# B-mode and contrast-enhanced ultrasonography for intestinal assessment in dogs: A review

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**ABSTRACT**
Ultrasound (USG) is a valuable diagnostic tool for evaluating the gastrointestinal tract of small animals, providing noninvasive and dynamic information. This review discusses intestinal evaluation in dogs using B-mode ultrasound and advanced contrast-enhanced ultrasound (CEUS) with microbubbles. B-mode USG allows the examination of the thickness and stratification of the intestinal wall, motility, and adjacent structures, such as lymph nodes and peritoneum to be examined. Reference values for intestinal wall thickness in dogs are based on body weight. The duodenum, jejunum, and ileum can be differentiated sonographically by location wall stratification, and relationship to other intestinal segments. Intestinal motility is assessed by counting peristaltic waves, and abnormalities indicate various diseases. CEUS uses microbubble-based contrast agents, such as sulfur hexafluoride, which remain stable in the vascular system and are eliminated by the lungs. This technique improves the perception of ultrasound echoes, making it possible to quantify tissue perfusion using parameters such as entry time, peak enhancement, and exit time. In human medicine, CEUS is used to assess intestinal perfusion in conditions such as Crohn's disease and ischemia. In veterinary medicine, this technique has been used to assess duodenal perfusion in healthy dogs and those with chronic enteropathies or alimentary lymphoma. Although CEUS shows potential in differentiating dogs with chronic enteropathies from healthy controls, more research is needed to standardize the methodology, establish reference values, and define clinical applications in various canine intestinal diseases. The combination of B-mode USG and CEUS provides a noninvasive assessment of intestinal morphology and function, contributing to the understanding of the pathophysiology of enteropathies in dogs.
**Keywords:** Canine, Contrast, Ultrasound, Intestinal wall thickness, Microbubbles.

## Introduction

Intestinal ultrasound evaluation provides relevant information on topography, wall thickness, layer stratification, echogenicity, motility, luminal content, and characteristics of adjacent structures such as lymph nodes, peritoneum, and other organs attached to the gastrointestinal tract (GIT) (Larson and Biller, 2009). USG can be helpful in cases where patients present symptoms of gastrointestinal disease, but clinical confrontation of the findings is always required (Larson and Biller, 2009; Penninck and D'anjou, 2015). However, there is a description of the limitations of USG in terms of its potential to define the diagnosis in some situations, especially when the examination is carried out based exclusively on clinical signs such as vomiting and diarrhea (Leib *et al.*, 2010; Leib *et al.*, 2012; Mapletoft *et al.*, 2018).

Diffuse or focal thickening, loss of parietal stratification, and increased or decreased echogenicity are findings on USG in dogs with GIT diseases (Penninck and D'anjou, 2015). Contrast-enhanced ultrasound (CEUS) is a sensitive method for assessing the vascular perfusion of organs (Nyman *et al.*, 2005). This innovative and noninvasive test can be useful for the intestinal evaluation of dogs (Cordella *et al.*, 2021). In medicine, CEUS is used to assess intestinal vascularization in inflammatory (Lassau *et al.*, 2006) and neoplastic diseases (Serra *et al.*, 2007). In Crohn's disease, CEUS revealed an increase in the enhancement pattern of the intestinal vessels, which was correlated with the clinical activity of the disease. Thus, the test is a tool for monitoring patients and has prognostic potential (Robotti *et al.*, 2004).

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In veterinary medicine, only a few studies have assessed CEUS for intestinal assessment. More recent research has been based on evaluating intestinal perfusion in dogs (Nisa *et al.*, 2018) and cats (Diana *et al.*, 2011), studying intestinal ischemia in cats (Linta *et al.*, 2020), canine inflammatory bowel disease, differentiating canine chronic enteropathy from alimentary lymphoma (Linta *et al.*, 2021), and a case report of histiocytic colitis in a dog (Cordella *et al.*, 2021).

Ultrasound modalities integrate and provide important information about the intestine (Larson and Biller, 2009; Heilmann, 2015). Given the scarcity of studies, the aim of this literature review was to address the intestinal evaluation of dogs using B-mode USG and the advanced CEUS technique.

**B-*mode ultrasound for the intestinal assessment of dogs***
USG is an important diagnostic imaging modality for assessing the gastrointestinal tract of small animals, as it is a noninvasive test that provides dynamic information and can be recommended for acute and chronic enteropathies (Haber *et al.*, 2002; Bhavani *et al.*, 2021). It is recommended for dogs showing clinical signs of gastrointestinal disease (Delaney *et al.*, 2003; Seiler *et al.*, 2022). However, Leib *et al.* (2010) found that in dogs with chronic vomiting, the diagnostic usefulness of USG was greater in older animals, in cases where there was a greater number of vomiting episodes and when there was weight loss. In dogs with chronic diarrhea, the usefulness of this imaging test was greater in cases of weight loss, alterations in abdominal palpation, and suspected GIT neoplasia (Leib *et al.*, 2012). In another study, ultrasound was considered counterproductive in some situations where only clinical signs justified its indication (Mapletoft *et al.*, 2018). We believe that USG has some limitations and that its use should be based on not only clinical manifestation but also anamnesis, the evolution of the presenting symptom, and the results of laboratory tests. In the intestinal evaluation of dogs, USG has been used in cases of gastrointestinal foreign bodies (Dandrieux, 2016; Bhavani *et al.*, 2021) and to suggest a differential between inflammation and neoplastic processes (Simeoni *et al.*, 2020). The evaluation focused on the intestine of dogs provides information on the thickness of the organ wall layer, motility, parietal stratification, and peripheral structures, such as lymph nodes and peritoneum (Larson and Biller, 2009).

Gladwin *et al.* (2014) determined reference values for the thickness of the layers of the duodenum, jejunum, and colon in dogs with the aim of providing support in the management of disorders that alter the thickness and stratification of these intestinal layers. This evaluation was the subject of a study that correlated the ultrasound intestinal layer of canine cadavers with histological aspects, except for the serous layer. There was no correlation between USG and histology; however, there was a hyperechoic line in the muscle, referring to the interface of the circular and longitudinal fibers (Le Roux *et al.*, 2016).

Abdominal USG provides dynamic information that reflects the physiology and anatomy of the structures assessed (Malancus and Malancus, 2017). The method was performed in a systematic evaluation pattern with an organized analysis of the components (Nyland *et al.*, 2015). Knowledge of normal and altered ultrasound features is advantageous in the diagnosis of GIT diseases (Larson and Biller, 2009).

***Basic principles of bowel ultrasound***
To perform the examination, the patient must be properly positioned in relation to the operator, USG equipment, and transducer (Carvalho, 2004). These considerations are related to obtaining consistent images and the comfort of the animal and examiner. The projected image is classified in the sagittal plane into ventral, cranial dorsal, and caudal, while in the transverse plane, the images formed are divided into medial and lateral (Nautrup, 2000).

For evaluating the GIT, high-frequency transducers (7.5–10 MHz) improve image resolution and allow for greater detail, as well as the measurement of the layers (Penninck and D'anjou, 2015). In addition to the frequency mentioned above, linear transducers are the most recommended. For a more detailed analysis, the scans should be in the longitudinal and transverse planes (Penninck and D'anjou, 2015).

Preparing the animal for the examination is of fundamental importance and reflects on the quality of the images generated. For ultrasound examinations focused on intestinal assessment, it is necessary to perform a wide trichotomy of the ventral region of the abdomen, from the seventh intercostal space to the caudal inguinal region. Because the ultrasound beam does not penetrate the air trapped between the hairs, it generates reverberation artifacts that impair the interpretation of the findings. The close contact between the skin and the transducer facilitates image formation without interference (King, 2006).

To remove air and fat from the surface of the skin, which could potentially meet the transducer, in addition to the trichotomy technique, acoustic gel, and 70% isopropyl alcohol should be used before using any other material (Carvalho, 2004). The acoustic gel should be applied abundantly to the skin to favor contact and prevent the formation of reverberation artifacts (Nyland *et al.* 2015).

Ideally, an 8-hour food fast is recommended in dogs without water restriction to help reduce gas inside the GIT, as this gas in the GIT promotes the formation of elements that hinder the assessment of abdominal structures (Penninck and D'anjou, 2015). The order of assessment varies, but in general, for intestinal assessment, the duodenum is assessed halfway through the procedure, while the general assessment of other intestinal segments (jejunum, ileum, cecum, ileoceocolic valve, ascending, descending, and

transverse colon) along with lymph nodes is carried out at the end of the USG (Nautrup, 2000).

The sequence is individual to the practitioner, but all structures should be categorically assessed (Nyland *et al.*, 2015). The thickness of the intestinal segments in dogs can be measured using cursors in the USG software at the outer edge of the serous layer and the inner portion of the mucosal layer (Riedesel, 2015). However, it is known that there is a positive correlation between the measurement of the intestinal wall and the dog's body weight, so reference values are divided based on weight ranges (Gladwin *et al.*, 2014). The reference values for the total measurements of the intestinal walls of dogs are presented in Table 1 and stratified in Table 2.

### Ultrasound bowel anatomy

The normal topography of the duodenum is in the right cranial abdominal quadrant starting between the last ribs and distal along the right abdominal wall, and the ultrasound evaluation of the intestinal segments is done by moving the transducer from right to left and the other way around, then cranial and caudal (Agut, 2009). These maneuvers were performed to obtain images of the intestines in all sections, including sagittal transverse, and oblique images (Penninck and D'anjou, 2015).

The duodenum (Fig. 1B) of a healthy dog can be differentiated from the jejunum (Fig. 1C) by USG because of its more superficial topography in the right epigastric region and because it is cranially close to the stomach (Fig. 1A). In addition, it is possible to observe the duodenal papilla as an anatomical reference if the image resolution is of high quality (Riedesel, 2015).

The ileum (Fig. 1D) was anatomically located in the craniomedial abdomen to the right of the ascending colon and cecum (Goggin *et al.*, 2000). This intestinal segment can be visualized by USG in the right abdominal quadrant and is the most robust and bright submucosal layer in the image (Prestes *et al.*, 2019). The jejunum can be found and evaluated by the US in practically every abdominal cavity (Goggin *et al.*, 2000). Although the jejunal loops can be found and

**Table 1.** Reference values for the wall thickness of intestinal segments in dogs divided by region and body weight.

| Species | Weight | Duodenum | Jejunum | Ileum | Cecum/colon |
|---------|--------|----------|---------|-------|-------------|
| Canine | < 15 Kg | 0.38 cm | 0.30 cm | 0.30 cm | 0.15 cm |
| | 15-30 Kg | 0.41 cm | 0.35 cm | 0.35 cm | |
| | > 30 Kg | 0.44 cm | 0.38 cm | 0.38 cm | |

Source: Adapted from Larson and Biller, 2009.

**Table 2.** Reference values for the thickness of the intestinal wall layers (in cm) of healthy dogs according to body weight with segmentation of values with variations in measurements according to each layer (mucosa, submucosa, muscle and serosa).

| Segment | Layer | <15 Kg | 15–30 Kg | >30 Kg |
|---------|-------|--------|----------|--------|
| Duodenum | Mucosa | 0.24 (0.15–0.35) cm | 0.26 (0.15–0.37) cm | 0.28 (0.2–0.39) cm |
| | Submucosa | 0.06 (0.03–0.08) cm | 0.06 (0.03–0.1) cm | 0.06 (0.03–0.1) cm |
| | Muscular | 0.05 (0.02–0.08) cm | 0.05 (0.03–0.08) cm | 0.06 (0.02–0.09) cm |
| | Serous | 0.04 (0.02–0.06) cm | 0.04 (0.03–0.06) cm | 0.04 (0.02–0.07) cm |
| | Total | 0.38 (0.29–0.47) cm | 0.41 (0.3–0.55) cm | 0.44 (0.31–0.57) cm |
| Jejunum | Mucosa | 0.18 (0.12–0.26) cm | 0.2 (0.15–0.32) cm | 0.22 (0.11–0.32) cm |
| | Submucosa | 0.05 (0.03–0.09) cm | 0.06 (0.03–0.1) cm | 0.05 (0.03–0.08) cm |
| | Muscular | 0.04 (0.02–0.07) cm | 0.05 (0.03–0.08) cm | 0.05 (0.03–0.09) cm |
| | Serous | 0.04 (0.02–0.06) cm | 0.04 (0.03–0.06) cm | 0.04 (0.03–0.06) cm |
| | Total | 0.3 (0.22–0.41) cm | 0.35 (0.24–0.48) cm | 0.38 (0.27–0.47) cm |
| Colons | Mucosa | 0.04 (0.02–0.06) cm | 0.04 (0.02–0.05) cm | 0.05 (0.03–0.07) cm |
| | Submucosa | 0.04 (0.02–0.06) cm | 0.03 (0.02–0.04) cm | 0.04 (0.02–0.05) cm |
| | Muscular | 0.04 (0.02–0.07) cm | 0.03 (0.02–0.05) cm | 0.04 (0.02–0.07) cm |
| | Serous | 0.04 (0.02–0.05) cm | 0.04 (0.02–0.05) cm | 0.04 (0.02–0.05) cm |
| | Total | 0.15 (0.12–0.16) cm | 0.14 (0.11–0.19) cm | 0.16 (0.11–0.26) cm |

Source: Adapted from Gladwin *et al*. (2014).

form an image during the evaluation of the other structures during the complete examination, systematic and detailed scanning of the jejunal segments is recommended (Larson and Biller, 2009).

There is difficulty in scanning the colon due to the large amount of gas that is naturally present, plus the fecal material that is stored is an additive factor in the difficulty of evaluation (Larson and Biller, 2009). The transverse colon lies immediately caudally to the stomach and duodenum, while the descending branch can be found caudally and dorsally to the urinary bladder (Goggin *et al.*, 2000). The segments of the colon are shown in Figure 2.

When the colon is seen to be distended on ultrasound, it can be mistakenly characterized as a full uterus. However, the differentiation between the abdominal organs and the intestines on ultrasound, in addition to differential ultrasound diagnosis of diseases, can be obtained by checking intestinal peristalsis, the pattern of stratification of layers, and the topography of the intestine and its adjacencies (Drost, 2015). In dogs, the segments of the colon are often visualized on USG with gas or fecal distension and have a thinner wall than the other parts of the intestine in dogs (Penninck and D'anjou, 2015).

The duodenum, jejunum, and ileum can be characterized on USG and differentiated based on some image findings, topography, mural stratification, and syntony with other intestinal segments (Gaschen, 2011). This information is detailed in Table 3.

On USG, the five parietal layers can be assessed (Fig. 3), with an interface between the intestinal lumen and the mucosa that is hyperechoic and forms a bright, central line on the image. The mucosal layer is associated with this and is hypoechogenic, followed by the hyperechogenic submucosa. The thin muscular
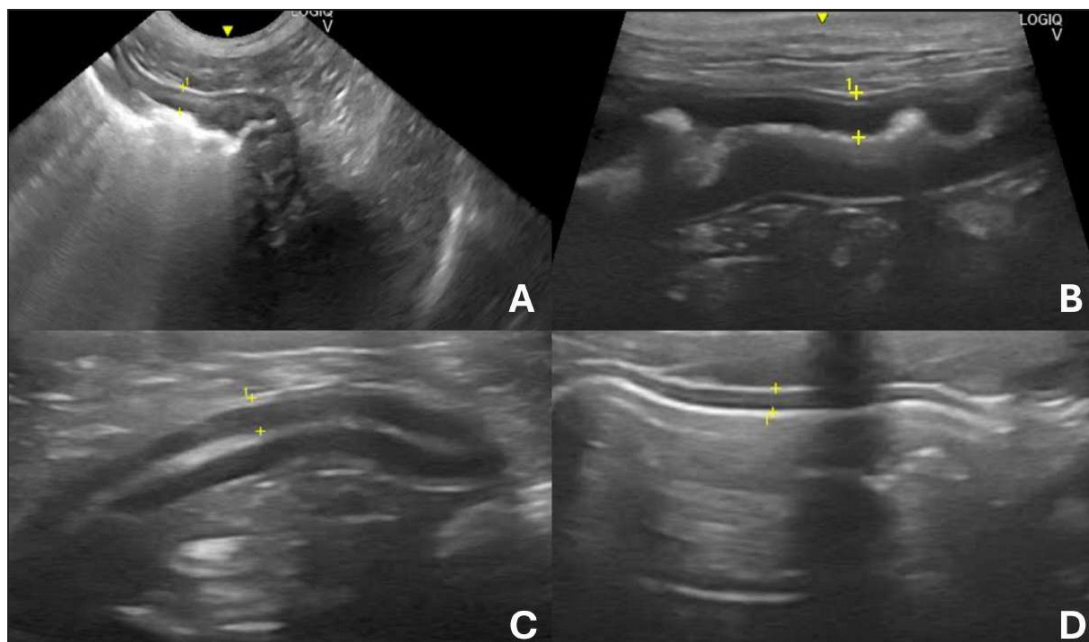


**Fig. 1.** Ultrasound images of the gastrointestinal tract of healthy dogs. A: gastric body, B: duodenum, C: jejunum, D: ileum. All segments with wall thickness were measured using a cursor in the ultrasound machine software. 8 MHz microconvex transducer.
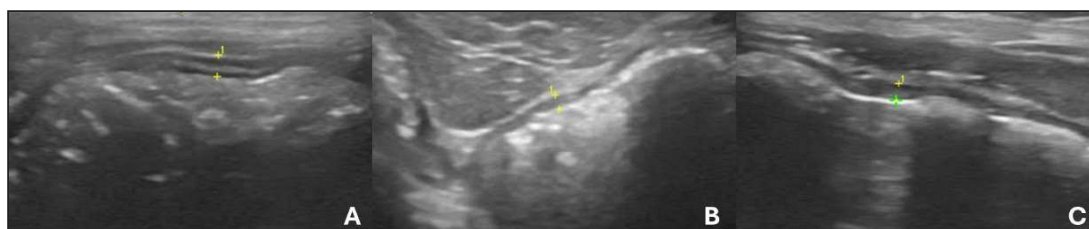


**Fig. 2.** Ultrasound images of the colon in healthy dogs. A, ascending colon; B, transverse colon; C, descending colon. All segments with wall thickness were measured using the ultrasound machine's software cursor. An 8-MHz microconvex transducer was used, and longitudinal section images.

**Table 3.** Intestinal ultrasound properties of dogs separated by region, anatomical location and specific characteristics.

| Region | Anatomical localization | Specific characteristics |
|---|---|---|
| Duodenum | Right epigastric | Peyer's patches on the antimesenteric border |
|  |  | Major and minor duodenal papilla |
| Jejunum | Mesogastric and hypogastric | - |
| Ileum | Right mesogastric, medial to duodenum | Ileocolic junction with thicker submucosa |
|  |  | Rosette appearance in cross-section |

Source: Adapted from Gaschen (2011).
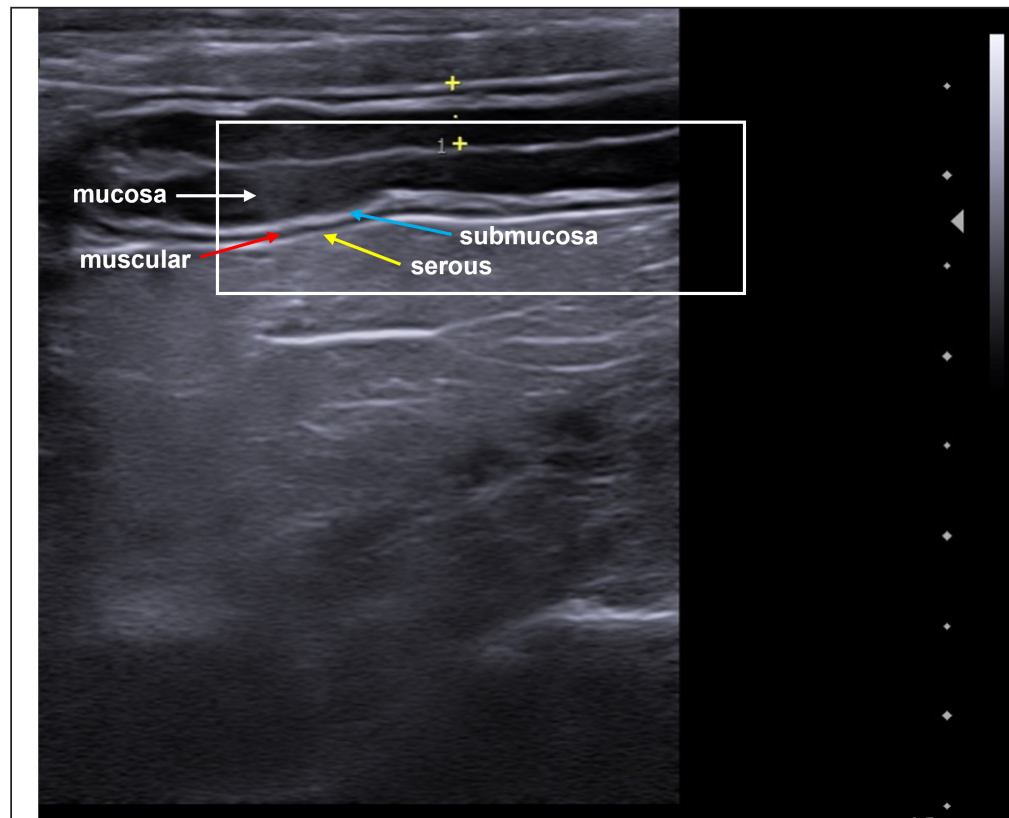


**Fig. 3.** Ultrasound image of the intestinal segments of a dog. The image shows transverse and longitudinal sections. The stratified layers are the mucosal layer (white arrow), submucosal layer (blue arrow), muscular layer (red arrow), and serosal layer (yellow arrow).

layer is hypoechogenic, and the outermost layer is the serosa, which is hyperechogenic (Goggin *et al.*, 2000; Agut, 2009; Larson and Biller, 2009).

The content in the intestines, as well as its quantity, is directly related to the ultrasound appearance, and when there is little intestinal filling, it is possible to observe a mucous pattern with a hyperechogenic nucleus surrounded by a hypoechogenic halo of the intestinal wall (Penninck and Nyland, 1990; Agut, 2009). The nucleus formed generally represents the mucoid content and air bubbles that are trapped at the interface between the lumen and mucosa. However, when the intestine is filled with fluid, an anechoic area can be observed

between the intestinal walls (Agut, 2009). Gaseous filling of the intestinal loops leads to the formation of a highly echogenic interface with posterior acoustic shadowing (Penninck and Nyland, 1990).

***Ultrasound evaluation of intestinal motility***
Intestinal motility can be assessed by counting peristaltic waves, and the expected contractions for the intestines are 3–5 movements per minute (Penninck and Nyland, 1990). This assessment can be performed using USG in different segments of the GIT (Riedesel, 2015).

Inflammatory, infectious, and neoplastic intestinal diseases can cause functional alterations with

compromised motility, and the ileum is the segment most frequently affected. The dysmotility that occurs in the adynamic ileus must be differentiated from mechanical obstructions, and USG is essential in the differential diagnosis, since the sonographic characteristics of the functional condition are mild to moderate, generalized dilation, predominantly liquid filling, reduced or absent motility with movement of the intraluminal intestinal contents (Penninck and D'anjou, 2015).

In the case of altered intestinal motility due to mechanical causes, USG can show dilatation of varying proportions in the intestinal portions, but most commonly, there is dilatation cranial to the obstructive point or stenosis, with increased peristalsis in the region, moderate to severe distension of the intestine, reduced diameter, and motility in the section caudal to the point of obstruction. In these cases, the foreign body can usually be visualized by a hyperechoic surface that promotes posterior acoustic shadowing (Gaschen, 2011).

Intestinal hypermotility can be detected using ultrasound and occurs secondary to the presence of an obstructive or non-obstructive foreign body, dietary changes, intestinal parasitism, enteritis, and previous intestinal surgery. This increased mobility can lead to intussusception (Oliveir , 2019).

Intestinal motility was assessed using USG with the influence of fasting in healthy dogs, and it was found in this study that the test is effective in assessing the motility of the GIT and that food restriction of 12–24 hours led to a reduction in the contractility of the stomach, duodenum, jejunum, and ileum. However, there was a need for further studies with longer fasting times postoperatively and in dogs with gastrointestinal diseases (Sanderson *et al.*, 2017).

In addition to B-mode USG, the use of pulsed Doppler ultrasound makes it possible to graphically observe the movements of the small intestine in dogs, and these values can be analyzed for qualitative and quantitative data, which provides a noninvasive way of assessing intestinal kinetics (An *et al.*, 2001).

***Basic principles and physical properties of contrast-enhanced ultrasonography***

The CEUS is a diagnostic imaging technique based on the use of contrast-enhanced images associated with ultrasound examination. It is a current tool applied to routine veterinary medicine and has led to the modernization and optimization of the diagnosis of various conditions in animals, especially in diseases that compromise the vascular perfusion of the organ (Nyman *et al.*, 2005).

CEUS uses microbubbles (Fig. 4) as a contrast medium to intensify the perception of ultrasound echoes and quantify tissue perfusion. Microbubbles have a high reflective capacity and are therefore able to increase the Doppler signal, allowing the perception of flows that cannot be verified by other techniques. The presence of these microbubbles in each region visualized by USG

generates a contrast effect in the structure analyzed, which favors the observation of microcirculation (Nyman *et al.*, 2005).

Microbubbles are gases with a high molecular weight and a diameter of between 1 and 6 μm, which favors and allows them to pass through the bed of the blood capillaries into the intravascular space without diffusing into the interstitium and renal excretion, making them safe to use with no hemodynamic effects (Kalantarinia and Okusa, 2007). The use of microbubble contrast was made unfeasible for a time because there was no commercially available product that could remain in the intravascular space after passing through the pulmonary circulation. Subsequently, more resistant media capable of remaining in the systemic circulation were created (Ignee *et al.*, 2016).

Currently, most contrast mediums are stable because they contain microbubbles that are covered by a capsule, usually a thin membrane of phospholipids arranged in a monolayer with hydrophilic poles on the outside toward the circulating blood and lipophilic chains arranged toward the encapsulated gas (Baun, 2017; Takahashi *et al.*, 2021). These contracts are currently classified as first-generation, when the gas used has a lower diffusion coefficient and is, therefore, less stable, and second-generation, which is more modern because the gas has a higher diffusion coefficient and is, therefore, more stable (Takahashi *et al.*, 2021).
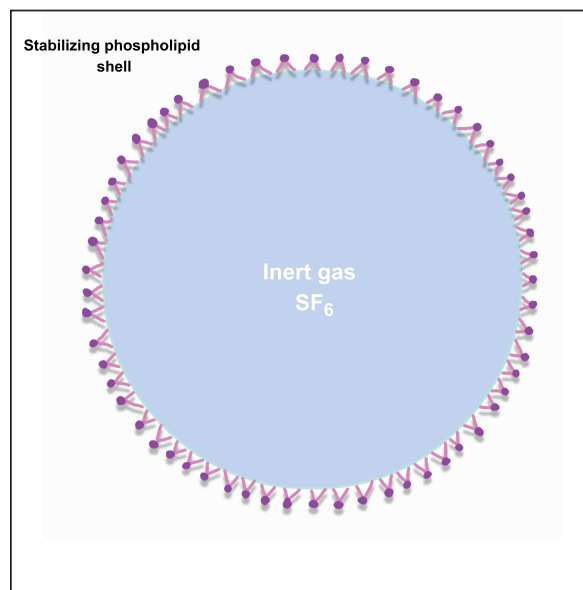


**Fig. 4.** Representative image of a microbubble used as an ultrasound contrast agent. The material consists of a lipid-coated membrane filled with gaseous content. The lumen is filled with sulfur hexafluoride (SF6). The figure was created using the Penup Samsung® application (Windows 11, Ldta, Samsung Electronics, Suwon, South Korea) and Canva Pro® (Canva Pty Ltd, Sydney, Australia).

The product most commonly used in this technique is sulfur hexafluoride, a contrast element capable of remaining stable inside the vessels without diffusing through the vascular bed. The medium also has the potential to remain in the bloodstream for prolonged periods due to its low solubility in water and the high molecular weight of its gaseous molecules, which makes it resistant to external pressure (Volta *et al.*, 2014) and is excreted by the lungs (Nyman *et al.*, 2005). It can be administered mainly by two routes: intravenous and intracavitary (Takahashi *et al.*, 2021). Contrasts have a low capacity to cause adverse reactions. In veterinary medicine, syncope and emesis are described after 24 hours of use (Seiler *et al.*, 2013). In medicine, there are few reports of headaches, nausea, and increased body temperature after CEUS (Haers *et al.*, 2010). The product is contraindicated for individuals with hypersensitivity reactions to any component of the contrast medium (Takahashi *et al.*, 2021). The safety profile of the material used to perform CEUS has been extensively studied in children and has been shown to be highly safe with discrete, self-limiting, or easily reversible reactions (Rosado and Riccabona, 2016; Mao *et al.*, 2019).

The intravenous administration of CEUS is indicated for assessing intestinal perfusion. After application, the microbubbles are restricted to the intravascular space without entering the organ parenchyma (Fig. 5). Contrast media classified as second generation can remain in the vascular bed for approximately 10 minutes, and as the bubbles break, they are gradually eliminated through respiration (Takahashi *et al.*, 2021). The physical properties of using microbubbles are due to the nonlinear detection that bubbles of different sizes produce when they are diluted in the patient's blood and meet ultrasound echoes and, consequently, produce harmonic frequencies. The oscillation of this material in the evaluation region generates an intense contrast enhancement on the surface of the organ, as shown in the USG image, allowing microcirculation to be observed (Nyman *et al.*, 2005).

The image parameters used to evaluate this imaging method are related according to the degree of homogeneous or heterogeneous filling of the organ's microcirculation by the microbubbles, mainly in specific circular regions within the tissues being examined. This defines the intensity patterns as "hyperintense" in tissues with greater intensity of visualization of the contrasted region, "isointense" in which it is not possible to distinguish the tissue adjacent to the evaluated region even with the use of contrast and "hypointense", which determines a pattern of low intensity (Volta *et al.*, 2014).
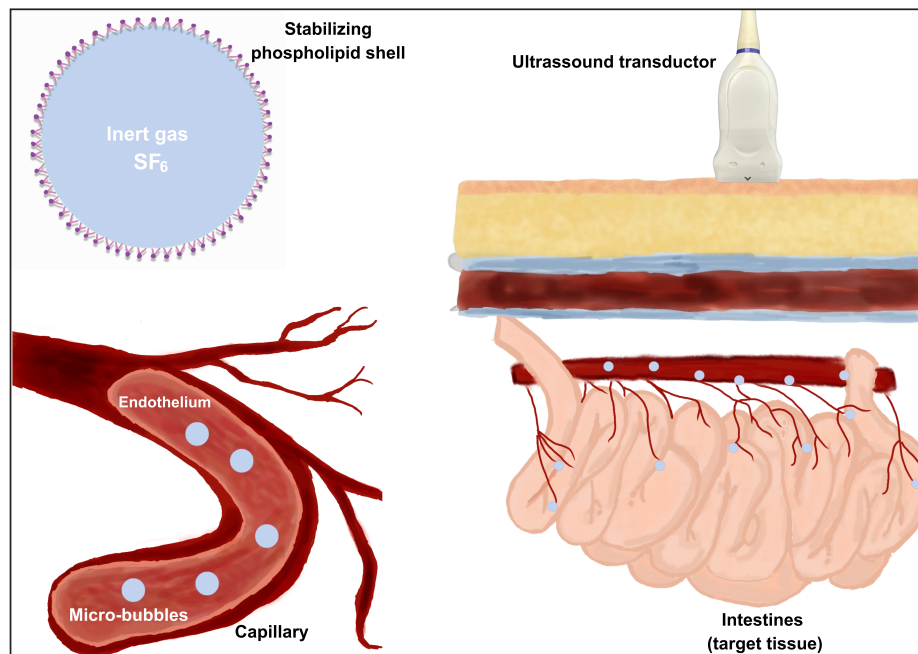


**Fig. 5.** Representative images of the basic principles of ultrasound with microbubble contraction. When exposed to ultrasound echoes, the microbubbles containing gas and coated with lipid membrane oscillate in expansion and generate vascular markings in the tissue. The migration of the contrast into the intestinal vessels can be observed. The figure was created using the Penup Samsung® application (Windows 11, Ldta, Samsung Electronics, Suwon, South Korea) and Canva Pro® (Canva Pty Ltd, Sydney, Australia).

In addition, CEUS can be used to assess vessel filling time from the intravenous application of contrast to the start of perfusion of the organ under analysis, known as wash-in. The peak of contrast enhancement corresponds to the moment of greatest perfusion after wash-in and the time of total contrast elimination, known as wash-out (Lock *et al.*, 2011).

***Contrast-enhanced ultrasound for intestinal evaluation in dogs***

In medicine, among the various organs evaluated using this technique, there have been studies on intestinal ischemia (Kalantarinia and Okusa, 2007). Furthermore, the intestinal perfusion of people with Crohn's disease was assessed using CEUS, and changes were observed in the contrast enhancement pattern and intestinal perfusion parameters when compared with healthy control subjects (Quaia, 2013). In addition, CEUS has been used for intestinal assessment in medicine to monitor the activity and response to treatment of Crohn's disease (Saevik *et al.*, 2014).

Some CEUS studies used in the intestinal evaluation of dogs are presented in Table 4. The evaluation of intestinal perfusion in dogs provides important information for the diagnosis and monitoring of

**Table 4.** Contrast-enhanced ultrasonography studies for intestinal evaluation in dogs organized by author, study design, number of samples and main results.

| Reference | Study design | *N* | Results on accuracy of CEUS |
|---|---|---|---|
| Jiménez *et al.* 2011 | Original research article | 9 dogs | The CEUS technique and the normal jejunal perfusion pattern described can provide useful data for the evaluation of vascular, inflammatory and neoplastic intestinal diseases in dogs. |
| Johnson-Neitman *et al.* 2012 | Original research article | 8 dogs | The results suggest that contrast-enhanced ultrasound can be a useful and minimally invasive method for assessing duodenal perfusion in dogs. The data obtained in healthy dogs can help with enteropathies and response to clinical treatment. |
| Caivano *et al.* 2017 | Preliminary Findings | 17 dogs | The study demonstrated that CEUS of the duodenum in dogs is feasible and highlights the presence of different vascular patterns and contrast enhancement characteristics in dogs with inflammatory bowel disease. |
| Nisa *et al.* 2017 | Prospective original research article | 6 dogs | CEUS was viable for assessing duodenal perfusion in healthy sedated dogs. |
| Nisa *et al.* 2018 | Original research article | 6 dogs | The combination of butorphanol and midazolam had no significant influence on the perfusion parameters of duodenal CEUS and could be a good option for sedation before duodenal CEUS in debilitated dogs. |
| Nisa *et al.* 2019 | Cross-sectional original research article | 47 dogs | No significant differences were found in perfusion parameters between the lymphoma and chronic inflammatory enteropathy groups or between the lymphoma and control groups. Peak intensity and the area under the curve can detect duodenal inflammation and are potentially useful for excluding a diagnosis of canine chronic inflammatory enteropathy. |
| Linta *et al.* 2021 | Original research article | 62 dogs | CEUS allows discrimination between dogs with inflammatory bowel disease and healthy dogs by evaluating time-intensity curves, but did not provide useful information for monitoring the response to treatment. |
| Cordella *et al.* 2021 | Case report | 1 dog | CEUS showed increased and heterogeneous vascularization of the colon in histiocytic colitis. |
| Simeoni *et al.* 2021 | Prospective original research article | 41 dogs | CEUS can be useful in the evaluation of canine gastric disorders to distinguish between gastritis and gastric neoplasms, but there are no specific aspects that allow this differentiation. |
| Chhoey *et al.* 2023 | Case report | 1 dog | Evaluation of perfusion by CEUS can help diagnose and characterize gastrointestinal stromal turmoil in dogs. |
| Chhoey *et al.* 2024 | Restrospective study | 3 dogs | Intraluminal CEUS evaluations helped assess lumen permeability, the extent of luminal dilation, rule out luminal narrowing, determine the presence of a mass within the lumen and identify rupture sites. |

N: Number of samples; CEUS: Contrast-enhanced ultrasonography.

chronic intestinal diseases (Nisa *et al.*, 2019). Jiménez *et al.* (2011) published quantitative and qualitative data that allowed the intestinal vascularization of dogs without enteropathy to be characterized, but clinical applicability had not been established at the time of the study.

One study evaluated the repeatability and reproducibility of perfusion parameters using CEUS of the duodenum of healthy dogs under sedation. The results showed that in this experimental unit profile and under sedation, the parameters were stable, namely the time taken for the contrast to reach the duodenum until maximum enhancement, the degree of intensity of this enhancement, and the entry and exit rates of the microbubbles. Despite these results, a further study in dogs with intestinal disorders was suggested to determine the clinical applicability of the test (Nisa *et al.*, 2019).

Subsequently, the duodenum of dogs with CE and alimentary lymphoma was evaluated using CEUS. The CEUS-derived data showed that the regional blood volume in the duodenal mucosa was increased in dogs with CE compared with the control group. These results also suggest that perfusion parameters can be used as predictive values for duodenal inflammation in symptomatic animals. However, the modality was not sufficient to differentiate CE from alimentary lymphoma given the similarity of CEUS findings in both conditions (Nisa *et al.*, 2019).

In a more recent study, CEUS was effective in distinguishing dogs with CE from those without intestinal disease using the results of the microbubble contraction time and intensity curve. However, qualitative evaluation could not determine this differentiation. Furthermore, the method is not capable of evaluating the monitoring of response to treatment independently if qualitative and quantitative metrics are considered (Linta *et al.*, 2021).

In a case report of granulomatous colitis in a French Bulldog multiple diagnostic imaging techniques were used, such as B-mode USG, elastography, and CEUS. Contrast enhancement showed greater demarcation in the serosa and muscular region and, to a lesser extent, in the mucosa and submucosa, as well as an accelerated wash-in and wash-out pattern, which determined increased and heterogeneous circulation in the colon (Cordella *et al.*, 2021).

In general, in the veterinary medical literature, the application and use of CEUS are more restricted to intestinal perfusion studies, specifically in the duodenum. In some cases, the technique has been described in the use of dogs with CE and alimentary lymphoma (Nisa *et al.*, 2019). Considering the scarcity of publications on the subject, a recent study aimed to prospectively evaluate the feasibility of CEUS in the duodenal perfusion of dogs, to observe the enhancement pattern and, in addition, to follow up dogs treated in a standardized way for intestinal affection using this imaging exam (Linta *et al.*, 2021).

We emphasize that although USG can provide important information about the GIT in dogs, it is important to consider that its indication should not be based solely on isolated clinical signs. It is necessary to associate clinical information, physical examination findings, and correlations with laboratory results. In some situations, ultrasound examination may be inconclusive because of the lack of specificity of clinical gastrointestinal signs in dogs. Therefore, we believe that USG is an important diagnostic tool, but it must be used based on well-defined clinical criteria.

## Conclusion

The anatomical and functional complexity of GIT in dogs, together with the frequent diseases that affect the intestines of these animals and the nonspecific clinical signs, highlight the importance of auxiliary diagnostic methods. This underscores the relevance of basic and advanced CEUS. These dynamic procedures help assess both functionality and morphology.

We emphasize that USG results can be nonspecific if the underlying cause of the disease is not related to the gastrointestinal tract. Due to the nonspecificity of gastrointestinal clinical signs, ultrasound examination should be indicated based on the entire clinical context, history, and other complementary tests.

B-mode ultrasound has limitations for gastrointestinal assessment, such as the experience of the examiner, the presence of gas in the GIT, the depth of the organs assessed, and anatomical variations between species and breeds. CEUS for intestinal assessment has limitations, such as being restricted to use in animals with heart disease and respiratory disorders and the lack of studies with reference parameters for different enteropathies.

An analysis of the descriptions of the sources used in this study shows that B-mode and CEUS are valuable approaches for noninvasive intestinal assessment. These findings emphasize the growing importance of combining different complementary tests to understand the pathophysiology of enteropathies.

It is worth noting that studies are still needed to fill the gaps in the literature on the methodology of applying these imaging tests, to standardize reference values and measurements, to systematize execution, and to provide practical clinical applications for the different diseases affecting the intestines of dogs.

*Data availability*

All data supporting the findings of this study are available in the manuscript.

*Authors' contribution*

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