

## ORIGINAL ARTICLE

# Evaluation of cold atmospheric microwave plasma on skin physiological parameters and tolerability in dogs

Na-Eun Lee<sup>1</sup> | Yeong-Hun Kang<sup>1</sup> | Soon-Young Song<sup>1</sup> |  
Seung-Joon Baek<sup>2</sup> | Cheol-Yong Hwang<sup>1</sup>

<sup>1</sup>Laboratory of Veterinary Dermatology and the Research Institute for Veterinary Science, College of Veterinary Medicine, Seoul National University, Seoul, Korea

<sup>2</sup>Laboratory of Signal Transduction, College of Veterinary Medicine, Seoul National University, Seoul, Korea

**Correspondence**

Cheol-Yong Hwang, Laboratory of Veterinary Dermatology and the Research Institute for Veterinary Science, College of Veterinary Medicine, Seoul National University, Seoul 08826, Korea.  
Email: [cyhwang@snu.ac.kr](mailto:cyhwang@snu.ac.kr)

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**Abstract**

**Background:** Cold atmospheric microwave plasma (CAMP) is a promising therapeutic option for treating skin infections and wounds. Changes in biophysical skin parameters and the tolerability in dogs after applying CAMP is unknown.

**Objective:** This study aimed to evaluate the *in vivo* effects of CAMP on skin biophysical parameters [hydration, transepidermal water loss (TEWL) and surface temperature] and tolerability in dogs.

**Animals:** Twenty client-owned dogs with normal skin.

**Materials and methods:** Cold atmospheric microwave plasma treatment was performed for 30 s and 1, 2 and 4 min, respectively, at different sites of normal canine skin in the inguinal area. Hydration, TEWL and surface temperature were measured five, three and three times, respectively, before and after CAMP application. After treatment, pain and adverse effects were evaluated using a modified Melbourne Pain Scale and the modified short form Glasgow Composite Measure Pain Scale (modified CMPS-SF).

**Results:** Transepidermal water loss values significantly decreased with 4 min of treatment, and hydration decreased significantly with 2 min of treatment. Temperature increased significantly with increasing treatment time. For other parameters, no significant changes were observed. No significant pain response or adverse effects were observed in most dogs, aside from mild erythema in the treatment area after 4 min.

**Conclusion and clinical significance:** Cold atmospheric microwave plasma treatment was well-tolerated and did not significantly change canine skin biophysical parameters. CAMP achieves basic recommendations for safe use and is a potential therapeutic option for various skin diseases in dogs.

## INTRODUCTION

Plasma is generated when a gas is ionised by electric fields and is called the fourth state of matter (solid, liquid, gas and plasma).<sup>1,2</sup> Air-generated plasma contains a reactive mixture of atoms, reactive oxygen and nitrogen species, ultraviolet photons and charged particles, all of which can be modulated by adjusting the gas composition and gas output methods.<sup>2,3</sup> Cold atmospheric plasma (CAP) is of particular interest in medicine because reactive chemical processes can modify

DNA, proteins and cell membranes while treating heat-sensitive living tissue at low temperatures. CAP can be generated with relatively low gas pressure and electric power.<sup>4</sup> Furthermore, it has great potential for improving wound healing, treatment of various skin infections and tissue regeneration, making it widely applicable in human dermatological treatment.<sup>5</sup> The antibacterial effect of plasma is achieved by two mechanisms: an electrostatic field that penetrates and damages bacterial cell walls, and high oxidative stress that directly destroys bacterial DNA and protein.<sup>3,6</sup>

Different types of CAPs have been developed based on various power supply sources. Among them, previous studies have demonstrated that neither the atmospheric pressure plasma jet (APPJ) or the dielectric barrier discharge (DBD) method damage the skin barrier or cause pain or discomfort in humans.<sup>7</sup> The cold atmospheric microwave plasma (CAMP) used in this study was a 2.45 GHz microwave coaxial cavity resonator (CCR) streamer glow. This method is advantageous as it can effectively deliver more reactive oxygen species compared with other conventional CAP methods that do not use microwave power and has a strong sterilisation effect even at relatively low temperatures and power supply.<sup>8–11</sup> To the best of our knowledge, no study has evaluated the efficacy and safety of CAMP in dogs.

Several biophysical skin parameters such as transepidermal water loss (TEWL), hydration, erythema and pH are used to reflect skin condition.<sup>12</sup> The measurement of TEWL provides valuable information for assessing damage to skin barrier function and for estimating water retention. TEWL increases as moisture loss increases, and occurs with damage to the stratum corneum (SC), such as during exposure to irritating substances, self-excoriation or allergic dermatitis.<sup>13</sup> Skin hydration, measured using an electrical capacitance method, represents the moisture content of the SC which has been shown to affect the balance between epidermal cell proliferation and differentiation.<sup>14, 15</sup>

The present study aimed to demonstrate the safety and tolerability of CAMP as a new medical treatment tool in dogs, and to evaluate changes in skin physiological parameters.

## MATERIALS AND METHODS

### Ethics

Procedures used in this study were approved by the Institutional Animal Care and Use Committee of Seoul National University (SNU-IACUC, permit no. SNU-201224-1). Owners were informed of the entire

experimental process and signed consent-to-participate forms before treatments were carried out.

### Study population

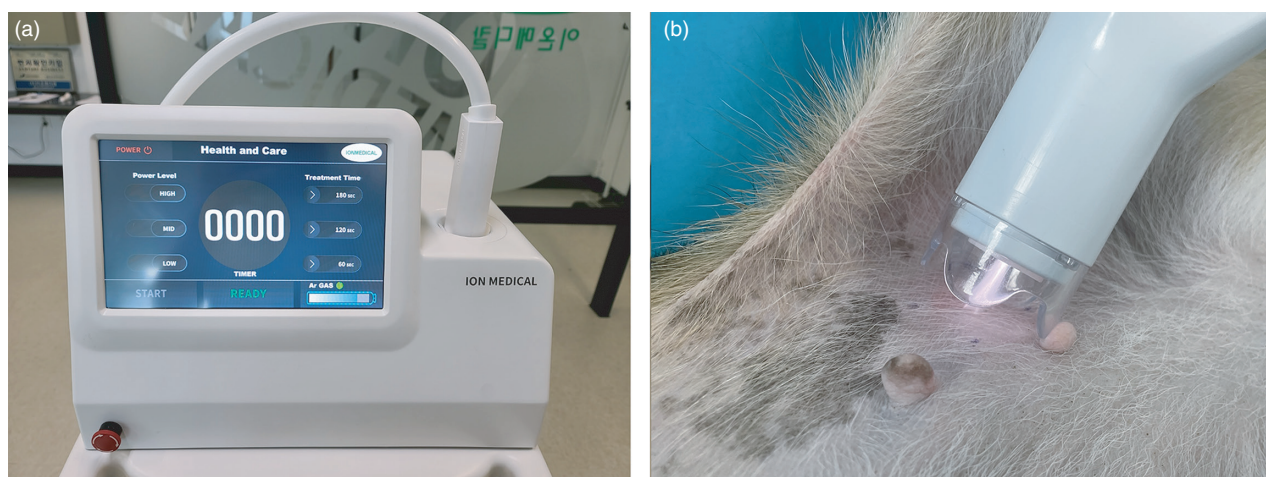
The owners of the dogs submitted a participation application to the Department of Dermatology, Seoul National University Veterinary Medical Teaching Hospital (SNU-VMTH), between April 2021 and July 2021. Clinically healthy dogs with no previous medical history or clinical signs of disease were recruited. All dogs underwent basic physical and dermatological examinations before measurements were performed. If skin lesions were found, the dogs were excluded. Dogs that had received any medications within four weeks before enrollment were excluded.

### Plasma device

A CAMP device (Bio Stimulation Microwave Plasma v1.0, IonMedical Inc.) was used in this experiment (Figure 1a). Using nonionised argon gas as a carrier gas, plasma was produced inside a cylinder by microwave energy application (30–50W, 2450MHz).<sup>9, 16, 17</sup> Subsequently, plasma was ignited and emitted through the nozzle and delivered to the target site. The plasma emission could be changed depending on the power consumption and gas velocity. The microwave energy could be adjusted to 30–50W, and the flow rate of the argon gas could be adjusted 10–20L/min. The length of the plasma jet also was influenced by the power consumption, varying between 3 and 15mm.<sup>16, 17</sup> At a gas flow rate of 15L/min and with a power consumption of 30W, the effluent had a visible length of 10mm and the surface temperature of the plasma was maintained at <40°C.

### Plasma treatment

The locations of the treatment sites are illustrated in Figure 2. Before treatment, the dogs were placed on



**FIGURE 1** Cold atmospheric microwave plasma (CAMP). (a) Photograph of the CAMP device. Plasma is released in a torch-like form, and output and treatment time can be altered. (b) Application of plasma on dog skin using CAMP

a cushion in dorsal recumbency, and assistants gently restrained the dogs to prevent excessive movement. Four 15 × 15 mm areas on the inguinal area were marked with a surgical pen.<sup>18</sup> CAMP was set at a gas flow rate of 15 L/min, with 2450 MHz, 3.5 kV microwave energy, and power consumption at 30 W. The tip of the CAMP handle was fixed above the skin surface at a distance equal to the length of the jet (10 mm), and was moved manually and repeatedly in a meandering pattern at an average velocity of approximately 10 mm/s.<sup>19</sup> Each treatment site was in contact with the plasma several times.<sup>20</sup> Treatments were performed for 30 s, 1, 2 and 4 min, respectively, in the inguinal area (Figure 1b).

## Assessment of skin physiological parameters

All dogs were acclimatised for ≥15 min before testing in an environmentally controlled room kept at 20–24°C with 40%–60% relative humidity (Table S1). Hair at the treatment sites was clipped using an electric clipper 5 min before CAMP treatment. The hair length was kept at 5 mm to prevent epidermal layer damage using the clippers. Skin physiological parameters were evaluated by the same dermatologist. Skin hydration, TEWL and skin surface temperature were measured five, three and three times, respectively, before and after CAMP irradiation. Skin hydration and TEWL measurements were obtained by lightly placing the probes perpendicular to the treatment sites. The distance of the thermometer sensor was 2–3 cm from the measuring point, as instructed in the device manual.<sup>21</sup> All measurements were performed by the same investigator, and all biophysical measurements were completed within 5 min after plasma treatment.<sup>22</sup> Readings were recorded manually and subsequently transferred to a

computerised spreadsheet. The mean and median of the measurement values were calculated.

## Measuring instruments

All devices were calibrated before the beginning of the study according to the manufacturers' instructions. TEWL was measured using an evaporimeter (VapoMeter SWL-3, Delfin Technologies Ltd) based on the guidelines provided previously<sup>23</sup> and the results were expressed as evaporation rate (g/m<sup>2</sup>/h).<sup>24</sup> Skin hydration was measured with a corneometer (MoistureMeterSC, Delfin Technologies Ltd) and expressed in arbitrary units (a.u.). The surface temperature (°C) was measured with a noncontact infrared thermometer (FS-300, HuBDIC Ltd).<sup>21</sup>

## Treatment tolerance

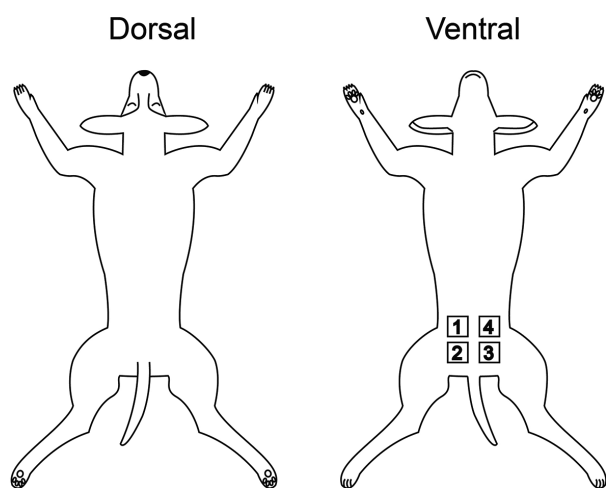
Pain was monitored using the modified Melbourne Pain Scale during or right after CAMP treatment.<sup>25</sup> We evaluated the dogs' pain on a multidimensional scale by measuring their behaviour (activity level, posture, vocalisation and reaction after palpation) and physiological response (cardiac and respiratory rate change). In addition, to evaluate the adverse effects, evaluation for erythema and pruritus in the treatment area were included. The score was calculated as the sum of scores for the eight individual categories. The possible score ranged from 0 to 24. More detailed evaluation criteria are presented in Table 1.

## Treatment tolerance as evaluated by owner

In order to assess for chronic pain and adverse effects from the treatment, owners were asked to evaluate their dog's behavioural changes with the modified short form of the Glasgow Composite Measure Pain Scale (modified CMPS-SF) 2 h after CAMP treatment.<sup>26</sup> Erythema and pruritus also were evaluated. The survey was conducted using an online survey platform (<https://office.naver.com>).

## Statistical analysis

All statistical analyses were performed under the independence assumption. Comparisons of measurements between CAMP treatments were made with Student's paired *t*-test or permutation test, as appropriate. Analysis of the mean difference over plasma application time was performed with the permutation test. Normality of data was determined using the Shapiro–Wilk test. All paired tests were one-sided tests. A *p*-value of <0.05 was considered statistically significant. The number of replications in the permutation test was fixed at 10,000. Statistical analyses were carried out using R v3.6.3 for Linux [R Core Team (2021)<sup>27</sup>]. The



**FIGURE 2** Schematic figure of treatment sites measured for transepidermal water loss (TEWL), skin hydration and skin surface temperature. Skin barrier function and body surface temperature were evaluated in the inguinal area before and after plasma application. Each test area was divided into 15 × 15 mm<sup>2</sup> separated by a surgical pen. Cold atmospheric microwave plasma (CAMP) irradiation was applied for 30 s, 1, 2 and 4 min to areas 1, 2, 3 and 4, respectively

**TABLE 1** Modified University of Melbourne Pain Scale for assessing pain and side effects in dogs during or after cold atmospheric microwave plasma (CAMP) treatment

Variable	Criteria	Score	Results
Heartbeat change (After treatment)	<20% increase	0	19/20 (95%)
	20% to ≤50% increase	1	1/20 (5%)
	50% to ≤100% increase	2	0/20 (0%)
	100% increase	3	0/20 (0%)
Respiratory rate change (After treatment)	<20% increase	0	11/20 (55%)
	20% to ≤50% increase	1	2/20 (10%)
	50% to ≤100% increase	2	3/20 (15%)
	100% increase	3	4/20 (20%)
Activity level (During treatment)	Resting or asleep	0	10/20 (50%)
	Awake	1	9/20 (45%)
	Restless	2	1/20 (5%)
	Rolling or thrashing	3	0/20 (0%)
Posture (After treatment)	Sleeping or calm	0	7/20 (35%)
	Sternal or sitting up	1	13/20 (65%)
	standing up with head down	2	0/20 (0%)
	guarded posture	3	0/20 (0%)
Vocalisation (During treatment)	None	0	16/20 (80%)
	During palpation of the surgical site	1	0/20 (0%)
	Intermittent (<25% of total time)	2	4/20 (20%)
	Continuous (>25% of total time)	3	0/20 (0%)
Erythema (During and after treatment)	None	0	10/20 (50%)
	Partial mild erythema of treatment site	1	9/20 (45%)
	Partial moderate erythema of treatment site	2	1/20 (5%)
	Entire moderate erythema of treatment site	3	0/20 (0%)
Reaction after palpation (After treatment)	No response	0	19/20 (95%)
	Turn head	1	1/20 (5%)
	Guarded position	2	0/20 (0%)
	Cries out or aggressive behaviour	3	0/20 (0%)
Pruritus (After treatment)	No response	0	20/20 (100%)
	Licking or scratching after palpation	1	0/20 (0%)
	Intermittent licking or scratching	2	0/20 (0%)
	Continuous licking or scratching	3	0/20 (0%)

Evaluation criteria for pruritus and erythema were added to the conventional Melbourne Pain Scale. Pain score was calculated as the sum of scores for the eight individual categories, with possible scores ranging from 0 to 24.

*t*- and Shapiro–Wilk tests were performed in R/STATS, and the permutation test was performed in R/WPERM v1.0.1.

## RESULTS

### Study population

Twenty client-owned dogs were recruited, including nine females (seven spayed), and 11 males (all castrated), with ages ranging from 6 months to 12 years old (mean age 4.67 years). The breed population included seven mixed breed dogs, four miniature poodles, three bichon frises, and one each of dachshund, Chihuahua, Yorkshire terrier, shih tzu, beagle and Maltese.

### Transepidermal water loss changes

Transepidermal water loss showed no significant increase until 2 min of CAMP treatment [11.51 to 11.91 g/m<sup>2</sup>/h at 30 s (*p* = 0.245), 14.21 to 13.41 g/m<sup>2</sup>/h (*p* = 0.892) at 1 min, and 13.63 to 13.39 g/m<sup>2</sup>/h (*p* = 0.566) at 2 min] (Figure 3a). The mean TEWL significantly decreased from 15.02 to 10.97 g/m<sup>2</sup>/h at 4 min of treatment. The differences in TEWL were compared among the four measurement sites. No significant differences were observed up to 2 min of treatment (*p* > 0.05), yet significant differences were observed at 4 min application (*p* < 0.05). In addition, the TEWL values were compared among the different plasma application times, and no significant time-dependent differences were found (*p* > 0.05).

## Skin hydration changes

No significant decrease in hydration was observed at 30 s (12.96 to 12.54 a.u.;  $p = 0.38$ ), 1 min (15.84 to 13.85;  $p = 0.1184$ ) or 4 min (13.51 to 12.39;  $p = 0.1928$ ) (Figure 3b). However, 2 min of CAMP treatment resulted in a significant decrease in hydration from 14.55 to 11.70 ( $p = 0.006$ ). Significant differences in hydration levels among treatment sites ( $p > 0.05$ ) and plasma application times ( $p > 0.05$ ) were not observed.

## Skin surface temperature changes

Skin surface temperature at the treatment site did not increase significantly until 2 min of treatment (Figure 3c). CAMP treatment for 4 min produced a significant increase in temperature from 36.72 to 36.88°C ( $p = 0.011$ ). The differences in skin surface temperature by treatment site were not significant ( $p > 0.05$ ). Skin surface temperature increased with increasing plasma application time ( $p < 0.05$ ).

## Plasma tolerance and adverse effects

### Plasma tolerance after treatment

Outcome evaluations performed immediately after the treatment are described in Table 1. For physiological pain indicators, nine of 20 dogs (45%) showed increased respiratory rates and two of 20 dogs (10%) showed increased heart rates. The most observed indicator of change was erythema, with 10 of 20 dogs (50%) showing partial mild erythema and one of 20 dogs (5%) showing partial moderate erythema. All 11 dogs that showed erythema did so only at 4 min of treatment. Most dogs were asleep or awake during the procedure and did not show movement, such as struggling. None of the dogs showed pain-induced behaviour after the plasma treatment. Some dogs (four of 20, 20%) whined at the end of the procedure, and one of 20 dogs (5%) turned its head immediately after palpation of the treatment site. None of the dogs showed signs of pruritus. There was no significant difference in terms of tolerability among the different breeds.

### Plasma tolerance after 2 h of treatment

Based on the owner's survey responses, two of 20 dogs (10%) showed licking or scratching behaviour after palpation. Three of 20 dogs (15%) showed partial mild erythema of the treatment site. Two of 20 dogs (10%) glanced at the treatment site after gentle pressure was applied. No unfavourable changes were identified for the rest of the indicators, such as overall condition, posture and activity.

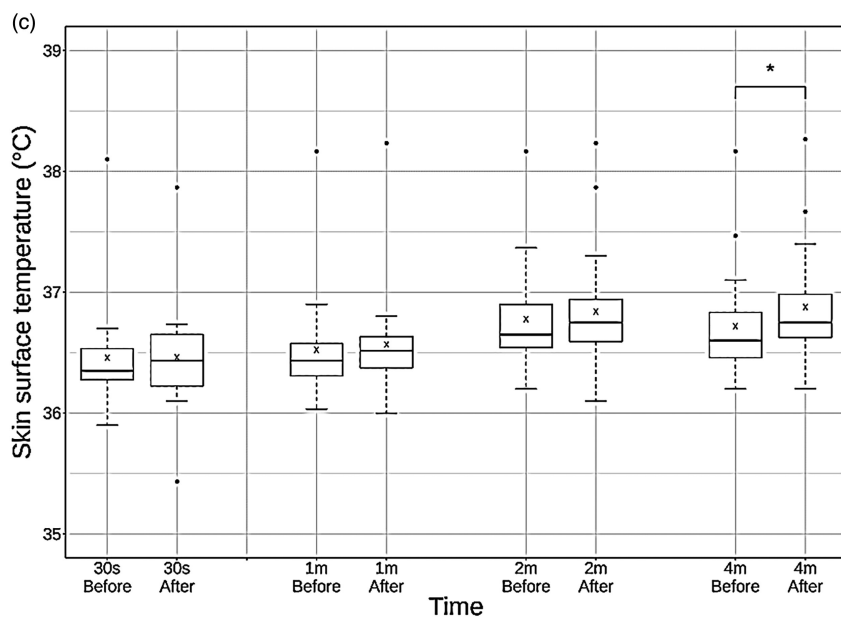
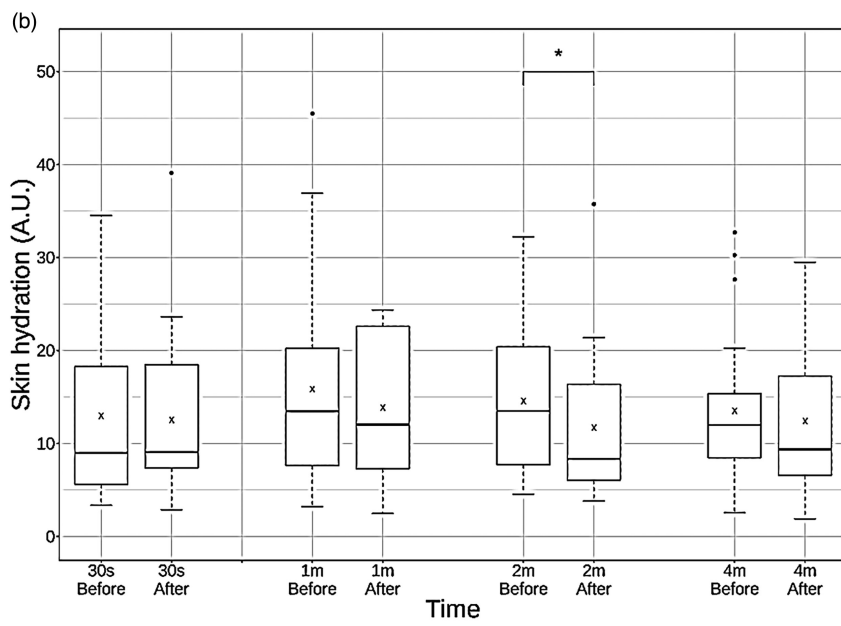
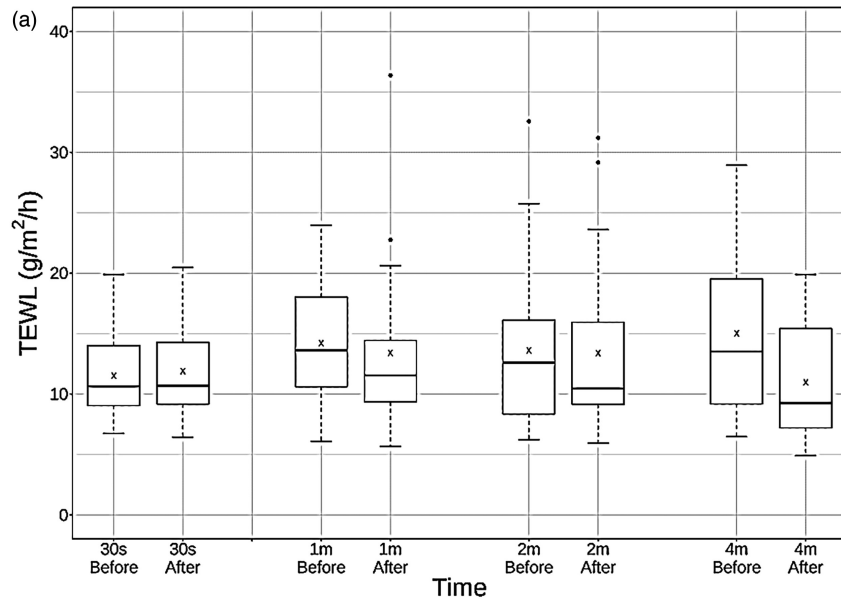
## DISCUSSION

The current study measured changes in skin physiological parameters, and evaluated patient tolerance and adverse effects of CAMP treatment in healthy dogs. A significant decrease in TEWL was observed after 4 min of treatment, contrary to the assumption that TEWL would increase as a result of the heat and gas flow emitted by CAMP.<sup>22, 28</sup> The observed decrease in TEWL presumably was a result of variations between the measurement sites, as noted in a previous study.<sup>29</sup> Because measurements were obtained at different sections of the inguinal area to prevent cumulative effects of CAMP, deviations between the measurement sites may have occurred.

No significant differences were observed in hydration and surface temperature among measurement sites. In the case of skin hydration, a slight decrease occurred only with 2 min of treatment. This is consistent with previous results reported in human medicine, which showed temporary moisture loss in the SC immediately after CAP exposure.<sup>22</sup> In subsequent longer treatments, skin hydration increased and helped restore skin barrier function.<sup>28, 30</sup> No significant differences were observed for TEWL and skin hydration, indicating no time-dependent changes in skin barrier function after plasma application.

Increased surface temperature was observed with increasing CAMP treatment time. Because the measurements were made in order of skin hydration, TEWL, then temperature, there was a time interval of approximately 3 min between plasma treatment and temperature measurement. This interval makes it difficult to know the temperature immediately after CAMP application and how rapidly it might have changed. The antimicrobial effects of CAMP were demonstrated previously<sup>16</sup> in a shorter time period and a larger area (<60 s, 9.6 cm<sup>2</sup>) than used herein (<4 min, 2.25 cm<sup>2</sup>). Therefore, it appears that with a short application time per area, simultaneously obtaining sufficient antimicrobial effects and preventing thermal damage is possible.

Assessing pain in animals has become more important in veterinary medicine, yet sensitive, objective and specialised criteria to measure the degree of pain in animals are difficult to determine.<sup>31</sup> Many of the pain scoring systems used in veterinary medicine have focused on acute pain after surgery. We modified the Melbourne Pain Scale by adding criteria for erythema and pruritus. CAMP treatment did not cause much discomfort, as no significant changes were found in pain indicators. Interestingly, 11 of 20 dogs showed erythema only in sites exposed to CAMP for 4 min. Given the increase in skin surface temperature and erythema only in the 4 min treatment area, thermal damage is expected to occur when applied for 4 min to a relatively small area. In most cases, the respiratory rate increased during the change of posture after the experiment, and not during the experiment. Thus, we assume no significant association between CAMP application and pain response.



**FIGURE 3** Skin barrier function and skin temperature changes in dogs before and after cold atmospheric microwave plasma (CAMP) treatment. Changes in (a) transepidermal water loss (TEWL), (b) skin hydration, and (c) skin surface temperature before and after the application of CAMP on the skin at exposure times of 30 s, 1, 2 and 4 min. (a) TEWL decreased significantly with 4 min of CAMP exposure, and did not show significant changes with 30 s, 1 and 2 min of treatment. (b) Skin hydration decreased significantly with 2 min of CAMP exposure, and not with 30 s, 1 and 4 min of treatment. (c) Skin surface temperature increased significantly with 4 min of CAMP exposure, and not with 30 s, 1 and 2 min of treatment. The values are expressed as boxplots: middle horizontal line, median value; x, mean value; n = 20

Finally, we assessed long-term adverse effects of CAMP application. Based on the online survey responses of owners 2 h after CAMP treatment, no complications were observed except in one dog. This dog immediately showed erythema in the area treated for 4 min. The day after the treatment, erythema and licking behaviour were observed, resulting in a prescription for topical steroid ointment. These symptoms are believed to have been caused by thermal damage, and further study by laser microscopy or histological analysis<sup>32</sup> is needed to determine whether the actual skin surface structure is damaged by this duration of therapy.

Significant advantages of CAP treatment are that it is painless, noninvasive, and less likely to cause antimicrobial resistance or toxic reactions than other therapies.<sup>33</sup> Despite the well-investigated decontamination effects of CAP in human medicine, few papers have been published on the applications of CAP in veterinary medicine. A previous study has proven its antibacterial activity against bacterial strains frequently encountered in canine bite wounds *in vitro*.<sup>20</sup> In addition, a study confirmed the considerable antibacterial and antibiofilm effect of CAMP against bacteria frequently associated with pyoderma and otitis externa in dogs.<sup>16, 17</sup>

There are several advantages of using CAMP compared with previous commercial CAP methods, which use discharge sources such as radio frequency (RF) or dielectric barrier discharge (DBD). DBD sources need relatively high voltages and deliver low time-averaged plasma densities.<sup>34</sup> Besides, RF sources require relatively lower voltages but require detailed control to prevent transitions between operating modes.<sup>4</sup> By contrast, CAMP device only requires a power supply such as that commonly used for a microwave oven (2.45 GHz). It does not require electrodes to operate and is easy to handle, as particles are released in a torch-like manner.<sup>34</sup> Furthermore, it effectively produces a wider variety of reactive species compared with other CAP devices, thereby causing a significant sterilisation effect.<sup>8, 35</sup>

This study has several limitations. First, because this study was conducted on normal skin, tolerance of multiple daily procedures on larger areas of diseased skin cannot be predicted. The investigation of plasma tolerance should be pursued in clinical situations such as skin infections or wounds. Second, we fixed the output and the moving velocity of CAMP, yet there may be more clinically appropriate outputs and application times per unit area that will need to be established.

## CONCLUSIONS

The present study demonstrated that CAMP could be a safe and well-tolerated therapeutic option in dogs. Because the antibacterial efficacy *in vitro* and safety *in vivo* have been confirmed, future evaluations of

the clinical efficacy of CAMP in treating various skin conditions, such as infections and wounds, should be conducted.

## ACKNOWLEDGMENTS

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## CONFLICT OF INTEREST

Cheol-Yong Hwang and Seung-Joon Baek are co-founders at Ion Medical Inc.

## AUTHOR CONTRIBUTIONS

**Na-Eun Lee:** Conceptualization; data curation; methodology; visualization; writing – original draft. **Yeong-Hun Kang:** Investigation. **Soon-Young Song:** Investigation. **Seung-Joon Baek:** Investigation; methodology. **Cheol-Yong Hwang:** Conceptualization; methodology; project administration; supervision; writing – review and editing.

## ORCID

Na-Eun Lee  <https://orcid.org/0000-0001-9295-1249>  
 Yeong-Hun Kang  <https://orcid.org/0000-0001-9524-024X>  
 Seung-Joon Baek  <https://orcid.org/0000-0001-7866-7778>  
 Cheol-Yong Hwang  <https://orcid.org/0000-0001-7113-0361>

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## SUPPORTING INFORMATION

Additional supporting information may be found in the online version of the article at the publisher's website.

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## Résumé

**Contexte** - Le CAMP (Cold Atmospheric Microwave Plasma) est une option thérapeutique prometteuse pour le traitement des infections cutanées et des plaies. Les modifications des paramètres biophysiques de la peau et la tolérance chez les chiens après l'application de CAMP sont inconnues.

**Objectif** - Cette étude visait à évaluer les effets *in vivo* du CAMP sur les paramètres biophysiques de la peau [hydratation, perte d'eau transépidermique (TEWL) et température de surface] et la tolérance chez le chien.

**Animaux** - Vingt chiens de propriétaires à peau normale.

**Matériels et méthodes** - Le traitement CAMP a été effectué pendant 30 s et 1, 2 et 4 min, respectivement, sur différents sites de peau canine normale dans la région inguinale. L'hydratation, la TEWL et la température de surface ont été mesurées cinq, trois et trois fois, respectivement, avant et après l'application de CAMP. Après le traitement, la douleur et les effets indésirables ont été évalués à l'aide d'une échelle de douleur de Melbourne modifiée et de la forme courte modifiée de l'échelle de mesure de la douleur composite de Glasgow (CMPS-SF modifiée).

**Résultats** - Les valeurs de TEWL ont diminué de manière significative après 4 minutes de traitement et l'hydratation a diminué de manière significative après 2 minutes de traitement. La température a augmenté de manière significative avec l'augmentation du temps de traitement. Pour les autres paramètres, aucun changement significatif n'a été observé. Aucune réponse significative à la douleur ni aucun effet indésirable n'ont été observés chez la plupart des chiens, à l'exception d'un léger érythème dans la zone de traitement après 4 minutes.

**Conclusion et signification clinique** - Le traitement CAMP a été bien toléré et n'a pas modifié de manière significative les paramètres biophysiques de la peau canine. CAMP répond aux recommandations de base pour une utilisation sûre et constitue une option thérapeutique potentielle pour diverses maladies de la peau chez les chiens.

## Resumen

**Introducción**- el plasma de microondas atmosférico frío (CAMP) es una opción terapéutica prometedora para el tratamiento de infecciones y heridas de la piel. Se desconocen los cambios en los parámetros biofísicos de la piel y la tolerabilidad en perros después de aplicar CAMP.

**Objetivo**- este estudio tuvo como objetivo evaluar los efectos *in vivo* de CAMP en los parámetros biofísicos de la piel [hidratación, pérdida de agua transepidermica (TEWL) y temperatura superficial] y la tolerabilidad en perros.

**Animales** - Veinte perros de propietarios particulares con piel normal.

**Materiales y métodos** - El tratamiento CAMP se realizó durante 30 s y 1, 2 y 4 min, respectivamente, en diferentes sitios de piel canina normal en el área inguinal. La hidratación, el TEWL y la temperatura superficial se midieron cinco, tres y tres veces, respectivamente, antes y después de la aplicación de CAMP. Después del tratamiento, el dolor y los efectos adversos se evaluaron mediante una escala de dolor de Melbourne modificada y la escala de dolor de medida compuesta de Glasgow de forma abreviada modificada (CMPS-SF modificada).

**Resultados**- los valores de TEWL disminuyeron significativamente con 4 min de tratamiento y la hidratación disminuyó significativamente con 2 min de tratamiento. La temperatura aumentó significativamente con el aumento del tiempo de tratamiento. Para otros parámetros no se observaron cambios significativos. En la mayoría de los perros no se observaron reacciones significativas de dolor ni efectos adversos, aparte de un leve eritema en el área de tratamiento después de 4 min.

**Conclusión y significado clínico**- el tratamiento con CAMP fue bien tolerado y no cambió significativamente los parámetros biofísicos de la piel canina. CAMP obtuvo recomendaciones básicas para un uso seguro y es una opción terapéutica potencial para diversas enfermedades de la piel en perros.

## Zusammenfassung

**Hintergrund** - Kaltes atmosphärisches Mikrowellen Plasma (CAMP) ist eine vielversprechende therapeutische Option zur Behandlung von Hautinfektionen und Wunden. Veränderungen der biophysischen Hautparameter und der Toleranz bei Hunden nach Anwendung von CAMP ist bisher nicht bekannt.

**Ziel** - Diese Studie zielte darauf ab, die *in vivo* Wirksamkeit von CAMP auf biophysische Parameter der Haut [Hydratation, Transepidermaler Wasserverlust (TEWL) und die Oberflächentemperatur] und die Verträglichkeit bei Hunden zu evaluieren.

**Tiere** - Es wurden zwanzig Hunde in Privatbesitz, alle mit normaler Haut, inkludiert.

**Materialien und Methoden** - Eine CAMP Behandlung wurde für 30 s und 1, 2 bzw 4 Minuten an verschiedenen Stellen normaler Hundehaut in der Inguinalgegend durchgeführt. Hydratation, TEWL und die Oberflächentemperatur wurde fünf, drei bzw dreimal vor und nach der CAMP Anwendung gemessen. Nach der Behandlung wurden Schmerz und Nebenwirkungen mittels modifizierter Melbourne Pain Scale sowie der modifizierten Kurzform der Glasgow Measure Pain Scale (modifizierte CMPS-SF) evaluiert.

**Ergebnisse** - Die TEWL Werte nahmen nach 4 Minuten der Behandlung, die Hydratation nach 2 Behandlungsminuten signifikant ab. Die Temperatur nahm mit zunehmender Behandlungsdauer signifikant zu. Für die anderen Parameter konnten keine signifikanten Veränderungen beobachtet werden. Es wurden bei den meisten Hunden

keine signifikanten Schmerzreaktionen oder Nebenwirkungen beobachtet, außer einem geringen Erythem nach 4 Minuten in der behandelten Körperregion.

**Schlussfolgerungen und klinische Bedeutung** – Die Behandlung mit CAMP wurde gut toleriert und veränderte die biophysischen Parameter der Hundehaut nicht signifikant. Daher kann die CAMP grundsätzlich zur sicheren Anwendung bei Hunden empfohlen werden und stellt eine potenzielle Therapieoption für verschiedene Hauterkrankungen bei Hunden dar.

## 要約

**背景** – 低音大気圧マイクロ波プラズマ (CAMP) は、皮膚の感染症や創傷の治療法として有望である。イヌにCAMPを適用した場合の皮膚の生物物理学的パラメータの変化と忍容性は不明である。

**目的** -本研究は、犬の皮膚の生物物理学的パラメータ (水和性、経皮水分蒸散量 (TEWL)、表面温度) および忍容性に及ぼすCAMPの *in vivo* 効果を評価することを目的とした。

**供試動物** - 正常な皮膚を持つオーナー所有犬20頭。

**材料と方法** – 正常なイヌの鼠径部皮膚の異なる部位に、CAMP処理を30秒、1分、2分、4分それぞれ実施した。水和性、TEWL、表面温度はCAMP塗布の前後にそれぞれ5回、3回、3回測定した。治療後、修正メルボルン疼痛尺度及び修正短形式グラスゴー複合測定疼痛尺度 (修正CMPS-SF) を用いて、疼痛および副作用を評価した。

**結果** - TEWL値は4分間の治療で有意に減少し、水和性は2分間の治療で有意に減少した。表面温度は治療時間が長くなるにつれて有意に上昇した。その他のパラメータについては、有意な変化は認められなかった。4分後の治療部位の軽度の紅斑を除けば、ほとんどの犬で有意な疼痛反応や副作用は観察されなかった。

**結論と臨床的意義** – CAMP治療は忍容性が高く、犬の皮膚の生物物理学的パラメータに有意な変化を与えなかった。CAMPは安全使用のための基本的な推奨事項を満たしており、犬の様々な皮膚疾患に対する潜在的な治療オプションであると考えられる。

## 摘要

**背景**-冷大気微波等离子体(CAMP)是治疗皮肤感染和伤口的一种有前景的治疗选择。尚不清楚应用CAMP后犬的生物物理皮肤参数变化和耐受性。

**目的**-本研究旨在评价CAMP对犬皮肤生物物理参数[水合作用、经表皮水分流失 (TEWL)和表面温度]和耐受性的体内影响。

**动物**-20只皮肤正常的私家犬。

**材料与方方法**-在腹股沟区正常犬皮肤不同部位分别进行CAMP治疗30s和1、2和4 min。应用CAMP前后分别测量5次、3次和3次水化、TEWL和表面温度。治疗后采用改良墨尔本疼痛量表和改良简明格拉斯哥综合测量疼痛量表(改良CMPS-SF)评价疼痛和不良反应。

**结果**-治疗4 min后,TEWL值显著下降,治疗2 min后,水合作用显著下降。随着治疗时间的增加,温度显著增加。对于其他参数,未观察到显著变化。除4 min后治疗区域出现轻度红斑外,在大多数犬中未观察到显著的疼痛反应或不良反应。

**结论和临床意义**-CAMP治疗耐受性良好,未显著改变犬皮肤生物物理参数。CAMP实现了安全使用的基本建议,是犬各种皮肤病的潜在治疗选择

## Resumo

**Contexto** – O plasma frio atmosférico de micro-ondas (CAMP) é uma opção terapêutica promissora para o tratamento de infecções cutâneas e feridas. Não se sabe a respeito das alterações nos parâmetros biofísicos da pele e a tolerabilidade de cães após a aplicação de CAMP.

**Objetivo** – Este estudo tem como objetivo avaliar os efeitos *in vivo* de CAMP nos parâmetros biofísicos da pele [hidratação, perda de água transepidermica (TEWL) e temperatura da superfície] e a tolerabilidade em cães.

**Materiais e métodos** – O tratamento com CAMP foi realizado por 30s e 1, 2 e 4 min, respectivamente, em diferentes locais da pele canina normal na região inguinal. Hidratação, TEWL e temperatura da superfície foram medidas cinco, três e três vezes, respectivamente, antes e após a aplicação do CAMP. Após o tratamento, a dor e os efeitos adversos foram avaliados usando uma escala de dor de Melbourne modificada e a escala de medida composta de dor de Glasgow modificada (CMPS-SF modificada).

**Resultados** – Os valores de TEWL reduziram significativamente com o tratamento de 4 min, e a hidratação reduziu significativamente com dois minutos de tratamento. A temperatura aumentou significativamente com o aumento do tempo de tratamento. Não foram observadas alterações significativas para outros parâmetros. Não se observou

uma resposta de dor significativa ou efeitos adversos na maioria dos cães, além de eritema leve na área tratada após 4 min.

**Conclusão e significância clínica** – O tratamento com CAMP foi bem tolerado e não alterou significativamente os parâmetros biofísicos da pele canina. CAMP requer recomendações básicas de segurança na sua utilização e é uma opção terapêutica potencial para várias dermatopatias em cães.