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# LYMPHATIC CONNECTIONS BETWEEN THE NASAL MUCOSA AND INTRACRANIAL SPACE

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It is known from clinical experience that during anaesthesia for surgery on the nasal septum vertigo, sweating, or eventually loss of consciousness may occur. It is also well-known that common cold is often associated with headache. In single cases of retrobulbar neuritis recovery was achieved by painting cocaine on the nasal mucous membrane (GABÁNYI [4]). REAGEN et al. [13] have reported that the intranasally instilled Newcastle disease virus produced symptoms similar to those seen after intracerebral administration. These facts made us to study the existence of an eventual close connection between the nasal and the cranial cavity.

As early as 1875, KEY and RETZIUS [9] demonstrated the connection between subarachnoidal space and nasal lymphatics, by injecting a dye into the CSF space. IVANOW and ROMODANOWSKY [7] injected China ink into the subarachnoidal space of live and dead dogs and found considerable amounts of the dye in the nasal mucosa. The experiments of SCHWALBE [14] also proved the existence of a circulatory connection between subarachnoidal space and nasal mucosa; his results, however, could not be reproduced by QUINCKE [12].

The divergence in results appears to have been due to differences in the molecular size of the various dyes used. According to WEED [17], ferri ammonium citrate and potassium ferrocyanide injected into the cerebrospinal fluid appears in the perineural space of cranial nerves, in the cervical lymph nodes and in the nasal mucosa. TCHOUDNOSOVYETOV [3] has obtained evidence in live animals and fresh cadavers concerning the existence of connections between subarachnoidal space and nasal cavity. These findings have been substantiated by the results reported by SPERANSKY [15]. In contrast with the retrograde experiments, VENDÉG [16] used the so-called anterograde technique in cadavers and found that methylene blue injected into the nasal mucosa appeared in the base of the skull, expecially so in the region of the sella turcica, lamina cribrosa and clivus. BAUM and TRAUTMANN [2] have demonstrated in detail the lymphatic system of the nasal cavity in live and dead horses, dogs, calves, pigs and suggested that the nasal lymphatics may be connected with the intracranial space.

The mentioned data originate from studies made partly in live, and partly in dead subjects. In live subjects exclusively the retrograde injection technique has been employed for demonstrating the communication between intracranial space and nasal mucosa. Therapeutic considerations have made it necessary to demonstrate the existence of this connection in the live subject, by the anterograde technique. An account of the first part of our investigations has been given at the 20th Meeting of the Hungarian Physiological Society, in 1954 [10].

### Materials and methods

Albino rats, rabbits and cats were studied.

In the rat experiments, 65 albino rats were involved. Under ether or Evipan anaesthesia, a hypodermic needle was introduced into the nose and China ink or trypan blue, diluted 1 : 10, was injected under the nasal mucosa, in a volume of ½ ml. In the 10 control experiments the dyes were injected either intravenously (into the femoral or the tail vein), or were administered into the carotis communis artery. Both the controls and the test animals were killed within 15 to 120 minutes after injection. To avoid passing of the needle through the nasal cavity, and injection of the dye directly into the brain we have determined the optimum site of injection. This was found to be in the central region of the nasal septum, 12 mm from the nostril, taking the distance betwen nostril and lamina cribrosa to be 22 mm. A relatively large volume (500 cu. mm) of dye had to be applied to provide constant pressure. During injection pressure did not exceed 100 mm water. In a few cases pressure was measured by a physicist. The animals were killed by giving an excess of anaesthetic or by exposure to coal gas. The nasal septum, the walls of the nasal cavity and the brain were fixed in 4 per cent formaldehyde, Carnoy's fluid, or 80 per cent alcohol. The specimens were then either embedded in paraffine, or were frozen, cut up into sections and stained with haematoxylin-eosin, azan, or carmine.

In 5 rabbits we used a similar method, whereas in 2 rabbits frontal craniotomy was performed under Evipan anaesthesia and aseptic conditions. The frontal lobe was raised,1, 5 ml. China ink was injected into the septal mucosa and the coloration of the base of the skull was examined. In cats the method was similar to that described for rats.

## Results

*Gross findings*

After injection into the *anterior* one-third of the nasal septum the dye appeared in the anterior one-third of the septum, in the mucosa of the conchae nasi, of the lateral wall of the nasal cavity, as well as in the submandibular, sublingual, para- and retropharyngeal, cervical lymph nodes and in the two tonsillae palatinae. No coloration was visible in the base of the skull and on the basal aspect of the brain.

After injection into the *middle* one-third of the septum, the septum turned black in an area reaching to the lamina cribrosa, just as it happened with the scala anterior, particularly with the lamina cribrosa. On the basal surface of the brain, along the olfactory lobe, on both sides of the chiasma opticum and hypothalamus, up to the pons black lines were visible, just like in the two aspects of the bilateral frontal lobes facing each other (Fig. 1). In the rabbit gross examination after frontal craniotomy revealed the presence of a black coloration at the lamina cribrosa, the finer distribution of which could not be observed.

*Histological findings*

In stained preparations of the nasal septum the following were seen.

I. Respiratory region. On the surface there was multinucleated villous cylindrical epithelium below which in the tunica propria lymphocytes predominated. The tunica propria had no acute outlines toward the submucosa. In this area were found the mixed glands of the nasal mucosa. In deeper layers,


*Fig. 1.* Gross appearance of the basal aspect of the brain in the rat. The passage of China ink can be followed along the olfactory lobe, in the subarachnoidal cavum, on both sides of the hypothalamus, to the pons. Magnification, 1 : 5

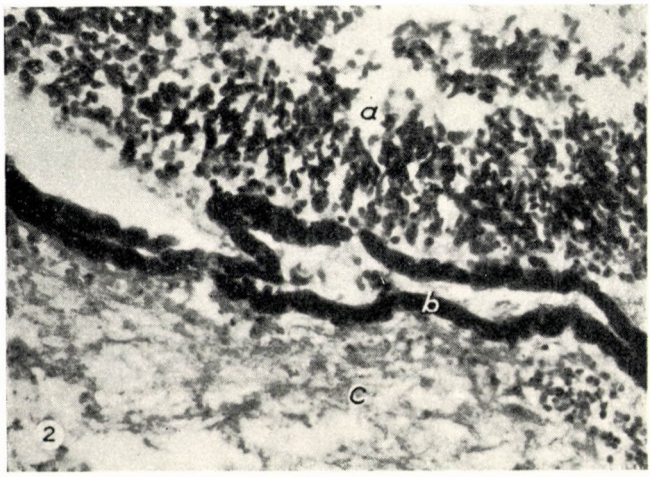


*Fig. 2.* Microphotographic appearance of the respiratory region of the nasal septum. *a)* Tunica propria. *b)* Lymph vessel filled with China ink. *c)* Submucosa. Haematoxylin-eosin, × 480

at the submucosal-perichondrial or periosteal junction, dilated venous sinuses were seen in loose connective tissue. Above the lymphocytic zone, as well as

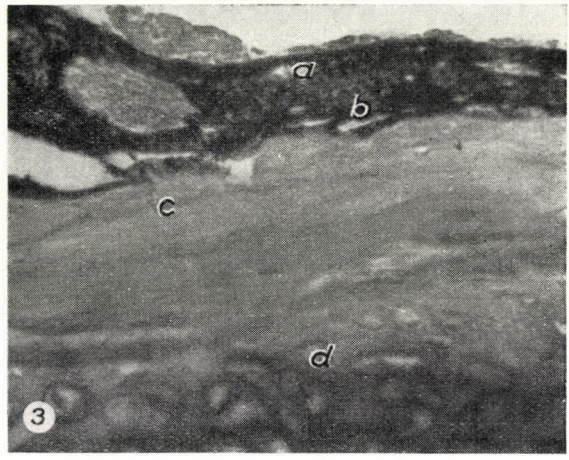


*Fig. 3.* Microphotographic appearance of the respiratory region of the nasal septum. *a)* System of lymphvessels under tunica propria. *b)* System of lymphvessels between submucosa and perichondrium. *c)* Perichondrium. *d)* Cartilage. The network of communication between the two lymphatic systems is visible in the submucosa. Haematoxylin-eosin, × 250

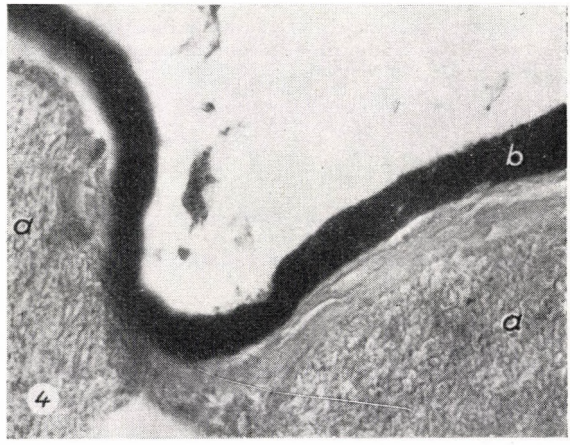


*Fig. 4.* Microphotographic appearance of the olfactory region of the nasal septum. *a)* Olfactory lobe. *b)* China ink perineurally. Haematoxylin-eosin, × 480

at the perichondrial or periosteal and submucosal junction the dye appeared in the lymph plexi. The two layers communicated in the form of a network surrounding the end chambers of glands and the veins, but no dye was visible either in the lumina of glands or in the veins (Figs. 2, 3).

II. Olfactory region. In this region the dye was distributed mainly as in the respiratory region. It was also found along the fila olfactoria, perineurally (Fig. 4).

III. Subarachnoidal cavity. The brain complete with leptomeninx was worked up systematically. In the subarachnoidal space the dye was detectable both on the basal and the convex aspect. From the subarachnoidal space the passage of dye into the brain could be followed alone the vessels (Fig. 5). But

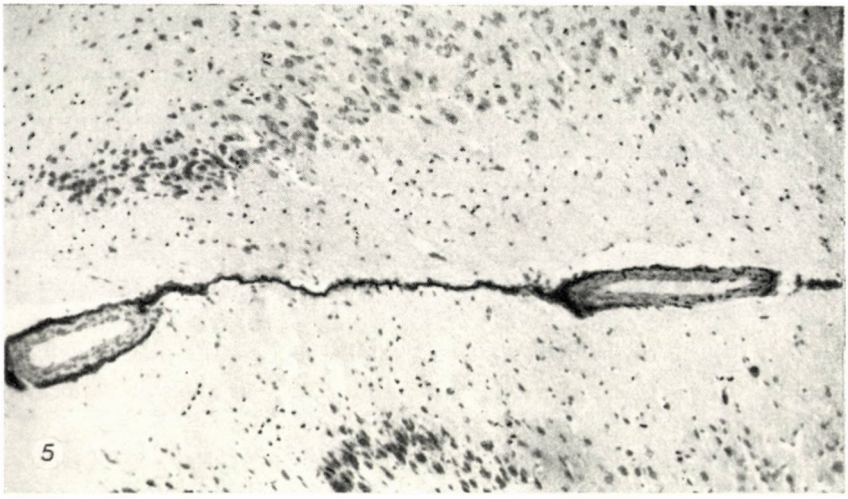


*Fig. 5.* Microphotographic appearance of a section cut parallel with the lamina cribrosa, from the frontal lobe. Perivascularly China ink is visible. Haematoxylin-eosin, × 120

from the cerebral matter proper it was only in the hypothalamus that dye could be demonstrated. Here fine China-ink granules were found extracellularly. To exclude interference by formaldehyde crystals, specimens fixed in 80 per cent alcohol and Carnoy's fluid were also examined.

*Control tests*

After intraarterial and intravenous injection of the dye the brain turned black in colour. The dye was in the cerebral blood vessels ; no dye was demonstrable in the cerebral matter, or subarachnoidally.

## Discussion

The evidence obtained confirmed the results of other authors, according to which a lymphatic connection exists between nasal mucosa and intracranial cavity. As regards these results, they were gained partly on dead material

(VENDÉG [16], BAUM and TRAUTMANN [2], KEY and RETZIUS [9], SPERANSKY et al. [15]), and partly on live animals (BAUM and TRAUTMANN [2], SPERANSKY et al. [15], HOFFMANN and THIEL [20], YOFFEY [18, 19]). The method used for unmasking the communication was mostly the retrograde one, i. e. perfusion from the subarachnoidal and subdural spaces. However, the results were not unequivocal. The cause of this has been suggested to be in the variability of CSF pressure in the subarachnoidal space (SPERANSKY et al [15]). This statement calls attention to two factors. 1. It is known that if CSF pressure is increased, for example during an epileptic seizure, the CSF is conducted also toward the nasal cavity, thus injection of dye into the subarachnoidal space gives rise to a pathological condition. 2. The pressure of the CSF must overcome the resistance of the valves in the nasal lymph vessels. This is shown, among others, by the fact that in cases with low pressure only little, or even no, dye appears in the nasal cavity. For this reason perfusion from the nasal mucosa seems to be more physiological. In perfusion from the nasal mucosa pressure is not the sole factor. This was shown by the investigations of KROMPECHER et al. [10], who found that after introducing a cotton swab with streptomycin on the septum of normal children the resulting concentration of streptomycin in the CSF was much higher than after parenteral administration. The investigations with this anterograde mode of administration yielded information as to the optimum site of injection. BAUM and TRAUTMANN [2] already made attempts at dividing the nasal cavity into areas with regard to lymph outflow. Our studies in rats revealed that the lymphvessels in the anterior one-third of the nasal septum communicate first of all with the cervical, submandibular, para- and retropharyngeal lymph nodes, whereas they are in no direct communication with the intracranial cavity. In the middle third is found the most dense lymphatic network, located in the area of the tunica propria next to the epithelium and especially in the submucosa. These networks communicate with each other and extend toward the lamina cribrosa. According to YOFFEY [18, 19], as well as HOFFMANN and THIEL [20], this lymphatic network communicates with the perineural spaces, creating thereby the connection between nasal mucosa and intracranial cavity. We have been unable to confirm the existence of a connection between the venous and lymphatic networks claimed by SPERANSKY [15] and by BAUM und TRAUTMANN [2]. No dye was visible in the lumina of vessels 15 to 60 minutes after injection. However, the dye did appear in the lymphatic spaces of the conchae nasales, too. From the hind third of the septum the outflow toward the intracranial cavity is mechanical ; in this the perineural route appears to be also involved beside the flow through the lymphatics. The lymphvessels of the nasal mucosa pass through the lamina cribrosa and empty into the subarachnoidal space. This is confirmed by the variable intensity of coloration of the nasal mucosa after injecting the dye into the subarachnoidal or subdural space.

From the subarachnoidal space the dye enters the cerebral matter alongside the cerebral blood vessels. If the dye granules are small, the brain is probably entered also by diffusion, or *via* the plasma bridges described by LEONHARDT [11]. The dyes used by us appeared extracellularly in the basal part of the hypothalamus but no elsewhere in the brain. This phenomenon is due to the well-known difference in permeability existing between the hypothalamus and other parts of the brain (BAKAY [1], HEVESY [5,6]).

Accordingly, in our experiments the dye was demonstrable in the brain. The methods used by us revealed granules of dye only in the hypothalamus. Nevertheless, the possibility is given to carry out not only qualitative, but also quantitative studies in other parts of the brain by some improved technique.

**Summary**

In view of its practical significance, we have subjected to study the lymphatic communication between nasal septum and intracranial space in 65 albino rats, 7 rabbits and 3 cats. 10 control experiments were also carried out. China ink and trypan blue, injected under the mucosa of the nasal septum, were used for demonstrating communications. In paraffine-embedded or frozen sections, stained with haematoxylin-eosin, carmine or azan the following were found.

1. The dye injected into the anterior one-third of the septum appeared in the submandibular, sublingual, parapharyngeal lymph nodes and in the tonsilla palatina.
2. The dye injected into the middle third of the septum was found in
   *a)* the lymphatic network of the nasal septum,
   *b)* the subarachnoidal space,
   *c)* in the hypothalamus, extracellularly.
3. The dye was never demonstrable in blood vessels.
4. Communication is by the following routes:
   *a)* Direct lymphatic connection from the nasal mucosa to the subarachnoidal space.
   *b)* Perivascularly.
   *c)* Perineurally.

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## СВЯЗЬ МЕЖДУ ЛИМФООБРАЩЕНИЕМ СЛИЗИСТОЙ ОБОЛОЧКИ НОСА И ЧЕРЕПНОЙ ПОЛОСТЬЮ

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Ввиду большого значения для практики, авторы исследовали связь между лимфообращением носовой перегородки и черепной полостью. Подопытными животными обыли: 65 белых крыс, 7 кроликов и 3 кошки. Одновременно проводились также и 10 контрольных экспериментов. Для выявления связи авторы применяли тушь, трипановую синьку, вводя эти краски под слизистую оболочку носовой перегородки. На залитых в парафин или же замороженных срезах получались после окрашивания гематоксилин-эозином, кармином или азокрасителем следующие результаты:

1. Введенные в переднюю треть носовой перегородки краски появляются в субмандибулярных, подъязычных, парафарингеальных лимфатических узлах и в небной миндалине.
2. Введенные во вторую треть носовой перегородки краски обнаруживаются:
   *а)* в лимфатической системе носовой перегородки,
   *б)* в подпаутинном пространстве,
   *в)* в гипоталамическом веществе внеклеточно.
3. В кровеносных сосудах появления краски никогда не наблюдалось.
4. Связь осуществляется несколькими путями:
   *а)* непосредственным лимфатическим путем исходящим из слизистой носа в подпаутинное пространство,
   *б)* периваскулярно,
   *в)* периневрально.

## VERBINDUNG ZWISCHEN DEM LYMPHKREISLAUF DER NASENSCHLEIMHAUT UND DEM DER SCHÄDELHÖHLE

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Es wurden — in Anbetracht ihrer praktischen Bedeutung — die Verbindungen zwischen dem Lymphkreislauf der Nasenscheidewand und jedem der Schädelhöhle an 65 weissen Ratten, 7 Kaninchen und 3 Katzen untersucht. Es wurden 10 Kontrollversuche vorgenommen. Um die Verbindung zu bezeichnen, wurde Tusche oder Trypanblau unter die Schleimhaut der Nasenscheidewand injiziert. An Paraffin- oder Gefriersschnitten wurde nach Haematoxylin-Eosin, Karmin oder Azanfärbung das folgende beobachtet:

1. In das erste Drittel der Nasenscheidewand eingeführter Farbstoff erscheint in den submandibulären, sublingualen und parapharingealen Lymphknoten und in der Gaumentonsille.

2. Der in das zweite Drittel der Nasenscheidewand injizierte Farbstoff erscheint :
   *a)* im Lymphgefässnetz der Nasenscheidewand,
   *b)* im Cavum subarachnoideale,
   *c)* extrazellular im Hypothalamus.
3. In den Blutgefässen wurde niemals Farbstoff beobachtet.
4. Die Verbindung kommt auf mehreren Wegen zustande :
   *a)* direkter Lymphweg ausgehend von der Nasenschleimhaut im Cavum subarachnoideale,
   *b)* perivaskulär,
   *c)* perineural.

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