THE ACTIONS OF SUBSTANCES INTRODUCED INTO THE CEREBROSPINAL FLUID AND THE PROBLEM OF INTRACRANIAL CHEMORECEPTORS

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PART I. THE ACTIONS OF SUBSTANCES INTRODUCED INTO THE CEREBROSPINAL FLUID

A. Historical introduction

The cerebrospinal fluid (CSF), alone with the cavities containing it, was probably the last system to be investigated physiologically. The extent to which its importance was underestimated is shown by the fact that neither Nagel's nor Luciani's textbook, both of which contain several volumes and were published at the beginning of this century, nor even the seventh edition of Höber's excellent textbook of physiology, published in 1934, makes any mention of this system at all. The function of the CSF was thought to be purely mechanical. It is true that it protects the central nervous system (CNS) which "swims" in it

against mechanical injuries. However, apparently no attention was paid to the fact that this function could not be ascribed to the internally situated ventricles.

The CSF first became an object of greater concern when the *blood-brain barrier* was discovered. It was observed that many substances introduced into the bloodstream either did not reach the brain at all or reached it only very slowly. But if they were introduced directly into the CSF, either the substances or their effects were easily detectable. L. Stern, whose fundamental investigations of the effects of substances introduced into the CSF will be discussed in detail later, believed that all substances were obliged to pass the "barrière hémato-encéphalique" in order to reach the brain.

This problem, the high points of which have been summarized by Friedemann (69), Winterstein (174), and Bakay (5), need not concern us. The blood-brain barrier is of interest to us only insofar as it was the point of departure for the question to be treated here: namely, the effects of substances introduced directly into the cerebral ventricles. Such investigations were initially undertaken to study the passage of substances through the barrier. For this reason, dyes were mainly used since their presence was very easy to detect. Next followed the investigation of substances which, when introduced into the circulation, produced no effects because of their inability to cross the barrier. In the final stages of these investigations the problem arose whether those substances which *did* have an effect when introduced into the bloodstream, had the same or a different effect when they were injected into the CSF.

Before the fundamental research work of Stern and Gautier (1921–1923) the effects of substances introduced into the CSF had seldom been investigated. But as early as 1875 Claude Bernard (13) observed in anesthetized dogs that morphine chloride, subdurally injected, produced hardly any depressant effects, but caused the animals to become excited and to cry as if in pain, as if they had not been anesthetized at all.

At the end of the nineteenth century, Jacob (86) injected potassium iodide subdurally into the lumbar region of dogs and observed marked signs of pain followed immediately by paraplegia of the hind legs (sometimes also of the forelegs), salivation, tachypnea, and increase in temperature, symptoms which lasted twelve to twenty-four hours.

In 1899 Bier (18) introduced lumbar anesthesia by injecting small amounts (5 mg) of cocaine into the CSF of the lumbar region. In the same year Bruno (21) injected small amounts of morphine into the brain matter and observed, instead of the usual tranquilizing effect, violent clonic and tonic convulsions. Whereas he found sodium ferrocyanide given intravenously to be without effect, Lewandowsky (115) observed that when this substance was injected subdurally into the lumbar region, or suboccipitally, violent signs of excitation and vocalization resulted. Injections into the region of the central cerebral convolutions evoked restlessness, chewing, and salivation, phenomena which could not be evoked by intravenous administration, even with doses one hundred times as great.

In a short paper Dixon and Halliburton (44) reported on the rapidity of ab-

sorption of drugs introduced into the CSF. A small dose of atropine (1 ml of a 0.1 % solution) produced paralysis of the vagus in a very short time, usually within a few seconds. Epinephrine (1 ml of a 0.0001 % solution) equally rapidly produced a typical rise of arterial blood pressure, "almost as rapidly as if it were injected intravenously." It should be noted that this result is in direct contradiction to that which all later investigators have found, and suggests that epinephrine may have actually been introduced directly into the bloodstream, perhaps *via* the sinus venosus, as was observed by Stern and Gautier (153). For example, Becht (7) observed that adrenaline and nicotine, when injected into the cisterna magna, did not produce their characteristic effects upon the blood pressure; furthermore, epinephrine could be detected for at least five hours after its injection.

Baglioni and Magnini (4b) appear to have been the first (1909) to investigate the central effect of curare, a problem which has continued to occupy many investigators up to the present. It was indeed a surprising discovery that this substance, which when introduced intravenously produces the well-known paralysis at the neuromuscular junction, produced distinct excitation when applied to the surface of the cortex (2b, 4b) or injected into the spinal cord or the fourth ventricle (McGuigan, 124). The last found in his experiments a noticeable increase in reflex action, development of convulsions, and finally death from paralysis of respiration.

Stern and Rothlin (157) investigated the action of curare when applied directly to the different parts of the cerebellum of dogs, cats, and guinea pigs. They observed a special state of excitation only if the substance reached the fourth ventricle. In dogs, but not in the other two species, this effect was associated with a noticeable increase in temperature.

Stern and Gautier (153–155) performed an extensive investigation during the years 1921–1923. They started with the above-mentioned assumption that all substances, in order to have an effect upon the CNS, were obliged to pass into the CSF within the ventricles. By injecting a large number of substances into the CSF they sought to establish the relationship between the CSF and the circulatory system. We shall summarize briefly the results of their experiments in the order in which they were originally published. The experimental animals changed as well as the sites of injections, which were made partly in the general direction of the fourth ventricle, partly under the dura parietalis, and partly into the lateral ventricles.

Sodium ferrocyanide in guinea pigs produced apnea, sodium thiocyanate in rabbits strong excitation and convulsions, sodium picrate in cats clonic convulsions and dilatation of the pupils.

In agreement with the previously mentioned observations of Becht (7), the investigators did not observe any increase in blood pressure when they introduced 0.5 ml of a 1 % solution of epinephrine into the CSF of dogs. The same held for cats and one rabbit; in a completely analogous experiment on another rabbit there was a sudden increase in blood pressure and the animal died immediately. The investigators mentioned that the injection had been made in the

vicinity of the sinus venosus and that the substance had probably made its way into the blood. Atropine produced the first sign of mydriasis in the cat only after 10 minutes; it became maximal after 17 minutes. Pilocarpine did not produce marked salivation until one and one-half hours after introduction into the lateral ventricle of a rabbit.

In a second paper Stern and Gautier (154) compared in dogs, cats, rabbits, and guinea pigs the effects of a large number of substances when they were introduced into the blood circulation, and into the CSF or the brain matter. By the latter route, sodium ferrocyanide, which was ineffective intravenously, gave rise to convulsions in all four species; subdural injections were much less effective than injections into the lateral ventricles. In contrast, the convulsant action of sodium thiocyanate was approximately the same whether injected subdurally, intravenously, or into the lateral ventricles. Similar results were obtained with sodium picrate. Sodium salicylate was without effect in all three instances. Suitable doses of morphine, curare, strychnine, bile salts, and atropine were found to be convulsant when injected into the CSF, whereas the ineffectiveness of subdurally or intraventricularly injected epinephrine was again confirmed. Marked signs of excitation could be evoked by way of the CSF (either by subdural injection or by injection into one of the lateral ventricles) with dyes such as fluorescine and methylene blue, as well as with sera containing neurotoxins.

The problem of whether the effects of substances introduced into the CSF cavities at different locations are the same or not was the subject of Stern and Gautier's third paper (155). The very important result, which will be considered later in more detail, was that nearly all substances investigated, whether they produced signs of excitation and convulsions (*e.g.*, sodium citrate, KCl, NaF, morphine, pilocarpine, bile) or depression (*e.g.*, $CaCl_2$), were incomparably more effective, even in much smaller doses, when they were injected into the ventricles than into the subarachnoid cavity. Only curare proved to be an exception; in this case no such difference was observed.

In the best interests of a clear survey, the historical-chronological sequence will now be abandoned, and experimental results will be considered according to the nature of the substances investigated. First, let us consider the methods involved. In the great majority of experiments, the substances to be investigated were simply introduced by an injection needle, either into the cisterna magna or through the cranium into one of the lateral ventricles of animals which were in most instances anesthetized. Adam and collaborators (1) were apparently the first to substitute the method of simple injection by a *perfusion method.* A Ringer's solution containing the substance entered the cerebral cavities through one cannula introduced into one of the lateral ventricles and flowed out by way of a second cannula placed into the cisterna magna. The same general method was used later in many experiments by Leusen, who took care to make the composition of the solution as far as possible exactly like that of the CSF (98). Loeschcke (118) also used this method, limiting the perfusion mainly to the fourth ventricle. The perfusion method of Bhattacharya and Feldberg (15) is

essentially identical with that of Leusen. On the other hand, Feldberg with other collaborators (47) led the perfusion only as far as the aqueduct in order to eliminate the fourth ventricle and the subarachnoid cavity.

Bouckaert and Leusen (20) modified the simple perfusion method according to the method which Heymans developed for artificial circulation of blood through the brain of dogs (*cf.* Winterstein, 173). They transfused the CSF of a donor dog into the cerebral ventricles of a smaller dog where it could exert its action.

Apart from its use for the study of special problems, it appears doubtful that the perfusion method is better for investigation of the intraventricular effect of a substance than the simple injection method. The substitution of a normally very slow moving fluid, such as CSF, by a relatively fast flowing perfusion fluid must produce abnormal conditions. Besides, it is impossible to determine the exact site of the effect, a fact which can lead to erroneous conclusions, as we shall see later. The best procedure seems to be that of Feldberg and Sherwood (61), in which a permanent cannula is introduced into one of the lateral ventricles of the brain. The cannula is covered with a rubber membrane, which allows the introduction of substances at any time without the conscious animal's being aware of it.

B. Cations

1. Potassium, calcium, and magnesium. It has already been mentioned that Stern and Gautier (155) observed in rabbits and guinea pigs strong signs of excitation when KCl (1 to 4% solution) was introduced intraventricularly. On the contrary, $CaCl_2$ (1.7% solution) produced a condition of torpor. Later, Stern and Chvoles (152) repeated these experiments on dogs and demonstrated that solutions containing Ca^{++} and K^+ could counteract each other. The same prolonged effect as with KCl could also be produced with Na_2HPO_4, and this too could be counteracted by $CaCl_2$. In investigations made on dogs, Stern *et al.* (156) tried to determine more exactly the mechanism of these K^+-Ca^{++} effects. They found that the decrease of blood pressure, the slowing of the heart, and the dilatation of the blood vessels of the spleen, all produced by stimulation of the pressoreceptors of the carotid sinuses, could be increased by intraventricular injection of small amounts of Ca^{++}. In the same manner the rise of blood pressure caused by clamping the common carotid arteries was diminished.

These effects of K^+ induced Stern (150, 151) to try intraventricular injections of potassium phosphate in treating artificially produced shock in animals, and to recommend this method of treatment for man on the battlefield, since the main feature of shock consists in a considerable reduction in tone of the sympathetic nervous system, whereas K^+ produces exactly the contrary effect. Suboccipital injection of mixtures of K_2HPO_4 and KH_2PO_4 with a pH of 7.6 is supposed to have been very successful. In contrast Ainslie and Dax (2) found that following 11 suboccipital injections of potassium phosphate made in 3 physically normal mental patients, only in one instance did an increase in blood pressure appear in connection with a slowing of the pulse rate. Smolik (147, 148) repeated Stern's

experiments on dogs and found that in normal dogs potassium phosphate produced a marked increase in blood pressure and respiration, but in hemorrhagic hypotension or shock the response was not uniform. With large doses, the blood pressure fell abruptly after its initial rise, respiration became irregular, and death ensued. Downman and Mackenzie (45), using unanesthetized rabbits, found that no matter whether bleeding had preceded the experiment or not, increase in blood pressure, in respiratory movements, and in muscle tone appeared. All these effects were found to be independent of the vagus nerves.

According to Vleeschhouwer (164), the increase of blood pressure produced by the intracisternal injection of KCl was strongly depressed by intravenous injection of F 883 (diethylaminomethyl-3-benzodioxane).

Calma and Wright (23), experimenting on cats, also investigated the effects of intrathecal injection of KCl and $CaCl_2$, usually at the level of the first sacral vertebra. The stimulating effect of the KCl was, according to them, conditioned by the excitation of the afferent fibers in the posterior nerve roots. Hilarowicz and Szajna (82) had already observed that injection of a 1 to 1.5% solution of KCl, particularly into the fourth ventricle, evoked an enormous rise in blood pressure, even up to double the initial value. In addition, after some time the heart rate became slower and the pulse amplitude greater. At the same time they observed a regulatory effect upon the respiration: previously depressed breathing was accelerated, and shallow frequent breathing became slower and deeper. In the case of unanesthetized dogs, KCl, injected at the level of the lumbar vertebrae or the seventh cervical vertebra, at first induced characteristic reflex movements of the forelegs and afterwards an almost complete muscle paralysis. Resnik and collaborators (138) investigated the effect of suboccipital injection of various electrolytes upon the blood pressure of dogs. They found that KCl, as well as all salts which diminished the ionization of calcium (phosphate, oxalate, citrate), caused a distinct increase in blood pressure. Calcium chloride brought about only a slight decrease, but produced a distinct and long-lasting capacity to counteract the stimulating effect of potassium. Magnesium chloride exhibited the same, but a weaker effect than $CaCl_2$. Lead ion evoked a delayed but prolonged increase in blood pressure. All these results were produced by doses which when given intravenously were without effect. The site of action appeared to lie in or near the floor of the fourth ventricle. Goats reacted similarly but were less sensitive. Potassium ion and salts which diminish the ionization of Ca^{++} initially increased the respiration; larger doses led to death by respiratory paralysis.

Increase in blood pressure and secretion of epinephrine following suboccipital injection of KCl were also observed by Hermann and collaborators (78).

The idea of Stern and her co-workers, that the increase in blood pressure is probably conditioned by an influence on the vasomotor center situated on the floor of the fourth ventricle, was confirmed by the investigation of von Euler (50), who found that in cats an intralumbar injection had a longer latency than an intracisternal one. The increase of blood pressure could be blocked by intravenous or intracisternal injection of ergotoxin, and the effect of ergotoxin, *i.e.*, decrease in blood pressure, by KCl. Von Euler observed also the antagonism between the

actions of K^+ and Ca^{++} on the blood pressure, but could not observe any significant effect on the respiration.

In contrast, Smolik (147) observed in dogs an increase in respiration as well as in blood pressure with potassium phosphate. In further investigations, in collaboration with Walker and Gilson (167), he found an increase in blood pressure only when large doses (0.12 ml/kg of a M/6 solution) were used, whereas low doses (0.3 ml) produced a decrease followed by a return to the normal level. The results were exaggerated after vagotomy. The effects on respiration consisted in an initial interruption of inspiration followed by its marked stimulation. The restoration of regular rhythm was associated with an increase in amplitude and rate. Vagotomy resulted in apneustic respiration.

Huggins and Hastings (85) observed, with intracisternal injections in dogs, the contrast between an excess of Ca^{++} and Ca^{++}-lack produced by citrate solutions. In the second instance, violent contractions of the voluntary muscles of the whole body appeared immediately, and lasted from 5 to 15 minutes, after which they were replaced by a still longer period of ataxia accompanied by a wide-based gait. If $CaCl_2$ or $MgCl_2$ was injected in equivalent amounts as soon as the motor reaction was obtained, the muscles relaxed in a few seconds, and within a few minutes the animal appeared to be normal.

Mullin *et al.* (132) also investigated the muscular responses to variations in Ca^{++} and K^+ concentration in the CSF. Lowering the Ca^{++} concentration by washing the cisterna magna with Ca^{++}-free solutions or by injecting small amounts of calcium citrate into the cisterna caused the onset of a typical tetanic syndrome. Increase of the K^+ concentration of the CSF also produced a marked increase in muscular tension. In both cases the signs were associated with rise in blood pressure and increase in respiratory rate. All changes were reversible when the normal Ca^{++} and K^+ concentrations were restored.

Merlis (130), using a method of continuous perfusion of the spinal subarachnoid space on barbital-anesthetized dogs, the spinal cords of which had been sectioned at T-10, observed that solutions free from Ca^{++} or containing sodium citrate produced an augmentation of the spinal flexion reflex and an increase in muscle tone. High Ca^{++} concentrations were without effect.

The relaxing effect of calcium aroused interest in the investigation of its relation to natural *sleep*. But the injections made for this purpose were mostly intracerebral and not into the CSF. Demole (40) injected $CaCl_2$ into the vicinity of the tuber cinereum of the cat. The observed "sleep" was dependent upon the dose (0.0002 to 0.002 g) and could be deepened to anesthesia. Invariable signs of this sleep, which lasted up to several hours, were: closing of the eyes, relaxation of the nictitating membrane, slowing of heart beat and respiration, and relaxation of the muscles. The animal could be aroused but went right back to sleep again. Injection of KCl, on the contrary, evoked excitation.

Marinesco and collaborators (127) repeated Demole's experiments on cats which had been revived out of anesthesia. They found that simple puncture into the wall of the third ventricle in the region of the tuber cinereum evoked typical and reversible signs of sleep. Its appearance was accelerated and its

duration increased by injection of Ca^{++}. Injection of KCl produced at first violent excitation and delayed the sleep. After Ca^{++} injection, the sleep curve often showed two maxima interrupted by a period of wakefulness. The first maximum was attributed to the Ca^{++}, the second to the puncture. Injection of $CaCl_2$ into one of the lateral ventricles also resulted in typical sleep which appeared after 50 to 55 minutes and lasted 3½ to 5½ hours. Under these circumstances, K^+ also gave rise to violent excitation, followed later by sleep.

On the basis of an increase in Ca^{++}, Cloetta in particular has attempted to build a theory of sleep. Together with his pupils Fischer and van der Loeff (29, 30), he observed that injections of Ca^{++} into the infundibular region produced typical conditions of sleep in rats, cats, rabbits, and dogs, which exhibited relaxation of muscles, constriction of pupils, slowing of pulse and respiration, and signs of dreaming; the animals could be aroused readily. Recently Feldberg (54), using his method of permanent cannulation, was able to produce a condition resembling sleep in conscious cats by injection of $CaCl_2$.

The results obtained with the simple injection method, which in general agree with each other, were essentially confirmed and extended in a large number of publications by Leusen (98–102) and his co-workers who used the perfusion method. In experiments on dogs they first established that after bilateral vagotomy and isolation of the carotid sinuses, perfusion of the cerebral ventricles with an isotonic solution of a composition similar to that of the normal CSF caused no great change in blood pressure, carotid sinus reflexes, and the effects of faradic stimulation of the proximal end of the vagus. Along with the other above-mentioned investigators, Leusen established that an excess of K^+ or lack of Ca^{++} increases the vasomotor tone and the vasomotor reflexes. An excess of Ca^{++} could be neutralized by an excess of K^+. Absence of K^+ did not seem to have any influence on the vasomotor system. A solution from which both K^+ and Ca^{++} were absent had the same effect as one with K^+ but without Ca^{++}. According to Bekaert and Leusen (11), the effect of K^+ in the perfusion solution becomes significantly increased and accelerated by the presence of veratrine.

In addition to the increase of vasomotor tone, a rise in K^+ concentration or decrease in Ca^{++} concentration produces a strong contraction of the spleen. This takes place also when the spleen has been denervated, and is therefore probably conditioned also by the secretion of epinephrine (Leusen, 99). Magnesium chloride has the same depressant effect as $CaCl_2$, but lack of it causes little or no change (Leusen, 100–102). Lithium, which was found to be poisonous to animals and humans (literature cited by Leusen and Demeester, 114), proved to be poisonous also when injected suboccipitally. In unanesthetized dogs it produces signs very similar to those observed in patients: muscle tremor, disturbance of sight, adynamia and apathy, confusion, coma, and even death.

Devos (41), using the perfusion method, has investigated particularly the effects of K^+, Ca^{++}, and Mg^{++} on the cardiac activity of anesthetized dogs. According to him an excess of K^+ seems to produce, on one hand cardiac acceleration by stimulation of the accelerator and medullo-suprarenal centers and, on the other hand, a slowing of the heart, due to a reflex influence of the increase in

blood pressure and to stimulation of the cardio-inhibitory center. A decrease in the K^+ concentration did not affect the heart rate. A high concentration of Ca^{++} seemed to stimulate the cardio-inhibitory center and to depress the cardio-accelerator center. On the contrary, a decrease of the Ca^{++} concentration produced stimulation of the cardio-accelerator centers and probably also of the medullo-suprarenal center.

Increase in the Mg^{++} concentration produced a slowing of the heart beat, but this was less pronounced than with excess of Ca^{++}. Decrease of Mg^{++} did not affect the heart rate. Obstruction of the aqueduct of Sylvius and perfusion of the third or the fourth ventricle alone with solutions containing an excess of K^+ or Ca^{++} gave the same result as the perfusion of all the ventricles together (Devos, 42).

Bekaert (9) investigated the influence of the cation content of the perfusion fluid on the movement of the stomach. Increase of K^+ or decrease of Ca^{++} concentration caused an increase in stomach movements, rise in Ca^{++} or Mg^{++} concentration decreased them, and lack of K^+ or Mg^{++} had no effect. Here too the action of increased K^+ could be counteracted by increased Ca^{++}. Denervation of the adrenals had no influence on this phenomenon. On the other hand, it could be removed by cutting or atropine-blockade of the vagi, indicating that it was brought about by way of these nerves.

The effects of K^+, Ca^{++}, and Mg^{++} on pulmonary ventilation were investigated by Verstraeten (160, 162) with the perfusion method. The results proved to be completely analogous to those observed on the other organ systems. Increase of Ca^{++} concentration caused a decrease in respiration. The initial period of excitation, described by Stern and collaborators who used subdural injections, could not be observed. The depression of respiration described by Verstraeten lasted a long time after the initiation of perfusion with normal fluid. Lack of Ca^{++}, as well as increase of K^{++}, evoked increased respiration. Here, on the contrary, the initial depression described by Resnik and collaborators could not be observed. Lack of K^+ had no effect, lack of Ca^{++} and K^+ had the same effect as lack of Ca^{++} alone. Here, too, the effect of excess Ca^{++} could be compensated by an excess of K^+, and excess Mg^{++} had an effect similar to but weaker than increased Ca^{++} concentration; lack of Mg^{++} had no effect at all. In later papers, Verstraeten (161, 163) investigated the effect of these cations on the oxygen consumption. Whereas normal perfusion fluid produced no change, an increase in Ca^{++} concentration produced a reduction in oxygen uptake which persisted long after the return to normal perfusion fluid. An excess of K^+ or lack of Ca^{++} increased the oxygen consumption. This effect was not influenced by thyroidectomy or parathyroidectomy, or by denervation of the adrenals, or by combinations of such. On the contrary, it disappeared after curarization and therefore appeared to be conditioned by the nervous influence on muscle metabolism. Excess Mg^{++} had the same effect as that of Ca^{++}; lack of K^+ or Mg^{++} was without effect.

With their method of injection through a permanent cannula inserted into one of the lateral ventricles, Feldberg and Sherwood (66) investigated the effects

of Ca^{++} and K^+ on unanesthetized cats. $CaCl_2$ (2 mg) produced initially a short period of tachypnea, muscular weakness, and ataxia, followed by bradypnea and an anesthesia-like condition lasting about one and one-half hours. Potassium chloride (0.5 mg) increased alertness and accelerated movements; in larger doses (2 to 3 mg) it produced tonic seizures of short duration with or without a few clonic convulsions. A period of increased muscle tone followed.

John and his co-workers (89) investigated with Feldberg's method the effects on conditioned responses in cats of injections of cations into the cerebral ventricles. The animals were trained: 1) to leap a hurdle on an elevated runway in order to obtain visible food; 2) to discriminate visual patterns (a black circle or a black square on a white background) for food; 3) to avoid shock on the presentation of visual or auditory stimuli. Potassium ion produced contralateral somatic overactivity which could be blocked by Ca^{++}. The latter produced a dynamic immobilization. Strangely, the K^+-Ca^{++} antagonism, which has been observed in all organ systems, was not to be found in conditioned responses. The performance of the various responses deteriorated after injection of K^+ as well as of Ca^{++}. Avoidance responses to visual stimulation were more affected than those to auditory stimuli, and the responses to avoid shock more than pattern discrimination or runway performance, but the deficit in performance was the same whether K^+, Ca^{++}, or a mixture of both had been injected.

2. Ammonium. The NH_4^+-ion deserves special consideration because it forms complex ions. The attention of physiologists has been drawn to it especially on account of the exceptionally small excitatory effect of NH_4Cl acidosis on the respiratory system. Winterstein and Gökhan (179) thought they had found the explanation for this when they discovered that the CSF became alkaline, in spite of the strong acidification of the blood. This led them to the idea that pulmonary ventilation is controlled also by the pH of the CSF. They were strengthened in this belief by the discovery that after removal of the carotid and aortic chemoreceptors by cutting of the vagi and the sinus nerves, NH_4Cl produced, instead of the usual increase, a decrease in respiration, in spite of the fact that the pH of the blood and the CSF remained unchanged. Since the lowering of the pH, which formerly stimulated the respiration, was no longer effective after removal of the chemoreceptors, the authors concluded that the H^+-ions of the blood exercised their influence by way of the chemoreceptors, and not directly because they were not able to pass the blood-brain barrier. In order to confirm this hypothesis, Winterstein and Gökhan (179) in a similarly deafferented dog injected a dose of NH_4Cl, which intravenously produced a decrease in respiration of 33%. Suboccipitally, however, this produced an increase of 80%. They explained it by the assumption that by intracisternal injection the H^+-ions had avoided the blood-brain barrier and were therefore effective.

Bersaques and Leusen (14) studied with their perfusion method the effect of NH_4Cl solutions, the pH of which had been adjusted to 7.3 with NaOH. Initially an increase in respiratory amplitude and frequency was observed; this was regularly followed by respiratory depression. The respiratory movements became more and more irregular until the appearance of complete apnea and death.

Loeschcke and Katsaros (117) perfused the fourth ventricle of lightly anesthetized cats with solutions containing 0.01, 0.03, or 0.1 mg of ammonium chloride per l, and observed the appearance of two stages: (1) rapid increase in tidal volume without change in respiratory rate (this effect increased with the NH_4Cl concentration), and (2) a delayed depression of tidal volume and rate and disturbance of rhythm, which at higher concentrations ended in respiratory arrest. Breathing could be started again by artificial respiration. Both effects, the stimulant as well as the depressant, were also observed when the pH shift caused by NH_4Cl was compensated for. The authors explained these phenomena by the assumption that NH_4Cl acts on two different structures. We shall discuss this in Part II.

3. Hydrogen-ions. The importance which these ions seem to have, not only in the blood but also in CSF, for the control of respiration, has already been mentioned in the discussion of the NH^+-ion. The first scientist who investigated the importance of artificial variations of the pH in the interior of the cerebral ventricles with the perfusion method was Leusen. We shall discuss these effects in connection with the variations in CO_2 pressure, since they have generally been investigated together. At first Leusen (103), using the method of Bouckaert and Leusen (20), demonstrated that hyperventilation of donor dog A produced in dog B, which had had its carotid and aortic chemoreceptors removed, a depression of respiration, whereas artificial respiration of dog A with a mixture of O_2 and CO_2 stimulated respiration in dog B. Perfusion of the ventricles with a solution, the pH of which had been reduced by addition of CO_2, caused hyperventilation and therefore decreased the pH of the blood. On the contrary, rise in pH of the CSF caused hypoventilation and therefore increased the blood pH (Leusen, 105, 107, 110). Investigation of the comparative effectiveness of a change in the CO_2 concentration when on the one hand, the carotid bodies, and on the other, the cerebral ventricles were perfused showed that the latter were less sensitive (106). Hypercapnia produced by breathing CO_2 caused a decrease in the pH of both blood and CSF at the same rate, and when normal air was again breathed, the pH of both returned to normal at the same rate. On the contrary, decrease in pH caused by hyperventilation appeared earlier in the blood than in the CSF, and when normal breathing returned, the CSF remained alkaline for a longer time (Leusen, 108, 112). So far, the results of Leusen agreed completely with the results to be expected from the reaction theory of Winterstein (171, 172, 173). But when Leusen (104, 109, 111) used a borate buffer instead of CO_2 to change the pH of the perfusion fluid between 6.9 and 7.8, he could not observe any influence on the respiration. He found further, that if perfusion was carried out with solutions containing different amounts of CO_2 and $NaHCO_3$, the pH of which was adjusted to 7.3, larger concentrations of bicarbonate initially produced an increase in respiration which, however, returned after a short time to its normal level, and sometimes finally became depressed (Leusen, 109, 111). These results led Leusen (111, p. 51) to the conclusion that "intraventricular alterations in H^+ concentration, independently from CO_2 or HCO_3^- do not influence the respiratory activity," but "the CO_2/HCO_3 ratio in the cerebral ventricles influences

the respiratory centres." These results, which are in contrast not only with all former experimental evidence but also with the physico-chemical law of Henderson $\left(cH = K \frac{[H_2CO_3])}{[HCO_3^-]}\right)$, will be discussed and explained in Part II when we consider the intracranial chemoreceptors.

Exactly opposite results were obtained by Loeschcke and his collaborators (121), which gave fundamentally new knowledge about the control of respiration.

In lightly anesthetized cats with denervated peripheral chemo- and presso-receptors, the fourth ventricle and the adjacent areas of the brainstem were perfused with isotonic bicarbonate buffers, the pH and CO_2 tension of which were varied separately. Acid shift of pH at constant CO_2 tension was followed by an increase, alkaline shift by a decrease of tidal volume. The respiratory rate remained unchanged. On the other hand, increase of CO_2 tension at constant pH was accompanied by a slight but highly significant diminution, decrease by a slight augmentation of the tidal volume.

These results are in complete agreement with Winterstein's reaction theory, according to which pulmonary ventilation is controlled by pH, and the stimulating effect of CO_2 is only due to its influence on the pH. In addition to this, Loeschcke and his collaborators obtained the extraordinary result that acid buffers and hydrochloric acid up to a concentration of 10^{-3} N locally applied to the floor of the fourth ventricle, *i.e.*, to a region which is generally considered to include the so-called respiratory center, had absolutely no effect on pulmonary ventilation. An effect could be obtained only when the acid solution was injected into the lateral recess of the fourth ventricle. This direct demonstration of the existence of intracranial receptors will be discussed in Part II.

It might be mentioned that Winterstein and Wiemer (181) in two experiments found that suboccipital injection of Ringer's solution, the pH of which was brought to a value between 5 and 6, was without effect. Wiemer (168), in experiments with injections of CO_2-free solutions into the cisterna pontis of cats and rabbits, observed that even a solution of pH 2.8 had no effect on respiration.

C. Organic substances

Before consideration of the actions of intraventricular injections of organic substances, it will be useful to discuss the experiments of Loeschcke and collaborators, conducted with *procaine* in direct connection with their studies on H^+-ions, just mentioned.

By perfusing the cerebral ventricles of unanesthetized dogs with Ringer's solution containing procaine, or injecting it into the third and fourth ventricles of cats, Loeschcke and Koepchen (119) observed an inhibitory effect on the chemoreceptor drive as well as on the central drive. In sufficient concentration it arrested the respiratory movements. It also diminished arterial pressure. Local application of a 2% procaine solution in a sponge to the floor of the fourth ventricle in lightly anesthetized cats was without effect on respiration and circulation (120). On the contrary, local application by injection of 0.01 ml into the lateral recess diminished tidal volume and arterial pressure. These results, as will be seen later, are evidently in perfect agreement with the above-mentioned

observations of Loeschcke and collaborators concerning the site of the excitatory effect of H^+-ions.

Loeschcke and Koepchen (118) performed, in addition, perfusion experiments on cats and dogs using solutions containing *veratridine*. The results were completely analogous to those obtained by using solutions of decreased pH. When introduced in small doses into the CSF of the base of the brain, veratridine caused an increase in tidal volume and arterial pressure, but not when applied to the floor of the fourth ventricle.

1. Chemoreceptor stimulators. Winterstein (180) has defined chemoreceptor-stimulators as those substances which, according to the investigations of Heymans and his collaborators, produce their effects by way of the peripheral chemoreceptors.

a. Lobeline. One of the most important of these substances is lobeline. When injected intravenously, it stimulates the respiration, an effect which is often made use of in medical practice. In 1925 Janossy (87) reported the effect of suboccipital injection of lobeline in man. In one case of morphine-poisoning, injection of 10 mg had a favorable effect producing deep, slow, regular respiration and complete recovery. In a second case, of Wilson's disease, the same dose also increased the amplitude and rate of respiration, but caused at the same time very disagreeable side-effects, such as profuse perspiration, bradycardia, intestinal cramps, vomiting, and restlessness. In further cases smaller doses were administered. With injection of 3 to 4 mg, all results and side-effects were the same; however, the side-effects could be avoided by subcutaneous injections of atropine, whereas respiration was even further increased. According to Bakucz (6) there is no doubt that 1.5 to 2 mg lobeline given intracisternally to children with respiratory paralysis is life-saving. Loos (122) has recommended suboccipital injections of lobeline in cases of opiate-poisoning and drowning. Hazama (74) investigated the action of suboccipitally injected lobeline in experimental animals. He found it to be nearly without effect in normal rabbits, but very effective after severe hemorrhage, and even more so in animals poisoned by morphine. In a case of the latter type, for example, the respiration, which had fallen to $\frac{2}{3}$ of its volume, was increased within ten minutes to above normal. Also, in two neonatal animals with poor respiration, suboccipital injection of $\frac{1}{10}$ of the normal intravenous dose had excellent results.

Clementi (27), in applying α-lobeline HCl in a 0.3 to 2.0% solution to the floor of the fourth ventricle in dogs, claimed to have observed strong tachypnea, sometimes with a tendency to periodic respiration. According to him, the accelerating effect of a 0.3% solution of lobeline can be blocked by cutting the vagi, and also by injection of morphine or chloral.

Clementi (28) investigated in dogs the effects of lobeline derivatives, applied to the floor of the fourth ventricle, and observed with lobelanine an increase in respiratory frequency and vomiting, with lobelan only the latter.

Kasahara and Niizu (90) observed with intraspinal injection of 0.18 mg lobeline in rabbits an increase in respiratory amplitude and rate, and with smaller doses (0.06 mg) a decrease of blood pressure.

According to Huang (83), subarachnoid injection of lobeline has a considerable

excitatory effect on normal respiration as well as on respiration impaired by subarachnoid injection of cocaine and other local anesthetics. The effects of the latter drugs, according to him, were diminished, and their paralyzing effect prevented if lobeline had been injected previously. Mercier and Delphaut (129) found that the effect of suboccipitally injected lobeline resembled that of nicotine, as far as it too increased the blood pressure; after a short period of apnea it produced an increase in respiration.

Nicholson and Sobin (134) also found a similarity between the effects of nicotine and lobeline, but following application of lobeline to the floor of the fourth ventricle they found, in contrast to all the other papers mentioned above, that its effect was depressant; definite augmentation of respiration was never observed. This result is in agreement with that of Bekaert and Leusen (12) who, with suboccipital injection of lobeline in a dose which intravenously produced tachypnea, observed in anesthetized dogs a depression of respiration. An increase did not appear, even in unanesthetized dogs. Winterstein and Gökhan (180) injected intravenously ineffective doses of lobeline suboccipitally into dogs deafferented by cutting the vagi and carotid nerves and found an enormous increase in respiratory volume. But it must be mentioned that in these investigations, just as in the others which we have already discussed, it was not taken into consideration that commercial lobeline is strongly acidic.

Loeschcke and Koepchen (118), perfusing the fourth ventricle with solutions containing 10^{-5} g/ml to 10^{-4} g/ml lobeline, confirmed the results of Bekaert and Leusen and obtained a reduction of the tidal volume, and, with solutions containing $3 \cdot 10^{-4}$ g/ml lobeline, respiratory arrest. On account of this, Winterstein in collaboration with Wiemer (181) repeated in cats and rabbits, deafferented by cutting the nerves from the carotid and aortic chemoreceptors, the experiments with suboccipital injection of neutralized lobeline solutions. In 23 % of the cases they found a distinct increase, but never a decrease in respiration. Injecting lobeline into the cisterna pontis, situated on the base of the brain, Wiemer (168) observed regularly a diminution of blood pressure and a decrease in respiratory amplitude, only occasionally preceded by an increase.

b. Cyanide. The effect of NaCN, applied in different ways, has already been thoroughly studied by Winder *et al.* (169). They found that, when injected into an uninjured carotid artery, 0.3 μg/kg was sometimes sufficient to cause an effect on the respiration, whereas after the removal of the carotid body, 75 times this dose produced only a minute effect. Injection of the first dose into the fourth ventricle of a deafferented animal can be effective. Winterstein and Gökhan (180) also obtained a transitory increase of respiration in deafferented dogs, with suboccipital injection of a fraction of the dose which was ineffective intravenously. In perfusing the fourth ventricle of anesthetized cats with weak solutions of NaCN (10^{-6} to 10^{-5}) Loeschcke and Koepchen (118) generally obtained a depressant effect on the respiration. With intermediary concentrations (10^{-4} to 10^{-3}) they found mostly an increase in respiration associated with convulsions, and with still higher concentrations arrest of breathing. The blood pressure was increased with all concentrations. After local application to the floor of the

fourth ventricle, weak concentrations remained without effect, and strong concentrations produced respiratory arrest.

Winterstein together with Wiemer (181) repeated the experiments with suboccipital injection of 0.05 to 0.15 mg/kg NaCl on deafferented cats, and obtained in nearly half of the cases (48.5 %) a distinct increase in respiration, twice preceded by a short decrease. Convulsions appeared only once. Wiemer (168) finally made exhaustive investigations on the effect of cyanide injections into the cisterna basalis of anesthetized and deafferented cats and rabbits. With doses between 0.0125 and 0.1 mg/kg, all animals showed respiratory changes. In the majority of cases, they consisted in an initial decrease in tidal volume and increase in rate, followed by a long-lasting increase of amplitude. Sometimes the initial diminution was preceded by a short period of increase. Besides this, there were some cases in which only an increase or (very seldom) only a decrease was observed. In most cases, the blood pressure also showed two phases. During the diminution of respiration, it often rose, but sometimes fell; during the increase of respiration it also mostly rose, but these changes in respiration and blood pressure were independent of each other. An attempt to explain all these different results of Loeschcke, Winterstein, and Wiemer will be made in Part II.

2. Endogenous compounds. a. Epinephrine. The hormones and the substances influencing their action are of particular interest on account of the extraordinary differences between their effects when they are introduced into the general circulation, and when injected into the CSF. We have already mentioned that Dixon and Halliburton (44) were the only ones who, after injecting epinephrine into the cerebral ventricles, observed the same increase of blood pressure regularly obtained by introducing it intravenously; and the possibility that this may have resulted from inadvertent introduction of the compound into the venous circulation has been noted (*cf.* pp. 72–73).

Baas (4) observed with subdural or intracerebral injection of 6 to 8 mg epinephrine (Suprarenin; Merck) into dogs a sleeplike state lasting from two to five hours. The tendon and corneal reflexes remained intact, and the pupils were small and reacted to light. The investigator considered these phenomena as direct effects on the brain, and not on the brain vessels. Indeed Fog (68), after applying epinephrine to the brain surface could not observe any change in the caliber of the arterioles.

According to Heller (75), intracisternal injection of epinephrine in anesthetized and unanesthetized dogs in doses up to 1.5 g produces no changes of blood pressure, or only minute ones. In cats he observed a small to fatal decrease in blood pressure depending on the dose. Cutting the vagi or atropinization did not abolish this effect. According to him, treating animals with doses of pituitrin which are ineffective alone protects them against an otherwise fatal dose of epinephrine. The depressor effect of epinephrine on the blood pressure changes under these conditions into a pressor effect.

More detailed investigations on the action of epinephrine, when applied intracisternally, have been made by Leimdorfer and collaborators. Leimdorfer and Metzner (96) observed with injections of larger doses (1 mg/kg) analgesia,

drowsiness, and sometimes sleep. The blood pressure, electrocardiogram (ECG), and electroencephalogram (EEG) remained normal, and only the respiratory movements were increased, mainly in amplitude. Leimdorfer *et al.* (95) observed that epinephrine injected into the cisterna magna caused "a rapid, high and sustained rise in the concentration of the glucose in blood." Leimdorfer (97) investigated, in addition, the effects of intracisternally applied sympathomimetic amines. The catechol compounds [N-*iso*propylarterenol, ethylnorepinephrine (Butanefrine), *l*-arterenol] exhibited an analgesic, anesthetic, and hypnotic action, and produced a more or less long-lasting hyperglycemia. Whereas the injection of epinephrine produced a stimulation of the respiratory center without changes in blood pressure, the injection of phenol compounds was followed by a large rise in blood pressure and unfavorable disturbances in the EEG. The phenyl compounds [ephedrine, amphetamine (Benzedrine), phenylpropanolamine (Propadrine)] produced no analgesia, no anesthesia, no sleep, no change in blood glucose, but great excitement, marked rise in blood pressure, and severe disturbances in the ECG. The effects of the 2-amino-heptanes (Teramine and Benethyl) resembled those of the phenyl compounds. These investigations, according to the authors, "indicate a definite relationship between chemical structure of sympathomimetic amines and the mode of their action on the central nervous system."

Reitter (137) also observed in dogs the anesthetizing action of intracisternally injected epinephrine without distinct effects on the circulatory system.

Feldberg and Sherwood (63) injected epinephrine or norepinephrine into one of the lateral ventricles of the cat and observed a slight anesthesia. Sherwood (146) injected epinephrine suboccipitally into catatonic patients and observed general relaxation, lowering of muscle tone, and later, drowsiness or sleep. Feldberg and Malcolm (58) observed the antagonistic effects of epinephrine or norepinephrine on the excitant effects of tubocurarine to be described later.

Palmer (136) investigated in sheep the effects of epinephrine and norepinephrine by the Feldberg method, and observed restlessness and incoördination, and later a state of partial anesthesia, *i.e.*, effects comparable to those described by Feldberg and Sherwood in the cat. The latest investigator to concern himself with intraventricularly injected epinephrine was Rothballer (139). He used Feldberg's method and implanted electrodes in order to take EEG recordings. Shortly after the injection, the cat retched or vomited several times, the latency varying inversely with the dose. Respiration was stimulated. Gradually the cat became inactive, showing none of its normal affectionate behavior. At the height of the effect the animal lay with open eyes, immobile except for panting. It appeared to be extremely weak. There was a conspicuous analgesia or at least absence of response to painful stimuli. Analgesia and stupor seemed to appear hand in hand. While there were no specific EEG changes accompanying these phenomena, it was usual to see varying periods of slow activity characteristic of drowsiness which persisted during the height of the stupor. But these periods alternated with long and conspicuous periods of marked EEG activation, characteristic of the alert cat, even though the animal was lying apparently unconscious.

b. Acetylcholine and acetylcholinesterase inhibiting substances. Suh *et al.* (159) examined the effects of acetylcholine when introduced into the CSF of dogs. Applied intracisternally, as well as directly, to the floor of the exposed fourth ventricle, it produced a rise in blood pressure which was not abolished by sectioning the vagi or atropinizing the animal.

Dikshit (43) introduced small doses of acetylcholine (0.1 to 0.5 μg) into the lateral ventricles of the brain or deeper into the hypothalamic region. Such injections produced a condition closely resembling sleep. The effect came on from ten to thirty minutes after the injection and lasted from two to three hours.

Henderson and Wilson (77) injected 2.5 to 7.5 mg acetylcholine into one of the lateral ventricles of human subjects, and observed such effects as would be expected from stimulation of peripheral cholinergic nerves: *i.e.*, vomiting, intestinal peristalsis, and sometimes sweating. Sleep was an inconstant phenomenon despite the large amounts given compared with those given to cats by Dikshit. But the same amount of acetylcholine which, when introduced into the ventricles, produced an intense and prolonged disturbance, was entirely devoid of action when injected intravenously. Atropine, when injected intraventricularly or subcutaneously, completely prevented or abolished all responses to intraventricular acetylcholine.

Von Euler (50) confirmed the observation of Suh and collaborators that acetylcholine introduced intracisternally causes an increase in blood pressure which can be blocked by ergotoxine, but not by vagotomy or atropinization. Feldberg and Sherwood (62, 63), after injecting 0.1 to 0.5 μg acetylcholine into one of the lateral ventricles of unanesthetized cats, observed within a few seconds as immediate signs retching, high-pitched phonation, and a state resembling an akinetic seizure. Afterwards the animals remained more or less stuporous. Large doses of acetylcholine (1 mg) produced convulsions followed by sleep and stupor, sometimes reaching a catatonia-like condition.

Bhawe (17) injected 500 μg acetylcholine intraventricularly into cats, and observed sometimes an increase but mainly a decrease in blood pressure, effects abolished by intravenous administration of atropine.

It is interesting to compare these observations with the effects of di*iso*propylfluorophosphate (DFP) and other drugs which inhibit the action of acetylcholinesterase and therefore increase the amount of active endogenous acetylcholine. Henderson and Wilson (77) explained in this way the delay of about twelve minutes before the effects of physostigmine (eserine), injected into the human cerebral ventricles, were manifest, and the very close resemblance of these effects to those produced by acetylcholine.

Calma and Wright (23) introduced eserine into the subarachnoid space of cats at the level of the seventh or eighth thoracic vertebra. The effects were limited to the distal part of the neuraxis, and did not develop more rapidly than after intravenous injection.

Feldberg and Sherwood (64) injected eserine and DFP into cats through a permanent cannula and found the results in good agreement with the supposi-

tion that the effects were produced by inhibition of the action of acetylcholinesterase, and therefore by the accumulation of acetylcholine in the diencephalon near the surface of the ventricles. The phenomena observed consisted in severe itching and irritation, as evidenced by intense scratching and vigorous wiping with the foreleg over the face and head; in a second stage, there were changes in gait and posture, and finally, alteration of awareness, and stupor, with signs of catatonia.

Sherwood (146) gave his psychotic patients preparations of cholinesterase intraventricularly and obtained very interesting results: fully catatonic patients began to react, to respond to instructions, or to reply to simple questions. One patient, in a perpetual state of violent paranoid excitement, calmed down and spoke rationally. Of fifteen patients treated in this way, six showed a recovery, apparently lasting sometimes for several years.

Recently Palmer (136) investigated with Feldberg's permanent cannula method the effects of eserine and DFP in sheep. Both drugs evoked excitement, incoordination, and abnormal movements of the head and ears, followed by paralysis of the muscles innervated by cranial nerves. With DFP the period of recovery was longer. The effects in sheep were quite similar to those described in cats. However, stupor and catatonia were not observed in sheep.

c. Extracts of the neurohypophysis. The effects of posterior pituitary extract (Pituitrin) when introduced into the lateral ventricles of men were carefully investigated by Cushing (31). He found definite signs and symptoms of parasympathetic excitation, similar to that which could be produced by the intravenous injection of pilocarpine, *i.e.*, vasodilatation, rise in pulse rate, dilatation of pupils, sensations of warmth, drenching perspiration, excessive salivation, and prolonged retching and vomiting. The blood pressure did not change. Heller and Kusunoki (76), who experimented on dogs with intralumbar injections of intravenously ineffective doses of posterior pituitary, observed a marked increase in blood pressure. Atropine, whether given subcutaneously or previously introduced into the cerebral ventricles, appeared to counteract completely the usual effects of both pilocarpine and Pituitrin, when administered by way of the ventricles (Cushing, 33).

Whereas pilocarpine, injected intravenously, showed the same effects as when injected intraventricularly, the effects with Pituitrin were entirely different according to the route of administration. This suggests that Pituitrin does not produce its effects after absorption into the bloodstream, but directly on the subependymal and diencephalic nerve centers (Cushing, 34). Also the fact that tribromethanol (Avertin), introduced rectally, antagonizes the excitatory effects of intraventricularly injected Pituitrin, is consistent with this conclusion (Cushing, 35).

Heller and Kusunoki (76) found in dogs that an intracisternal injection of the whole extract of the hypophysis (Pituitrin) or Pitressin (vasopressin) was much more effective in unanesthetized dogs than in dogs anesthetized with urethane.

d. Histamine. Bedford (8) found that histamine injected into the subarachnoid space did not cause a fall in blood pressure. Feldberg and Sherwood (63) found

with their method of permanent cannulation of unanesthetized cats violent retching, defecation, swallowing, salivation, tachypnea, and profound muscular weakness.

Bhawe (17) observed secretion of gastric juice in cats after intraventricular injection of histamine (500 μg/kg). It persisted after cutting the vagi and extirpation of the adrenals. It could be proved that it was not a central effect, but the result of the absorption of histamine into the bloodstream. Draškoci and collaborators (46, 47) investigated the absorption of histamine from the cerebral ventricles into the bloodstream with the perfusion method, and confirmed Bhawe's conclusions. The onset of secretion and its increase during the one-hour period were similar whether the histamine was infused intravenously or perfused through the cerebral ventricles.

3. Alkaloids. a. Ergotamine and ergotoxine. Hess (79) observed in 1925 that injection of ergotamine tartrate into the third or one of the lateral ventricles in cats produced sleep, a phenomenon that he considered to be the result of a shift in the excitation-equilibrium from the sympathetic to the parasympathetic nervous system. Marinesco and co-workers (127) confirmed this observation. According to von Euler (50), intracisternal injection of ergotoxine (a mixture of ergocornine, ergocrystine, and ergocryptine) produces, after a short latency, a fall in blood pressure, which counteracts the rise caused by injected potassium.

b. Atropine. Feldberg and Sherwood (63), with the method of permanent cannulation, observed increased liveliness and restlessness in unanesthetized cats following injections of small amounts of atropine (up to 150 μg). The animals became unusually affectionate. With larger doses (200 to 300 μg) this state was preceded by defecation, vomiting, salivation, and tachypnea. The antagonizing action of atropine on the effects of acetylcholine has already been mentioned.

c. Curare and its derivatives have been the object of many investigations. Those made by Baglioni (4b), Amantea (2b), and McGuigan (124) and by Stern and Gautier (155) were already mentioned in the historical introduction. The appearance of convulsions was later observed by von Euler and Wahlund (51) after intracisternal injection of curare and curarine, and by Everett (52) with *d*-tubocurarine and other curare fractions.

Subsequently, Salama and Wright (141, 142) studied the effects of intraventricular, intracisternal, and intrathecal injections of *d*-tubocurarine and many derivatives on cats anesthetized with chloralose or decerebrated. In the first case they observed excitation of the vasomotor, respiratory, cardiac and the other autonomic centers, especially those innervating the salivary glands and the bronchi, and, in addition, increased reflex excitability and generalized convulsions. The quaternary curare alkaloids, *d*-tubocurarine and calabash curare, and the quaternary compound, curarine dimethyl ether dimethiodide, had central excitatory actions, and the tertiary compounds, *l*- and *d*-bebeerines, and the erythroidines, had none.

According to experiments of Chennels (24) on cats and rats with intraventricular, intracisternal, and intrathecal injections, the "competitive" blocking agents [*d*-tubocurarine, *l*-tubocurarine, gallamine triethiodide (Flaxedil), and

o-methoxy-acanthine dimethiodide] all enhanced respiration. In contrast, decamethonium and succinylcholine had little central action.

Reitter (137) described the antagonistic effect of intracisternally injected *d*-tubocurarine on the anesthetizing action of intracisternally given epinephrine, as mentioned before.

Feldberg and his collaborators investigated with the method of permanent cannulation the effects of tubocurarine on anesthetized and unanesthetized cats (summarized in 54, 55, 56). In the latter case, a few minutes after injection convulsions resembling centrencephalic epileptic seizures appeared. The behavior of the cats suggested that the animals were not aware of the convulsions. In cats anesthetized with pentobarbital or chloralose, Feldberg *et al.* (59, 60) investigated the effects of tubocurarine on the electrical activity of the brain. Small doses (15 to 20 μg) which evoked clonic contractions and increase of muscle tone, which could be abolished by an intraventricular injection of norepinephrine, produced inconstant changes in the electrical record of the cerebral cortex. None of the changes could be reproduced by intravenous injections. The effect of topical cortical application of tubocurarine, whether applied locally with a filter paper disk or by flooding the cortex, differed from the effect seen after its intraventricular injection. This important observation will be discussed more thoroughly in Part II.

Apart from the changes in evoked electrical responses, the intraventricular injection of tubocurarine produced episodes of abnormal "spontaneous" activity in cortical and subcortical regions. The flushing of the cortex which occurred regularly on intraventricular injection of large doses of tubocurarine could not be due to a direct action of it since it did not occur when the cortex was flooded with tubocurarine solution.

Feldberg and Malcolm (58) tried to determine more exactly the site of action of intraventricularly injected tubocurarine with the perfusion method of Bhattacharya and Feldberg (16), in which the outflow from the cisterna magna or the aqueduct was collected or the fourth ventricle perfused with a double cannula; they found that, in the case of unilateral injection, the electrical activity of the brain appeared with the same latency on both sides, and argued that this excluded structures lining the lateral ventricles as the sites at which the activity originated. When perfusing with tubocurarine it could be shown that the aqueduct was the most sensitive region from which to elicit muscular effects. The augmentation of the knee-jerk reflex, and the tremor-like activity produced by tubocurarine were prevented or abolished by adding epinephrine or norepinephrine to the perfusion fluid.

Salama and Wright (144) investigated those drugs which, when injected intraventricularly in cats, antagonized the effect of intraventricularly given curare. They found the following to be effective: atropine, acetylcholine, neostigmine, eserine, hexaethyltetraphosphate, tetramethylammonium, and lobeline. Nicotine could delay or diminish the central excitatory effect of subsequently injected *d*-tubocurarine, but was comparatively ineffective in annulling convulsions that had already started. Salama and Wright (143) also made comparative

studies of a great number of drugs applied intraventricularly, intracisternally, intrathecally, and intravenously. RP 3565 (dimethiodide of bis-dimethylaminophenoxy-1:5-pentane), RP 3697 [triethiodide of tris-(β-diethyl-aminoethoxy-) benzene], and tetraethylammonium had a central excitant, and tetramethylammonium a central depressant effect.

d. Strychnine. As early as 1900, it was established by Lewandowsky (115) that the action of subdurally injected strychnine was the same as when a much higher dose was injected intravenously. This was confirmed by Stern and Gautier (154) and by Mercier and Delphaut (129) with suboccipital injections. The latter investigators observed the convulsant effect also with caffeine and nicotine. Many investigators used the local signs of excitation produced by application of minute doses of strychnine to specialized areas of the spinal cord (Baglioni, 4a; Dusser de Barenne, 47b) or the cortex (Baglioni, 4b; Amantea, 2a; Dusser de Barenne, 47a, 47b; Giachetti, 70a) and even in the interior of the brain (Dusser de Barenne, 47c) in the studies on sensory localization.

e. Morphine. On the contrary, the anesthetizing effect of subcutaneously induced morphine changed into a convulsant one when the drug was applied by way of the CSF, as we have already mentioned in the historical introduction (cf. p. 72) (Bernard, 13; Bruno, 21; Stern and Gautier, 154).

Méhes (128) made the curious observation that small amounts of morphine, injected suboccipitally, evoked a violent itching reflex in the region of the head and neck. Similar effects have also been obtained with codeine and paracodeine, but not with dionine or heroin. Königstein (91) tried to localize more exactly the origin of the "scratch action" which he produced in animals not only with morphine, but also with suboccipital injections of caffeine, camphor, acetylcholine, and physostigmine. It remained after extirpation of the whole brain down to the acoustic nucleus. According to Winiwarter (170) a center situated in the medulla oblongata produced this phenomenon spontaneously when it was excited by morphine, or by impulses conducted by way of sensory nerves.

f. Veratridine. Perfusion of the fourth ventricle of anesthetized deafferented (vagi and carotid nerves sectioned) cats with solutions containing veratridine produced an increase of blood pressure and tidal volume, effects which were not obtained by applying the solutions to the floor of the fourth ventricle (Loeschcke and Koepchen, 118).

g. Bulbocapnine. Among the many drugs studied by Feldberg and Sherwood (63, 64) for their effects on unanesthetized cats, bulbocapnine merits special attention. Intraventricularly injected, it showed its effect with $\frac{1}{40}$ of the subcutaneously effective dose. Large doses (1 mg) evoked catatonic stupor, whereas smaller doses (100 to 200 μg) produced short-lasting changes in behavior; for example, a previously docile cat might show hostility.

4. Drugs producing local and general anesthesia. Besides morphine, the local anesthetics cocaine and procaine (Novocaine), when introduced into the CSF, also show effects which are generally different from those obtained with local application. But investigators do not agree on this. According to Huang (83), subarachnoid injection of lethal doses of local anesthetics quickly reduced respira-

tory rate and amplitude, an effect which could be abolished by the intravenous injection of lobeline. According to Nicholson and Sobin (134), cocaine in solution or mixed with three parts of petrolatum to form an ointment, when applied to the obex of the medulla oblongata of dogs produced in some cases sudden arrest of breathing, and otherwise variable effects such as temporary decrease of respiratory rate and amplitude.

Detailed investigations have been performed by Loeschcke and Koepchen (119, 120). As described previously (*cf.* p. 82), local application of 2% procaine in a sponge to the floor of the fourth ventricle was without effect on respiratory movements and arterial pressure. In contrast, injection of 0.01 ml to the lateral recess diminished the blood pressure and had an inhibitory effect on the chemoreceptor as well as on the central drive. After denervation, a sufficient concentration arrested respiration. All effects were fully reversible. The results are in full agreement with the authors' investigations on the central excitatory effects of H^+-ions (*cf.* p. 82), and will be discussed in Part II as the most cogent argument for the existence of intracranial chemoreceptors.

Very interesting results were obtained by Feldberg (57) when anesthetic drugs (barbiturates, magnesium chloride, chloral, chloralose) were injected intraventricularly in doses which were ineffective intravenously. Under these conditions, all these substances, before producing an anesthesia-like condition, evoked *hyperphagia*. The author explained this strange effect by the supposition that, according to Anaud and Brobeck (3), the lateral areas of the diencephalon must be regarded as the actual feeding centers, since bilateral destruction of them is followed by a complete absence of eating. On the contrary, the ventromedial nuclei exert an inhibiting influence on these centers. "The injected anaesthetics impinge on these medially situated nuclei and by anaesthetizing them, remove the inhibitory control over the feeding center and produce hyperphagia" (Feldberg, 57, p. 27).

5. Glucose. The reason why a considerable number of scientists investigated the effect of injection of glucose into the CSF was the assertion of Marinelli and Giunti (125, 126) that suboccipital injection of 10% glucose produced a transient but considerable hypoglycemia in dogs and in one man. The maximum was to be observed after 15 minutes, and the return to normal was complete after two hours. None of the investigators who tried to confirm this fact was able to do so—neither Lackey (92) with suboccipital injection into anesthetized or unanesthetized dogs, nor Weiland and collaborators (167b) with lumbar injection in men, nor Sack and collaborators (140) by performing ventricular, suboccipital, or lumbar injection in patients.

Leusen and Demeester (113) injected glucose suboccipitally in unanesthetized dogs, and observed even a slight hyperglycemia, caused by systemic absorption, but never hypoglycemia. Suboccipital injection of insulin did not produce hyperglycemia. This result was confirmed by Vuylsteke (166) who, using Leusen's method of perfusing the cerebral ventricles of dogs, did not observe any change in blood sugar.

6. Miscellaneous drugs. Finally, the effects of various drugs which are dif-

ficult to classify into special groups may be mentioned. With the method of permanent cannulation of unanesthetized cats, Feldberg and Sherwood (63) obtained effects more or less like those of histamine, such as muscular weakness, vomiting, salivation, and tachypnea with various drugs: *e.g.*, hexamethonium, decamethonium, and methantheline (Banthine). Adenosinetriphosphate (but not sodium pyrophosphate) caused muscular weakness, ataxia, and a tendency to sleep. With 5-hydroxytryptamine (serotonin) they observed muscular weakness, tachypnea, and profuse salivation. The cat showed a pronounced tendency to sit or lie down. This depressant action on the brain is, according to Gaddum and Vogt (70), antagonized by intraventricularly injected lysergic acid diethylamide (LSD), ergometrine, morphine, methadone, and amphetamine, but not by 2-bromo-LSD, 5 benzyloxygramine or methylmedmain. The sedation produced by *reserpine* is, according to them, antagonized by LSD, morphine, and methadone. The action of reserpine intraventricularly administered to unanesthetized cats by Feldberg's method has been studied by Dasgupta and Haley (37). They found relaxation of the nictitating membrane, miosis, narrowing of the palpebral fissure to a slit, avoidance of light, diarrhea, anorexia, and tranquilization. The metabolites of reserpine were not involved in these reactions.

John *et al.* (88) investigated the effect of reserpine, intraventricularly injected through Feldberg's permanent cannula, on the conditioned responses of unanesthetized cats, a method which we have already described in the discussion of the effects of cations (p. 80). Reserpine attenuated avoidance responses while leaving approach responses relatively unaffected. With sufficiently low doses, it was possible to block visual and avoidance responses, leaving the response to auditory stimuli and the approach responses unaffected. They studied also the interactions of centrally injected serotonin, iproniazid, epinephrine, norepinephrine, and atropine. Only epinephrine appeared to attenuate the effects of reserpine. Methamphetamine consistently reversed the effects of reserpine when injected peripherally. Central injection of this drug reversed only the autonomic effects; the reserpine-induced block of conditioned responses were not affected as much.

Bekaert and Kluyskens (10), who also collected the clinical bibliography about the damaging or even fatal effects of the intracisternal injection of streptomycin, investigated the effects of streptomycin sulphate, dihydrostreptomycin, and chloralhydrate-streptomycin in dogs. They found a progressive depression of respiratory rate and amplitude. The blood pressure also fell progressively after an initial slight rise.

At the end of this section, it may be mentioned that Munck (133) observed the rapid onset of depression in dogs into which the CSF of uremic patients had been injected suboccipitally. The animals were not able to stay upright and lost their sensitivity to pain. After one and one-half hours recovery was complete. These phenomena resembled closely those of the patients from whom the CSF had been taken. The CSF of chronically ill patients was much more effective than that taken from acutely ill ones. Normal CSF never produced such effects.

Part II. The Problem of Intracranial Chemoreceptors

In the first part we reviewed the effects obtained by introducing various substances into the CSF. Now the question to be discussed is the way in which these effects were produced, a question seldom posed until now. There are three main possibilities: 1) The substances introduced into the CSF may be reabsorbed into the bloodstream; 2) they may penetrate into the CNS and have direct effect on it; and 3) they may stimulate the chemosensitive endings of some receptor nerves.

As we have already mentioned on different occasions, L. Stern (149) believed that all substances in order to reach the brain and to be effective there, were obliged to permeate the fluid contained in the ventricles. Surely nobody would support this idea any more. The speed with which some substances, especially the anesthetics, are effective, excludes the possibility of diffusion through the slowly moving CSF. Under these conditions it would be impossible indeed to understand the importance of the rich capillary net of the brain, if there were no direct exchange of material between blood and brain substance. Hauptmann (73) proposed a weaker form of Stern's theory supposing that all substances were obliged to pass the "Weg über den Liquor," understanding by this not necessarily the CSF contained in the ventricles but also the part filling the so-called Robin-Virchow cavities between the brain vessels and the brain substance. But in this form also the theory can no longer be maintained, since anatomical research has established that these cavities surround only the small arteries and not the capillaries, which immediately border on the brain substance. Indeed, there can be no doubt that substances such as anesthetics for which no barrier exists, can pass directly into the brain and be effective. On the contrary, the substances which are not able to permeate the barrier between the blood, on the one hand, and the CSF or the brain on the other hand, are—as Stern rightly recognized—effective only when they are introduced directly into the CSF or the brain matter, thus bypassing the barrier.

The idea that such substances produce their effects by special receptors ending in the CSF has been rarely considered in spite of the fact that there are several observations pointing clearly in this direction. If, for example, passing the blood-brain barrier by direct introduction into the CSF were the only condition for making a substance effective, the site of introduction into the CSF should be without importance. Stern and Gautier (155) have already established that this is not the case. In several experiments they observed that subarachnoid injections were much more slowly effective than intraventricular ones, or even not effective at all. On account of this, these investigators were the first to discuss the possibility that the effects of such substances "sont dus à une action réflexe déclanchée par l'excitation des partis ventriculaires."

This idea has not been pursued until recently, when it was first suspected that inside the cranium "intracranial" or within the brain centers "intracentral" receptors for chemical stimuli exist, analogous to the peripheral chemoreceptors discovered by Heymans in the carotid and aortic bodies. The basis of this idea was found to be fundamentally different from, and to have no connection at all with, the experiments described in the first part of the present review.

The term "medullary chemosensitive receptors" within respiratory centers was first used by C. von Euler and Söderberg (48) as a result of the following observations. They recorded the action currents of the central end of the phrenic nerve in decerebrated and curarized cats, which were hyperventilated with oxygen containing 6.5% CO_2. If these animals were anesthetized with chloralose, the action currents diminished considerably, whereas the electrical potentials evoked reflexly by electrical stimulation of the superior laryngeal nerve, or the central trunk of the vagus, or some other nerve remained unchanged. They concluded that within the centers some receptors, especially sensitive to CO_2, or the pathways of respiratory reflexes starting from these receptors had been paralyzed by chloralose. In further experiments they recorded action currents from the completely denervated rhombencephalon, and found that this was possible only when the animal was ventilated with oxygen containing CO_2 and not with pure oxygen. This also seemed to the authors to be a proof of a CO_2-effect produced by central receptors.

In a second series of experiments von Euler and Söderberg (49) recorded slow potential changes from the medullary respiratory centers in decerebrate cats. The amplitudes of these potentials depended upon the CO_2 content of the inspired air, and were specifically depressed by chloralose, whereas they remained unimpaired by respiratory reflexes. The investigators concluded again that these "chemo-potentials" were signs of the activity of special medullary chemosensitive receptors.

In our opinion neither the observations of von Euler and Söderberg nor the numerous other experiments on the "loss of CO_2-excitability" (summarized by Winterstein, 172), justify the conclusion that intracranial chemoreceptors exist. The conclusion rests on the assumption that CO_2 produces a specific excitation of the respiratory centers analogous to those induced by nerve impulses. This is contradicted by the "reaction theory" (Winterstein, 171, 173) according to which the CO_2 has an excitatory effect only by lowering the pH, whereas its specific effect on nervous tissues is a paralyzing one, at least if its concentration rises above a certain level. From the bibliography reviewed by Winterstein (172) we shall mention only a few experiments in which this depressing effect has been directly proved. Lorente de Nó (123) found that in frog nerves all concentrations of CO_2 between 0.5 and 100% increase the threshold for electrical stimulation, and according to the observations of Laget and Lavigne (93) the threshold concentration for this depressant effect of CO_2 was found to be as low as 0.1%. We have already mentioned the more recent experiments of Loeschcke and his collaborators (121) (*cf.* p. 82) according to which, in perfusion of the fourth ventricle with solutions of constant pH, every increase of CO_2 concentration produces a decrease, every diminution of it produces an increase in pulmonary ventilation. Therefore it is easy to understand why the addition of the depressant effect of CO_2 to that of chloralose induces a diminution of reactivity where the simple transmission of reflexes does not. In addition, the dependence of action potentials on the CO_2 concentration observed by von Euler and Söderberg need not be explained only by specific chemosensitive receptors. It can be explained by the fact that the respiratory centers need a certain concentration of hydrogen

ions to be able to work rhythmically and their activity ceases if this level is not reached. The effects of CO_2 are therefore not sufficient to prove the existence of intracranial chemoreceptors. Winterstein and Gökhan (180) evolved new arguments founded upon the way the substances designated by them as "chemoreceptor stimulating substances" act. These are substances which, according to the classical investigations of Heymans and his collaborators, produce their central effects by means of the peripheral chemoreceptors, and remain without effect after deafferentation (summarized by Heymans and Bouckaert (80) and Heymans and Neil (81)). Winterstein and Gökhan (179) concluded from their investigations on NH_4Cl-acidosis (p. 80) that the H^+-ions of the blood belong to these chemoreceptor-stimulating substances which lose their effects after deafferentation. Since they found that all these substances regained their activity after having been introduced directly into the CSF, it was a short step to the conclusion that within the CSF, too, their effects were initiated through some intracranial chemoreceptors.

The proof that this conclusion was correct, or at least very probable, has been provided for the hydrogen-ions by the investigations of Loeschcke and his collaborators (121). As we have already mentioned above, they showed that acid buffers up to 10^{-3} N were completely without effect when applied locally to the region of the bulbar respiratory center, whereas when injected into the lateral recesses of the fourth ventricle, they influenced pulmonary ventilation according to the hydrogen-ion concentration. This phenomenon can scarcely be explained in any other way than by the supposition that some chemoreceptors which influence the respiration reflexly exist at these sites.

This supposition also gives an excellent explanation of Loeschcke and Koepchen's (120) observations of the opposite effect of procaine (p. 82). Just as hydrogen-ions have no excitatory effect on the floor of the rhombencephalon, procaine has no paralyzing effect there, whereas, when injected into the third or fourth ventricle of deafferented dogs, it arrests pulmonary ventilation, evidently on account of its effect on some chemoreceptors. This may also be the explanation of the strange observation of Leusen (*cf.* p. 81), who perfused the cerebral ventricles with CO_2-free solutions, and did not observe any effect on pulmonary ventilation when he varied the pH. If it is assumed that there was no specific effect of the borate, used as the buffer, the probable explanation is that the intracranial chemoreceptors for hydrogen-ions could not be reached directly by the perfusion fluid, but only indirectly by the quickly diffusing CO_2.

According to Loeschcke and Koepchen (118) the effects of *veratridine* also give evidence of intracranial chemoreceptors (*cf.* p. 91). Application of this substance to the floor of the fourth ventricle in solutions up to a concentration of $5 \cdot 10^{-4}$ g/l has no effect on respiration and blood pressure, whereas perfusion of the ventricle with solutions of 10^{-7} g/l increases tidal volume and arterial pressure.

The numerous papers of Feldberg and his collaborators, discussed in the first part of this review, concerning the effects of widely different substances introduced into the CSF, contain in our opinion many convincing arguments in

favor of the presence of intracranial chemoreceptors. Although Feldberg (53, 54, 56) does not explicitly mention this idea, he had already (in his first communication on intraventricular administration of drugs) designated the periventricular gray matter as "an as yet scarcely explored area of high pharmacological sensitivity." For example, Feldberg and Sherwood (64) found the effect of intraventricular injections of bulbocapnine to be at least forty times as effective as subcutaneous injections, and this led them to the conclusion that "the relative smallness of the dosage and the speed with which the bulbocapnine effects appear indicate that structures close to or directly in contact with the ventricular surface are likely to be implicated" (p. 373).

In the same way Feldberg and Sherwood (64) concluded from their experiments on acetylcholine and cholinesterases: "We must therefore accept the view that acetylcholine can produce motor phenomena from some region close to the ventricular surface" (p. 497). By application of this view to the results obtained with eserine and DFP they concluded: "The motor phenomena produced by these anticholinesterases are thus thought to result from inhibition of cholinesterase and accumulation of acetylcholine in the diencephalon near the ventricular surface" (p. 497).

In using drugs which pass the blood-brain barrier with difficulty the difference in doses and in speed with which an effect appears may not be a reliable argument in favor of the existence of intracranial chemoreceptors; but on the other hand, with substances acting from the CSF as well as from the bloodstream, the extraordinary qualitative difference in the effects can certainly be a sign of a difference in the mechanisms of action. Let us recall from the first part the differences in the effects of intravenously and intraventricularly applied epinephrine, morphine, curare, *etc.* The differences in action mentioned above, depending on the locality in the CSF into which a substance has been introduced, speak also in favor of specific receptors at certain sites of the CSF cavities. For example, Feldberg *et al.* (59, 60) observed that the effect of tubocurarine, when the exposed cerebral cortex was flooded with a solution of 1:1000, could be produced by intraventricular doses as small as 20 to 40 μg! Besides this, the first application did not elicit the characteristic features of the second, *i.e.*, the intense flushing of the cortex, the widespread muscular contractions, and the generalized increase in electrical activity. Therefore the investigators concluded that tubocurarine "probably has a selective action on nerve cells near the ventricle lining and the fact that the changes spread over wide areas of the cortex with varying latencies signifies only that some focus is probably acting as a powerful 'pacemaker' " (p. 144). This means evidently that tubocurarine acts on specific chemoreceptors, which transmit their excitation to some other parts of the brain.

Winterstein and Wiemer (181) repeated Winterstein and Gökhan's experiments on the action of lobeline and of sodium cyanide (*cf.* p. 84) with solutions of physiological pH, and found lobeline to be effective in 23%, and NaCN in 45.5% of the experiments in increasing pulmonary ventilation. The most probable explanation of this behavior seems to be that these drugs in order to be effective were obliged to diffuse from the site of application to the receptors.

Therefore the cyanide ion, with its smaller size, succeeded more easily in reaching the place of action than the larger lobeline molecules. Analogous considerations are valid for Wiemer's (168) experiments, already described, on injection of NaCN and lobeline into the cisterna basalis (p. 85). Whereas suboccipital injections of NaCN produced an increase in respiration in about half of the experiments, basal intracisternal injections were always effective, but in varying forms. In most of the cases there appeared at the beginning a short diminution of respiratory amplitude, followed by a long-lasting increase, which was interrupted by a second diminution if a second injection was made. Lobeline, which suboccipitally had no effect at all or only an excitatory one, had only a depressant action if injected into the cisterna basalis. How could a substance like the cyanide ion, the water-solubility of which, according to Loeschcke (117), is 500 times as great as that of CO_2, evoke, when injected at intervals of a few millimeters, such different or even opposite effects if its action were always produced uniformly on the respiratory center? According to Winterstein (176), a detailed analysis of Wiemer's experiments leads to the probable conclusion that the partly depressant and partly stimulating effects of cyanide do not succeed each other, but occur at the same time and partially interfere with each other. The interruption of the stimulant effect by a second depression following a second injection indicates this. All these events could only be explained by the presence of numerous chemoreceptors which excite the different parts of the respiratory center by means of their individual connections with it, in the same way as the various tactile, optic, and acoustic receptors of the sense organs produce quite different effects in spite of being relatively near to each other.

The recent experiments of Loeschcke and Katsaros (117) with perfusion of NH_4Cl solutions through the fourth ventricle and adjacent areas (p. 81) led them to the conclusion that the respiratory and vasomotor effects which can be observed during the initial period are due to superficial responsive elements which may be identical with the structures sensitive to hydrogen-ions. Here too it may be a matter of chemoreceptors spatially separated from the respiratory center and united with it by specific nerve fibers.

GENERAL REMARKS AND ANATOMICAL CORRELATIONS

If the material collected in this review is surveyed, it will be apparent that the existence of special intracranial chemoreceptors is highly probable. By this term we do not mean, as several investigators have proposed, the fact known long ago, that chemical stimuli are able to produce central or centrogenic effects by actions directly on the brain. We mean by this term specific nervous formations which are especially adapted to receive chemical stimuli and to transmit these excitations to other centers, in the same way as the nerve endings of the sensory organs are adapted to their specific stimuli. It must be admitted that it will rarely be possible to draw definite conclusions as to the correctness of this supposition. Other explanations may be possible. The most favored is a "central chemical excitability" attributed to nerve cells, axons, or synapses. The question why certain cells, axons, or synapses should be endowed with such an excitability

whereas quite similar neighboring ones are not apparently has not troubled the scientists who have proposed this explanation.

An excellent example of this kind of problem is presented by the "vomiting center." Wang and Borison (167a) established in the medullary surface of the lateral reticular formation the presence of a specialized chemoreceptor trigger zone, after the destruction of which intravenously or orally administered apomorphine, which generally had been considered as a "central" excitant of the vomiting center, had no longer any effect. By their experiments and their detailed study of the bibliography (summarized in 18a), the investigators came to the conclusion that "the concept of a direct action of 'central emetics' on the vomiting center is no longer tenable. Indeed there is, at present, no good evidence that any substance which causes emesis, as its chief or side-effect, does so by direct stimulation of the vomiting center" (18a, p. 209). "All emetic responses, as far as is known, are mediated via reflex arcs which pass through the vomiting center regardless of whether these responses are initiated at peripheral or central receptor sites" (p. 225).

Of course an incontestable proof for the existence of a chemoreceptor will be delivered only when we succeed in demonstrating these receptors histologically, and in proving their functions by stimulating and by removing them. Loeschcke and his collaborators, who showed that hydrogen-ions are effective only on the surface and the lateral parts of the fourth ventricle and not near the respiratory center, and that paralysis of respiration by procaine is also possible only at the same place, have come closest to attaining this aim.

Histological research has demonstrated the possibility of the existence of such nervous receptors, especially in the ventricles. As early as 1922 Stöhr (158), investigating in men the innervation of the pia mater and the choroid plexus, established the presence of a great number of various nerve endings. According to him they were mainly sensitive to variations in pressure of the CSF; however, this does not exclude their possible sensitivity to changes of its chemical composition. He referred especially to the richness of nerve fibers in the tela choroidea not to be observed in any other place, which certainly points to some important function of this part.

Clark (25, 26) emphasized that the nerve endings of the choroid plexus of the fourth ventricle in which, according to Loeschcke, the main seat of the chemoreceptors is to be found, exhibit the structure of sensory nerves except for those leading to the smooth muscle of the blood vessels. Voetmann (165), who in more recent times investigated the structure of human choroid plexus, also considered the nerve fibers of it to be sensory. Schaltenbrand (145) mentioned in his review of the structure of plexuses the enormous richness in probably sensory nerves of the telae of the third and fourth ventricles. Recently Fleischhauer (67) studied the glia of mammals with the help of the fluorescence microscope, and described the exceedingly complicated structure of the third and the lateral ventricles, which points to a manifold function of their walls. Unfortunately the innervation was not investigated.

Appendix: The Theory of Intracranial Chemoreceptors and the Central Effect of Oxygen Lack

For more than a century it has been well known that chemical factors, especially the content of CO_2 and O_2 in the inspiratory air, influence pulmonary ventilation. It was customary to distinguish between nervous and chemical regulations of respiration. The first was supposed to be performed by impulses transmitted through nerve fibers, and the second to be directly active on some part of the CNS, considered to be the origin of the activation of pulmonary ventilation, the so-called respiratory center. The latter was thought to differ from the other centers since it not only integrated and transmitted nerve impulses, but could also be directly excited by chemical factors such as the composition of the blood. This theory of a central excitability of the respiratory center was modified by Heymans' discovery that a great part of chemical control of respiration is not central but peripheral in origin. Numerous experiments (summarized in Winterstein, 172) proved that increase in CO_2 pressure was partly, and lack of oxygen perhaps completely, effective by means of reflexes which had their origin in specific sense organs, the aortic, and mainly the carotid bodies. In spite of this, there seemed to be no doubt that a direct central response to CO_2 and H^+-ions existed also. Meanwhile, Loeschcke discovered the existence of intracranial chemoreceptors which, in agreement with the reaction theory, are sensitive only to hydrogen-ions. Thus, the second category of respiratory stimuli also appeared to have its origin in peripheral formations from which the excitations are transmitted to the centers. There no longer appears to be any need to assign to the respiratory center an exceptional position; a proper central chemical control of respiration does not necessarily exist. The respiratory center is a center, like all the others of the central nervous system, the function of which consists in the integration and transmission of the innumerable afferent impulses coming from all parts of the body (Winterstein, 175). Among them are also the chemical excitations which, as all others, are conducted by nerve fibers, from the peripheral chemoreceptors of Heymans (80), especially sensitive to lack of oxygen, as well as from the intracranial chemoreceptors of Loeschcke (121), especially sensitive to H^+-ions.

From an analogous standpoint, it may be possible to gain insight into the problem, until now unsolved, of the central effect of oxygen lack. The experiments with anesthetized animals on the effect of oxygen lack have shown that the increase of pulmonary ventilation, normally observed, does not occur after deafferentation of the carotid and aortic chemoreceptors. But, at least in dogs, this appeared to be a result of the anesthesia. Moyer and Beecher (131) observed that O_2-lack under light hexobarbital or thiopental anesthesia produced hyperpnea after a long latency period. Davenport and his collaborators (38) described in unanesthetized deafferented dogs the effect of O_2 lack as an initial respiratory depression followed by a period of increased respiration. The authors explained this phenomenon by the hypothesis that, during the hypoxia, a strongly exciting chemical respiratory stimulus developed in the CNS.

This idea agreed perfectly with that of Liljestrand (116) about the action of

cyanide, which depends on an inhibition of the oxidation processes and approaches, therefore, that of hypoxia. Liljestrand thought that cyanide, by producing O_2-lack, initially depressed the respiration which was then increased by hypoxic formation of acid.

The question as to how the inhibition of oxidation produces a respiratory depression, which is the only effect to be observed in anesthetized animals and leads to respiratory arrest and to death, has not been explained by any of these investigators. Since, in unanesthetized dogs, the respiratory apparatus is able to perform on an even greater scale in the second period than before, the hyperpnea cannot be the result of an insufficient production of energy due to lack of oxygen.

Gökhan and Winterstein (71) tried to explain the weakening of respiration by the hypoxic shift of the pH of the CSF to the alkaline side which they had discovered. Wiemer (168), as already mentioned, found that the injection of NaCN into the cisterna pontis produced mostly an initial depression followed by an increase of respiration, and tried to explain this according to Liljestrand's theory. We have just shown (p. 98) that the phenomena are in much better agreement with the assumption that a simultaneous excitation of several partly inhibitory, partly excitatory, intracranial chemoreceptors takes place. Indeed Noell and Kornmüller (135) succeeded in showing that O_2-lack, as well as cyanide, is able to produce an increase of action currents and of spontaneous electrical potentials of the cortex, *i.e.*, excitatory phenomena without a secondary accumulation of hypoxic acids.

According to the assumption just explained, the initial hypoxic depression of pulmonary ventilation after removal of the peripheral chemoreceptors is not a passive event, but active inhibition. The biological function of such an inhibition might be that if a lack of O_2 is so great that it can no longer be compensated by reflex increase of respiration, but reaches the centers themselves, a reduction of respiratory work and oxygen consumption might be of advantage.

The existence of inhibitory processes through the vagus nerves in the pulmonary control of respiration has been known for a long time. Winterstein and Frömter (177) have recently shown that this inhibition not only is conditioned by the tension of the lung walls, as had been believed, but also is always present even independently of it. After removal of the peripheral chemoreceptors, pulmonary ventilation arrested by lack of oxygen can be temporarily revived by a simple blockade of the vagi. As they have demonstrated in a second paper (178), this tonic inhibition concerns mainly the reflex apparatus of the peripheral chemoreceptors. The depression of respiration which is produced by a blockade of these chemoreceptors is much greater when the vagi, too, are blocked at the same time. This means that under these conditions the part of respiratory control due to the carotid and aortic bodies had increased.

All these observations speak in favor of the hypothesis that the central reactions to lack of oxygen are not the result of a direct chemical excitability of the respiratory center, but of reflexes starting from inhibitory and excitatory *intracranial* chemoreceptors, as in the case of peripheral chemoreceptors and intra-

cranial hydrogen-ion receptors. The study of their morphology and the localization of their function is an important task for future research.

REFERENCES

1. Adam, H. M., McKail, R. A., Obrador, S. and Wilson, W. C.: Acetylcholine in the cerebro-spinal fluid. J. Physiol. 93: 45P, 1938.
2. Ainslie, J. A. and Dax, E. C.: Blood pressures after injection of K salts. Brit. med. J. 2: 768, 1942.
2a. Amantea, G.: Azione della stricnina e del fenolo sulle diverse zone della corteccia cerebrale del cane. Arch. Farmacol. sper. 14: 3, 1912.
2b. Amantea, G.: Sull azione del curaro applicato direttamente sui centri nervosi. Arch. Farmacol. sper. 14: 3, 1912.
3. Anaud, B. K. and Brobeck, J. R.: Localisation of a "feeding center" in the hypothalamus of the rat. Proc. Soc. exp. Biol., N. Y. 77: 323, 1951.
4. Baas, A.: Über eine Wirkung des Adrenalins auf das Gehirn. Z. ges. Neurol. Psychiat. 26: 600, 1914.
4a. Baglioni, S.: Physiologische Differenzierung verschiedener Mechanismen des Rückenmarks. Arch. Anat. Physiol., Lps., 1900, suppl., p. 193.
4b. Baglioni, S. and Magnini, M.: Azione di alcuni sostanze chimiche sulle zone eccitabili della corteccia cerebrale del cane. Arch. Fisiol. 6: 240, 1909.
5. Bakay, L.: The Blood-Brain Barrier, with Special Regard to the Use of Radioactive Isotopes. Charles C Thomas, Springfield, Ill., 1956.
6. Bakucs, J.: Über Zisternenpunktion im Kindesalter. Klin. Wschr. 6: 1379, 1927.
7. Becht, F. C.: Studies on the cerebrospinal fluid. Amer. J. Physiol. 51: 1, 1920.
8. Bedford, T. H. B.: The absorption of histamine from the subarachnoid space of the dog. J. Physiol. 120: 62P, 1953.
9. Bekaert, J.: Influence de la teneur en potassium, calcium et magnésium du liquide céphalo-rachidien sur les mouvements de l'estomac. Arch. int. Physiol. 58: 69, 1950. (Preliminary communication: Arch. int. Pharmacodyn. 77: 54, 1948.)
10. Bekaert, J. and Kluyskens, P.: Influence de l'injection sous-occipitale de streptomycine sur la respiration et la pression artérielle. Brux. méd. 30: 1603, 1950.
11. Bekaert, J. and Leusen, I.: Influence de la vératrine sur l'action du potassium du liquide céphalo-rachidien. Arch. int. Physiol. 57: 229, 1949.
12. Bekaert, J. and Leusen, I.: Influence of the suboccipital injection of lobeline upon respiration. XVIII Int. Physiol. Congr., p. 99, Copenhagen, 1950. (Same subject in French: Schweiz. med. Wschr. 80: 1236, 1950.)
13. Bernard, Cl.: Leçons sur les anésthesiques et sur l'asphyxie, p. 213. Paris, 1875.
14. Bersaques, J. de and Leusen, I. R.: The direct influence of ammonium chloride on the respiratory center. Arch. int. Pharmacodyn. 97: 13, 1954.
15. Bhattacharya, B. K. and Feldberg, W.: Perfusion of the ventricular system of the brain in the anaesthetized cat. J. Physiol. 135: 4P, 1956.
16. Bhattacharya, B. K. and Feldberg, W.: Perfusion of cerebral ventricles. Effects of drugs on outflow from the cisterna and the aqueduct. Brit. J. Pharmacol. 13: 156, 1958.
17. Bhawe, W. B.: Experiments on the fate of histamine and acetylcholine after injection into the cerebral ventricles. J. Physiol. 140: 169, 1957.
18. Bier, A.: Versuch über Cocainisierung des Rückenmarks. Dtsch. Z. Chir. 51: 361, 1899.
18a. Borison, H. L. and Wang, S. C.: Physiology and pharmacology of vomiting. Pharmacol. Rev. 5: 193, 1953.
19. Bouckaert, J. J.: Au sujet des fonctions du liquide céphalo-rachidien. Brux. méd. 31: 1528, 1951.
20. Bouckaert, J. J. and Leusen, I.: Technique pour l'étude des influences directes du liquide céphalo-rachidien sur les centres nerveux supérieurs. Experientia 5: 452, 1949.
21. Bruno, J.: Über die Injektion von Giften ins Gehirn. Dtsch. med. Wschr. 25: 369, 1899.
22. Calma, I. and Wright, S.: Action of intrathecal injected eserine on the spinal cord of the cat. J. Physiol. 106: 80, 1947.
23. Calma, I. and Wright, S.: Effects of intrathecal injection of KCl and other solutions in cats. Excitatory action of K ions on posterior nerve root fibres. J. Physiol. 106: 211, 1947.
24. Chennels, M. H. D.: Patterns of respiratory discharge in the cat and rat. J. Physiol. 138: 33P, 1957.
25. Clark, S. L.: Nerve endings in the chorioid plexus of the fourth ventricle. J. comp. Neurol. 47: 1, 1929.
26. Clark, S. L.: Innervation of the chorioid plexuses and the blood vessels within the central nervous system. J. comp. Neurol. 60: 21, 1934.
27. Clementi, A.: La esistenza del centro respiratorio nel bulbo dimonstrata mediante la lobelinizzazione circoscritta del pavimento del 4° ventricolo. Arch. Fisiol. 27: 299, 1929.
28. Clementi, A.: Über die emetische Wirkung von zwei Nebenalkaloiden der Lobelia inflata, nämlich des Lobelanins und des Lobelanidins. Arch. exp. Path. Pharmak. 181: 265, 1936.
29. Cloetta, M. and Fischer, H.: Über die Wirkung der Kationen Ca, Mg, Sr, Ba, K und Na bei intracerebraler Injektion. Arch. exp. Path. Pharmak. 158: 254, 1930.
30. Cloetta, M., Fischer, H. and van der Loeff, M. R.: Die Biochemie von Schlaf und Erregung mit besonderer Berücksichtigung der Bedeutung der Kationen. Arch. exp. Path. Pharmak. 174: 589, 1934.
31. Cushing, H.: I. The reaction to posterior pituitary extract (pituitrin) when introduced into the cerebral ventricles. Proc. nat. Acad. Sci., Wash. 17: 163, 1931.

32. CUSHING, H.: II. The similarity in the response to posterior lobe extract (pituitrin) and to pilocarpine when injected into the cerebral ventricles. Proc. nat. Acad. Sci., Wash. 17: 171, 1931.
33. CUSHING, H.: III. The action of atropine in counteracting the effects of pituitrin and of pilocarpine injected into the cerebral ventricles. Proc. nat. Acad. Sci., Wash. 17: 178, 1931.
34. CUSHING, H.: IV. The method of action of pituitrin introduced into the ventricle. Proc. nat. Acad. Sci., Wash. 17: 239, 1931.
35. CUSHING, H.: V. The counteractive effect of tribromethanol (avertin) on the stimulatory response to pituitrin injected in the ventricle. Proc. nat. Acad. Sci., Wash. 17: 248, 1931.
36. CUSHING, H.: VI. Concerning a possible "parasympathetic center" in the diencephalon. Proc. nat. Acad. Sci., Wash. 17: 253, 1931.
37. DASGUPTA, S. R. AND HALEY, T. J.: Intraventricular administration of reserpine and its metabolites to conscious cats. Brit. J. Pharmacol. 12: 529, 1957.
38. DAVENPORT, H. W., BREWER, G., CHAMBERS, A. H. AND GOLDSCHMIDT, S.: The respiratory responses to anoxemia of unanesthetised dogs with chronically denervated aortic and carotid chemoreceptors and their causes. Amer. J. Physiol. 148: 406, 1947.
39. DAVSON, H.: Physiology of the Ocular and Cerebrospinal Fluids. Little, Brown & Co., Boston, 1956.
40. DEMOLE, V.: Pharmakologisch-anatomische Untersuchungen zum Problem des Schlafes. Arch. exp. Path. Pharmak. 120: 229, 1927.
41. DEVOS, J.: Central influences on heart rate. Arch. int. Physiol. 59: 385, 1951.
42. DEVOS, J.: Localisation of the action of the potassium and calcium ions on the central nervous system. Arch. int. Physiol. 59: 388, 1951.
43. DIKSHIT, B. B.: Action of acetylcholine on the "sleep centre." J. Physiol. 83: 42P, 1935.
44. DIXON, W. E. AND HALLIBURTON, W. D.: The rapidity of absorption of drugs introduced into the cerebro-spinal fluid. J. Physiol. 44: vii, 1912.
45. DOWNMAN, C. B. B. AND MACKENZIE, C. C.: Intracisternal injection of potassium phosphate. Lancet 2: 471, 1943.
46. DRAŠKOCI, M., FELDBERG, W. AND HARANATH, P. S. R. K.: Passage of circulating adrenaline into perfused cerebral ventricles and subarachnoidal space. J. Physiol. 150: 34, 1960.
47. DRAŠKOCI, M., FELDBERG, W. AND HARANATH, P. S. R. K.: Absorption of histamine into the blood stream on perfusion of the cerebral ventricles and its uptake by brain. J. Physiol. 150: 50, 1960.
47a. DUSSER DE BARENNE, J. G.: Experimental researches on sensory localisations in the central cortex. Quart. J. exp. Physiol. 9: 355, 1916.
47b. DUSSER DE BARENNE, J. G.: Experimentelle Untersuchungen über die Lokalisation des sensiblen Rindengebietes im Grosshirn der Affen (Macacus). Dtsch. Z. Nervenheilk. 83: 273, 1924.
47c. DUSSER DE BARENNE, J. G. AND SAGER, O.: Über die sensiblen Funktionen des Thalamus opticus der Katze. Z. ges. Neurol. Psychiat. 133: 231, 1931.
48. EULER, C. VON AND SÖDERBERG, U.: Medullary chemosensitive receptors. J. Physiol. 118: 545, 1952.
49. EULER, C. VON AND SÖDERBERG, U.: Slow potentials in the respiratory centers. J. Physiol. 118: 555, 1952.
50. EULER, U. S. VON: Reflektorische und centrale Wirkung von Kaliumionen auf Blutdruck und Atmung. Skand. Arch. Physiol. 80: 94, 1938.
51. EULER, U. S. VON AND WAHLUND, H.: Über sentrale Kurarewirkung. Acta physiol. scand. 2: 327, 1941.
52. EVERETT, G. M.: Pharmacological studies of *d*-tubocurarine and other curare fractions. J. Pharmacol. 92: 236, 1948.
53. FELDBERG, W.: Intraventricular administration of drugs in the cat. Proc. R. Soc. Med. 48: 853, 1955.
54. FELDBERG, W.: Pattern of excitation and inhibition produced by injection of substances into the cerebral ventricle of the conscious cat. Abstr. XX Int. Physiol. Congr., p. 18 (summary). Brussels, 1956.
55. FELDBERG, W.: The action of drugs injected into the cerebral ventricles. International Symposium on Psychotropic Drugs, p. 303 (summary). Milan, 1957.
56. FELDBERG, W.: Behavioral changes in the cat after injection of drugs into the cerebral ventricle. Brain and human Behaviour 36: 401 (summary), 1958.
57. FELDBERG, W.: A physiological approach to the problem of general anaesthesia and of loss of consciousness. Brit. med. J. 2: 771, 1959.
58. FELDBERG, W. AND MALCOLM, J. L.: Experiments on the site of action of tubocurarine when applied via the cerebral ventricles. J. Physiol. 149: 58, 1959.
59. FELDBERG, W., MALCOLM, J. L. AND SHERWOOD, S. L.: Some effects of tubocurarine on the electrical activity of the cat's brain. J. Physiol. 132: 130, 1956.
60. FELDBERG, W., MALCOLM, J. L. AND SMITH, I. D.: Effect of tubocurarine on the electrical activity of the cat's brain under chloralose. J. Physiol. 138: 178, 1957.
61. FELDBERG, W. AND SHERWOOD, S. L.: A permanent cannula for intraventricular injections in cats. J. Physiol. 120: 3P, 1953.
62. FELDBERG, W. AND SHERWOOD, S. L.: Intraventricular injections of acetylcholine and of 5-hydroxytryptamine (serotonin) into the conscious cat. J. Physiol. 120: 12P, 1953.
63. FELDBERG, W. AND SHERWOOD, S. L.: Injections of drugs into the lateral ventricle of the cat. J. Physiol. 123: 148, 1954.
64. FELDBERG, W. AND SHERWOOD, S. L.: Behaviour of cats after intraventricular injections of eserine and DFP. J. Physiol. 125: 488, 1954.
65. FELDBERG, W. AND SHERWOOD, S. L.: Injections of bulbocapnine into the cerebral ventricles of cats. Brit. J. Pharmacol. 10: 371, 1955.

66. FELDBERG, W. AND SHERWOOD, S. L.: Effects of calcium and potassium injected into the cerebral ventricles of the cat. J. Physiol. 139: 408, 1957.
67. FLEISCHHAUER, K.: Fluorescenzmikroskopische Untersuchungen an der Faserglia I. Z. Zellforsch. 51: 467, 1960.
68. FOG, M.: Cerebral circulation. I. Reaction of pial arteries to epinephrine by direct application and by intravenous injection. Arch. Neurol. Psychiat. 41: 109, 1939.
69. FRIEDEMANN, U.: Blood-brain barrier. Physiol. Rev. 22: 125, 1942.
70. GADDUM, J. H. AND VOGT, M.: Some central actions of 5-hydroxytryptamine and various antagonists. Brit. J. Pharmacol. 11: 175, 1956.
70a. GIACHETTI, A. AND PIVA, G.: L'azione della colinesterase sul clono corticale stricninico. Rivista critica e recerche sperimentali. Arch. Fisiol. 58: 188, 1958.
71. GÖKHAN, N. AND WINTERSTEIN, H.: Die chemische Reaktion der Cerebrospinal-Flüssigkeit im Sauerstoffmangel. Hoppe-Seyl Z. 295: 71, 1953.
72. HAMPERL, H. AND HELLER, H.: Die Ausbreitung intrazisternal injizierter Stoffe im Liquorraum. Arch. exp. Path. Pharmak. 173: 283, 1933.
73. HAUPTMANN, A.: Der "Weg über den Liquor." Klin. Wschr. 4: 1297, 1925.
74. HAZAMA, F.: Über die Verwendung des Lobelin-Ingelheim zur intraspinalen Injection. Münch. med. Wschr. 74: 1418, 1927.
75. HELLER, H.: Über die zentrale Blutdruckwirkung des Adrenalins. Arch. exp. Path. Pharmak. 173: 291, 1933.
76. HELLER, H. AND KUSUNOKI, G.: Die zentrale Blutdruckwirkung des neurohypophysären Kreislaufhormons (Vasopressin). Arch. exp. Path. Pharmak. 173: 301, 1933.
77. HENDERSON, W. R. AND WILSON, W. C.: Intraventricular injection of acetylcholine and eserine in man. Quart. J. exp. Physiol. 26: 83, 1936.
78. HERMANN, H., JOURDAN, F., MORIN, G. AND VIAL, J.: Mécanisme de l'action hypertensive de l'ion potassium. C. R. Soc. Biol., Paris 129: 593, 1938.
79. HESS, W. R.: Über die Wechselbeziehungen zwischen psychischen und vegetativen Funktionen III. Schweiz. Arch. Neurol. Psychiat. 16: 285, 1925.
80. HEYMANS, C. AND BOUCKAERT, J. J.: Les chémo-récepteurs du sinus carotidien. Ergebn. Physiol. 41: 28, 1939.
81. HEYMANS, C. AND NEIL, E.: Reflexogenic Areas of the Cardiovascular System. Churchill, London, 1958.
82. HILAROWICZ, H. AND SZAJNA, M.: Über den Einfluss der subduralen Darreichung von Kaliumchlorid auf die Vasomotoren, Hershemmungs- und Atemzentren. Z. ges. exp. Med. 64: 772, 1929.
83. HUANG, Y.: Über den Einfluss des intravenös injizierten Lobelins, Hexetons und Coramins auf die Atemwirkungen der in den Subarachnoidealraum eingespritzten Lokalanaesthetica. Folia pharm. jap. 17: 12, 1934. (*Cf.* Ber. ges. Physiol. 78: 170, 1934.)
84. HUANG, Y.: Über den Einfluss der subarachnoidealen Injektion des Lobelins, Hexetons und Coramins auf die Atemwirkungen der in den Subarachnoidealraum eingespritzten Lokalanaesthetica. Folia pharm. jap. 18: 69, 1935. (*Cf.* Ber. ges. Physiol. 83: 469, 1935.)
85. HUGGINS, C. B. AND HASTINGS, A. B.: Effect of calcium and citrate injections into cerebrospinal fluid. Proc. Soc. exp. Biol., N. Y. 30: 459, 1933.
86. JACOB, P.: Duralinfusion. Berl. klin. Wschr. 35: 461, 488, 1898.
87. JANOSSY, J.: Über die Wirkung des intrazisternös verabreichten Lobelins. Med. Klinik 21: 1009, 1925.
88. JOHN, E. R., WENZEL, B. M. AND TSCHIRGI, R. D.: Differential effects on various conditioned responses in cats caused by intraventricular and intramuscular injections of reserpine and other substances. J. Pharmacol. 123: 193, 1958.
89. JOHN, E. R., TSCHIRGI, R. D. AND WENZEL, B. M.: Effects of injections of cations into the cerebral ventricles on conditioned responses in the cat. J. Physiol. 146: 550, 1959.
90. KASAHARA, M. AND NIIZU, R.: Tierexperimentelle Untersuchungen der intraspinalen Injection von Lobelin-Ingelheim. Z. ges. exp. Med. 80: 206, 1931.
91. KÖNIGSTEIN, H.: Über Lokalisationsversuche des durch Morphium ausgelösten "Kratswerkes" im Centralnervensystem. Arch. int. Pharmacodyn. 62: 1, 1939.
92. LACKEY, R. W.: Effect of the injection of glucose into the cerebrospinal fluid. Science 106: 618, 1947.
93. LAGET, P. AND LAVIGNE, S.: Précisions sur les limites d'action de l'anhydride carbonique dans le fonctionnement du nerf de Batracien. C. R. Soc. Biol., Paris 144: 810, 1950.
94. LEIMDÖRFER, A.: Über die Wirkung intralumbal eingeführter Hypophysenpräparate auf den Blutdruck. Arch. exp. Path. Pharmak. 118: 253, 1926.
95. LEIMDORFER, A., ARANA, R. AND HACK, M. H.: Hyperglycemia induced by the action of adrenaline on the central nervous system. Amer. J. Physiol. 150: 588, 1947.
96. LEIMDORFER, A. AND METZNER, W. R. T.: Analgesia and anesthesia induced by epinephrine. Amer. J. Physiol. 157: 116, 1949.
97. LEIMDORFER, A.: The action of sympathomimetic amines on the central nervous system and the blood sugar: relation of chemical structure to mechanism of action. J. Pharmacol. 98: 62, 1950.
98. LEUSEN, I.: Influence de la teneur en calcium et en potassium du liquide céphalo-rachidien sur le système vasomoteur. Arch. int. Pharmacodyn. 75: 422, 1948. (Preliminary communication: Experientia 4: 154, 1948.)
99. LEUSEN, I.: Mécanisme de l'action centrale du potassium et du calcium sur le tonus vasomoteur. Arch. int. Pharmacodyn. 77: 48, 1948.
100. LEUSEN, I.: Influence de la concentration en magnésium du liquide céphalo-rachidien sur le système vasomoteur. Arch. int. Pharmacodyn. 77: 50, 1948.
101. LEUSEN, I.: The influence of calcium, potassium and magnesium ions in cerebrospinal fluid on vasomotor system. J. Physiol. 110: 319, 1949. (Same subject as 98–100 in English.)

102. Leusen, I.: Balancement entre les influences vaso-motrices des concentrations intraventriculaires de calcium, potassium et magnésium. J. Physiol., Paris 42: 157, 1950. (Same subject.)
103. Leusen, I.: Au sujet de l'influence du pH céphalo-rachidien sur la respiration. Arch. int. Physiol. 57: 432, 1950.
104. Leusen, I.: Au sujet de la spécificité de l'action du CO_2 sur le centre respiratoire. Arch. int. Physiol. 57: 456, 1950.
105. Leusen, I.: Influence du pH du liquide céphalo-rachidien sur la respiration. Experientia 6: 272, 1950. (Same subject.)
106. Leusen, I.: Importance comparative de l'action centrale directe et de l'influence réflexe du CO_2 sur la respiration. Experientia 6: 390, 1950.
107. Leusen, I.: Influence du pH du liquide céphalo-rachidien sur le pH sanguin. Arch. int. Physiol. 58: 112, 1950.
108. Leusen, I.: pH sanguin, pH du liquide céphalo-rachidien et fonction respiratoire. Arch. int. Physiol. 58: 115, 1950.
109. Leusen, I.: Influence of different concentrations of CO_2 and bicarbonate on the respiratory center. Arch. int. Pharmacodyn. 87: 248, 1951.
110. Leusen, I. R.: Chemosensitivity of the respiratory center: influence of CO_2 in the cerebral ventricles on respiration. Amer. J. Physiol. 176: 39, 1954. (Same subject as 105 and 107.)
111. Leusen, I. R.: Chemosensitivity of the respiratory center. Influence of changes in the H^+ and total buffer concentrations in the cerebral ventricles on respiration. Amer. J. Physiol. 176: 45, 1954. (Same content as 104 and 109.)
112. Leusen, I. R.: Acid-base equilibrium between blood and cerebrospinal fluid. Amer. J. Physiol. 176: 513, 1954. (Same content as 108.)
113. Leusen, I. and Demeester, G.: Au sujet de l'action centrale du glucose. Arch. int. Physiol. 57: 227, 1949.
114. Leusen, I. and Demeester, G.: Au sujet de la toxicité du chlorure de lithium. Acta med. scand. 138: 232, 1950. (Preliminary communication: Arch. int. Pharmacodyn. 81: 390, 1950.)
115. Lewandowsky, M.: Zur Lehre von der Cerebrospinalflüssigkeit. Z. klin. Med. 40: 480, 1900.
116. Liljestrand, G.: The action of certain drugs on respiration. Brit. med. J. 2: 623, 1951.
117. Loeschcke, H. H. and Katsaros, B.: Die Wirkung von in den Liquor cerebralis eingebrachtem Ammoniumchlorid auf Atmung und Vasomotorik. Pflüg. Arch. ges. Physiol. 270: 147, 1959.
118. Loeschcke, H. H. and Koepchen, H. P.: Über das Verhalten der Atmung und des arteriellen Drucks bei Einbringen von Veratridin, Lobelin und Cyanid in den Liquor cerebrospinalis. Pflüg. Arch. ges. Physiol. 266: 586, 1958.
119. Loeschcke, H. H. and Koepchen, H. P.: Beeinflussung von Atmung und Vasomotorik durch Einbringen von Novocain in die Liquorräume. Pflüg. Arch. ges. Physiol. 266: 611, 1958.
120. Loeschcke, H. H. and Koepchen, H. P.: Versuch zur Lokalisation des Angriffsortes der Atmungs- und Kreislaufwirkung von Novocain im Liquor cerebrospinalis. Pflüg. Arch. ges. Physiol. 266: 638, 1958.
121. Loeschcke, H. H., Koepchen, H. P. and Gertz, K. H.: Über den Einfluss von Wasserstoffionenkonsentration und CO_2-Druck im Liquor cerebrospinalis auf die Atmung. Pflüg. Arch. ges. Physiol. 266: 569, 1958.
122. Loos, J. W.: Ned. Tijdschr. Geneesk. 93: 3273, 1949. (*Cf.* Bekaert, J. and Leusen, I., ref. 11.)
123. Lorente de Nó, R.: A Study of Nerve Physiology, chap. III, p. 148: Carbon dioxide and nerve function. Rockefeller Institute, New York, 1947.
124. McGuigan, H.: The central action of curare. J. Pharmacol. 8: 471, 1916.
125. Marinelli, L. and Giunti, V.: Concentration en glucose de la liqueur c.r. et centres nerveux glycorégulateurs. Schweiz. med. Wschr. 77: 1231, 1947.
126. Marinelli, L. and Giunti, V.: Hypoglycemic effect of intraspinal glucose injection. Proc. Soc. exp. Biol., N. Y. 65: 23, 1947.
127. Marinesco, G., Sager, O. and Kreindler, A.: Experimentelle Untersuchungen zum Problem des Schlafmechanismus. Z. ges. Neurol. Psychiat. 119: 277, 1929.
128. Méhes, J.: Experimentelle Untersuchungen über den Juckreflex am Tier. II. Auslösen heftiger Juckanfälle bei der Katze durch intracisternale Injektionen von Morphium und einiger seiner Derivate. Arch. exp. Path. Pharmak. 188: 650, 1938.
129. Mercier, F. and Delphaut, J.: Sur l'action expérimentale de la strychnine, de la caféine, de la nicotine, de la lobéline administrées par voie sous-occipitale. C. R. Soc. Biol., Paris 121: 1509, 1936.
130. Merlis, J. K.: The effect of changes in the calcium content of the cerebrospinal fluid on spinal reflex activity in the dog. Amer. J. Physiol. 131: 67, 1940.
131. Moyer, C. A. and Beecher, H. K.: Central stimulation of respiration during hypoxia. Amer. J. Physiol. 136: 13, 1942.
132. Mullin, F. J., Hastings, A. B. and Lees, W. M.: Neuromuscular responses to variations in calcium and potassium concentrations in the cerebrospinal fluid. Amer. J. Physiol. 121: 719, 1938.
133. Munck, P. de: Symptômes d'urémie par injection intracisternale de liquide céphalo-rachidien d'urémiques ches le chien. Schweiz. med. Wschr. 79: 907, 1949.
134. Nicholson, H. C. and Sobin, S.: Respiratory effects from the application of cocaine, nicotine and lobeline to the floor of the fourth ventricle. Amer. J. Physiol. 123: 766, 1938.
135. Noell, W. and Kornmüller, A. E.: Zur Sauerstoffmangelwirkung auf die Hirnrinde: eine bioelektrische Untersuchung. Pflüg. Arch. ges. Physiol. 247: 685, 1944.
136. Palmer, A. C.: Injection of drugs into the cerebral ventricle of sheep. J. Physiol. 149: 209, 1959.
137. Reitter, H.: Narkoseeffekt durch intrazisternale Adrenalin-Injektion. Anaesthesist 6: 131, 1957.

138. Resnik, H., Mason, M. F., Terry, R. T., Pilcher, C. and Harrison, T. R.: The effect of injecting certain electrolytes into the cisterna magna on the blood pressure. Amer. J. med. Sci. 191: 835, 1936.
139. Rothballer, A. B.: The effects of catecholamines on the central nervous system. Pharmacol. Rev. 11: 494, 1959.
140. Sack, H., Simon, K. and Wild, H.: Zur Frage der zentralnervösen Regulation des Kohlenhydratstoffwechsels. Schweiz. med. Wschr. 79: 1030, 1949.
141. Salama, S. and Wright, S.: Action of d-tubocurarine chloride on the central nervous system of the cat. Brit. J. Pharmacol. 5: 49, 1950.
142. Salama, S. and Wright, S.: Action of calabash curare and related curariform substances on the central nervous system of the cat. Brit. J. Pharmacol. 6: 459, 1951.
143. Salama, S. and Wright, S.: Action on central nervous system of compounds RP 3565 and RP 3697 and of tetramethylammonium and tetraethylammonium. Brit. J. Pharmacol. 7: 1, 1952.
144. Salama, S. and Wright, S.: Influence of various drugs on the action of curare on the central nervous system of the cat. Brit. J. Pharmacol. 7: 14, 1952.
145. Schaltenbrand, G.: Plexus und Meningen. Handb. mikrosk. Anat. 4: pt. II, 1955.
146. Sherwood, S. L.: The response of psychotic patients to intraventricular injections. Proc. R. Soc. Med. 48: 855, 1955.
147. Smolik, E. A.: Effect of intracisternal injection of potassium phosphate in hemorrhagic hypotension and shock in the dog. Proc. Soc. exp. Biol., N. Y. 53: 70, 1943.
148. Smolik, E. A.: Observations on the intracisternal injection of potassium phosphate in the dog. Surgery 15: 460, 1944.
149. Stern, L.: Le Liquide céphalorachidien au point de vue de ses rapports avec la circulation sanguine et avec les éléments nerveux de l'axe cérébrospinal. Schweiz. Arch. Neurol. Psychiat. 8: 215, 1921.
150. Stern, L.: Shock treatment by direct action on vegetative nerve centres. Lancet 2: 572, 1942.
151. Stern, L.: Treatment of shock by direct action on the vegetative nervous centres. Brit. med. J. 2: 538, 1942. (Same subject.)
152. Stern, L. and Chvoles, G.-J.: Effet de l'injection intraventriculaire des ions Ca et K. C. R. Soc. Biol., Paris 112: 568, 1933.
153. Stern, L. and Gautier, R.: Recherches sur le liquide céphalo-rachidien. I. Les rapports entre le liquide céphalo-rachidien et la circulation sanguine. Arch. int. Physiol. 17: 138, 1921/2.
154. Stern, L. and Gautier, R.: Recherches sur le liquide céphalo-rachidien. II. Les rapports entre le liquide céphalo-rachidien et les éléments nerveux de l'axe cérébrospinal. Arch. int. Physiol. 17: 391, 1921/2.
155. Stern, L. and Gautier, R.: Recherches sur le liquide céphalo-rachidien. III. Rapports entre le liquide céphalo-rachidien des espaces ventriculaires et celui des espaces sous-arachnoidiens. Arch. int. Physiol. 20: 403, 1922/3.
156. Stern, L., Rossine, J.-A. and Chvoles, G.-J.: Le mécanisme d'action du K et du Ca injectés dans les ventricules cérébraux. C. R. Soc. Biol., Paris 114: 674, 1933.
157. Stern, L. and Rothlin, E.: Effets de l'application directe du curare sur les différentes parties du cervelet. Schweiz. Arch. Neurol. Psychiat. 3: 234, 1918.
158. Stöhr, P.: Über die Innervation der Pia mater und des Plexus chorioideus des Menschen. Z. Anat. EntwGesch. 63: 562; 64: 555, 1922.
159. Suh, T. H., Wang, C. H. and Lim, R. K. S.: Effect of intracisternal injections of acetylcholine. Proc. Soc. exp. Biol., N. Y. 32: 1410, 1935.
160. Verstraeten, J. M.: Influences centrales du calcium et du potassium sur la respiration. Arch. int. Pharmacodyn. 77: 52, 1948.
161. Verstraeten, J. M.: Influence de la concentration en calcium, potassium et magnésium du liquide céphalo-rachidien sur la consommation d'oxygène. Experientia 5: 251, 1949.
162. Verstraeten, J.-M.: Influences de la concentration en ions potassium, calcium et magnésium du liquide céphalo-rachidien sur la respiration. Revue Belge Path. 20: 1, 1950. (Same subject as 160.)
163. Verstraeten, J.-M.: Influence de la concentration en ions calcium, potassium et magnésium dans les ventricules cérébraux sur la consommation d'oxygène. Revue Belge Path. 20: 22, 1950. (Same subject as 161.)
164. Vleeschhouwer, G. de: Au sujet de l'action du diéthylaminométhyl-3-benzodioxane (F 933) et du pipéridométhyl-3-benzodioxane (F 933) sur le système circulatoire. Arch. int. Pharmacodyn. 50: 251, 1935.
165. Voetmann, E.: On the structure and surface area of the human choroid plexuses. Acta anat. 8: suppl. 10, 1949.
166. Vuylsteke, Ch. A.: Glycorachie et glycémie. Arch. int. Pharmacodyn. 78: 456, 1949.
167. Walker, S. M., Smolik, E. A. and Gilson, A. S., Jr.: The effects of intracisternal injection of potassium phosphate on the rate and rhythm of the heart and on the blood pressure and on the respiration of the dog. Amer. J. Physiol. 145: 223, 1945.
167a. Wang, S. C. and Borison, H. L.: A new concept of organisation of the central emetic mechanism: recent studies on the sites of action of apomorphine, copper sulfate and cardial glycosides. Gastroenterology 22: 1, 1952.
167b. Weiland, H. I., Broh-Kahn, R. H. and Mirsky, I. A.: Lack of a hypoglycemic response to intrathecal injection of glucose. Proc. Soc. exp. Biol., N. Y. 67: 171, 1948.
168. Wiemer, W.: Die Wirkung von Chemoreseptoren-Reizstoffen vom Liquor aus. Z. Biol. 111: 287, 1959.
169. Winder, C. V., Winder, H. O. and Gesell, R.: The seat of action of cyanide on pulmonary ventilation. Amer. J. Physiol. 105: 311, 1933.
170. Winiwarter, F.: Über die pharmakologische Analyse des Ohrkratzreflexes. Arch. int. Pharmacodyn. 62: 42, 1939.

171. WINTERSTEIN, H.: The "reaction theory" of respiratory regulation. Experientia 5: 221, 261, 1949.
172. WINTERSTEIN, H.: Die chemische Steuerung der Atmung. Ergebn. Physiol. 48: 328, 1955.
173. WINTERSTEIN, H.: Chemical control of pulmonary ventilation. Dunham Lectures. New Engl. J. Med. 255: 216, 272, 331, 1956.
174. WINTERSTEIN, H.: Die Blut/Hirn-Schranke. Acad. of Sciences of the Georgian SSR, 1956.
175. WINTERSTEIN, H.: Probleme der Atmungsfunktion im Lichte neuerer Forschung. Ärztl. Praxis 10: 1045, 1062, 1958; Pflüg. Arch. ges. Physiol. 268: 16, 1958.
176. WINTERSTEIN, H.: Die Entdeckung neuer Sinnesflächen für die chemische Steuerung der Atmung. Naturwissenschaften 47: 99, 1960.
177. WINTERSTEIN, H. AND FRÖMTER, E.: Die Wirkung der Vagi und des O_2-Mangels auf das Atmungszentrum. Z. Biol. 111: 421, 1960.
178. WINTERSTEIN, H. AND FRÖMTER, E.: Der zentrale Angriffspunkt der Vagushemmung. Z. Biol. 112: 58, 1960.
179. WINTERSTEIN, H. AND GÖKHAN, N.: Ammoniumchlorid-acidose und Reaktionstheorie der Atmungsregulation. Arch. int Pharmacodyn. 93: 212, 1953.
180. WINTERSTEIN, H. AND GÖKHAN, N.: Chemoreseptoren-Reizstoffe und Blut/Hirn-Schranke. Arch. exp. Path. Pharmak. 219: 192, 1953.
181. WINTERSTEIN, H. AND WIEMER, W.: Die Wirkung von Lobelin und Natriumcyanid bei suboccipitaler Injektion. Arch. exp. Path. Pharmak. 235: 235, 1959.