

## Spontaneous Tumours in Guinea Pigs

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### Abstract

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The aim of the study is to describe spontaneous tumours in guinea pigs. Twenty neoplasias from 19 guinea pigs were examined histologically. In 15 cases biopsy samples were examined, samples from four animals were collected during autopsy. Except for one, all animals were kept as pets. Skin tumours were diagnosed in five of them. They appeared in different locations - abdomen, plantar side of hind leg, back (in two animals), and rump, and were of different sizes, the largest one was five cm in diameter. All tumours were of follicular origin - two trichofolliculomas, two trichoepitheliomas, one malignant pilomatricoma. The age of affected animals ranged from two to 7.5 years. Tumours of the mammary gland were present in five guinea pigs. Adenocarcinoma was diagnosed in two males, sarcoma of myoepithelial origin was found in one female. Tubular adenoma was present in one two-year-old female, and adenomatous hyperplasia of the mammary gland was observed in another female of the same age. In six guinea pigs, three females and three males, between three and five years of age, there were tumours in subcutaneous tissue. Three were lipomas, in one animal the lipoma was multiple. Liposarcoma was found in one male, myxoid liposarcoma was diagnosed in another one. Ossifying fibroma was histologically diagnosed in one female. Lymphatic leukaemia was observed in three males. All animals were 4-year-old. Hepatocellular adenoma was found in a 5-year-old female suffering also from trichofolliculoma as mentioned above. Data about tumours in guinea pigs are relatively rare, and therefore information along this line is useful both for clinical practice and comparative pathology.

*Histopathology, neoplasia, skin, mammary gland, soft tissues, haemopoietic tissue, comparative pathology*

Spontaneous tumours in guinea pigs are, according to literature data, rather rare. With exception of leukaemia in certain inbred strains, neoplasias are practically non-existent in animals less than 1 year of age (Wagner and Manning 1976). In animals surviving three years the frequency of tumours is as high as 15% (Blumenthal and Rogers 1965). In some laboratory strains, animals older than three years, had tumour incidence ranging from 14.4% to 30% (Wagner and Manning 1976). General overview of tumours in guinea pigs is presented by Blumenthal and Rogers (1965), Wagner and Manning (1976), Squire et al. (1978), and Percy and Barthold (1993). Report of 14 spontaneous tumours was published by Kitchen et al. (1975), lymphoblastic leukaemia in two strains was described by Hong et al. (1970). Zwart et al. (1981) have described three cutaneous tumours in guinea pigs.

Guinea pigs, like rabbits and rats, are becoming more and more popular as pet animals; e.g. they live in 8.2% of Czech pet-keeping households along with dogs, as indicated by a recent survey (Baranyiová et al. 2001). Guinea pigs usually survive for about three years, and when adult or old they suffer, in addition to other diseases, also from neoplasias. Therefore, knowledge about tumours in guinea pigs is increasingly important for both clinical practice and comparative pathology.

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## Materials and Methods

All but one animal were kept as pets in households. They were both short-haired and long-haired, mainly tricolor, but also black, without any genetic identification. One guinea pig, suffering from leukosis, originated from a laboratory animal colony and belongs to strain C2BB/R+.

Samples submitted for histological examination were predominantly biopsies obtained by surgical extirpation. Only in four cases they were collected in the course of necropsy of euthanized guinea pigs carried out by veterinarians. Samples were fixed in 10% buffered formalin and processed by common paraffin technique. Histological sections 4 mm of thickness were, after deparaffinization, stained with haematoxylin and eosin. In indicated cases were performed staining after Giemsa, PAS reaction, alcian blue at pH 2.5 with PAS reaction, and impregnation after Gomori. Malignant pilomatricoma in skin and four tumours of mammary gland were examined also by means of immunohistochemistry. Cytokeratins were proved by monoclonal antibody MNF 116 (DAKO), cytokeratin K18, expressed in monolayer epithelium, was identified by monoclonal antibody DC-10 (EXBIO), smooth muscle actin was determined by antibody HHF35 (DAKO), vimentin by monoclonal antibody clone V9 (DAKO), and S 100 protein was identified by polyclonal antibody (DAKO). Immunohistochemistry was performed on sections from paraffin material by the common immunoperoxidase method. Endogenous peroxidase activity was quenched by 3% peroxide at room temperature for 15 min. For detection of cytokeratins the slides were digested with trypsin (0.1% in 0.1% calcium chloride) for 15 min at 37°C. The remaining antigens were demasked by boiling of the slides in 0.1 M citrate buffer pH 6.0 for 10 min. Binding of antibodies was performed in humid chambers at room temperature for 60 min. The reaction was visualized by means of streptavidin-biotin universal detection system (Immunotech). The sections were counterstained with haematoxylin.

## Results

### Tumours in the skin

Table 1  
Tumours of the skin

No.	Age – years	Sex	Diagnosis
1	7	F	Malignant pilomatricoma
2	Unknown	F	Trichofolliculoma
3	2	M	Trichoepithelioma
4	2.5	M	Trichoepithelioma
5	5	F	Trichofolliculoma

In guinea pig No. 2 there were two tumours. One was in rump, the second was in shoulder region. In animals No. 3 and 4 the tumours were located in back and rump regions, respectively and in animal No. 5 the neoplasia was in abdominal region. The tumours were 1-5 cm in diameter. On cut surface of trichoepitheliomas and trichofolliculomas there was macroscopically apparent pasty material.

Trichofolliculomas were composed of more primary follicular formations that were cystically dilated and keratinized through a granular cell layer. From the primary follicles multiple secondary follicles of different stage of maturation radiated outward.

Trichoepitheliomas were composed of random admixture of budding epithelial islands and cystic structures. The islands were composed of basaloid cells with peripheral palisading. In addition to infundibular keratinization there was also matrical keratinization. Epithelial islands were encompassed with fibrous or myxomatous stroma.

Malignant pilomatricoma was located on the plantar side of hind leg. The tumour was dome-shaped, approximately 2 cm in diameter. According to histological structure the tumour consisted of three parts. One was well differentiated pilomatricoma, composed of multiple cystic formations of different size that were lined predominantly by basaloid keratinocytes. Zones of squamocellular epithelium, mainly without granular cell layer, were also present. The basaloid cells had scant cytoplasm and ovoid, hyperchromatic nuclei. Mitotic activity was mild to moderate. Among and inside of the cells there were many apoptotic bodies. Lumina of cysts contained predominantly masses of keratinized ghost cells but lamellar keratin was also present.

The second part of the tumour consisted of islands of epithelial cells separated by connective tissue. Many islands were solid, others contained keratin or ghost cells in their central part and small solid foci or thin bands of epithelial cells grew out from their periphery. In the islands the epithelium was differentiated into smaller cells of matrical nature and larger cells with clear or slightly eosinophilic cytoplasm and ovoid nuclei that contained fine chromatin. Nucleoli in neoplastic cells were apparent and mitotic activity was fairly high. Many apoptotic bodies were either among the epithelial cells or they were phagocytized by them. Rudimentary sebaceous glands were attached to the periphery of many epithelial islands. The third part was composed of spindle, basaloid cells arranged in irregular islands and bundles. Cytoplasm of the neoplastic cells was rather basophilic, nuclei were ovoid with fine chromatin and with one or more conspicuous nucleoli. Mitotic activity was rather high and atypical figures were also present. Among and inside of the cells there were many apoptotic bodies. Matrical keratinization was minimal. Multiple rudimentary sebaceous glands or cells were inside the islands of neoplastic cells. Moreover there were multiple foci of metaplastic lamellar bone tissue (Plate VII, Fig. 1). In this neoplastic tissue were multiple necrotic foci and the periphery of the tumour was necrotic with mixed inflammatory cellulation.

Using monoclonal antibody MNF 116 (DAKO), positivity of cytokeratins was proved in majority of epithelial cells in the first part of the tumour. In the second part predominantly the cells in outgrowths of the islands were positive (Plate VII, Fig. 2). In the third part, the neoplastic cells were negative with exception of single small groups of differentiated epithelial cells. Positivity of S100 protein was almost identical with the reaction of pancytokeratin antibody. Cytokeratin K 18, typical of simple epithelium, was not observed in neoplastic cells, only in the sebaceous glands there was mild positivity. Vimentin was present in fibrocytes of the connective tissue and in the cells at the periphery of metaplastic bone tissue in the third part of the tumour. In accordance with Goldschmidt et al. (1998) the tumour was diagnosed as a malignant pilomatricoma.

## Tumours of the mammary gland

Table 2  
Tumours of the mammary gland

No.	Age – years	Sex	Diagnosis
1	4	M	adenocarcinoma
2	7	M	adenocarcinoma
3	4	F	myoepithelial sarcoma
4	2	F	adenomatous hyperplasia
5	2	F	tubular adenoma

In male No. 1, a tumour of left mammary gland, size of 2 ¥ 1 ¥ 1 cm, had been observed for four months. In male No. 2 the tumour was dome-shaped, 2 cm in diameter. Both tumours were histologically diagnosed as adenocarcinomas. They were arranged in tubular and cystopapillar formations, and in the second case there were also foci of squamous metaplasia and conspicuous inflammatory cellulation. Angioinvasion was not apparent.

Tumour from animal No. 2 was examined by means of immunohistochemistry. Reaction with pancytokeratin antibody (MNF 116, DAKO) was only slight in the columnar epithelium whereas in the squamous epithelium it was strong. Reaction with antibody to cytokeratin K 18 (Exbio) was positive in the columnary epithelium in tubular and papillar structures (Plate VIII, Fig. 3). Detection of smooth muscle actin revealed nodular proliferations of the myoepithelium. In tubular formations the myoepithelial cells were only scarce.

The dimensions of the sarcoma of myoepithelial origin were 2 × 1 × 1 cm. The tumour was rather well limited. Histologically, it consisted of interlacing bundles of spindle-shaped cells. The nuclei were oval, round or irregular with fine chromatin and only in some of them were conspicuous nucleoli. Mitotic figures were rather frequent, some of them were atypical. Cytoplasm was pale basophilic and reticulated, cytoplasmic membrane was not well visible. Angioinvasion was not observed. Immunohistochemistry revealed positivity for smooth muscle actin and S100 protein in the neoplastic cells. The reactions for cytokeratins and vimentin were negative.

In animal No. 4, a tumourous formation, 2 cm in diameter, was present. Histologically it consisted of tubular glandular structures arranged in islands of different sizes and shapes that were disseminated in loose connective and fat tissues. Tubuli of different diameter with proteinaceous secretion in their lumina prevailed in the adenomatous tissue. The epithelium was predominantly cubic, here and there with sign of apocrine type of secretion. No mitotic figures and no cytologic abnormalities were observed. Some rudimentary hair follicles with sebaceous glands or cells were also present. The lesion was diagnosed as adenomatous hyperplasia of the mammary gland. Immunohistochemical examination for cytokeratins, using pancytokeratin antibody MNF 116 (DAKO), revealed positivity only in the epithelium of hair follicles. Epithelium of the tubular formations was negative.

Reaction for smooth muscle actin was positive in majority of the interstitial cells and in some epithelial cells in the adenomatous formations. This reaction revealed well the myoepithelial cells situated at the base of the tubuli.

In guinea pig No. 5, the nodule in the mammary gland was approximately 1 cm in diameter. Its histological structure was characteristic for tubular adenoma. The tubules were small in diameter and contained proteinaceous material in the lumina. Results of immunohistochemistry were the same as in the above mentioned case, only the reaction for actin revealed more myoepithelial cells.

## Tumours in the subcutaneous tissue

Table 3  
Tumours of the subcutaneous tissue

No.	Age – years	Sex	Diagnosis
1	4	M	Lipoma
2	5	M	Liposarcoma
3	5	F	Lipoma
4	4	F	Lipoma
5	5	F	Ossifying fibroma
6	3	M	Myxosarcoma

Lipomas. In guinea pig No. 1, the tumour was located on the right side of the thoracic wall and its size was 2 × 1 × 1 cm. In animal No. 3, the tumour was 2-3 cm in diameter and it was located in the pubic region. Multiple lipomas were in guinea pig No. 4. They were located on the ventral side of the body, in the right axilla, on the right side of thorax, and they were of different sizes. Unfortunately, the clinician did not determine their size.

Liposarcoma was situated in the left groin and its dimensions were 5 × 3 × 3 cm. This tumour reached this size in the course of one month. Histologically, it was relatively well differentiated liposarcoma. The majority of neoplastic cells contained in the cytoplasm large fat vacuol surrounded by narrow rim of cytoplasm. In the nuclei was fine chromatin and anisokaryosis was apparent. Mitotic activity was low (Plate VIII, Fig. 4).

Myxoid liposarcoma was located on dorsal part of the neck. The tumour had been observed for one month and during this period it reached 1 cm in diameter. Histology revealed tumourous tissue which consisted of large quantity of vacuolated amorphous intercellular substance. Neoplastic cells were polymorphous with cytoplasmic processes. The amorphous intercellular substance contained acid glycoproteins. Vacuoles were residui of fat that was extracted during histological processing. Infiltration of tumourous tissue into the surrounding tissues was well apparent. Diagnosis was made on basis of description by Hendrick et al. (1998).

Ossifying fibroma was situated on ventral side of thorax. It was 10 cm long and its transversal dimensions were 2 × 1 cm. Histological structure consists of proliferative collagen connective tissue with conspicuous bone metaplasia. The neoplastic tissue was not encapsulated but it was well limited and no histological signs of malignity were apparent.

## Tumours of the haemopoietic tissue

Table 4  
Tumours of the haemopoietic tissue

No.	Age – years	Sex	Diagnosis
1	4	M	Generalized lymphocytic leukosis
2	4	M	Generalized lymphocytic leukosis
3	4	M	Generalized lymphocytic leukosis

In all three cases the neoplastic cells were of lymphocytic nature. Compared to normal lymphocytes they were rather large, the nuclei were round, oval or irregular with indentations or clefts and contained fine chromatin. Nucleoli were apparent only in small proportion of the cells. Cytoplasm was slightly basophilic in form of narrow rim around the nucleus. Cohesivity among the cells was low. In the first case there was high mitotic activity and in some cells there were atypic mitotic figures. In the remaining two cases the mitotic figures were almost absent.

Gross pathology and histopathology were similar in all three animals. Necropsy revealed general lymphadenopathy, including mesenteric lymph nodes, moderate splenomegaly and multiple little light foci in many organs including intestine. In case No. 2 there was also hydrothorax and hydropericard, in guinea pig No. 3 was anasarca, milk-turbid effusion in thoracic and abdominal cavities and haemorrhages in the lymph nodes.

Histopathological examination revealed diffuse infiltration of lymph nodes and their perinodal connective tissue with neoplastic lymphocytes. The original structure of the lymph nodes was entirely effaced or only some remnants of original structure persisted. In guinea pig No. 1, also in the spleen was diffuse infiltration with the neoplastic cells and multiple foci of necrosis were present. In animals No. 2 and 3, the original structure of the spleen was preserved but the sinuses and cords of the red pulp were infiltrated with neoplastic cells. In mucosa associated lymphatic tissue (MALT) of the intestine there was diffuse infiltration of lymphoma cells. In subepicardial connective tissue of the heart there were segments of infiltration with leukaemic cells. In these segments the epicardium was absent or damaged and reparative processes characterized by proliferation of blood capillaries and fibroblasts were observed. Myocardium of all three animals was free of neoplastic cells. In the lungs were sheaths of neoplastic cells around the blood vessels and mild diffuse infiltration of pulmonary interstitium. Perivascular sheaths of lymphoma cells were also in the portal fields of the liver. Besides this, mild infiltration of sinusoids or foci of neoplastic lymphocytes were present in the liver parenchyma.

Large or small aggregates of neoplastic cells were observed also in the kidneys, adrenals, and epididymis. The femoral bone marrow was examined histologically only in guinea pig No. 3 but infiltration with neoplastic lymphocytes was not observed.

In accordance to a short course of clinical disease the high grade leucosis could be considered. From the cytological point of view, the cells were similar to centroblasts and centrocytes. Immunohistochemistry was not performed because our laboratory did not possess appropriate antibodies.

### Tumour of the liver

This tumour was revealed accidentally in one female, five years of age, by necropsy done by one clinician. In the same animal also trichofolliculoma was found. The tumour submitted for histological examination was globoid, 1.5 cm in diameter. It consisted of hepatocytes arranged in lobules with central vein but portal triads and interlobular biliary ducts were not developed. Based on histological examination, the tumour was diagnosed as hepatocellular adenoma.

## Discussion

Blumenthal and Rogers (1965) have reported about 140 tumours in guinea pigs. Only in one animal they recorded a tumour of the skin that was diagnosed as *epithelioma adenoides cysticum*. Kitchen et al. (1975) examined 14 spontaneous tumours in guinea pigs and in three cases trichoepithelioma was diagnosed. Wagner and Manning (1976) reported about 29 trichofolliculoma and one trichoepithelioma. They did not observe any other types of skin tumours. The authors state that skin tumours are the most frequent of all reported tumours in guinea pigs. Of these, the trichofolliculomas are probably the best known. Zwart et al. (1981) reported about two trichofolliculomas and one sebaceous gland adenoma. In our collection there were two trichofolliculomas, two trichoepitheliomas and one malignant pilomatricoma. In the available literature no information on pilomatricoma or malignant pilomatricoma in guinea pigs was found. Immunohistochemistry revealed different grades of differentiation of the neoplastic cells in the malignant pilomatricoma. Especially in the second part of the tumour was well visible distinction between the cells inside and at periphery of neoplastic islands. Poorly differentiated cells in the third part did not express cytokeratins. In three of five our cases, the skin tumours were situated in dorsal region of the body. Similar predilection exists also in dogs (Gross et al. 1992).

In five cases of mammary gland tumours there were two adenocarcinomas and both appeared in males of middle age category. In one female of similar age category malignant tumour of myoepithelial histogenesis was diagnosed. In two young females were benign processes – one adenoma and one adenomatous hyperplasia of the mammary gland. Blumenthal and Rogers (1965) reported about 11 cases of neoplasia in mammary gland. Three were benign (adenoma and cystadenoma) and eight were adenocarcinomas. Three of them were in males. Kitchen et al. (1975) observed two adenocarcinomas, one in female and one in male – both of middle age, one adenoma in male, one malignant mixed tumour with pulmonary metastases in a female 7.5-year-old. In accordance with literature and my own experience, mammary neoplasias appear in males of different animal species, including humans, rarely. Some species of rodents are exception, and, e.g., in old rat males the frequency is relatively high. In 16% of males, over 24 months of age, Wistar strain, bred in the Czech Republic, the author diagnosed mammary tumours (unpublished data). Negative demonstration of cytokeratins in mammary adenocarcinoma, by means of antibody MNF 116 (DAKO) was surprising. In accordance to own experience this antibody is very well usable in mouse, rat, hamster, dog and cat. Demonstration of cytokeratin K18 in the guinea pigs by means of EXBIO antibody was successful.

From six tumours located in subcutaneous tissue three were lipomas, one of them was multiple. In case of liposarcoma the histopathological diagnosis was in good relation to growth rate even though mitotic activity was fairly low. Blumenthal and Rogers (1965) diagnosed two fibrolipomas, two neurilemmomas, seven fibrosarcomas, three fibroliposarcomas and one neurogenic sarcoma. Altogether they observed neoplasia in subcutaneous tissue in 15 cases from 140 animals. Kitchen et al. (1975) diagnosed, among 14 tumours in guinea pigs, two lipomas and one schwannoma in the subcutaneous tissue. Wagner and Manning (1976) observed eight cases of fibrolipoma, and by one fibroma, fibrosarcoma and lipoma. Percy and Barthold (1993) presented only a list of the subcutaneous tumours – lipoma, fibrosarcoma, fibroma and carcinomas without further characterization. Ossifying fibroma, a rare tumour, was diagnosed in accordance with description by Palmer (1993). In the available literature no information about this tumour and myxoid liposarcoma in guinea pigs was found. Squire et al. (1978) state that skin tumours and tumours in subcutaneous tissue are rare but it is not in accordance with the results of other authors. E.g. in the collection of Wagner and Manning (1976) neoplasias of the skin and subcutis range to 15.4%.

In three males of our collection generalized leukosis of lymphocytic nature was diagnosed. This neoplasia was clinically manifested by lymphadenopathy and shortly before death of the animal by alteration of general health state. In our cases the clinical course of the disease was short. In guinea pigs kept as pets it is impossible to determine the frequency of neoplasias, including the ones of haemopoietic tissue and lymphomas but in accordance with veterinary physicians these neoplasias are not rare. Majority of these cases are not exactly diagnosed because the owners have no interest in laboratory examination with regard to poor prognosis of the disease. From 140 neoplasias collected by Blumenthal and Rogers (1965) there were 10 cases of leukosis and 3 cases of malignant lymphoma. Kitchen et al. (1975) observed only one case of histiocytic lymphosarcoma among 14 neoplasias. Wagner and Manning (1976) diagnosed leukemia in 13, and lymphosarcoma in nine guinea pigs. Squire et al. (1978) state that lymphomas and lymphocytic leukaemias are not uncommon in middle-aged guinea pigs. In laboratory breedings the incidence of lymphomas and leukemias is related to the strain of guinea pigs. Hong et al. (1980) reviewed spontaneous lymphoblastic leukaemia in several laboratory strains. These authors have diagnosed only nine cases of lymphoblastic leukaemia in 4,500 examined guinea pigs. Seven cases occurred in strain 2/N, one in 13/N and one in Dunkin-Hartley/FD strain. Both gross pathology and histopathology were similar to our cases. Similarly to cited authors, we did not do further classification of leukosis by immunohistochemistry because we did not possess the appropriate antibodies.

No liver tumour is presented in the collections of Blumenthal and Rogers (1965) and Kitchen et al. (1975). Wagner and Manning (1976) have reported liver cell adenoma, cavernous haemangioma and gallbladder papilloma. General opinion is that in guinea pigs tumours of the liver are rare.

In our collection of neoplasias, no lung tumour was observed though, according to Squire et al. (1978), they are not rare in form of adenomas and adenocarcinomas. According to Percy and Barthold (1993) the pulmonary tumours form 35%, and tumours of reproductive organs 25% of all spontaneous tumours in guinea pigs. Also neoplasias of the cardiovascular system are, according to the literature, not uncommon. In other organs frequency of tumours is low. In spite of the differences in the literature concerned of tumours frequency and classification, there is general agreement that in guinea pigs older than three years neoplasias are not rare. According to Wagner and Manning (1976), the frequency of neoplasias is ranging from 14.4% to 30% in guinea pigs of this age category. The aim of this paper is to contribute to so far scarce literature concerning the tumours in guinea pigs.

## Spontánní tumory u morčat

Histologicky bylo vyšetřeno 20 nádorů od 19 morčat. V 15 případech byly tumory, zasláné k vyšetření, biopsie, ze čtyř morčat byly vzorky orgánů odebrány při pitvě. Kromě jednoho zvířete, pocházejícího z laboratorního chovu, byla morčata chována v domácnostech. U pěti morčat byly diagnostikovány tumory kůže. Byly lokalizovány na rozdílných místech těla – břicho, plantární plocha pánevní končetiny, zád a ve dvou případech na hřbetě. Nádory byly rozdílné velikosti, největší měl průměr 5 cm. Všechny tumory vycházely z chlupových folikulů – dva trichofolikuly, dva trichoepiteliomy a jeden maligní pilomatricom. Postižená zvířata byla ve věku 2-7 roků. Tumory mléčné žlázy se vyskytly u pěti morčat. U dvou samců byl diagnostikován adenokarcinom, a u samic byl po jednom případě sarkom myoepiteliální histogeneze, tubulární adenom a adenomatozní hyperplazie. Nádory v podkožním vazivu byly zjištěny u šesti morčat – tří samců a tří samic, věkové kategorie 3-5 roků. Ve třech případech to byly lipomy, z toho u jednoho morčete vícečetné, a po jednom případě byl diagnostikován liposarkom, myxoidní liposarkom a osifikující fibrom. Lymfocytární leukóza byla zjištěna u tří samců, u všech ve věku čtyři roky. Hepatocelulární adenom byl nalezen při pitvě u jedné samice u které byl současně trichofolikulom uvedený výše.

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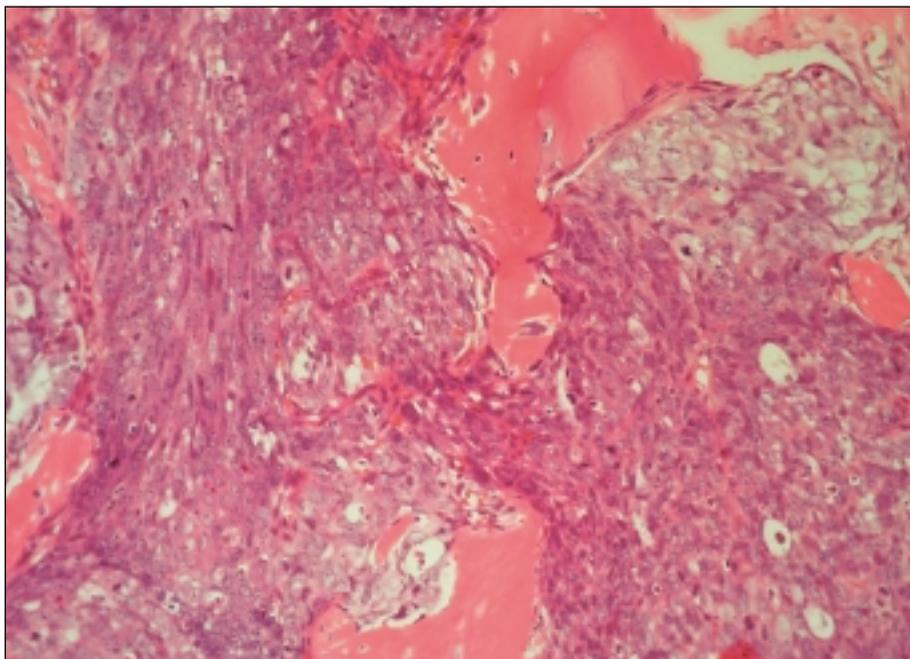


Fig. 1. Solid tumour consisting predominantly of spindle cells with some mitotic formations and metaplastic bone tissue. Staining with HE, original magnification  $\times 75$ .

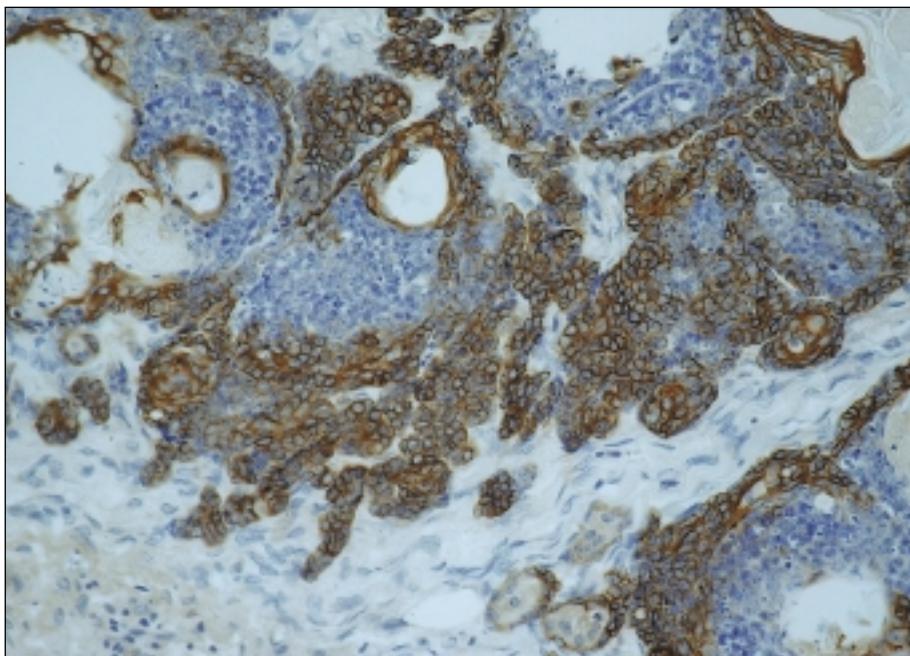


Fig. 2. Malignant pilomatricoma. Positivity for cytokeratins in cells at periphery of epithelial islands and their outgrowths. Immunoperoxidase reaction, original magnification  $\times 75$ .

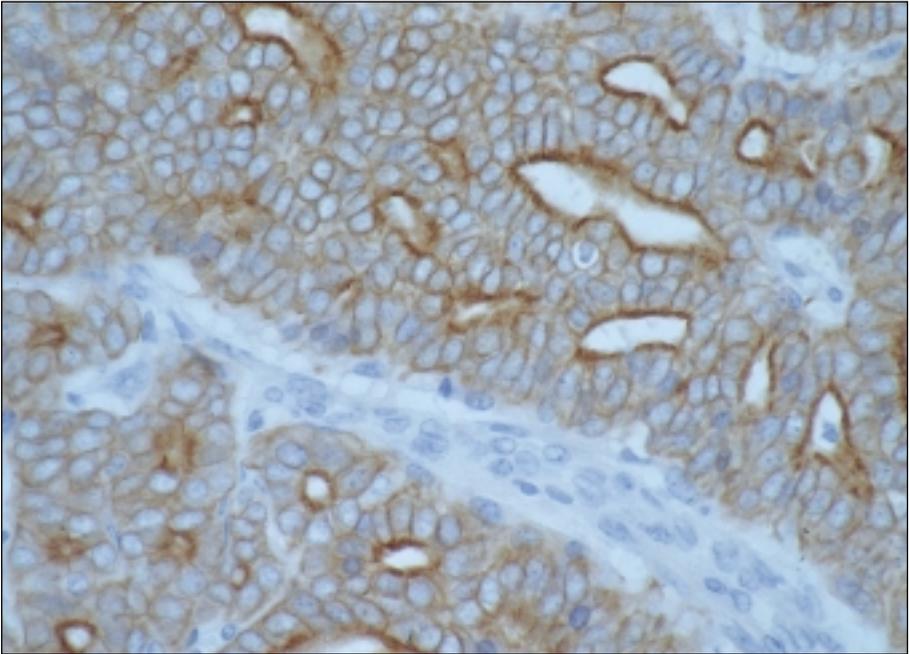


Fig. 3. Tubular adenocarcinoma of the mammary gland. Detection of cytokeratin K18. Immunoperoxidase reaction, original magnification  $\times 150$ .

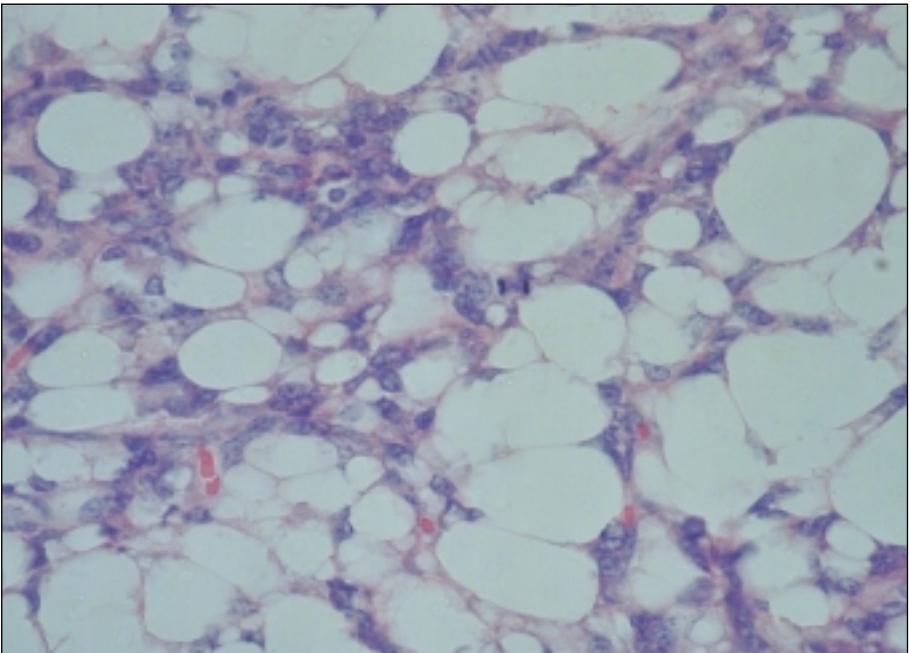


Fig. 4. Liposarcoma in the subcutaneous connective tissue. In the neoplastic tissue there are large vacuoles of fat, mild polymorphy of the tumourous cells and one mitotic figure. Staining with HE, original magnification  $\times 150$ .