

Gastrointestinal Hemodynamics in Dogs with Nonfood Induced Atopic Dermatitis

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Background: Canine atopic dermatitis can be a result of exposure to aeroallergens or trophallergens. Hemodynamic alterations occur in dogs with food hypersensitivity.

Hypothesis/Objectives: To evaluate if hemodynamic alterations occur in dogs with NFICAD with lowered resistance to diastolic flow at fasting, after feeding, or both.

Animals: Ten healthy dogs and 22 dogs with NFICAD were included from the hospital population.

Methods: Blinded prospective study. Peak systolic velocity (PSV), end diastolic velocity (EDV), mean velocity (MV), pulsatility index (PI), resistive index (RI) and PSV/EDV ratio were measured at fasting for both arteries (cranial mesenteric artery [CMA], celiac artery [CA]) and at 40 minutes after feeding in CMA and at 60 minutes in CA. The results were analyzed statistically with a mixed model.

Results: There was no difference detected between groups of dogs for any variable except EDV during fasting ($P = .01$).

Conclusions and Clinical Importance: There is no decrease in resistance in NFICAD to diastolic flow. This observation could be explained by the absence intestinal inflammation in NFICAD.

Key words: Atopy; Intestinal permeability; Skin; Vascular flow.

Canine atopic dermatitis is a genetically predisposed inflammatory and pruritic allergic skin disease with characteristic clinical features associated with IgE antibodies.¹ Controversies exist regarding the routes of allergen exposures in canine atopic dermatitis.² The inhalation route has long been highlighted as the major route of sensitization of dogs.³ More recently, the epicutaneous route was demonstrated experimentally by patch test and theoretically by the observation of many defects of the skin barrier in atopic dogs.^{4,5} The oral route is a pathway of sensitization leading to the expression of clinical signs in nonfood allergic dogs that could be associated with gastrointestinal hemodynamics changes.⁶ Splanchnic blood flow could be used as a biomarker of these local changes as shown in other diseases involving the intestinal tract such as food allergy, inflammatory bowel diseases.⁷

Currently, there are data on the parameters of blood flow in normal dogs and dogs with gastrointestinal disorders, including food allergy.⁷⁻⁹ As in humans, Doppler ultrasound of the cranial mesenteric artery (CMA) and the celiac artery (CA) in food allergic dogs with intestinal signs showed a significant decrease in Doppler hemodynamic parameters (resistive index [RI] and pulsatility index [PI]) between a maintenance diet and one containing an allergen.⁷ These effects could be a local acute inflammatory response with increased and

Abbreviations:

PSV	peak systolic velocity
EDV	end diastolic velocity
MV	mean velocity
PI	pulsatility index
RI	resistive index
CMA	cranial mesenteric artery
CA	celiac artery
NFICAD	nonfood induced atopic dermatitis
FICAD	food induced atopic dermatitis

prolonged vasodilatation because of local vasodilatory mediators.

Only one pilot study investigated modifications in nonfood allergic pruritic dogs.¹⁰ In this pilot study, the low number of dogs included (8 nonfood allergic pruritic dogs) and the choice of the control group (5 food allergic dogs and not normal dogs) limit the conclusions drawn from the results.

The aim of this study was to identify whether dogs with nonfood induced atopic dermatitis show changes in gastrointestinal hemodynamic responses in comparison with normal dogs before or after feeding. Therefore, spectral waveform analysis of the celiac artery and cranial mesenteric artery was assessed. We hypothesized that dogs with nonfood induced atopic dermatitis would have a lowered resistance to diastolic flow compared with normal dogs before and/or after feeding. Doppler ultrasound of gastrointestinal vessels could represent a noninvasive tool for the assessment and monitoring of various treatments or dietary therapy in atopic dermatitis in dogs.

Materials and Methods

Study Populations

This blinded prospective study was done in the dermatology clinic of the veterinary teaching hospital of Oniris, Nantes-Atlantic National College of Veterinary Medicine, Food Science and Engi-

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Submitted July 5, 2012; Revised January 16, 2013; Accepted February 7, 2013.

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10.1111/jvim.12072

neering (Nantes, France) between January and June 2011. The study protocol was reviewed and approved by the animal care and use committee of Oniris and owner consent was obtained for each case.

Inclusion of dogs with nonfood-induced canine atopic dermatitis (NFICAD group) was based on the fulfillment of the Prelaud's criteria (at least 3 criteria), a negative response to a rigorous limited-allergen diet of 6–8 weeks and positive responses to intradermal testing for aeroallergens, and the exclusion of other causes of pruritic dermatitis (parasitic or infectious diseases). At the time of inclusion, dogs could not have received either glucocorticoids, ciclosporin or antihistamines for at least 1 month, or antibiotics and/or antifungal medications for at least 9 days. All dogs were negative for intestinal parasites in fecal examination.

Control dogs (Control group) were individually owned healthy dogs with normal clinical examination, normal biological parameters (complete blood count, glucose, urea, creatinine, alkaline phosphatase, alanine transferase, albumin, total protein) without signs of gastrointestinal disease and without history of chronic or recurrent gastrointestinal disease. These dogs were also without any history or signs of pruritic dermatitis and negative for parasitic elements in fecal examination.

For the ultrasound examination the food fed was the dog's normal diet with a standardized size of the half of the daily volume. The quality and the composition of the diet were variable between dogs and not precisely identified.

Ultrasound Examination

Dogs were fasted overnight (at least 12 hours) and examined the next morning. They were not exercised before examination to avoid exercise-induced changes. They did not receive any sedative or anesthetic. Dogs were placed in dorsal recumbency, the hair was clipped on the ventral abdomen, and the skin was cleaned with 70% ethanol. Coupling gel was applied and the CA and CMA were imaged. Measurements were made at fasting and 40 minutes after ingestion of food for CMA and after 60 minutes for CA.

The timings of the arteries' measurements were based on the literature.⁷ In dogs with food hypersensitivity, the maximal difference of RI and PI was at 20 and 40 minutes for CMA and at 60 minutes for CA between regular and challenge diet.⁷ Thus, on the assumption that NFICAD could react as food hypersensitive dogs, examination was started at 40 minutes for the CMA, and the CA was examined in second, at 60 minutes.

Each dog was examined by 1 sonographer measuring blinded (control versus NFICAD group) the parameters by the same method modified from Riesen.⁹ Two sonographers participated to the protocol. The examinations were performed with an ultrasound unit (Esaote, Mylab 70) with a 9-MHz micro-convex transducer. Pulsed spectral Doppler was used to trace the flow in each vessel with a sampling gate size of 2 mm. The vessels were identified by 2-dimensional gray-scale and color Doppler ultrasound. Doppler spectral waveform tracings of flow in the CA and CMA were made close to their origins to limit turbulence and vortex shedding. The angle between the ultrasound beam and direction of the vessel was $< 60^\circ$. The peak systolic velocity (PSV), the end-diastolic velocity (EDV), and the mean velocity (MV) of 5 waveforms from each vessel (obtained from 2 or 3 recordings) at each time point were calculated by the ultrasound computer. The resistive index ($RI = (PSV - EDV)/PSV$) and the pulsatility index ($PI = (PSV - EDV)/MV$) were calculated and the mean of 5 waveforms was used for further analysis. The percentage deviation from to for RI and PI was calculated for each time point.

Statistical Analysis

XLSTAT 2011[®] software (Addinsoft, Paris) was used. The results were analyzed statistically with a mixed model. The different measurements (PSV, EDV, MV, EDV/PSV, PI, RI) were integrated in a model, including several factors: the age, the sex, the operator, the timing (preprandial versus postprandial), the group (NFICAD versus control). Each measurement was compared by pairwise with 1 explicative factor. Significant difference was considered as $P < .05$.

Results

Twenty-two dogs with nonfood induced atopic dermatitis and 10 control dogs were included in the study.

There were 14 males (5 neutered), 8 females (2 spayed) in the NFICAD group and 6 males (2 neutered), 4 females (2 spayed) in the control group. There was not significant difference of the sex ratio between both groups (P -value: .13).

Fourteen breeds (included mixed-breed) were recorded in the NFICAD: 6 West Highland White Terriers, 2 Boxers, 2 Labrador Retrievers, 2 German Shepherds, and 1 dog of each of the following breeds: Golden Retriever, Fox Terrier, Jack Russell Terrier, German Pointer, Dalmatian, Standard Poodle, Fauve of Brittany, Cavalier King Charles Spaniel, Shih Tzu, mixed breed. Nine breeds were recorded in the control group: 2 Dachshund Terriers and 1 dog of each of the following breed: Golden Retriever, Siberian Husky, Rottweiler, Yorkshire Terrier, Boxer, Great Dane Dog, Cocker Spaniel, mixed breed.

The mean age was 4.6 years for the NFICAD group and 4.3 years for the control group. There was not significant difference of the age repartition between both groups (P -value: .83).

The mean PSV, EDV, MV, PSV/EDV, PI, and RI values for the CA and CMA for each time point are shown in Table 1.

Only EDV after fasting was significantly affected (EDV, P -value: .01) (Table 1). The others parameters observed were not statistically different (Table 1).

The acceleration of EDV in the control group was the only statistical change observed (P -value: .03) (Table 1). In both group, a decrease in indices was observed for all measures except for the PI and RI of CA (Table 1). The decreases in RI and PI of CMA in both groups were statistically significant (RI CMA, P -value: .03, PI CMA, P -value: .02 in control group; RI CMA, P -value: .01, PI CMA, P -value: .02 in NFICAD) (Table 1).

The intersonographer results in function of the artery measured, the time point, the group, the age, and the sex were not statistically different (P -values not shown).

Discussion

Whereas the intestine is the main site of contact with allergens, our results suggest an absence of involvement of gastrointestinal hemodynamics in nonfood induced atopic dermatitis in dogs. No differences in

Table 1. Mean values ($\pm$ percentage deviation) for the peak systolic velocity (PSV), the end diastolic velocity (EDV), the mean velocity (MV), the ratio PSV/EDV, the resistive index (RI) and the pulsatility index (PI) of the celiac (CA) and cranial mesenteric arteries (CMA) both preprandial and postprandial with the *P*-values (mixed model) of the RI, the PI, the PSV, the EDV, the MV, the ratio PSV/EDV of the comparisons within each group (preprandial [PreP] versus postprandial [PostP]) and between groups (control group versus NFICAD group).

Variable	Group	Artery	Timing	Measurements ($\pm$ SD)	<i>P</i> -value PreP vs PostP	<i>P</i> -value NFICAD vs Control
PSV	NFICAD	CMA	PreP	140.3 ($\pm$ 44.0)	.87	.06
			PostP	142.6 ($\pm$ 44.5)		.20
		CA	PreP	164.8 ($\pm$ 59.4)	.69	.17
			PostP	150.7 ($\pm$ 47.8)		.2
	Control	CMA	PreP	104.7 ($\pm$ 23.4)	.40	
			PostP	118.7 ($\pm$ 39.0)		
		CA	PreP	130.6 ($\pm$ 47.7)	.81	
			PostP	125.0 ($\pm$ 43.0)		
EDV	NFICAD	CMA	PreP	23.9 ($\pm$ 7.7)	.07	.01
			PostP	29.4 ($\pm$ 10.1)		.07
		CA	PreP	31.3 ($\pm$ 12.9)	.78	.07
			PostP	28.6 ($\pm$ 11.7)		.74
	Control	CMA	PreP	15.7 ($\pm$ 4.9)	.03	
			PostP	22.1 ($\pm$ 5.6)		
		CA	PreP	22.2 ($\pm$ 5.7)	.31	
			PostP	26.9 ($\pm$ 11.4)		
MV	NFICAD	CMA	PreP	40.0 ($\pm$ 9.2)	.51	.17
			PostP	42.7 ($\pm$ 8.2)		.84
		CA	PreP	50.6 ($\pm$ 15.0)	.32	.95
			PostP	44.2 ($\pm$ 23.6)		.53
	Control	CMA	PreP	33.8 ($\pm$ 7.1)	.10	
			PostP	43.7 ($\pm$ 11.8)		
		CA	PreP	51.1 ($\pm$ 22.3)	.68	
			PostP	50.0 ($\pm$ 11.7)		
PSV/EDV	NFICAD	CMA	PreP	6.2 ($\pm$ 1.8)	.08	.24
			PostP	5.1 ($\pm$ 1.6)		.63
		CA	PreP	5.7 ($\pm$ 2.4)	.85	.75
			PostP	5.6 ($\pm$ 1.2)		.11
	Control	CMA	PreP	7.1 ($\pm$ 2.6)	.18	
			PostP	5.4 ($\pm$ 1.1)		
		CA	PreP	6.0 ($\pm$ 2.2)	.18	
			PostP	4.8 ($\pm$ 0.8)		
PI	NFICAD	CMA	PreP	2.69 ($\pm$ 0.47)	.02	.77
			PostP	2.31 ($\pm$ 0.56)		.86
		CA	PreP	2.47 ($\pm$ 0.46)	.78	.12
			PostP	2.44 ($\pm$ 0.34)		.33
	Control	CMA	PreP	2.75 ($\pm$ 0.49)	.02	
			PostP	2.27 ($\pm$ 0.29)		
		CA	PreP	2.18 ($\pm$ 0.30)	.62	
			PostP	2.29 ($\pm$ 0.34)		
RI	NFICAD	CMA	PreP	0.83 ($\pm$ 0.05)	.01	.54
			PostP	0.79 ($\pm$ 0.05)		.38
		CA	PreP	0.82 ($\pm$ 0.04)	.82	.32
			PostP	0.81 ($\pm$ 0.04)		.65
	Control	CMA	PreP	0.84 ($\pm$ 0.04)	.03	
			PostP	0.81 ($\pm$ 0.03)		
		CA	PreP	0.80 ($\pm$ 0.03)	.60	
			PostP	0.81 ($\pm$ 0.02)		

Bold values are statistically significant.

CAM and CA PI, RI and PSV/EDV were observed between the NFICAD group and the control group.

This absence of changes of gastrointestinal hemodynamics could be explained either by a moderate, or an absence of, local release of proinflammatory or vasoactive mediators. This hypothesis is supported by

a recent publication in which dogs with food induced canine atopic dermatitis (FICAD) did not have any changes detected in mucosal inflammatory cells profiles in the duodenum (PCR assay and immunohistochemical staining).¹¹ Thus, the intestinal mucosa in NFICAD, as in FICAD, does not seem to be the primary

site of inflammatory cell activation. No histopathological data from the intestine of dogs with NFICAD are available in the literature to support or refute the presence of local inflammation.

Moreover, this lack of significant differences in hemodynamic changes between the 2 groups in this report could be because of the location of allergic inflammation in the intestine. Indeed, the celiac artery supplies blood flow to the stomach, liver and spleen while the cranial mesenteric artery supplies the entire small intestine, proximal portions of the colon and the pancreas.¹² A release of vasoactive mediators in the distal colon could thus explain the lack of observing hemodynamic changes in the blood flow measurements of the CMA and the CA. In fact, this portion of intestine is supplied by the caudal mesenteric artery.¹² However, the involvement of the distal colon seems unlikely because the duodenum is the initial place of digestion and absorption of food constituents including allergens.

The major limitations of this study could be the population used, the meal, and the timing.

The route of sensitization in the natural allergic dogs used in this study was not identified (inhalant, transcutaneous, digestive). These dogs were sensitized to a variety of allergens (dust mites, pollens). Thus, the digestive way of sensitization was perhaps not involved. An allergenic challenge by suspected allergens in the diet of each dog could have yielded different results.

The composition of the meal may influence the duration of the blood flow changes during digestion.¹³ After a fatty diet, there is a tendency to have the greatest and longest changes. Carbohydrate-rich diets, on the contrary, have a tendency to have the fastest and shortest effect.¹³ Proteins provoke a decrease of PI and RI of CMA and CA in ranges between the 2 other diets.¹³ The heterogeneity of diet composition of the dogs could mask a difference between both groups. However, the diets of the dogs were not modified in order to limit interference because of changes in diet (inflammation, mucosal absorptive capacity, osmolarity, electrolyte concentrations) and to not influence the clinical signs of NFICAD animals.^{14,15} This limitation is minimized by data from a follow-up study (results not shown). Both groups of dogs were reassessed after being fed solely with 1 commercial diet for 8 weeks and no difference was noted between the NFICAD and the control groups.

The 3rd limitation could be the choice of the time points. Our choice was based on the study made by Kircher on food hypersensitivity with gastrointestinal signs. In this study, the maximal difference of RI and PI was at 20 and 40 minutes for CMA and at 60 minutes for CA between regular and challenge diet. In our study, the measurements at 40 minutes for CMA and at 60 minutes for CA were based on the assumption that NFICAD could react as food hypersensitive dogs (gastrointestinal signs), but these time points were perhaps inadequate for dogs with atopic dermatitis. Measurements with more time points should be done to respond to these interrogations.

Despite the absence of difference of the indices (RI, PI) between groups, the EDV of CMA in NFICAD group was modified. However, currently, for assessing blood flow in small abdominal arteries, the indices are preferred to the velocity measurements because their use allow to reduce errors associated with angle between the blood stream of the vessel and the Doppler beam and size of the vessel.¹⁶ Moreover, the ratio PSV/EDV without any difference between both groups allow to definitively rule out the possible decreased resistance at fasting of dogs in NFICAD.

In conclusion, it appears that the 2 vascular beds react in NFICAD dogs as in normal dogs with no difference of pulsatility index and resistive index. The local intestinal hemodynamics and the conditions of splanchnic blood flow do not seem modified in dogs with nonfood induced canine atopic dermatitis. In the conditions of this study, dogs with nonfood induced atopic dermatitis do not have a lowered resistance to diastolic flow compared with normal dogs at fasting and after feeding. This observation could indirectly rule out an intestinal inflammation in NFICAD.

Acknowledgment

Conflict of Interest: Authors disclose no conflict of interest.

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