 1920, has added greatly to our means for diagnosis and therapeusis. The most recent contributions to the diagnostic use of the different punctures are the air-injection methods of Dandy (1918) and the lipiodol injection method of Sicord in Paris and of Forestier (1921). Sicord used the injection of lipiodol into the cord in order to facilitate X-ray examination.

Since 1774, when Contugno has generally been credited with the discovery of the cerebrospinal fluid, an immense amount of work has been directed to an

understanding of the fluid. However, even today dissenting answers are given to its most elementary problems and agreement is often just within the school of a particular investigator. Various results have been obtained where identical methods have been used and various interpretations have been given to equivalent results.

## III. Anatomy

A correct understanding of the anatomy of the ventricular canties and of the envelope of the brain is essential before we can consider the formation, circulation and absorption of the cerebrospinal fluid.

The ventricles really represent simply modifications of the primitive neural canal. Due to the high state of development which the brain has reached in man and the manner in which the cerebrum has folded itself back upon the diencephalon the embryological relationships have been considerably distorted but the ventricular cavities remain continuous with the canal of the spinal cord, the foraminaof Monro and the aqueduct of Sylvius being only constrictions of the primitive neuralcanal.

## Anatomy

The lateral ventricle consists of a body, anterior, posterior and inferior horns. It communicates with the third ventricle through the interventricular foramen of Monro. The anterior horn projects forward from the interventricular foramen to the genu of the corpus callosum. The body extends posteriorly from the interventricular foramen as far as the spleniam of the corpus callosum. It contains choroid plexas which is continuous with that of the third ventricle caudally and with that of the inferior horn rostrally. The body of the lateral ventricle near the splenium bifurcates into the posterior and inferior horns. The posterior horn extends into the occipital lobe and the inferior horn into the temporal lobe. The posterior horn contains no choroid plexus.

The third ventricle lies between the two thalami and just dorsal to the hypothalamus. Its roof is formed by thin ependyma containing the vascular tufts of pia mater known as the choroid plexus of the third ventricle. It communicates anteriorly with the lateral ventricles through the foramina of Monro and posteriorly with the fourth ventricle through the aqueduct of Sylvius. It's choroid plexus is continuous with the choroid plexus of

the lateral ventricle but not with that of the fourth ventricle.

The fourth ventricle is situated in front of the cerebellum and behind the pons and upper half of the medulla. It is continuous superiorly with the aqueduct of Sylvius and inferiorly with the central canal of the spinal cord. The choroid plexus of the fourth ventricle is located in the inferior portion of the roof of the ventricle. It consists of two vertical inflexions and one horizontal inflexion of highly vascular pia mater so that it has the appearance of a T. Its shape suggests the fundamental bilateral character of the plexuses.

The fourth ventricle is prolonged laterally on the dorsal surface of the restiform body and just inferior to the brachium pontis to form the lateral recesses. The tips of these recesses are open and communicate with the subarachnoid space. These foramina are known as the foramina of Luschka. There is another questionable opening at the inferior tip of the ventricle in the mid-line known as the foramen of Magendie.

The ventricles are lined throughout by ependyma which in the embryo is ciliated but which in the adult

tends to become non-ciliated. The choroid plexuses consist of highly vascular tufts of the pia mater which invaginate this ependymal lining in the regions mentioned above. The ventricles, it must be understood are completely closed cavities at all places except at the foramina of Luschka and Magendie. A double epithelial lining is interposed between the blood stream and the cerebrospinal fluid (the capillary endothelium and the ependymal epithelium).

The meninges, as we all know, consist of three layers of modified mesoderm, the dura, the arachnoid and the pia mater. The dura is tough and adherent to the cranium. The subdural space is more of a potential space than an actual space and has no connection with the cerebrospinal fluid as will be shown later. The subarachnoid space is the space which interests us most in the study of the cerebrospinal fluid as it is directly continuous with the ventricles through the foramina of Luschka and Magendie. The arachnoid is a thin lacy network (arachnoid=spider form) bridging across the sulci of the brain. The pia mater (tender mother) is a very delicate, highly vascular membrane following the surface of the brain in all its convolutions. Thus the subarachnoid

space varies in thickness depending upon the particular location involved. The space is traversed by numerous fibrous trabeculae so that instead of being a single continuous space it is really a series of little spaces, which, however, all communicate with one another.

In certain areas around the brain the subarachnoid space becomes of considerable magnitude. These areas are called cisterns and the ones of most importance are as follows: 1. the cisterna magna (cerebellomedullaris), located beneath the cerebellum and posterior to the medulla; 2. the cisterna pontis, located on the ventral surface of the pons and containing the basilar artery; 3. the cisterna interpeduncularis, extending between the two temporal lobes and containing the circle of Willis; 4. the cisterna chiasmatis, the anterior part of the interpeduncular cistern; 5. the cisterna fossae cerebri lateralis, located in front of the temporal lobes and formed by the arachnoid bridging across the lateral fissure; 6. the cisterna venae magnae cerebri, situated between the splenium of the corpus callosum and the superior surface of the cerebellum and containing the great vein of Galen.

The arachnoid membrane in certain areas projects

into the venous sinuses forming structures known either as Pacchionian granulations or arachnoid villi. The Pacchionian granulations are simply pathological hypertrophic arachnoidal villi (Weed, 69). The arachnoidal villi are evaginations of the arachnoid, and consist of fine reticular fibers arranged in delicate web-like manner. They stain like myxomatous tissue and Weed considers them to be true myxomatous tissue. These villi frequently have a vein forming their core; the vein traverses the villus on its way to the sinus. The villi are capped by mesothelial cells which usually are one layer thick but which may become several layers thick and quite redundant. This mesothelial cap is derived from the outer layer of the arachnoid with which it is continuous. There is considerable doubt as to whether the endothelial lining of the venous sinus is continuous over the cap of the villus or not. Consensus of opinion favors the view that it is continuous, however. The inner wall of the dura, ie: the wall nearest the arachnoid, is not continuous, however. Thus the cerebrospinal fluid in passing through an arachnoid villus must traverse two layers of epithelial cells - those of the villus and those of the endothelium of the blood vessel.

## Anatomy

The villi are most numerous in the areas overlying the sulci, a fact which intimates that the villi are placed in the main channels of circulation of the cerebrospinal fluid for a purpose. They are not present in the spinal region but are found in the superior longitudinal sinus, the transverse sinus, the cavernous sinus, the superior petrosal sinus, and the middle meningeal veins.

The Pacchionian granulations are not present in infants and in some animals. The subarachnoid space is prolonged for varying distances along the cranial and spinal nerves. It's relationship to the nerves will be taken up under a discussion of the "Sites of absorption of the cerebrospinal fluid".

We have one other anatomical point of importance to discuss. It concerns the perivascular spaces and their connections. These spaces were first described by Pestalozzi [99] in 1849. Subsequently Virchow[116](1851), Robin[103](1859), His[92](1865) and Roth[105](1869) made contributions to the subject. The perivascular spaces are now sometimes called the Virchow-Robins spaces.

As the vessels enter the brain substance they carry with them a cuff of the leptomeninges, the pia mater being on the outside and the arachnoid on the inside.

Anatomy

It is really thus an invagination. The perivascular spaces are those lying between the pia and arachnoid. Whether or not these spaces are lined by true mesothelial cells or merely semi-specialized fibroblasts has caused considerable debate but for our purposes we may consider them mesothelially lined. The pia mater of the space lies in close apposition to a layer of glial cells, the two together being called the pia-glial membrane. Just how far the leptomeningeal cuff follows the blood vessels is not known but at any rate a continuous space is present from the nerve cell clear to the subarachnoid space. The arachnoid probably blends with the adventitia of the blood vessels. Where the leptomeninges cease the glial membrane continues on inward to form the boundary of the perineuronal spaces. Here it is called the membrana glia limitans. The pericapillary spaces thus are probably lined with glial cells and by the wall of the capillaries. The important point is that between the blood and the brain tissue proper there is always interposed a membrane of some sort - a barrier, as it were, to the passage of certain substances into the brain cells. This barrier, called the hemato-encephalic barrier is composed of choroid

plexes and ependyma in the ventricles and in the depths of the central nervous system itself of capillary endothelium histiocytes and glial cells. In the larger perivascular spaces the pia mater also enters into the formation of the barrier.

The brain has no lymphatics as such, the perivascular spaces possibly playing the role of lymphatics. This point will be discussed later, however. The meninges possess no lymphatics, either.

The choroid plexuses are supplied by branches from the internal carotids. They are drained by the choridal veins which form the great vein of Galen; this vein has adequate collateral circulation.

The arachnoid is thought by many to have no veins. The veins in the subarachnoid space, which seem to be called arachnoid veins by many, probably possess no capillaries. The pia mater is highly vascular, but its vessels contain no capillaries either.

## IV. Sites of Formation of the Cerebrospinal Fluid

### Intraventricular Sources

Clinical observations, although not proving that the choroid plexus is the site of formation of the fluid,

nevertheless to me prove that the ventricles, and especially the lateral one, are intimately concerned in the formation of the cerebrospinal fluid. Magendie [97] described cases of hydrocephalus associated with obliteration of the orifice of Magendie (many now deny the existence of a normal opening in the region where Magendie's foramen is supposed to be) or with tumors compressing the fourth ventricle or the aqueduct of Sylvius. Hilton [93] records findings similar to those of Magendie. Mott [51] has reported cases of hydrocephalus due to slow growing non-malignant tumors of the third ventricle and to a chronic basilar meningitis. Zange[124] in cases of internal hydrocephalus found very little fluid in the subarachnoid spaces. This is very strong evidence that the ventricles play a part in the formation of the cerebrospinal fluid. Dandy and Blackfan[15] have reported two cases of hydrocephalus due to congenital malformations of the ventricular system. In one the foramina of Luschka and Monro were lacking and in the other there was agensis of the aqueduct of Sylvius.

The work of Dandy[13] has been especially illuminating in regard to the role the ventricles play in the

production of hydrocephalus. He conducted experimental work on dogs under ether anesthesia and induced artificial hydrocephalus by blockage at various points along the pathway of the cerebrospinal fluid. By placing a gelatine capsule in the aqueduct of Sylvius he was able to produce a hydrocephalus in both the third and lateral ventricles. From these experiments in which the aqueduct of Sylvius was blocked. Dandy reaches the following conclusions: 1. That the cerebrospinal fluid forms in the cerebral ventricles. 2. That absorption of the fluid in the ventricles is less than the production. 3. That the aqueduct of Sylvius is a necessary outlet from the third and both lateral ventricles and 4. That there are no collateral channels which assume the function of the iter when it is occluded. By placing a piece of fascia or peritoneum in the foramen of Monro of a dog a cicatrix forms which occludes the foramen. The foramen is reached through a transcortical incision well posteriorly and opening into the lateral ventricle. The foramen of Monro may be seen quite anteriorly at a point where the choroid plexus makes a sharp bend into the third ventricle. The foramen is slightly scarified before the fascia is placed. The cortical defect is sutured with fine silk. A uni-

lateral hydrocephalus develops on the occluded side. Curiously enough the cortical incision heals without fistula or hernia formation.

Spiller[63] has published a case of unilateral hydrocephalus with occlusion of one foramen of Monro. The occlusion of the foramen was due to a tuberculous process in which a cicatrix formed obliterating the foramen.

Further proof that the ventricles are the site of production of the cerebrospinal fluid lie in experiments or in observations of cases in which hydrocephalus resulted from occlusion of the vena magna Galeni. The vein of Galen drains the entire cerebrum including the blood supply to the ventricles. As regards clinical observations on an obstructed vein of Galen the cases are few and far between. Newman[53] reports a case and Browning[5] another. Experimentally, Dandy[13] has been able to prove the relationship of obstruction of the vein of Galen to hydrocephalus and hence the relationship of the ventricles to the formation of the fluid. He found that obstruction of the vein of Galen at any point but at its very origin did not produce hydrocephalus. Obstruction at the origin, however, did produce hydrocephalus. This is to be explained simply

on the basis of an adequate collateral circulation being present when the vein was not obstructed at its origin and an inadequate collateral circulation being present when obstruction was present at its origin. Hence in obstruction at its origin we can explain the hydrocephalus only by increased production of fluid by the ventricles since the vein of Galen drains them. This experiment in no way throws light upon what part of the ventricles is responsible for production of the fluid but does show the intimate connection the ventricles have with the formation of the fluid.

## Choroid Plexus

Actual formation of the fluid by the choroid plexus was observed by Cushing in a human case in 1914 and in animals by Schaltenbrand and Putnam(1927)[60, 108] and Howe (1928).[32] Schaltenbrand and Putnam also injected a dye intravenously and noticed the appearance of this dye in the choroid plexus. Dandy was able to prevent his artificially induced hydrocephalus by excision of the choroid plexus. Putnam in 1935[54] introduced the method of coagulation of the choroid plexus by electrical means for relief of hydrocephalus in infants. Although the choroid plexus lies on the roof of the third and fourth

ventricles as well as in the lateral ventricles it appears that the portion of it in the lateral ventricles is responsible for formation of most of the fluid. According to Merritt and Fremont-Smith[48] the first one to suggest that the choroid plexuses were the site of formation of the cerebrospinal fluid was Faivre, in 1853. However, Katzenelbogen[37] claims that as far back as 1664, Willis credited the choroid plexuses and the pineal gland with its elaboration.

Besides the observations noted above in regard to hydrocephalus and the site of formation of the cerebrospinal fluid the following examples may be sited as evidence for the theory that the choroid plexuses elaborate the cerebrospinal fluid. Claisse and Levi,[83] in 1899, noted upon post-mortem examination of a hydrocephalic child a marked hypertrophy of the choroid plexuses. In the same year, Haushalter and Thiry reported a case of hydrocephalus ascribed to congenital lues in which the plexuses were abnormally developed. Davis[17] described a case of non-obstructive hydrocephalus with enormously hypertrophied plexuses.

Against the theory that the choroid plexuses are the site of formation of the cerebrospinal fluid are

the findings of Coupin[84], who in his anatomopathological studies on fish found a striking discrepancy between the amount of cerebrospinal fluid and the size of the choroid plexuses in various fishes. That is, some species of fish with large, well-developed choroid plexuses have but little cerebrospinal fluid and others with poorly developed plexuses have considerable cerebrospinal fluid. Coupin thus came to the conclusion that the choroid plexuses had nothing to do with the formation of the fluid. Hassin[29] refers to observations recorded in the literature and to his own material, which shows that large amounts of cerebrospinal fluid as seen in hydrocephalus may co-exist with sclerosis or other pathological changes in the choroid plexuses. He also records a case of hydrocephalus in an infant in whom the anatomopathological examination revealed absence of choroid plexus in the lateral and third ventricles and but fibrous vestiges of a choroid plexus in the fourth ventricle.

Although the findings just sited argue against the fact that the choroid plexus are the site of formation of the fluid the observation of Cushing of seeing the choroid plexus form the fluid in a living human case seems irrefutable to me.

## Sites of Formation of the Cerebrospinal Fluid

Further evidence that the choroid plexuses are concerned in the formation of the cerebrospinal fluid is to be found in embryological studies on the development of the cerebrospinal system. Weed,[71] noted that the first extraventricular spread of spinal fluid begins with the tufting of the plexus. This finding is only suggestive that the choroid plexuses plays some part in the formation of the cerebrospinal fluid. Flexner[19] in Amphibian studies corroborated Weeds findings. Also, especially interesting to me were his experiments dealing with transplantation of the brain of these animals to other protions of the body of the embryo. He noted that portions of the transplanted brain containing choroid plexus developed sealed off vesicles, which, after about two weeks became greatly distended with fluid. Similar transplants of spinal cord, in which fluid formation without choroid plexus, would have to take place, were never dilated to the degree that those containing plexus tufts were.

## Ventricular Ependyma

The role which the ventricular ependyma plays in the life history of the cerebrospinal fluid has been and still is a debated problem. Three possibilities are

present. The ependyma may play no part in the cerebrospinal fluid cycle. It may help secrete the cerebrospinal fluid. It may help absorb the fluid. Possibly, a combination of the last two may be present.

Arguments in favor of the the secretory nature of the ependyma hinge largely on histological studies of the ependyma. Luschka,[96] although recognizing the ventricles as the main source of the cerebrospinal fluid also emphasized the importance of the ventricular ependyma as a contributor. He based this conclusion on the observation of metaplastic changes in the ependyma similar to those seen in the choroid plexuses. He noticed clear vesicles in the ependyma and in the cerebrospinal fluid likewise. Studnicka,[115] in a comprehensive histological study of the ventricular ependyma in various animals as well as in man found that very frequently the whole surface of the ependyma was covered with droplets and vesicles, which, he claimed, represented outpouring fluid. Hence he was led to the conclusion that the ependyma represented a secretory membrane. Kafka[94] injected uranin into the general circulation and detected it later in the ependyma. However, Goldmann[90] in similar studies in which dyes were injected into the general circulation for

the purpose of determining their distribution in the central nervous system found no dye in the ependyma. Stern[65,66] believes that the ependyma controls the passage of crystalloids into the cerebrospinal fluid. Fuchs,[88] like Studnicka,[115] has studied the ependyma in various vertebrates and, like the latter, has come to the conclusion that the ependyma is a secretory organ. Jacobi and Magnus[34] have observed directly the exudation of fluid from the ependyma.

Wislocki and Putnam[78] in studies on the areae postremae found that Prussian blue granules accumulated in the ependymal cells of this area after intravenous injection of potassium ferrocyanide and Ferric ammonium citrate. The tissue was fixed with hydrochloric acid. The areae postremae are two mounds of extremely vascular tissue protruding into the lumen of the fourth ventricle in the region of the calamus scriptorius. When the reagents were injected in the subarachnoid space under normal conditions of pressure no absorption of the dye appeared to have taken place by the ependymal cells of the areae postremae. Likewise, on intravenous injection of the reagents the dye accumulated nowhere except in the ependymal cells of the areae postremae. Thus these men

suggest that the ependymal cells make a slight contribution to the cerebrospinal fluid.

The fact that Weed[71] found the presence of cerebrospinal fluid in embryos before the choroid plexus developed seems to point to an ependymal source of the fluid.

From the above we see that fairly strong arguments in favor of secretion by the ependyma are advanced. Arguments against the theory that the ependyma is a secretory epithelium and hence in favor of the choroid plexus theory of formation may be discussed as follows:

Dandy and Blackfan[16] have shown that removal of the choroid plexuses of one ventricle while the ependyma remained intact, conbined with occlusion of the corresponding foramen of Monro, instead of producing hydrocephalus, caused a collapse of the ventricle. From this they conclude that "we have the only absolute proof that cerebrospinal fluid is formed from the choroid plexus. Simultaneously, it is proven that the ependymal lining of the ventricles is not concerned in the production of the cerebrospinal fluid." Likewise, Dandy has removed the choroid plexus from both lateral ventricles and obstructed the aqueduct of Sylvius. This resulted in the slow development of a hydrocephalus. The development

of hydrocephalus was much slower than when the aqueduct was occluded without preceding plexectomy. The results of these experiments may be explained by saying that the choroid plexus of the third ventricle produces fluid at a slightly faster rate than the absorption from the three ventricles can take place. From Dandy's experiments it would seem that the ependyma played little part in the formation or absorption of the cerebrospinal fluid. His experiments also point quite conclusively to the fact that the choroid plexuses are the main source of the cerebrospinal fluid - at least so says Dandy.

Before accepting Dandy's work without reservation let us turn to the critical review of it made by Flexner.[20] Flexner says that Dandy has no proof that the foramen of Monro was entirely blocked. Dandy apparently relied solely upon macroscopic examination of the plugged hole. Flexner feels that more rigid examination of the block should have been made either by demonstration of a complete block by use of dyes or by microscopic examination of the foramen. Also as has been mentioned, Dandy had to make a cortical incision to reach the foramen of Monro, and Flexner feels that perhaps fluid escaped out of this incision, accounting for the collapse of the ventricle. He

says that microscopical examination of the incision should have been made showing that the ventricular ependyma was intact at the sight of the wound. To me these objections which Flexner raises do not seem well grounded for this reason. Dandy performed a similar operation on a dog in which the choroid plexus was not removed. The same cortical incision had to be made in order to reach the foramen of Monro and plug it. In this case the hydrocephalus developed much quicker than when the choroid plexus was removed. Surely, there would be just as much opportunity for the fluid to escape from the cortical incision in one as in the other, and for the foramen to be blocked as thoroughly in one as in the other. Flexner again finds fault with Dandy for concluding that the fluid must come only from the choroid plexus of the third ventricle when most of the choroid plexus of the lateral ventricle is removed. It is this word "most" that Flexner finds fault with. Without a definite statement that all of the choroid plexus of the lateral ventricles was removed Flexner will not accept Dandy's conclusion. However, Flexner does not mean to say that Dandy's conclusions may not be correct. He merely means to indicate points which he feels were not fully

controlled at the time of the experiments and which, therefore, render the conclusions liable to error. He does admit that Dandy's results are generally accepted, however.

## Choroid Plexus an Organ of Absorption?

It has been claimed by certain men that the choroid plexus was an organ of absorption not of elaboration of the cerebrospinal fluid. Askanazy[1,80] based his conclusions on the finding of hemosiderion pigment in the epithelial cells of the choroid plexus in cases of intraventricular hemorrhage. Hassin, Isaacs, and Cottle[30] in post-mortem examination of a case of pons hemorrhage discovered similar findings. Wullenweber in 1924[123] corroborated these findings stating that they constitute definite evidence of absorption on the part of the choroid plexus. Klestadt[40] injected carmine, fat and glycogen into the ventricle and found all substances in the epithelial cells of the plexus. He claimed that this indicated absorption on the part of the plexus. Hassin, mentioned above, held the extreme view that the choroid plexuses played no part in the formation of the cerebrospinal fluid but were simply an organ of absorption. His

views were based on histopathological studies which Flexner claims were highly questionable. Moreover, Flexner states that the finding of particulato matter in the epithelium of the choroid plexus after this matter has been injected into the ventricles is no evidence for absorptive function of the cerebrospinal fluid but rather probably represents simply phagocytic action on the part of the cells concerned.

From the above we will agree I believe that under normal conditions the choroid plexus absorbs no cerebrospinal fluid. However, under non-physiological conditions this statement may not always hold as I shall point out by the following experiment. Forbes, Smith and Wolff[23] using potassium ferrocyanide and ferric ammonium citrate, injected the materials into the subarachnoid spaces of a cat. They used barbiturate derivatives for anesthesia. Accompanying the injection of the reagents they also injected hypertonic glucose intravenously. Concomitantly they recorded pressure readings on a manometer connected to the subarachnoid spaces. Pressure of course fell after injection of the hypertonic solution. In a short time the aorta of the cat was opened and formaldehyde solution in H CL was

injected directly into the aorta. In some cases, instead of the injecting the formaldehyde into the aorta it was poured directly on the brain. This was done to avoid the possibility of washing out the Prussian blue granules from the choroid plexus by addition of fluid to the blood stream. The cat was then killed and placed in formaldehyde solution for twenty four hours. Blocks of tissue in paraffin were then sectioned and stained with Bismarck brown. Histological examination of the choroid plexus then showed Prussian blue granules not only in the epithelium of the choroid plexus but also in the lumen of the choroid vessels. Since it was possible that the material might have been absorbed by the lymphatus and then carried by the blood stream to the choroid plexus control sections were alsorun on heart muscle, intercostal muscle, kidney, liver and spleen. No granules were found in the capillaries of these structures. It must be remembered that this experiment was conducted with hypertonic glucose solution and not under physiological condition. Weed[70,73] in 1914 and 1923 using a method similar to the one described had already shown that no Prussian blue granules accumulated in the choroid plexus under normal osmotic

pressure relationships. Foley[22] in 1923 using methods similar to those of Forbes, Smith and Wolff but without injecting hypertonic glucose found Prussian blue granules in the choroid plexus after subarachnoid injection. At first glance these results might seem contradictory to those of Weed but when we observe the strength of solutions used we find that Weed used a one per cent solution of ferrocyanide and citrate whereas Foley used a four per cent solution of the same substances. The one per cent solution is approximately isotonic with the blood. Thus, on the basis of the hypertonic solution used by Foley can we account for granules in the choroid plexus. The fact that osmotic relationships of the fluids concerned influenced the direction of flow through the choroid plexus is quite strong evidence that the plexus acts as purely a semi-permeable membrane rather than as an actual secretory organ. This will be taken up in greater detail in following paragraphs, however.

## Summary of Intraventricular Sources of Formation

From the evidence reported above I believe that we may reasonably reach the conclusion that the choroid

plexus is the main source of the cerebrospinal fluid but that it has not definitely been proved to be the sole intraventricular source, the ependyma possibly adding small amounts to the fluid. Under normal osmotic and hydrostatic pressure the direction of flow through the choroid plexus is from the blood to the cerebrospinal fluid but under abnormal conditions of osmotic and hydrostatic pressure the direction of flow may to some extent be reversed. An important clinical application of these conclusions is the procedure of coagulation of the choroid plexus for relief of hydrocephalus and the injection of hypertonic solutions to relieve brain edema and acutely raised intracranial pressure.

## V. Mechanism of Formation by the Choroid Plexus

We come now to a consideration of the mechanism of formation of the cerebrospinal fluid by the choroid plexuses. Is it a secretion? Or is the choroid plexus purely a semipermeable membrane and the cerebrospinal fluid simply a dialysate? We must admit that this problem is as yet unsolved. It is a fundamental problem of biological chemistry and physics and may be likened

unto the discussion which raged for many years as to whether the glomerulus of the kidney was simply a dialysing membrane or an actively secreting structure.

In a consideration of this problem, as well as in all problems relating to the cerebrospinal fluid, there is a need for the closest correlation between anatomy and physiology. The cerebrospinal fluid is the result of a delicately balanced mechanism and may vary in pressure, composition and amount from hour to hour. Coincidently with its functional changes go changes in the anatomy of the choroid plexuses and ependyma. For this reason experiments conducted on the cerebrospinal fluid although perhaps giving similar results have been interpreted differently by different investigators because the conditions under which the experiments were conducted varied. That is, the physiology of the cerebrospinal fluid was not carefully controlled by various investigators.

## Arguments in favor of the Secretory Theory and Answers to these Arguments

A discussion of the two theories mentioned above resolves itself largely into statements and conclusions made by various investigators with criticisms of the

same by others. Riser and Meriel[101] produced hyperglycorrhachia by the intravenous injection of hypertonic dextrose solutions into men. They found that in some cases the ratio between the cerebrospinal fluid sugar and the blood sugar was not the same. Yet the experiments were all conducted under similar conditions. From these results they claim that the laws of physics can not be used to explain the discrepancies in the various ratios. Also, those favoring the dialysis theory must imply the existence of a Donnan theory of equilibrium, Walter.[118] It is well known that certain substances will not pass into the cerebrospinal fluid from the blood stream. Hence the term Hemato-encephalic barrier ("Barriere-Hemato-encephalique" suggested by Stern[65] in 1923) has been applied to this prevention of the passage of certain materials into the cerebrospinal fluid from the blood stream. Yet when these same substances are placed in the subarachnoid spaces they are readily absorbed into the blood stream. These results can surely not be explained simply on the basis of the Donnan theory of equilibrium. For example, iodide is denied entrance to the cerebrospinal fluid according to Stern[112] whereas when placed in the subarachnoid spaces it is readily

absorbed. This alone confuses the dialysate theory but as if this were not enough the results obtained by individual workers varies. Wittgenstein and Krebs[122] report findings almost the exact opposite of Stern. They claim that all anions pass readily from the blood into the cerebrospinal fluid. Cations, however, do not pass into the cerebrospinal fluid according to them. They account for this by saying that the positively charged ions are bound in the blood by the negatively charged proteins. The amphoteric proteins as we know are negatively charged in blood where pH is slightly on the alkaline side. If their results are true then the experiments of Miss Stern would be invalidated. Wesselkin,[121] to still further complicate the problem has gotten results contradicting Wittgenstein and Krebs. He found that certain acid dyes failed to pass the barrier whereas basic dyes stained the central nervous system.

Walter[119] in a series of rather extensive experiments found that Donnan's theory of equilibrium could not be used to explain completely the blood-spinal fluid relationship. Using bromide he measured blood and spinal fluid levels. He found that only about one third of the bromide was present in the cerebrospinal fluid, the rest

being in the blood. The one third refers to concentration of course. It may be assumed at first thought that this one third in the spinal fluid represents only the diffusible portion of the bromide, the rest being bound in the blood. This assumption Walter proved incorrect by the following means, however. Blood serum containing bromide was placed in contact with a 7.2% sodium chloride solution for twenty four hours. Only a dialysing membrane separated them. The Na Cl solution was changed three times. At the end of the twenty four hours the reaction of the blood was negative for bromide. The inference from this experiment is that the entire amount of bromide in the blood was diffusible. Not content with this one experiment he repeated the experiment, this time dialysing for only ten hours and not changing the salt solution. Similar results obtained. This experiment showed conclusively that the bromide in the blood was diffusible. Walter cites as further evidence against the blanket application of the Donnan theory to blood-spinal fluid relation the fact that dextrose is of a different concentration in the blood from its concentration in the spinal fluid. Also, in cases of tumor, an increase in the spinal fluid protein may occur with no

change in the electrolytes of the blood. Walter concludes that if the physico-chemical processes do take part in the formation of the cerebrospinal fluid they do not tell the complete story. Like Hober he concludes that vital activity on the part of the cells must be assumed to completely satisfy the experimental facts. The term "physiological permeability" has been suggested as covering both the physicochemical reactions and the vital cellular processes.

As is known a small amount of protein is present in the cerebrospinal fluid. This presence of protein has been of no small concern to those favoring the dialysate theory. Normally, we know that proteinsdo not pass through a dialysing membrane, the dialysate remaining free from protein. Thus if the proponents of the dialysate theory are to validate it, they must give some explanation for its presence.

Mestrezat, who was the first to propound the theory of dialysis explained the presence of the protein by saying that there was some looseness on the part of the cells of the choroid plexus, the protein passing through the intercellular spaces. Lange says that if Mezetrat's explanation were true then ferments and

agglutinins would be found in the cerebrospinal fluid also. Lange says that none are present. Lange might be answered by saying that just because up to the present we have not found other colloids in the cerebrospinal fluid does not mean that they might not be there. Indeed Kafka[35,36] has claimed in fact to have found ferements in the cerebrospinal fluid. Lange offers another suggestion for the presence of protein in the spinal fluid. As we know, a very few cells may normally be found in the cerebrospinal fluid. They have the appearance of blood lymphocytes but according to Lange are not from the blood at all but are from the desquamation of the endothelium lining the cerebrospinal fluid spaces. It is from the cytolysis of these cells that the cerebrospinal fluid protein originates, he claims, and that none comes from the blood stream. The reason he says that the cells must be from the endothelium of the cerebrospinal fluid spaces and not lymphocytes from the blood is because if they are lymphocytes that have gotten there by diapedesis then why are granular leucocytes, which have motility not present in the fluid also. This explanation of the origin of the cells seems rational

to me but I do not favor his interpretation as to the origin of the protein. It was stated above that Lange denies the presence of ferments in the cerebrospinal fluid. Yet he explains the presence of protein on the basis of cytolysis of the cells in the spinal fluid. To me cytolysis of this nature implies the presence of digestive engymes. Walter[119] also finds fault with Lange's explanation. In fact, according to Walter it defeats his own theory of dialysis. If, as Lange assumes, no protein whatsoever gets into the spinal fluid directly from the blood, how does any of it get back out into the blood stream after being formed by the cytolysis of cells in the cerebrospinal fluid? If Lange claims that the Donnan theory holds in the formation of the fluid he must also assume that it holds in the absorption. If it held in the absorption of the fluid, and Lange's hypothesis on the origin of the protein in the cerebrospinal fluid was true, then we would soon have a terrifying accumulation of protein in the cerebrospinal fluid, Walter asserts.

Further arguments against the theory that the cerebrospinal fluid is a dialysate hinge on histological studies of the choroidal cells. Meek[46] in 1907

injected pilocarpine and muscarine intravenously into animals and found that the choroidal cells became larger and that the cells differentiated into an outer clear zone and an inner granular zone. However, this is the exact opposite of the findings in cells of other glands; they become smaller and the differentiation is reversed, during secretion. Grynfeltt and Euziere (1919) studied the comparative activity in the choroidal cells in animals killed by hanging and in those killed by bleeding to death. In the histological examination of those killed by bleeding to death they found the picture of active secretion. In those killed by hanging they found no such activity on the part of the cells. They interpreted the state found in the animals killed by bleeding to represent an attempt on the part of the choroid plexus to maintain normal intracranial pressure by excessive secretion of the fluid.

I have already mentioned previously under a discussion of the origin of the cerebrospinal fluid the presence of vacuoles, fat droplets and granules in the ependyma a choroid plexuses. These observations have been made by numerous investigators. The opponents of the secretion theory describe these histological

secretory features in the epithelium of the plexuses as artificial, post-mortem formations and as products of resorption.

The increased secretion of fluid after injection of substances like pilocarpine, muscarine, adrenalin, etc. has been offered as evidence for the secretory theory also. This argument is answered by the dialysists by saying that any increased secretion of the cerebrospinal fluid after the administration of drugs is not due to any specific action of the drug on the plexus itself but is due to an indirect effect that the drug has by virtue of it's ability to change the hemodynamics in the skull. Such are the arguments for the secretory theory. It will be seen that most of them are answered by the dialysists in a fairly, yet not too, convincing fashion.

Before going on and discussing the dialysate theory I want to mention a theory held by but a few and which I believe has been conclusively refuted in the preceding discussion on the origin of the cerebrospinal fluid. Its main proponent is Fleischmann.[86] He believes that the choroid plexus seems to form the fluid first, as a pure filter. Then it absorbs the

substances from the cerebrospinal fluid which would prove toxic to the nervous system. This theory I think we will agree is a bit far fetched. The plexus becomes both the site of origin of the fluid and the site of its' absorption.

## Arguments in favor of the Dialysate Theory and Answers to these Arguments

The theory of dialysis was first propounded by Mestrezat.[49] By dialysing horse plasma through collodion he obtained a fluid of a compositon characteristic of the cerebrospinal fluid. Also by introducing dialysing sacs into the peritoneal cavity of animals he obtained a colorless, limpid, non-albuminous fluid, similar to the cerebrospinal fluid in composition. Mestrezat claimed that it was exactly similar but we know now that it was not exactly similar to cerebrospinal fluid because the latter fluid contains protein and Mestrezat's did not. However, Mestrezat's experiments did contribute the following finding in support of the dialysate theory. He found that the content of chlorides in his dialysate was higher than that in his serum in the collodion bag, a finding corresponding with relationship of the cerebrospinal fluid chloride and blood plasma. This he accounted for on the basis of the osmotic

pressure of the proteins in the horse serum, the chloride having to be in greater concentration in the dialysate than in the collodion bag to preserve equilibrium. Dailey[12] has confirmed Mestrezat's experiments. Nevertheless, although the chlorides in dialysates agree qualitatively with the chlorides of the cerebrospinal fluid, they do not agree quantitatively(Merritt-Fremont-Smith,[48] 1938).

In other experiments Mestrezat sought to eliminate the reasoning by analogy which naturely must be present in his experiments described above. He reasoned that if the blood and spinal fluid were in dialytic equilibrium then the composition of the two fluids should not change when subject to dialysis "in vitro". Hence, he placed a collodion sac containing 50 ce of cerebrospinal fluid in 500 ce of blood plasma and allowed them to dialyse for seventy three hours. After this time he found the sugar, the chlorides, the protein, the ash and the alkalinity to have remained as they were "in vivo", ie. within experimental error. Not satisfied with this experiment he further demonstrated that heating of the collodion membrane so changed its character that the cerebrospinal fluid

inside it became more and more similar in composition to the blood plasma outside of it. He reasoned that this experiment proved the identification of the anatomical nature of the anatomical sub-stratum (the choroid plexus and any other barriers between blood and cerebrospinal fluid) with the composition of the cerebrospinal fluid. That is, a histological change in the choroid plexus would account for a change in the composition of the cerebrospinal fluid.

I have mentioned under the arguments against the dialysate theory that with certain substances in the blood and spinal fluid the Donnan theory does not appear to hold - at least according to the investigators mentioned. However, on the other side of the fence we have those which favor the application of the Donnan theory in most cases. Mestrezat, Fremont-Smith and Dailey[24] have observed that under pathological conditions in the cerebrospinal fluid such as in meningitis, the chloride content of the cerebrospinal fluid goes down. I have already mentioned that it is normally higher than the blood. Both the findings in normal cases and the pathological findings point to at least a qualitative agreement with Donnan's theory

of equilibrium. McClenden[98] (quoted by Katzenelbogen) found that the concentration of sugar, urea, and diffusible salts in the cerebrospinal fluid was the same as in the ultrafiltrate of blood plasma. In other words, he confirmed Mestrezats observations. He also pointed out that the alkali reserve of the cerebrospinal fluid varied with the alkali reserve of the blood. If the cerebrospinal fluid was an active secretion how can we account for this he argued? The gastric juice and other glandular secretions do not depend, at least so minutely, upon the blood for their alkali reserve or even for their pH. Marrack and Thacker[44] have found that the calcium of the cerebrospinal fluid is slightly higher than the diffusible (ionized) portion of it in the blood. This would be qualitatively in accordance with Donnan's theory. An interesting sidelight which I might mention here is that the calcium of the cerebrospinal fluid of epileptics was found to be normal by Hamilton[28] in 1925. Thus it was proved the low ionized calcium in the cerebrospinal fluid could not be the cause of epilepsy. All the calcium in the cerebrospinal fluid since there is but little protein in the cerebrospinal fluid, is in the ionized form of course.

## Mechanism of Formation by the Choroid Plexus

Katzenelbogen has obtained results similar to Marrack and Thacker. Out of seventeen patients with normal cerebrospinal fluid the concentration of the spinal fluid Ca was equal to that of the diffusible blood calcium in fourteen (82.3%) slightly higher in one (5.9%) and slightly lower in two (11.8%).

I have already mentioned the work of Wittgenstein and Krebs as contradicting Miss Sterns results regarding the passage of iodide through the barrier. Their work may be summed up here as follows. They find that all anions, inorganic as well as organic (excluding colloids) pass from the blood into the cerebrospinal fluid. Under similar conditions, cations inorganic and organic, except the endogenous inorganic blood cations do not pass from blood into cerebrospinal fluid. The permeability of the barrier for colloids such as Congo red and Trypan blue is practically "nil" under physiologic conditions. From these results they conclude that the passage of substances into the cerebrospinal fluid from the blood depends solely upon their charge and upon the size of their molecules. The reason that cations do not pass into the cerebrospinal fluid (except the endogenous blood cations of course) is because they are bound with

the blood proteins. The storage of colloidal dyes by the plexuses, they say, can be explained on the physical principles just outlined. No postulation of vital action by the cells themselves need be brought into the picture.

From the above, we see that the theory that the cerebrospinal fluid represents a dialysate determined by physical laws must lay its foundations for correctness in experiments designed to show that its composition resembles the composition of a dialysate artificially produced. As I see it the reason that no definite conclusion has been reached as to whether the cerebrospinal fluid is a dialysate or not is because different workers carrying out similar experiments obtained contradictory results. For example, Mestrezat claimed to have gotten a filtrate by artificial dialysis of blood plasma which resembled in every way the cerebrospinal fluid. But let us now turn to the work of Stary, Kral and Winternitz whose observations are so splendidly summarized by Katzenelbogen. These men found that the ultrafiltrate of blood serum differed from the cerebrospinal fluid in the following ways. Inorganic phosphorus in the cerebrospinal fluid is less

than half of its concentration in the blood serum whereas in the ultrafiltrate the inorganic phosphorus is equal to that in the blood serum. Thus there is apparently no chemical combination of the phosphorus with the blood proteins. Why is it not found in greater quantity in the cerebrospinal fluid? The spinal fluid potassium is only 53% of the blood potassium whereas the ultrafiltrate of the blood serum contains over 100% of the blood potassium. The spinal fluid magnesium is about 137% of the serum magnesium yet the ultrafiltrate contains approximately 70% of the serum content.

After the cerebrospinal fluid and blood had been separated from each other by a semi-permeable membrane for a time, the following changes had taken place. The difference between the calcium content in the cerebrospinal fluid and in the blood serum was smaller after than before dialysis. Potassium and phosphorus were higher in the serum than in the cerebrospinal fluid before, and after dialysis were practically equal in both fluids. The magnesium of the cerebrospinal fluid which in cerebrospinal fluid within the body is slightly higher than in the serum was lowered by dialysis. If the cerebrospinal fluid was a true dialysate these

changes would not have taken place the authors argue. Their findings are in direct contradiction to those of Mestrezat's.

## Summary of Mechanism of Formation

From the above, we see that a state of confusion exists as to the mode of formation of the cerebrospinal fluid. To me certain findings absolutely rule out the possibility of pure dialysis. On the other hand I believe that osmotic forces and hydrostatic pressures play the major role in the formation of the cerebrospinal fluid. This idea finds clinical application in the use of hypertonic solution to decrease the volume of cerebrospinal fluid. Thus, the term "physiological permeability" is best applied to the membranes separating the blood and cerebrospinal fluid. This implies that the permeability varies with the physiology of the cells concerned and that metabolic products of cells may also be added to the cerebrospinal fluid.

The two opposing theories are very concisely and expertly summed up by Meritt and Fremont-Smith[48] as follows:

"Observations in favor of the dialysate theory are:

## Mechanism of Formation by the Choroid Plexus

1. The cerebrospinal fluid is isotonic with the blood plasma and tends to remain in osmotic equilibrium with the blood when the composition of the latter is changed experimentally (Fremont-Smith et al., 1931) or by disease (Fremont-Smith, 1932).

2. Variations in the plasma level of glucose, urea, chloride, lactic acid and alcohol are followed by parellel changes in the cerebrospinal fluid.

3. The pressure and volume of the cerebrospinal fluid may be changed at will by varying the osmotic or hydrostatic pressure of the blood. (Weed and McKibben, 1919)

4. The direction of flow through the choroid plexus can be reversed by making the blood hypertonic. (Foley, 1923; Forbes, Fremont-Smith and Wolff, 1928)

5. The distribution of chloride and sodium ions (in water content of plasma and cerebrospinal fluid) is qualitatively in accordance with the Donnan theory of membrane equilibrium. (Pincus and Kramer, 1923; Dailey, 1931; Fremont-Smith et. al., 1931)

6. There is no known substance in the cerebrospinal fluid which is not a normal constituent of the blood plasma.

## Mechanism of Formation by the Choroid Plexus

7. The balance between the osmotic and hydrostatic pressures of the capillary blood in the choroid plexus and the venous blood in the dural sinuses will account for the known facts concerning the formation, reabsorption, and pressure of the cerebrospinal fluid both in normal and pathologic conditions. (Fremont-Smith, 1927)
8. The chief histologic changes in the cells of the choroid plexus accompanying increased formation of fluid are the exact opposite of those occurring in a gland cell during secretion.

Observations against the dialysate theory are:

1. The unequal distribution of calcium, potassium, phosphate, uric acid, creatinine, amio-acid, glucose, and magnesium between blood plasma and cerebrospinal fluid is not easily explained on the dialysate theory. (Cockrill, 1931).
2. The distribution ratio of sodium, chloride, and bicarbonate between blood plasma and cerebrospinal fluid does not satisfy the Donnan theory quantitatively.
3. The composition of the cerebrospinal fluid is not quantitatively identical with that of artificially produced filtrates or dialysates of plasma.
4. The dialsate theory does not account for the

histologic structure of the choroid plexus cells nor for the changes which take place in these cells during conditions which induce a more rapid formation of cerebrospinal fluid.

5. The distribution of injected bromide between blood plasma and cerebrospinal fluid is not readily explained by the dialysate theory.

6. According to Flexner (1934) the cerebrospinal fluid cannot be considered an ultrafiltrate because the pressure in the capillaries of the choroid plexus is too small to account for the free energy change which takes place in the formation of the fluid.

"We believe that the weight of evidence favors Mestrezat's theory of dialysis. The composition of the cerebrospinal fluid as a whole corresponds to what a dialysate should be, i.e., it is isotonic with the blood and varies in tonicity with variations in the blood. Moreover, the volume and pressure changes which occur in response to hydrostatic and osmotic variations in the blood leave no doubt as to the influence of osmotic and hydrostatic forces in the formation and absorption of the cerebrospinal fluid. The reversal of flow through the choroid plexus after

hypertonic intravenous injections could hardly be accounted for by secretion. The fact that variations in concentration of chloride, sodium, glucose, and urea in the plasma are followed by parallel changes in the level of these substances in the cerebrospinal fluid shows that diffusion between the two fluids is active. There can be no doubt that hydrostatic and osmotic forces play a dominant role in the formation, reabsorption, and composition of the cerebrospinal fluid nor that the fluid is in approximate hydrostatic and osmotic equilibrium with the blood plasma, constantly tending to maintain such equilibrium in spite of variations in the composition of the blood. It would not be justifiable, however, in the present state of our knowledge to assume that no other forces play a part in the elaboration of the fluid. If, in the course of their metabolism, the cells of the choroid plexus, the ependyma, or the subarachnoid space elaborate any such cells withdraw substances or utilize energy in the formation of the fluid or any portion thereof, the cerebrospinal fluid cannot be considered as a pure dialysate. We do not doubt that some of these cells exert an influence, for it is hard to imagine fluid

passing through a layer of living cells without being modified at least to some extent by their activity. But there is no clear-cut evidence for such modifications at present. We are inclined to believe that the chief function of the choroid plexus cells is to maintain the characteristic permeability of the choroid plexus as a dialyzing membrane. It should be remembered in this regard that, from a plasma of given composition, several dialysates of different composition may be obtained by the use of suitable membranes. Each of these dialysates will be in hydrostatic and osmotic equilibrium with the plasma, but their exact composition will depend upon the particular permeability of the membranes used."

## VI. Extraventricular Sources of the Cerebrospinal Fluid

Whether or not the perivascular spaces of the brain contribute any appreciable amount of material to the cerebrospinal fluid has long been and still is a debated problem.

The classical experiments of Spina[64] in 1899 seem to me to offer the best evidence in favor of an extraventricular source of the fluid. Spina used dogs in his experiments. The dog was treated with curare and subjected to artificial respiration. A trephine opening was made in the skull and the brain exposed. Adrenalin was then injected into the general blood stream. A rise in blood pressure of course followed. When the blood pressure reached 280 mm. Hg. Spina observed the formation of droplets upon the surface of the brain. This he considered to be conclusive evidence of formation of cerebrospinal fluid by the intracerebral capillaries or nerve tissue. It would also corroborate the view that the cerebrospinal fluid moves in a direction away from the perivascular spaces and towards the subarachnoid space. Riser and Sorel,[102] in conducting experiments similar to Spinas but differing in that the atloido-ociepital membrane was cut so as to allow the escape of cerebrospinal fluid before injection of

adrenalin found no accumulation of fluid on the brain surface. They thus concluded that the droplets seen on the surface of the brain in Spina's experiments were due to the increased volume of the brain caused by vessel dilatation, which compressed the cerebrospinal fluid reservous at the base, so that the cerebrospinal fluid was squeezed out on the surface. Hence, they claim that Spina's experiments prove nothing.

Mott[51] in his studies on cerebral anemia came to the conclusion that the cerebrospinal fluid flowed in a direction from the subarachnoid spaces towards the perivascular and pericapillary spaces. This view would imply the belief that the cerebrospinal fluid was important in nutrition of the nerve cells. Spina's experiments I think definitely rule out Motts views. Weed[74] irrigated the subarachnoid space for several hours with Prussian blue without finding any of the material in the perivascular spaces. This is also quite conclusive evidence against Mott's view.

From the above, I think that the weight of evidence favors the theory that the perivascular spaces do play a part in the formation of the cerebrospinal fluid. Logical reasoning would also lead one to a

similar conclusion. The brain, as we know, has no lymphatics. Nerve cells as other cells must throw off products of metabolism which must in some manner be carried away. Is it not reasonable that some of their metabolic products be poured into the perineuronal spaces and eventually end up in the subarachnoid spaces. The perineuronal spaces connect with the pericapillary spaces which in turn empty into larger and larger spaces, which are called perivascular spaces. These latter then empty into the subarachnoid space. Of course it is possible that all the metabolic products of nerve cells are carried off directly by the venules and hence empty directly into the blood stream.

Granting that the perivascular spaces do make a contribution to the cerebrospinal fluid to what degree to they do so? In other words is it possible to measure the amount of fluid flowing from the perivascular spaces into the subarachnoid space. Weed (1922) said: "The perivascular spaces pour a certain amount of fluid into the subarachnoid space, where this fluid mixes with the liquid produced in the cerebral ventricles." Schaltenbrand[59] concluded that there was no measurable amount of flow into the subarachnoid space from the

perivascular spaces. His view is based on the following experiments. Fluorescine was injected intravenously and then the surface of the brain observed with a microscope to see if any of the dye came out from the perivascular spaces. Apparently, none did. Secondly, they occluded experimentally the subarachnoid space. Were there any appreciable flow from the perivascular spaces normally, then a considerable dilatation of them would expect to be found. No such dilatation was found. On these two pieces of evidence they base their view. However, there is no absolute proof that the subarachnoid space was completely occluded. Also could not the blocking of the subarachnoid space have been compensated for by an increased absorption on the part of the venules? Kubie and Schultz,[42] in studying the cells of the cerebrospinal fluid and the meninges in the cat observed that after intravenous injection of hypotonic solutions, leucocytes lying in the perivascular spaces appeared to be washed out into the subarachnoid space. Probably they simply accentuated the rapidity of flow which normally takes place. This would be in opposition to the findings of Schaltenbrand. As is well known the protein in the intraventricular

spinal fluid is less in amount than that in the subarachnoid space whereas the sugar level in the two locations is the exact opposite. This difference has been said to be due to the admixture of a fluid from the perivascular spaces relatively rich in protein and poor in sugar with the fluid coming from the ventricles. This would imply an appreciable flow from the perivascular spaces. Of course, the diminished sugar level in the subarachnoid space could be accounted for by supposing that the cells of the arachnoid membrane and pia mater use sugar in their metabolism. The raised protein level in the subarachnoid space could be accounted for by the absorption of water.

Some have claimed (Schaltenbrand and Bailey,1928)[107] that the vessels of the subarachnoid space also contribute to the cerebrospinal fluid by the process of filtration. However, so far as anatomical studies show there are no arterioles, venules or capillaries in the subarachnoid space and Rous, Gilding and Smith[56] in studies on vascular permeability in skeletal muscle failed to find any vessels larger than arterioles or venules that were permeable. Other information on the vascular system in general confirms their findings so

it is unlikely that the subarachnoid vessels contribute to any degree at all to the cerebrospinal fluid.

### Summary of Extraventricular Sources of Cerebrospinal Fluid

From the brief discussion on extraventricular sources of cerebrospinal fluid I think that it will be agreed that the perivascular spaces probably make a small but rather insignificant contribution to the cerebrospinal fluid. In their contribution are probably included some of the products of nervous metabolism.

## VII. Hematoencephalic Barrier

The hematoencephalic barrier as previously mentioned consists probably of histiocytes, glia, ependyma and the epithelium of the choroid plexuses. It has been claimed to have remarkable powers of preventing the passage of injurious substances from the blood stream into the central nervous system. For example, Tetanus toxin does not enter the cerebrospinal fluid. If we postulate the presence of a hematoencephalic barrier then we must assume that substances must first pass through the cerebrospinal fluid to reach the central nervous system. If we accept this assumption it must follow it seems to me that the cerebrospinal fluid must

be an important organ of nutrition for the nervous system. This latter point is certainly not accepted by all men. Many more accept the theory of the presence of a hematoencephalic barrier. I think the discrepancy lies in the definition of the term cerebrospinal fluid. If it includes the fluid which exudes from the capillaries deep in the central nervous system, then the cerebrospinal fluid would certainly be the source of nutrition to the nerve cells. If it includes only the fluid returning from the pericapillary and perivascular spaces then it would certainly not be instrumental in nervous nutrition. This represents my opinion only.

Probably the most important part of the hematoencephatic barrier is the choroid plexus. Next comes the ependyma. Of considerable importance also, however, are the histiocytes and glial cells lining the pericapillary and perineuronal spaces. The permeability of the barrier to various substances varies in different parts of barrier. For example, the occurrence of meningitis may be explained by saying that the choroid plexuses or ependyma were permeable to the toxin or bacterium. In general paralysis, on the other hand, the choroid plexus and ependyma have maintained their defense front but the histiocytes, capillary endothelium and"membrana glia limitans"

deep in the central nervous system have allowed the spirochaete to pass.

Very little is known concerning the exact mode of function of the hematoencephalic barrier. Nothing is known regarding its regulation. The study of it offers a luminous field, however, as perhaps in the future we may be able to control its permeability at will, decreasing its permeability when we want to protect the central nervous system against something and increasing it when we want certain substances to reach the nervous system for therapeutic purposes. I shall discuss this more in a later section.

## VIII. Sites of Absorption of Cerebrospinal Fluid

The location of absorption of the cerebrospinal fluid, strange as it may seem, is still a moot question. From the dawn of history there have been various theories as to the location of its absorption. Galen thought that it passed down the stalk of the pituitary and thence through the sphenoid bone into the nasal cavity. Later investigators thought that there was no absorption but that the cerebrospinal fluid moved back and forth in an ebb and flow manner. Magendie, in 1825, held this view.

## Sites of Absorption of Cerebrospinal Fluid

### Pacchionian Granulations

The beginning of modern concepts of absorption may be said to begin with the work of Quincke, Key and Retzius.

Quincke in 1872[55] reported his observations made following injections of powdered cinnabar (red mercuric sulfide) into the spinal subarachnoid space. This material is finely granular. At varying periods after injection he killed the animals and examined the tissues microscopically for the presence of the granules. The greatest accumulations of powder were found in the basilar portions of the subarachnoid space. However, on microscopic examination the granules were found to be distributed throughout the subarachnoid space and many were found in the Pacchionian granulations. With injections made into the spinal cord subarachnoid space the granules could be traced a short distance out into the perineural spaces. However, in practically all cases the granules were lying within lymphocytes or other larger phagocytic cells. Some lay in the meshes of fine fibrin. Thus in effect his powder had stimulated a mild aseptic meningitis and as far as I can see contributed nothing to the problem of absorption of

the cerebrospinal fluid. Nevertheless, Quincke concluded that the pathway of absorption to some extent was through the Pacchionian granulations. Quincke also found the cinnabar in the cervical and sub-maxillary lymph nodes but here again the material was within the lymphocytes. To me this proves nothing about the absorption of the cerebrospinal fluid through lymphatic channels but simply that the phagoceptic cells of the meninges end up in the cervical and sub-maxillary nodes following a meningitis.

It is interesting to note that Quincke found the cinnabar extending out along the olfactory nerve as far as the cribiform plate; along the optic nerve as far as the episcleral space of Schwalbe and clear to the perilymphatic spaces of the internal ear along the eighth nerve. Also, when he injected the subdural space he found that the granules passed from this space to the subarachnoid space. The reverse did not take place. He was thus led to conclude that fluid could pass from the subdural space to the subarachnoid space but not in a reverse direction. So far as I know no confirmation or contradiction of this has yet been offered.

## Sites of Absorption of Cerebrospinal Fluid

In 1876 Key and Retzius[39] published an extensive monograph on the anatomy of the meninges and nervous system in which they gave a very extensive discription of the cerebral envelopes that up to that time were really very poorly understood. Using gelatin solutions colored with Berlin blue and injecting them into the spinal subarachnoid space they were able to demonstrate the continuity of the spinal with the cerebral subarachnoid space. More important though, their injection mass passed through the Pacchionian granulations into the venous sinus. Studies of their preparations led them to believe that the mass passed, not directly through the arachnoid membrane into the venous sinus, but first into the subdural space around the arachnoid membrane and thence through the stomata between the endothelial cells. In addition to this passage through the Pacchionian granulations they also noticed the passage of the gelatin out along the cerebral nerves and into the lymphatic vessels accompanying these nerves. Key and Ketzius were aware of the fact that the pressure under which they injected the solutions could have ruptured the cells in the Pacchionian granulations and forced the dyed gelatin into abnormal places. However, all things

considered they accepted the Pacchionian granulations as the source of exit of the cerebrospinal fluid under normal pressures also. Following the publication of Key and Retzius' work the Pacchionian granulations were for a time considered to be the chief source of escape of the fluid. However, with more extensive anatomical studies it was observed that the Pacchionian granulations were variable structures. They did not seem to be present in infants, nor were they present in the lower animals and in the anthropoids. Hence the opinion swung back to the older view that they were pathological lesions. Calmeil (1826)[81] and Rokitansky (1844).[104] With this change of events new sites were sought for the absorption.

Reiner and Schnitzler[100] furnished further proof that the Pacchionian granulations were not the pathway of absorption of the cerebrospinal fluid. They injected solutions of potassium ferrocyanide into the subarachnoid spaces of various animals and then tapped the jugular vein at varying intervals following the injection, testing the blood for potassium ferrocyanide. Ferrocyanide was found to be present in the animals they used, no Pacchionian granulations were present.

## Sites of Absorption of Cerebrospinal Fluid

Consequently, they argued that by analogy that the Pacchionian granulations in man served no function.

### Venous Absorption and Lymphatic Absorption

Hill[31] traced methylene blue solution "straight into the venous sinuses" after subarachnoid injection. He found that the dye colored the urine and could be found in the stomach long before the dye was noticed in the lymphatics of the neck. This seemed a fairly good argument that the venous system furnished the chief source of exit of the cerebrospinal fluid.

Spina[110] made important contributions also. Conducting his experiments first on dead animals recently killed he injected milk and dyestuffs into the subarachnoid spaces. For the most part his work was done under pressures of about 160 mm. He concluded that the dyestuffs and milk passed directly into the venous circulation and, under normal conditions, to a lesser extent into the lymphatic system. He used young animals containing no Pacchionian bodies or older ones in which the Pacchionian bodies were destroyed. The noteworthy point of his experiments is that as increasing pressures on injection were

made more and more of the dye became absorbed by the lymphatic system and less and less by the venous system. He later confirmed his results on living animals. These results led him to the conclusion that under normal conditions of pressure very little cerebrospinal fluid is absorbed by the lymphatic system; but as intracranial pressure rises more and more comes to be absorbed by the lymphatics and less by the veins. Spina explained this on the basis that the lymphatics by virtue of their direct connection with the cerebrospinal fluid spaces were forced to absorb more fluid under heightened intracranial tension whereas the veins, not being in direct communication with the cerebrospinal fluid were compressed by the raised intracranial pressure. Reasoning from Spina's experiments we then see that a rise in intracranial pressure instead of being a simple vicious circle, is really a twofold vicious circle, if that expression may be used. We are all cognizant of the fact that a rise in intracranial pressure causes a rise in blood pressure through action of the medullary centers by virtue of their threatened anoxemia. This rise we further know tends to lead to greater

formation of cerebrospinal fluid which leads to further increased intracranial tension. Here is our one vicious circle. The other one as brought out by Spina's experiments is that raised intracranial pressure causes diminished venous absorption which will again cause the intracranial pressure to rise. This circle, is compensated for to some extent by increased absorption on the part of the lymphatics. Hence the great value of hypertonic solutions in all cases of raised intracranial pressure.

Cushing,[10,11] whose illustrious name will find its way into any bibliography on the nervous system, through a series of ingenious experiments on living subjects with hydrocephalus came to the conclusion that the venous system must be the main source of absorption of the cerebrospinal fluid and that the Pacchionian bodies did not play an important part in that absorption. It was in the late 90's and early 1900's that Cushing first began his work on hydrocephalus. It was his clinical observations on hydrocephalus that stimulated him to seek the location of exit of the cerebrospinal fluid. As he put it, "...too much attention had been paid to the bath itself and too little to the tap, to

the construction of the tub as a container of the bath, and to the place and mechanism of its outlet." Cushing had noticed numerous examples of hydrocephalus in infants that had come under his observation in the clinic, and it was found that in most of them the fluid could be withdrawn from them by lumbar puncture. (Quincke, in 1891, introduced the lumbar puncture as a routine measure). Moreover, insertion of needles connected with manometers into the ventricles and spinal subarachnoid spaces concomitantly showed in most cases equal pressure in the two manometers, said pressure remaining the same under all conditions such as laughing, coughing and straining. Also, he observed, as many others had, that even though large amounts of fluid (30 cc and over at times) might be withdrawn and the ventricles temporarily collapsed they would within 24 hours refill to their utmost with fluid practically the same in composition to that which they had withdrawn. From these simple clinical observations Cushing became convinced that there must be some defect in the development of the pathways of exit of the cerebrospinal fluid rather than a block, in most of these cases observed in infants. At the time that he was conducting his

experiments on hydrocephalus the prevalent opinion was either that the Pacchionian bodies were the absorbing bodies or that the lymphatics (extracranial and extraspinal along the sheaths of the nerves) carried the fluid off. Cushing, not being able to find Pacchionian bodies in infants dismissed them as a possible exit of the fluid. Acting on the theory that the lymphatics were the source of exit he attempted to cure hydrocephalus by draining the subarachnoid lumbar fluid directly into the retroperitoneal tissues through a trephine opening in the body of the 5th lumbar vertebra. The proximity to the thoracic duct was thought to be especially advantageous. Previously he had tried draining the fluid into the subaponeurotic space of the scalp; this had met with failure. His second site of drainage met with no more success than his first site.

Abandoning the idea that the lymphatics normally served as the source of exit of the cerebrospinal fluid, Cushing next decided that to cure hydrocephalus the fluid would have to be drained directly into the venous system. Previous attempts by others to transplant a vein between the subarachnoid space and a vein in the neck had failed so Cushing devised a silver tube which

he placed in such a manner that it led directly from the dilated 3rd ventricle to the superior longitudinal sinus. Only mediocre success followed this procedure, however. One important bit of knowledge was gained, nevertheless, and that was that no blood flowed from the vein back into the cerebrospinal fluid. This showed that the cerebrospinal fluid pressure was at least higher than the venous pressure according to Cushing.

Cushing noted one or two other points which also showed that the veins under normal conditions furnished the main outlet for the cerebrospinal fluid. He found that digital compression of the jugular veins caused a marked increase in the amount of fluid in the brain substance and in its membranes. This strongly indicated that the veins were a pathway of escape. Also mercury dispersed in the subarachnoid space was found to collect within the venous sinuses. Injection of the substance directly into the venous sinuses was not followed by the appearance of the material in the subarachnoid spaces. These findings led Cushing to believe that there was some valve like arrangement present between the blood and cerebrospinal fluid. Weed later demonstrated, that at least histologically,

there was no evidence of valvular arrangement.

Sicard and Cestan[109] obtained findings similar to Quincke as far as I can see. They found that injections in the subarachnoid space passed out along the spinal nerves as far as the posterior ganglia in the dorsal roots and to a shorter distance in the ventral roots. Dye (India ink) would pass from the subarachnoid space to the subdural space but not in a reverse direction. They did not venture to suggest a mechanism for absorption although their findings suggested a lymphatic route.

Cathelin[82] in 1912 claimed (on rather insufficient evidence according to Weed) that the chief pathway of absorption was through the perineural sheaths and thence into the lymphatics and thoracic duct.

Dandy[14] confirmed the findings of Hill, Reiner and Schnitzler and others that the venous absorption of dyes was much greater than the absorption along lymphatic channels. He concluded however, that the dye was absorbed as rapidly from the spinal subarachnoid space as from the cerebral subarachnoid. They based their conclusions on the observations that with the spinal fluid subarachnoid space separated from the

cranial space absorption took place just as rapidly from the spinal fluid space as from the cerebral space. In 1914 Dandy and Blackfan[18] expressed the opinion that absorption of the cerebrospinal fluid was a diffuse process, all of the vessels of the subarachnoid space participating in it. Howe,[39] in 1929, without adding any experimental evidence of his own concurred in this opinion. For their experiments Dandy and Blackfan used phenolsulfonphthalein and made quantitative estimations of the rapidity of absorption. Becht[2] criticizes this work of Dandy's on the basis that determination, quantitatively, of the absorption of dye from the subarachnoid spaces is no indication of the rate of absorption of the cerebrospinal fluid itself. Dandy and Blackfan had stated that the cerebrospinal fluid was replaced by new liquid every 4 - 6 hours. From Dandy's experiments, however, has evolved the Dandy phenolsulfonphthalein test for differentiation between internal and external hydrocephalus. The dye is injected into the spinal subarachnoid space and in internal hydrocephalus will not be found in the ventricles on ventricular puncture. In external hydrocephalus the dye will be found in the ventricles after spinal subarachnoid injection. As will

be shown later the circulation of the cerebrospinal fluid normally is in an opposite direction to which the dye must go in order to reach the lateral ventricles in internal hydrocephalus. Does the appearance of the dye in the lateral ventricles indicate a reversal of flow of the cerebrospinal fluid in hydrocephalus or simply a diffusion of the dye on a physical basis? This question has not been answered. The percentage of dye found in the urine is also used as an indication of whether the hydrocephalus is internal or external. Normally, 30-40% of the dye is absorbed from the combined cerebral and spinal subarachnoid spaces. In internal hydrocephalus this full amount will be absorbed of course; but in external hydrocephalus where the blockage is in the cerebral subarachnoid spaces only 10-20% will be absorbed (that absorbed by the spinal subarachnoid spaces.) Weed and Becht have severely criticized Dandy's conclusions on dye absorption, however, on the basis that a considerable degree of experimental error may be present. Also, Weed[57(4)] failed to find in his replacement experiments in the subarachnoid spaces any Prussian blue granules in the spinal subarachnoid vessels. Flexner,[20] however, claims that

## Sites of Absorption of Cerebrospinal Fluid

Weed's objections to Dandy's results are invalid because it may be possible that the spinal subarachnoid vessels are permeable to cerebrospinal fluid and not to Prussian blue reagents. As proof of his objection Flexner states that the choroid plexus and ventricular ependyma are impermeable to ferrocyanide although obviously permeable to cerebrospinal fluid. More will be mentioned along the line of Flexner's statement later.

In 1929 Dandy[14] approached the problem from a different angle, and from an exceedingly difficult one at that. He claims to have separated the pia and arachnoid from the superior longitudinal sinus, transverse sinus and circular sinus in young dogs. The pia and arachnoid remained intact he states. Following the dogs for a period of 4 - 6 months, he observed that they developed no hydrocephalus whatsoever. This according to him proves that there is no absorption into the venous sinuses but that it is entirely absorbed into the vessels of the subarachnoid spaces. However, Elman,[18] in 1923, has described arachnoid granulations as being present in the subarachnoid spaces and perhaps the fluid was absorbed by these structures, acting as a compensating mechanism when the normal mechanism of

absorption through the venous sinuses was destroyed. Such seems a valid criticism at least. Furthermore, to assume that the subarachnoid spaces remained disconnected with the venous sinuses for six months following the operative procedure is no small assumption. We are all aware of the remarkable regenerative power of these membranes. Microscopic examination, which Dandy did not do should be done to confirm the break in connection. Nevertheless, we have no definite proof against the correctness of his results and assumptions.

## Weed's Work

We come now to the experiments of Weed.[69] It is to him that we owe the most widely accepted conception of the normal mechanism of absorption of the cerebrospinal fluid. Since Weed[73] has contributed so much to the knowledge of absorption and has controlled his experiments probably better than any other man working on the same problem I believe that we may profitably consider his work in some detail.

In his earliest work Weed used injections of finely ground carbon particles. These were injected into the subarachnoid space of unesthetized dogs. The

injections were made at pressures as near normal as possible ie just enough higher than normal pressure to prevent leakage of the cerebrospinal fluid. The injections were made over long periods of time one injection requiring four to six hours. Following the injection the tissue was removed "en bloc", the meninges being kept intact. The block was then hardened in ten percent formol. Following his experiments with finely ground carbon Weed next used lamp black in which the carbon particles were even smaller. On microscopical examination the particles were seen to be caught in the subarachnoid spaces. None of the particles were found to be phagocytosed as Quincke had found because Weed had not allowed time for phagocytosis to occur. Weed did not find but a very few particles in the walls of the longitudinal sinus - too few to be of any significance he concluded. The lungs of the animals were studied for the presence of granules, the presence being taken as an indication that the particles had attained entrance into the venous system at some point. Here, however, he ran into an insurmountable obstacle. The dogs had anthracosis! Furthermore, Weed realized that the carbon particles were too coarse and that to

really demonstrate pathways of absorption he was going to have to find some substance of a finer nature. Thus he next used carmine in slightly acid solution so that the dye was precipitated. Findings similar to those with the carbon obtained, however. Following carmine he tried various true solutions such as silver nitrate and alkaline carmine with subsequent precipitation during fixation. Two objections were raised to all the substances he tried. One was that many of the materials were too toxic; the other was that those that weren't toxic were diffuse tissue stains and as such were of course of no benefit. At last he hit upon the use of potassium ferrocyanide and ferric ammonium citrate - the two reagents that I have mentioned in previous discussions and ones that are considerably used now in the study of the dynamics of the cerebrospinal fluid.

Using the same technique as he did with the carbon granules and being careful not to inject the reagents at too high a pressure he obtained the following findings.

No precipitated granules were found in the subdural space. This, as will be recalled, is in accordance with the findings of Quincke. When injected into the subdural

space the materials quickly passed into the subarachnoid space, however. Why this should be true is a mystery. There is as yet no explanation as to why this selective direction of flow is present. Weed considers that the subarachnoid spaces are the normal outlet for the fluid secreted by the epithelial cells lining the subdural spaces, however.

When injections were made into the spinal subarachnoid space over a brief interval of time the granules were found to accumulate in greatest quantity in the basilar regions of the space and especially in the arachnoid villi of the cavernous sinus. However, when the injection was made above the tentorium the granules accumulated in the arachnoidal villi of the superior longitudinal sinus. This thus indicates different pathways for the fluid from various parts of the system. It will be noticed that Weed mentions that the granules passed through the arachnoid villi. This was a view which up to this time had not been definitely stated. Many investigators, as I have previously mentioned, concluded that the drainage of the cerebrospinal fluid was into the venous system but Weed was the first one to definitely prove through which structures it went.

## Sites of Absorption of Cerebrospinal Fluid

Weed noticed that there was a precipitation of granules within the cytoplasm of the outside cells of the arachnoid villus. Whether or not a lining of endothelium derived from the blood sinus wall cap the villus he is not sure. It is his opinion, though, that such a layer is present. Besides finding the granules intracellularly they were also present extracellularly which would indicate that the passage through intercellular spaces as well as through the cytoplasm of the cells.

With the concept established that the cerebrospinal fluid passes through the arachnoidal villi one must next ask why it passes through them. It is probably due to the factors of osmosis, diffusion and filtration. Which of these processes play the greatest part it is hard to say. It is well known that the osmotic pressure of the blood is higher than that of the cerebrospinal fluid because of the former's higher protein content. Also, granting that the choroid plexuses elaborate a certain amount of the cerebrospinal fluid all the time there must be some degree of pressure built up, hydrostatic pressure we may call it, which would favor diffusion and filtration. Weed in 1914 stated that the pressure in the arachnoid villus

remained constantly at a higher pressure than the blood in the venous sinus. However, in 1935,[72] he raised this question. Dogs and other four footed animals have a horizontal posture. Thus their cerebrospinal fluid hydrostatic pressure in the region of the sagettal sinus would be expected to be considerably more then in man. Their venous pressure would also be higher. On assuming an erect posture man's cerebrospinal hydrostatic pressure in the region of the sagettal sinus must surely have dropped. Yet his venous pressure probably did not drop so much because of the fact that it is dependent to some degree upon the heart beat. According to the Monro-Kellie doctrine the cranium acts as a closed box and any increase in volume of one of its contained structures must be compensated for by a decrease in volume of the other contained structures. Using complicated formulae Weed calculated the coefficient of elasticity of the structures which must be put under tension when an animal assumes an erect posture and from these calculations concluded that the structures would allow quite a drop in hydrostatic pressure upon attaining erect posture. In fact such a large drop in pressure would be expected

to be present that the pressure in the venous sinus would far outweigh the pressure in the cerebrospinal fluid. From these theoretical considerations he now wonders why the cerebrospinal fluid is absorbed at all in the sagettal sinus. No answer has as yet been offered according to Weed but I believe Cushing's silver tube experiments already discussed should be repeated as perhaps they would throw some light on hydrostatic pressure differences.

When Weed injected solutions into the spinal subarachnoid spaces which had been isolated from the cranial spaces he found the absorption to be only about one third as rapid as when the entire system was intact. Thus he concluded that absorption from the cranial spaces played a much more important part than the subarachnoid spaces. Dandy and Blackfan[15] it will be recalled reached just the opposite conclusion on their experiments with phenolsulfonphthalein as they believed the spinal absorption to be as great if not greater than that from the cranial spaces.

Dandy and Blackfan also claimed that the process of absorption was a diffuse one throughout the whole arachnoid, the cerebrospinal fluid entering the capil-

laries of the pia mater. Now the vessels of the pia have no cappillaries at all as previously mentioned. The arachnoid is entirely avascular. Weed indeed found no passage of the granules through the walls of the veins of the pia and hence takes issue with Dandy.

So far we have considered only Weed's findings on absorption of the cerebrospinal fluid as related to the venous system. Let us now turn to his findings on the relation of the cerebrospinal fluid to the lymphatic system. He found that his injections of ferrocyanide and citrate solutions passed for varying distances out along the subarachnoid space which, as many anatomists had previously noted, and as I have mentioned, passed out along the nerves for a short distance before ending as a blind pouch. Especially interesting are his findings in regard to certain of the cranial nerves. Granules were found in great concentration along the olfactory nerve as far as the cribiform plate. Often the coloring, however, passed through the cribiform plate and was to be found within the tissue spaces and lymphatics of the nasal mucous membrane. If the pressure of the injection was high enough fluid was observed to drip from the nose - a cerebrospinal-rhinorrhea.

## Sites of Absorption of Cerebrospinal Fluid

This shows that in certain cases considerably raised intracranial pressure may cause a "fistula." Along the optic nerve the granules could be traced into the episcleral space (space of Schwalbe, space beneath Tenon's capsule and upon the sclera) as far as the filtration angle of the eye. This thus indicates a close relationship between the aqueous humor and the cerebrospinal fluid. Along the auditory nerve the fluid was traced into the perilymph of the saccule, utricle and cochlea again demonstrating continuity of the subarachnoid space with the specialized organs of sense. Along the vagus the Prussian blue could be found as far down as the middle of the neck.

Weed also found considerable staining of the lymphatics of the neck, these findings being in accordance with the views of others which I have previously mentioned. Weed thus confirmed the postuation that the lymphatics might serve as an accessory pathway of drainage for the cerebrospinal fluid. It must be clearly understood, however, that there are no lymphatics to be found in the meninges or brain (Sabin[57]). The absorption takes place into the lymphatics accompanying the nerves and hence is an extracraniospinal process.

## Sites of Absorption of Cerebrospinal Fluid

Weed believes this lymphatic drainage to be of greater importance in the spinal area than in the cranium because in the spinal region it represents the only avenue of escape of the fluid. (Weed believed there to be no upward current of spinal fluid.)

From Weed's extensive experiments was laid the modern, yet not uncriticized, concept of cerebrospinal fluid absorption. Through his work and the experiments of Dandy the different types of hydrocephalus have been recognized and rational, yet unfortunately often futile, therapy established.

Weed's experiments have been accepted by the majority of investigators. Halliburton[27], Cushing[10], Winkelman and Fay[77] have all adopted it. W. E. Clark[7] has confirmed Weed's statements on the distribution of the arachnoid villi.

## Modifications and Additions to Weed's Theory

Wislocki and Putnam[78] produced an artificial internal hydrocephalus in young rabbits and kittens by injections of lamp black into the cisterno-cerebello-medullaries. Subsequently, a readily diffusible dye was injected into the dilated ventricle. The animals were then killed in periods varying from eight to

twenty four hours. Grossly, on cutting the brain the dye was visible in the tissue surrounding the ventricle. The dye could be traced in the nervous tissue as far out as the cortex and as far inferiorly as the lumbar cord! Following these experiments with the dye they then used potassium ferrocyanide and ferric ammonium citrate injecting them into the ventricles of hydrocephalic animals. The distribution of the precipitated granules after the animals had been killed and the tissues fixed was similar to the distribution of the dye except that the granules could be seen within the cells of the ependyma but not within the cells of the choroid plexus. This observation is of considerable importance because it indicates that the choroid plexus is impermeable to the Prussian blue reagents and hence throws some doubt on the validity of Weed's conclusions. As I have mentioned previously this finding of the absence of Prussian blue in the choroid plexus forms the basis for Flexner's objection to Weed's statement that the meningeal vessels are not concerned in the absorption of the cerebrospinal fluid. Flexner rightly asks, "If the choroid plexuses are impermeable to the reagents, may not the meningeal veins also be impermeable

to the reagents?" Just because the reagents do not pass through the structures concerned is no proof that these structures might not absorb cerebrospinal fluid.

Dandy and Blackfan[15,16] found that when phenolsulfonphthalein was injected into the ventricles of human subjects with internal hydrocephalus the dye appeared in the urine.

Nanagas[52] in experiments similar to Wislocki found in normal kittens a very small amount of absorption from the ventricles - ie the dye was found surrounding the ventricles for only a short distance. Hypotonic solutions injected into the venous system decreased the absorption from the ventricles to almost nothing. Hypertonic solutions increased the absorption from the ventricles. He found no evidence of absorption by the choroid plexus. In hydrocephalic kittens he could trace injections of dye and Prussian blue reagents into the ventricles down through the ependyma into the underlying capillaries. In addition to these findings which are identical with those of Wislocki he also tested the influence of hypertonic solutions on the absorptive power of the ventricles in hydrocephalus. Hypertonic solutions, although causing at first a brief rise in

cerebrospinal fluid pressure (probably due to increased blood volume), in the main caused a marked drop in the cerebrospinal fluid pressure with increased absorption from the ventricles.

Scholz and Ralston[61] used a method similar to Weed's but instead of fixing the tissue in an aqueous solution, they immediately froze the tissue with liquid air and dehydrated it to avoid any post-mortem diffusion which might have taken place by Weed's method. Their findings were similar to Weed's except that they also found granules in the pia-arachnoid veins. Benninghoff states that there are arachnoid veins but that they contain no muscle. It will be remembered that under the discussion of Weed's views he claimed that the arachnoid was entirely avascular but admitted that the pia had vessels. Scholz and Ralston's views constitute a compromise view between those of Weed and Dandy.

## IX. Mechanism of Absorption

I do not propose to discuss this in detail as a complete discussion would be repitition of the theories proposed on the mechanism of formation of the fluid.

## Mechanism of Absorption

Cushing[10] thought that some valve like action of the villi was present permitting the flow of cerebrospinal fluid in but one direction, that direction being from the arachnoidal villi into the venous sinuses. After Weed's experiments,[69] however, in which Weed showed that there was no histological evidence for any valvular action and that the fluid actually could flow in the reverse direction under the influence of hypotonic solutions he retracted this view.

Weed believes that the factors of filtration, osmosis and diffusion all play a part in the absorption. These factors are dependent upon the difference in hydrostatic pressure between the cerebrospinal fluid and the venous system and upon the concentration of colloids and crystalloids in the cerebrospinal fluid and blood. I have already mentioned Weed's rather interesting theoretical consideration on the effect an erect posture should have on hydrostatic pressure.

The only way that we have been able to modify the rate of absorption up to the present time is by the injection of hypertonic and hypotonic solutions, the former increasing the rate of absorption, the

latter decreasing it.[75] The effects of these solutions last but a short time and hence to be effective clinically must sometimes be repeated. If we could but discover some substance that would increase absorption over a long period of time.

The essential difference between the theories on the absorption of the cerebrospinal fluid and on its formation is that nowhere have I found any article which stated that the absorption of the cerebrospinal fluid was on a secretory basis, whereas many have felt that its formation was on a secretory basis. Another peculiar finding is that very few investigators have concerned themselves with the mechanism of absorption yet those same investigators have spent considerable time in trying to prove their theories on the mechanism of formation. It seems to me that much could be learned on the mode of formation of the fluid if more were known concerning its mode of absorption. For example, those favoring as the mode of formation, the dialysate theory point to the fact that when cerebrospinal fluid is separated by only a semipermeable membrane from the blood plasma there is no change in volume of either nor any appreciable change in composition proving that

the cerebrospinal fluid is in equilibrium with the blood. Yet these same ones will accept the theory that the cerebrospinal fluid passes back into the venous sinuses through the arachnoid villi by a process of diffusion and osmosis! If they do not accept this theory they ignore their inconsistencies. I cannot see how the dialysists can so completely ignore the absorption of the fluid in a discussion of its formation. The only way that the dialysists can still stick to their theory it seems to me is to definitely prove that hydrostatic pressure alone can account for the absorption of the cerebrospinal fluid. At the present time there is no evidence available on the comparative differences in hydrostatic pressure in the venous sinuses and in the cerebrospinal fluid just adjacent to it.

## X. Summary of Sites of Absorption and Mechanism of Absorption of the Cerebrospinal Fluid

I believe that from the evidence presented it will be agreed that there are at least two distinct views on the location of the absorption of the cerebrospinal fluid. The one, whose main proponement is Dandy, holds that the absorption of the cerebrospinal

## Summary of Sites of Absorption and Mechanism of Absorption of the Cerebrospinal Fluid

fluid takes place in a diffuse manner throughout the cranial and spinal subarachnoid space. The veins of the pia and possibly the veins of the arachnoid (if there be any) along with the lymphatics draining the perineural sheaths form the essential avenues of escape. In hydrocephalus appreciable absorption may take place through the ventricular ependyma. The absorption through the spinal avenues is greater than that by the cranial avenues. The other theory, whose main proponement is Weed, states that the arachnoid villi are the structures through which the greater part of the cerebrospinal fluid drains into the venous sinuses. These villi are most numerous in the sagittal and circular sinuses and are absent in the spinal cord. Thus the absorption from the cranial portion of the subarachnoid cavity far exceeds the absorption from the spinal part. The perineural spaces draining into the lymphatics account for the escape of some of the fluid. Under conditions of increased intracranial pressure this latter pathway of absorption may assume greater importance. This theory denies the absorption of cerebrospinal fluid directly into the veins of the

pia-arachnoid. The Pacchionian granulations Weed considers to be merely pathological, hypertrophic arachnoid villi.

No one can say which theory is correct. If one could, then there would not be two theories. Further experimentation will have to be done to clarify the situation; the question is by no means settled. However, Weed's theory is the one generally accepted.

## XI. Circulation of Cerebrospinal Fluid

The circulation of the cerebrospinal fluid has really been indirectly covered in a study of its formation and absorption. Hence I shall not devote too much space to its consideration. This discussion will serve, more or less, to bring together isolated facts and evidence which have been mentioned in the previous sections on formation and absorption with the addition of important, relevant facts.

Magendie, in 1825,[97] was the first to hint that the cerebrospinal fluid underwent movement. He advocated the "ebb and flow" theory. In the work of Key and Retzius, in 1876, which has already been

mentioned the presence of a true circulation was strongly hinted. Cathelin,[82] in 1903 was the first to state that the cerebrospinal fluid circulated in a system of well defined pathways originating from the blood and returning to the place from whence it arose. From the time that Cathelin first advanced the idea that the cerebrospinal fluid was an actively circulating medium an argument has been raging pro and con. Most all observers do admit some movement of the fluid, however, but the rate of movement, the direction of the movement, and the variation in movement in the different parts of the subarachnoid space has been a matter of much controversy, as yet unsettled.

Weed[71] through his embryological studies on the pig could trace Prussian blue reagents in pig embryos from the ventricular cavities through the roof of the fourth ventricle into the perimedullary mesenchymal syncytium. This indicated that the fluid passed out of the roof of the fourth ventricle. Anatomists have long argued about the presence of a medial foramen of Magendie and lateral ones of Luschka in the region of the fourth ventricle. Apparently, the existence of the lateral foramina is fairly well established. The

presence of a medial one is in doubt. Meritt and Fremont-Smith deny the presence of the foramen of Magendie. The appearance of the fluid in the mesenchymal syncytium takes place about the fifth week of intrauterine life. As development continues the fluid may be seen to find its way through the leptomeningeal channels in the mesenchyme, to finally be absorbed through the arachnoid villi.

Dandy in his experiments on hydrocephalus, which I have fully dealt with proved quite conclusively that the fluid must escape from the foramen of Monro, pass down the aqueduct of Sylvius and out the foramina of Magendie and Luschka. Dandy[14] also found that phenolsulfonphthalein introduced into the ventricles quickly appeared in the subarachnoid spaces indicating an active circulatory process.

On the other hand Solomon, Thompson and Pfeiffer[62] using the same dye as Dandy found that injections into the ventricles was followed by very little change in location of the dye. Similar results were obtained when they injected the dye into the cisternal subarachnoid space or the lumbar subarachnoid space i.e., the dye did not very readily move from its site of injection.

## Circulation of Cerebrospinal Fluid

The concept of an active circulation was also combatted by Sachs, Wilkins and Sams.[58] Using trypan blue they injected it into the subarachnoid spaces at the levels of the third lumbar, first thoracic and parietal regions. The injections of the dye into these various levels was of course done on separate animals, only one location being used on any particular animal. Following the injection they exposed the dura without killing the animal and followed the course of the dye "in vivo." Along with these experiments "in Vivo" they also carried out experiments "in vitro" in which the dye was injected into a column of cerebrospinal fluid through a rubber diaphragm. The diffusion in "vitro" was then observed. Comparing the movement of the dye "in vivo" and in "vitro" they have come to the conclusion that the concept of a true circulation in the cerebrospinal fluid is incorrect but that substances introduced into the cerebrospinal fluid spread by diffusion only. Lowering of the cerebrospinal fluid pressure by lumbar puncture or by artificial drainage elsewhere, of course, they admit establishes an artificial circulation with the direction of flow towards the point of drainage. Gartner[89] claims that the pressure gradient between the

fluid in the ventricles and that in the subarachnoid spaces is too small to account for any more than a minimal circulation through the meshes of the subarachnoid space.

Such are the arguments against the theory that the cerebrospinal fluid has an active circulation. Most of them hinge on the attempt to prove that the movements of dyes which observers have found to take place have been due to mere diffusion of the dye and not to active circulation. Their arguments to me fall by the wayside when compared with Dandy's experiments on hydrocephalus and to Flexner's and Winter's[21] in which they noticed a small flow from a cannula introduced into the aqueduct of Sylvius in etherized cats.

It is probably in these experiments of Flexner's and Winter's that we find anexplanation for some investigators concluding that the cerebrospinal fluid has no active circulation. They found that the flow from the cannula was not constant but varied with the cat and with the physiological conditions present. At all times the flow was small. Surely, when Flexner and Winters found such a small flow through an opening through which all of the cerebrospinal fluid must pass

attempts to measure the spread of a dye introduced into the lacy subarachnoid space, where the fluid has spread out over a large area would lead to conclusions that the dye spread only by diffusion. An analagous situation exists in the blood vascular system. The movement through the large arteries is easily visible due to their small volume. Yet the movement through the capillaries is very slow due to the fact that the same volume of blood has come to occupy a much greater area. Yet no one denies active circulation in the blood vascular system.[33]

In contrast to the usual view of circulation Monakow[50] has proposed a rather unique method of circulation. He states that the cerebrospinal fluid is formed by the choroid plexuses, passes into the ventricle and then back through the ventricular ependyma into the nervous tissue in its perineuronal spaces. From here it goes into the perivascular spaces and thence into the subarachnoid spaces and then into the arachnoid villi. Stern and Gautier[113] and Stern and Rapoport[114] accepted Monakows theory on the basis of the following experiments. Injections of toxic solutions into the ventricles have caused much more

pronounced effect than injections of the same substances into the subarachnoid spaces. Crystalloid dyes introduced into the subarachnoid space failed to produce any coloration of the nervous system. The same dyes injected into the ventricles produced marked staining of the ependyma, neuroglia and ganglion cells and dye was also found in the pericellular and perivascular spaces and within the arterioles and capillaries, not only of the ventricular wall, but of the cortical surface as well. I have already commented on similar findings by Nanagas and Putnam.

The chief objection to Monakow's theory, outside of Dandy's contradictory results with hydrocephalus, seems to me to lie in the fact that there is as much evidence pointing to the elaboration of cerebrospinal fluid by the ependyma as their is evidence that is absorptive in function. To ascribe both functions to it seems hardly plausible.

Mott, in his experiments on cerebral anemia that I have previously discussed came to the conclusion that the direction of flow was towards the pericellular spaces from the subarachnoid space - the exact opposite of the findings of Monakow. It must be remembered that

## Circulation of Cerebro-spinal Fluid

Mott's experiments were done on cerebral anemia and that very likely in cerebral anemia the flow of fluid can reverse and pass down into the pericellular spaces and hence into the cerebral capillaries, from the sub-arachnoid spaces. The majority of evidence seems to indicate though that under normal conditions the flow from the perivascular spaces is centrifugal. Monakow's theory that the pericellular fluid arises from the ventricular fluid by virtue of its passage through the ependyma, however, I believe to be untrue for reasons stated above.

We have yet to consider the movement of fluid in the spinal cord and spinal subarachnoid spaces.

Weed denied that there was any upward movement of fluid in the spinal canal. His theory is supported by various bits of evidence demonstrating the difference in composition of the spinal fluid and cranial fluid. In jaundice bile may be present in the spinal fluid and absent in the ventricular fluid. The cell count is higher in the lumbar region than in ventricular fluid (Weigeldt, 1923)[20] Dahlstrom and Wideroe[85] in similar observations on syphilitics with paresis or tabes found the lymphocyte count to be much higher in

the lumbar fluid than in the ventricular fluid. In six out of seven patients in which they tested the Wassermann they found it to be negative in the ventricular fluid but positive in the lumbar fluid.

On the other hand there is considerable evidence to show that there is a movement of fluid present in the spinal spaces. Kramer,[41] whose work on spinal circulation is perhaps the most significant found that methylene blue injected into the lumbar central canal reached the subarachnoid spaces of the base of the brain, the cerebellum, the whole ventricular system and sheaths of the cranial nerves. The whole central canal of the spinal cord was stained. This would certainly indicate an upward movement of the dye and hence an upward movement of the cerebrospinal fluid. Especially significant was the fact that dye introduced into the ventricular system above the cerebellum did not pass down into the spinal canal. Rather contradictory to this evidence, however, is Kramer's findings that the central canal of the spinal cord is frequently closed near its upper opening in adults. Perhaps the mode of exit of the cerebrospinal fluid from the spinal canal is different in different people.

## Circulation of Cerebro-spinal Fluid

Concerning movement in the spinal subarachnoid spaces Dahlstrom and Vidroe[85] found that phenolsulfonphthalein injected into the subarachnoid spaces in the lumbar region revealed the presence of the dye in the ventricular fluid within five minutes. Stepleanu-Horbatsky[111] in methylene blue experiments in which the dye was injected into the spinal subarachnoid spaces found the medulla, the isthmus, the base of the brain, and the ventricular lining stained with the due.

I have often wondered what effect the arterial pulsation would have on the flow of the cerebrospinal fluid. Observations on the exposed brain of a living animal viewed through a glass plate show that the fine subarachnoid strands of tissue are jerked with each arterial pulsation causing an ebb and flow of the fluid. (Vehrs)[68]

## XII. Summary of the Pathway of Circulation of the Cerebrospinal Fluid

I have endeavored to give a picture that shows how incomplete our knowledge is on the circulation of the cerebrospinal fluid . Since the circulation is intimately bound up with the formation and absorption of the fluid, subjects about which we know but little,

this is the picture which we would expect. The influx, efflux, eddys and currents of the fluid, the trabeculated path which it must follow, the effect physiological states and variations in osmotic pressure of the blood must have on its direction of flow all make its trail difficult to follow. The consensus of opinion at the present time, however, is that the fluid leaves the foramina of Luschka and possibly the foramen of Monro, passes upward through the incisura tentorium cerebelli and finally enters the arachnoid villi and venous sinuses. Fluid probably ascends in a leisurely manner from the spinal canal and spinal subarachnoid spaces entering the cavernous sinus and possibly undergoing some diffuse absorption into the arachnoid veins.

## XIII. Criticism of the Methods of Investigation

The presence of vacuoles, granules and fat droplets in the choroid plexus and ependyma has been offered as evidence of the secretory nature of these structures. The criticism of this contention lies in the fact that it is very possible that these inclusions represent artifacts and are post mortem changes.

## Criticism of the Methods of Investigation

Dandy's extensive experiments with hydrocephalus to me have very little about them to criticize. Some investigators feel that he should have examined his points of blockage microscopically so that he could definitely prove that there was complete blockage. Also the cerebrospinal fluid might have leaked out his incision woulds they say. I have answered this criticism earlier in the paper, however, and shall not again discuss it.

Granular injection masses have been used in studying the circulation and absorption of the cerebrospinal fluid. These same substances have been frequently used by the anatomists and morphologists in studying vascular and lymphatic channels. For this purpose they are admirably suited; but for the study of the cerebrospinal fluid they have but little value. They, as we know, are subject to phagocytosis. The process of phagocytosis is but little understood but depends we know on surface tension and absorption phenomena a great deal. Also we know how great the variance is in various substances concerning their susceptibility to phagocytosis. Thus different substances might show different pathways of escape from the cerebrospinal fluid. Furthermore, and most important, we have no assurance whatsoever

that the pathway traveled by the phagocytic cells represents the true pathway that is followed by the fluid itself. MacCallum[43] in his study on the absorption of granular material from the peritoneal cavity fully realized that the pathway followed by his granular material might not be the pathway for fluid absorption. For this same reason granular masses that are injected into the ventricles and appear in the ependymal cells is no proof that the ependyma is absorbing cerebrospinal fluid. It may be pure phagoc y tic action.

Viscous injection fluids have been used in the methods of investigation . As far as I can see they are to be condemned as practically useless in the study of the cerebrospinal fluid. They must be either agonal or post mortem. They must change the osmotic tension of the cerebrospinal fluid considerably and thus change its absorption remarkably. They pass with difficulty into finer spaces and if injected with the force which is often necessary for their use they may rupture delicate barriers and create artificial pictures. Their use must be limited to the study of the gross confines of the cerebrospinal fluid.

The use of oily emulsions has somewhat the same

criticism of it that the use of granular materials has, namely, that they may be subject to phagocytosis. Other than this they may be useful in identifying pathways of fat absorption.

The most recent method of studying the physiology of the cerebrospinal fluid is by the use of dyes and true solutions. True solutions it seems to me are better than dyes because of the smaller division of the solute. The chief objection to both materials is that they may be toxic to the cells concerned and hence give erroneous results. The easiest way to rule out this possibility is by the use of large numbers of substances with similar physical properties. If their pathway which they follow represents the true pathway of the cerebrospinal fluid then all the substances should be distributed alike. So far this has not been done simply because a large number of suitable substances has not yet been found. A second objection to the use of true solutions and dyes lies in the fact that possibly pure diffusion enters into their spread as a factor as much as the actual circulation and absorption of the fluid. This objection has not completely been ruled out but is minimized by the immediate freezing of the tissues "en

bloc" following death. I have previously commented on this new approach.

Attempts have been made to completely separate the arachnoid spaces from any venous sinus connection for the purpose of studying absorption (Dandy[14]). The chief objection to this method lies in the fact that it is such a technically difficult procedure that we have no surety that it has been completely accomplished.

## XIV. Clinical Importance of a Correct Knowledge of the Mechanism of Formation and Absorption of the Cerebrospinal Fluid

The value of a clear understanding of the mode of formation and absorption of the cerebrospinal fluid cannot be overestimated. Would that we had such an understanding. Whole new therapeutic methods would be opened up and answers to many unsolved problems furnished. In this brief discussion I shall endeavor to point out some of the unsolved problems and to indicate what the future may have in store for us.

The post-concussional and contusional states present symptoms of a varied nature. Persistent headache is common; asthenia, dizziness, etc. may be present. The intracranial pressure, curiously enough, is

frequently raised. (Greenfield)[26] Why is it raised? We don't know. Its presence has been explained on the basis of a pathological alteration of the choroid plexus causing it to secrete more. Persistent brain edema has been blamed for the raised pressure. At any rate the raised intracranial pressure has probably some connection with the symptoms.

How may we modify barrier permeability? If this question could be completely answered I will wager that many organic neurological and possibly functional neurological diseases could be combatted with ease. I have previously shown how barrier permeability varied depending on the substance concerned. Most investigators feel that any substance which is to reach the central nervous system must first pass the so-called hemato-encephalic barrier. This barrier, is composed of the following elements; choroid plexus, ventricular ependyma, histiocytes vascular endothelium and glial cells; its composition of course varies with its location. Now if we could but change its permeability so that substances which we wished to enter the central nervous system could enter at will; or if we could raise its permeability so that toxic substances could be kept out, then

our methods of therapy would be tremendously simplified. Everyone is conscious of the fact that neurosyphilis is often quite refractory to treatment. This is probably because arsenic but poorly passes the blood-brain barrier. Now if we could give some substance with the arsenic that would lower barrier permeability then the arsenic would probably be much more beneficial. As yet no method of changing barrier permeability has been worked out. However, a method which depends on the induction of a reversal of flow of the cerebrospinal fluid for its effectiveness consists in injecting hypertonic glucose with the arsenic. The hypertonic solution causes a reversal of flow of the cerebrospinal fluid in the perivascular and pericapillary spaces so that the fluid, instead of passing from the depths of the nervous tissue outwardly in a centrifugal manner, passes inward from the subarachnoid tissue along the perivascular spaces to the perineuronal spaces. Theoretically, this is supposed to result in a transportation of the arsenic inward along with the cerebrospinal fluid. The only trouble with this method as I see it is that the arsenic must get into the cerebrospinal fluid first through the choroid plexuses

and these plexuses are notoriously impermeable to arsenic - the barrier effect demonstrated.

Russell[106] has shown that the permeability to arsenic and trypan blue increases when brain tissue is injured and decreases again when repair has taken place. His experiments show that barrier permeability may be modified but unfortunately would not have a clinical action. Friedman and Elkeles[87] have shown that the barrier permeability apparently varies with the electrical charge of the molecules. In dogs with crossed circulation, cobra venom, and the toxin of lamb's dysentery passed directly into the brain as they were either positively charged or neutral. Tetanus toxin, botulinus toxin and diphtheria, being negatively charged did not pass the barrier. They had to reach the central nervous system along the nerves. Perhaps the solution of the problem of barrier permeability lies in the field of biophysics?

Heilig and Hoff[91] have made some interesting discoveries which may have some therapeutic value, however. They find that thyroid administration or the administration of bacterial toxins hasten the passage of substances from the blood to the cerebrospinal fluid. Raising or

lowering the $H^t$ ion concentration has the same effect. Theophylline sodium acetate, urotropin and salicylates also increase permeability. Arsenic and antibodies when given with these drugs appear to pass more readily into the central nervous system.

Forced drainage of the cerebrospinal fluid has been found to be of benefit in various conditions, notably acute meningitis. The common explanation for its beneficial effect is that it serves to wash out bacterial toxins and bacteria. However, this may not be its real mode of action at all. It may work by increasing the permeability of the barrier to antibodies.

Walter[118] has worked out a test which he considers a fairly accurate measure of permeability of the barrier. Bromide, being not very easily eliminated by the kidneys, was chosen as the substance through which the permeability of the barrier is measured. 20 mgm. per kilogram of body weight are fed for five days and the blood and cerebrospinal fluid are examined the morning following the fifth day. The stomach must be in a fasting condition at the time of examination. The normal ratio of blood bromide to cerebrospinal fluid bromide is 3:1. 3.3 is

considered as the upper limit of normal. A low ratio means a greater permeability; a high ratio a lowered permeability. Quotients below normal are found in acute meningitis, luetic meningitis, general paralysis, cerebral arteriosclerosis, acute fevers, uremia and alcoholism. Elevated quotients are found in schizophrenia, mental deficiency and at times in post-encephalitic states. The test is not of clinical value as a diagnostic procedure but possibly it may have some value in the future. Perhaps some substance other than bromide will give a more delicate test? Will it serve to throw light on the etiology of the functional states and if it does will it lead to a new method of therapy in them?

Mehrtens[47] and West in attempting to elaborate a test proceeded in an opposite manner from that of Weed. They injected phenolsulfonphthalein into the subarachnoid spaces in people with various pathological conditions. They found that the dye was longer in making its appearance in the urine in people with disease of the central nervous system than in normal people. Diseases of the meninges especailly lengthened the interval between injection and urinary appearance. Weston[76]

had findings similar to those of Mehrtens and West. However, Gardner and Nosik[25] in conducting similar experiments claim that such an experiment is of no value as the variability of rate of absorption in normal people is so great as to make interpretations most difficult. Perhaps some other substance will be found more satisfactory?

Various theories have been advanced on the etiology of the functional psychoses which had for their basis some postulation of alteration in the secretory or absorptive mechanism. Von Monakow[117] proposed the theory that a pathological change in the choroid plexus led to an increase in their permeability to toxic products of protein metabolism which thus entered the ventricular fluid and subsequently diffused into the hypothalamus. Here they caused changes resulting in one of the various functional psychoses. Kafka[95] thinks that all the cerebral diseases (including the functional state) will be found to have their own characteristic protein level in the cerebrospinal fluid when finer methods of analysis are elaborated. The fundamental fault in the functional diseases would thus be in either

the secretory or absorptive mechanism of the cerebrospinal fluid.

The headache of hypertensive encephalopathy is often a most distressing condition. Other symptoms such as insomnia and irritability are often associated with it. These symptoms have been blamed on cerebral anemia but probably equally responsible for them are the alterations in the physiology of the cerebrospinal fluid. A rise in intracranial pressure is most always found whenever a high diastolic pressure is present. The sytolic pressure seems to have no effect on the intracranial pressure. The rise in arterial pressure causes a rise in the venous pressure which interferes with absorption of the cerebrospinal fluid and raises intracranial pressure in accordance with the Monro-Kellie doctrine. Coincidently with rise in venous pressure we have an increase in the permeability of the choroid plexus due to capillary damage. This further tends to cause a rise in intracranial pressure because more cerebrospinal fluid is formed. The amount of increase in permeability of the barrier as determined by Walter's method has been used as an index of the severity of the hypertension.(Kessler,

Moschowitz, and Savitsky[38]). The papilledema of hypertension is due to the same two factors as the headache, namely, increased permeability of the choroid plexus and increased venous pressure, both factors tending to cause a rise in intracranial pressure.

Since Weed discovered that hypertonic solutions cause a drop in intracranial pressure numerous solutions have been tried in an attempt to find one which would cause a sustained drop without a secondary rise. Unfortunately no ideal solution has yet been found. Most hypertonic solutions when given intravenously, although they cause a temporary drop in pressure, are followed by a secondary rise. This secondary rise is due to the fact that the solute of the solution passes through the choroid plexus into the cerebrospinal fluid and in time the osmotic pressure of the cerebrospinal fluid exeeds that of the blood and an increased production of the cerebrospinal fluid results which continues until the two systems have come to equilibrium. The situation is much like a pendulum swinging with it gradually coming to rest. If some substance could be found that was non-toxic and at the

same time did not pass into the cerebrospinal fluid then this objectional feature would be eliminated.

Hypertonic glucose was the first substance used to reduce intracranial pressure. Sucrose has also been used and appears to be better than glucose because it does not pass the barrier so readily and hence does not cause such marked rebound phenomena. (Young)[79] Blau[3] feels that high concentrations of saline are as good as any of the substances yet found. Caffeine he feels is better than glucose but not as good as saline. (15% sol.) Brouder and Bragedorn[4] infer that the individuals upon which the solution is being used plays a part in the effectiveness of the solution. Some people's choroid plexuses are more permeable to certain substances than others. In general, though, they feel that sucrose is the best dehydrating substance. Sorbitol they place on a par with glucose.

I think from this brief survey of substances which have been used to lower intracranial pressure and reduce brain edema we will agree that the perfect substance has not yet been found. It is my opinion that the eventual solution to the problem of raised

intracranial tension will not lie along the line of hypertonic solutions, because these at best can have but temporary effect, but rather will center about the finding of some substance which will, through a selective action on the choroid plexus, reduceits permeability over a considerable period of time. Of course, at present no such substance with such a highly selective action has been found but there is no doubt that the choroid plexus and its modified covering of ependyma are highly specialized structures and thus should be subjects to the action of some specific drug.

I do not intend to discuss hydrocephalus here but simply want to call attention to the fact that it might be more successfully treated if we knew more about the mechanism of formation and absorption of the cerebrospinal fluid. Especially would the idiopathic type benefit by a better understanding of the mechanism; in fact, it probably would then cease to be idiopathic.

The occurrence of meningismus in a variety of conditions has led to numerous explanations for its presence. The most satisfactory explanation for this

phenomenon perhaps lies in the possible difference in osmotic tension between blood and cerebrospinal fluid in acute fevers. In acute fevers there is a sharp drop in the blood chlorides causing a diminished osmotic pressure in the blood. Now there is a certain "lag" in the cerebrospinal fluid chloride, it not dropping as rapidly as that in the blood. Perhaps, during this discrepancy between the osmotic tensions of the two fluids there is an increased formation of the cerebrospinal fluid. This leads to raised intracranial pressure and the symptoms of "meningism."

## XV. Summary of Clinical Importance of a Correct Knowledge of the Mechanism of Formation and Absorption of the Cerebrospinal Fluid

I think that all will agree that a more complete knowledge of the mechanism of formation and absorption of the cerebrospinal fluid is necessary before many puzzling problems in the treatment of neurological diseases may be solved. Much of the work in the future must center around the hemato-encephalic barrier with an attempt to modify its function through changing its permeability to various substances.

## Conclusion

The paucity of very recent articles on the formation and absorption of the cerebrospinal fluid might lead one at first glance to believe that the subject was a settled one and that there was no more work to be done on it. Such, however, is certainly not the case and I have attempted to show in the previous sections how really little we do know about the mechanism of formation and absorption and yet how very important it is that we learn more about these phases of the life cycle of the cerebrospinal fluid.

If we had answers to but a few of the following questions then perhaps the therapy of many neurological conditions would be made many times more successful. Is the fluid a dialysate or a secretion or a combination of the two? If a combination of the two mechanisms which is the more important? How may we modify barrier permeability to our advantage? What effect do different ions have on barrier permeability? (Cohen[8]) Do changed pressure relations within the skull such as in hyrocephalus lead to new routes of formation and absorption of the fluid? If so are these new routes of any importance? Why do the chlorides fall in the cerebrospinal fluid in meningitis? Is it due to the drop in blood chlorides? Is it due to an alteration

## Conclusion

in the cellular precesses forming the fluid? Is it due to the increased protein in the cerebrospinal fluid? Why do the arachnoid villi hypertrophy to form Pacchionian granulations in the adult? Why will fluid pass from the subdural space into the subarachnoid space but not in a reverse direction?

I have attempted to give the impression that it is along the line of barrier permeability modification that future progress will be made for I feel that when and if we can control the permeability of the hematoencephalic barrier we will have a whole new field of approachopened up in the treatment of neurological diseases. Especially will it become more and more necessary that we be able to modify this permeability as the number of chemotherapeutic drugs increases. They can not exert their action if they are denied entrance to the central nervous system. To solve the problems pertaining to the question of barrier permeability I feel that it is necessary that we gain a more fundamental understanding of the mechanism of formation and absorption and how these must vary under changing physiological conditions.

It must be emphasized that the many equivocal results obtained by various investigators may probably

be explained by virtue of the fact that they conducted their experiments under different physiological conditions.

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