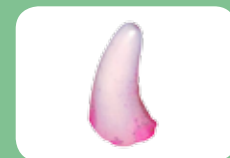


## Accurate measurement of dental plaque extent is now possible on conscious dogs

When all the commonly used techniques require anaesthesia, this promising new method based on image-analysis allows dental plaque coverage assessment without any sedation.

Accurate assessment of the extent of dental plaque coverage on tooth surfaces is essential in periodontal research. In veterinary medicine, several conventional indexes such as Logan & Boyce plaque index are used to assess the dental plaque coverage over the crown surface, but they have a major limitation as they all require general anaesthesia. Indeed, a conventional "clean tooth model" procedure requires at least 3 successive anaesthesia. A gingival plaque contour index was recently described through the measurement of plaque accumulation on conscious dogs, but it relies on visual evaluation.



After contouring the tooth, a plaque/tooth surface ratio is automatically calculated.

The group was composed of 2 dolichocephalic and 6 mesocephalic dogs, from different size and breeds (Fox terrier, Shetland, Brittany spaniel, English cocker, Golden retriever, White shepherd). Repeatability (intra-operator variation) was assessed on 8 teeth from each dog, by the same trained duo of operators, on 6 repetitions of the whole method. The average coefficient of variation was 12.39% (+/- 2.73), showing that the complete method was repeatable. Reproducibility (inter-operator variation) was performed by 2 different duos of operators on 8 teeth from each dog, with 3 repetitions per duo. No difference was detected between the two duos of operators for the mean of plaque/tooth ratio, showing that the global method was reproducible.

*This study validated a new method of dental plaque coverage assessment on trained conscious dogs. This new image-analysis system allows an accurate evaluation and monitoring of dental plaque deposition, avoiding anaesthesia. Compared with conventional indexes, this new method helps quantify the dental plaque coverage providing continuous data and allows archiving of pictures.*

Mariani C, Boutoille F, Shrum B, Biourge V, Warnery R, Hennet P. Validation of a new method based on an image analysis system for the measurement of dental plaque accumulation in conscious dogs. Proceedings of the 22nd ECVD Congress, Prague (Czech Republic), May 23-26, 2013.

The purpose of this study was to validate a new method for dental plaque coverage assessment in conscious dogs based on an image-analysis system, previously validated (see News from Research N°5).

This method relies on image-analysis method and consists of 3 steps:

- After applying a plaque disclosing solution (erythrosine) on the crown surface of the teeth, standardized pictures of the maxillary and the mandibular dental arches are taken.
- Then, using a graphic tablet and a photo editing software, crown surface are contoured.
- Finally, using a specific algorithm analyzing the disclosed plaque surface, a plaque/tooth surface ratio is calculated.

To assess the repeatability and reproducibility of this method in conscious dogs, 8 healthy adult dogs with full dentition were included.



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## Intro

### Homemade diets, commercial diets and Health Nutrition.

A recent study published in the JAVMA<sup>1</sup> in cooperation with the American College of Veterinary Nutrition has shown that 95% of home-made diets found in veterinary textbooks, pet care books for owners and websites are nutritionally unbalanced, with at least one essential nutrient at concentrations that did not meet NRC or AAFCO guidelines!

Royal Canin is going further than just ensuring the complete nutritional profile of our diets. As a responsible company, we also have to show the benefits of our nutritional answers on cats and dogs health. In

this objective, one of the motto of our Research team is that: "we are only able to improve what we are able to measure". In this issue of News from Research, this motto is illustrated by the article introducing a new method to quantify dental plaque extent on conscious dogs. Developing new ways to assess the benefits of nutrition on health is at the foundation of our Research. It allows us to assess the impact of nutrition on cats and dogs, but these new methods can also be shared more widely with other teams to conduct their own evaluation studies.

<sup>1</sup>STOCKMAN, J., FASCETTI, A. J., KASS, P. H. & LARSEN, J. A. 2013. Evaluation of recipes of home-prepared maintenance diets for dogs. J Am Vet Med Assoc, 242, 1500-5.  
NRC: National Research Council  
AAFCO: Association of American Feed Control Officials

## Mobility

### A diet enriched in Green-lipped mussel reduces clinical signs of osteo-arthritis in dogs

The effect of Royal Canin Veterinary Diet Mobility Canine was assessed in 23 client-owned osteoarthritic dogs.

Green-lipped mussel is a complex natural product comprised of several potent bioactive compounds. GLM extracts have been shown to have chondro-modulator and anti-inflammatory properties. This prospective study was conducted to evaluate the effects of a diet enriched in GLM and fish oil (source of EPA and DHA fatty acids) on pain behaviour and functioning in dogs with clinical osteo-arthritis (OA).

Thirty adult dogs, with no health concerns other than chronic lameness were included. An orthopedic examination and peak vertical force (PVF) gait analysis, using a biomechanical force platform, established the severity of lameness in each joint and limb. A radiographic diagnosis confirmed osteoarthritis. Six dogs had one OA lesion associated with lameness in a limb. This lame limb was evaluated throughout the study. For dogs with multisite OA lesions (n=24), the limb most affected by lameness, as shown by gait analysis and orthopedic examination, was followed throughout the study. Amongst exclusion criteria, the dogs could not receive oral nutraceuticals or sporadic non-steroidal anti-inflammatory drugs (NSAIDs) for a 4-weeks prior to the trial.

To standardise food regime, all dogs were first fed the same commercial control diet (\*) from inclusion (day 0) to day 30. Then, they were fed exclusively ROYAL CANIN Mobility Support Large Breed Canine from day 31 to day 90. The daily food rations were calculated using the manufacturer's recommendations based on the body weight of the dog measured at day 30 and day 90. Data acquisition was performed at day 0, day 30, and day 90. Plasma omega-3 fatty acid (EPA and DHA) were measured and gait was analysed using peak vertical force (PVF), which is considered to be the gold standard method. Owners also completed a pain questionnaire once a week (Client Specific Outcome Measure questionnaire: CSOM), where they recorded up to 5 activities that were

the most impaired by the painful condition of OA. Twenty-three dogs completed the study (7 dogs were removed because their deteriorated condition required the use of NSAIDs (n=4) or because the owners did not comply with the guidelines (n=3)). The GLM diet resulted in an increase in concentrations of plasma EPA and DHA (p<0.016), and a significant improvement of PVF (p=0.003), whereas PVF did not significantly change from day 0 to day 30, when dogs were fed the control diet. Pain behaviour, as assessed with the CSOM questionnaire, also significantly improved from day 0 to day 90.



*The GLM-enriched diet improved gait in dogs with clinical osteoarthritis, with a significant increase in peak vertical force, which is the gold standard method to assess the severity of lameness. This diet also contributed to reduce pain behaviour over the study period.*

	Inclusion (Day 0)	Day 30	Day 90
Mean body weight (kg)	40.4 kg (SD 8.9)	40.3 kg (SD 8.5)	41.4 kg (SD 8.38)
Mean Peak vertical ground reaction force adjusted to body weight change (% BW)	65.4% (SD 17.2)	67.3% (SD 19.8)	69.9% <sup>a,b</sup> (SD 21.3)
Median pain score (CSOM questionnaire with scores from 0 to 4)	2.0	1.5 <sup>a</sup>	1.0 <sup>a</sup>

a: significant difference from day 0 (p<0.05)  
b: significant difference from day 30 (p<0.05)

Riolland P, Bichot S, Lussier B, Moreau M, Beaudry F, Del Castillo J, Gauvin D, Troncy E. Effect of a diet enriched with green-lipped mussel on pain behavior and functioning in dogs with clinical osteoarthritis The Canadian Journal of Veterinary Research 2013;77:66-7

(\*)Purina Dog Chow for adult dogs, without GLM, fish oil or glucosamine-chondroitin

## Urinary pH: not a good criterion to assess the risk of Calcium Oxalate uroliths

Although in epidemiological studies, acidifying diets have been identified as risk factors for the development of Calcium Oxalate (CaOx) in cats, this prospective study supports that urinary pH has no impact on CaOx relative supersaturation (RSS\*) for pH ranges encountered in feline diets.

This study examined the effects of 4 similar diets inducing different acidic urinary pH (<6.5) on several urinary ions excreted (including calcium) and on CaOx RSS, in 13 healthy adult cats. Four feline maintenance dry diets were manufactured. They contained identical ingredients and nutrient content, except for sulfur and chloride: for gradual acidification, sodium bisulfate substituted sodium chloride with levels of 0%, 0.6%, 1.3% and 1.9% as fed, respectively. All cats tested the 4 diets in a sequential order. Each diet was fed to weight maintenance for 9 days, followed by 5 days of individual urine collection. Urine samples were pooled and kept at 4°C until laboratory analysis. Urinary volume was recorded for each cat. Urinary pH, 24-hour ion excretions (µmol/kg/day), and CaOx RSS were calculated.

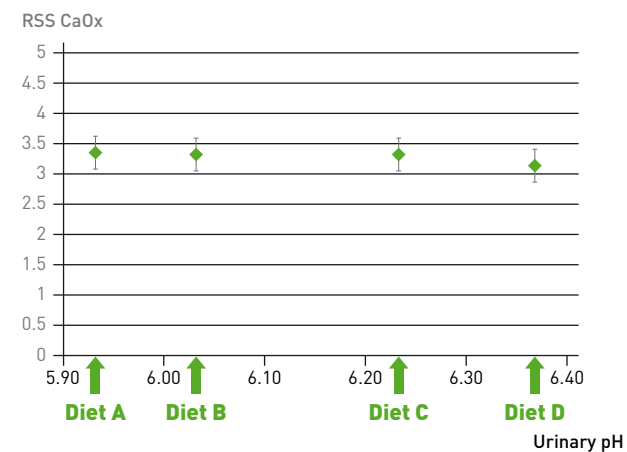
Urinary volumes (mL/kg/day) produced during the collection period did not differ between diets (p=0.45). Diets A, B, C and D induced urinary pH of 5.93±0.03, 6.02±0.03, 6.23±0.03 and 6.37±0.04, respectively. A significant effect of diet on urinary excretions was found on the following ions: calcium, ammonium, citrate, sulfate and oxalate. Excretions of calcium, ammonium and sulfate were higher as the diet induced a lower pH (p<0.0001), and inversely, excretions of oxalate and citrate were lower with diets inducing lower pH (p<0.001). However, CaOx RSS was not affected by urinary pH (p=0.63).

Despite higher calcium excretion, CaOx RSS remained unaffected by urinary pH. This may be explained by the fact that RSS is a multifactorial calculation that includes additional urinary parameters. Lower oxalate excretion seen with diets inducing lower urinary pH, may be one hypothesis. This study reinforces the fact that a diet can be acidified to prevent and dissolve struvite uroliths, without adverse effects on the risk of CaOx formation.

Queau Y, Van Hoek I, Feugier A, Le Verger L, Soulard Y, Biourge V. Urinary pH affects urinary Calcium excretion but not calcium oxalate relative supersaturation in healthy cats. J Vet Intern Med 2013;27:738-739 (abstract).

\*RSS: Relative Supersaturation (RSS) is a method to assess the risks of forming urinary crystals based on the level of saturation of poorly soluble salts such as calcium oxalate or struvite. It is the most widely used method in humans and has been validated for dog and cat urine.

«Least Square» Means (+/- SEM) of CaOx relative supersaturation in 13 cats fed 4 similar diets inducing different urinary pH



## Feline idiopathic cystitis: a blood test for diagnosis?

A diagnostic test for Feline Idiopathic Cystitis (FIC) is lacking. This study evaluated the use of a test based on infrared microspectroscopy of blood collected using blood spot cards.

FIC is a urinary tract disorder in cats linked to increased stress levels. The diagnosis of FIC currently remains one of exclusion, relying on combinations of clinical signs, urine analysis and culture, abdominal imaging to rule out urolithiasis, and cystoscopy. This study evaluated the diagnostic use of IRMS (InfraRed MicroSpectroscopy) of blood collected from 42 cats and applied to blood spot cards.

Twenty-two healthy adult cats (median age 7.0 years), and 20 cats with FIC (median age 9.9 years) from the same colony were included. Blood was collected, applied to bloodspot card, left to dry and subjected to IRMS. Absorbance spectra were obtained and analysed with a pattern recognition method, based on Principal Component Analysis.

This method revealed significant difference between groups (p<0.05). The separation was predominantly attributable to the band at 1546 cm<sup>-1</sup>, which is related to metabolites of tryptophan.

This study confirmed differentiation between healthy cats and cats with FIC, based on specific infrared spectra. Further evaluation is needed to understand the potential role of tryptophan metabolites in cats with Feline Idiopathic Cystitis.



van Hoek I, Rodriguez-Saona L, Biourge V, Buffington CA. a diagnostic test for feline idiopathic cystitis based on infrared microspectroscopy. J Vet Intern Med 2013;27: 741(abstract)

van Hoek I, Rodriguez-Saona L, Plans M, Buffington CA. A dried blood-spot based test for diagnosis of feline idiopathic cystitis based on infrared microspectroscopy and pattern recognition analysis. Proceedings of 16th International Conference on Near Infrared Spectroscopy, La Grande Motte (France) June 2-7 2013

Differences in spectra between Healthy and FIC cats (A) and discriminating power of transformed IRMS (B) spectra

