Feline Injection Site Sarcomas: An Evidence-Based Approach

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Injection Site Sarcomas - History

1970’s: MLV without adjuvants in use
1985: New Al-adjuvanted FeLV vaccine released
1985: MLV rabies vaccine replaced by killed adjuvanted rabies vaccine because of clinical signs of rabies in 1:500,000 cats
1987: Rabies vaccination mandatory for cats
1996: Vaccine-Associated Feline Sarcoma Task Force
2002: Gobar and Kass: Incidence of vaccine site-associated sarcomas is low and decreasing
2003: Kass et al.: Vaccine brand, type, practices not associated with increased or decreased risk; long acting injections implicated
2003: Injection site sarcomas can also occur in ferrets and dogs

Unique susceptibility to injection site sarcomas in the cat?
Cats have increased susceptibility to oxidative stress for a number of reasons, including:
(1) Cats have 8 weak sulfhydryl groups on their hemoglobin compared to only 2 for other species;
(2) They have a non-sinusoidal spleen, which reduces their ability to remove Heinz bodies (denatured precipitated hemoglobin on the erythrocyte surface);
(3) They have increased lipid peroxidation of the erythrocyte membrane;
(4) Feline hemoglobin easily converts from tetramers to dimers; and
(5) Cats have a limited concentration of enzymes that catalyze glucuronide conjugation, thereby causing increased exposure of feline erythrocytes to metabolites and increased risk of oxidative injury.

Injection Site Sarcomas – Incidence Studies
The true incidence of feline injection site sarcomas is unknown, but published estimates are based on data from retrospective epidemiologic studies, surveys of biopsy specimens submitted to diagnostic labs, historical records and veterinarian/cat owner recollections linking tumor sites with injection sites. The denominator in the rate equation i.e. (# of sarcomas ÷ # cats at risk) per unit time, is based on current estimates of the US cat population and the average number of annual visits to veterinarians. Additionally, since injection site sarcomas usually develop from 3 months to over 3 years after an injection is administered, data collection is confounded by memory bias and unreported/unknown factors that might have been relevant to tumor development over the period. Because of the uncertainty with which much of this data is collected, current estimates vary from 1-10 cases/10,000 medicalized cats.
One large Canadian epidemiologic study reporting prevalence data showed that despite the widespread use of strategies designed to reduce the risk of injection site sarcomas, such as recommendations to administer vaccines in the lower limbs (1996) and the introduction of recombinant feline vaccines (2000), the prevalence of injection site sarcomas has not reduced over the period 1992-2010 (Wilcock, et al., 2012 Canadian Veterinary Journal 53:430-434). Similar annual statistics from the UK (UK Suspected Adverse Reaction Surveillance Scheme) demonstrate that injection site sarcomas are reported in association with inactivated (adjuvanted) vaccines, recombinant vaccines, and non-vaccine injections every year. However, it is important to remember that there are a variety of possible factors that could impact on the incidence of injection site sarcomas, and these factors can change over time.
due to differences in protocols and population dynamics. It is impossible for any one study to examine all of the relevant factors simultaneously, leading to inherent sources of bias. The most recent published incidence data comes from a 2013 UK study that estimated that the incidence risk of feline injection site sarcomas per year was estimated was -

- 1/16,000 -50,000 cats registered by practices,
- 1/10,000-20,000 cat consultations;
- 1/5,000-12,500 vaccination visits.

Searching for a cause and effect relationship

It is important to remember that retrospective studies, such as those usually used to investigate rare diseases such as injection site sarcomas, can only produce evidence for associations between the disease and the risk factors under study. In these studies, models are constructed using data collected from a sample population and the data is tested using statistics. The statistical significance of the findings is based on the assumption that the model is correct, but there are numerous potential sources of bias, such as selection bias, memory bias, and loss to follow-up over the extended study periods required to collect data. By contrast, prospective studies are required to produce cause-and-effect data. A sample size calculation performed by the University of Nottingham Centre for Evidence-Based Veterinary Medicine, based on 1 case of injection site sarcoma/10,000 vaccination events calculated that for a cohort (prospective) study to be performed, over 4 million cats would need to be enrolled and study for at least 10 years to produce meaningful data about the cause/s of this disease (Assoc. Prof. R. Dean, University of Nottingham, Personal communication, 2016).

Which injections?

Vaccines

Many kinds of injections have been associated with injection-site sarcomas. Although vaccines remain the predominant injection type associated with injection-site sarcomas, even recombinant vaccines have been associated with injection-site sarcoma development. There is no published evidence to date to show that recombinant vaccines carry less risk of injection-site sarcoma development than modified live vaccines. One paper (Srivastav, 2012) demonstrated an association only between adjuvanted (killed) vaccines and injection-site sarcoma development, in the rear limb region only and for rabies and FeLV vaccines only (not FVRCP vaccines). In this study, cats with injection-site sarcomas were less likely to have received a recombinant vaccine than a killed vaccine. The use of modified live vaccines of any kind (rabies, FeLV, or FVRCP) and at either site (interscapular or rear limb) did not carry any increased risk of injection-site sarcoma development when compared to recombinant vaccines in this study. Interestingly, for the interscapular region only, the use of depot glucocorticoids injections was associated with higher risk of injection-site sarcoma development in this study. Regarding adjuvants, both adjuvanted and non-adjuvanted vaccines have been reported in association with sarcomas. With respect to adjuvants, three large epidemiologic studies failed to find evidence of increased risk in aluminum containing vaccines compared to those without aluminum. One study reported that adjuvanted (killed) FeLV and rabies vaccines carried increased risk for injection-site sarcoma development, but only in the rear limb region. There is no published evidence to date to show that recombinant vaccines are any safer than modified live vaccines. Multiple injections with vaccines at a single site have been associated with increased risk compared to no vaccine administration as follows: 1 injection increases risk by 50%; 2 injections at the same site increase risk by 127%; and 3 injections at the same site increase risk by 175%. The administration of cold vaccines was associated with mild increased risk in another study. One large epidemiologic study was unable to demonstrate any increased risk in association with specific vaccine brands or manufacturers, vaccine administration factors (e.g. syringe reuse), history of trauma at the tumor site, or concurrent viral infection.
Non-Vaccines
Other injection types with reported associations with injection site sarcomas include long-acting antibiotics or steroids, lufenuron and meloxicam. Additionally, non-absorbable suture material, microchip implants (cats and 1 dog), subcutaneous fluid port placed long-term, retained surgical sponge material after spay (1 cat, 1 dog) and surgical implants in laboratory animals, including polymers, cellophane, glass and plastics have also been implicated.

Information from the UK Suspected Adverse Reaction Surveillance Scheme demonstrates that feline injection site sarcomas are reported in association with inactivated (adjuvanted) vaccines, recombinant (vectored) vaccines and a variety of non-vaccine injections every year (Table 1).

Table 1: Suspected Adverse Reactions, UK
(Dyer et al., Veterinary Record (2006-2012) and Davis et al., Veterinary Record (2013). UK Suspected Adverse Reaction Surveillance Scheme (SARSS))

<table>
<thead>
<tr>
<th>Year</th>
<th>Total (n)</th>
<th>Vaccine related * (n)</th>
<th>Feline Injection Site Sarcomas (n)</th>
<th>Inactivated/ Recombinant vaccine/ Non-vaccine (n)</th>
<th>Inactivated/ Recombinant vaccine/ Non-vaccine (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2005</td>
<td>694</td>
<td>155</td>
<td>34</td>
<td>9/ 1</td>
<td>26/ 3</td>
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<tr>
<td>2006</td>
<td>802</td>
<td>164</td>
<td>39</td>
<td>6/ 1</td>
<td>15/ 3</td>
</tr>
<tr>
<td>2007</td>
<td>857</td>
<td>191</td>
<td>59</td>
<td>10/ 4</td>
<td>17/ 7</td>
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<tr>
<td>2008</td>
<td>926</td>
<td>230</td>
<td>42</td>
<td>8/ 5/ 4</td>
<td>19/ 12/ 10</td>
</tr>
<tr>
<td>2009</td>
<td>850</td>
<td>179</td>
<td>40</td>
<td>10/ 4/ 4</td>
<td>25/ 10/ 10</td>
</tr>
<tr>
<td>2010</td>
<td>882</td>
<td>223</td>
<td>53</td>
<td>11/ 9/ 1</td>
<td>21/ 17/ 2</td>
</tr>
<tr>
<td>2011</td>
<td>962</td>
<td>239</td>
<td>34</td>
<td>3/ 2/ 2</td>
<td>9/ 6/ 6</td>
</tr>
<tr>
<td>2012</td>
<td>1027</td>
<td>295</td>
<td>26</td>
<td>4/ 3/ 0</td>
<td>15/ 12/ 0</td>
</tr>
<tr>
<td>2013</td>
<td>1256</td>
<td>264</td>
<td>22</td>
<td>6/ 4/ 1</td>
<td>27/ 18/ 5</td>
</tr>
</tbody>
</table>

*Modified live/inactivated/mixed/vector vaccine/unidentified vaccine

Comparison of feline sarcomas at vaccination sites and non-vaccination sites
Cats with vaccination site sarcomas were younger (median 8 years vs. 11 years old for non-vaccination site cases) in one study. The same study showed a declining risk of sarcoma development at a vaccination site with increasing age. Another study reported local recurrence in up to 45% cases and metastasis in 0-28% cases. Regarding the histological characteristics of injection site sarcomas, the same two studies reported that their location was mainly subcutaneous location and that there was often an inflammatory infiltrate consisting of lymphocytes and macrophages at the periphery of the tumor. In some cases, macrophages contained blue-gray/brown foreign material, which was hypothesized to be the remnants of aluminum adjuvants, although this has never been confirmed. The characteristics of aggressive tumors included increased mitotic activity, pleomorphism and tumor-associated necrosis.

Clinical Management of Injection Site Sarcomas
Owners should monitor all injection sites and incisional, wedge, or multiple tru-cut biopsy specimens should be submitted for histopathology if the 3-2-1 rule applies i.e.

- Mass is present 3 months post-injection
• Mass is ≥ 2cm in diameter
• Mass increasing in size after 1 month

Report all injection site sarcomas to the manufacturer and United States Pharmacopeia -


Tumor factors - The location, shape and size of the tumor should be recorded.

Cat factors – Record full details of signalment, history, clinical signs, treatment etc.

Injection factors – For all injections administered, record details of the product name, manufacturer, serial/lot #, expiration date, administration date, vaccine type, location of injection, person administering vaccine. Unnecessary injections and previous injection sites should be avoided.

Vaccine factors - Injectable vaccines should be administered SC not IM, to facilitate the detection of a mass. Multiple injectable vaccinations at single sites should be avoided, as should unnecessary vaccinations and previous vaccination sites.

Administer vaccines within 30 minutes of reconstitution and keep refrigerated at all times throughout transport & storage (2-7 °C – in the middle of the refrigerator). Follow the manufacturers instructions regarding reconstitution, administer 1 vaccine/single-use syringe and needle, dissolve completely and administer immediately. If spillage occurs, clean fur with alcohol swabs and surfaces with a 1:32 bleach solution.

Coordinated Management – For all cases, baseline laboratory data including CBC, serum biochemistry and FIV/FeLV status should be obtained. Chest radiographs (3 views) and abdominal ultrasound should be performed to check for metastases; imaging with CT/MRI prior to biopsy is optimal.

Therapy of Injection Site Sarcomas

Referral to a specialist surgeon is recommended, since aggressive surgical resection tumor removal with 5cm margins and 2 muscle planes is required to minimize the chance of tumor recurrence. In some cases, small sarcomas (<3cm) can be controlled with surgery alone, but a combination therapeutic approach, including surgery, radiation therapy and chemotherapy is thought to provide the best outcome. However, pre-operative radiation therapy can be associated with wound healing complications and post-operative radiation therapy has been associated with increased adverse effects in some studies.

Prognosis

The published prognosis for injection site sarcomas varies according to treatment regimen, study design and the presence or absence of recurrence and/or metastases; medial survival times from 9 months to 4.2 years have been reported.

Injection Site Sarcomas - Take-home messages

• Many types of injections have been associated with sarcomas, especially long-acting injections.
• Adjuvanted (killed) FeLV and rabies vaccines carry increased risk for injection-site sarcoma development, in the rear limb region only, in one study.
• No vaccines are risk-free.
• There is no evidence of reduced prevalence of injection site sarcomas in response to changes in vaccine formulation or recommended changes in feline vaccination protocols.
• Veterinarians should document injection sites and details of injections administered.
• Cat owners should monitor injection sites.
• Combination therapy helps provide the best outcome.

References
Available on request (Email: Annette.Litster@zoetis.com).