Exceptional *in vivo* wound healing following destruction of cutaneous and subcutaneous tumours in companion animals treated with the novel epoxy-tigliane drug EBC-46

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EBC-46 is a novel small molecule discovered from an Australian rainforest plant

EBC-46 is currently being developed, by Australian biotechnology company Gilbitics Ltd, for intratumoral treatment of cutaneous and subcutaneous tumours in humans and companion animals.

To date, more than 300 client-owned companion animals (dogs, cats and horses) with spontaneous tumours have been treated with EBC-46 in veterinary case studies.

In these studies EBC-46 has:
- caused significant local ablation of tumours in more than 80% of cases;
- shown efficacy against a wide range of tumour types including sarcomas, carcinomas, mastocytomas and melanomas; and,
- been well tolerated with a lack of any significant side effects at efficacious doses.

Case study 1: 11-year-old Beagle, spindle cell tumour

Case study 2: 11-year-old Whippet, soft tissue sarcoma

Case study 3: 8-year-old Shih Tzu, mast cell tumour

Case study 4: 8-year-old Stock horse, equine sarcoid

Minimal tissue deficit and minimal scarring were consistent features of the resolved wounds

EBC-46, and related epoxy-tiglianes, have significant potential as novel pharmaceuticals to aid in wound healing

The exceptional dermal wound healing observed in companion animals following tumour destruction by EBC-46 provides strong evidence that epoxy-tiglianes can directly facilitate wound repair. We hypothesise that the initial, acute pro-inflammatory response is one of the key mechanisms underlying the enhanced healing initiated by EBC-46.

Studies currently in progress are:
- (a) examining the direct effects of topically formulated epoxy-tiglianes on chronic wounds in companion animals; and,
- (b) investigating the apparent multi-factorial mode of action of epoxy-tiglianes in initiating favourable wound outcomes.

Epoxy-tigliane drugs may have significant future application in addressing chronic wounds in companion animals; and,

Examining the direct effects of topically formulated epoxy-tiglianes on chronic wounds in companion animals; and,

**Rapid formation of granulation tissue and enhanced re-epithelialisation characterise wound resolution following tumour destruction by EBC-46**

Time to closure and basic macroscopic wound characteristics were assessed following destruction of cutaneous tumours by EBC-46 in case studies of 29 dogs and 10 horses. In all instances there were no active wound management interventions (e.g. dressings, suture, antibiotics, or other concurrent medications) following the initial treatment of the tumour with EBC-46.

Time for full wound closure was strongly related to wound size (Figure 1). Closure rates were favourable compared to data available in the veterinary literature (e.g. Bohling et al. 2004, Vet Surg. 33: 579; Weikert & Weeren 2000, Vet Clin Equine 27: 151). Healing of canine and equine trunk wounds was primarily by contraction, whereas re-epithelialisation was more important for wounds on extremities (and consequently these wounds are more representative of healing processes in humans).

Case studies 2 to 4 below provide typical examples of the time course and features of wound resolution that followed tumour sloughing.

**EBC-46 is under development to treat cancers in humans and companion animals**

EBC-46 is a signalling molecule, rather than a cytotoxic, and causes rapid haemorrhagic necrosis of treated tumours through a combination of:

- local recruitment and activation of leukocytes, especially neutrophils and macrophages;
- initiation of an acute, but highly localised, pro-inflammatory response immediately surrounding the tumour; and,
- direct disruption of the integrity of tumour vasculature.

Short-term activation of specific isoforms of protein kinase C is fundamental to the mode of action of the drug in destroying tumours.

EBC-46 initiates rapid haemorrhagic necrosis of tumours followed by exceptional wound healing

Dependent on tumour type and position on the body, necrotic remnants of cutaneous tumours treated with EBC-46 generally slough within 4 to 14 days of injection leaving open wounds. Without further intervention, these wounds show exceptional spontaneous resolution characterised by:

- rapid formation of granulation tissue development;
- enhanced re-epithelialisation;
- minimal tissue deficit; and,
- minimal scarring.

Case study 1 illustrates the course of tumour destruction and subsequent wound healing following a single intratumoral treatment with EBC-46.

**Figure 1:** Wound size versus days to complete closure of open wounds that developed from excision of a spontaneous tumour by EBC-46 in 29 dogs (blue symbols) and 10 horses (green symbols).

<table>
<thead>
<tr>
<th>Case studies</th>
<th>Wound location</th>
<th>Dogs</th>
<th>Horses</th>
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<tbody>
<tr>
<td>Case study 2</td>
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<td>4</td>
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<tr>
<td>Case study 3</td>
<td>Extremity</td>
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<td>1</td>
</tr>
<tr>
<td>Case study 4</td>
<td>Extremity</td>
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**Table 1:** Tissue and hair features of treated wound sites at 30 consecutive annual following sloughing of spontaneous tumours treated with EBC-46.

Epoxy-tiglianes in initiating favourable wound outcomes.