

STANDARD ARTICLE

Cerebrospinal fluid lactate concentrations in dogs with seizure disorders

Christopher L. Mariani^{1,2,3}  | Carolyn J. Nye¹  | Laura Ruterbories^{1,2} |
 Debra A. Tokarz⁴ | Lauren Green² | Jeanie Lau² | Natalia Zidan² |
 Peter J. Early^{2,3}  | Karen R. Muñana^{2,3}  | Natasha J. Olby^{2,3}  |
 Chun-Sheng Lee² | Julien Guevar² 

¹Comparative Neuroimmunology and Neuro-oncology Laboratory, College of Veterinary Medicine, North Carolina State University, Raleigh, North Carolina

²Department of Clinical Sciences, College of Veterinary Medicine, North Carolina State University, Raleigh, North Carolina

³Comparative Medicine Institute, North Carolina State University, Raleigh, North Carolina

⁴Department of Population Health and Pathobiology, College of Veterinary Medicine, North Carolina State University, Raleigh, North Carolina

Correspondence

Christopher L. Mariani, Department of Clinical Sciences, College of Veterinary Medicine, NC State University, 1060 William Moore Drive, Raleigh, NC 27607.

Email: clmarian@ncsu.edu

Present address

Carolyn J. Nye, Carolyn J. Nye, Palm Beach Veterinary Specialists, 3884 Forest Hill Boulevard, West Palm Beach, FL

Debra A. Tokarz, Debra A. Tokarz, Experimental Pathology Laboratories Inc., 615 Davis Drive #500, Morrisville, NC

Jeanie Lau, Jeanie Lau, Small Animal Specialist Hospital (SASH), 1 Richardson Place, North Ryde, Sydney, Australia

Jeanie Lau, Jeanie Lau, Small Animal Specialist Hospital (SASH), 1 Richardson Place, North Ryde, Sydney, Australia

Chun-Sheng Lee, 6F, No. 23, Ln. 61, Guangrong Road, Luzhou District, New Taipei City, Taiwan

Julien Guevar, Julien Guevar, Department of Clinical Veterinary Sciences, Vetsuisse Faculty, University of Bern, Langgasstrasse 128, Bern, Switzerland

Funding information

Clinical Research, Education and Treatment of Encephalitis (CREATE) Fund; Department of

Abstract

Background: Cerebrospinal fluid (CSF) lactate concentrations increase after seizure activity in many human patients independent of the underlying disease process. The effect of seizure activity on CSF lactate concentration in dogs is unknown.

Hypothesis/Objectives: Cerebrospinal fluid lactate concentration is unaffected by seizure activity in dogs and is more dependent on the underlying disease process causing the seizures.

Animals: One-hundred eighteen client-owned dogs with seizure disorders.

Methods: Case series. Cerebrospinal fluid lactate concentration was determined using a commercially available lactate monitor. Seizure semiology, time from last seizure to CSF collection, number of seizures within the 72 hours preceding CSF collection, and clinical diagnosis were recorded.

Results: Dogs with focal seizures had higher CSF lactate concentrations than did those with generalized seizures ($P = .03$). No differences in lactate concentrations were found among dogs with single seizures, cluster seizures or status epilepticus ($P = .12$), among dogs with CSF collection at different time points after the last seizure activity ($P = .39$) or among dogs having different numbers of seizures within the 72 hours preceding CSF collection ($P = .42$). A significant difference ($P = .001$) was found in CSF lactate concentrations among diagnostic groups, and dogs with inflammatory and neoplastic disease had higher concentrations than did dogs with idiopathic or unknown epilepsy.

Abbreviations: CNS, central nervous system; CSF, cerebrospinal fluid; MRI, magnetic resonance imaging; ROC, receiver-operating characteristic.

This is an open access article under the terms of the Creative Commons Attribution-NonCommercial License, which permits use, distribution and reproduction in any medium, provided the original work is properly cited and is not used for commercial purposes.

© 2020 The Authors. *Journal of Veterinary Internal Medicine* published by Wiley Periodicals LLC on behalf of American College of Veterinary Internal Medicine.

Conclusions and Clinical Importance: Cerebrospinal fluid lactate concentration is minimally affected by seizure activity in dogs and increased concentrations are more likely associated with the underlying disease process.

KEYWORDS

acute repetitive seizures, biomarker, cluster seizures, epilepsy, status epilepticus

1 | INTRODUCTION

Measurement of lactate concentration in the cerebrospinal fluid (CSF) is potentially useful as a biomarker of disease in the central nervous system (CNS) of dogs. Several investigators have published normative data and reference intervals for this analyte in the CSF of dogs,¹⁻⁴ but only a few reports of CSF lactate concentration in dogs with CNS disease are available.^{1,5-8} Cerebrospinal fluid lactate concentration in humans is increased in a number of CNS disorders including mitochondrial disorders or other inborn errors of metabolism,^{9,10} inflammatory conditions,^{9,11,12} neoplastic disease,⁹ cerebrovascular disease⁹ and head trauma,¹³ and CSF lactate concentration is particularly useful in discriminating bacterial from aseptic meningitis.^{14,15} A handheld lactate monitor recently has been validated for quantification of lactate in canine blood¹⁶ and CSF,⁴ and a CSF reference interval has been established for this analyte (1.0-2.5 mmol/L).

A number of studies also suggest that CSF lactate concentration increases in human patients after seizure activity, independent of the underlying cause of the seizures.^{9,17-20} Such increases have been described to persist for 24 to 72 hours and confound the diagnostic utility of this analyte in identifying underlying etiologies.⁹ We had not observed a consistent increase in CSF lactate concentration in dogs after seizures.

Our objectives were to evaluate CSF lactate concentrations in dogs with seizure disorders, to determine if these concentrations were associated with the underlying disease process and evaluate if these concentrations were associated temporally with the seizure activity. We hypothesized that seizure activity would have minimal effects on CSF lactate concentration and that this concentration would be more dependent on the underlying disease process causing the seizures.

2 | MATERIALS AND METHODS

2.1 | Animals, sample collection, and lactate analysis

Cerebrospinal fluid samples were collected from dogs that were referred to the NC State Veterinary Hospital for epileptic seizures as part of a routine diagnostic evaluation. The samples were obtained from the cerebellomedullary cistern or lumbar subarachnoid space in a routine manner. Most samples were analyzed immediately using a commercially available lactate monitor (Lactate Plus, Nova Biomedical, Waltham, Massachusetts) although some were evaluated after being frozen at -80°C , which has little effect on lactate concentration.⁴

2.2 | Seizure classification

Although the samples primarily were analyzed in a prospective manner, medical records of dogs with CSF lactate concentration also were retrospectively reviewed to determine diagnosis and inclusion into the study. Dogs were included if seizures were a component of the presenting complaint. Seizure activity was established based on owner or referring veterinarian reports before admission or direct observation by veterinary clinicians or technicians while dogs were hospitalized. Additional data required for inclusion in the study were CBC, serum biochemical analysis, CSF analysis, and a magnetic resonance imaging (MRI) study of the brain. Seizures were classified as generalized, focal, or undetermined based on a review of the medical records (seizure semiology). Dogs having both generalized and focal seizures were placed in the generalized group for analysis purposes. Cases also were categorized as having had a single seizure, cluster seizures (>1 seizure within 24 hours) or status epilepticus (>5 minutes of continuous seizure activity or >1 seizure without return to baseline neurological status between events), which was termed "seizure presentation." The number of seizures (if clustered) and the time from the last seizure to CSF collection also were recorded and dogs were grouped as <24 hours, 24 to 72 hours, and > 72 hours for comparison. Dogs having seizures within the 72 hours preceding CSF collection were further categorized according to the number of observed seizures in this time period as 1 to 2, 3 to 9, or >10 seizures. Dogs in status epilepticus were included in the latter group. Cases with an incomplete medical record were excluded but cases with incomplete information regarding seizure semiology, seizure presentation, and seizure timing were permitted.

2.3 | Disease categorization

Dogs were classified with idiopathic (suspected genetic, primary) epilepsy if they had their first seizure between 6 months and 6 years of age, a normal interictal neurologic examination and normal CBC, serum biochemistry, intracranial imaging, and CSF analysis.^{21,22} Mild increases in CSF protein concentration (<50 mg/dL) were permitted in this category.²³ Dogs with abnormalities on brain imaging or CSF analysis were classified as having structural epilepsy. Those with unremarkable diagnostic testing but falling outside of the age range determined for idiopathic epilepsy were classified with unknown epilepsy. For comparison with other diagnostic groups, dogs with idiopathic and unknown epilepsy were combined into a single cohort. Finally, dogs determined to have

metabolic or toxic causes of their seizures were considered to have reactive seizures.

Dogs with structural epilepsy were further subdivided into presumptive neoplastic, inflammatory, vascular, and miscellaneous subgroups. Some cases were classified using histopathological reports after biopsy or necropsy examination. Dogs were diagnosed with presumptive neoplastic CNS disease if they had a well-defined mass lesion on brain imaging or neoplastic cells visible on cytological evaluation of the CSF. Dogs were considered to have presumptive inflammatory CNS disease (i.e., meningoencephalitis) if they had an increased CSF nucleated cell count (>5 cells/ μ L) or brain imaging results consistent with meningoencephalitis (multifocal or diffuse T2-weighted and fluid-attenuated inversion recovery [FLAIR] hyperintensities with variable contrast enhancement). Dogs were diagnosed with a presumptive cerebrovascular disorder if they had acute onset of neurological signs without subsequent progression of these signs, changes consistent with either ischemic injury or hemorrhage on MRI (including T2* and diffusion-

weighted studies) and a CSF nucleated cell count within the reference interval. Small diameter (<6 mm), round-ovoid, hypointense T2* lesions (putative cerebral microbleeds), which often are seen incidentally in dogs,^{24,25} were not considered sufficient for a diagnosis of a cerebrovascular disorder. Additional neurological signs, improvement of signs with or without therapeutic intervention and partial or complete resolution of lesions on repeat brain imaging were other factors considered in assigning cases to a diagnostic category. Dogs in the miscellaneous category included those diagnosed with congenital malformations, neurodegenerative disorders, or undefined lesions not easily categorized as described above.

2.4 | Statistical analysis

Descriptive statistics were generated for age at presentation, weight, CSF nucleated cell count, CSF red blood cell count, and CSF protein and lactate concentrations. D'Agostino and Pearson normality testing

TABLE 1 Physical characteristics of dogs with seizures

Diagnostic category	Number of dogs	Age (y)	Weight (kg)	Sex			
				M	MC	F	FS
Idiopathic	21	3.9 (1.1-5.9)	20.5 (5.6-46.5)	1	8	2	10
Unknown	26	7.7 (0.3-12.3)	26.9 (1.8-72.0)	2	13	2	9
Idiopathic/unknown ^a	47	5.9 (0.3-12.3)	21.0 (1.8-72.0)	3	21	4	19
Presumptive neoplastic	37	9.9 (5.0-13.8)	26.7 (2.8-50.4)	0	17	0	20
Presumptive inflammatory	16	5.5 (2.4-13.3)	6.3 (2.5-30.6)	1	3	1	11
Presumptive vascular	3	10.8 (8.2-12.2)	26.7 (23.1-32.6)	0	1	0	2
Miscellaneous	12	6.4 (0.1-13.4)	14.3 (1.6-45.5)	2	3	1	6
Reactive	3	12.5 (6.8-12.5)	7.4 (6.8-24.6)	0	1	0	2
All cases	118	7.4 (0.1-13.8)	20.6 (1.6-72.0)	6	46	6	60

Note: Age and weight values are expressed as median (range).

Abbreviations: F, female; FS, female spayed; M, male; MC, male castrated.

^aIdiopathic/unknown is a combination of the idiopathic and unknown categories.

TABLE 2 Cerebrospinal fluid parameters in dogs with seizures

Diagnostic category	Collection site		Nucleated cell count (cells/ μ L)	Red blood cell count (cells/ μ L)	Protein (mg/dL)
	CCSF	LCSF			
Idiopathic	20	1	1 (0-3)	10 (0-2630)	14.8 (10.8-34.5)
Unknown	26	0	1 (0-5)	7.5 (0-8500)	17.3 (8.9-48.4)
Idiopathic/unknown	46	1	1 (0-5)	10 (0-8500)	16.0 (8.8-48.4)
Presumptive neoplastic	34	3	1 (0-10 244)	5 (0-15 000)	23.8 (12.0-146.8)
Presumptive inflammatory	13	3	92 (2-3858)	12.5 (0-5260)	79.0 (18.5-300.8) ^a
Presumptive vascular	3	0	1 (0-1)	0 (0-0)	24.2 (22.4-32.6)
Miscellaneous	11	1	1 (0-116)	2 (0-1183)	20.0 (9.1-2018.0)
Reactive	3	0	6 (2-44)	358 (0-518)	18.7 (13.5-101.3)
All cases	110	8	1 (0-10 244)	5 (0-15 000)	20.2 (8.8-2018)

Note: Nucleated cell count, red blood cell count, and protein values are expressed as median (range).

Abbreviations: CCSF, cerebellomedullary cistern cerebrospinal fluid; LCSF, lumbar cerebrospinal fluid.

^aIn 2 samples, the quantity was not sufficient to analyze protein concentrations.

TABLE 3 Cerebrospinal fluid lactate concentrations in dogs by seizure presentation, seizure semiology, time from last seizure to CSF collection, and number of seizures within 72 hours of CSF collection

Diagnostic category	Number	Median lactate (range) (mmol/L)	Mean lactate \pm SD (mmol/L)	Samples above reference interval ^a
Single seizure	39	2.0 (1.1-8.4)	2.5 \pm 1.6	6 (15.4%)
Cluster seizures	64	1.8 (0.6-7.0)	1.9 \pm 0.8	6 (9.4%)
Status epilepticus	12	2.0 (1.2-2.9)	2.0 \pm 0.5	2 (16.7%)
Total seizure presentation	115^b	1.9 (0.6-8.4)	2.1 \pm 1.2	14 (12.2%)
Generalized seizures	93	1.9 ^a (0.6-8.0)	2.0 \pm 1.0	8 (8.6%)
Focal seizures	17	2.1 ^b (1.2-8.4)	2.7 \pm 1.9	5 (29.4%)
Total seizure semiology	110^c	1.9 (0.6-8.4)	2.1 \pm 1.2	13 (11.8%)
<24 h	47	1.9 (0.6-7.0)	2.0 \pm 1.0	7 (14.9%)
24-72 h	27	1.9 (0.9-8.4)	2.2 \pm 1.5	2 (7.4%)
>72 h	40	2.0 (1.2-8.0)	2.2 \pm 1.1	5 (12.5%)
Total seizure timing	114^d	1.9 (0.6-8.4)	2.1 \pm 1.2	14 (12.2%)
1-2 seizures	30	2.0 (0.9-8.4)	2.3 \pm 1.5	5 (16.7%)
3-9 seizures	23	1.7 (0.6-2.6)	1.7 \pm 0.4	1 (4.3%)
10+ seizures	20	1.9 (1.1-7.0)	2.1 \pm 1.3	3 (15%)
Total seizure number	73^e	1.9 (0.6-8.4)	2.1 \pm 1.2	9 (12.3%)

Note: Within a column, median values with different superscript letters differ significantly ($P < .05$).

Abbreviation: CSF, cerebrospinal fluid.

^aReference interval is 1.0 to 2.5 mmol/L.

^bIn 3 dogs, seizure presentation could not be established from the medical record.

^cIn 8 dogs, seizure semiology could not be established from the medical record.

^dIn 4 dogs, time from last seizure to CSF collection could not be established from the medical record.

^eIn 1 dog, the number of seizures could not be established from the medical record.

showed that the lactate concentrations for most groups did not follow a normal distribution and therefore nonparametric evaluations were used to compare groups. Dogs with idiopathic and unknown epilepsy diagnoses were combined for further analyses. Lactate concentrations in dogs with generalized and focal seizures were compared using a Mann-Whitney test. A Kruskal-Wallis test followed by Dunn's multiple comparison test was used to compare lactate concentrations among different seizure presentations, among dogs with different times from last seizure to CSF collection, among dogs with different numbers of seizures within 72 hours before CSF collection and among diagnostic groups. A receiver-operating characteristic (ROC) analysis was used to compare lactate concentrations in dogs with structural epilepsy to those with idiopathic or unknown epilepsy. Spearman correlation was used to examine the relationship between CSF red blood cell counts and lactate concentrations. P values $< .05$ were considered significant. All analyses were conducted using commercial software (Prism version 8.4.2, Graphpad Software, Inc, La Jolla, California).

3 | RESULTS

A total of 118 dogs presenting with seizures were included, with a variety of breeds represented. Descriptive statistics for age, weight and sex of the individual diagnostic groups and the whole cohort are shown in Table 1. One-hundred ten CSF samples (93.2%) were collected from the cerebellomedullary cistern and 8 (6.8%) from the

lumbar subarachnoid space. Characteristics of the CSF analysis for each diagnostic group are shown in Table 2. Because of limited sample volumes, total protein concentrations were not available for 2 cases. No correlation was found between CSF red blood cell count and CSF lactate concentrations ($P = .33$, $r = -0.09$). Infectious disease testing and necropsy examinations were performed in some cases, and these results are included in a supplemental document outlining details from dogs with structural epilepsy (Table S1).

Lactate concentrations for dogs categorized according to seizure presentation, semiology, timing, and number of seizures within 72 hours of CSF collection are shown in Table 3. A significant difference in lactate concentrations ($P = .03$) was noted between dogs with seizures noted to be generalized versus those with exclusively focal seizures. In 8 dogs, seizure semiology was undetermined. No difference was found in lactate concentrations among dogs with single seizures, cluster seizures, or status epilepticus ($P = .12$). In 3 dogs, the seizure presentation could not be determined from the medical record. Seventy-four dogs had seizures within 72 hours of CSF collection, but in 1 dog the number of seizures could not be determined. No difference was found in lactate concentrations among dogs with CSF collection at different time points after the last seizure activity ($P = .39$, Figure 1A) or among dogs having different numbers of seizures within the 72 hours preceding CSF collection ($P = .42$, Figure 1B).

Lactate concentrations for dogs categorized by presumptive diagnosis are shown in Table 4. A significant difference ($P = .001$) was

found in CSF lactate concentrations among diagnostic groups, with dogs with presumptive inflammatory and neoplastic disease having higher concentrations than dogs with idiopathic or unknown epilepsy.

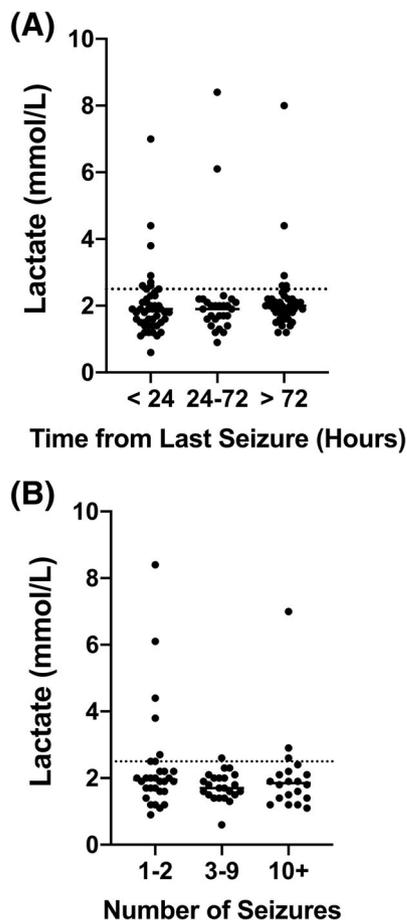


FIGURE 1 A, Cerebrospinal fluid (CSF) lactate in dogs ($n = 114$) grouped by time from last observed seizure to CSF collection. B, Cerebrospinal fluid lactate in dogs with at least one observed seizure within 72 hours of CSF collection ($n = 73$) grouped by number of observed seizures in this time period. Dogs in status epilepticus were categorized in the >10 seizure group. Note: Solid lines represent median values and dotted lines represent the upper limit of the reference interval

The ROC curve is shown in Figure 2 and indicated that a concentration exceeding the upper limit of the established lactate reference interval (0-2.5 mmol/L) was 100% specific in identifying a dog with structural epilepsy but this concentration had a sensitivity of only 21%.

4 | DISCUSSION

We found a statistically significant difference in CSF lactate concentrations between dogs with generalized seizures versus those with focal seizures, but no differences in lactate concentrations related to seizure presentation, duration from last seizure to CSF collection, or number of seizures within the 72 hours preceding CSF collection. Significant differences in CSF lactate concentrations were found among different diagnostic groups, with presumptive neoplastic and inflammatory groups having higher concentrations. Last, a lactate concentration above the established reference interval (>2.5 mmol/L) was 100%

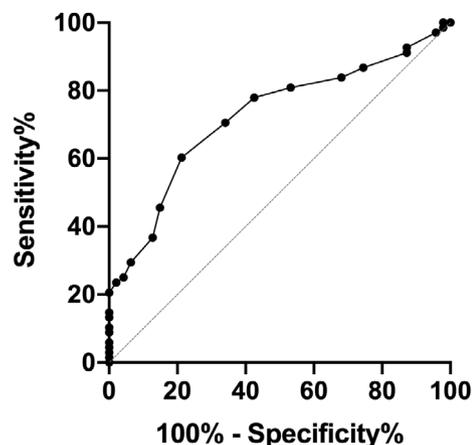


FIGURE 2 Receiver-operating characteristic (ROC) curve comparing cerebrospinal fluid lactate in dogs with idiopathic/unknown epilepsy ($n = 47$) to those with structural epilepsy ($n = 68$). The area under the ROC curve is 0.72 (95% confidence interval = 0.63-0.82)

TABLE 4 Cerebrospinal fluid lactate concentrations in dogs with seizures

Diagnostic category	Number	Median lactate (range) (mmol/L)	Mean lactate \pm SD (mmol/L)	Samples above reference interval*
Idiopathic	21	1.6 (1.1-2.4)	1.7 \pm 0.3	0 (0%)
Unknown	26	1.7 (0.6-2.5)	1.7 \pm 0.4	0 (0%)
Idiopathic/unknown	47	1.7 ^a (0.6-2.5)	1.7 \pm 0.4	0 (0%)
Presumptive neoplastic	37	2.0 ^b (0.9-6.1)	2.1 \pm 0.8	4 (10.8%)
Presumptive inflammatory	16	2.3 ^b (1.2-8.4)	3.0 \pm 2.0	7 (43.8%)
Presumptive vascular	3	2.5 (1.9-2.6)	2.3 \pm 0.4	1 (33.3%)
Miscellaneous	12	1.7 (1.1-8.0)	2.3 \pm 2.0	2 (16.7%)
Reactive	3	2.1 (1.2-2.2)	1.8 \pm 0.6	0 (0%)
All cases	118	1.9 (0.6-8.4)	2.1 \pm 1.2	14 (11.8%)

Note: Within a column, median values with different superscript letters differ significantly ($P < .05$).

*Reference interval is 1.0 to 2.5 mmol/L.

specific for predicting structural epilepsy, albeit with a sensitivity of only 21%.

In humans, several studies have suggested that CSF lactate concentration can be increased after seizures, independent of the underlying seizure etiology.^{9,17-19} This finding has been documented after both isolated seizures and status epilepticus^{17,19,26} and occurs in 14% to 28% of patients.^{18-20,26} Some studies have shown a greater increase in CSF lactate concentration with status epilepticus when compared with isolated seizures^{18,26} or with motor seizures versus nonmotor seizures.²⁰ Other studies have shown less clarity in these areas.¹⁷ Similarly, the duration of seizure activity was a factor in lactate concentration increases in some studies,^{26,27} but not in others.¹⁷ One report found no difference in CSF lactate concentrations in children having seizures from various causes compared with controls, but discovered that a higher proportion of those with recurrent or prolonged seizures (>30 minutes) had increased lactate concentrations.^{28,29} Another study found that increases in CSF lactate concentrations were more likely associated with a shorter latency from status epilepticus to CSF collection¹⁹ and a number of reports have documented increased CSF lactate concentration for up to 72 hours after seizure events.^{9,26,27} Recommendations for normative CSF lactate data on humans suggest excluding patients with refractory epilepsy or those having recent seizures (within 24 hours).³⁰

We could not find evidence to support a role for seizure activity causing an increase in CSF lactate concentration in our study. In fact, somewhat counterintuitively, dogs with focal seizures had higher lactate concentrations than did those with generalized seizures in this cohort. In addition, no difference was found in lactate concentrations between dogs with single seizures and those with cluster seizures or status epilepticus and no evidence was found that recent seizures were more likely to be associated with a higher CSF lactate concentration than those with a longer interval between the seizure event and CSF collection. It is possible the CSF lactate was higher in animals with focal seizures because dogs with structural epilepsy were more likely to have focal seizures (i.e., that the lactate differences were driven primarily by the disease process and not the nature of the seizures). After eliminating those dogs in which seizure semiology was undetermined, 6/46 (13.0%) idiopathic or unknown cases and 11/61 (18.0%) structural cases had focal seizures. However, these data on seizure semiology must be viewed cautiously because the differences in lactate concentration are relatively small, and we considered dogs with both focal and generalized seizures in the generalized category in the analysis of seizure semiology. In addition, it is very difficult to distinguish dogs that have a focal onset of their seizures with rapid secondary generalization from those having true generalized seizures, particularly in a retrospective study relying mainly on owner-reported events. We suspect that this finding has no clinical relevance, but it should perhaps be further investigated in future studies.

Limited published information is available regarding CSF lactate concentrations in dogs with seizures, and we are not aware of other clinical studies investigating them in veterinary patients. Experimental studies in various species, including dogs, have shown that prolonged seizures lead to increased cerebral blood flow and an increase in cerebral consumption of oxygen and glucose that is 2 to 3 times normal

throughout sustained seizure activity.³¹⁻³³ Concurrently, a number of studies have shown increases in brain tissue lactate concentrations with experimental seizure activity.^{31,34-36} The conclusion of many of these studies is that the mechanism of increased CSF lactate concentration with seizure activity is related to increased metabolic activity resulting from intense and coordinated neuronal firing and not to the presence of hypoxic conditions.^{31,32,36,37} Instead, it is thought that glucose is diverted from mitochondrial oxidative degradation and instead is shunted to anaerobic glycolysis pathways, with large amounts of lactate being released into the brain parenchyma and potentially into the CSF.^{32,38,39}

However, an experimental study of dogs induced to have seizures using IV pentylentetrazol or an electroconvulsive instrument showed that brain tissue and CSF lactate concentrations remain unaffected in dogs having seizures when anesthetized and ventilated on 100% oxygen.⁴⁰ In dogs allowed to become hypoxemic (PaO₂ of 21-48 mm Hg), brain tissue lactate concentration increased (approximately 2-fold) whereas CSF lactate concentration showed no changes.⁴⁰ Notably, these CSF measurements were made during seizure activity and changes in brain lactate concentration may not have had time to be reflected in the CSF. Nonanesthetized dogs having spontaneous seizures are likely to have blood oxygen tensions that fall between those reported in the 2 groups in this experimental study. A previous study showed a modest increase in mean CSF lactate concentration (3.8 mmol/L from a baseline of 3.1 mmol/L) when sampled immediately after 8 chemically induced seizures in anesthetized dogs ventilated with 100% oxygen.³³ A study of experimentally induced seizures in rats showed that CSF lactate concentration was increased as early as 10 minutes after administration of a proconvulsant compound, but had returned to normal by 6 hours after seizures.⁴¹

The reason for a lack of increased CSF lactate concentration in the dogs without structural epilepsy in our study is unclear. Discrepancies in experimental methods, species evaluated, anesthetic protocols, arterial oxygen saturation, methods of seizure induction, duration of seizure activity, and number of seizures make comparison among experimental studies challenging. However, the available data suggest that any increase in CSF lactate concentration in dogs with normal to increased arterial oxygen tensions is modest at best and may be transient. Unlike the scenario for most humans, CSF collection in clinical canine patients is performed under general anesthesia, typically in scenarios with increased oxygen tension in the blood. Therefore, if hypoxemia is a contributory factor in the development of increased brain and potentially CSF lactate concentrations, as suggested previously,⁴⁰ this change may have been masked by the anesthetic protocol, because dogs in our study were maintained on isoflurane or sevoflurane in oxygen and CSF was collected after brain imaging, potentially allowing additional time for normalization of lactate concentrations. Because CSF from dogs invariably is collected under general anesthesia and oxygen supplementation, increased blood oxygen concentrations may abrogate some of the potential effects of seizures on CSF lactate concentrations in clinical canine patients.

Other mechanisms aside from relative oxygen debt have been advanced to explain increased lactate production during seizures.

Excess synaptic glutamate associated with excessive neuronal firing is taken up by astrocytes, and stimulates glycolytic activity in astrocytes and subsequent release of lactate into the extracellular space.^{41,42} In addition, although somewhat controversial, evidence now suggests that lactate can be used as a fuel source for neurons.^{38,39} Relative differences in brain volume, the number of neurons and supporting cells involved in the seizure and the mechanisms of seizure initiation or propagation between dogs and humans all may play a role in explaining the discrepancies noted in CSF lactate concentration between dogs and human patients.

Many of the studies in humans described above excluded patients with infectious or inflammatory meningoencephalitis or meningeal carcinomatosis but included patients with solitary brain tumors, cerebrovascular accidents, and other forms of structural epilepsy.¹⁸⁻²⁰ Several of these studies failed to detect differences in CSF lactate concentrations when comparing patients with structural (symptomatic) epilepsy to those with unknown (cryptogenic) epilepsy.^{20,26} However, similar to our study, 2 previous studies found higher CSF lactate concentrations in human patients with structural epilepsy.^{18,19} Ultimately, CSF lactate concentrations in dogs with seizure activity may depend on the oxygenation state of the dog in question, additional comorbidities, and the underlying etiology initiating seizure activity.

Our study had several limitations. Although most of the CSF samples in our study were collected and analyzed prospectively, medical records were reviewed retrospectively, which contributed to some incomplete information with respect to seizure presentation, timing, and semiology. Samples from both the cerebellomedullary cistern and lumbar subarachnoid space were used, and the effect of sampling site on lactate concentration is unknown. It is possible that CSF lactate concentration changes secondary to seizures are diminished in samples collected from the lumbar subarachnoid space because of the relative distance of this collection site from the brain. One study comparing clinically healthy dogs found higher lactate concentrations in CSF collected from the lumbar subarachnoid space compared to the cerebellomedullary cistern, suggesting concentration of this analyte with caudal flow of CSF, although the difference was small (means of 1.58 mmol/L and 1.44 mmol/L respectively).⁴³ The number of samples collected from the lumbar region in our study was small (8/110) and of these, 3/8 (0/1 idiopathic, 1/3 neoplastic, 1/3 inflammatory, and 1/1 miscellaneous) had lactate concentrations above the reference interval. We therefore suspect that the site of collection had little influence on the results and conclusions of our study, although this possibility might be worthy of future investigation. We did not exclude cases based on blood contamination of CSF, which may have led to alterations of CSF lactate concentrations in some cases. However, we found no correlation between CSF red blood cell counts and CSF lactate concentrations. In the entire cohort of 118 dogs, only 13 dogs had a CSF RBC count > 500/ μ L, and of these 2/13 (15.4%) had CSF lactate concentrations that exceeded the high end of the reference interval. We did not consistently measure matched blood lactate concentrations in these dogs and cannot more rigorously investigate this relationship. However, it appears unlikely that blood contamination spuriously increased CSF lactate concentrations in our study

although falsely decreased lactate concentrations from a dilutional effect are possible.

We used clinical presentation, examination results, blood tests, neuroimaging findings, results of CSF analysis and response to treatment to categorize dogs into diagnostic groups, and it is possible that diagnoses in some dogs were incorrect. In addition, seizures themselves can create imaging abnormalities within the brain that are transient in nature,⁴⁴⁻⁴⁶ which also may have complicated diagnostic accuracy in our study. Although such postictal changes could be confused with several etiologies, such as neoplastic disorders or cerebrovascular disease, the primary concern is with meningoencephalitis. In these previous studies, the results of CSF analysis have been unremarkable or have shown albuminocytologic dissociation. Few dogs in our study had repeat brain imaging to better define any potential transient nature or improvement of the lesions noted. Of the 16 dogs diagnosed with presumptive inflammatory CNS disease, only 2 had normal CSF nucleated cell counts, and 1 of these was diagnosed with distemper encephalitis after necropsy examination. Of the 3 dogs diagnosed with cerebrovascular disorders in our study, 1 had what appeared to be an old thalamic infarct and 2 had lesions consistent with hemorrhagic infarcts, findings that are not typical of what has been reported for transient post-ictal changes in dogs. In the dogs categorized with miscellaneous structural lesions, most had MRI abnormalities that were also inconsistent with described transient post-ictal changes (Table S1). However, 2 dogs had MRI and CSF findings that could represent such changes, although both had normal CSF lactate concentrations. Thus, we suspect few dogs in our cohort had lesions solely attributable to their seizures, and some incorrect diagnoses, although possible, should not substantially change the main conclusions of our study. Finally, the CSF lactate concentration reference interval used was developed using dogs without identifiable CNS pathology, and in part using dogs with idiopathic and unknown epilepsy, because CSF from clinically normal dogs was not available for that study.⁴ Although dogs having seizures within 24 hours of CSF collection were eliminated in development of these variables, it is possible that dogs with refractory idiopathic or unknown epilepsy could have CSF lactate concentrations that are truly increased compared to clinically normal dogs, and future adjustment of this reference interval might be indicated. However, we found no effect of proximity of seizures to CSF collection on CSF lactate concentration in our study. Regardless, our study indicates that a CSF lactate concentration exceeding the current reference interval (>2.5 mmol/L) is highly predictive for structural epilepsy.

In conclusion, seizures appear to have minimal effect on lactate concentration in CSF obtained by conventional methods, and increased CSF lactate concentrations in dogs presenting with seizures are more likely related to the underlying disease process. Finding a CSF lactate concentration above the current reference interval (>2.5 mmol/L) may be a useful, rapid way to identify dogs at risk for structural epilepsy.

ACKNOWLEDGMENT

Funding for this study was provided by the Clinical Research, Education and Treatment of Encephalitis (CREATE) Fund and the Department of Clinical Sciences, NC State University.

CONFLICT OF INTEREST DECLARATION

Authors declare no conflicts of interest.

OFF-LABEL ANTIMICROBIAL DECLARATION

Authors declare no off-label use of antimicrobials.

INSTITUTIONAL ANIMAL CARE AND USE COMMITTEE (IACUC) OR OTHER APPROVAL DECLARATION

This study was conducted on samples obtained during the course of a routine diagnostic evaluation; IACUC approval is not required for such studies at our institution. However, written, informed consent was obtained from all owners for the use of these samples for research purposes.

ORCID

Christopher L. Mariani  <https://orcid.org/0000-0003-3228-6536>

Carolyn J. Nye  <https://orcid.org/0000-0003-3412-2260>

Peter J. Early  <https://orcid.org/0000-0003-3399-8500>

Karen R. Muñana  <https://orcid.org/0000-0001-9592-6276>

Natasha J. Olby  <https://orcid.org/0000-0003-1349-3484>

Julien Guevar  <https://orcid.org/0000-0001-9868-5703>

REFERENCES

- Caines D, Sinclair M, Wood D, et al. Evaluation of cerebrospinal fluid lactate and plasma lactate concentrations in anesthetized dogs with and without intracranial disease. *Can J Vet Res.* 2013;77:297-302.
- Galan-Rodriguez A, Carletti BE, Morgaz-Rodriguez J, et al. Short communication: cerebrospinal fluid lactate and pyruvate concentrations and ratio in healthy adult dogs. *Vet Rec.* 2013;173:249.
- Lobert V, Mischke R, Tipold A. Lactate and pyruvate levels in blood and cerebrospinal fluid. *Kleintierpraxis.* 2003;48:735-743.
- Nye CJ, Mariani CL. Validation of a portable monitor for assessment of cerebrospinal fluid lactate in dogs. *Vet Clin Pathol.* 2018;47:108-114.
- Benedicenti L, Gianotti G, Galban EM. Comparison between cerebrospinal fluid and serum lactate concentrations in neurologic dogs with and without structural intracranial disease. *Can J Vet Res.* 2018;82:97-101.
- Mariani CL, Nye CJ, Tokarz DA, et al. Cerebrospinal fluid lactate in dogs with inflammatory central nervous system disorders. *J Vet Intern Med.* 2019;33:2701-2708.
- Pugliese M, Carrasco JL, Andrade C, et al. Severe cognitive impairment correlates with higher cerebrospinal fluid levels of lactate and pyruvate in a canine model of senile dementia. *Prog Neuropsychopharmacol Biol Psychiatry.* 2005;29:603-610.
- Portero M, Martinez de Merlo E, Perez C, et al. Cerebrospinal fluid and blood lactate concentrations as prognostic biomarkers in dogs with meningoencephalitis of unknown origin. *Vet J.* 2019;254:105395.
- Chow SL, Rooney ZJ, Cleary MA, et al. The significance of elevated CSF lactate. *Arch Dis Child.* 2005;90:1188-1189.
- Magner M, Szentivanyi K, Svandova I, et al. Elevated CSF-lactate is a reliable marker of mitochondrial disorders in children even after brief seizures. *Eur J Paediatr Neurol.* 2011;15:101-108.
- Jarius S, Paul F, Franciotta D, et al. Cerebrospinal fluid findings in aquaporin-4 antibody positive neuromyelitis optica: results from 211 lumbar punctures. *J Neurol Sci.* 2011;306:82-90.
- Lutz NW, Viola A, Malikova I, et al. Inflammatory multiple-sclerosis plaques generate characteristic metabolic profiles in cerebrospinal fluid. *PLoS One.* 2007;2:e595.
- DeSalles AA, Kontos HA, Becker DP, et al. Prognostic significance of ventricular CSF lactic acidosis in severe head injury. *J Neurosurg.* 1986;65:615-624.
- Huy NT, Thao NT, Diep DT, et al. Cerebrospinal fluid lactate concentration to distinguish bacterial from aseptic meningitis: a systemic review and meta-analysis. *Crit Care.* 2010;14:R240.
- Sakushima K, Hayashino Y, Kawaguchi T, et al. Diagnostic accuracy of cerebrospinal fluid lactate for differentiating bacterial meningitis from aseptic meningitis: a meta-analysis. *J Infect.* 2011;62:255-262.
- Nye CJ, Musulin SE, Hanel RM, et al. Evaluation of the Lactate Plus monitor for plasma lactate concentration measurement in dogs. *J Vet Emerg Crit Care.* 2017;27:66-70.
- Calabrese VP, Gruemer HD, James K, et al. Cerebrospinal fluid lactate levels and prognosis in status epilepticus. *Epilepsia.* 1991;32:816-821.
- Chatzikonstantinou A, Ebert AD, Hennerici MG. Cerebrospinal fluid findings after epileptic seizures. *Epileptic Disord.* 2015;17:453-459.
- Malter MP, Choi S, Fink GR. Cerebrospinal fluid findings in non-infectious status epilepticus. *Epilepsy Res.* 2018;140:61-65.
- Suesse M, Saathoff N, Hannich M, et al. Cerebrospinal fluid changes following epileptic seizures unrelated to inflammation. *Eur J Neurol.* 2019;26:1006-1012.
- Berendt M, Farquhar RG, Mandigers PJ, et al. International veterinary epilepsy task force consensus report on epilepsy definition, classification and terminology in companion animals. *BMC Vet Res.* 2015;11:182.
- De Riso L, Bhatti S, Munana K, et al. International veterinary epilepsy task force consensus proposal: diagnostic approach to epilepsy in dogs. *BMC Vet Res.* 2015;11:148.
- Coelho AM, Maddox TW, Sanchez-Masian D, et al. Diagnostic value of cerebrospinal fluid analysis in a population of dogs with suspected idiopathic epilepsy. *Vet Rec.* 2019;185:539.
- Fulkerson CV, Young BD, Jackson ND, et al. MRI characteristics of cerebral microbleeds in four dogs. *Vet Radiol Ultrasound.* 2012;53:389-393.
- Kerwin SC, Levine JM, Budke CM, et al. Putative cerebral microbleeds in dogs undergoing magnetic resonance imaging of the head: a retrospective study of demographics, clinical associations, and relationship to case outcome. *J Vet Intern Med.* 2017;31:1140-1148.
- Tumani H, Jobs C, Brettschneider J, et al. Effect of epileptic seizures on the cerebrospinal fluid—a systematic retrospective analysis. *Epilepsy Res.* 2015;114:23-31.
- Imuekemhe SO, Obi JO, Sykes RM. Cerebrospinal fluid/serum lactic acid in febrile convulsions. *East Afr Med J.* 1989;66:589-593.
- Simpson H, Habel AH, George EL. Cerebrospinal fluid acid-base status and lactate and pyruvate concentrations after short (less than 30 minutes) first febrile convulsions in children. *Arch Dis Child.* 1977;52:836-843.
- Simpson H, Habel AH, George EL. Cerebrospinal fluid acid-base status and lactate and pyruvate concentrations after convulsions of varied duration and aetiology in children. *Arch Dis Child.* 1977;52:844-849.
- Zhang WM, Natowicz MR. Cerebrospinal fluid lactate and pyruvate concentrations and their ratio. *Clin Biochem.* 2013;46:694-697.
- King LJ, Lowry OH, Passonneau JV, et al. Effects of convulsants on energy reserves in the cerebral cortex. *J Neurochem.* 1967;14:599-611.
- Meldrum BS, Nilsson B. Cerebral blood flow and metabolic rate early and late in prolonged epileptic seizures induced in rats by bicuculline. *Brain.* 1976;99:523-542.
- Plum F, Posner JB, Troy B. Cerebral metabolic and circulatory responses to induced convulsions in animals. *Arch Neurol.* 1968;18:1-13.

34. Bain JA, Pollock GH. Normal and seizure levels of lactate, pyruvate and acid-soluble phosphates in the cerebellum and cerebrum. *Proc Soc Exp Biol Med.* 1949;71:495-497.
35. Bain JA, Pollock GH, Stein SN. Lactate, pyruvate, and acid-soluble phosphates in monkey brains treated with carbon dioxide and electric shock. *Proc Soc Exp Biol Med.* 1949;71:497.
36. Stone WE, Webster JE, Gurdjian ES. Chemical changes in teh cerebral cortex associated with convulsive activity. *J Neurophysiol.* 1945;8:233-240.
37. Bolwig TG, Quistorff B. In vivo concentration of lactate in the brain of consious rats beford and during seizures: a new ultra-rapid technique for the freeze-sampling of brain tissue. *J Neurochem.* 1973;21:1345-1348.
38. Schousboe A, Bak LK, Sickmann HM, et al. Energy substrates to support glutamatergic and GABAergic synaptic function: role of glycogen, glucose and lactate. *Neurotox Res.* 2007;12:263-268.
39. Waitt AE, Reed L, Ransom BR, et al. Emerging roles for glycogen in the CNS. *Front Mol Neurosci.* 2017;10:73.
40. Beresford HR, Posner JB, Plum F. Changes in brain lactate during induced cerebral seizures. *Arch Neurol.* 1969;20:243-248.
41. Oses JP, Muller AP, Strogulski NR, et al. Sustained elevation of cerebrospinal fluid glucose and lactate after a single seizure does not parallel with mitochondria energy production. *Epilepsy Res.* 2019;152:35-41.
42. Pellerin L, Magistretti PJ. Glutamate uptake into astrocytes stimulates aerobic glycolysis: a mechanism coupling neuronal activity to glucose utilization. *Proc Natl Acad Sci USA.* 1994;91:10625-10629.
43. Galan A, Seisdedos-Benzal A, Carletti BE, et al. Cisternal versus lumbar cerebrospinal fluid lactate concentration in healthy dogs. *Vet Med.* 2020;65:297-300.
44. Mellema LM, Koblik PD, Kortz GD, et al. Reversible magnetic resonance imaging abnormalities in dogs following seizures. *Vet Radiol Ultrasound.* 1999;40:588-595.
45. Smith PM, Talbot CE, Jeffery ND. Findings on low-field cranial MR images in epileptic dogs that lack interictal neurological deficits. *Vet J.* 2008;176:320-325.
46. Viitmaa R, Cizinauskas S, Bergamasco LA, et al. Magnetic resonance imaging findings in Finnish Spitz dogs with focal epilepsy. *J Vet Intern Med.* 2006;20:305-310.

SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of this article.

How to cite this article: Mariani CL, Nye CJ, Ruterbories L, et al. Cerebrospinal fluid lactate concentrations in dogs with seizure disorders. *J Vet Intern Med.* 2020;34:2562–2570.
<https://doi.org/10.1111/jvim.15953>