Oral Carcinoma in a Monkey Colony
A Report of Two Additional Cases*

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A search of the literature discloses the extreme rarity of oral carcinomas in domesticated and captive wild animals. Only 15 cases of carcinoma of the tongue in domesticated animals (horse, cow, cat, dog) reported from various countries have come to our attention (14). The autopsy records of the 10,298 mammals and birds that died in the Philadelphia Zoological Garden during a period of about 30 years show that only one animal, a male bear, with a carcinoma of the tongue was found (11). As far as subhuman primates are concerned, the incidence of neoplastic diseases, according to Ratcliffe, has been and still is lower in the Philadelphia Zoological Garden than for any comparable group of mammals (12), and until 1947 no carcinomas of the tongue in monkeys were discovered (13). Nor have we seen any references to malignant neoplasms of the tongue in autopsy reports issued from other laboratories and zoological gardens in which monkeys have come to autopsy. The occurrence of 2 additional oral carcinomas in our laboratory colony of monkeys is of particular interest not only because of the previous occurrence of 3 carcinomas of the tongue in 2 other monkeys of the same colony (14), but also because the 2 cases to be reported involve different sexes and different divisions of the suborder Pithecoidea. Furthermore, we believe that the information available on these animals and on the environmental conditions under which the disease occurred is somewhat more detailed than is usually the case in studies of monkeys that have died in laboratories or menageries.

Case Reports

These 2 monkeys were members of a colony of catarrhine and platyrrhine monkeys kept by one of us (H. K.) for neurophysiological and behavioral investigations.

Monkey No. 1.—A male rhesus monkey (Macaca mulatta), purchased from an animal dealer in New York City, was received in the laboratory on October 7, 1939. Its age at the time of arrival was estimated to be approximately 8 years. It was kept continuously in the laboratory until its death on February 23, 1945. It is worth noting that throughout this period of observation, whenever the animal was found sitting quietly, its lower jaw drooped (Fig. 1). This behavior is not characteristic of normal rhesus monkeys. During January 1945 the monkey started losing weight and confined itself more and more to eating soft food only. Saliva drooled frequently from its mouth and an occasional twitching in the region of the left corner of the mouth was observed. An examination of the mouth under nembutal anesthesia, on February 22, 1945, revealed an ulcerating lesion at the tip of the tongue measuring about 1.8 cm. in diameter and extending transversely about 2.1 cm. at the under surface of the tongue. Biopsy disclosed a squamous cell carcinoma.

On February 23, 1945, the monkey was anesthetized with ether and then sacrificed by an incision into the heart. Eyes, ears, nose and throat were grossly normal. The tongue had an ulcerating lesion on the left tip extending from the papillary margin down to the floor of the mouth. The submaxillary gland as well as the muscular tissues and vessels on the left side seemed to be directly infiltrated by the tumor. A small isolated nodule was noted in the hard palate. The other submaxillary and sublingual glands as well as the main cervical lymph nodes of the upper part of the neck were involved to a greater or lesser degree by metastatic tumor. The thyroid was not identified. The lungs were normal except for anthracotic pigmentation. The mediastinum and its contents and the heart were normal. Lymph nodes were not involved. The thymus was not identified. The abdominal cavity seemed grossly normal, except for a few adhesions between the viscera, and the spleen was rather small and shrunken. The liver, gallbladder, extrabiliary ducts, pancreas, stomach and small bowel seemed normal. The

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large bowel was normal except for a cecal diverticulum. There was a moderate amount of retroperitoneal fat. The kidneys, adrenals, bladder, testes and prostate seemed grossly normal. The teeth were in good alignment and showed no visible caries with the exception of the maxillary third molar on the right side. Body weight: 7.7 kgm.

Microscopic examination of the primary lesion showed a squamous cell carcinoma irradiating deeply into the tongue musculature (Fig. 2). There were regions of anaplasia and regions where pearl formation was quite definite. Sections through the enlarged regional cervical nodes showed practically complete replacement of lymphoid tissue by metastatic squamous cell carcinoma (Fig. 3). The following tissues were examined and found to be negative: striated muscle, parotid gland, stomach, small bowel, gallbladder, kidney, adrenal, urinary bladder, pancreas, spleen and lung. The testes showed normal configuration and relationship of tubules and stroma. However, there was definitely less spermatogenesis apparent than would be expected in normal monkeys at the height of sexual potency. The liver showed no cirrhosis or cholangitis. There was extensive fatty infiltration of the liver cells.

*Monkey No. 2.*—A female squirrel monkey (*Saimiri sciurea*) was received from the Brookfield Zoological Garden (Illinois) on June 3, 1938, and kept under study in the laboratory until its death on March 1, 1944. It was estimated to be between 2 and 2½ years old at the time of its arrival. On November 6, 1943, a small reddish lesion which at first was not encrusted appeared at the right angle of the mouth. About two weeks later the animal was no longer able or willing to shell peanuts, but only in February 1944 was it apparent that the visible lesion had definitely increased in size with a progressive swelling of the right cheek. The animal was often seen rubbing its face against the boards of the cage and scratching its right cheek very gently. On March 1, 1944, the swelling extended from the angle of the mouth to the right ear and was about the size of half a walnut. During the preceding week the animal had not been able to consume any food except milk and milk-soaked bread. When an examination revealed a large ulcerating lesion of the right buccal mucosa emitting an offensive odor, the monkey was killed by means of ether. Body weight: 568 gm. Brain weight: 22 gm. Owing to other demands on the laboratory, no detailed postmortem examination could be made. All organs, including the tongue and the teeth, appeared grossly normal.

Microscopic examination of the lesion of the right buccal mucosa showed a typical squamous cell carcinoma (Fig. 4).
CONDITIONS ASSOCIATED WITH OCCURRENCE OF CANCERS

Age factor.—At the time of death monkey No. 1 had lived approximately 5 years and 4 months and monkey No. 2, 5 years and 9 months under the conditions of this laboratory. If our estimate of the age on arrival is approximately correct, monkey No. 1 (rhesus) was about 13½ years old at the time of death and must, therefore, be considered aged in view of what is known about the life span of macaques. Published records provide no definite information on the duration of life in squirrel monkeys. Flower (3) discovered that a statement to the effect that a squirrel monkey lived as long as 9 years in the London Zoological Gardens was "founded on a clerical error." However, it is a fact and not a statement based on a clerical error that a female squirrel monkey lived 9 years and 9 months in our laboratory. This animal was apparently in good health and pregnant when an unknown disease suddenly brought about its death and, within a few days, the death of 3 other squirrel monkeys and of 3 night monkeys housed in the same room. Bacteriological examinations did not reveal the causative organisms. Since this particular female was at least 3 to 4 years old on arrival, we consider it likely that squirrel monkeys may reach an age of 15 to 20 years. It appears, therefore, that monkey No. 2 (squirrel) which was only about 8 years old at the time of death cannot be considered aged.

Hereditary factor.—Since monkey No. 1 was an Old World monkey and No. 2 a New World monkey, a common hereditary factor may be safely excluded.

Diet.—The diet used for the monkey colony, except for supplementing it with brewers' yeast, was the same as previously described (14) and appears to have been adequate even for South American monkeys. In view of the fact that in the course of a century only 4 births occurred among South American monkeys in the London Zoolog-
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ical Gardens (17), it deserves emphasis that in our laboratory 9 such births occurred during a period of 5 years (1940-1945). Mention should be made of the fact that the daily supply of vegetables and fruits obtained for the animals always included some spoiled and decayed products. Only after cutting off the rotten parts and rinsing or washing the rest under running water was such food offered to the animals. However, there is no doubt that the removal of the visible rotten parts was not always complete and that the monkeys, throughout the years of their existence in the laboratory, often chewed and even consumed decomposed matter of plant origin. Although no hot food was ever offered, boiled food (potatoes) was at times part of the diet of the Old World monkeys.

When received in the laboratory, monkey No. 1 appeared to be somewhat emaciated and in rather poor health. It was listless and frequently had diarrhea, but with the care and diet given in the laboratory its general condition rapidly improved during the first 3 months. In fact, the gain in weight continued so that about 3 years later the monkey was fairly obese and started losing weight only during the last weeks of its life. Nevertheless, it did not always enjoy perfect health since it was at intervals subject to diarrhea and was now and then seen lying on the seat of its cage. Its dull expression and its slow movements when undisturbed made an observer easily forget that it was a powerful and aggressive animal ever ready to exhibit startling speed and agility in attacking any human being within reach and even succeeding in seriously injuring an experienced caretaker. As a rule, normal rhesus monkeys do not accept or eat meat offered to them although fully mature or old animals recently arrived from India may occasionally do so. In testing the food preferences of monkey No. 1 it was observed that the animal accepted and slowly ate pieces of sausages or boiled tongue. It seems likely that it had been forced at times to rely on animal products for food in its natural habitat, where it probably also ac-

Fig. 3.—Photomicrograph of metastatic squamous cell carcinoma in cervical lymph node from primary lesion shown in Fig. 2. Mag. X 125.

Fig. 4.—Photomicrograph of primary lesion of the buccal muco a of monkey No. 2 showing typical infiltrating squamous cell carcinoma. Mag. X 77.
Zuckerman (18) has considered the possibility that seems to be similar to that seen in females (16, 19). Except masturbation during the week following of the sexual skin in normal male rhesus monkeys juvenile monkeys (1). The response of the region male rhesus monkeys of our colony. In monkey at times there was also a reddening as intense as swelling, of the "sexual skin," including the scrotal exhibited a red coloration, unaccompanied by ing and swelling of the sexual skin in the female rhesus monkey develops progressively during the follicular phase (1, 18, 19) and that the inhibitory occurrence of births, the growth curves and general condition of the offspring, it seems unlikely that the lesion at the right angle of the mouth in monkey No. 2 was the result of nutritional deficiency.

Brain lesions.—Since the previously reported carcinomas occurred in monkeys with brain lesions (14), it is of special interest that one of the two cases reported here had also undergone a brain operation. On January 24, 1941, monkey No. 1 underwent extirpation of the left temporal lobe including most of the amygdala, hippocampus and uncus. Anesthesia was induced by an intraperitoneal injection of nembutal. The Bovie high frequency current and blunt spatulae were used in connection with the operation. There was an uneventful recovery.

Hormonal and sex factors.—Throughout the years of its laboratory existence monkey No. 1 exhibited a red coloration, unaccompanied by swelling, of the "sexual skin," including the scrotal skin. There was only an occasional blanching, but at times there was also a reddening as intense as that seen in pregnant females of this species. Incidentally, the same intensive reddening may at the present time be observed in all aged or old male rhesus monkeys of our colony. In monkey No. 1, no overt sexual behavior was ever observed except masturbation during the week following the brain operation. It is known that the reddening and swelling of the sexual skin in the female rhesus monkey develops progressively during the follicular phase (1, 18, 19) and that the inhibitory effects of progesterone on the effects of estrogen on the sexual skin are particularly marked in juvenile monkeys (1). The response of the region of the sexual skin in normal male rhesus monkeys seems to be similar to that seen in females (16, 19). Zuckerman (18) has considered the possibility that the activating agent of the sexual skin in male monkeys is not an estrogenic substance. It is of interest in this connection that Dr. F. C. Koch found 6 international units of androgens (1.22 units/kgm. of body weight) per 24 hours and no estrogens in capon assays on the urine of a sexually mature male rhesus monkey of our colony. However, there was no reddening of the sexual skin at the time the assays were made and red coloration developed only years later. It will be necessary to study the daily urinary excretion of androgens and estrogens in male rhesus monkeys exhibiting continuously a brilliant sexual skin.

In the case of monkey No. 2, the period of visible development of lesion and tumor fell into a period of gestation and lactation. The first signs of pregnancy became manifest 7 weeks before the appearance of the small lesion at the right angle of the mouth. Monkey No. 2 gave birth to a male baby on November 24, 1943 (Fig. 5). We have not seen any published records indicating that birth of a squirrel monkey has ever previously occurred in captivity. Nursing and maternal behavior as well as growth of the baby seemed to be normal in all respects. The infant was only about 14 weeks old when it became necessary to sacrifice the mother, but its development was such that it continued to thrive on solid food and milk.

Infectious and contagious factors.—The 2 monkeys were housed in different rooms of the same building. The cage in which monkey No. 1 was always kept alone stood in a room used as quarters for rhesus monkeys and a number of monkeys belonging to other species. Monkey No. 2 always occupied a cage together with the other squirrel monkeys of the colony. The same room contained cages housing night monkeys and rats. All animals were exposed to the same caretakers and investigators. There was no evidence of tuberculosis or other diseases in the colony during the years monkeys No. 1 and No. 2 lived in the laboratory.

Porphyrins.—In both monkeys the ulcerated portions of the carcinomas exhibited a striking red fluorescence when they were illuminated by light from a mercury vapor lamp passing through a Corning filter No. 5874. The fluorescence spectra of these tissues suggested the occurrence of free porphyrins. In monkey No. 1 the red fluorescence as well as a fluorescence spectrum characteristic of porphyrins were seen only in the cancer tissue of the tongue, but not in the metastatic tumor. In monkey No. 2 it was the surface of the lesion involving the right buccal mucosa which exhibited the most brilliant red fluorescence. When an examination of a cross section through the right cheek was made, it was apparent that the red

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The fluorescence became less intense with increasing distance from the ulcerated surface and that it finally changed into a violet and whitish-violet fluorescence. The lesion at the right angle of the mouth, even after removal of its encrustation, showed a deep red fluorescence. In both monkeys, small portions of the tissues exhibiting red fluorescence were removed for spectrochemical investigation. An extraction of the tissues provided evidence, chiefly based on solubilities and the measurements of fluorescence spectra in different solvents, that the cancer tissue from monkey No. 1 contained deuteroporphyrin and that from No. 2, protoporphyrin. In addition, traces of other ether-soluble porphyrins were present, but the amounts were not sufficient for identification (probably protoporphyrin in the case of monkey No. 1 and coproporphyrin in No. 2). The fluorescence of cancer tissues has engaged the attention of several investigators (2, 4, 5, 7-10, 15). Although porphyrins have been held responsible for the red fluorescence occasionally seen in cancers, it seems that such porphyrins have not been isolated and identified by extracting the cancerous tissues. In examining rat sarcomas, Policard (10) observed that the necrotic center of the inoculation tumor showed an intense red fluorescence surrounded by a narrow violet zone which was again encircled by a peripheral zone showing white fluorescence. Körlber (7, 8) described and photographed the red fluorescence of human ulcerated carcinomas. He was not able, and Policard did not even try, to isolate the porphyrin supposedly responsible for the red fluorescence. Both investigators believed hematoporphyrin to be the agent responsible for the effects observed, but we know at the present time that it is not one of the naturally occurring porphyrins. In considering the origin of the porphyrins found in the ulcerated cancer tissues of monkeys No. 1 and 2, it seems safe to consider, first of all, such factors as bacterial action and the degradation of blood and food remnants. We have previously reported that an intense red fluorescence may be observed in the necrotic wounds of monkeys and that various porphyrins may be extracted from necrotic tissue (6). Neither the extractions of portions of the metastatic tumor in monkey No. 1 nor the spectroscopic examination of the non-ulcerated portions of the tumor in monkey No. 2 provided any definite evidence for the occurrence of porphyrins. It is worth mentioning that the tartar found on the molars of monkey No. 1 exhibited a red fluorescence and a fluorescence spectrum with an emission band at about 620 mμ. Since we have made similar observations on many normal monkeys it is doubtful whether any special significance can be attached to the finding. Extractions of such deposits always furnish one or several ether-soluble porphyrins.

Various environmental factors.—The conditions prevailing in the laboratory as to temperature, illumination, housing, and food utensils have been previously described (14). The animals were under close observation, but no injuries of the tongue were ever noticed. The food never included such items as oats and barley. With exception of the anesthetics mentioned, no drugs were ever administered. Other animals present at various times in the monkey quarters of the laboratory were cockroaches, mice, cats and human beings. Since the laboratory building was once used for work in chemistry, it was considered desirable to check on the presence of radioactivity. Dr. Nickson from the Argonne National Laboratory reported that the spot checks around monkey cages, sinks, door knobs, floors, walls and cabinets failed to reveal any alpha, beta or gamma radioactivity exceeding 100 alpha counts per minute per 100 sq. cm., or 0.1 mμ. per hour at 1 inch.

SUMMARY

Two squamous cell carcinomas occurred in 2 monkeys of a laboratory colony in which 3 squamous cell carcinomas of the tongue in 2 other monkeys had previously been observed. A squamous cell carcinoma of the tongue with local tumor metastases occurred in a male rhesus monkey and a squamous cell carcinoma of the right cheek in a female squirrel monkey. The period of life in the laboratory was approximately 5½ years for both monkeys. The South American squirrel monkey was not considered aged at the time of death. The male rhesus monkey, an animal with an experimental brain lesion of long standing, exhibited an almost continuous reddening of the "sexual skin." In the squirrel monkey the period of the visible development of the tumor fell into a period of gestation and lactation. In both monkeys the ulcerated portions of the carcinomas showed a striking red fluorescence. The porphyrins isolated from the ulcerated tissues were deuteroporphyrin and protoporphyrin. The occurrence of the disease could not be definitely related to any of the etiological factors supposedly responsible for oral carcinomas in man. The fact that of the 30 monkeys in the stock colony of the laboratory 4 have been found to have oral carcinoma (a gross incidence of 13 per cent of this population) and that monkeys belonging to species as different as Macaca mulatta and Saimiri sciurea have been among the affected animals strongly suggests that an extraneous factor may have played an etio-
logical role although there is no indication as yet what this factor may be.

REFERENCES
Oral Carcinoma in a Monkey Colony. A Report of Two Additional Cases

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