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# Reaction pattern of xenografted human salivary glands in nude mice

## An immunohistological and autoradiographical study

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**Summary.** 35 specimens of human parotid gland and 37 of submandibular gland were transplanted into athymic nude mice. At distinct time intervals, from 1 day to 8 months the transplants were collected and examined.

The transplanted glands were studied by light microscopy, immunohistology and autoradiography. The following changes were detectable:

1. acute injury to the xenograft and inflammatory reaction (day 1–7),
2. regeneration of the transplant and the beginning of adaptation to the "mouse milieu" (day 8–30),
3. completion of adaptation (day 30 and later).

The presence of the following substances was analysed: amylase, lactoferrin, secretory component, tissue polypeptide antigen (TPA).

Amylase was only detected in the early transplants. Lactoferrin was seen only in the small duct system. TPA was present during all transplantation periods and was quantitatively correlated with the $^{3}H$ thymidine labeling index.

From our observations we can say that the salivary glands show two different reacting compartments: a large and a small duct system. The histogenesis of the xenografts, and the relationships of the changes observed to human salivary gland diseases were discussed.

**Key words:** Nude mouse – Salivary glands – Heterotransplantion – Immunohistology – Ductal proliferation – Autoradiography.

The nude mouse is an important model system for experimental tumor research. Rygaard and Povlsen (1969) transplanted the first human tumor into this animal, the thymuic deficiency of which was demonstrated by Pantelouris (1968). Povlsen et al. later (1974) demonstrated that normal human tissue could grow in nude mice. Because this animal species offered

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a new approach to study human tumors under "in vivo" conditions, the number of transplantations grew rapidly (see for review: Fogh and Giovanella 1978; Houchens and Ovejera 1978; Shimosato et al. 1979; Bastert et al. 1981).

Morphological studies have been based on conventional analysis generally by H + E staining, showing that the tumor tissue did not undergo much change during the transplantation (Lindenberger 1981; Thomsen et al. 1981; and others).

We intended to study normal human salivary gland tissue during different periods of transplantation. Our objective was primarily a morphological one. The following questions were proposed:

1. Which histological and immunohistological changes are found in normal salivary gland tissue transplanted for different periods?
2. What histogenetic implications be deduced from the reaction pattern of the salivary gland tissue?
3. Can our observations be correlated with pathological conditions in salivary glands?

## Material and methods

Female NMRI nude mice, 6–8 weeks old, were used (Zentrale Versuchstieranstalt Hannover). The animals were kept under normal animal house conditions with addition care, including infection prophylaxis, which was performed every 2 weeks with antibiotics (veterinary control: Dr. med. vet. Dimigen, UKE).

The human tissue was collected in sterile physiological NaCl immediately after surgical excision[1]. The tissue was transplanted into the nude mice, anaesthezied by ether. The site of transplantation was the right and left dorsolateral thoracic wall (Kyriazis and Kyriazis 1980). The size of the xenografts was measured weekly.

The mice were killed at distinct times after transplantation and the following organs were removed:

1. Transplant
2. Mouse tissue
   heart, lung, liver, kidney, intestine, skin and salivary glands.

The material was fixated in Bouin's solution (Romeis 1968), in glutaraldehyde and frozen in liquid nitrogen. The immunohistology was performed on paraffin embedded and on frozen material. In general, we used the "Triple Layer" variant of the immunperoxidase technique (Burns 1978; Sternberger 1979; DeLellis 1981).

The following substances were studied:

1. Enzymes and other cell products: Amylase, lactoferrin, secretory component.
2. "Tumor markers": tissue polypeptide antigen.

The triple layer method is based on following steps:

1. Preincubation with goat normal serum
2. Blocade of the endogenous peroxidase
3. Incubation with the first (specific) antibody
4. Incubation with the second "link" antibody (e.g. goat anti-rabbit antibody)
5. Incubation with the rabbit peroxidase-antiperoxidase complex
6. DAB reaction.

The steps are separated by washings in PBS (phosphate buffered saline; for details see: Burns 1978).

1 We thank Prof. Dr. C. Herberhold, ENT-clinics of the University of Hamburg, for providing the specimens.

**Table 1.** Transplants of human salivary glands in athymic mice

| Time of transplantation | Parotid gland | Submandibular gland |
|---|---|---|
| Day 1–4 | 5 | 2 |
| Day 5–10 | 3 | 5 |
| Day 11–15 | 3 | 3 |
| Day 16–25 | 4 | 6 |
| Day 26–36 | 5 | 5 |
| Day 37–60 | 5 | 9 |
| Over 2 months | 8 | 3 |
| Total | 33 | 33 |

The xenografts were also investigated by autoradiography. At different time intervals after transplantation the animals were injected intraperitoneally 4 × 2 μCi Methyl $^3$H thymidine body weight (specific activity: 25 Ci/mmol, Amersham Buchler).

The mice were killed two hours after the last $^3$H thymidine injection ($^3$H thymidine incorporation time: 8 h). Immediately after the death of the animals, the material was fixed in Bouin's solution. The usual histological treatment followed.

The autoradiography was performed after the immunhistological staining (Stripping film technique, with Kodak Fine Grain Autoradiographic Stripping Plate AR 10). Film exposition time was 21 days.

By this combined immunhistological-autoradiographical technique we could mark TPA (a "proliferation antigen") and determine the $^3$H thymidine labeling index (l.i.) in one section. This index is a quantitative measure of cell proliferation.

Furthermore we analysed the correlation between the labeling index and the TPA positive cells (for method see: Fitzgerald et al. 1968; Schmiegelow et al. 1982).

*Antibodies*

Antibodies were available from the following sources: Anti-TPA antibody from the rabbit was a kind gift of Prof. Dr. B. Björklund, Stockholm.

Anti-amylase antibody from the rabbit was a kind gift of Dr. Grenner Behringwerke/Marburg.

Antibodies against lactoferrin, secretory component were purchased from Dako/Copenhagen.

The anti-rabbit antibody from the goat, the rabbit peroxidase-antiperoxidase complex were purchased from Medac, Hamburg.

## Results

35 pieces of parotid and 37 pieces of submandibular gland were transplanted (see Table 1).

6 animals died before the experiment was completed and so we examined 33 parotid and 33 submandibular xenografts.

Macroscopically, the transplants were nodules on the back of the animals. After the first 14 days, the size of the transplant remained relatively constant.

Microscopically the general reaction pattern of the two glands was similar. The changes in the parotid gland and submandibular gland are described together, were relevant differences between the two glands are indicated.

*Light microscopy*

*Day 1–7.* The main features in this period were extensive necrosis (up to 95 percent of the transplant) and intense inflammation (Fig. 1).

By the 3rd day a large part of acinar cells and duct cells was necrotic. At the end of this period a zonal organization in the xenograft appeared with a marginal zone of 10–15 cells layers, an "intermediate zone" of 8–10 cells layers and central necrosis.

The cells at the *margin* showed only slight signs of alteration. In the *intermediate zone* the cells were in a necrobiotic state, were swollen, with some pyknotic nuclei. In the *center* the cells were entirely necrotic and inflammatory cells (lymphocytes and granulocytes) of mouse origin were generally found.

Myoepithelial cells and mucous cells were relatively resistant to these changes.

Totally intact acinar cells, laying in the marginal zone, were only found in the very early part of this period. At day 7 most of the acinar cells were severely damaged or even necrotic. The lumen of ducts showed the beginning of enlargement.

*Day 8–30.* The following changes were present:

More and more collagenous connective tissue was found in the necrosis. The original structures of the gland had disappeared. The large number of lymphocytes, granulocytes and mast cells diminished. The "intermediate zone" disappeared slowly.

In the marginal zone there were important changes in the gland tissue:

The acinar cells were necrotic at about the 11th day. The small ducts were arranged in small groups surrounded by fibrous tissue similar to intercalated duct cells (Fig. 2a). The small ducts generally showed a double cell layer of cuboidal duct and myoepithelial cells.

The large ducts were surrounded by cylindrical epithelium, generally in two layers, with large duct cells and myoepithelial cells. They were generally cystically enlarged. Squamous cells, goblet cell and sebacous cell metaplasia were seen. The implanted tissue was surrounded by a capsule of connective tissue, up to the 12th day. This well established capsule remained during the following observation periods.

*Day 30 and later (up to 8 months).* From the 30th day, the "period of changing" was finished and the xenografts were adapted to the mouse milieu.

The pattern of long term xenografts were:

1. The necrosis had disappeared, only connective tissue with fibroblasts and fibrocytes was seen.
2. An "intermediate zone" could not be separated any longer.
3. The marginal zone was larger, in this zone large and small duct systems were situated. Acinar cells could not be detected.

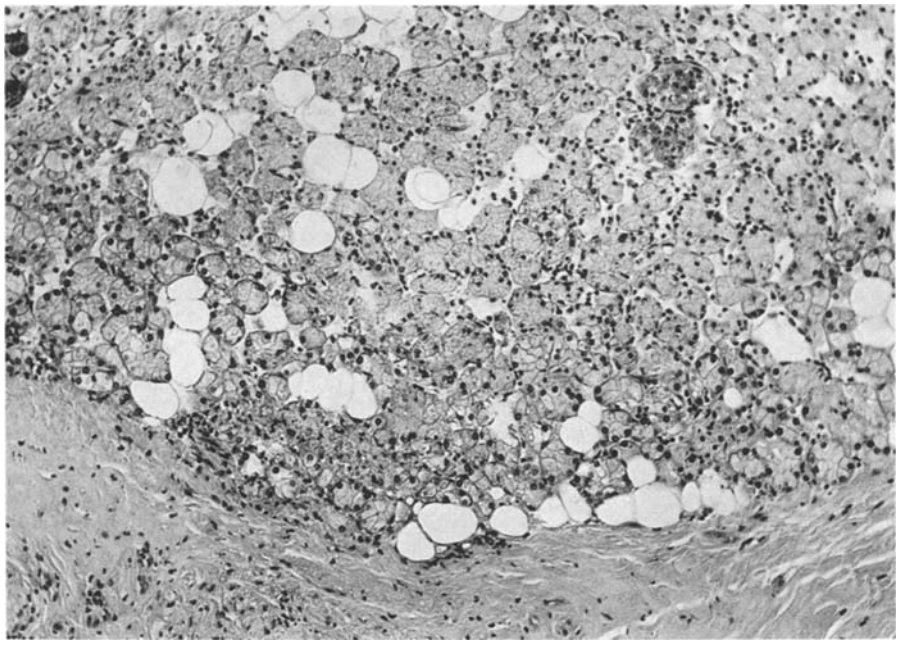

**Fig. 1.** Parotid gland in athymic mouse. Day 1. *Top*, necrotic acini; at the border of the transplant (*bottom*) some intact structures. HE. Magnification ×120

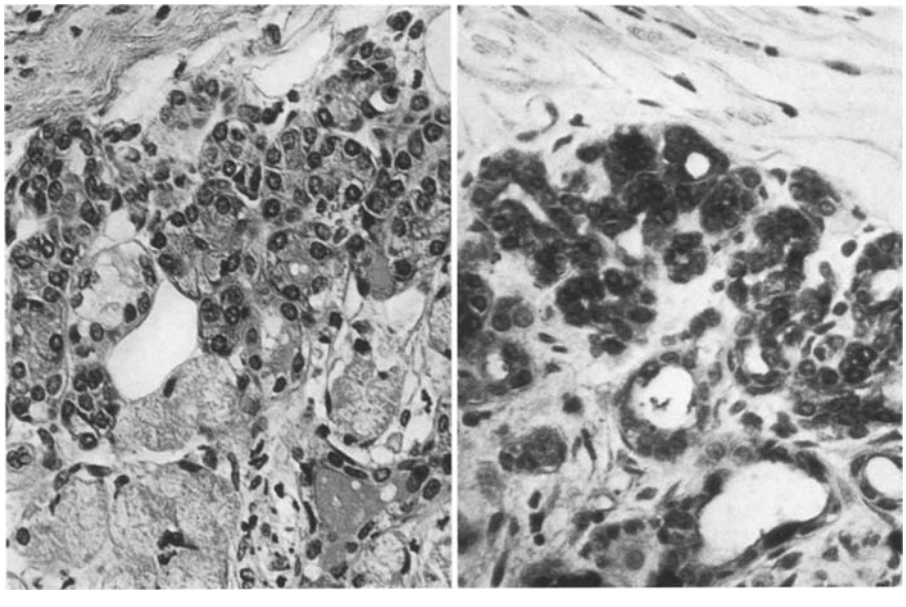

**Fig. 2.** **a** (left) Submandibular gland in athymic mouse. Day 7. In the regeneration zone, there are some small ducts lined by a cuboidal epithelium. HE. Magnification ×300. **b** (right) Submandibular gland in athymic mouse. Day 15. Small ducts in regeneration zone are lactoferrin positive. Immunoperoxidase reaction for lactoferrin. Magnification ×300

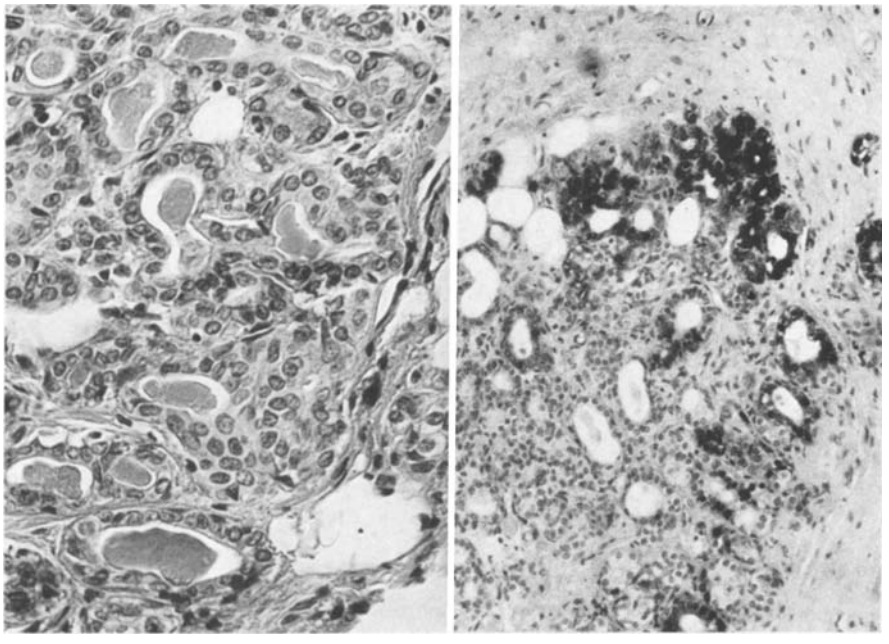

**Fig. 3. a** (left) Parotid gland in athymic mouse. More than 30 days. Predominantly small ducts. HE. Magnification ×300. **b** (right) Parotid gland in athymic mouse. More than 30 days. Lactoferrin positive small ducts. Immunoperoxidase reaction for lactoferrin. Magnification ×120

The small duct system was constructed of two cell layers, cuboidal duct cells, myoepithelial cells and "clear cells" (Fig. 3a). The lumen was either obstructed by solid epithelial cell sheats or by squamous cell metaplasia. In a few sections the lumen was dilated, in this lumina we found detritus and secretory material.

The large ducts showed two or more cell layers (Fig. 4), following types of cells were found: large duct cell, myoepithelial cell and "clear cell". Goblet cell and sebacous cell metaplasia were seen. The lumen was dilated and cystes were formed, filled with detritus and secretory products. Through epithelial cell proliferation, the ducts acquired a cribriform aspect.

All the ducts were surrounded by a fibrosis, the large ducts were laying singularly, the small ones were laying in a lobular group.

The whole xenograft was surrounded by a connective tissue capsule.

These characteristic pictures did not change in subsequent periods time.

*Immunohistology*

*Amylase* was only found in acinar cells of the marginal zone (day 1 to 7). In the beginning, about 30% of the cells were positive. After the 10th day, no amylase was detected.

*Secretory component* disappeared even faster. It was found in the duct only during the first 3 days.

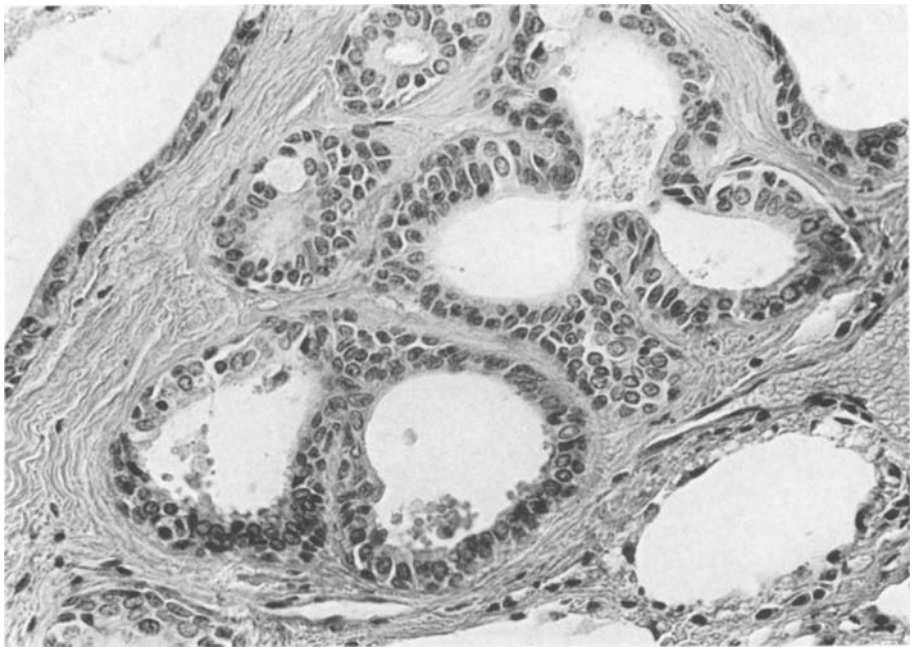

**Fig. 4.** Parotid gland in athymic mouse. More than 30 days. Predominantly large ducts. HE. Magnification ×300

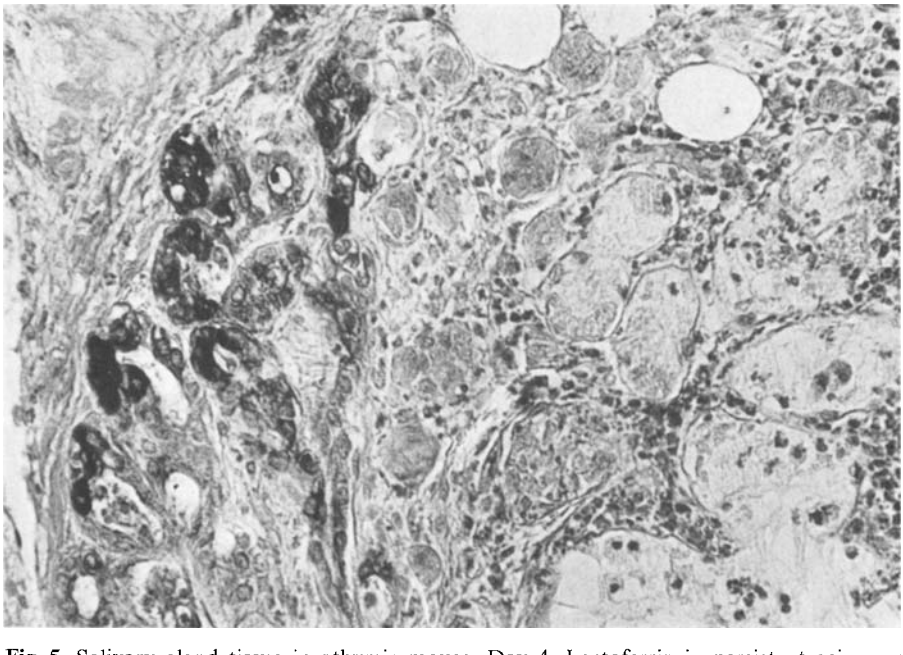

**Fig. 5.** Salivary gland tissue in athymic mouse. Day 4. Lactoferrin in persistent acinar cells and cells of the intercalated ducts. To the right, necrotic area. Immunoperoxidase reaction for lactoferrin. Magnification ×300

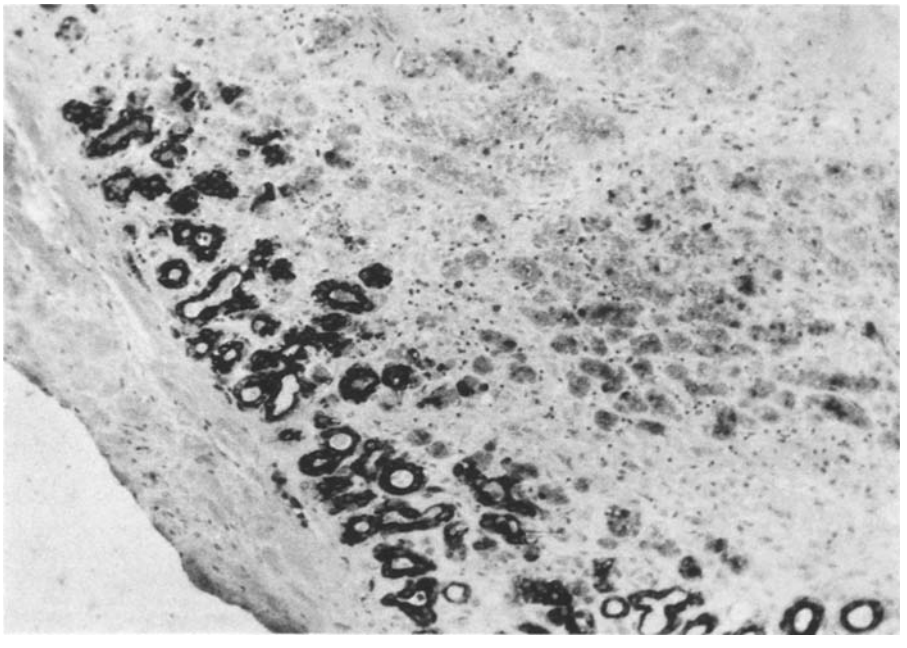

**Fig. 6.** Salivary gland tissue in athymic mouse. Day 6. TPA-positive small ducts at the margin of the transplant. Immunperoxidase reaction for TPA. Magnification ×120

*Lactoferrin* was detected in all periods. From day 1 to 7, up to 90% of all marginal cells were stained. In the next periods, only the small duct cells located at the margin were positive (Fig. 2b, 3b, 5).

*TPA* was present in the surviving duct cells (up to 40%). The acinar cells were generally negative. The positive duct cells sometimes showed a diffuse cytoplasmic staining of a larger number of cells (Fig. 6). Sometimes there was a "mosaic-like" pattern of strongly positive and negative cells. Some cells showed a predominant staining of the apical cytoplasm.

*Autoradiography*

For autoradiography, we used 25 salivary glands (12 parotid glands and 13 submandibular glands). Our main interest was to study the first and the second period (9 parotid glands and 10 submandibular glands). For determination of the labeling index (l.i.) we counted 250 small duct cells and 250 large duct cells. There were the following results:

Up to the *5th day*, the xenografts showed a small l.i., maximum 2.5 percent of the cells counted.

From the *5th day*, there was a strong increase of the l.i. At the 6th day 8 percent and at the 7th day 10 percent of the cells were labeled. This day showed the highest range of labeled cells.

Associated with the increase of labeling index, the marked cells became TPA positive (double labeling technique).

From the *7th day*, the l.i. decreased. At the 8th day 5 percent and at the 11th day only 3 percent were labeled. After this time up to 8 months, the l.i. remained constant between 1.5 percent and 2.5 percent.

Beginning at the 5th day, the part of TPA positive cells was about 80 percent.

Differences between the two salivary glands were not evident.

## Discussion

Shortly after the discovery of the typical features of the nude mouse, tumor transplantations in large numbers were performed (Giovanella et al. 1978; Reid et al. 1978; Fogh et al. 1980; Kyriazis and Kyriazis 1980; Lindenberger 1981; Caselitz 1982a, b; Barkla and Tutton 1983; and others). The morphological studies done on xenografted tumors were only undertaken to control the integrity of the histological structure. A small number of these studies were performed on tissue of the head and neck region, and these did not include normal salivary gland tissue nor use modern immunohistochemical techniques (Lindenberger et al. 1978; Wennerberg et al. 1983).

We wanted to study the changes of normal human salivary glands in athymic mice during a long-term transplantation. Our purpose was to discover the typical reaction pattern of this tissue under these conditions. In particular the application of immunohistochemical techniques should answer the question of whether the functional/structural integrity of the tissue is impaired.

We chose the parotid and submandibular glands and examined their behaviour in the nude mouse.

Depending on the time of transplantation, the reaction pattern of the salivary glands can be divided into three groups:

1. Day 1 to 7
2. Day 8 to 30
3. Day 30 and longer.

In the first period, necrosis was the leading feature. In the second period, we found the beginning of regeneration phenomena and after 30 days the transplant is entirely adapted to the nude mouse. In the following months, no additional features are seen if there is no infection or mechanical irritation.

In the first period, glandular structures in the center of the transplant are destroyed. This is explicable by the long diffusion pathways in the xenograft which is not yet vascularized. The well differentiated acinar cells are very sensitive to transplantation and light microscopy showed vacuolisation of their cytoplasm and nuclear pyknosis. The disappearance of amylase staining is correlated with this finding.

The same alterations are observed in immunohistological stainings for secretory component. However, the presence of lactoferrin seems to be quite stable, especially in the small duct system, which is related to the intercalated ducts.

These histological patterns correlate with acute sialadenitis (Seifert 1966). We see necrosis of the acinar cells and duct cells and massive inflammation with granulocytes. In our model, the source of the granulocytes is the host organism, the mouse. As a consequence of transplantation of small pieces, we see changes in saliva production and transportation (Proteo-dyschylia) (Seifert 1966). This aspect of the pathogenesis is explained by obstruction. Parallel experimental data are available from rodents. Duct ligation in all major salivary glands have been performed (Standish and Shafer 1957; Berger 1973; Donath et al. 1973; Tamarin 1979) and disappearance of acinar cells at the 10th day correlates with our results. The resistance of myoepithelial cells and of the mucous cells is found both in obstructive experiments and in the transplantation model.

The alterations in the xenografts may be influenced by vascular disorders. Thus, there is a certain relation to necrotizing sialometaplasia (Abrams et al. 1973; Donath 1979; Dunley and Jacoway 1979; Lynch et al. 1979; Walker et al. 1982). Necrotizing sialometaplasia is currently explained as an infarct of the parotid gland (Donath 1979) but other explainations have been made (Forney et al. 1977; Gad and Willen 1980).

The changes after parotid infarction (Donath 1979) resemble those of the first week after transplantation. Therefore vascular disturbance may be an additional factor in this period.

Acute changes have also been found by other groups studying carcinomas of the large intestine (Barkla and Tutton 1983).

The main changes after the first week of transplantation are regeneration phenomena. In early stages there is still some necrosis with granulocytic reaction, but signs of regeneration in the transplant are evident in addition.

There are now three zones in the xenograft. The regeneration phenomena are seen in the marginal zone of about 10 to 15 cell layers, where the transplant has the best vascularity. In the second zone, again 8 to 10 cell layers, there are necrobiotic changes. In the center, we observe complete necrosis where no intact glandular structures can be seen.

Regeneration in the marginal zone shows that the terminal duct segments, including acinar cells and intercalated duct cells, and the larger ductal segments, including the striated ducts and large excretory ducts, are reacting as two systems. The small ducts retain the appearance of "small" ducts, resembling the intercalated ducts and similar to the findings in obstructive sialadenitis. The cytology of the cells resembles those of the intercalated duct cells. Myoepithelial cells and some clear cells are seen. Mitoses are relatively frequent about the first week. The striated and the excretory duct retain the aspect of a "large duct". The cytology of the duct cells shows a more cylindrical type with some relation to the striated duct cells. Clear cells and myoepithelial cells are regularly seen. Mitoses are sometimes observed.

Immunohistologically, again, we can distinguish between small and large duct systems by the staining for lactoferrin. The lack of lactoferrin in the large duct system may be interpreted as showing a different histogenetic origin for this system (Caselitz et al. 1982a). This interpretation is in accor-

dance with the theory of Eversole (1971) that there are two different reacting compartments of reserve and stem cells: one for the terminal duct system and one for the large duct system.

Another interesting aspect concerns TPA which is considered to be a proliferation or differentiation antigen (Holyoke and Chu 1979; Björklund 1980). TPA is seen in the duct cells of the salivary glands, but not in the acinar cells, this fact identifying the proliferative compartment. The duct cells show a different staining (Caselitz et al. 1983). The most intense staining for TPA is found in regenerating small and large duct systems. This pattern correlates with the $^3H$ thymidine labeling index. At the 7th day we have the highest l.i., and the $^3H$ thymidine positive cells were also marked by TPA.

These histological alterations of the xenograft finished at the 30th day. Acinar cells were no longer seen, having only small and large ducts. The small ducts have a cubicoidal epithelium, a myoepithelial cell layer and some "clear cells". The small ducts were stained with lactoferrin and TPA.

The large ducts had a cylindrical epithelium of multiple layers, myoepithelial cells and "clear cells". They were generally dilated and sometimes cystic. Squamous cells, goblet cells and sebaceous cell metaplasia were seen.

This terminal state is similar to chronic obstructive sialadenitis (Seifert 1966; Berger 1973; Donath et al. 1973; Seifert and Donath1976), where small and large ducts with periductal fibrosis are seen.

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## References

Abrams AM, Melrose RJ, Howel FV (1973) Necrotizing sialmetaplasia: A disease simulating malignancy. Cancer 32:130–135

Barkla DH, Tutton PJM (1983) Temporal morphologic changes in human colorectal carcinoma following xenografting. Am J Pathol 110:315–321

Bastert GBA, Fortmeyer HP, Schmidt-Mathiesen H (1981) Thymusaplastic nude mice and rats in clinical oncology. Gustav Fischer, Stuttgart New York

Berger R (1973) Zur Ultrastruktur der Submandibularis nach Unterbindung des Wharton'schen Ganges. Med Dissertation. Universität Hamburg

Björklund B (1980) On the nature and clinical use of tissue polypeptid antigen (TPA). Tumor Diagnostik 1:9–20

Burns J (1978) Immunohistological methods and their application in the routine laboratory. In: Anthony PP, Woolf N (eds) Recent advances in histopathology, No 10. Churchill Livingstone, Edingburgh London New York, pp 337–350

Caselitz J, Becker K, Wustrow J, Thayssen G (1982a) Analyse von menschlichen Speicheldrüsentumoren auf thymusaplastische Mäuse. Immunhistochemische und elektronenoptische Untersuchungen. Verh Dtsch Ges Path 88:466

Caselitz J, Sendler A, Schmiegelow P, Salfelder A (1982b) Morphologische Reaktionsmuster von transplantiertem normalem Speicheldrüsengewebe in thymusaplastischen Mäusen. Verh Dtsch Ges Pathol 66:615

Caselitz J, Seifert G, Björklund B, Björklund V (1983) Detection of TPA (Tissue polypeptide antigen) in salivary glands and salivary gland tumours. Appl Pathol 1:115–120

DeLellis RA (1981) Diagnostic immunohistochemistry. Masson Publishing USA Inc., New York Paris Barcelona Milan Mexico City Rio de Janeiro
Donath K (1979) Pathohistologie des Parotisinfarktes (necrotizing sialometaplasia). Laryng Rhinol 58:70–76
Donath K, Hirsch-Hoffmann KU, Seifert G (1973) Zur Pathogenese der Parotisatrophie nach experimenteller Gangunterbindung. Virchows Arch [Pathol Anat] 359:31–48
Dunley RE, Jacoway JR (1979) Necrotizing sialometaplasia. Oral Surg 47:169–172
Eversole LR (1971) Histogenetic classification of salivary tumors. Arch Pathol 92:433–443
Fitzgerald PJ, Carol B, Lipkin L, Rosenstock L (1968) Pancreatic acinar cell regeneration. V. Analysis of variance of the autoradiographic labeling index (thymidin-$H^3$). Am J Pathol 53:953–970
Fogh J, Giovanella BC (1978) The nude mouse in experimental and clinical research. Academic Press, New York San Francisco London
Fogh J, Orfeo T, Tiso J, Sharkey FE, Fogh JM, Danies WP (1980) Twenty-three new human tumor lines established in nude mice. Exp Cell Biol 48:229–239
Forney SK, Foley JM, Sugg WE, Oatis GW (1977) Necrotizing sialometaplasia of the mandible. Oral Surg 43:720–726
Gad A, Willen H (1980) Necrotizing sialometaplasia of the lip simulating squamos cell carcinoma. Histopathology 4:111–121
Giovanella B, Stehlin JS, Williams LJ, Lee SS, Shepard RC (1978) Heterotransplantation of human cancers into nude mice. Cancer 42:2269–2281
Holyoke ED, Chu TM (1979) Tissue polypeptide antigen. In: Heberman RB, McIntire KR (eds): Immundiagnosis of cancer. Dekker, New York, pp 513–521
Houchens DP, Ovejera AA (1978) Proceeding of the symposium on the use athymic (nude) mide in cancer research. Gustav Fischer, New York Stuttgart
Kyriazis A, Kyriazis AP (1980) Preferential sites of growth of human tumors in nude mice, following subcutaneous transplantation. Cancer Res 50:4509–4511
Lindenberger J, Ganzer U, Fortmeyer HP (1978) Die Heterotransplantation bösartiger Kopf- und Halsgeschwülste auf thymusplastisch nackte Mäuse. Arch Otorhinol 220:117–128
Lindenberger J (1981) Aspects of xenografted tumor of the ear, nose and throat. Morphology, cell kinetics, growth. Behavior and immunology. In: Bastert GBA, Fortmeyer HP, Schmidt-Matthiesen H (eds) Thymus-aplastic nude mice and rats in clinical oncology. Gustav Fischer, Stuttgart New York, pp 449–466
Lynch DP, Crago CA, Martinez MD (1979) Necrotizing sialometaplasia. Oral Surg 47:63–69
Pantelouris EM (1968) Absence of thymus in a mous mutant. Nature 217:370
Povlsen CO, Skakkebaek NE, Rygaard J, Jensen G (1974) Heterotransplantation of human foetal organs to the mouse mutant nude. Nature 248:247–249
Reid LM, Holland J, Jones C, Wolf B, Niwayama G, Williams R, Kaplan NO, Sato G (1978) Some of the variables affecting the success of transplantation of human tumors into the athymic nude mouse. In: Houchens DP, Ovejerra AA (eds) The use of athymic (nude) mice in cancer research. Gustav Fischer, New York Stuttgart, pp 107–121
Romeis B (1968) Mikroskopische Technik. Ouldenburg, München
Rygaard J, Povlsen CO (1969) Heterotransplantation of a human malignant tumor to "nude mice". Acta Pathol Microbiol Scand 77:758–760
Schmiegelow P, v Dahl M, Löning T (1982) Zur Zellkinetik und Wundheilung mehrschichtiger Plattenepithelien. Experimentelle autoradiographische, immunhistologische und ultrastrukturelle Untersuchungen. Verh Dtsch Ges Pathol 66:453
Seifert G (1966) Mundhöhle, Mundspeicheldrüsen, Tonsillen und Rachen. In: Doerr W, Seifert G, Uehlinger E Spezielle pathologische Anatomie, Bd I. Springer, Berlin Heidelberg New York, pp 1–415
Seifert G, Donath K (1976) Die Morphologie der Speicheldrüsenerkrankungen. Arch Otolaryngol 213:111–208
Shimosato Y, Kameya T, Hirohashi S (1979) Growth, morphology, and function of xenotransplanted human tumors. Pathol Annu 14:215–257
Standish SM, Shafer WG (1957) Serially histologic effects of rat submaxillary and sublingual salivary gland duct and blood vessel ligation. J Dent Res 366:866–879

Sternberger LA (1979) Immunohistochemistry. John Wiley and Sons, New York Chichester Brisbane Toronto

Tamarin A (1979) The leukocytic responce in ligated rat submandibular glands. J Oral Pathol 8:293–304

Thomsen D, Neville AM, Phelan K, Schartz-Sconzano R, Vandevoorde JP (1981) Human cancers transplanted in nude mice retain the expression of their organ specific neoantigen. Eur J Cancer Cell Oncol 17:1191–1197

Walker GH, Fechner RE, Johns ME, Teja K (1982) Necrotizing sialometaplasia of the larynx secondary to atheromatous embolization. Am J Clin Pathol 77:221–223

Wennerberg J, Tropè C, Björklund A (1983) Heterotransplantation of human head and neck tumors into nude mice. Acta Otolaryngol 95:183–190

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