# EX VIVO SENSITIVITY AND ROBUSTNESS OF A NOVEL HOME SCREENING TEST FOR FELINE GLUCOSURIA





## Introduction

Diabetic cats are late to exhibit visible clinical signs, and glucosuria is one of the first detectable signs of feline diabetes. A novel screening test for feline glucosuria has been developed. It consists of white, absorbent granules, which are sprinkled on mineral-based cat litter, turning blue when in contact with glucose-containing urine.

## Objectives and hypothesis

Hypothesis	The test can consistently detect clinicall glucosuria
Objectives	<ul> <li>determine the lower limit of detection glucosuria</li> <li>verify the product robustness in altere composition</li> </ul>

## Materials and methods

- Granules chromogenic reaction evaluation: in triplicate, by applying 50 µL of glucose-fortified feline urine onto granules at glucose concentrations ranging from 0 – 1000 mg/dL.
- **Different urinary compositions**, with various urinary specific gravities (USG), pH, proteinuria, ketonuria and hematuria.
- Time of reading: coloration intensities noted at 5, 10, 20, 30, 60 minutes and at 2, 4, 8, 24 and 48 hours after exposure to the ex vivo-fortified urine.
- Color intensity of the chromogenic reaction (Fig. 1): scored with an ordinal visual scale from 0 (none) to 3+ (intense)



Figure 1: Chromogenic intensity scale

- Variables verification/measurement methods:
  - glucosuria: chemistry analyzer (DxC600®, Beckman Coulter)
  - urinary pH: pH-meter (Hi 2222<sup>®</sup>, Hanna Instruments)
  - USG: refractometer (Reichert VET 360®) ketonuria: urinary strips (Chemstrip9® Roche Diagnostics)
  - hematuria: microscopic evaluation
- Data analysis: mixed logistic regression model for repeated measures, taking into account glucosuria, time, USG, pH, proteinuria, ketonuria and hematuria.

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## The test adequately detects glucosuria at clinically relevant levels. The color remains stable for at least for 48 hours.

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### Lower limit of detection (LLD): **20mg/dL** (1,1mmol/L).

Appearance of chromogenic reaction usually between 1 and 3 minutes after contact with glucosuria. LLD significantly increased with the time of reading, i.e. LLD = 30-50 mg/dL at 48 hours; p-value < 0.05 (Table 1).

Glucosuria (mg/dL)

Granules score 10 min

Granules score 30 min Granules score 24h

Granules score 48h



<u>Table 1:</u> Chromogenic responses of the granules (over 48 hours), urinary sample with SG=1,013, pH=6.

### LLD significantly increased with:

## Discussion

### **Relevance of this test:**

- Diabetes mellitus (DM) is one of the most common feline endocrinopathies, occurring in an estimated 0.4%-0.7% of cats<sup>1,2,3,4</sup>, with evidence that this prevalence is increasing<sup>3,4</sup>.
- Glucosuria is one of the first clinical signs of DM<sup>1,2</sup>.
- Relevance of an early diagnosis of DM<sup>1</sup> and relevance of urine glucose measurement in the monitoring of DM<sup>5</sup>
- **Relevance of this limit of detection:**
- Mean glucosuria among DM cats = 864mg/dL<sup>6</sup>,
- Mean glucosuria among healthy cats  $\leq 7 \text{mg/dL}^6$ , Glucosuria > 25mg/dL is considered as pathologic<sup>6</sup>.
- With a reading at 48 hours, the granules detect as low as 30-50mg/dL, over the pathological limit and far below DM cats mean glucosuria levels.
- Limits of this study: ex vivo controlled conditions. Parallelly presenting a test in clinics and households with feline patients.

## Results

0	20	30	50	75
0	1+	2+	2+	3+
0	1+	1+	2+	3+
0	0	1+	2+	3+
0	0	1+	2+	3+

increasing pH (i.e. LLD = 100 mg/dL at pH  $\geq$  8.2; p-value < 0.0001),

**USG** (i.e. LLD = 100mg/dL at USG  $\geq 1,060$ ; p-value < 0.0001),

**High proteinuria** (i.e. LLD = 100 mg/dL at proteinuria  $\geq$  5g/L; p-value < 0.0001).

No significant effect of ketone bodies and hematuria on the reactivity (p-value > 0.05).

1. Sparkes AH, Cannon M, Church D, Fleeman L, Harvey A, Hoenig M, et al. ISFM consensus guidelines on the practical management of diabetes mellitus in cats. J Feline Med Surg. 2015;17(3):235-50. 2. O'Neill DG, Gostelow R, Orme C, Church DB, Niessen SJ, Verheyen K, et al. Epidemiology of Diabetes Mellitus among 193,435 Cats Attending Primary-Care Veterinary Practices in England. J Vet Intern Med. 2016;30(4):964-72.

3. Banfield. State of Pet Health Report. 2016. 4. Baral R, Rand J, Catt M, Farrow H. Prevalence of feline diabetes mellitus in a feline private practice (abstract). Journal of Veterinary Internal Medicine. 2003;17(3):p. 433-4. 5. Behrend E, Holford A, Lathan P, Rucinsky R, Schulman R. 2018 AAHA Diabetes Management Guidelines for Dogs and Cats. J Am Anim Hosp Assoc. 2018;54(1):1-21. 6. Zeugswetter FK, Polsterer T, Krempl H, Schwendenwein I. Basal glucosuria in cats. J Anim Physiol Anim Nutr (Berl). 2019;103(1):324-30.



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## Conclusion

The test adequately detected glucosuria at clinically relevant levels and is suitable for both clinical and household trials.

## Literature cited

## For further information