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Efficacy of treatment of equine anhidrosis with acupuncture and Chinese herbs is low but higher in treated horses compared with placebo

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OBJECTIVE
To assess the therapeutic efficacy of acupuncture in combination with Chinese herbs for treatment of horses affected with anhidrosis.

ANIMALS
44 horses affected with anhidrosis for up to 3 years' duration were enrolled. Inclusion required both compatible clinical signs and results of a quantitative intradermal terbutaline sweat test.

METHODS
Study horses were randomly allocated into 2 groups. Group 1 (n = 19) was treated with daily Chinese herbs and 4 weekly acupuncture sessions. Group 2 (n = 25) was given daily hay powder as a placebo and 4 weekly sham acupuncture sessions. Horses were tested by quantitative intradermal terbutaline sweat test within 2 days after treatment completion and again 4 weeks following treatment.

RESULTS
Terbutaline-induced sweat responses (mg) were not different between groups within 2 days and 4 weeks after treatment. Two days after treatment, ratios of sweat responses (compared to baseline) were higher ($P < .05$) in the treatment group compared to the placebo group at terbutaline concentrations of 1.0, 100, and 1,000 µg/mL. The number of horses responding to treatment was higher in the treatment group (5/19 [26%]), compared to horses in the placebo group (1/25 [4%]) for 1 of 5 terbutaline concentrations 2 days (10 µg/mL) or 4 weeks (0.1 µg/mL) after treatment.

CLINICAL RELEVANCE
Ratios of sweat responses were higher in treatment horses 2 days after treatment, compared to baseline, but not 4 weeks later. The efficacy of a traditional Chinese veterinary medicine protocol for anhidrosis treatment with acupuncture and Chinese herbs was low but higher in treated horses compared with placebo.

Keywords: horses, anhidrosis, acupuncture, sweat, terbutaline sweat test

Anhidrosis is a serious condition of horses characterized by decreased or absent ability to produce sweat in response to increased body temperature.[1] This condition is reported primarily in hot and humid climates, although it can occur seasonally in more temperate areas.[2–4] Anhidrosis is a performance-limiting condition and can be life-threatening in the most severe cases.[5,6] Within the US, anhidrosis is commonly seen in southeastern states and Texas. In 1 study,[6] the prevalence of anhidrosis in Florida was 56 of 500 (11.2%) at the farm level and 83 of 4,620 (1.8%) at the animal level; in that study, family history of anhidrosis was identified as the most important exposure factor associated with this condition in study horses.

The pathophysiologic aspects of the disease are not well understood. There is inconclusive evidence that supports the hypothesis that anhidrosis initially results from epinephrine-induced downregulation of β2 adrenergic receptors on sweat glands.[7] Although initially reversible, progressive degenerative changes in chronically anhidrotic sweat glands eventually render them permanently nonfunctional.[1] Therefore,

with no evidence-based studies to support any purported or anecdotal remedy, it is not surprising that treatment is also unsuccessful.[7] Although moving horses with acute anhidrosis to cooler and drier climates is an option for management of this condition, this option is often not feasible.

Acupuncture combined with Chinese herbal medication has been used with ostensible success for treatment of horses affected with anhidrosis.[8,9] Chinese herbal medicine is often used in combination with acupuncture to prolong treatment effect and reduce the need for multiple and frequent acupuncture sessions. Determining the acupuncture treatment protocol for a specific patient is often dictated by a traditional Chinese veterinary medicine (TCVM) pattern diagnosis and selection of acupoints based on that diagnosis. To provide adequate uniformity for this research, a single treatment plan was used for all patients based on the most common TCVM diagnosis made for horses with anhidrosis.[8] To our knowledge, the efficacy of acupuncture combined with Chinese herbs for treatment of horses affected with anhidrosis has not been evaluated by use of objective research methods. The purpose of the study reported here was to assess the therapeutic efficacy of acupuncture in combination with Chinese herbs for treatment of horses affected with anhidrosis. Our hypothesis was that acupuncture in combination with Chinese herbs would be an effective treatment for horses with anhidrosis of < 3 years' duration.

Methods

All study procedures were approved by the University of Florida's IACUC.

Study horses

Horses with anhidrosis were identified and recruited for study through word of mouth and mailed requests to equine veterinarians located within 50 miles of the authors' institution. Based on the observation by Evans[10] that the condition may be irreversible in horses that have been anhidrotic for > 2 or 3 years, 60 horses with owner-reported clinical signs of anhidrosis that began within the previous 2 years were initially considered for inclusion, excluding those with a chronic history of anhidrosis. Forty-eight horses were enrolled into the study on the basis of compatible clinical signs of anhidrosis and inadequate response to a quantitative intradermal terbutaline sweat test (QITST).[11] Horses with net sweat weight lower than 1 SD below the mean values established by MacKay[11] at 3 of the 5 terbutaline concentrations tested were considered anhidrotic and qualified for enrollment (cutoffs: 0.1 µg/mL, < 31 mg; 1 µg/mL, < 89 mg; 10 µg/mL, < 149 mg; 100 µg/mL, < 245 mg; and 1,000 µg/mL, < 343 mg). Twelve horses were excluded due to normal QITST response.

Sample size

A previous study of sweating in anhidrotic horses in north-central Florida (separate anhidrotic group from horses used to validate QITST procedure) found a 2-fold difference between mean ± SD sweat weights of anhidrotic horses (226 ± 190 mg) and their free-sweating pasture-mates (450 ± 287 mg) in response to 1,000 µg/mL of intradermal terbutaline.[12] These values were used to model a clinically relevant response to treatment for anhidrosis. Thus, a sample of 40 study horses (19/group) provided 95% confidence and 80% power to declare differences in sweat weights between 226 ± 190 mg (placebo group) and 450 ± 287 mg (acupuncture/herbs group) as statistically significant.[13] In addition, a sample size of 20 horses/group provided 95% confidence and 80% power to declare differences in the number of horses that responded to treatment (50% treatment vs 10% control) as statistically significant. Eight additional horses were enrolled for a total of 48 horses to allow for possible attrition during the study period.

Allocation of horses into experimental groups

A free randomizer software (www.randomizer.org) was used to generate random numbers and treatment assignments. Group 1 (acupuncture/herbs group; n = 22) were horses treated with the combination of daily Chinese herbs top-dressed on feed and acupuncture once every week for 4 weeks. Group 2 (placebo; n = 26) were horses that were fed hay powder daily and subjected to sham acupuncture once every week for 4 weeks.

Outcomes

One outcome of interest was terbutaline-induced sweat production (mg) in response to serial dilutions of terbutaline injected intradermally in study horses according to the previously described QITST procedure.[11] Briefly, the QITST is performed by injecting 0.1 mL of serial dilutions of terbutaline (0, 0.1, 1, 10, 100, and 1,000 µg/mL) intradermally in the neck of the study horse. Absorbent pads are taped over the injection sites, and sweat produced in response to terbutaline is collected for 30 minutes. The sweat weights are quantified and compared to normal values for free-sweating horses.[11] Sweat responses were measured at the time of enrollment (baseline) and 1 to 2 days and 4 weeks after treatment was completed. Testing was repeated twice following the treatment period to measure both immediate and prolonged response to treatment. In addition, among horses in the acupuncture/herbs group, terbutaline-induced sweat responses showing a 2-fold increase from baseline levels at 3 or more terbutaline concentrations (1 to 2 days or 4 weeks after treatment) were considered an indication that study horses responded to therapy.

Treatments

Acupuncture treatment was administered by 1 of 4 certified veterinarian acupuncturists at a specific set of acupoints selected to treat the TCVM diagnosis of Summer Heat (the typical pattern diagnosis made in cases of equine anhidrosis).[8] In TCVM, Summer Heat is considered a Yang pathogenic factor and can exhaust Qi and Yin, resulting in a chronic Yin deficiency that manifests as anhidrosis. Treatments are targeted to-

ward releasing excess Heat and restoring both Qi and Yin.[14] Treatment included dry needling of KID-7, SP-6, HT-7, TH-1, LU-11, SI-1, HT-9; hemoacupuncture of Er-Jian, Wei-Jian, Tai-Yang, Wei-ben, and Xiong-tang; and electroacupuncture for 20 minutes (80 to 120 Hz) of BL-13 + Gei-shu, GV-14 + GB-21, and BL-15 + BL-22. Total time of treatment was 30 minutes from the initiation of needle placement. Sham acupuncture consisted of all needles placed 2 cm lateral to each treatment point, with the electroacupuncture unit attached to the relevant needles but not turned on. The herbal formula contained dolichoris seed (bian dou), magnolia (hou po), lonicera (jin yin hua), forsythia (lian qiao), and elsholtzia (xiang ru; Modified Xiang Ru San; Jing Tang Herbal Pharmacy). Placebo peanut hay powder was packaged and dosed in the same fashion as the herbal medication.

Bias control

Attending veterinarians and horse owners were blinded to study groups. Acupuncture treatments and sweat testing were performed by separate teams to maintain blinding of all personnel performing diagnostic tests. The acupuncture team was aware of treatment assignments; all other study personnel were blinded.

Data analysis

Baseline comparisons were performed to establish the comparability of experimental groups. The frequencies of horses by month of enrollment (June, July, or August) and breed (Thoroughbred, Quarter horse, or other) were compared between groups by use of a χ^2 test. Distributions for the variables of age (years), duration (years) of clinical signs at enrollment, and baseline sweat responses were compared between groups by use of the nonparametric Wilcoxon rank sum test.

Outcome comparisons were conducted to assess treatment efficacy. The null hypotheses that sweat responses (mg; ranked data) would not be different between groups at baseline and 1 to 2 days and 4 weeks after treatment at specific terbutaline concentrations were tested by use of repeated-measures ANOVA. The null hypotheses that median ratios of increase in sweat response (mg) would not be different between groups (1 to 2 days after treatment vs baseline or 4 weeks after treatment vs baseline) at specific terbutaline concentrations were tested by use of the nonparametric Wilcoxon signed rank test. The null hypotheses that the number of horses that responded to treatment (≥ 2-fold increase in sweat response compared to baseline) would not be different at specific terbutaline concentrations were tested by use of a χ^2 test. Finally, in this study, treatment efficacy was measured using the following formula for attributable fraction: ([response rate in treatment horses – response rate in placebo horses] / [response rate in treatment horses]) X 100 or ([50% – 10%] / [50%]) X 100 = 80%.

Results

Forty-eight horses were initially enrolled in the study. Four horses (3 in the acupuncture/herbs group and 1 in the placebo group) were excluded. Three horses were removed from the study due to owner compliance with treatments and 1 was excluded because of illness unrelated to treatment.

The study sample included 14 mares, 24 geldings, and 6 stallions **(Table 1)**. Breeds were Thoroughbred

Table 1—Baseline comparisons of horses treated or not treated with acupuncture and Chinese herbs.

	Acupuncture/herbs n = 19	**Placebo group n = 25**	***P* value**
Month of enrollment			.790
June	6	7	
July	8	9	
August	5	9	
Breed			ND*
Thoroughbred	5	9	
Quarter horse	4	7	
Paso fino	1	2	
Draft	3	1	
Pony	1	3	
Warmblood	2	2	
Arab	1	0	
Standardbred	2	0	
Mini	0	1	
Breed			.543
Thoroughbred	5	9	
Quarter horse	4	7	
Other	10	9	
Age (y)	15 (9, 22)**	11 (4, 18)**	.230
Duration of anhidrosis (y)	2 (0, 2)**	2 (1, 2)**	.210
Sweat response (mg) dose			
0 µg/mL terbutaline	0.041 (0.024, 0.062)**	0.049 (0.028, 0.057)**	.981
0.1 µg/mL terbutaline	0.049 (0.030, 0.91)	0.066 (0.041, 0.085)	.484
1.0 µg/mL terbutaline	0.092 (0.060, 0.113)	0.083 (0.052, 0.112)	.484
10 µg/mL terbutaline	0.118 (0.068, 0.156)	0.128 (0.098, 0.150)	.569
100 µg/mL terbutaline	0.165 (0.096, 0.229)	0.156 (0.095, 0.238)	.962
1,000 µg/mL terbutaline	0.229 (0.208, 0.332)	0.186 (0.079, 0.294)	.209

*ND = Not determined. **Data are reported as median (first quartile, third quartile).

(n = 14), quarter horse (11), warmblood (4), draft horse (4), Paso Fino (3), pony (4), Arabian (1), Standardbred (2), and miniature horse (1). Median age was 13 years (minimum, 8 years; maximum, 21 years). Months of study enrollment were June (n = 13 horses), July (17), and August (14) of 2010, 2011, and 2012.

Baseline comparisons

Experimental groups were comparable on the basis of number of horses enrolled in June, July, or August; breed; age; duration of clinical signs of anhidrosis; and sweat response at enrollment ($P \geq .209$; Table 1).

Outcome comparisons

Using repeated-measures ANOVA, terbutaline-induced sweat responses were not different between groups at baseline and 1 to 2 days and 4 weeks after treatment at selected terbutaline concentrations of 0 µg/mL ($P = .798$), 0.1 µg/mL ($P = .915$), 1 µg/mL ($P = .458$), 10 µg/mL ($P = .760$), 100 µg/mL ($P = .885$), and 1,000 µg/mL ($P = .381$; **Figure 1**).

Sweat responses 1 to 2 days after treatment, compared to baseline—Ratios of sweat responses were higher ($P < .05$) among horses in the treatment group, compared to horses in the placebo group at selected terbutaline concentrations of 1.0, 100, and 1,000 µg/mL **(Table 2)**. In addition, the number of horses that responded to treatment was higher in the treatment group (5/19 [26%]), compared to horses in the placebo group (1/25 [4%]) when using a terbutaline concentration of 10 µg/mL, but this difference did not reach statistical significance ($P = .07$). The observed treatment efficacy is [26% – 4%] / [26%] X 100 = 85%.

Sweat responses 4 weeks after treatment, compared to baseline—Ratios of sweat responses were not different ($P \geq .28$) among horses in the treatment group, compared to horses in the placebo group at all 6 investigated terbutaline concentrations **(Table 3)**. However, the number of horses that responded to

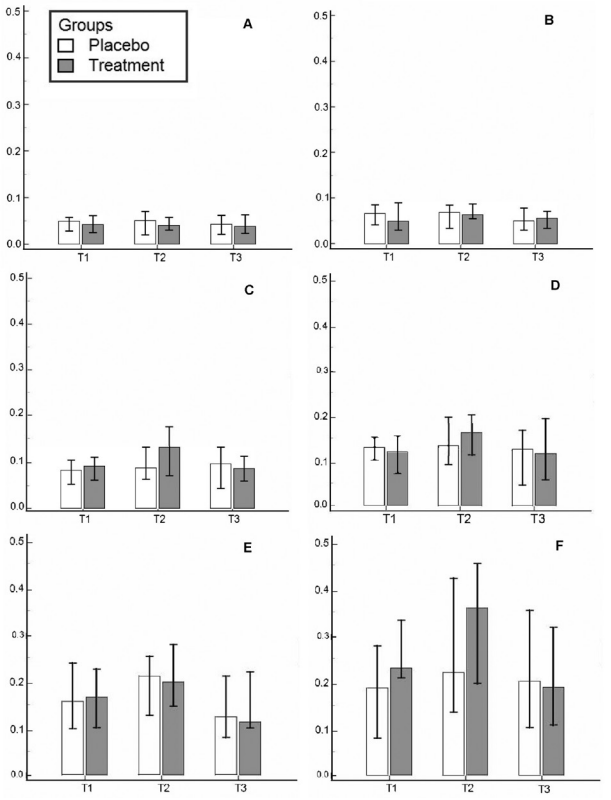

Figure 1—Comparisons of sweat responses (mg of sweat) induced by 0 µg/mL terbutaline (A), 0.1 µg/mL terbutaline (B), 1 µg/mL terbutaline (C), 10 µg/mL terbutaline (D), 100 µg/mL terbutaline (E), and 1,000 µg/mL terbutaline (F) between anhidrotic horses treated with acupuncture and Chinese herbs (treatment) versus sham acupuncture and hay powder (placebo). Sweat tests were conducted at T1 = enrollment (baseline), T2 = 1 to 2 days after completion of treatments, and T3 = 4 weeks after treatment. Each bar is median sweat weight; error bars indicate first and third quartile values.

Table 2—Outcome comparisons of (A) sweat response (mg) ratios 1 to 2 days after treatment compared to baseline and (B) number of horses that responded to treatment: ≥ 2-fold increase in sweat 1 to 2 days after treatment.

	Acupuncture/herbs n = 19	Placebo group n = 25	*P* value
(A) Ratios of sweat responses (mg) compared to baseline*			
0 µg/mL terbutaline	0.91 (0.73, 1.94)	1.06 (0.29, 1.81)	.923
0.1 µg/mL terbutaline	1.17 (0.97, 2.36)	0.93 (0.50, 1.53)	.402
1.0 µg/mL terbutaline	1.54 (0.98, 2.23)	1.31 (0.81, 1.95)	.010
10 µg/mL terbutaline	1.22 (0.82, 2.11)	1.04 (0.86, 1.75)	.059
100 µg/mL terbutaline	1.32 (0.84, 1.63)	1.10 (0.96, 1.46)	.006
1,000 µg/mL terbutaline	1.33 (0.88, 2.01)	0.96 (0.79, 2.11)	.031
(B) No. (%) of horses that responded to treatment (≥ 2-fold increase in sweat)**			
0 µg/mL terbutaline	4 (21)	5 (20)	1.000
0.1 µg/mL terbutaline	6 (31)	4 (16)	.286
1.0 µg/mL terbutaline	5 (26)	6 (24)	1.000
10 µg/mL terbutaline	5 (26)	1 (4)	.070
100 µg/mL terbutaline	3 (15)	4 (16)	1.000
1,000 µg/mL terbutaline	5 (26)	6 (24)	1.000

*Data are reported as median ratio (first quartile, third quartile). **Data are reported as n (%).

Table 3—Outcome comparisons of (A) sweat response (mg) ratios 4 weeks after treatment compared to baseline and (B) number of horses that responded to treatment: ≥ 2-fold increase in sweat 4 weeks after treatment.

	Acupuncture/herbs n = 19	Placebo group n = 25	*P* value
(A) Ratios of sweat responses (mg) compared to baseline*			
0 µg/mL terbutaline	1.20 (0.37, 1.99)	0.82 (0.34, 1.76)	.617
0.1 µg/mL terbutaline	1.21 (0.38, 2.83)	0.83 (0.38, 1.24)	.284
1.0 µg/mL terbutaline	1.05 (0.68, 1.27)	0.95 (0.51, 1.56)	.997
10 µg/mL terbutaline	0.97 (0.59, 1.58)	0.88 (0.40, 1.25)	.553
100 µg/mL terbutaline	0.98 (0.52, 1.72)	0.73 (0.57, 1.61)	.482
1,000 µg/mL terbutaline	0.71 (0.38, 1.21)	1.13 (0.65, 2.12)	.895
(B) No. (%) of horses that responded to treatment (≥ 2-fold increase in sweat)**			
0 µg/mL terbutaline	4 (21)	4 (16)	.709
0.1 µg/mL terbutaline	5 (26)	1 (4)	.071
1.0 µg/mL terbutaline	3 (15)	5 (20)	1.000
10 µg/mL terbutaline	4 (21)	1 (4)	.148
100 µg/mL terbutaline	3 (15)	5 (20)	1.000
1,000 µg/mL terbutaline	3 (15)	7 (28)	.474

*Data are reported as median ratio (first quartile, third quartile). **Data are reported as n (%).

treatment was higher in the treatment group (5/19 [26%]) compared to horses in the placebo group (1/25 [4%]) when using a terbutaline concentration of 0.1 µg/mL, but this difference did not reach statistical significance (*P* = .07).

Discussion

This study suggested that the use of acupuncture in combination with Chinese herbs might be efficacious for treatment of anhidrosis in study horses. The investigation was designed as a randomized clinical trial to mitigate investigator bias during treatment allocation. Furthermore, baseline comparisons of variables measured in this study (ie, breed, age, duration of anhidrosis, and baseline terbutaline-induced sweat responses) showed that study groups were comparable. Finally, this study used the QITST to produce a standard measure for sweat responses in study horses, thus reducing the risk of bias for outcome measurements.

Overall, terbutaline-induced sweat responses (directly comparing pre- and post-treatment sweat weights) were not different from baseline in study horses within 2 days after treatment and 4 weeks after treatment with acupuncture and Chinese herbs. At 2 days following treatment, ratios of sweat responses were higher (*P* < .05) among horses in the treatment group, compared to horses in the placebo group at selected terbutaline concentrations of 1.0, 100, and 1,000 µg/mL. The ratio of sweat response pre- and post-treatment provides a relative comparison of sweat weight as an additional measure of effectiveness. The number of horses that responded to treatment was higher in the treatment group (5/19 [26%]), compared to horses in the placebo group (1/25 [4%]) for 1 of the 5 terbutaline concentrations at both 2 days and 4 weeks after treatment. This result highlights the need for additional objective validation of perceptions of treatment effectiveness. Although current compendia of equine acupuncture treatments claim success in treating anhidrosis with combinations of acupuncture and Chinese herbs,[8,9] objective evidence only marginally supports such claims.

To the best of our knowledge, this study was the first randomized clinical trial of any putative anhidrosis remedy. A wide variety of empirical approaches to anhidrosis have been reported, including antihistamines, vitamins B and C, salt/electrolytes administered IV or PO, thyroid supplements/iodinated casein/organic or inorganic iodine, vitamin E/germinated wheat, adrenal powder/ACTH, prostaglandin F2α, and α-methyl dopa.[7] A popular feed supplement containing vitamin C, niacin, tyrosine, and cobalt is marketed for treatment of anhidrosis on the basis of an uncontrolled clinical trial.[15] It is not clear that any of these purported remedies improve sweating in anhidrotic horses, and it is hoped that future claims of efficacy will be supported by properly designed clinical studies.

This study had several limitations. Most of the horses (28/44) enrolled in the study had been anhidrotic for 2 or 3 years at the time of enrollment, and it is conceivable that more robust responses to acupuncture and Chinese herbs would have been achieved if the study had been limited to horses with shorter histories of nonsweating. However, when we examined sweat responses in 16 study horses with a history of anhidrosis for 1 year or less at the time of enrollment, the study results did not change. In addition, it is possible that the acupuncture control procedure used in this study could have obscured treatment success. Sham acupuncture points were 2 cm from verum acupoints, and needles were fully inserted at these points although not stimulated electronically. A recent systematic review by Choi et al[16] summarized contradictory evidence from a number of pain studies that have led some investigators to question the concept of point specificity of acupuncture and therefore call into question the validity of sham acupuncture as a

placebo control. If, as a result of ambiguous point specificity, needling of sham acupoints in this study elicited partial stimulation of sweating, it is possible that this would reduce our ability to detect a difference in resulting sweat weights between groups. Finally, it is possible that despite sample size calculations completed as part of the study design, the study groups were too small to detect an effect of treatment.

In summary, use of an established TCVM protocol for treatment of anhidrosis by acupuncture and Chinese herbs was marginally effective in increasing sweat production in horses that were classified as anhidrotic on the basis of quantitative sweat response tests. Treatment efficacy was low but higher in treatment horses, compared to placebo horses at low terbutaline concentrations 2 days and 4 weeks after treatment. Ratios of sweat responses (mg; compared to baseline) were higher in treatment horses 2 days after treatment but not 4 weeks after treatment. Future work should aim to use this testing approach to evaluate other putative anhidrosis remedies.

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Disclosures

Dr. Huisheng Xie owns the Chi Institute and Jing Tang Herbal companies (Reddick, Florida). Herbal medication and placebo powder were donated by Jing Tang Herbal for this study.

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