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Original article

Epidemiological and morphological analysis of feline injection site sarcomas

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Abstract

Feline injection site sarcomas (FISS) are malignant neoplasms of mesenchymal origin which arise in sites of injections in cats. The prevalence is estimated between 1 in 1000 and 1 in 10 000 vaccinations in the United Kingdom. The aim of this study was to estimate the incidence of FISS in Poland and to analyse clinical aspects and histological and cytological features of injection site sarcomas. In our study the prevalence of FISS was 0.16% (16 FISS on 10.000 of cats) in feline patients in one of a veterinary surgery which conducts the general practice and 85 on 10.000 cats in a practice focused on veterinary oncology. The most typical microscopic features of FISS found in the present analysis were: the presence of perilesional scarring and inflammation, aggregates of lymphocytes at the tumour periphery, moderate but usually marked cellular pleomorphism and intralesional necrosis. The most typical cytologic features of FISS found in present study were: the presence of neutrophils, marked cellular pleomorphism, the presence of lymphocytes and macrophages, the presence of extremely large nuclei in the neoplastic cells, and high sample cellularity.

Key words: feline injection site sarcomas, FISS, FISS prevalence, cat, histopathology, cytology

Introduction

Sarcoma which arises in the site of injection in cats is called injection site sarcoma (ISS) or feline injection site sarcoma (FISS) and comprises a particular kind of soft-tissue sarcomas. This kind of neoplasm used to be called vaccine-associated sarcoma (VAS). This term is no longer appropriate because sarcoma can arise after injections performed for reasons other than vaccinations, for example to administer long-acting antibiotics and steroids, lufenuron, meloxicam, cisplatin (Esplin et al. 1999, Kass et al. 2003, Munday et al. 2011, Martano et al. 2012) or even after microchip implantation (Carminato et al. 2001, Daly et al. 2008). FISS comprises about 13% of all examined histopathologically and cytopathologically skin tumors in cats (Wilcock et al. 2012). The age of affected cats ranges from 8 to 11 years without breed or sex predilection (Couto et al. 2002, Wilcock et al. 2012). In the North America the incidence is estimated at 1 to 10 per 10000 vaccinations yielding 300-500 ISS every year in Canada and roughly 2000 per year in the USA and

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it has not changed since 1992. In the United Kingdom the incidence was 1-2 per cat in 10000 vaccinations. These estimations have been based only on tumors submitted for histological confirmation so the actual incidence is certainly higher (McEntee and Page 2001, Kirpensteijn 2006).

The most detailed data about changes in tumor location come from the USA. There had been a shift in FISS location from 1996 when Vaccine-Associated Feline Sarcoma Task Force recommended vaccination sites for FeLV and rabies vaccines, adopted later by the American Association of Feline Practitioners in 1997 (Shaw et al. 2009). Before 1996 feline ISS had been found 5 times more frequently in the interscapular region than in any other location. After 1996, there was a significant shift in tumor location to the hind legs and the lateral abdomen (Shaw et al. 2009). Proportion of FISS decreased in the interscapular region from 53.4% to 39.5% and increased on the right hind leg (recommended place for rabies vaccination) from 1.1% to 9.5% and on the left hind leg (recommended place for leukemia vaccination) with the left lateral abdomen region from 11.4 to 13.8% (Shaw et al. 2009). In the United Kingdom the most commonly affected site is still the interscapular region (84%), followed by femoral (6%), flank (5%), lumbar (3%) and gluteal (2%) regions (Kass et al. 2003).

Microscopic features of FISS are so characteristic that they allow the pathologist to identify them only with the microscopic examination. Unfortunately, histopathological gradation system proposed by Kuntz et al. (1997) or Powers et al. (1995) used for microscopical examination of „classical" soft tissue sarcomas is unsuitable for evaluation of ISS in cats. Therefore, on the basis of high number of FISS, 10 histopathological features typical for tumors which developed in sites of injection have been established. The features include for example: the presence of aggregates of lymphocytes, infiltrative margins, intralesional necrosis, perilesional scarring/inflammation, adjuvant-like material in macrophages, medium-high mitotic rate, neoplastic giant cells and types of cellular differentiation among neoplastic cells. It is assumed that the presence of 7 from 10 these features allows to classify sarcoma as ISS (Dean et al. 2013). FISS emerge a few months to 5 years after an injection so the exact type, number and site of performed injections are very hard to establish in most cases. The aim of this study was to analyze (1) the prevalence of FISS in cats, (2) clinical aspects of animals with these neoplasms and (3) histological and cytological features of collected specimens.

Materials and Methods

Cases were identified retrospectively by searching the medical records of the Division of Animal Pathomorphology, Faculty of Veterinary Medicine, Warsaw University of Life Sciences (Poland) for the period 1998-2013, Veterinary Surgery Białobrzeska in Warsaw and Veterinary Surgery in Piaseczno (Poland) for the period 2008-2014.

Including criteria. Cases were classified as FISS if they were located in the areas where injections are routinely made (caudal neck, scapular region, lumbal and sacral region, flanks, sides of the chest, hind legs) and results of microscopic examination of cellular (cytology) and/or tissue (histopathology) samples were available and recognized as sarcoma.

Epidemiological analysis. To evaluate the prevalence of FISS in cats population four analyses were made: (a) percentage of cats with FISS recognized among population of all cats presented to the Veterinary Surgery Białobrzeska (example of veterinary practice where oncologic patients are significant group of all patients), (b) the percentage of cats with FISS recognized among population of all cats presented to the Veterinary Surgery in Piaseczno (example of veterinary practice conducting the general practice), (c) the percentage of FISS recognized histologically and (d) cytologically among all cats that were tested by microscopic examination in the laboratory of the Division of Animal Pathomorphology, Faculty of Veterinary Medicine, Warsaw University of Life Sciences.

Clinical features. Location of the mass/masses was described as: caudal neck, scapular region, lumbal and sacral region, flanks, sides of the chest, and hind legs. Macroscopic description of FISS included clinical features of malignancy: firm consistency, fixation to surrounding tissues, the presence of cystic spaces within mass (detected during clinical examination and confirmed by ultrasonography or puncture), and the presence of superficial ulceration. Finally, size of the mass was evaluated as small (the longest diameter shorter than 2 cm), medium-sized (the longest diameter between 2-5 cm) and large (the longest diameter longer than 5 cm).

Microscopic analysis. Tissue samples of 35 tumour masses collected during surgery or necropsy were fixed in 10% neutral buffered formalin, embedded in paraffin wax, cut in sections (3 fm) and stained with haematoxylin and eosin. Based on generally accepted criteria tumours were classified as sarcoma and then based on additional histochemical stains (Van Gieson method, Trichrome stain, PAS method, Sudan method) into specific types of sarcomas (fibrosarcoma, liposarcoma, leiomyosarcoma, myxosarcoma,

undifferentiated sarcoma). Cytological samples were collected during fine-needle aspiration biopsy from 26 histopathologically confirmed injection site sarcomas and smeared in routine manner. Smears of good quality were dried, fixed in 70% methanol, stained with Giemsa solution, and examined by light microscope. To establish histopathological features of FISS, 26 randomly selected cases were compared to 9 cases of sarcomas from location other than the injection site (non-FISS; oral cavity, external ear canal, cheek, larynx, footpad, perivulvar region) in cats with no history of previous neoplastic tumours. We evaluated the presence of following microscopic features: infiltrative margins (yes/no), perilesional scarring and/or inflammation (yes/no), aggregates of lymphocytes (yes/no), multinucleated giant cells (yes/no), pleomorphism (low/ medium/ high), the presence of adjuvant-like material in cells (yes/no), intralesional necrosis (no/ medium/ high) and mitotic rate (average number of mitotic figures in 10/high power field -HPF). To establish cytological features of FISS, fine-needle aspiration biopsy samples collected from 26 histopathologically confirmed cases of injection site sarcomas were analyzed under light microscope. Cytological features analyzed included: general cellularity (low or high); number of mitotic figures in entire slide (no/ low/ high); and the presence of features of necrosis (including the presence of proteinaceous fluid, cellular debris, naked and ruptured nuclei; 0, +, ++). Furthermore, the presence of: cellular clusters (yes/no), numerous erythrocytes (yes/no), mononuclear inflammatory cells (yes/no), neutrophils (yes/no), macrokaryosis (yes/no), neoplastic multinucleated giant cells and marked cellular pleomorphism of neoplastic cells (yes/no) was recorded.

Statistical analysis. 95% confidence intervals (CI) for percentages were calculated using Wilson score method (Altman et al. 2000). Data presented in nominal scale were compared between groups with a chi-square or Fisher's exact test. A Mann-Whitney U test was used to compare data in ordinal scale. A two-tailed p-value below 0.05 was considered to indicate statistical significance.

Results

Epidemiological data. Based on the mass location and results of histopathology and/or cytopathology 135 FISS were diagnosed between 2008 and 2014 in the Veterinary Surgery Białobrzeska; cats with these tumours accounted for 1.45% (95% CI: 1.23%, 1.71%) of all feline patients in this period (135 of 9318 cats). Based on the mass location and results of histopathology and/or cytopathology 8 FISS were diagnosed between 2008 and 2014 in the Veterinary Surgery in Piaseczno; cats with these tumours accounted for 0.16% (95% CI: 0.08%, 0.32%) of all feline patients in this period (8 of 4894 cats). Based on the mass location and results of histopathology 140 FISS were diagnosed between 1998 and 2013 in the laboratory of the Animal Pathomorphology Division. These tumors accounted for 13.13% (95% CI: 11.24%, 15.29%) of all microscopically examined tissue samples collected from cats (140 of 1066 feline samples) and 45.08% (95% CI: 37.81%, 48.51%) of all skin and subcutaneous tissue samples in this species (140 of 325 feline samples of dermal and subcutaneous lesions). Based on the mass location and results of cytopatological examinations 29 FISS were diagnosed between 2009 and 2013 in the laboratory of the Animal Pathomorphology Division. These tumors accounted for 7.59% (95% CI: 5.34%, 10.69%) of all cytopathological examinations performed in this period in cats (29 of 382 feline samples), 23.39% (95% CI: 16.81%, 31.57%) of samples collected from dermal and subcutaneous lesions of any type (non-neoplastic and neoplastic) (29 of 124 feline samples of dermal and subcutaneous lesions), moreover, 45.31% (95% CI: 33.73%, 57.43%) of cases of dermal and subcutaneous neoplasms diagnosed in cats in this period (29 of 64 feline samples collected from dermal and subcutaneous neoplasms).

Clinical features. Particular data on location of FISS in cats were available for 149 cats and are presented in Table 1. Clinical examination (supported by ultrasonography and mass puncture) revealed firm consistency in 89.2% of cases, fixation to surrounding tissues in 82.2% of cases, the presence of cystic spaces containing sero-mucinoid fluid material within mass in 28.6% of cases, and the presence of ulceration was not observed in any case. Twenty five per cent of masses were small, 37.5% medium and 37.5% large.

Histopathological analysis. Among all 140 FISS examined by histopathology 72.1% were recognized as fibrosarcomas, 17.1% as undifferentiated sarcomas or spindle-cell sarcomas, 7.1% as liposarcomas, and 3.7% as other types of sarcomas (leiomyosarcoma and myxosarcoma). Detailed histopathological analysis of the randomly selected FISS cases is presented in Table 2. Selected pictures of histopathological features are presented in Fig. 1.

Cytological analysis. Detailed data on cytological features examined during microscopic analysis of 26 histologically confirmed FISS are presented in Table 3. The most typical cytological features of FISS were: features of intralesional necrosis, the presence of neutrophils, marked cellular pleomorphism of neoplastic cells (every features present in all 26 cases), the presence of mononuclear inflammatory cells and macrokaryosis (both present in 25 of 26 cases) (Fig. 2).

Table 1. Location of FISS in particular groups of cats included into study.

	Percent of cases in particular localization					
	Scapular	Lumbal/sacral	Flanks	Neck	Chest wall	Hindlegs
FISS examined in Division of Animal Pathomorphology n=103	45	13	12	13	9	8
FISS examined in Veterinary Surgery Białobrzeska n=46	67	9	2	4	11	7
All FISS included into study n=149	52	12	10	10	9	7

Table 2. Results of histopathological analysis of FISS and non-FISS; microscopic features with statistically significant differences between groups of tumors are labeled with*

	Infiltrative margins	Perilesional scarring or inflammation	Aggregates of lymphocytes	Giant cells	Cellular pleomorphism			Adjuvantlike material	Intralesional necrosis			Mitotic rate
					low	medium	high		no	medium	high	
FISS n=26	92% (24)	96% (25)	88% (23)	50% (13)	0%	31% (8)	69% (18)	42% (11)	0%	54% (14)	46% (12)	2.96
non-FISS n=9	100% (8)	56% (5)	22% (2)	11% (1)	11% (1)	78% (7)	11% (1)	22% (2)	22% (2)	67% (6)	11% (1)	2.16
p-value	1.00	0.011*	<0.001*	0.056		0.007*		0.431		0.038*		0.427

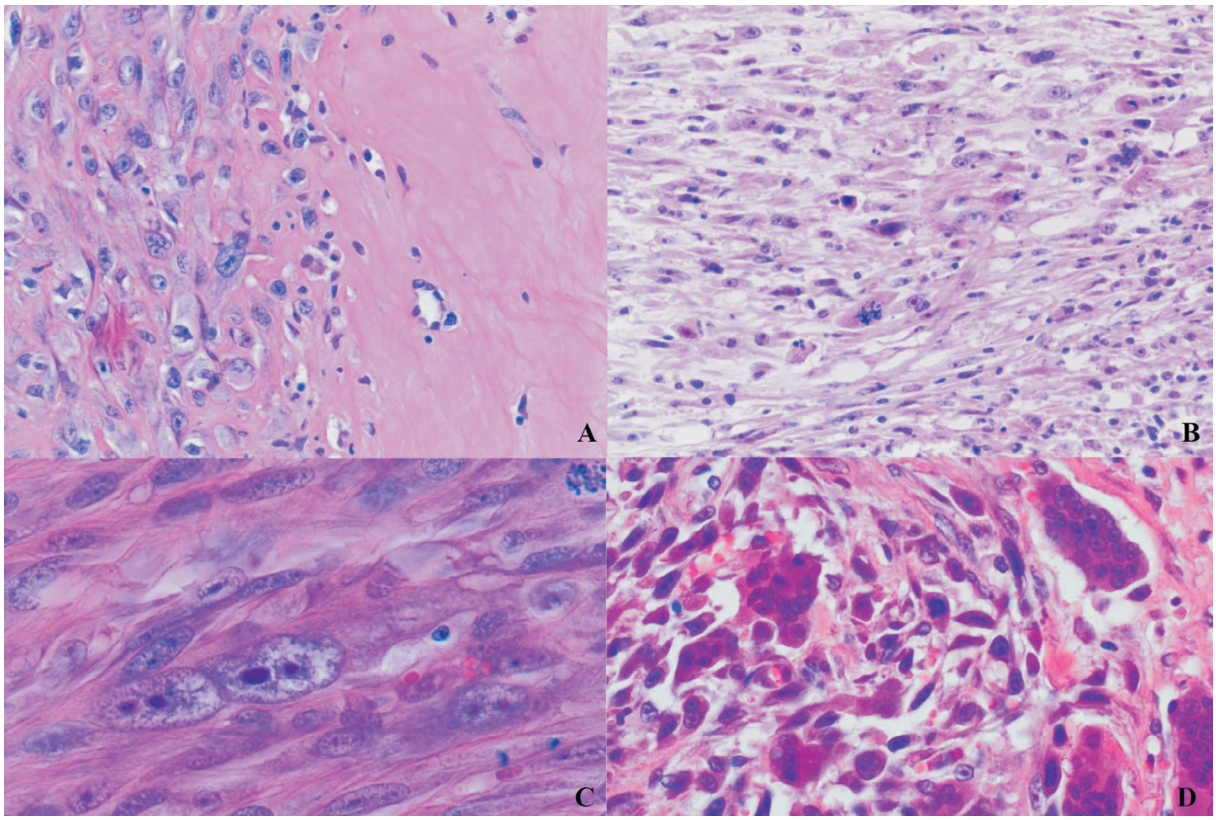

Fig. 1. Histopathological features of FISS. H-E stain. A – Large field of necrosis on the right side of the slide. Magnification 200x. B – High cellular pleomorphism of neoplastic cells. x200. C – Macrokaryosis in neoplastic cells. x400. D – Giant cells are often present in FISS. x200.

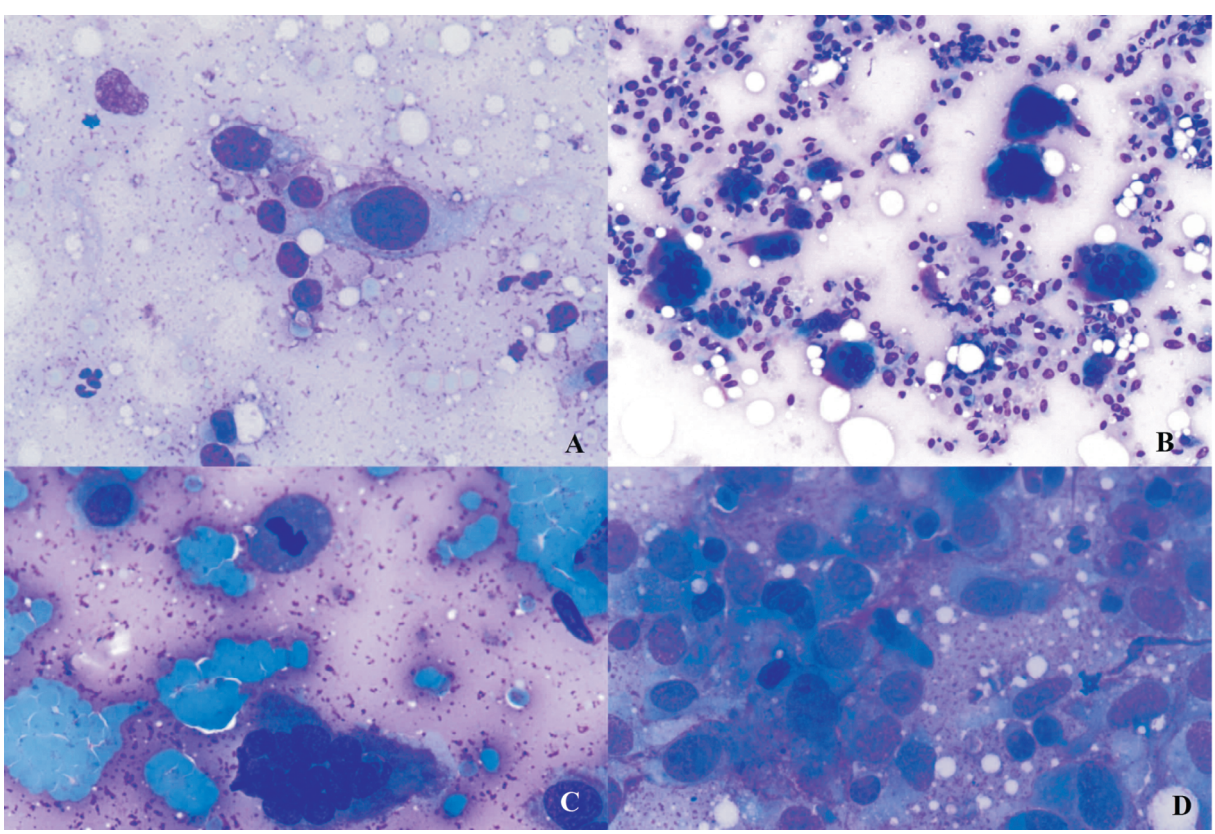

Fig. 2. Cytopathological features of FISS. Giemsa stain. A – Macrokaryosis. x400. B – Giant cells. x200. C – Mitotic figure (on the top) and giant cell (on the bottom of the picture). x400. D – Large amount of neoplastic cells with high cellular pleomorphism. x400.

Table 3. Results of cytological examination of histopathologically confirmed FISS; N- not present, L – low, H – high.

No.	Cellularity of sample	Cellular clusters	Numerous erythrocytes	Features of necrosis	Mononuclear cells	Neutrophils	Macrokaryosis	Giant cells	Marked cellular pleomorphism	Mitoses
1	H	+	+	++	+	+	+	+	+	H
2	H	+	+	+	+	+	+	–	+	L
3	L	+	–	++	+	+	+	+	+	N
4	L	–	–	++	+	+	+	–	+	N
5	H	+	+	++	+	+	+	–	+	N
6	H	+	–	++	+	+	+	+	+	L
7	H	+	+	++	+	+	+	+	+	L
8	L	+	+	++	+	+	+	–	+	N
9	H	+	–	++	+	+	+	–	+	N
10	L	+	+	++	+	+	+	–	+	L
11	H	+	–	++	+	+	+	+	+	L
12	H	+	+	+	+	+	+	+	+	L
13	H	+	+	+	+	+	+	+	+	H
14	H	+	+	++	+	+	+	–	+	L
15	H	+	–	++	+	+	+	+	+	L
16	H	+	–	++	+	+	–	–	+	N
17	H	+	+	++	+	+	+	–	+	N
18	L	–	+	+	+	+	+	+	+	N
19	H	+	–	++	+	+	+	–	+	H
20	H	+	+	+	–	+	+	–	+	L
21	H	+	+	++	+	+	+	+	+	N
22	H	–	+	+	+	+	+	+	+	N
23	H	+	+	+	+	+	+	–	+	H
24	L	–	–	++	+	+	+	–	+	N
25	H	+	+	++	+	+	+	+	+	H
26	H	+	+	++	+	+	+	–	+	N

Discussion

The problem of malignant mesenchymal tumours growing in sites of previous vaccinations in cats has been known since 1991 (Hendrick and Goldschmidt 1991). In the past, criteria required for VAS recognition were very strict and included among others specific location of the mass, the presence of vaccine adjuvant-like material within intratumoral macrophages and finally information about administration of certain vaccines and the site of vaccination (Hendrick et al. 1992, Hendrick et al. 1994). When recognized this way, VAS were considered to be very rare problem – from 1 to 13 per 10000 cats. In one report involving 31671 cats the number of vaccine site-associated sarcomas was even lower – 0.63 on 10000 animals (Gobar and Kass 2002). Unfortunately, in most cases neither site of vaccination nor type of administered vaccine can be determined. Moreover, there have been many reports of sarcomas arisen in sites of injections other than vaccinations thus the term „feline injection site sarcoma" should be used in such cases (Esplin et al. 1999, Carminato et al. 2001, Kass et al. 2003, Daly et al. 2008, Munday et al. 2011, Martano et al. 2012). Hence two criteria need to be addressed to recognize FISS. First, histopathological character of a tumor has to be typical for malignant mesenchymal tumor, and secondly, a tumour should grow in a site where injections are routinely performed. On these criteria the prevalence of FISS in cats population turned out higher than before. We based our analysis on a population of feline patients of two veterinary clinics. In our study the prevalence of FISS in feline patients of general practice veterinary surgery was 0.16% (16 FISS out of 10000 cats). On the other hand, the prevalence estimated for the oncological veterinary surgery was five-fold higher. These appear to be high values, especially when compared to the prevalence of FISS in the United Kingdom, where only 1 FISS was recognized per 50 000 cats registered and per 5 000 vaccination visits (Dean et al. 2013). These differences are consequences of a few factors, firstly, we examined the incidence only in two selected veterinary surgeries, and secondly, the analysis embraces period from 2008 to 2013. In study of Dean et al. (2013) only cases recognized in 2007 were examined, additionally numerous veterinary practices were enrolled in their research. However, our analysis although tentative allows to approximately estimate the significance of the problem in Poland, and it has shown that FISS prevalence is considerable in general cat population and can be considered high in feline oncologic patients. Additionally, FISS are serious problem in oncologic surgery. We have revealed that more than 13% of all tissue samples resected from cats and 45% of samples of feline cutaneous and subcutaneous tumors examined histopathologically in our laboratory were recognized as FISS. Study conducted by Wilcock et al. (2012) revealed that prevalence of FISS among cats with skin masses resected and submitted to histopathological analysis was from 6.8 to 21.1%. These values are lower than those obtained from our histopathological material but similar in regard to the cytological samples (23.3% of cytological samples collected from dermal or subdermal lesions were recognized as FISS). However, further analysis of cytopathological samples revealed that 45% of all skin and subcutaneous lesions examined were recognized as FISS. In the light of these results FISS must be considered as a very serious clinical problem because these tumors need very radical medical treatment.

Because of serious problems encountered during surgical intervention in cases of FISS, other sites of injection, especially neck, scapular/interscapular, sacral and lumbar regions are not recommended and should be avoided in cats. However, in our analysis FISS in these body regions were common (74% of cats included in this study). The results are worse than those obtained in the USA, were the proportion of FISS decreased from 1990 to 2006: 53.4% to 39.5%, respectively, in the interscapular region (Shaw et al. 2009). In our research, among 149 cats with precisely known location of a mass, in 52% of FISS tumor were growing in the interscapular or periscapular region. Fortunately, the situation in Poland is better than, for example, in the UK where the most affected site is still the interscapular region and the percentage of FISS arisen in this area of the body reaches 84% (Kass et al. 2003). Although the size of the tumor did not influence the outcome of surgical treatment in one study it seems that smaller sarcomas much more often allow for complete resection (Romanelli et al. 2008). The present study has revealed that medical diagnosis of FISS is commonly obtained in advanced stage, because in more than 35% of cases a diameter of the tumor was longer than 5 cm, and only 25% of tumors were considered to be small (diameter shorter than 2 cm). As it was shown in our research, histopathological analysis of randomly selected 26 archival cases revealed that free margins were present only in 2 sarcomas. Free margins were shown to be related to more favorable prognosis in cases of FISS (Giudice et al. 2010). The results obtained suggest that widespread and precise owners; education by veterinarians is necessary, because delaying diagnosis and surgery makes complete resection hard to perform or even impossible due to very fast growth of FISS. Beside rapid growth and large mass, in the majority of cases (more than 80% of FISS) direct examination of a mass revealed clinical features of malig-

nancy. FISS are usually firm and fixed to the underlying structures what is a typical characteristic of an infiltrative growth.

Histopathological features of sarcomas related to previous injections in cats are well known. However, we decided to compare histopathological picture of randomly selected cases of FISS with tumors growing in location that allow to excluded their connection with injections. In eariler studies microscopic features of vaccine-associated sarcomas rather than injection site sarcomas were usually described (Couto et al. 2002, Gobar and Cass 2002, Romanelli et al. 2008). Previous investigations have shown that the most common histological type of FISS is fibrosarcoma; it accounts for about 80-90% of these tumors (Couto et al. 2002, Romanelli et al. 2008, Giudice et al. 2010, Wilcock et al. 2012). Our findings were similar – fibrosarcoma was the most common histopathological type (72% of FISS), followed by undifferentiated spindle-cell sarcomas, liposarcomas and other types which were recognized incidentally. Although it is important from the morphologic standpoint, histopathological type of FISS is not related to biological behavior or treatment outcome (Romanelli et al. 2008, Giudice et al. 2010).

Regardless of histological type of recognized sarcomas the most typical microscopic features of FISS in the present analysis were: the presence of perilesional scarring and inflammation, aggregates of lymphocytes at tumor periphery, moderate but usually marked cellular pleomorphism and intralesional necrosis. These features were significantly over-represented in FISS, for example cellular pleomorphism in the majority of cases of FISS was marked in and in sarcomas not connected with injection sites was usually moderate. It is not surprising as these microscopic characteristics are typical for malignant mesenchymal tumors considered to be related to previous injections in cats (Giudice et al. 2010). As it was shown in other studies, histological grade based on tumor cells' differentiation, mitotic index and extension of necrosis was not correlated to recurrence rate after surgical excision of FISS (Romanelli et al. 2008, Giudice et al. 2010). However, poor histologic differentiation of tumour as a single parameter was related to local recurrences in one study (Giudice et al. 2010) and to the presence of distant metastases in the another (Romanelli et al. 2008). Unfortunately, currently there are no widely accepted histological criteria for histological grading of FISS, in the past grading system for canine soft tissue sarcomas was used but its usefulness in feline patients is still debatable (Kuntz et al. 1997, Couto et al. 2002).

The other microscopic features, for example the presence of adjuvant-like material in intratumoral macrophages was observed in less than 50% of FISS cases. Some authors consider the presence of this material as typical feature of FISS (Giudie et al. 2010, Dean et al. 2013). Theoretically, if a tumor had been caused by injection, remnants of vaccine or other drugs should be present in macrophages. However, during microscopic observation such material may also be observed in some non-FISS. Lack of significant differences between FISS and non-FISS in terms of adjuvant-like material present in macrophages is in our opinion negligible. It seems that definition „adjuvant-like material" is not precise, and without additional diagnostic methods pathologists are unable to differentiate contents of macrophages with certainty. Actually, macrophages filled with „adjuvant-like material" may as well be siderophages or macrophages that have phagocytized remnants of cellular debris abundant in necrotic areas of tumor. The presence of neoplastic giant cells is considered to be characteristic in many FISS, although in the present study we observed these cells only in a half of tumors examined. In our study the difference between FISS and non-FISS in this parameter was statistically insignificant. Interestingly, proliferation rate of FISS examined by mitotic index was the same as in cases of non-FISS, although other authors have revealed that sarcomas linked to injections usually have high mitotic indexes (Hendrick and Brooks 1994, Hendrick et al. 1994, Wilcock 2012).

Surprisingly, despite the fact that injection site sarcomas are widespread in cats, descriptions of their cytological picture are sparse. According to the available data cytology is diagnostic only in about 50% of FISS (Martano et al. 2011). The most typical cytologic features of FISS in the present study were the presence of neutrophils and marked cellular pleomorphism observed in all cases examined. Additionally, the presence of lymphocytes and macrophages, the presence of extremely large nuclei, and high sample cellularity were detected in vast majority of cases. Mitotic figures and multinucleated neoplastic giant cells were found in approximately 50% of cases. The presence of neutrophils results from necrotic changes in the tumour parenchyma, which are common in these neoplasms, as it was shown in our and other studies (Giudice et al. 2010, Martano et al. 2011, Dean et al. 2013). Lymphocytic infiltration is also a common finding in histopathological slides, especially on the border of a tumour, cytological examination revealed that mononuclear cells were usually encountered in cytological smears. In some cases solid tumour consistency or the presence of necrosis could be responsible for a very low cellularity of the sample. However, even in such cases highly pleomorphic cells, macrokaryosis and sometimes single cell clusters are

observed during cytopathological examination. Besides the presence of necrosis, tumors often contain caverns filled with fluid, so the clinicians must remember that it is very important to collect several samples from various sites of FISS or to perform ultrasonography-assisted biopsy. Information about typical cytological features of FISS is important because in our opinion in most cases clinical data (tumor location) supported by cytological examination of mass are sufficient to obtain reliable diagnosis.

Conclusions

Feline injection-site sarcomas (FISS) has become a very serious problem in daily veterinary practice in many countries, including Poland. Veterinary oncologists are particularly familiar with this problem because FISS account for over 40% of skin and subcutis neoplasms in cats. Clinicians and cat owners have to be particularly alert to tumors arisen in sites of previous injections because in case of FISS fast and radical treatment is necessary for favourable prognosis. Since it is not always possible to avoid FISS development, clinicians should respect international recommendations for preferable injections sites in cats which are hind legs and flanks. Differences in histopathological features as compared to previous reports have also been found. These differences included adjuvant-like material in intratumoral macrophages observed in less than 50% of FISS cases as well as giant cells present in only half of tumors examined. Both of these features were previously reported as characteristic for FISS. Proliferation rate examined by mitotic index was also the same in FISS and non-FISS, contrary to the results obtained by some authors who have revealed that FISS usually have very high mitotic indexes. Conclusion is that cythopathology can be very useful although often underestimated diagnostic method of FISS. Cytological picture supported by clinical data provide medically relevant information which enables clinicians to take more radical steps in the treatment of the affected cat.

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