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# Relaxin for pregnancy detection

### AMY WEEDEN

Gribbles Veterinary offers relaxin testing for the detection of pregnancy in both dogs and cats. This hormone is superior to progesterone measurement for pregnancy detection in both species.

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Progesterone cannot reliably distinguish between pregnancy and pseudopregnancy in dogs or cats, and similar levels of progesterone are produced in pregnancy and dioestrus in dogs. Relaxin does not increase during pseudopregnancy in dogs<sup>1</sup> or cats or in non-pregnant bitches in dioestrus.

Relaxin testing is most commonly used in dogs, but a 2010 study showed that the assay used by Gribbles also works well in cats. The study involved 162 female cats, including 24 queens from a breeding colony. The earliest date that pregnancy was detected in this study was gestational day 20. By gestational day 29, the sensitivity of the



test was 100%. False positives occurred in three queens, two of which had large ovarian cysts. Two non-pregnant queens with smaller ovarian cysts were negative for relaxin, suggesting that not all cats with ovarian cysts will have false positive results.<sup>2</sup>

Paws claws and inder things

An older study in Labrador Retrievers and Beagles (using a different assay) looked at plasma relaxin levels in pregnant and lactating dogs. This study showed that plasma relaxin is first detectable in week three or four of gestation, peaks 2-3 weeks before whelping, declines significantly until whelping, and remains at a low level for several weeks into lactation.<sup>1</sup>

There are some test limitations and stability issues to be mindful of. Similar to the findings in the studies above, the manufacturer of the Relaxin kit reports that the test may detect pregnancy as early as gestational day 20, but the test is not 100% sensitive until day 31 in either species.<sup>3</sup> Negative results before this date should be confirmed by retest or other diagnostic means (e.g. ultrasound).

In addition to timing of the test, accuracy may also be affected by size of the bitch and size of the litter (e.g. a large bitch with a small litter may have undetectable relaxin). Relaxin should remain positive through gestation.<sup>3</sup> Note relaxin does decline near parturition, so results could be negative at this time with a small litter size (anecdotal experience), though data from the manufacturer and the JAVMA study<sup>2</sup> do not bear this out.

Relaxin will degrade over time, and the test should be run within four hours of sample collection if the sample (serum or plasma) is kept at room temperature.<sup>4</sup> Samples can be kept refrigerated for up to two days, and longer term storage requires freezing for sample stability (samples should ideally be separated prior to refrigerated storage, and samples MUST be separated for frozen storage).<sup>4</sup> As mentioned above, ovarian cysts have caused false positives in rare documented cases in cats.<sup>2</sup>

A brief summary of our relaxin test can be found in the Gribbles Veterinary Handbook on our website here.

#### References:

- Steinetz BG, Goldsmith LT, Lust G. Plasma relaxin levels in pregnant and lactating dogs. *Biol Reprod*. 37:719-25, 1987.
- DiGangi BA, Griffin B, Levy JK, Smith BF, Baker HJ. Use of a commercially available relaxin test for detection of pregnancy in cats. J Am Vet Med Assoc. 237:1267-74, 2010.
- 3. <u>https://www2.zoetisus.com/content/\_assets/docs/</u> <u>Diagnostics/brochures/WITNESS-Relaxin-Sell-</u> <u>Sheet-WIT-00100.pdf</u>
- 4. Zoetis Witness® Relaxin package insert

# What's inside?

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## A tale of high drama

### **BERNIE VAATSTRA**

Loki is a lively two-year-old male neutered English Springer Spaniel with a strong interest in food of all kinds, edible or otherwise. On a Friday evening on returning from work, Loki's owner noticed him behaving unusually. He barked sharply at the car he usually recognises and approached with a swaying, uncoordinated gait.

### Clinical findings:

Closer examination revealed a horizontal head tremor, dilated pupils, sluggish pupillary reflexes, and hypermetria. Temperature was 38.5, heart rate and respiratory rates were within normal ranges.

On questioning the family members, it was ascertained that Loki had been taken for a walk around some council gardens and a local rugby club approximately three hours previously. During the walk, he was noticed to have snuffled around in a particular area and surreptitiously eaten a "strange green object". The children walking him were unable to prevent him swallowing the item. On return home, Loki was fed his usual dry biscuits half an hour prior to clinical signs first being noticed.

Suspecting exposure to a poison, and with Loki still alert and responsive, emesis was induced with a small dose of apomorphine. A small amount of dog food was brought up but nothing to suggest what might have been



ingested on the walk. Loki was then administered activated charcoal and placed in a warm cage for the night.

The following morning, Loki was extremely sedated with ongoing incoordination and head tremor. He was able to be roused and taken out for toileting, but quickly tired and stumbled. He also developed urine dribbling. Serum and urine samples were collected and further charcoal was administered.

Based on the history and clinical signs, a CNS toxicity was suspected. Differential diagnoses included tremorgenic mycotoxins, macrocyclic lactone toxicity, ethylene glycol (antifreeze) toxicity, metaldehyde (slug bait) toxicity, mushroom toxicity, and cannabis toxicity. A biochemistry panel was requested to assess for liver and renal damage and therefore help rule out ethylene glycol and metaldehyde exposure.

### Laboratory results:

Biochemistry was unremarkable aside from mild increases in ALT (103 IU/L – reference interval 0-88 IU/L), AST (125 IU/L – reference interval 0-51 IU/L) and CK (918 IU/L – reference interval 0-385 IU/L) concentrations. These findings suggested subtle hepatocellular and muscle damage, possibly explained by liver metabolism of an ingested toxin and tremors and ataxia, respectively. Urinalysis was unremarkable. Overall, these findings supported a primary CNS toxin and helped exclude ethylene glycol and metaldehyde poisoning.

A urine sample was sent to a human laboratory for analysis. The carboxy-THC concentration was 23  $\mu$ g/L (cut-off 15  $\mu$ g/L) and the carboxy-THC:creatinine ratio was 4  $\mu$ g/mmol, confirming the presence of cannabis metabolites in the urine. The final diagnosis was cannabis toxicity.

### Clinical progress:

Loki's clinical signs of incoordination and depression improved gradually over approximately 72 hours. He was noticeably hyper-reactive and hungry on the third day. After approximately four days, he seemed back to normal.

Photograph left: Loki enjoying subsequent good health and a delicious chewy.

#### Discussion:

Cannabis (marijuana) toxicity occurs due to ingestion of leaves, flowers and products made from the *Cannabis sativa* plant. The plant contains hundreds of chemicals although  $\delta$ -9-tetrahydrocannabinol (THC) is the major psychoactive agent. Given the widespread recreational use and increasing medicinal use of cannabis, poisoning in companion animals may increase in incidence. Fortunately, cannabis has a relatively high safety margin and deaths in dogs are rare. The minimum lethal dose of THC is >3g/kg (Fitzgerald et al, 2013).

Clinical signs of cannabis toxicosis develop within 60 minutes and may last for several days, mediated largely by cannabinoid receptors distributed throughout the brain. Common manifestations include sedation, hypersalivation, hypermetria, mydriasis, urine dribbling, head tremor, hypothermia, and bradycardia. Higher doses may cause hyperexcitability and seizures (Fitzgerald et al. 2013). Treatment is largely supportive since there is no specific antidote.

Diagnosis is through appropriate clinical signs, history of ingestion, and ruling out other causes through biochemistry, haematology and urinalysis. Detection of THC metabolites in urine may help confirm the diagnosis but many human assays have low sensitivity in dogs due to low concentrations of the specific THC metabolites in canine urine. A novel ultraperformance liquid chromatography–tandem mass spectrometer method with excellent sensitivity was recently described and may prelude more reliable diagnostic assays becoming available for domestic animals (Fitzgerald et al. 2021).

### References:

Fitzgerald KT, Bronstein AC, Newquist KL. Marijuana poisoning. *Top Companion Anim Med.* 28:8-12, 2013

Fitzgerald AH, Zhang Y, Fritz S, Whitehouse WH, Brabson T, Pohlman L, Cernicchiaro N, Tonozzi C, Ensley S. Detecting and quantifying marijuana metabolites in serum and urine of 19 dogs affected by marijuana toxicity. *J Vet Diagn Invest.* 33:1002-1007, 2021.

## Facial eczema season approaching

For every clinical case of facial eczema you see, there will be at least ten sub-clinically affected animals. Infected stock will fail to thrive, have reduced milk production, poor fertility, lose weight and possibly die.



Summer is just around the corner and it is never too early to ensure plans are in place to protect the herd. To be effective, preventative measures need to be in place **before** *Pithomyces chartarum* spores are found.

# It's what you can't see that you should be worried about.

We offer a range of cost effective solutions to assist with monitoring facial eczema (FE) risk, minimising the incidence of disease, checking your management programme is working and assessing the damage caused by sporidesmin toxin from ingested spores.

Testing options available include:

- > Spore counts (pasture and faeces)
- > Individual GGT and zinc testing

The national FE spore count monitoring service coordinated by Gribbles Veterinary is active during the summer and autumn months. With our <u>online Lab-portal</u>, you can start submitting your local spore counts as soon as you start doing them.

Data is shown in real-time on the graphs in the Lab-portal, so you will always be up-todate with the latest local and national trends. All veterinary clinics can register for a user account and we encourage you to do so. The more data that is received in the portal, the better the indication or risk in your region.

See full details of how you can keep on top of facial eczema in the article and information sheet located on our website.

# New - online shopping FAQs!

We frequently get calls from clients who strike problems when shopping online for consumables. Passwords or usernames are forgotten, does the clinic actually have a user account for our website etc. So in the last few issues of our newsletter, we have provided some tips on what to do when your online shopping doesn't quite go as planned.

We've now taken this a step further, and have added a **FAQ section** to our website <u>consumables ordering page</u>. This will hopefully cover all the common issues or questions you may have. Plus we have provided a direct contact link for our website administrator (should the FAQs not help) to save you ringing the laboratory.

So the next time you strike out, check through the FAQ list and see if they can resolve your issue. We hope they save you some time and effort, and make your shopping experience that much better!



## Let's talk turkey

Ready to talk Christmas yet? Just in case you do want to know, it is just roughly even weeks until Christmas day. So get making your cakes and fruit mince for pies already! Too early? Sorry, just skip this bit then!

This year we will be closed ALL of Christmas and New Year weekends, including the public holidays.

### **Christmas hours:**

24 December >	OPEN
25 - 28 December >	CLOSE
29 - 31 December >	OPEN

#### New Year hours:

1 - 4 January >

CLOSED

5 January onwards > OPEN

We will remind you again in December, but thought we'd get in early in case you were making plans.



# Consumable of the month

Do you order laboratory consumable items from us online or via our order form? If you need just one blood tube or swab, or enough for a herd, we've got you covered.

Our featured consumable item this month is **blood film spreader slides.** So if you really feel like upping your blood smear quality, this might just be the tool for you!

So what is a spreader slide? These slides are narrower than the smear slide to avoid spreading the cells over the edge of the slide. They are also slightly thicker so do not break as easily.

We recommend you keep your spreader slide with the working end in a small container of water. This will ensure it is free from blood and ready to use (after drying off).

These slides can be found in the "<u>General lab supplies</u>" area of our online

shop and are sold individually. If you would like to be in to win a couple of these, make sure you stay tuned to our <u>Facebook page</u> and enter the giveaway this month.

For more information on how to make the perfect blood film, head to the USEFUL FORMS & INFO section on our website under Veterinary Info, and download a copy of our blood film preparation guide.



If you follow us on Facebook, you'll be familiar with our regular Friday slot. Here's one of our recent popular posts to finish this newsletter off with.

If you don't follow us on <u>Facebook</u>, head over and hit the button now!









## Contact us

Contacting Gribbles Veterinary couldn't be easier.

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