

Clinical Studies

A Randomized, Blinded and Controlled Study Using Digital Thermal Imaging to Measure Temperature Change Associated with Acupuncture in Dogs with Back Pain

Patricia J. Collins DVM, MS-TCVM

ABSTRACT

Digital thermal imaging provides an objective and measurable evaluation of the changes in radiant energy emitted by the body. This study sought to determine the effects of acupuncture immediately after back pain treatment in dogs through digital thermal imaging. Dogs ≥ 6 months exhibiting clinical symptoms of back pain with pain scale 1 or 2 (0-4 scale) were recruited for the study. Subjects ($n=24$) were randomly assigned to the Acupuncture Group ($n=12$) which received dry needle acupuncture at GV-14, BL-23 bilateral, *Bai-hui* and *Shen-shu* for 15 minutes or the Control Group ($n=12$) which was not treated but waited 15 minutes. Digital thermal images were obtained before and after the acupuncture treatment (Acupuncture Group) or the waiting period (Control Group) by a person blinded to the group assignments. Maximal temperature was recorded within the affected surface area and absolute change of temperature was compared between study groups. Group comparison of subject signalment data, baseline pain score, and baseline (pre-treatment) temperature (all p -values > 0.05) suggested group comparability for the study. The mean \pm SD temperature change in the Acupuncture Group was $1.60\pm0.51^{\circ}\text{F}$, and in the Control group was $0.44\pm0.26^{\circ}\text{F}$. The overall temperature change difference between the two subject groups was statistically significant ($p = 2.96\times10^{-6}$). The results from this study indicate that local acupuncture treatment at a site of pain can lead to temperature changes in the location which may have effects on pain reduction. Future studies to investigate the association of acupuncture created temperature change and pain mitigation are warranted.

Keywords: acupuncture, digital thermal imaging, dogs, back pain

*Address correspondence to Dr. Patricia J. Collins (pcollins@vcah.org).

ABBREVIATIONS

EAP	Electro-acupuncture
DNAP	Dry needle acupuncture
IVDD	Intervertebral disc disease
TCVM	Traditional Chinese veterinary medicine

Pain is a subjective sensation that should be assessed by the individual, but dogs cannot provide this information verbally. Evaluation of chronic pain in dogs, therefore, is left to nonverbal observations such as gait abnormalities, behavior or attitude changes, radiographs and plasma hormone assays.¹ Each of these can be helpful distinguishing differences in pain levels of dogs but are not consistent due to individual variation. To complicate things further, dogs also exhibit breed specific and

training related differences in their response to pain.²

A study conducted by Hiem-Bjorkman demonstrated that chronic pain can be assessed better by thoughtful completion of a well-designed questionnaire by a pet owner than by reviewing radiographs or hormone assays alone.¹ Recognition of pain in dogs, therefore, can start with the patient's history and asking leading questions regarding subtle behavior patterns.³ Owner questionnaires can be detailed and may include: appetite, mood, interaction with family members, tail wagging, pacing, willingness to play/walk, ease of laying down/rising, negotiating stairs, excessive panting, lip licking, vocalization, aggression and different reactions to being touched. Even though subjective, the behavior assessment can then have a numeric pain scale assigned to questionnaire interpretation which improves standardization of the evaluation. The physical exam collects objective data such as temperature, heart rate, respiratory rate, pupil dilation, painful/tense areas and blood pressure but even these parameters are subject to variable dog responses to hospital or clinic environment.

From: Valley Cottage Animal Hospital, Valley Cottage, NY, USA

Author Professional Certifications: CVA, CVCH, CVFT, CVTP

It is also important to note that, while palpating painful areas, some dogs can be stoic which confounds pain recognition.

Today's digital thermal imaging innovations provide veterinarians with an efficient tool to monitor body function and injuries. Digital thermal imaging provides an objective and measurable evaluation of a patient's physiological state by documenting the surface temperature of the individual. Disease processes and tissue injury show different temperatures due to a change in blood flow. The metabolic heat from an animal is directly related to blood circulation. Increases in the body surface temperature are a result of increased blood flow due to inflammation, increased use or a result of injury, whereas decreases in body surface temperatures are a result of reduced blood flow due to nerve damage, atrophy, scarring, thrombosis or infarction.⁴

A study by Tunley and Henson showed that thermographic patterns are reproducible and topographical thermographic maps can be compared to pathological cases.⁵ The technique has also been shown to be useful for the diagnosis of cranial cruciate ligament disease in dogs without stifle laxity.⁶ Another study found thermal imaging to be a useful tool to diagnose pain in cats, which can be difficult and often goes undetected.⁷ The study, with a total of 103 cats, demonstrated that palpation of painful conditions and thermographic imaging correlated relatively well. The agreement between the owner's assessment of pain and thermographic imaging was low; however the agreement between palpation and thermographic imaging was moderate, making thermographic imaging a potential tool in clinical practice for screening cats for pain.

Acupuncture is an effective, safe, and minimally invasive treatment for acute and chronic pain control in companion animals which is steadily gaining popularity among veterinarians and pet owners.⁸ In traditional Chinese veterinary medicine (TCVM), *Qi* flow, or the flow of vital energy is the essence of health. When this *Qi* flow is obstructed, pain results. Acupuncture provides pain relief by restoring *Qi* and Blood flow thereby reducing painful Stagnation. Functional magnetic resonance imaging (MRI) studies investigating a conventional mechanism of action for pain relief associated with acupuncture treatment have shown that

stimulation of acupuncture points results in specific changes in the central nervous system such as the release of endogenous opioids.^{9,10} This is further supported by a study in equine colic which demonstrated that dry needle (DNAP) acupuncture and electroacupuncture (EAP) was associated with endorphin release which relieved clinical symptoms in affected horses.¹¹

The objective of this study was to measure thoracolumbar sacral skin temperature change associated with acupuncture treatment of canine back pain using digital thermal imaging. The hypothesis was that dogs treated with dry needle acupuncture would have greater back temperature change than the untreated controls.

MATERIALS AND METHODS

The study candidate population consisted of dogs exhibiting clinical signs of back pain. The presence of back pain was determined by a history of reluctance to exercise, stiffness, abnormal gait and/or pain exhibited during physical examination. In addition, some study subjects had previous radiographs showing spondylosis or narrowed disk spaces. The back pain of each individual study subject was graded according to the canine pain scale from 0-4, as classified by the Colorado State University Veterinary Medical Center (Table 1).¹² Dogs were recruited from the hospital population of client and staff owned dogs of the Valley Cottage Animal Hospital in Valley Cottage, New York, USA. Inclusion criteria were dogs (1) 6 months or older; (2) symptoms of mild back pain (pain score 1 or 2, scoring = 0 to 4) due to arthritis, disk disease, trauma or sports injuries; and (3) informed consent for study participation provided by owner. Subjects with any known cancer or infectious disease were excluded from the study.

A subject's baseline pain condition was considered a likely confounder to study outcome, therefore, to have good comparability between groups; a baseline pain score (grade 1 or 2 required) for each potential subject was assessed during the physical exam before the study. Qualified dogs were randomly assigned to one of two treatment groups: the Acupuncture Group or Control Group. The randomization assignment was conducted through an online tool^a using the block randomization method that would ensure equal sample sizes between study groups. For each subject in the Acupuncture Group,

Table 1: Canine pain scale used to grade severity of back pain in study dogs.¹²

Score	Conditions
0	Comfortable, happy and content
1	Content, but slightly unsettled with mild body tension
2	Uncomfortable with mild to moderate body tension
3	Unsettled, crying and guarding/flinching upon palpation
4	Constantly groaning/crying with moderate to severe body tension and extreme resistance to being touched

a baseline digital thermal image^b of the dog's back was obtained (Figure 1). The subject then received a dry needle acupuncture treatment (performed by author, certified in veterinary acupuncture) at GV-14, BL-23 (bilateral), *Bai-hui* and *Shen-shu* (bilateral). The needles (0.22×25 mm)^c were left in place without any further stimulation for 15 minutes, while the patient was allowed to move freely in a 10 by 12-foot exam room. Any needles that fell out were not replaced. After the 15 minute acupuncture treatment, the needles were removed and a second digital thermal image of the back was obtained. The dogs in the Control Group also underwent baseline digital thermal imaging of the back, waited for 15 minutes in the same exam room and then had a second digital thermal image obtained. All digital thermal imaging was performed by an investigator blinded to the group assignments.

To ensure consistency of thermal camera imaging, study dogs had limited exercise and were not bathed or otherwise submerged in water for a 24 hour period prior to treatment. All thermal images were taken in a room with a temperature range of 68 - 74°F and dogs were only minimally handled by trained technicians. In addition, the areas of the dogs' body to be analyzed were not touched, so instead of using black cloth gloves, the technicians were able to use the more hygienic one time use rubber gloves.

The anatomic region of the back to be imaged was defined as mid-thoracic to base of tail area. Two digital thermal images pre- and post-treatment were obtained per study participant. From each image, the average

temperature recorded within the selected surface area was used as the numerical data from each thermal temperature assessment (rectangle in image, Figure 1). The outcome measurement for each subject was the change of average temperature on the back from baseline to the time when the second thermal image was taken. The absolute value of the temperature change (increase or decrease) was used for group comparison since based on TCVM theory, an acupuncture treatment could modulate the body temperature at an injury site in either direction depending on the underlying cause of pain.

The study hypothesized dogs with symptoms of back pain who received a dry needle acupuncture treatment at protocol stipulated acupuncture points would have greater temperature change on the back compared to those who did not receive treatment. Without assuming a normal distribution of the temperature change data, two-sided nonparametric Wilcoxon rank sum test was used to test the hypothesis. The null hypothesis was rejected when the resulting *p*-value was less than 0.05. A study enrollment goal of 24 dogs with a sample size of 12 subjects in each group was sought to ensure that the applied Wilcoxon rank sum test had approximately 91% power to reject the null hypothesis with a significance level of 0.05. These parameters could be assumed under the condition that the probability that a subject in the Acupuncture Group has a larger temperature change than a subject in the Control Group was 85%. A commercial statistical software was used for all data graphic presentations and statistical analysis^d.

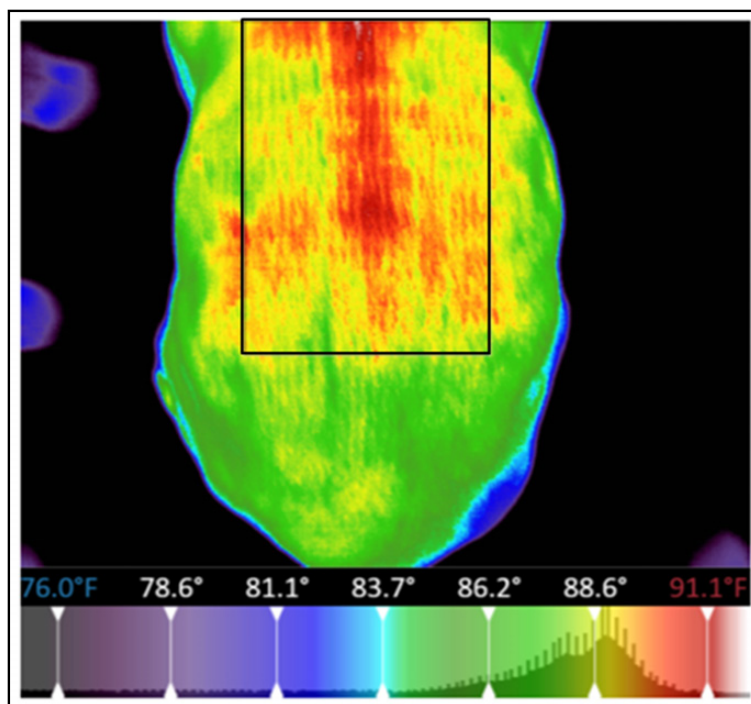


Figure 1: An example of a digital thermal image on a dog's back. The average temperature within the rectangle is the absolute temperature used for comparison between pre- and post-treatment values.

RESULTS

A total of 24 dogs exhibiting mild back pain (1 or 2 pain score) were enrolled and completed the study. Randomization resulted in 12 subjects in the Acupuncture Group and the remaining in the Control Group. In the acupuncture as well as the control groups, there were a total of 10 different breeds (Table 2). The diversity of breeds due to randomization of dogs within each treatment group reduced the possibility that the study outcome could be affected by a subject's breed.

The mean±SD age of subjects in the Acupuncture Group (11.2±3.4 years) were older than those in the Control Group (9.2±2.5 years) (Table 3). The group difference in age, however, was not statistically significant ($p = 0.079$; Wilcoxon rank sum test) between the two groups. The mean±SD weight among subjects in the Acupuncture Group was 44.3±34.7 pounds and was 46.5±32.9 pounds among those in the Control Group. Similarly, the weight difference between the two groups

was not statistically significant ($p = 0.932$, Wilcoxon rank sum test). The distribution of sex in the Acupuncture Group was 50% (6/12) female vs. 50% (6/12) male. In the Control Group, 66.7% (8/12) were female and 33.3% (4/12) were male. The proportions of male or female was not significantly greater than 0.5 [$p = 1.00$ (acupuncture) and 0.39 (control), exact binomial test]. Between the two treatment groups, the proportions of female (or male) were not significantly different ($p = 0.680$, Fisher's exact test).

All subjects had pain scores of either 1 or 2. Half of the subjects (6/12 = 50%) in the Acupuncture Group had pain score 1 and the remaining half had score 2. In the Control Group, 75% (9/12) of the subjects had pain score 1 and the remaining 25% had score 2. Between the two subject groups, the proportions of subjects with pain score 1 (or 2) were not significantly different ($p = 0.400$, Fisher's exact test), despite a 25% difference (Figure 2).

Table 2: Breed incidences occurring in each study group. The diversity of breeds, due to randomization within each study group, reduced the possibility that the study outcome could be affected by a subject's breed.

Breed	Control Group	Acupuncture Group
Golden Retriever mix	0	1
Miniature Poodle	0	1
Wirehair Dachshund	0	1
Cavalier King Charles Spaniel	1	1
German Shepherd	2	1
Miniature Dachshund	0	1
Havanese	1	1
Dachshund	0	2
Brittany mix	0	1
Labrador Retriever	1	2
Boxer mix	1	0
Chihuahua	1	0
Standard Poodle	1	0
Greyhound	2	0
Maltese	1	0
Brittany	1	0

Table 3: Summary table of study dog age, body weight and gender for the Control Group and Acupuncture Group. The study groups were comparable with no statistically significant differences in subject population.

	Control group (n = 12)	Acupuncture group (n = 12)	p-value
Age (mean±SD, years)	9.2±2.5	11.2±3.4	0.079
Weight (mean±SD, lbs)	46.5±32.9	44.3±34.7	0.932
Sex (Female %)	66.7%	50.0%	0.680

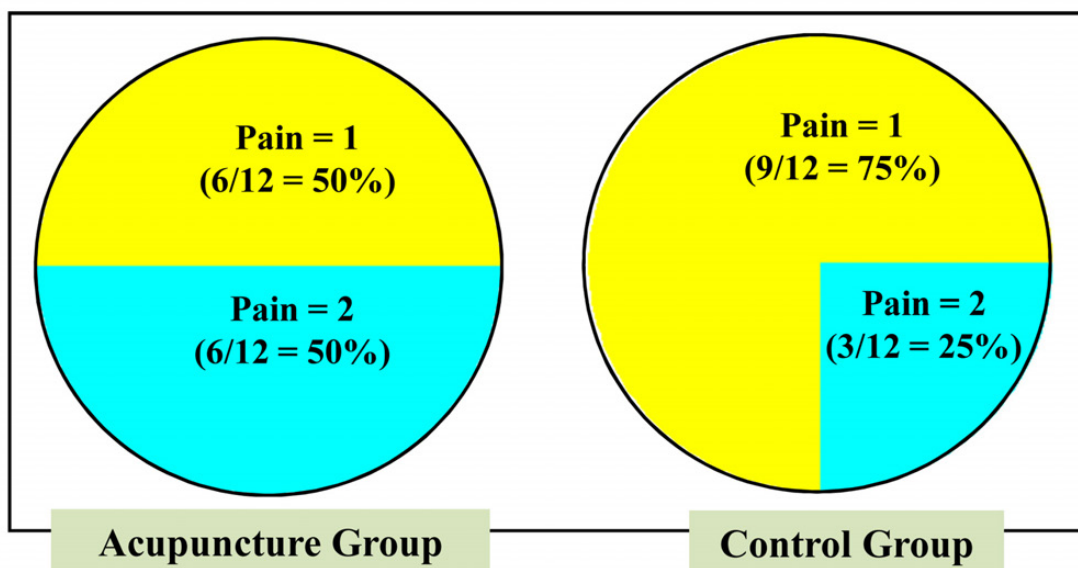


Figure 2: Distribution of pain score for study dogs before receiving study assigned treatment in each subject group (untreated control and acupuncture treatment). The groups were comparable with no statistically significant difference ($p=0.400$) in study population.

Thermal images were taken for each subject before and after the treatment period (Figures 3 and 4). The group mean \pm SD of the average temperature from the pre-treatment thermal imaging in the Acupuncture Group was $86.89\pm4.65^{\circ}\text{F}$, and in the Control Group was $86.15\pm3.92^{\circ}\text{F}$ (Figure 5). The baseline temperatures were comparable between the two subject groups ($p = 0.843$, no statistical significance, Wilcoxon rank sum test). Post treatment digital image temperature change was calculated as the absolute value of the difference between baseline and the post-treatment temperatures (Figure 6). Among the 12 subjects receiving acupuncture treatment, 9 subjects (75%) had reduced temperature and the remaining 3 (25%) had increased temperature. In the Control Group, 8 subjects (66.7%) had reduced temperature and the remaining 4 (33.3%) had increased temperature (Figure 7). The mean \pm SD temperature change in the Acupuncture Group was $1.60\pm0.51^{\circ}\text{F}$, and in the Control Group was $0.44\pm0.26^{\circ}\text{F}$. The overall temperature change difference between the two subject groups was statistically significant ($p = 2.96\times10^{-6}$, Wilcoxon rank sum test).

DISCUSSION

This is the first known randomized, blinded and controlled veterinary clinical trial to show the effects of acupuncture in a naturally occurring disease with digital thermal imaging. The study objective was to measure thoracolumbar sacral skin temperature change associated with acupuncture treatment of canine back pain. A total of 24 dogs divided into untreated controls ($n=12$) and DNAP treated ($n=12$) had pre- and post-treatment digital thermal images. Temperature change was calculated as the

absolute value of difference between baseline and post-treatment temperatures. The mean \pm SD temperature change in the Acupuncture Group was $1.60\pm0.51^{\circ}\text{F}$ versus Control Group at $0.44\pm0.26^{\circ}\text{F}$ (4X greater change for acupuncture). The overall temperature change difference between the two subject groups was statistically significant ($p = 2.96\times10^{-6}$). The results of the study supported the hypothesis, based on TCVM theory, that dogs treated with dry needle acupuncture would have greater back temperature change than the untreated controls under the experimental conditions of this study.

In this study, digital thermal imaging showed very clear differences between temperatures before and after acupuncture treatment, whereas, the Control Group had little temperature change. This suggests that acupuncture has a measurable effect on the radiated temperatures of the study subjects' bodies. It has been hypothesized that acupuncture derives its effects through increased production of endorphins and other neurotransmitters that influence pain sensation as well as other involuntary bodily functions.¹³ It also stimulates vascular and immunomodulatory factors.¹⁴ The beneficial effects, therefore, not only are associated with analgesia secondary to endorphin release, but also related to local vasodilation and anti-inflammatory effects.¹⁵

A characteristic of acupoint stimulation is that of dual effects. Stimulation of an acupuncture point can either inhibit excessive reactions or amplify deficient functions in the same target organ.¹⁶ For example, acupuncture needle insertion at an acupuncture point stimulates the nervous system which alters blood flow and humoral responses such that inflammation with heat is decreased while stimulation at the same acupoint in an

area that lacks perfusion will stimulate blood flow creating a temperature increase. In this way acupuncture harmonizes the biological constants of the body. Considering this, treated subjects have radiant temperature changes represented, at least partially, by changes in blood perfusion. Similar to findings in this

study, other studies have demonstrated acupuncture associated local changes in circulation and anti-inflammatory effects.¹⁷⁻²⁰ Acupuncture, therefore, starts as a local event of stimulation but then ripples through the nervous system of the body through multiple mechanisms.²¹

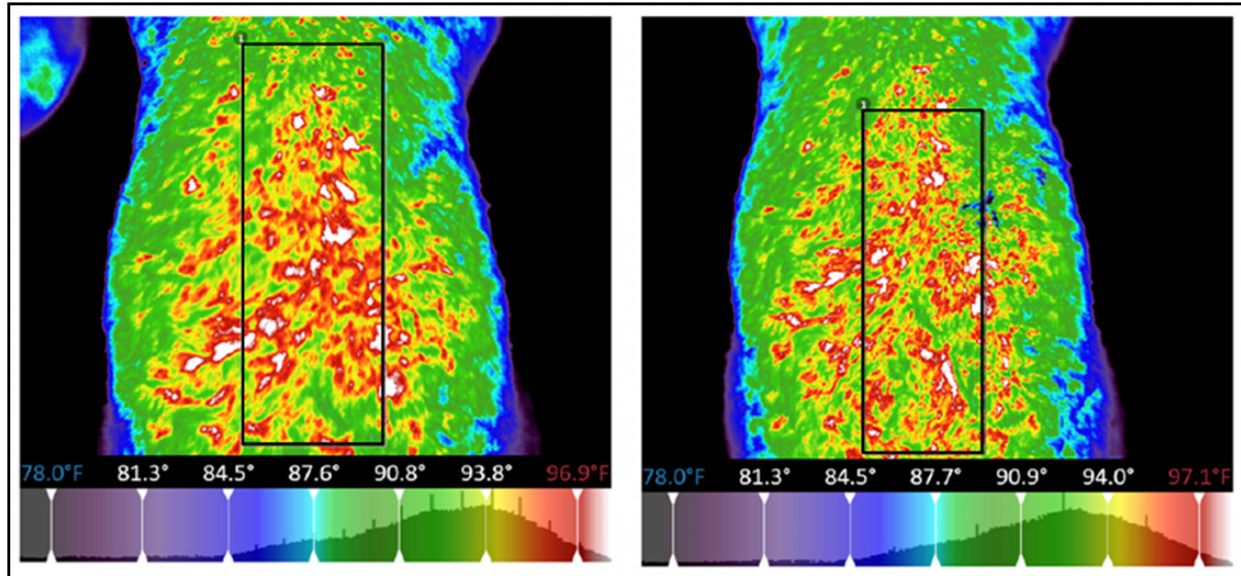


Figure 3: Thermal images on the back of a subject in the Control Group taken during the baseline period (left) and the post-treatment time (right). It is visually clear that the two images from the untreated control subject have similar color distributions. The average temperature within the focus area (rectangle) calculated by the device was 93.4°F in the baseline image and slightly increased to 93.6°F in the second image.

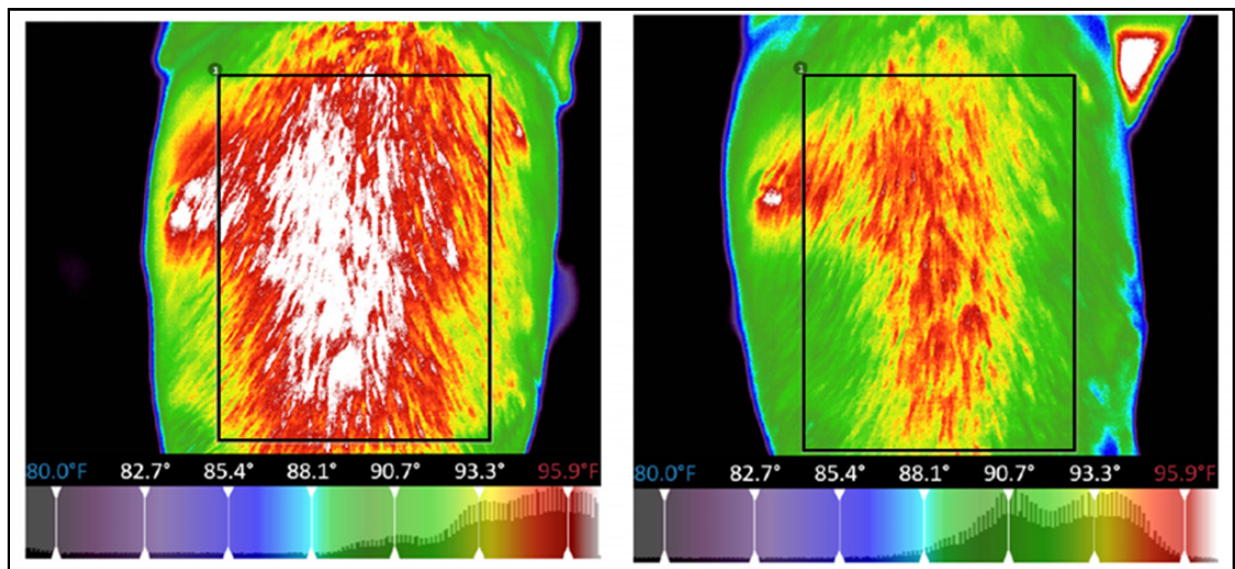


Figure 4: Thermal images on the back of a subject in the Acupuncture Group taken during the baseline period (left) and the post-treatment time (right). A significant difference in color distributions (rectangle) can be seen between the two images (94.8°F in the baseline image and reduced to 92.5°F in the post-treatment image).

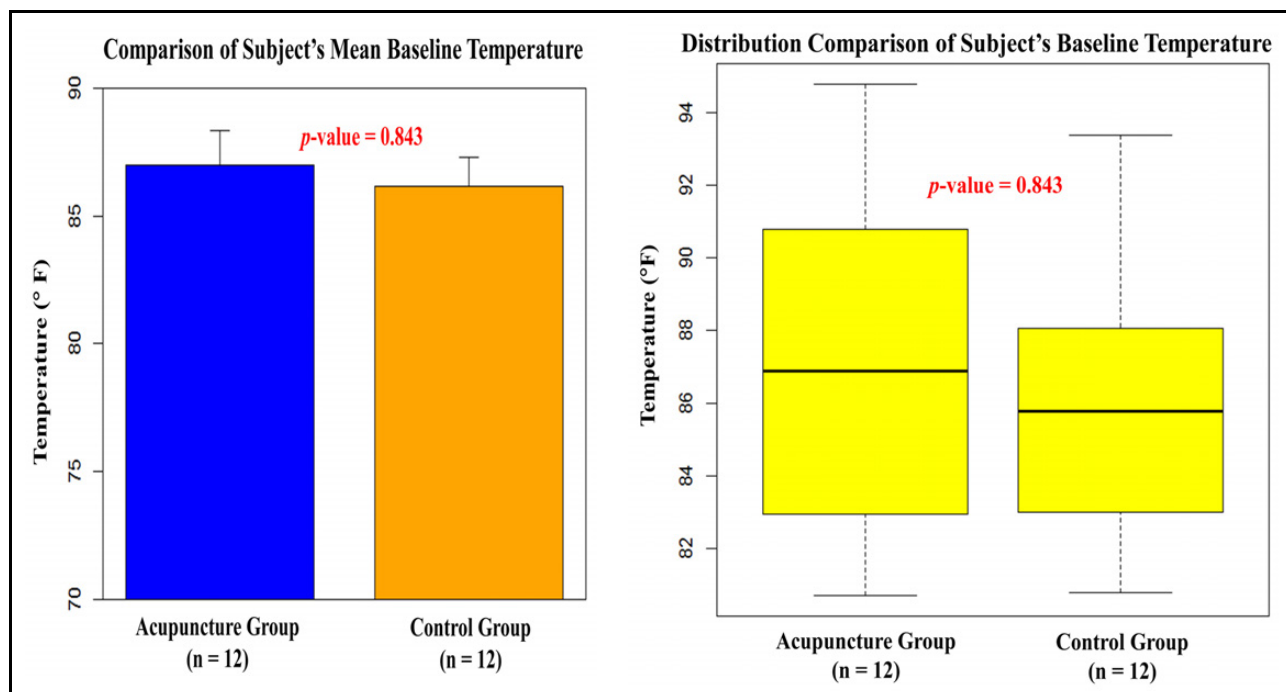


Figure 5: Pre-treatment average temperature mean and distribution of each study group; groups have similar baseline temperatures ($86.89 \pm 4.65^{\circ}\text{F}$ acupuncture vs $86.15 \pm 3.92^{\circ}\text{F}$ controls).

Digital thermal imaging has been used in humans to evaluate the efficacy of treatment of knee arthritis and has been shown to be a reproducible, sensitive and quantifiable method to document disease activity.^{22,23} Although the present study is the first randomized controlled veterinary clinical trial evaluating acupuncture effects in a naturally occurring disease with thermography, there are several case reports of natural disease documenting temperature change with thermography in individual acupuncture treated patients. For example, Dewey and Gucciardo reported on a 5-year old pit bull mix with intermittent hind leg lameness treated with acupuncture. Initial images revealed different temperature gradients in the right and left hind legs. Fifteen minutes following acupuncture treatment, thermal images demonstrated a dramatically decreased temperature accompanied by a rapid clinical response.²⁴ In another case report, a 4-year old French Bulldog with intervertebral disc disease (IVDD) was treated with EAP and saw an increase in thermal gradients correlating with the return of neurological function and circulation in the affected areas of the dog's back.²⁴ A study by Se-Wook et al. evaluated the efficacy of acupuncture on induced arthritis in dogs. Eight dogs were used in this experimental arthritis model which were randomly assigned into two groups [untreated control (n=4) and acupuncture treatment (n=4)]. After receiving acupuncture, once weekly, for a period of four weeks, the temperature in the arthritic joint returned to normal, whereas the temperatures in the non-treatment group remained high. The study concluded that thermography

would be useful to evaluate the treatment effect of acupuncture for canine induced chronic arthritis.²⁵

Limitations to this study included inability to evaluate long term clinical effects associated with acupuncture associated temperature change. Digital thermal imaging was only performed after the acupuncture treatment, and hence the longer-term (i.e. 30 or 60 minutes) effects from acupuncture treatment could not be assessed. In the clinical practice setting of this study, it was impractical to keep pets and their owners in a strictly controlled experimental environment for long periods of time. Other small issues that were considered to have minimal to no effect on study outcome were loss of acupuncture needles from dogs and movement of dogs during the 15 minute waiting period. Study dogs were generally selected as amicable dogs that had experienced acupuncture treatment before and therefore did not react with much body motion. Loss of needles only occurred in 2 animals (1 needle) and it is expected that retention of these needles would not have affected study conclusions.

Future investigations are warranted as the current study showed a statistically significant effect for acupuncture as measured by digital thermal imaging. By employing an objective measurement of the effects of acupuncture, studies in veterinary medicine could more closely track results in species that are not able to verbally self-report. It would also be important to compare images obtained in a longer time frame as well as linking the findings to clinical assessment of pain after treatment. Directly comparing study groups in the same environment and time frame would allow objective comparison of

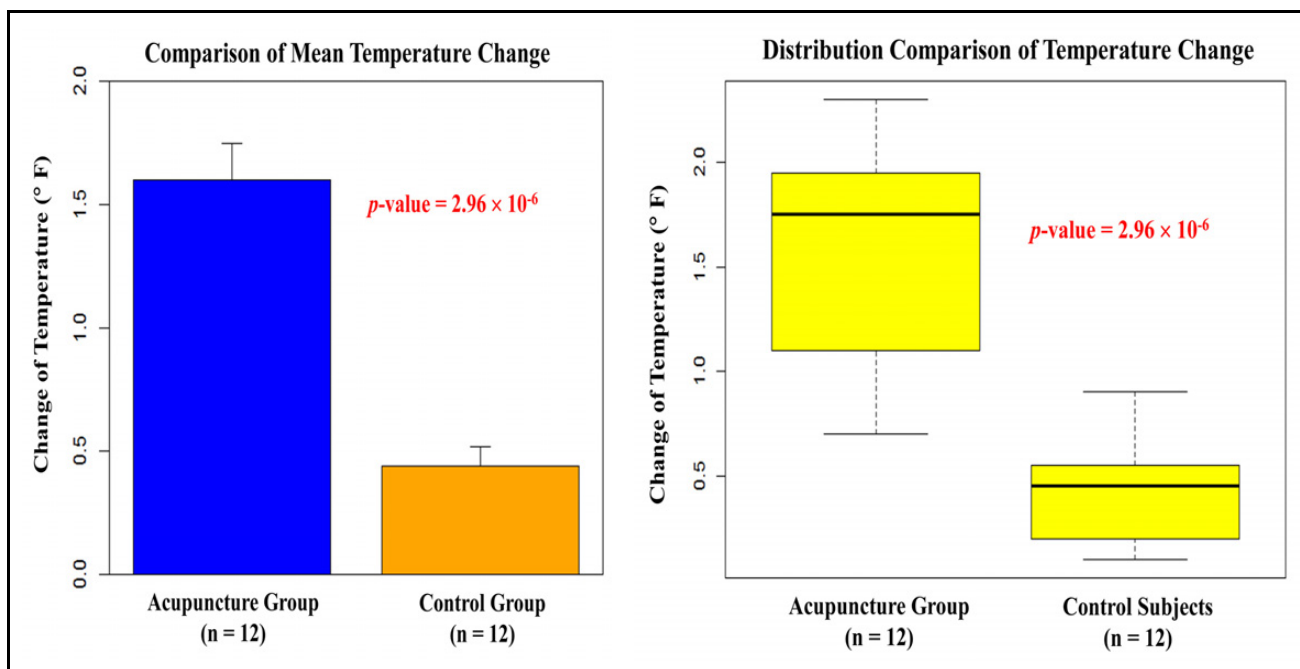


Figure 6: Post-treatment temperature change in each study group; mean and distribution of the temperature change from baseline to post-treatment. The mean temperature change in the Acupuncture Group was $1.60 \pm 0.51^\circ\text{F}$ versus Control Group of $0.44 \pm 0.26^\circ\text{F}$. The overall temperature change difference between the two subject groups was statistically significant ($p = 2.96 \times 10^{-6}$).

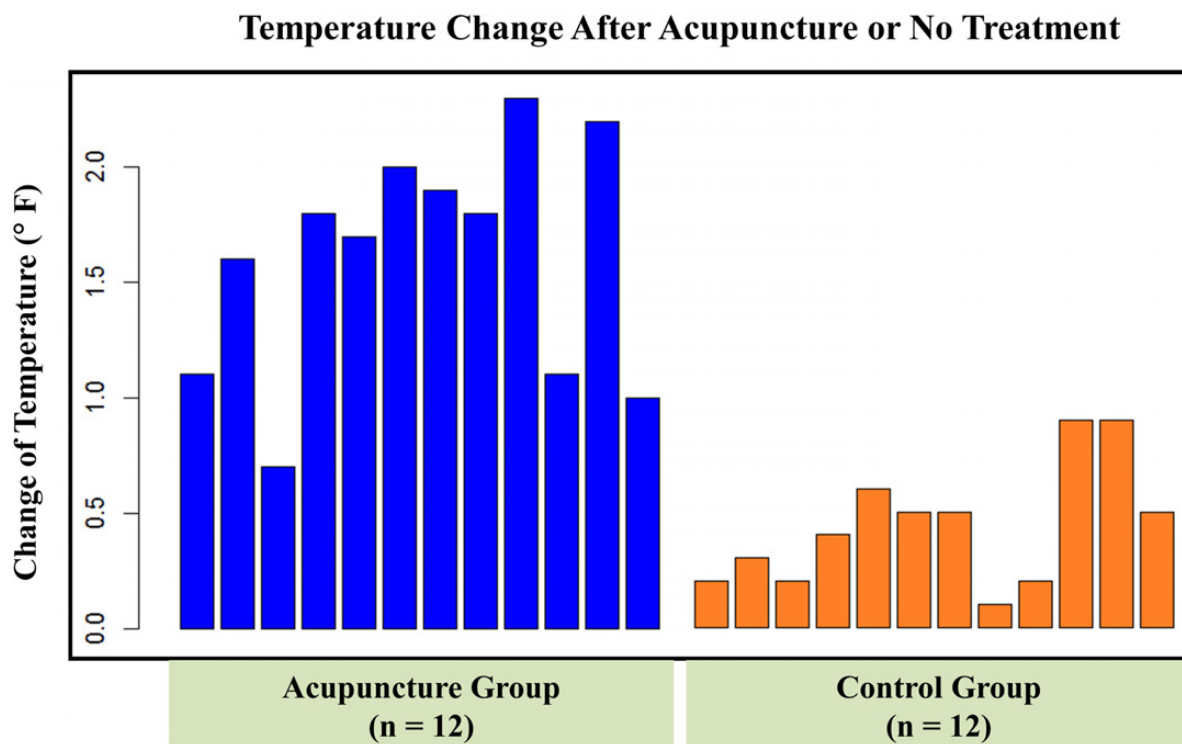


Figure 7: The absolute temperature change value for each individual subject is demonstrated. Temperature change was calculated as the absolute value of the difference between baseline and post-treatment temperature. Visual inspection reveals that individual dogs in the Acupuncture Group had greater temperature changes than those in the Control Group.

targeted acupuncture treatment, no treatment and an off-targeted therapy. Studies could also be designed to investigate the effects of acupuncture on specific conditions in dogs. The prevalent IVDD with its lack of function and perfusion would be a good disease to compare DNAP with EAP effects on the thermal gradients after treatment.

In summary, the results of this study demonstrated an immediate statistically significant difference regarding temperature change measured by digital thermal imaging between the Acupuncture Group and the Control Group. These findings objectively demonstrate that acupuncture has an immediate thermal effect on temperature gradients in dogs experiencing low grade back pain. In addition, this study supports the usefulness of thermography for monitoring therapeutic response to treatment in a non-verbal species and gives clinicians the ability to measure immediate acupuncture effects as well as demonstrate visual changes pet owners can easily appreciate.

ACKNOWLEDGEMENTS

The author thanks the entire staff of the Valley Cottage Animal Hospital for their support and greatly appreciates all clients and staff, as well as their lovely dogs who participated in this study.

Conflict of Interest

The author declares there is no conflict of interest that could be perceived as prejudicing the impartiality of this paper.

Funding

The author did not receive any specific grant of funding for this research study from any organization in the public, commercial or non-profit sectors.

FOOTNOTES

- a. www.randomization.com
- b. Digatherm IR Tablet 640, Digatherm, LLC, Ocala, FL, USA
- c. Jing Tang Herbal, Inc., Ocala, FL, USA
- d. R version 3.5.2. The R Foundation for Statistical Computing, Vienna Austria; <http://www.R-project.org>

REFERENCES

1. Hielm-Bjorkman A, Kuusela E, Liman A et al. Evaluation of methods for assessment of pain associated with chronic osteoarthritis in dogs. *J Am Vet Med Assoc* 2003; 222(11): 1552-1558.
2. Beaver B. Canine locomotive behavior. *Canine Behavior: A Guide for Veterinarians*, Beaver B (ed). Philadelphia, PA: WB Saunders Co 1999: 316.
3. Zimmermann M. Behavioral investigations of pain in animals. *Assessing Pain in Farm Animals*, Duncan I, Molony V (eds). Luxembourg, European Communities Directorate-General for Agriculture, Coordination of Agricultural Research 1986:16-27. V.2013/US/US2013_58 rdf.
4. Riegel R. Digital thermal imaging. *Veterinary Practice News* 2015; September: 44. Veterinarypracticenews.com/publications/de/201509/index.html.
5. Tunley B, Henson F. Reliability and repeatability of thermographic examination and the normal thermographic image of the thoracolumbar region in the horse. *Equine vet J* 2004; 36(4): 306-312.
6. Infernuso T, Loughin C, Marino D et al. Thermal imaging of normal and cranial cruciate ligament-deficient stifles in dogs. *Veterinary Surgery* 2010; 39(4): 410-417.
7. Vainionpaa M, Raekallio M, Junnila J et al. A comparison of thermographic imaging, physical examination and modified questionnaire as an instrument to assess painful conditions in cats. *J Feline Med Surg* 2012; 15(2): 124-131.
8. Levine J, Gromley J, Fields H. Preliminary clinical observations on the analgesic effect of needle acupuncture. *Pain* 1976; 2(2): 149-159.
9. Chiu J, Cheng H, Tai J et al. Electroacupuncture – induced neural activation detected by use of manganese-enhanced functional magnetic resonance imaging in rabbits. *Am J Vet Res* 2001; 62 (2): 178-182.
10. Madsen M, Gotsch P, Hrobjartsson A. Acupuncture treatment for pain: systematic review of randomized clinical trials with acupuncture, placebo acupuncture and no acupuncture groups. *BMJ* 2009; 338: doi:10.1136/bmj.a3115.
11. Skarda R, Teiwan G, Muir W. Cutaneous analgesia, hemodynamic and respiratory effects, and beta-endorphin concentration in spinal fluid and plasma of horses after acupuncture and electroacupuncture. *Am J Vet Res* 2002; 63(10): 1435-1442.
12. Zeltzman P. Download these Pain Scales. *Veterinary Practice News*. 2016. Veterinarypracticenews.com/download-these-pain-scales/.
13. Stux G, Berman B, Pomeranz B. *Basics of Acupuncture* 5th Ed. New York, NY: Springer-Verlag 2003: 17-26.
14. Shah J, Phillips T, Danoff J et al. An in vivo micro analytical technique for measuring the local biochemical milieu of human skeletal muscle. *J Appl Physiol* 2005; 99(5): 1977-1984.
15. Schoen A. *Veterinary Acupuncture-Ancient Art to Modern Medicine* 2nd Ed. St. Louis, MO: Mosby 2001: 161-169.
16. Qin Q, Mo Q, Liu K et al. Acupuncture at homotopic acupoints exerts dual effects on bladder motility in anesthetized rats. *BMC Complement Altern Med*. Biomedical Central (2015) 15:267, 6 pages. doi: 10.1186/s12906-015-0781-6
17. Min S, Lee H, Lim S-Y et al. Local changes in microcirculation and the analgesic effects of acupuncture: a laser doppler perfusion imaging study.

- J Alter Compl Med. 2015; 21(1): 46-52.
18. Zhang W, Wang L, Xie H et al. Comparison of acupuncture effect on blood perfusion between needling nonacupoint on meridian and needling nonacupoint off meridian. *Evid Based Compl Alter Med* 2013(7): 426052. doi:10.1155/2013/426052.
19. Kong P, Cao Y, Xu C et al. Detection of the therapeutic effect of acupuncture by laser speckle blood perfusion imaging. *Optik* 2018; 172: 1155-1161.
20. Zijlstra F, van den Berg-de Lange I, Huygen F et al. Anti-inflammatory actions of acupuncture. *Mediators of Inflammation* 2003; 12(2): 59-69.
21. Xie H, Preast V. *Xie's Veterinary Acupuncture*. Ames, Iowa: Blackwell Publishing 2007: 4, 15.
22. Kwon Y, Kim J, Yoon J et al. The Analgesic Efficacy of Bee Venom Acupuncture for Knee Osteoarthritis: A Comparative Study with Needle Acupuncture. *Am. J. Chin. Med.* 2001; 29(2): 187-199.
23. Deveraux M, Parr G, Thomas D et al. Disease activity indexes in rheumatoid arthritis; a prospective, comparative study with thermography. *Ann. Rheum. Dis.* 1985; 44(7): 434-437.
24. Dewey C, Gucciardo D. Electroacupuncture and digital thermal imaging. *Veterinary Practice News* 2017; February: 52. veterinarypracticenews.com/publications/de/201702/index.html.
25. Um S, Kim M, Lim J et al. Thermographic evaluation for the efficacy of acupuncture on induced chronic arthritis in the dog. *J. Vet. Med. Sci.* 2005; 67(12): 1283-1284.