Diagnosing Insulin Dysregulation &
Equine Metabolic Syndrome

Equine metabolic syndrome is characterised by insulin dysregulation, altered adipokine concentrations, dyslipidaemia and a predisposition to laminitis. Diagnosis of insulin dysregulation can be a real challenge in equine practice. As our understanding of the underlying disease improves, we recognise the limitations of the currently available tests and reference ranges.

This document summarises some of the currently available information, which should help when selecting and interpreting laboratory tests in clinical practice.

Collection tube requirements:
- Insulin: Serum tube
- Glucose: Fluoride oxalate tube

1a Basal insulin concentration
This is a convenient screening test. However, the test has a low sensitivity which means that a negative result does not rule out the presence of insulin dysregulation. A positive test result however, is really useful and confirms the presence of insulin dysregulation.

Basal insulin concentration is routinely measured after a 6 hour fast. A single serum sample is needed for analysis.

Fasting may further reduce the sensitivity of the test and in the future we may recommend measurement after feeding grass hay only (ideally hay low in non-structural carbohydrates NSCs <12%). Access to rich pasture, haylage or concentrated feed should be avoided before testing.

1b Interpreting the result
Our current recommendations after a 6 hour fast are:

< 10 iu/ml
A basal insulin concentration of less than 10 iu/ml is considered within an acceptable range. The large majority of horses with this result do not have insulin dysregulation. However, on oral sugar test is still required to fully rule out insulin dysregulation.

10 – 15 iu/ml
A basal insulin concentration in this range is equivocal for the presence of insulin dysregulation. An oral sugar test is recommended to more accurately confirm status.

15 - 20 iu/ml
A basal insulin of 15 - 20 iu/L is moderately accurate in assessing insulin dysregulation. The majority of horses with basal insulin >15 iu/L do have insulin dysregulation, but to confirm the status more accurately, an oral sugar test is required.

>20 iu/ml
Provided that the horse has been starved for at least 6 hours, this result has a good predictive value for ruling in insulin dysregulation, but still with occasional false positives. To confirm status most accurately, an oral sugar test is required.
Oral sugar tests

By challenging the horse with oral sugar either in the form of dextrose powder or corn sugar syrup a better evaluation of insulin function can be made. Suggested protocols are as follows:

Karo syrup test:

- Ideally fast horse for 6 hours or feed only low NSC forage.
- Administer 0.15mls/kg Karo syrup by dosing syringe (Alternative doses are 0.25ml/kg or 0.45ml/kg - see below) Karo syrup can be purchased at www.amazon.co.uk or in some supermarkets.
- 60 to 90 minutes after dosing, blood sample twice at 15-minute intervals for measurement of insulin and glucose.

Oral sugar test:

- Ideally fast horse for 6 hours or feed only low NSC forage
- Feed 1g/kg bodyweight powdered dextrose, mixed with 1g/kg bodyweight low sugar chaff and 1ml/kg bodyweight water.
- Collect a blood sample for measurement of insulin and glucose 2 hours later. It should be noted if/how much feed has been left.

At the current time it is not certain which of these tests is the most accurate. The advantage of the karo syrup test is that it does not rely on the horse eating dextrose powder that is not always palatable. Using a higher dose of Karo syrup may yield more accurate results but at this current time we still recommend the 0.15 ml/kg dose as the first line test, as this has been the most thoroughly investigated. A cut-off of 30 iu/ml may be better than the historically reported 60 iu/ml using this dose rate.

There is a lack of agreement about normal ranges at this time but the following table summarises currently available data:

<table>
<thead>
<tr>
<th>Test</th>
<th>Positive TEST cut-off iu/ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>Oral sugar challenge (1g/kg)</td>
<td>&gt;85</td>
</tr>
<tr>
<td>Karo syrup 0.15ml/kg</td>
<td>&gt;30</td>
</tr>
<tr>
<td>Karo syrup 0.25ml/kg</td>
<td>&gt;30</td>
</tr>
<tr>
<td>Karo syrup 0.45ml/kg</td>
<td>&gt;55</td>
</tr>
</tbody>
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Combined Glucose Insulin Test

This test is less practical to carry out in general practice but is a better test for insulin resistance (rather than function) and can be useful when the results of other tests have been equivocal. This is still the most accurate test for diagnosis of insulin dysregulation

A suggested protocol is below:

- Overnight fast then collect basal samples for measurement of glucose and insulin.
- Administer 150ml of 50% glucose per 500kg intravenously followed by 0.1 iu/kg soluble insulin (0.5 ml 100iu/mL per 500kg) intravenously.
- Collect blood samples for glucose at 1, 5, 15, 25, 35, 45, 60, 75, 90, 105, 120, 135, and 150 minutes.
- Collect a blood sample for insulin at 45 minutes.
- A normal response is a glucose concentration back to baseline by 45 minutes and an insulin concentration < 100 iu/ml at 45 minutes.
- If clinical signs of hypoglycemia (sweating, weakness, and muscle fasciculation) occur 60 mL of 50% dextrose should be given intravenously and repeated if necessary.

Other markers for Equine Metabolic Syndrome

Adiponectin - There is a great deal of interest in measuring the cytokines that are released from adipose tissue as a marker of Equine Metabolic Syndrome. Adiponectin is an anti-inflammatory cytokine that has a role in glucose regulation. High molecular weight adiponectin accounts for the majority of circulating adiponectin and its concentration is inversely proportional to body condition score and insulin concentration.

Early studies failed to correlate adiponectin with insulin concentration and laminitis risk. However, more recent work has shown a greater association between high molecular weight adiponectin concentration and laminitis risk.

A concentration of less than 2.5 ng/ml measured using a radioimmunoassay may be associated with an increased laminitis risk. However, further work is needed before measurement of adiponectin can be considered a first line test.

A good summary for further reading is The diagnosis of equine insulin dysregulation, Bertin FR, de Laat MA, Equine Vet J 2017 Sep;49(5):570-576
References:

1. Bamford NJ, Potter SJ, Harris PA, Bailey SR. Effect of increased adiposity on insulin sensitivity and adipokine concentrations in horses and ponies fed a high fat diet, with or without a once daily high glycaemic meal. Equine Vet J. 2016; 48:368-73


10. Rendle DR, Laboratory diagnosis of the endocrine causes of laminitis Livestock July/August 2017, Volume 22 No 4