

**SOP No:** AHP 89

**SUBJECT:** Induction of laminitis in horses by alimentary dosing with oligofructose carbohydrate

**POLICY:** This procedure may only be performed by veterinarians skilled in the technique. Horses must be monitored constantly throughout the experimental period by experienced veterinarians. Mild or temporary discomfort experienced during placement of catheters must be managed with intravenous pain relief drugs and local anaesthetic nerve blocks. At the onset of any persistent clinical discomfort shown by the horses, they must be euthanased immediately.

**PRECAUTIONS:** Boots, overalls, lead apron

**EQUIPMENT:** Oligofructose powder  
4L tap water  
Nasogastric tube  
2% Lignocaine  
Xylazine  
Ketamine  
Flunixin meglumine  
Romifidine  
Equine nappy  
Foot support blocks  
X-ray equipment

**PROCEDURE:**

**Jugular vein catheterization:**

Local anaesthetic (2% lignocaine, approximately 3 ml) is infused subcutaneously into the clipped, surgically prepared skin over the jugular vein. When desensitized the skin is incised (2-3 mm incision) and a catheter (14G flexible, extended use intravenous catheters (Milacath, Mila International) inserted into one vein and sutured in place. Collect jugular venous samples for biochemistry, haematology at time 0h and time 24h. Collect blood (12ml) for proteomic analysis every 2 h. Draw off the initial 5mls of blood and discard. Draw a total of 20ml for each sampling. Flush the catheter with heparinised saline after each sample to ensure patency.

**Digital vein catheterization:**

Local anaesthetic (2% lignocaine, approximately 2 ml) is infused subcutaneously into clipped, surgically

prepared skin over the lateral digital vein at the abaxial sesamoid site. When desensitized the skin is incised (2-3 mm incision) and a catheter (18G flexible, extended use intravenous catheter (Milacath, Mila International) with extension tubing inserted into one vein and sutured in place. The extension tubing is housed in a soft neoprene leg boot between usage. Collect digital venous blood samples (12ml)

1. To select laminitis free animals, sedate the horse with xylazine (0.6 mg/kg) by i/v injection into the left jugular vein. Stand the horse with both feet on a purpose- built wooden block 12 cm thick to align the feet parallel to the x-ray beam. Make exposures of the lateral to medial projection. Take a second radiograph at 24hr.
2. Fit the horse with an equine nappy to collect urine and faeces in separate compartments and to maintain stable hygiene
3. Administer 10g/kg body weight oligofructose (OF) powder dissolved in 4 litres of tap water via nasogastric tube.
4. After dosing, observe the horse continuously and collect data at 4 h intervals over a 24h study period. Make clinical observations regarding general demeanour, foot behaviour, oral mucous membrane capillary refill time, heart rate, digital pulse, faecal consistency, faecal pH, gut sounds (left colon and ileo-caecal sounds) and rectal temperature. Collect urine samples for urinalysis at time 0h and time 24h,
5. At the end timepoint (24hr) and prior to euthanasia anaesthetize the horse using the following regimen: administer 1.1mg/kg body wt xylazine i/v; 5min later administer 2.2mg/kg body wt ketamine i/v followed by 5mg/kg body wt thiopentone sodium administered i/v with further intravenous boluses of 2mg/kg body wt administered if required (to maintain deep general anaesthesia. Immediately following anaesthesia, apply Esmarch's Bandage to the fetlock of one fore limb.
6. Remove the fore and hind feet by disarticulation at the metacarpal/phalangeal joint when satisfactory deep general anaesthesia has been achieved (based on a lack of nystagmus, ventromedial or central eye position, lack of palpebral reflex and lack of any response to stimuli). Cut each foot on a bandsaw to harvest the dorsal hoof wall lamellae. Dissect each foot to obtain 5mm square lamellar explants and snap freeze in liquid nitrogen for

subsequent genomic/proteomic work. Immediately place other samples into 4% formaldehyde for 24 hours then embed in paraffin blocks. The estimated time for the processing of one fore foot is 3-5 min. The second fore foot is processed in the same way and the horse euthanased.

7. Euthanase the animal by overdosing with sodium pentobarbitone to effect.

**RECOMMENDATIONS:**

Administer finadyne (flunixin meglumine 50mg/ml) solution for relief of abdominal discomfort at a dose rate of 1.1mg/kg (10mls) iv if required. Administer 2% lignocaine for nerve blocks for pain relief, 1-2mls infused subcutaneously peri-neurally every 3-4 hours or as required if pain becomes evident. Administer Sedivet (Romifidine 1%) 1.5 – 5mls i/v if required for sedation for placement of catheters or nerve blocks

**DATE ISSUED:** 28 October 2009

**REVISED:**

**REFERENCES**

1. van Eps, A.W. and Pollitt, C.C. (2006) Equine laminitis induced with oligofructose. *Equine Vet J* 38, 203-208.