

Det Kgl. Danske Videnskabernes Selskab.

Biologiske Meddelelser **I**, 10.

INVESTIGATIONS ON THE SPIROPTERA CANCER IV

SPIROPTERA CANCER OF THE TONGUE IN RATS

BY

JOHANNES FIBIGER

WITH FOUR PLATES



KØBENHAVN

HOVEDKOMMISSIONÆR: ANDR. FRED. HØST & SØN, KGL. HOF-BOGHANDEL

BIANCO LUNOS BOGTRYKKERI

1918

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It is a well-known fact that cancer of the tongue is a disease of frequent occurrence in man, whereas, on the other hand, only a very few cases are known from the veterinary pathology. As regards wild living animals our knowledge of the occurrence of cancer on the whole is but very limited, and it may possibly be owing to this fact that no observation has been made of cancer in the tongue of these animals. On the other hand, cancer in domestic animals has been subjected to such extensive investigation that no doubt can exist now about cancer of the tongue being a disease of unfrequent occurrence amongst domestic animals, as it has been emphasized in 1889 by PLICQUE¹, in 1908 by ROGER WILLIAMS², in 1910 by PETIT³, and in 1917 by FØLGER⁴.

When in 1902 STICKER⁵ published his great statistics comprising more than 1200 cases of cancer in the horse, cow, swine, sheep, goat, cat, and dog, only one case of carcinoma of the tongue in a cat observed in 1890 by Mc.FADYEAN⁶ had evidenced that this kind of tumor may occur at all in animals.

¹ Les tumeurs chez les animaux. *Revue de chirurgie*. 9. année 1889.

² *The Natural History of Cancer*. 1908.

³ Généralités sur les tumeurs malignes des animaux domestiques. 11. Conférence internationale pour l'étude du Cancer. Paris 1910.

⁴ Geschwülste bei Tieren. *Lubarsch-Ostertag's Ergebnisse*. XVIII. Jahrg. 1917.

⁵ Über den Krebs der Tiere. *Archiv f. klin. Chirurgie*. Bd. 65. 1902.

⁶ Carcinoma of a Cat's Tongue. *The Journ. of Comparative Pathology and Therapeutics*. III. 1890.

The number of cases observed since then hardly exceeds 10, 4 of which have been found in the horse (GILRUTH¹, WIRTH², JOEST³, FÖLGER⁴), 2 in the cow (NAUTA⁵, GEHRIG⁶), 2 in the dog (BGHL⁷, MURRAY⁸) and 2 in the cat (BASHFORD⁹, MURRAY¹⁰).

The rarity of the disease does not, however, stand out in its proper light, before we have taken into consideration the enormous material and the numerous investigations, carried on for years and years, upon which the statements are based.

Thus, e. g. in the veterinary high school of Berlin alone more than 200,000 distempered horses were used for STICKER'S calculation of the frequency of cancer in this animal. PETIT'S statement of never having met with carcinoma of the tongue in domestic animals was based upon observations carried out for 12 years at the veterinary high school in Alfort, and FRØHNER¹¹ examined more than 60,000 dogs without pointing out cancer of the tongue, whereas this great material included no less than 1154 cases of carcinomatous tumors.

Nor has cancer of the tongue been traceable in birds,

¹ Report of the Principal Veterinary Offices. New Zealand Department of Agriculture. 1902—1903. Quoted by FÖLGER.

² Oesterreich. Monatsschrift f. Tierheilkunde. XXXV. 1910.

³ Ulzerirtes Zungenkarzinom beim Pferde. Bericht der Kgl. tierärztlichen Hochschule zu Dresden f. d. Jahr 1910. Dresden 1911.

⁴ Ulcererende Kankroid paa Tungeryggen hos en Hest. Landbohøjskolens Aarsskrift. 1917.

⁵ Carcinom der Zunge des Rindes. Tierärztliche Rundschau. Bd. XII. 1906.

⁶ Beitrag zur Kasuistik der Neubildungen des Rindes. Dissertation. Giessen 1912.

⁷ Zur Kasuistik der Neubildungen bei den Tieren. Abhandlungen des Kasan'schen Veterinärinstituts. Bd. XXI. Ref. in Ellenberger-Schütz's Jahresberichte f. d. J. 1909.

⁸ The Zoological Distribution of Cancer. Scientific Report on the Investigations of the Imperial Cancer Research Fund III. 1908.

⁹ Scientific Report on the Investigations of the Imperial Cancer Research Fund I—III. 1904—1908.

¹⁰ loc. cit.

¹¹ Monatsschrift f. praktische Tierheilkunde IV. 1895.

although the occurrence of cancer in domestic fowls has been subjected to wide-ranging investigations.

On the other hand, 3 observations (PICK¹, KOCH², JOEST and ERNESTI³) have been made of carcinoma of the oral cavity in poultry. Also among domestic mammals tumors of this kind have been met with, according to WILLIAMS, altogether in 16 cases, located principally in the mucous membrane of the jaws.

Also the extensive series of investigations which for the last decennium during the era of experimental cancer research have been devoted to the study of spontaneous carcinomatous growth in mice and rats are illustrative of the unfrequent occurrence of cancer of the tongue in animals.

Most extensive, of course, is the casuistry of the tumors in mice. As an example be it stated that in the Cancer laboratory (superintended by BASHFORD) of the Imperial Cancer Research Fund such wide-ranging experiments have been performed on mice for series of years that the stock of mice under simultaneous observation might sometimes comprise about 20,000 animals⁴. It required 4 years and the examination of 100,000 mice to discover the occurrence of 28 cases of spontaneous carcinoma of the mamma, and in 1906, only in investigations on the transplantability of JENSEN'S tumor 32,000 mice had been inoculated⁵. In the laboratories of BASHFORD and EHRLICH about 2,000 cases of spontaneous cancer in mice had been observed in 1913 for a period of some 10—12 years (APOLANT⁶, BASHFORD), and in 1917 another 2,000 sponta-

¹ Grosser Plattenepithelkrebs des Mundhöhlenbodens bei einem Huhn. Berliner klin. Wochenschrift. 40. 1903.

² Verhandlungen der Deutschen pathologischen Gesellschaft 1904.

³ Untersuchungen über spontane Geschwülste bei Vögeln. Zeitschrift f. Krebsforschung. Bd. XV. 1915.

⁴ Berliner klin. Wochenschrift. 1913. No. 1.

⁵ Fourth Annual Report of the Imp. Cancer Research Fund.

⁶ Die experimentelle Erforschung der Geschwülste. KOLLE-WASSERMANN'S Handbuch der pathogenen Mikroorganismen. III.

neous tumors in mice had been examined by SLYE, HOLMES and GIDEON WELLS¹.

Nevertheless, neither from these cancer institutes nor from other cancer laboratories has information been obtained as to cancer of the tongue occurring in mice, whereas carcinomatous growth taking its rise from mucosa of gingiva has been observed in a smaller series of cases (BORREL, HAALAND²).

Nor have observations been made of cancer in the rat's tongue. Although casuistry of the spontaneous tumors in rats is, to be sure, far less copious than is the casuistry of those in mice, the reports hitherto published are based upon a considerable number of examinations performed partly on white and black laboratory rats, partly on wild rats, procured by the systematic extermination of rats established in recent years. In MC. COY'S³ statistics comprising 100,000 wild rats (*Mus decumanus*, *M. rattus* and *M. alexandrinus*) allusion is not made to cancer of the tongue, any more than in WOOLEY'S and WHERRY'S⁴ report including 23,000 rats, nor in BORREL'S statement of the investigations carried out by BRIDRÉ on 8,000 rats.

According to information kindly given by the director of the Danish Ratin Laboratory, Mr. L. BAHR, no cancer of the tongue has been met with by postmortem examination of about 6,000 wild rats (*M. decumanus*) for ten years past. And Professor C. O. JENSEN too has told me that he has never observed cancer of the tongue in rats.

As a matter of course it cannot be precluded that cancer of the tongue may have been overlooked in examinations

¹ Studies on the Incidence and Inheritability of Spontaneous Tumors in Mice. *The Journal of Cancer Research*. 1917.

² Les tumeurs de la souris. *Annales de l'Institut Pasteur*. 1905. Spontaneous Tumors in Mice. Fourth Scientific Report of the Imperial Cancer Research Fund. 1911.

³ A Preliminary Report on Tumors Found in Wild Rats. *The Journal of Med. Research*. 1909.

⁴ Notes on Twenty-two Spontaneous Tumors in Wild Rats. *The Journal of Med. Research*. 1911.

which were not carried out for the special purpose of pointing out tumors of this kind; it should therefore be added that I myself while examining for the last ten years the tongues of about 2,000 rats (*M. decumanus*, *M. rattus* and black and white laboratory rats of different origin) have found no other cases of tumor of the tongue than those which will be recorded in this and in a later ensuing paper.

A series of experimental investigations have been carried out by H. STAHR who in a very short note of 1903¹ announced that, by feeding rats merely on oats, he had succeeded in producing development of fibro-epithelial tumor-like growths (Fibro-epithelioma) in radix linguæ of these animals. Destructive growth could not, however, be traced, any more than metastasis formation, and the tumor-like formations therefore were not interpreted as real carcinoma, but only as an approach to a real Blastoma of epithelial origin, as a stage preceding an Epithelioma malignum. In 1908—1909 I imitated these experiments of STAHR in a small number of animals, but only succeeded in provoking moderate ulcerating and inflammatory processes, which, according to my view, may most probably be due to a too short duration of my experiments. About this, however, I could at that time, not possibly know anything, as the report by STAHR contained no statements whatever as to further details. In a letter to STAHR 1914 I mentioned the negative results of my unpublished investigations from 1908—1909, which induced STAHR to issue in 1915² a great detailed report of his old experiments, quoting my short remarks without having applied to me for further particulars, and making various reflections — to some extent founded on the communications of my letter — on differences in predisposition. In a shorter communication in the "Centralblatt f. path. Anatomie

¹ Zur Aetiologie epithelialer Geschwülste. Centralbl. f. path. Anatomie u. allg. Pathologie. Bd. XIV. 1903.

² Durch andauernde Haferfütterung erzeugtes Epitheliom der Rattenzunge. Ziegler's Beiträge. Bd. 61.

and allg. Pathologie"¹ I have opposed STAHR, and shall not enter further into these examinations, as K. SECHER has given a detailed report of them², having at my request imitated and supplemented STAHR's experiments.

A short report of experimental investigation is due to YUTAKA KON² who in rabbits fed on lanoline has observed papillomatous growth on the lips and in mucosa of the mouth and of the tongue. In these experiments too, carcinoma did not occur.

In a lecture delivered at the Meeting of Scandinavian Naturalists in Kristiania 1916 and in my paper quoted above, I mentioned that during further investigations on the influence of the *Spiroptera neoplastica* (*Gongylonema neoplasticum*) upon rats, 4 cases of cancer of the tongue had been observed. Preparations of the carcinomatous tongues were exhibited at the meeting. These cases and one more, produced experimentally by feeding rats on oats in SECHER's experiments mentioned above, are the first cases of cancer of the tongue produced experimentally, and — as far as I know — on the whole the first cases of cancer of the tongue observed in rats. In rats infected with the Spiroptera I have later on met with 1 more case, and 1 case of papillomatous growth of the tongue; these cases and the 4 formerly observed will be described in the following; furthermore, carcinoma of the tongue has been observed in a series of cases in rats, fed on oats and barley, and finally, 1 similar case has occurred, the causal conditions of which are possibly lesions from deviated teeth. These last cases will be recorded in a later ensuing paper.

¹ Bd. 27. 1916.

² Hospitalstidende 1918, Nr. 43.

³ Weitere Mitteilung über die Adenom- und Papillombildung bei mit Lanolin gefütterten Kaninchen. "Gann". Bd. XI. 1917.

In my earliest reports¹ I briefly noted that, in some rats infected with the Spiroptera, not only the fundus of the stomach (or the fundus and the gullet simultaneously), but also the tongue was found to be the seat of inflammatory processes and epithelial hyperplasia due to the invasion of the nematode. In all cases observed the disease of the tongue was but slightly and far less pronounced than the corresponding changes of the fundus of the stomach in most of the cases. Papillary inflammations were not met with, neither was carcinoma or tumorlike growth.

In 1914, while beginning experiments reported in a preceding paper², I observed new cases of glossitis, most frequently in black and white rats which were not fed on cockroaches harbouring the nematode, but infected with free Spiroptera larvæ injected into their stomach or placed in their mouth. Some animals exhibiting more violent changes of the tongue made me subject this Spiroptera glossitis to a more thorough study.

For these investigations not only a considerable part of the experimental material of black and white rats — dealt with in the preceding paper² — and belonging to the proper stock of the institute, were used, but also black and white rats from London, some bastards of the Norway rat and black and white rats, furthermore black and white rats originating from other medical institutes of the University of Copenhagen. Altogether 217 animals were used, a smaller number of which were killed, whereas the great majority died spontaneously. Only white bread was used as feeding material.

The experiments may be divided into various groups according to the different technique used by the transmission

¹ Académie Royale des Sciences et des Lettres de Danemark. Extrait du Bull. de l'année 1913. Hospitalstidende and Zeitschr. für Krebsforschung 1913 and 1914.

² Det Kgl. Danske Videnskabernes Selskab. Biologiske Meddelelser I, 1918.

of the Spiropterae. Among 61 rats (group I), infected by feeding on parts of cockroaches, affection of the tongue was met with in 13 cases, among 120 (group II) infected with free larvæ, placed in the mouth or injected into the stomach, affection of the tongue occurred in 42 cases. These figures, however, only indicate the frequency of the disease at the time when the rats died. As will be discussed later on, the frequency, no doubt, has actually been greater, as in most cases the disease of the tongue is of limited duration and often disappears before the death of the rats.

But the greater frequency of the affections amongst rats infected by transmission of free larvæ cannot be referred to a very early death of these rats, nor to a later death of the rats fed on cockroaches, as the same difference of frequency showed itself independent of the space of time passed after the transmission of the Spiropterae. It is easily understood that the tongue must be more exposed to invasion of the nematodes, when free larvæ are placed in the mouth, than if feeding on cockroaches is employed as, in the latter instance, only a minor part of the contents of larvæ in the muscles is liberated in the mouth, the majority being not free until getting under the influence of the stomach. Affection of the tongue, however, also occurred frequently when free larvæ were injected through a caoutchouk catheter directly into the stomach. This, no doubt, is due to the fact that with this technique a disgorgement of the injected emulsion will take place in several cases, and that by the removal of the catheter larvæ may be led through the gullet into the mouth.

As a third group (III) I have set up a smaller number of rats (25) on the tongue of which 10—25 larvæ were applied every second or third day throughout various longer periods. By the application of this method severe affections of the tongue were produced in 17 cases. All animals employed for

these experiments died before 3 months had passed after the first transmission of the larvæ.

In the table below I have arranged the result of these experiments parallel to those of the two first groups

Group I and II			Group III		Total:	
Days from transmission of the Spiroptera to death of the rat	Number of rats	Affections of the tongue	Number of rats	Affections of the tongue	Number of rats	Affections of the tongue
4—20 days	16	4 G.	9	3 G.	125	64 G. 1 C.
21—44 —	38	26 - } 48 G.	8	8 - } 16 G.		
45—89 —	46	18 - }	8	5 - 1 C. } 1 C.		
90—119 days	22	3 G. 1 C.			81	4 G. 3 C.
120—179 —	31	1 - 1 - } 4 G 3 C				
180 and longer time	28	0 - 1 - }				
Total	181	52 G. 3 C.	25	16 G. 1 C.	206	68 G. 4 C.

In these tables as in those accompanying my previous report¹, the rats are arranged into groups according to their time of survival after the transmission of the nematodes — and if more than one transmission was employed — after the first of these. Affection of the tongue is marked with the letters *G.* and *C.*

As previously¹ mentioned this way of grouping need not represent the absolutely exact dates of the invading of the Spiroptera in cases where repeated transmissions were used. I, therefore, have set up the following tables, the first of which (A) contains rats which have survived the invasion of the nematodes for a period the length of which falls absolutely within the figures limiting the groups; the second table (B) comprises rats to which the Spiroptera were only transmitted once, and whose time of survival can therefore be fixed with absolute certainty (see the following page).

As will be seen from the table above affections of the tongue (*G.* and *C.*) were observed in 72 out of 206 black and white rats.

¹ Del kgl. Danske Videnskabernes Selskab Biologiske Meddelelser I, 1918.

Table A			Table B	
Days from transmission of the Spiroptera to death of the rats	Number of rats	Affections of the tongue	Number of rats	Affections of the tongue
4—20 days	11	5 G. }	10	4 G. }
21—44 —	11	9 - } 20 G.	11	9 - } 15 G.
45—89 —	18	6 - }	13	2 - }
90—119 days	6	0 G. }	6	0 G. }
120—179 —	14	1 - 1 C. } 1 G. 2 C.	9	0 - } 0 G. 1 C.
180 and longer time	14	0 - 1 - }	10	0 - 1 C. }
Total	74	21 G. 2 C.	59	15 G. 1 C.

Considering the smallness of the figures the results of the control tables will be seen to be in very good keeping with those of the chief table, and the approximate correctness of the latter will thus be corroborated.

Furthermore, affections were found in one out of 11 bastards of the Norway rat and black and white laboratory rats, infected with free prepared Spiroptera larvæ.

The disease appeared as 2 different types, partly as glossitis (in the tables marked *G.*) partly as tumor growth (marked *C.*)

These two types of disease occurred with very different frequency according to the length of the period in which the rats survived the transmission (resp. 1st transmission) of the Spiroptera.

As will be seen from the tables (Groups I and II) nematodes and glossitis were found most frequently in rats which died 3—6 weeks after the transmission. Among rats which died before 3 months had passed, the frequency strongly approached 50 pCt.

In accordance with this, Group III demonstrates that among 25 rats (on the tongue of which Spiroptera larvæ were applied every second or third day for a longer period, and which all died within 3 months after the beginning of the transmissions), affection of the tongue was met with in 17 rats, in

16 cases as a glossitis, in 1 case as glossitis and tumor-growth.

Among 7 of the bastards mentioned above which died within the same space of time, tumor growth was observed in 1 case, while the tongue in the other 6 rats was healthy.

On the other hand, glossitis only occurred in a very few cases, altogether in 4, among rats which survived the transmission (resp. 1st transmission) of the Spiroptera for 3—6 months and longer time. Neither was tumorgrowth of frequent occurrence in these rats, but in so far more frequent than in the shorter-lived rats, as it was observed in 3 out of 85 animals¹. In the great majority of these rats the tongue was found to be normal, harbouring no nematodes, whereas in rats which died earlier, it was often the seat of so many parasites and such violent changes that in some cases death would be caused by obliteration of the gullet and aditus laryngis.

The unfrequent occurrence of the glossitis among the long-lived rats does not, however, mean that these rats were not infected at all; as mentioned above, it must in several cases be ascribed to a healing of the affection before the rats died from the Spiroptera disease in the fundus of their stomach, or from other illness. Although it has caused me much trouble, even by means of small laryngoscopes constructed for that special purpose, to trace out the glossitis in living rats (that is to say, not in cases where the lesions — as will seldom be the case — were strongly pronounced in the tip of the tongue and not alone located in radix) — I have succeeded in observing in a series of cases the beginning, advancing, and ceasing of glossitis, a shorter or longer time before death of the animals.

The chief locality of the glossitis is the posterior part of radix linguæ, situated directly in front of the epiglottis; here most frequently the changes will be more pronounced than in any other part of the tongue. The surface of the mucous

¹ 4 bastard rats without affections of the tongue included.

membrane is then far more rough than in normal tongues (s. Plate I fig. 1) often strongly wrinkled, knotted or furrowed, covered with hyperplastical desquamating whitish layers of epithelium containing Spiropterae (s. Plate I fig. 2—3). Very frequently the mucosa of the soft palate and the pillars of fauces will prove affected too, even if less pronounced, and the process then passes directly into the corresponding changes of pharynx and oesophagus. Also the front side of epiglottis will sometimes be implicated as well as the mucosa of the hard palate and the inside of the cheeks.

As mentioned above, macroscopical examination — especially in rats which have been suffering for some time or a longer period — will show that the lesions are located merely in radix linguæ and in the pillars of fauces, from where they pass into the Spiroptera oesophagitis which is almost constantly incident to them. Far less frequently, macroscopical changes will be met with in other parts of the tongue, although microscopical examination may show these suffering too and harbouring a few nematodes. In the early stages of the disease these parts of the tongue may, however, also be affected, most frequently the dorsal surface, so that the changes may be seen advancing forwards surpassing papilla vallata. The epithelium of dorsum is then rough and thickened, sometimes diffusely, frequently in spots. Whitish isles of hyperplastical desquamating layers of epithelium contrast against more reddish areas in which the epithelial layer is desquamated or but very slightly thickened. The lateral sides of the tongue are sometimes affected so that the fimbriæ linguæ stand out strongly, and the process from here enters sulcus alveololingualis. The tip of the tongue and apex will only seldom be found suffering, sometimes, however, thickening of the epithelium, and strongly marked papillæ filiformes may be met with. In all parts of the tongue Spiropterae are found, most frequently and most numerous in the root and round

aditus pharyngis' which, as well as introitus laryngis, may be quite obliterated by fully developed large parasites and desquamating epithelium.

As already mentioned, these processes in some cases may be so far pronounced that death of the rat by suffocation is likely to be caused by them.

For the microscopical examination 115 tongues have been employed, including 70 out of the 72 put down in the tables in which already by macroscopical examination pathological changes were observed. The examination was carried out entirely in serial sections (at $10\ \mu$, every 10th to 6th or more of which were examined) comprising in some cases the tongue in toto.

The histological changes consist in inflammatory processes and proliferation of the epithelium. In the connective tissue of the mucous membrane a number of polymorphonuclear leucocytes and, more scantily, mononuclear lymphocytes will be met with, often gathered in abscess-like foci. Sometimes the connective tissue of mucosa, especially that of the pillars of fauces, will contain small abscesses or minimal superficial necrotical ulcers. The ducts of the mucous glands are often strongly dilated by deposits of secretion and leucocytes, while, on the other hand, the albuminous glands will most frequently be normal. The elements of the connective tissue of mucosa are proliferating and mixed up with plasmacells, sometimes in abundance.

The predominant phenomenon is the epithelial hyperplasia which is often exceedingly advanced and proves to have caused the furrows and wrinkles of mucosa observed macroscopically. In the radix linguæ it does not merely consist in hyperplasia of the superficial layers especially of the horny layer which, as a rule, is greatly thickened, but also in proliferation of the deeper layers, so that the papillæ of the connective tissue of mucosa, thickened and infiltrated with leuco-

cytes, are split up by vigorous downward penetrating epithelial projections s. Plate II (fig. 15—17). Also the epithelium of the ducts of the glands has often undergone hyperplasia. A common finding is parakeratosis. At times the keratinized desquamating cells are separated by glandular secretion and contain leucocytes in abundance, not seldom heaped up in minimal abscesses as in the connective tissue of mucosa. The epithelial proliferation is sometimes extremely pronounced, reaching into the upper layers of the muscles in which then leucocytes are abundant.

On the dorsal surface and apex linguæ as well as on the lateral sides of the tongue the changes prove to be essentially the same, most frequently less pronounced, often very slight. In some cases, however, remarkable, epithelial proliferation and typical downgrowth has been observed.

Spiropteræ are met with in varying numbers, often abundant, in the epithelium, as a rule in the upper layers, sometimes in the basal layers, in a few cases they were found invading into the ducts of the glands. They reach the same stage of development as in the fundus of the stomach, gain sexual maturity and produce eggs as in the latter place. In some cases a few nematodes were met with in the epithelium without having caused any pathological changes whatever here nor in other elements of the mucous membrane.

The upper layers of the epithelium, especially the desquamating horny cells, often contain bacteria and fungus threads in abundance, but neither in the downwards penetrating projections of the epithelium nor in the connective tissue of mucosa, nor in the muscles, will microbes generally be found, while in ulcers or deep necrosis they are traceable without difficulty.

In a few rats which survived the transmission of the Spiroptera for a longer period, the inflammatory processes were more chronic, the proliferation of the connective tissue of

mucosa being somewhat more advanced and some of its fibrillæ enlarged and thickened. Pronounced papillary excrescences or unquestionable cicatricial formation did not occur.

This first type of affections which may suitably be termed desquamative Spiroptera glossitis, perfectly harmonizes — as far as the morphological features are concerned — with the corresponding inflammations developed almost constantly in the gullet and in the fundus of the stomach, even if the process may often be much more violent in the latter place.

Only in 1—2 rats the glossitis appeared independently, in all other cases the fundus of the stomach was also the seat of inflammatory processes, in a considerable number of cases of carcinoma too. In most cases Spiroptera inflammation was also met with in the gullet.

The fact that the glossitis perfectly harmonized with the usual Spiroptera lesions of the fundus of the stomach would a priori render it probable that also the former might be followed by carcinoma, and this assumption was all the more natural as the glossitis in man, especially when associated with considerable epithelial hyperplasia (leukoplakia, keratosis linguæ) is frequently accompanied by cancer of the tongue.

In 2 cases of glossitis (occurring in rats which died resp. 43 and 46 days after the 1st transmission of the Spiroptera) I now succeeded in finding in mucosa on the limit between radix linguæ and the pillars of fauces slight heterotopical and invasive downgrowth, the upper bundles of the muscular fibrillæ being dispersed by abnormally keratinized atypical cells and small horny globes.

Similar changes although less pronounced were found on the front side of epiglottis in a third rat which survived the 1st transmission of the Spiroptera for 153 days.

That very early stages of carcinoma were present in these

cases hardly admitted of any doubt. Furthermore, pronounced carcinomatous growth was found in 1 rat (case I), and finally very extensive cancer was met with in 3 cases (III—V).

These four cases form the second group of Spiroptera affections in the tongue of black and white laboratory rats; to these may be added a 5th case observed in a bastard of the Norway rat and black and white laboratory rats (case II). Two of these cases were found in 1914, 2 in 1915 and 1 in 1917. The cases will be reported not in succession of time but according to shape and stage of development of the carcinoma.

Case I.

Black and white male rat. Weight 200 gram.

On the perfectly healthy tongue 25 Spiroptera larvæ are applied during the period: October 19th to November 16th 1917, every second or third day (13 times altogether.).

At the last application (November 16th) the tongue was found no more affected than at former transmissions. The rat died Decbr. 19th strongly emaciated (weight 120 gr.) 61 days after the first, 33 days after the last application of the larvæ.

Macroscopical examination now shows the tongue to be violently changed (s. Plate I fig. 4—5). The epithelium is everywhere, most pronounced in radix and aditus pharyngis, thickened, rough and frayed, containing numerous Spiroptera, some of which protrude freely into the cavity of the mouth and gullet (s. the figures). On the left lateral border of the tongue and on the dorsal surface, the process passes into a stronger marked tumorlike thickening which extends from the left side of radix c. 13 mm. towards the tip of the tongue, and takes up the whole left half of the dorsal central part (s. Plate I fig. 4—6).

The cavity of the mouth, especially the palate, is the seat of inflammatory processes, epithelial proliferation and Spiroptera. The teeth are healthy. No swelling of the submaxillar, retromaxillar or other lymphatic glands. Nowhere metastasis formation. In the gullet considerable inflammatory changes, pronounced epithelial proliferation and numerous Spiroptera.

The fundus of the stomach is also severely attacked. The wall is thickened (measuring up to 5 mm.), the mucous membrane exhibiting papillary changes. The epithelium is the seat of hyperplasia and numerous Spiroptera. All other organs normal. No metastasis formation.

Microscopical examination shows a strongly pronounced glos-sitis of the common type (s. Plate II fig. 17), located in the root of the tongue; on the left lateral border a marked carcinoma is found, the structure of which is identical with that of the carcinoma of the tongue

in man (s. Plate II fig. 18). Strands of atypical strongly keratinized epithelial cells mixed up with horny globes and epithelial pearls penetrate into the muscles. In a single nerve the carcinoma invades the lymphatic space, filling up totally the perineural lymphatic sheath. Spiropterae are met with in the glossitis as well as in the carcinoma.

Also the anterior part and the tip of the tongue contain Spiropterae and inflammatory processes, proliferation of the epithelium with downgrowth of epithelial columns, but carcinoma is not observed here.

In the gullet and in the fundus of the stomach (cut totally in serial sections) corresponding but stronger changes are met with, carcinoma being, however, not traceable.

The maxillar and retromaxillar lymphatic glands and the lungs are also cut in serial sections. Metastasis formation is not observed.

Thus, in this case a strongly pronounced Spiroptera glossitis combined with carcinomatous growth of the tongue was observed. In the cavity of the mouth the gullet and the fundus of the stomach advanced inflammatory processes, proliferation of the epithelium, and numerous Spiropterae were found, carcinoma being not traceable.

Case II.

☞ Adult male rat, bastard of black and white laboratory rat and wild Norway rat (*M. decumanus*) is fed on free Spiroptera larvæ November 6th, 19th and 30th 1914, c. 100 larvæ at a time, applied on white bread soaked with a normal saline solution (0,9 pCt.). December 18th the rat is splenectomized¹ under aseptic precautions. The laparotomy wound healed without reaction. The rat died emaciated (weight 140 gr.) on December 28th 1914, 52 days after the first, 28 days after the last transmission of the Spiropterae.

The tongue measures from the tip to epiglottis 2½ cm. Its largest width is 7 mm. On the dorsal surface a strongly transformed protruding area of tumorgrowth is found, its longitudinal extent being 11 mm., its posterior limit situated at a distance of 5 mm. from epiglottis, taking up dorsum in all its width (s. Plate I fig. 7—8). The epithelium of this area is rough, thickened, furrowed and folded, strongly proliferating and greyish white. Its colour is lighter than that of the surrounding epithelium, and its surface elevated above the level of the latter, separated from it by a deep furrow. The affection is almost fungiform, extending down on both sides of the tongue, deepest on the right side (see fig. 7) where the convex marginal limit almost reaches the sulcus alveololingualis. Sagittal sections of the tongue demonstrate the process

¹ S. my previous paper. Det Kgl. Danske Videnskabernes Selskab. Biologiske Medde'elser I, 1918 p. 18.

penetrating downward into the muscles. Spiropterae are not discovered. The mucous membrane of the mouth cavity and of the pillars of fauces is normal, containing here and there Spiropterae. The teeth healthy. The cervical, submaxillar and retromaxillar lymphatic glands normal. The epithelium of the gullet is proliferating and desquamating, containing some parasites.

The stomach is of normal size. The wall of the cardiac portion is greatly thickened owing to epithelial proliferation and inflammatory processes. The epithelium harbours numerous Spiropterae, some of which protrude freely into the cavity. The pyloric portion normal. The organs exhibit no abnormalities. No metastasis formation. Peritoneum normal. In the place of the spleen no particular changes are found.

Microscopical examination (s. Plate III fig. 19) shows the tumor of the tongue to consist of papillary excrescences of connective tissue lined with thick layers of hyperplastical squamous-celled epithelium and containing cysts filled with epithelium. The epithelial covering of these excrescences is here and there necrosed. Irregularly placed foci and strands of atypical, strongly keratinized epithelial cells mixed up with horny globes and epithelial pearls heterotopically invade the connective tissue of the mucosa splitting up the fibrillae which are often proliferating and admixed small masses of leucocytes and lymphocytes, — and penetrate downwards between the fibrillae of the muscles. The carcinomatous transformation of the epithelium is sharply marked off from the surrounding parts which exhibit only hyperplasia, desquamation, and some downgrowth, most pronounced immediately in front of epiglottis. Here slight inflammatory changes and some scattered Spiropterae are found as well as other parts of the tongue and the epithelium of the mouth harbour a few Spiropterae. The lingual and cervical glands and the lungs are cut in serial sections. Metastasis formation is not observed. In the gullet and in the fundus of the stomach a considerable inflammation and a great number of Spiropterae are noticed. In a smaller circumscribed area of the cardiac portion of the stomach, furthermore, a typical though little extensive carcinoma is met with, extending downwards into the upper layers of submucosa.

Thus, in this case, slight Spiroptera glossitis and pronounced papillary carcinoma of the tongue was observed. In the gullet and in the fundus of the stomach Spiroptera inflammation was also found, in the latter associated with a carcinoma of small extent.

Case III.

Black and white adult female rat, belonging to the proper stock of the institute. Weight 210 gr. July 18th 1914 the rat is splenectomized¹.

¹ S. case II.

The laparotomy wound healed without reaction. On September 17th c. 100 larvæ deposited in a hydrous saline solution (0.9 pCt.) are injected into the stomach of the rat through a caoutchouc catheter. Oct. 3rd some 100 larvæ are injected again, and Oct. 15th another 100. The rat died extremely emaciated Decbr. 29th 1914, 103 days after the first, 75 days after the last injection.

Post mortem examination showed the tongue remarkably changed (s. Plate I fig. 9), measuring from apex to epiglottis $3\frac{1}{2}$ cm. Radix linguæ is the seat of an ulcer which extends from epiglottis 13 mm. forward on the root of the tongue. It leaves the right border free, being here bounded by a sharp downward- and outward-bent line and taking up all the left side border. The greatest width is 1 cm. The surface is whitish, strongly furrowed and cleft, rough, almost cauliflower-shaped and rises hill-like 2—3 mm. above the level of the surface of the tongue. The pillars of fauces seem to be intact. On transverse sections of the tongue the epithelial proliferation is seen to extend about 6 mm. into the muscles. The cavity of the mouth exhibits no abnormalities. Spiroptera are not observed. The teeth normal. The submaxillar, retromaxillar and cervical glands normal. The stomach is of normal size. The outside smooth. The wall of the cardiac portion is considerably thickened owing to epithelial hyperplasia and inflammation of submucosa. In a single place the thickness amounts to 4 mm. caused by papillary excrescences. The pyloric portion normal. The gullet presents no macroscopical changes. The organs apparently healthy. Peritoneum normal. No inflammatory processes in the place of the spleen. No metastasis formation.

Microscopical examination demonstrates the changes of the tongue in the tumor it self and its borders to consist in rather advanced inflammatory processes, and pronounced proliferation and downgrowth of the epithelium. The connective tissue of mucosa which has undergone violent proliferation and contains lympho- and leucocytes in abundance, sends out irregular slightly ramified crateriform, leaf- or channel-shaped offshoots lined with enormous layers of strongly keratinized epithelium (s. Plate III fig. 20). This, however does not alone line the papillary processes of the connective tissue, filling out the crypts between these, but also penetrates downwards into the muscles, the fibrillæ of which are thus split up by long strands or columns of epithelial cells which partly belong to the type of stratum germinativum, partly are more or less keratinized and often arranged as epithelial pearls and horny globes (s. Plate III fig. 21—22). The affection from the surface downwards extends through more than the half of all the tongue, splitting up the fibrillæ of the muscles which are displaced, often subjected to atrophy, frequently infiltrated with round cells, here and there necrosed as in the cancerous tissue. The boundary line of the process in front is sharply marked off against the normal anterior part of the tongue, the transitional passage exhibiting hyperplasia of the epithelium. At the back it leaves off immediately in front of the pillars of fauces and epiglottis, the connec-

tive tissue of which is infiltrated with leucocytes and lined with hyperplastic desquamating epithelium on the front side. The part of the surface of the carcinoma immediately adjacent to epiglottis as well as the non-carcinomatous part of the surface of the tongue contain *Spiropteræ* with eggs. The lingual glands are normal.

Microscopical examination of the gullet shows hyperplasia, desquamation of the epithelium, inflammatory processes of mucosa and *Spiropteræ*. In the fundus of the stomach violent inflammation and *Spiropteræ* are met with, and in a small area, pronounced carcinomatous growth. All macroscopically distinguishable cervical, retromaxillar and submaxillar lymphatic glands, as well as the lungs and the liver are cut in serial sections, but no metastasis formation is traced.

Thus, also in this case, *Spiroptera glossitis* and papillary carcinomatous growth of the tongue was observed. In the gullet and the fundus of the stomach inflammatory processes due to the *Spiroptera* were also met with, in the latter place associated with carcinoma.

Case IV.

Black and white male rat (belonging to the proper stock of the institute), weight 120 gr. September 24th 1914 is infected by injection of c. 100 free *Spiroptera* larvæ into the stomach. October 7th and Oct. 17th repeated injection is performed, c. 100 larvæ each time. The rat dies March 14th 1915, 171 days after the first, 148 days after the last injection. Weight 110 gr.

Post mortem examination shows: The full length of the tongue from apex to epiglottis is 2,4 cm. its greatest width 8 mm. On the dorsal surface a large greyish white area is observed, taking up the surface in all its width; its front limit is situated 1 cm. behind the tip of the tongue, its hind border at a distance of 5—6 mm. from epiglottis (s. Plate I fig. 10). This area is elevated above the level of the other part of the dorsum, the surface is rough, mostly covered with a proliferating thickened epithelium, which in some places is desquamating and necrosed. A deep furrow, outside which the surrounding epithelium is somewhat thickened, marks out the limit of this area. The tip and the root of the tongue are normal. No parasites are observed. The cavity of the mouth exhibits no abnormalities. The teeth are healthy. The gullet contains a few *Spiropteræ*. The stomach is highly enlarged, and of dense consistence. The cardiac portion adheres to the left lobe of the liver and to diaphragma, communicating with these by a somewhat necrosed tissue, consisting partly of liver- and connective tissue, partly of necrotical masses, and encircling like a mantle the central part of the fundus of the stomach. The free outside is rough and wrinkled, between the gullet and the necrosed area an

epithelial crypt hardly as large as a cherry stone is observed. The wall of the cardiac portion is extremely thickened owing to wall-shaped papillary projections, epithelial cysts and crypts with septa of connective tissue between them, the length of which sometimes amounts to more than 1 cm. The pyloric portion is normal. No pathological changes in the other organs.

Microscopical examination of the tongue (cut completely in serial sections, every 3rd of which is examined) shows in the region of the tumor itself changes similar to those met with in the preceding cases (s. Plate IV fig. 23). From the connective tissue of mucosa which has undergone pronounced proliferation and contains leuco- and lymphocytes like the upper layers of the muscles, irregular crateriform, leaf- or channel-shaped offshoots are sent out, lined and filled with enormous layers of partly keratinized epithelium. This penetrates downwards and invades the muscles, the fibrillæ of which are split up, displaced and subjected to atrophy by masses or strands of cells, often keratinized, irregularly arranged and mixed up with epithelial pearls and horny globes. A part of the carcinomatous area contains glands which exhibit no pronounced changes. The carcinoma is marked off sharply from the adjacent neighbouring areas, the epithelium of the transitional zone being hyperplastical and desquamating, its processes are somewhat elongated and the connective tissue of mucosa proliferated and infiltrated with leucocytes and lymphocytes in moderate number. The tip and the root of the tongue are normal Spiropterae are not observed.

All macroscopically distinguishable submaxillar, retromaxillar and cervical lymphatic glands are cut in serial sections. No metastasis formation is found. The gullet is the seat of proliferation, desquamation and slight downgrowth of the epithelium, infiltration with leucocytes in the connective tissue of mucosa, and a few Spiropterae.

As already found at the macroscopical examination the thickening of the wall of the fundus of the stomach is due to enormous papillary excrescences and epithelial crypts filled with keratinized epithelium and often situated in submucosa. On the whole, the changes correspond to those described in previous papers, a detailed report being thus unnecessary. It must, however, be mentioned that the epithelial covering of the enormous crypts in a higher degree than is generally the case, is built up by cells which belong to the same type as the most basal cells of the normal epithelium. Their shape is more cylindrical, their reaction with staining agents more intensive and their tendency to keratinizing is found to be less pronounced than usually. The formation of crypts is extremely pronounced. Only a few Spiropterae are met with (the fundus of the stomach was cut completely in serial sections).

Real invasive carcinomatous proliferation of the common type is found in some areas, in which the cells in strands or foci, sometimes arranged as epithelial pearls and horny globes penetrate infiltratively into submucosa.

At the microscopical examination of the lungs (cut in serial sections), the kidney, spleen and cervical, maxillar and retromaxillar lymphatic glands no metastasis formation is found. The part of the left lobe of the liver adherent to diaphragma and the stomach is the seat of inflammatory processes and- without sharp demarcation passes into the necrosed tissue which covers the outside of the fundus. These changes will probably have developed in consequence of a preceding perforation of the highly dilated crypt in the wall of the fundus.

Thus, in this case, typical carcinoma and slight inflammatory changes, localized in the tumor itself and in the transitional zone have been observed in the tongue, the epithelium of which harboured no Spiropterae. In the gullet and the fundus of the stomach Spiropterae were met with, in the latter place the inflammatory processes were extremely violent and mucosa had undergone strong papillary transformation. Carcinoma was found in a limited area. The number of Spiropterae was only small.

Case V.

Adult black and white male rat (belonging to the proper stock of the institute) is fed June 2nd 1915 on the femora of 10 cockroaches (*P. americana*) infected with the Spiroptera. He dies December 20eth 1915, 201 days after the transmission of the Spiropterae. Post mortem examination shows the rat to be emaciated (weight 175 gr.). On the tip of the tongue a greyish white papillary fungiform tumor is found, sharply marked off from the other part of the tongue and taking up all the tip (s. Plate I fig. 11—12). It measures in lateral extension $\frac{1}{2}$ cm., from the surface downwards 6—7 mm., in longitudinal extension a little more than 2 mm. The tongue, for the rest, is normal, as well as the cavity of the mouth. The mucous membrane of the pillars of fauces is thickened, desquamating. No Spiropterae are found. The teeth normal. The gullet normal. The stomach exhibits violent changes, its fundus being enlarged and its wall strongly and unequally thickened (to about $\frac{1}{2}$ cm.). Papillary walls and processes consisting of strongly proliferated connective tissue and thick epithelial layers diminish the cavity to such an extent that its lumen is partly obliterated. Some few Spiropterae are found in the epithelium. The pyloric portion of the stomach is normal. The organs normal. No metastasis formation. Half of the tumor of the tongue is carefully washed out in a normal saline solution (0,9 pCt.), cut to pieces and minced. Through a hollow needle small particles are applied subepithelially on the tongue of 4 black and white young rats (weight 90—95 gr.) and subcutaneously on the front side of thorax of one young black and white rat. In

neither of these animals, which died 2—4 months after transplantation, development of cancer could be traced.

Microscopical examination: The half of the tumor which was not transplanted is cut completely in serial sections (every 3rd examined). The tumor on the tip of the tongue exhibits the common structure, typical of papillary carcinomata (s. Plate IV fig. 24—25). Irregular crateriform, leaf- or channel-shaped slightly ramified offshoots covered with enormous layers of strongly keratinized squamous celled epithelium are sent out by the connective tissue of mucosa, in which lymphocytes are abundant and proliferation strongly pronounced. The epithelium not only lines the papillary processes filling up the crypts between them, but also, partly in compact columns or strands, partly as pearls or small foci of cells, invades the underlying layers, forming laminated capsules of keratinized cells or horny globes. These strands or columns of cells force their way deep into the muscles, splitting up the fibrillæ of the latter and giving rise to atrophy. In some places carcinomatous growth is seen invading the lymphatic spaces of the nerve fibres. In serial sections these are then seen surrounded by epithelial cells partly of the stratum germinativum type, partly somewhat or completely keratinized, forming here and there laminated capsules or horny pearls (s. Plate IV fig. 26—27). The cancer cells are chiefly found in perineural — in a single place — moreover, in endoneural lymph spaces. The outer layers of the epithelium of the tumor are in some places desquamating or necrosed, but for the rest well conserved. Tumor is sharply marked off from the other part of the tongue which, on the whole, is healthy (examination in serial sections). Only on the limit of the carcinomatous area proliferation and desquamation of the epithelium is found, Spiropterae are not met with.

All submaxillar, retromaxillar and cervical lymphatic glands are cut in serial sections, every 5th—6th of which are examined. Metastases are not found. The gullet is normal, containing no Spiropterae. In the fundus of the stomach the following changes are observed: pronounced inflammation of the mucous membrane and of the connective tissue of submucosa, violent, partly heterotopical proliferation of the epithelium, limited development of cancer, and a few Spiropterae.

The lungs (cut completely in serial sections, every 5th of which is examined) contain no metastases, no more than the retroperitoneal lymphatic glands; the spleen, liver and kidney are normal. The urinary bladder normal, containing some *Trichodes crassicauda*. No other pathological changes.

Thus, in this case, as distinct from the preceding cases, the locality of the carcinoma was found to be the tip of the tongue. Outside the tumor no inflammatory

processes were met with. The epithelium of the tongue, for the rest, was normal, neither here nor in the normal epithelium of the gullet *Spiroptera* were found. On the other hand, the fundus of the stomach contained a few parasites, inflammatory changes and carcinomatous growth.

In a sixth case I have observed a tumor of the tongue in a rat infected with the *Spiroptera*.

Young black and white female rat, weight 90 gr. about 7½ months old was sent to the Anatomico-Pathological Institute from the Serum Laboratory of the Veterinary High School in Copenhagen, and belonged to a stock of rats imported from Holland. On February 1st 1918 10 fully developed and encysted *Spiroptera* larvæ, deposited in a normal saline solution are injected into the stomach of the rat through a catheter. The rat is killed June 10th 1918, 136 days after the transmission of the *Spiroptera*. On the right side of the dorsal surface adjacent to the right pillar of fauces a papillary tumor, about hemispherical in shape and measuring 5 mm. in diameter is observed (s. Plate IV fig. 13). It rises wart-like above the level of the surface, exhibiting no ulcerations. The other part of the tongue is normal. No *Spiroptera*. The gullet shows no abnormalities and contains no parasites. No swelling of the sub- and retromaxillary lymphatic glands. Slight *Spiroptera* inflammation and epithelial hyperplasia is found in the fundus of the stomach. In the liver small cysticercs. The organs normal. No metastasis formation.

Microscopical examination (serial sections) shows the tumor of the tongue to be a typical papillary fibro-epithelioma at the root of which pronounced inflammatory chiefly chronic processes are found in the mucous membrane. Carcinomatous growth is not traceable. The epithelium is normal, contains no *Spiroptera*. The gullet is normal. In the fundus of the stomach slight inflammation, epithelial hyperplasia and *Spiroptera* are met with. No signs of cancer.

Thus, in this case, a non-cancerous papilloma of the tongue was found in a black and white rat infected with the *Spiroptera*. The epithelium of the tongue contained no *Spiroptera*. Inflammatory changes were met with in the base of the papilloma. In the fundus of the stomach inflammation, epithelial hyperplasia and parasites were observed.

The structure of this papilloma was quite identical with that of the common papillomas in man (fibro-epithelioma)

occurring in such mucous membranes as are covered with squamous celled epithelium, and apparently corresponded also to the structure of the papillomata, observed by YUTAKA KON¹ in the tongue of rabbits fed on lanoline.

As seen from the above description, carcinomatous growth of the common type, arising from the squamous celled epithelium of the tongue was found in all 5 cases (I—V). In two cases (I and V) the carcinoma had invaded the peri- and endoneural lymphatic spaces of the nerves of the tongue, in no other case metastatic extension was found. The infection with Spiroptera in all 5 cases had produced slight or strong inflammatory changes of the fundus of the stomach, in 4 cases (II—V) associated with more or less extensive development of cancer in this organ.

The association of cancer of the tongue with cancer of the fundus of the stomach in these 4 cases, no doubt, as in analogous cases (e. g.: carcinoma of the tongue and stomach in horse communicated by FØLGER²) is to be ascribed to an independent development of cancer taking place coincidentally in both organs influenced by the same factor, in these cases the invasion of the Spiroptera. No other relationship can exist between these carcinomata. Setting aside that metastatic cancer of the tongue is of so unfrequent occurrence that the tongue has even been considered immune against metastatic deposits of tumor growth (WOLFF³), it would be unreasonable to suppose that cancer cells — and in 4 cases — should have been carried from the fundus of the stomach merely to the tongue, or, vice versa, from the latter alone to the stomach, without having affected lymphatic glands, lungs or other organs.

¹ l. c. p. 8,

² l. c. p. 4.

³ Die Lehre von der Krebskrankheit. Bd. II. 1911. p. 377.

Nor would it be reasonable to talk of implantation of cancer cells into the tongue from the fundus of the stomach, or implantation into the latter from the tongue, and merely because this way of extension in rats would meet with quite the same difficulties as have been emphasized in the discussion of the developing of similar multiple cancers in the alimentary canal of man, difficulties which have involved the denial of implantation of tumor growth occurring in the said organs at all.

The relation between the invasion of the Spiroptera and the development of cancer in the tongue is most conspicuously seen in case I. The greater part of the tongue, especially the radix, was here the seat of numerous Spiropterae and typical, strongly developed glossitis, which on the left border of the tongue passed directly into tumor-like thickenings, identified by microscopical examination as a pronounced carcinoma containing Spiropterae. The inflammatory changes could be traced through the gullet into the fundus of the stomach which was also highly affected but showed no carcinomatous transformation, any more than the gullet. The glossitis in this case was in perfect keeping with the corresponding changes of the gullet and the fundus of the stomach, and the total picture of the inflammation and the carcinoma of the tongue completely harmonized with the common findings in carcinomatous stomachs in rats infected with the Spiroptera.

In cases II and III where the carcinoma had obtained very considerable dimensions, the inflammatory processes in the tongue came out less strongly and were located essentially in the area immediately in front of epiglottis and in the neighbouring borders of the carcinoma. The Spiropterae, being only scantily present in the tongue, were found in greater number in the gullet and the fundus of the stomach.

In cases IV and V I did not succeed in tracing out Spiropterae in the epithelium of the tongue, the fundus of the stomach

contained only a very few specimens. In case IV the gullet contained a few, in case V no Spiropterae.

Neither was pronounced glossitis observed in as far as the inflammatory process was but slightly marked and located all but entirely in the carcinoma itself. Thus, a direct connection between carcinoma and Spiroptera glossitis was not traceable in the cases IV and V and an accidental association of carcinoma of unknown origin in the tongue with Spiroptera cancer in the fundus of the stomach might be supposed in these 2 cases. This assumption, however, would be but little reasonable only for the reason that spontaneous cancer or tumor growth of the tongue — as mentioned above (p. 7.) — anterior to these examinations has never been observed neither in the rats of the Institute, in spite of strenuous searching, nor in other rats. And furthermore, the non-appearance of glossitis and Spiropterae may quite naturally be explained by these rats having survived the transmission of the Spiroptera for about 6—7 months, and not like those in cases I—III only for about six weeks — 3 months. The tongue then may very probably have been the seat of inflammation and Spiropterae at an earlier date, and the fact that the parasites have disappeared later on, so that none could be traced at the death of the animals — perfectly corresponds to what is commonly found in rats which have survived the transmission of the Spiroptera for a longer period than 3 months.

It is then to be assumed that the earliest initial stage of the carcinoma in these cases has developed while the tongue was still the seat of Spiropterae and glossitis, and that, after this the carcinoma has continued its growth and manifested itself in spite of the retrogression of the inflammatory processes and the disappearance of the Spiropterae. The chances are, that the papilloma, described above, has developed in quite the same way. Besides, the relations of Spiroptera inflammation and Spiroptera cancer will be mentioned.

in an ensuing paper¹, where the disappearance of the nematodes will also be dealt with.

In no case cicatricial tissue which might have been put into causal relationship to the tumor growth, was observed, any more than abnormally sharp or deviated teeth.

Also in man carcinoma of the tongue may occur in certain cases after decrease or intermission of a preceding glossitis. Inspection of the literature concerning leukoplakia and cancer of the tongue will convey the impression that development of carcinoma is not only met with in cases where the leukoplakia is in active progression or florid, but may also occur in cases in which the leukoplakia is less pronounced or even retrogressive. In a few cases, reported by BUTLIN² and BORCHARD³, carcinoma of the tongue has developed after cessation of the initial leukoplakia.

The results of the investigations reported in this paper may be summed up as follows:

By transmitting *Spiroptera neoplastica* (*Gongylonema neoplasticum*) to black and white rats (and to bastards of these and wild Norway rats (*M. decumanus*)), invasion of the parasite into the epithelium of the tongue may give rise to a glossitis (most frequently when the rats are infected by transmission of free *Spiroptera* larvæ) which is mainly located in radix linguæ but is also likely to occur in other parts of the tongue. The glossitis as well as the contemporaneously developed *Spiroptera* inflammation in the gullet and the fundus of the stomach is associated with strongly pronounced proliferation, desquamation and downgrowth of the epithelium, which harbours *Spiroptera*. In numerous

¹ Det Kgl. Danske Videnskabernes Selskabs Biologiske Meddelelser I₁₁ 1918.

² British Medical Journal. 1906. May 26th.

³ Doppelttes Zungencarcinom bei Psoriasis linguæ. Deutsche Zeitschrift f. Chirurgie. 1914.

cases the glossitis decreases rather early so that pathological changes and Spiropterae will only unfrequently be met with in rats which survive the transmission of the Spiroptera for more than about 3 months.

The Spiroptera inflammation of the tongue, like that of the fundus of the stomach, may be associated with carcinoma which, fully developed, is traceable coincidentally with the glossitis, but, on the other hand, carcinoma may also be observed in cases where the latter has presumably ceased, inflammatory changes not being met with outside the carcinoma itself, and Spiropterae not found in the epithelium.

In one case a papilloma of the tongue has been observed, which was produced, most likely, in the same way as the other tumors.

According to these investigations Spiroptera cancer of the tongue seems to be of unfrequent occurrence in black and white rats. It cannot, however, be precluded that it may turn out to be more frequently produced in future experiments, especially if the rat's tongue be exposed to repeated invasion of Spiropterae during longer periods, as table III might perhaps suggest.

Possibly, cancer of the tongue might also prove to develop more easily in other stocks of black and white rats or in other species or races of rats than those hitherto employed. Up to now, I have only been able to produce glossitis, but no carcinomatous growth by transmitting the Spiroptera to Norway rats (*M. decumanus*), house rats (*M. rattus*), white laboratory mice, domestic mice (*M. musculus*) and forest mice (*M. sylvaticus*). In one out of the very few squirrels (*Sciurus vulgaris*) which have been at my disposal, a violent Spiroptera glossitis also appeared to be the only affection traceable.

It must, however, be mentioned that these experiments have not yet been completed and that only those concerning mice have comprised greater numbers of experimental animals.

The analogy existing between the development of cancer in the tongue associated with *Spiroptera glossitis* in rats, and that of cancer in the tongue associated with leukoplakia in man, has repeatedly been mentioned in the preceding pages. But also in other respects analogous conditions are met with. Firstly in the shape of the carcinoma of the tongue which in rats entirely corresponds to the papillary carcinoma of the tongue in man. Furthermore, in the microscopical structure which reveals perfect harmony, not merely in the histological details, but also in the demarcation of the tumors.

On studying comparatively the growth of spontaneous and transplanted carcinomata BASHFORD¹, for one thing, has emphasized the correspondence as regards the way of extension of the spontaneous carcinomata of the tongue in man and in cat. Also the carcinomata of the rat's tongue described in the preceding pages exhibit similar congruities which will be mentioned in an ensuing paper. This as well as the observations made by BASHFORD may be used as a confirmation of the assumption emphasized by RIBBERT a. o. that the propagation of the carcinoma takes place by its developing out of itself from small circumscribed foci of cells without increasing in size by a successive advancing of the process from the foci of cells originally carcinomatous to the adjacent neighbouring epithelium.

The result of my above investigations has furthermore proved the necessity of ascribing to the intestinal worms pathogenetical power in the development of cancer.

To assign to such parasites the ability of playing a prominent or a greater part in the development of cancer of the tongue in man, would, no doubt, be absurd, in view of our thorough knowledge of these carcinomata, but, on the other hand, it cannot be denied that they may occasionally act as causative factors.

¹ Scientific Report of the Imperial Cancer Research Fund. II. 1905.

Among the well known European parasites only the *Trichina spiralis* may be taken into consideration, other species of worms being of exceedingly rare occurrence in the tongue.

On the other hand, trichinæ are commonly found in the muscles of the tongue, and — as I have mentioned in previous reports — it has been emphasized as early as in 1863 (by KLOPSCH)¹, and also later on, that an etiological relationship may probably exist between the trichina and carcinomatous growth. Literature contains reports of altogether 4 cases of carcinoma mammæ (KLOPSCH², GROTH³, v. LINSTOW⁴, STRANDGAARD)⁵, 1 case of carcinoma of the skin (LANGENBECK)⁶, and 1 case of carcinoma pleuræ (BABÉS)⁷, in all of which cases the immediately adjacent muscles, in a few cases also the carcinoma itself contained trichinæ.

Greater interest in this respect attaches to 2 cases of carcinoma of the lip, reported by HEDINGER⁸ and BABLER⁹, in which trichinæ were also found. According to an accompanying table the case of BABLER was a carcinoma of apparently the same papillary structure as those met with in the rats described above. In literature, however, I have not been able to find out any other report of tumors containing trichinæ

¹⁻² Fall von Trichinenerkrankung im Jahre 1842. Virchows Archiv 1863.

³ Ein Fall von geheilter Trichinenkrankheit. Virchows Archiv 1864. Bd. 29.

⁴ Über das Vorkommen von *Trichina spiralis* in einem Scirrhus. Virchows Archiv Bd. 44. 1868.

⁵ Et Tilfælde af Trichinose som tilfældig Komplikation ved Cancer mammæ. Ugeskrift f. Læger 1900.

⁶ Verhandlungen der med. Gesellschaft. 20. Mai 1863. Deutsche Klinik. No. 24. 1863.

⁷ Ein 21 Jahre alter Fall von Trichinose mit lebenden Trichinen. Centralblatt f. Bakteriologie. 1906. Bd. 42.

⁸ Medizinische Gesellschaft. Basel. Sitzung 6. Mai. 1909. Correspondenzblatt f. Schweizerärzte. 1909. No. 13.

⁹ Trichinous Infection of a Carcinoma of the Lip. Annals of Surgery Vol. XLVII. 1908.

than one quoted by BUTLIN and observed by MILTON¹. In this case trichinæ were discovered in the muscular tissue round a "suspect" tumor in the tongue. The following observation, therefore, might be worthy of special interest. In a letter of 1913 Prof. HOLLÄNDER² of Berlin has communicated this case to me and permitted me to publish it. The patient in question had been operated for a carcinoma of the tongue 1898 by Prof. ISRAEL in Berlin. Microscopical examination carried out by Prof. HOLLÄNDER showed at the base of the carcinomatous ulcer, in the immediate neighbourhood of the carcinomatous area, a tissue infiltrated with round cells and containing some trichinæ. As far as could be seen from the microscopical preparations which comprised a rather large area, trichinæ were traceable in no other part of the tongue.

From this and the cases mentioned above it may be seen that observations have been made in the tongue, the lips, in mamma and elsewhere, in which the occurrence of trichinæ in or in the immediate neighbourhood of carcinomatous tissue can hardly be explained as an accidental coincidence. But the rarity of these cases makes it evident that carcinomatous growth produced by trichinosis only occurs under special circumstances. To find out these definitely is not yet possible. The main presupposition seems to be, that the parasites — as is sometimes the case — are located especially in certain areas of the muscles placed close to an epithelial tissue.

As will be discussed in a following paper, significance, no doubt, must be ascribed too to predisposition, viz: the susceptibility of the epithelial tissue to develop into carcinoma under the influence of the parasites; also possible va-

¹ The original paper (in Transactions of the Pathological Society of London 1849) most unfortunately has not been at my disposal.

² I desire to acknowledge my indebtedness to Prof. HOLLÄNDER also here.

riations of the ability of the trichinæ to produce poisonous excretions must be taken into consideration.

Finally, the chronic nature of the trichinosis may have played a rôle, the influence of the parasite being thus of greater duration. In GROTH'S case of carcinoma mammæ, in LANGENBECK'S case of cancer cutis, in BABÈS' case of carcinoma pleuræ, and in KLOPSCH'S and v. LINSTOW'S cases of carcinoma mammæ the patients concerned had lived infected with the trichinae for resp. 7, 18, 21, 24 and 25 years. In the cases reported by GROTH, BABÈS, KLOPSCH and v. LINSTOW the trichinae proved to be still alive.

I desire to acknowledge my indebtedness to the CARLSBREG Fund and the W. BENDIX Legacy for their support of these investigations.

PLATES

Plate I.

- Fig. 1. Normal tongue of a rat. $\times \frac{2}{1}$.
- 2. Spiroptera glossitis in a rat. $\times \frac{2}{1}$.
- 3. Spiroptera glossitis in a rat. $\times \frac{2}{1}$.
- 4-5. Spiroptera glossitis combined with carcinoma. Case I. $\times \frac{2}{1}$.
- 6. Transversal section of the tongue of the same rat. Case I. $\times \frac{2}{1}$.
- 7-8. Spiroptera carcinoma of the tongue in a rat. Case II. $\times \frac{2}{1}$.
- 9. — — — — Case III. $\times \frac{2}{1}$.
- 10. — — — — Case IV. $\times \frac{2}{1}$.
- 11-12. — — — — Case V. $\times \frac{2}{1}$.
- 13. Fibro epithelioma in the tongue of a rat. $\times \frac{2}{1}$.
- 14. Normal tongue of rat. Transversal section through radix. $\times \frac{4}{1}$.

PLATE I



Fig. 1.



Fig. 2.



Fig. 3.



Fig. 9.



Fig. 10.

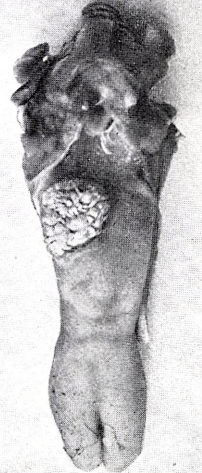


Fig. 13.



Fig. 4.

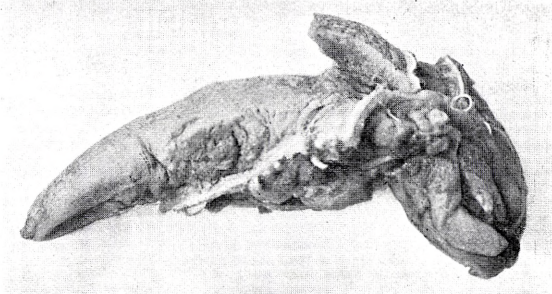


Fig. 5.

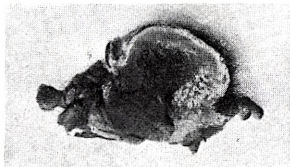


Fig. 6.



Fig. 8.



Fig. 11.

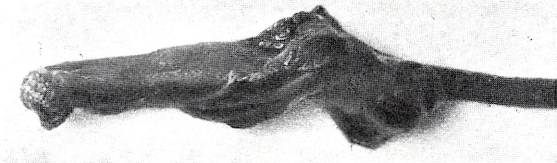


Fig. 12.

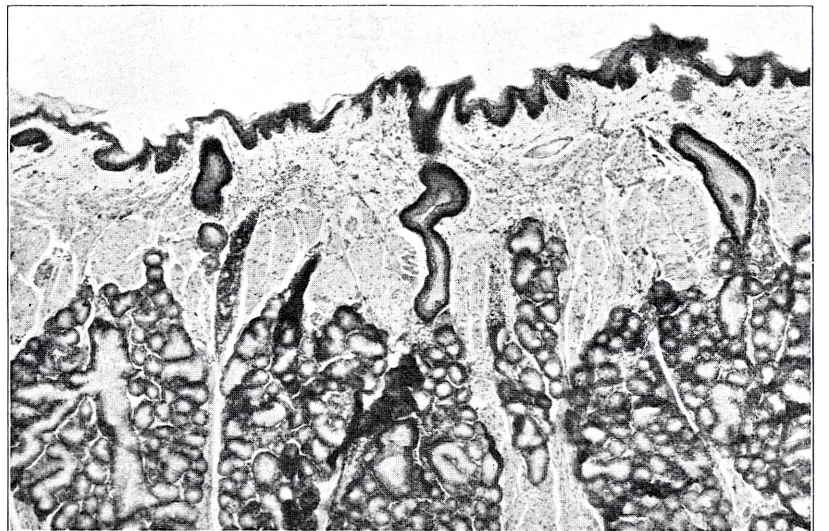


Fig. 14.

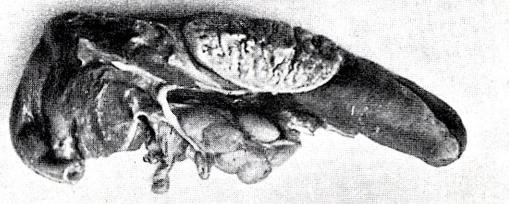


Fig. 7.

Plate II.

- Fig. 15. Spiroptera glossitis in a rat. $\times \frac{4.5}{1}$.
- 16. — — — $\times \frac{4.5}{1}$.
- 17. — — — Case I. $\times \frac{4.5}{1}$.
- 18. — carcinoma of the tongue a in rat. Case I. $\times \frac{2.4}{1}$.

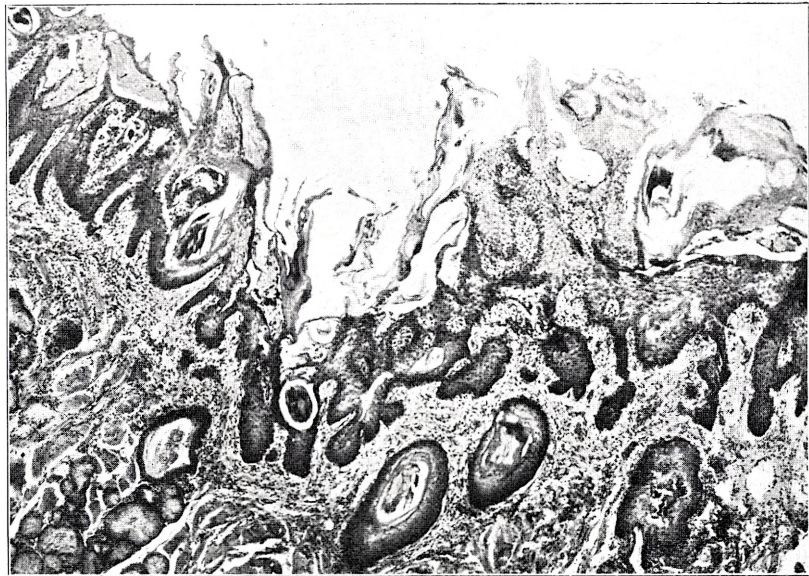


Fig. 15.

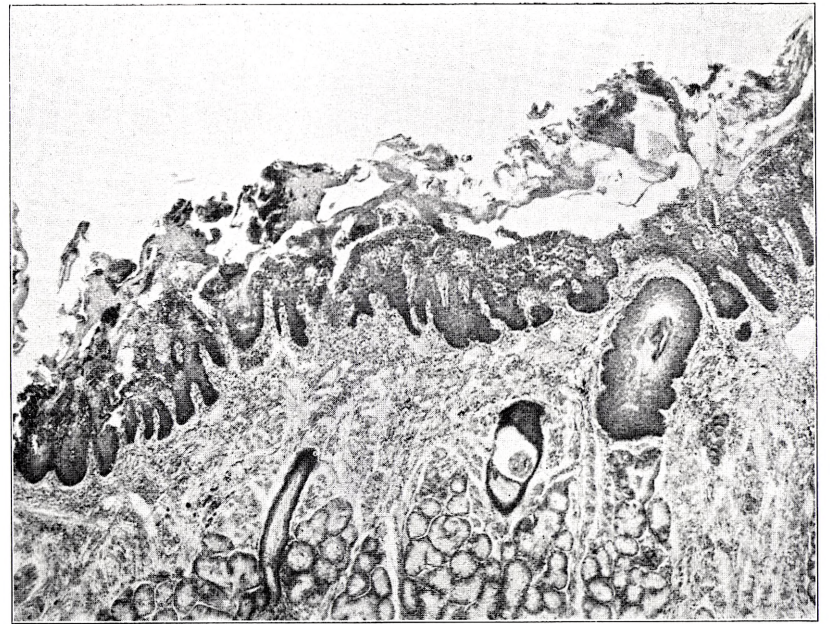


Fig. 17.

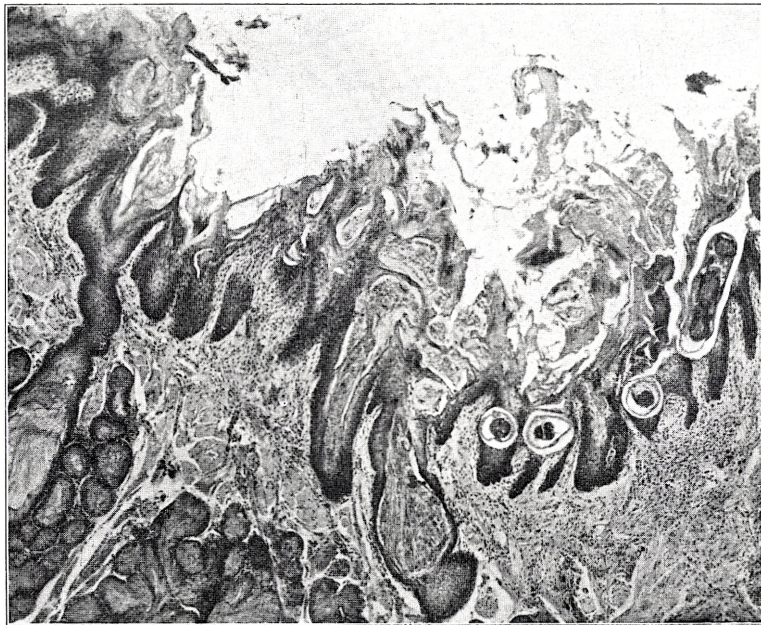


Fig. 16.

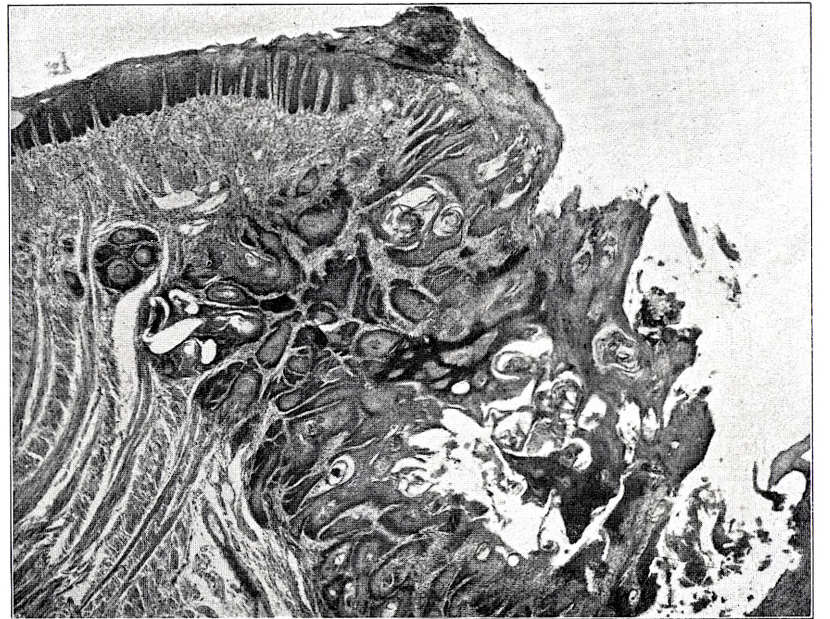


Fig. 18.

Plate III.

Spiroptera carcinomata of the tongue in rats

- Fig. 19. Case II. $\times \frac{5^0}{1}$.
- 20. Case III. $\times \frac{1^5}{1}$.
- 21. Case III. $\times \frac{5^2}{1}$.
- 22. Case III. $\times \frac{8^0}{1}$.

PLATE III

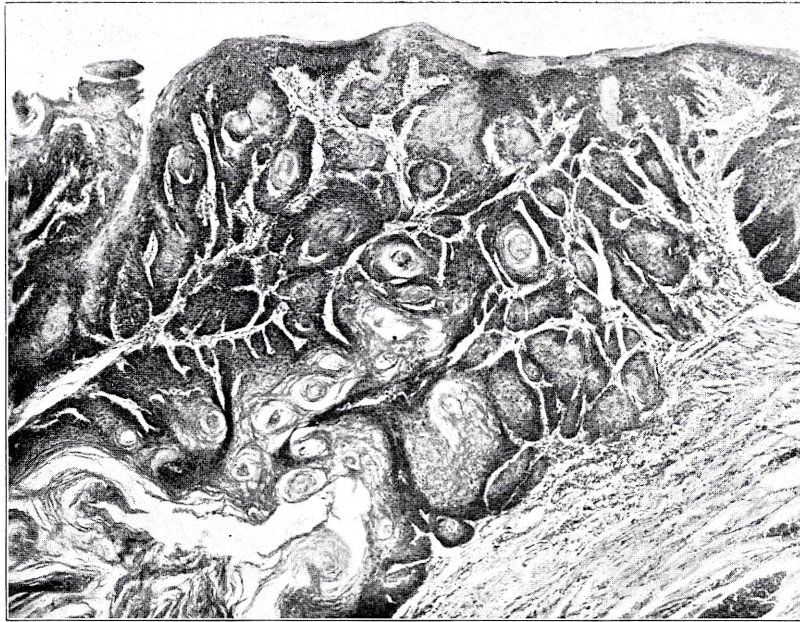


Fig. 19.



Fig. 21.



Fig. 20.



Fig. 22.

Plate IV.

Spiroptera carcinomata of the tongue in rats

- Fig. 23. Case IV. $\times \frac{4^2}{1}$.
- 24. Case V. Apex of the tongue. $\times \frac{1^5}{1}$.
- 25. Case V. $\times \frac{4^7}{1}$.
- 26. Case V. The carcinoma is seen invading the lymphatic spaces
of a nerve. $\times \frac{3^0}{1}$.
- 27. Case V. The same preparation. $\times \frac{13^0}{1}$.

PLATE IV

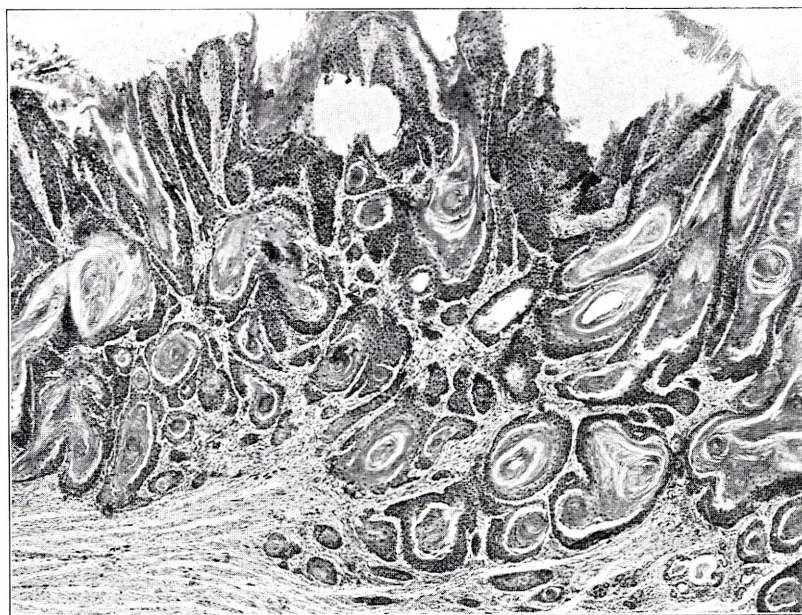


Fig. 23.

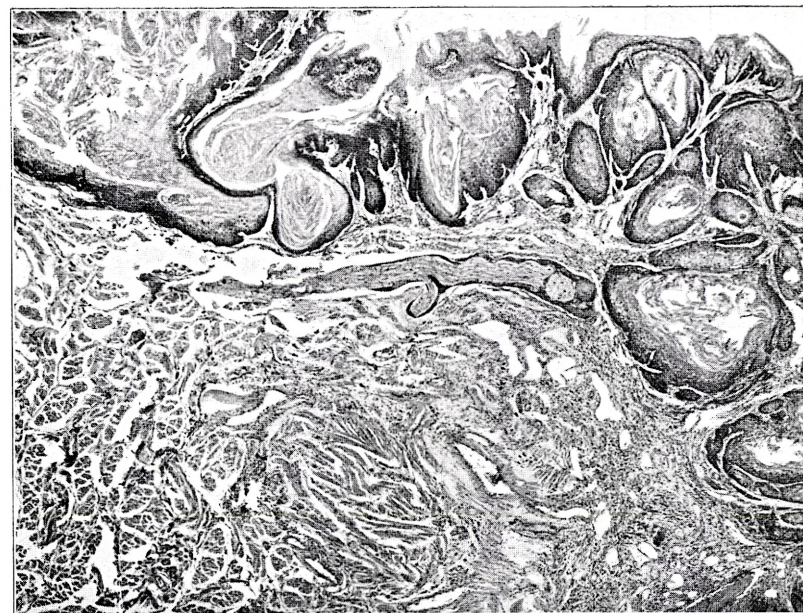


Fig. 26.



Fig. 24.

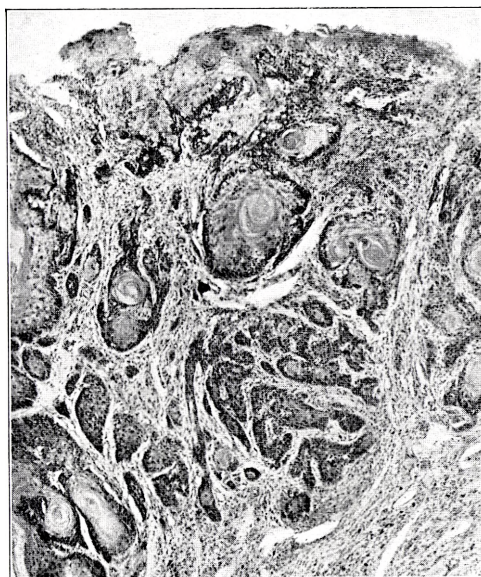


Fig. 25.

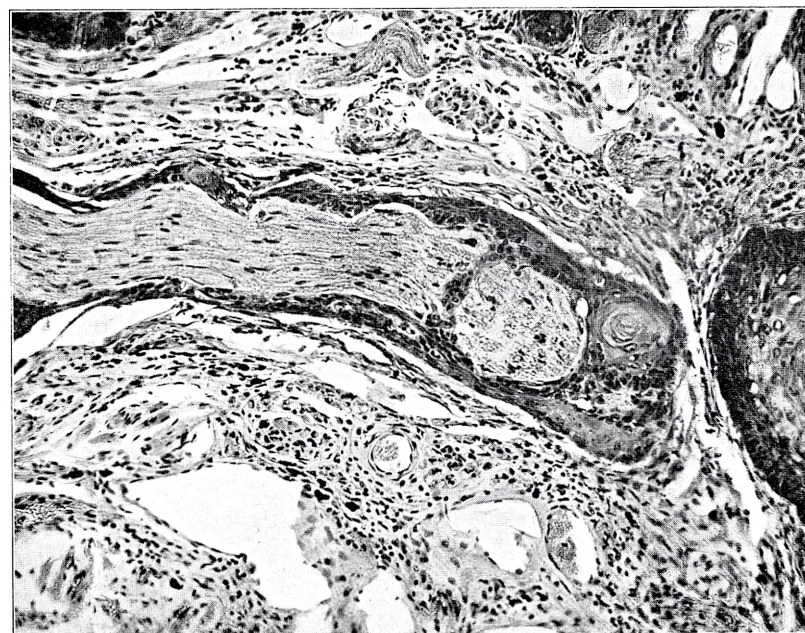


Fig. 27.

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8^{DE} RÆKKE

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