

Correlation Between Physiologic and Osteopathic Measures of Sympathetic Activity in Women With Polycystic Ovary Syndrome

Sarah E. Davis, DO; Jan Hendryx, DO; Stephanie Bouwer, OMS III; Christian Menezes, OMS IV; Heather Menezes, OMS IV; Vishesha Patel, OMS IV; Diana L. Speelman, PhD

From the Idaho College of Osteopathic Medicine in Meridian (Dr Davis) and the Lake Erie College of Osteopathic Medicine in Erie, Pennsylvania (Drs Hendryx and Speelman and Student Doctors Bouwer, C. Menezes, H. Menezes, and Patel).

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Address correspondence to
Diana L. Speelman, PhD,
Lake Erie College of Osteopathic Medicine, 2000 W Grandview Blvd, Erie, PA 16509-1029.

Email: dspeelman@lecom.edu

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Background: Polycystic ovary syndrome (PCOS) is a common hormone disorder that affects the reproductive, metabolic, and psychological health of reproductive-aged females, with a number of long-term health risks, including type 2 diabetes mellitus and cardiovascular sequelae. Sympathetic hyperactivity in affected persons may be involved in the pathogenesis of the disorder.

Objective: To determine whether physiologic and osteopathic measures of increased sympathetic tone correlate in a population of women with PCOS.

Methods: For this descriptive observational study, women with PCOS between the ages of 20 and 44 years were recruited. Physiologic measures of sympathetic tone (resting heart rate and blood pressure, resting heart rate variability, and postexercise heart rate recovery and blood pressure) were compared with osteopathic measures of sympathetic tone (Chapman points and viscerosomatic reflexes) for the heart, adrenal glands, and ovaries.

Results: Twenty-four women participated in the study. Overall, the participants exhibited both physiologic and osteopathic signs of increased sympathetic tone compared with reference values. In some participants, the number of osteopathic findings were greater than the number of physiologic findings of increased sympathetic tone.

Conclusions: Women with PCOS exhibit increased sympathetic tone by physiologic and osteopathic measures, indicating the utility of assessing sympathetic hyperactivity in these patients by osteopathic methods. Osteopathic structural examination is a valuable diagnostic tool that may allow detection of sympathetic hyperactivity in women with PCOS even before physiologic symptoms manifest. The osteopathic indicators of increased sympathetic tone may represent potential therapeutic targets to improve health in this population. (ClinicalTrials.gov NCT03383484)

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Polycystic ovary syndrome (PCOS) is the most common endocrine and metabolic disorder in females, affecting 5% to 15% of females of reproductive age.¹⁻⁴ The syndrome may manifest itself as early as the second decade of life, although it is often not diagnosed until the third decade, when the reproductive features of the disorder are more apparent.^{5,6} Polycystic ovary syndrome can affect endocrine, reproductive,

metabolic, and psychological health in females from puberty onward.⁶⁻⁸ Common clinical findings include menstrual irregularity secondary to oligo-ovulation, subfertility, greater incidence of miscarriage, hirsutism, acne, weight gain, anxiety, and depression. Females with PCOS often exhibit glucose intolerance and insulin resistance and are at greater risk for the development of type 2 diabetes mellitus and cardiovascular sequelae. Despite its prevalence and impact on health, the cause of the disorder remains unknown.⁹⁻¹⁴

Sympathetic nervous system hyperactivity, or increased sympathetic tone, is common in females with PCOS, yet, most patients are unaware of its occurrence. Increased sympathetic tone is associated with elevated androgen levels, anovulation, and menstrual irregularity, and it may play a role in the pathogenesis of the disorder.¹⁵⁻¹⁹ A generalized increase in sympathetic tone is evidenced by increased resting heart rate, lower heart rate variability, hypertension, exaggerated systolic blood pressure response to exercise, attenuated heart rate recovery after exercise, increased muscle sympathetic nerve activity, and increased levels of adrenergic metabolites in the serum and urine.²⁰⁻²² Chronic sympathetic nervous system hyperactivity is associated with central obesity, and obesity further exacerbates this hyperactivity.²³⁻²⁶ Local increased sympathetic tone has also been reported in females with PCOS and in rodent models of PCOS, with increased ovarian sympathetic outflow and expression of nerve growth factor, increased sympathetic tone of adipose tissue, and altered adrenoreceptor expression in the ovaries and in the periventricular nucleus in the hypothalamus.^{19,27-31} Furthermore, transection of either the superior ovarian nerve or the vagus nerve in a rat model for PCOS improved sympathetic tone, decreased ovarian androgen production, improved cyclicity, and restored ovulation.^{27,32} Together, these findings demonstrate an important role for the sympathetic nervous system in the pathogenesis of PCOS.

The osteopathic approach to health and medicine offers a unique set of tools with which to assess somatic dysfunction in females with PCOS and to

potentially treat these females. Current methods used to assess sympathetic hyperactivity in patients with PCOS rely on physiologic measurements, which can be time-consuming and impractical to use routinely in practice but which nonetheless provide important information in making a comprehensive clinical assessment of a patient's health.^{20,21} Increased sympathetic tone leads to tissue texture changes, such as the formation of Chapman points and viscerosomatic reflexes associated with specific organs.³³

The purpose of this study was to determine whether physiologic measurements of sympathetic tone correlate with osteopathic structural examination findings of sympathetic tone in women with PCOS.

Methods

Participants

Participants were recruited from the Erie, Pennsylvania, area by direct advertising through radio, billboards, websites, and a newspaper and magazine from October 2016 through January 2017. Physician referrals from Medical Associates of Erie clinics were also used to recruit participants. Potential participants signed a consent form to have relevant medical records released to confirm their PCOS diagnosis, as well as informed consent to participate in the study. All protocols were approved by the Lake Erie College of Osteopathic Medicine Institutional Review Board.

A participant questionnaire was given at the time of consent to determine eligibility. Inclusion criteria were aged 20 to 44 years and meeting the 2003 Rotterdam criteria² for PCOS diagnosis, defined as at least 2 of the following 3 features: (1) clinical or biochemical hyperandrogenism (moderate acne or a modified Ferriman-Gallwey score ≥ 8 or free testosterone >5 pg/mL); (2) polycystic ovaries (≥ 12 cysts on 1 ovary or ovarian volume >10 mL for 1 ovary); and (3) menstrual irregularity (≤ 8 menstrual periods per year or cycles averaging >35 days). Exclusion criteria were current smokers; use of hormone-based medications within the past 3 months (eg, hormonal contraception, ovulation inducers,

antiandrogens) or use of insulin-sensitizing agents; diagnosis of another endocrine disorder; pregnant or having breastfed within the past 6 months; diagnosis of a major psychiatric disorder or taking antipsychotic medication; or currently receiving osteopathic manipulative treatment, chiropractic manipulation, or acupuncture on a regular basis.

Assessment of Participants

In January 2017, eligible participants underwent physiologic assessment of sympathetic tone and osteopathic structural examination within the same week, each conducted by different investigators who were blinded to the findings of the reciprocal assessment. Physiologic and osteopathic measurements were made in different rooms in the same building, but each set of measurements (physiologic or osteopathic) was performed in the same temperature- and humidity-controlled rooms for each participant. A whole blood sample was collected and analyzed by outside laboratories. Free testosterone was calculated from a mathematical model using sex hormone-binding globulin, albumin, and total testosterone and quantified by liquid chromatography with tandem mass spectrometry.

Physiologic Assessment

For each participant, resting heart rate (measured by pulse oximeter) and blood pressure (standard method using the antecubital artery) were recorded while the participant was seated and quiet. Heart rate variability was determined by a continuous 5-minute electrocardiogram (EKG) recording while the participant was lying quietly in a supine position. Next, participants exercised on a stationary bike for 15 minutes at 70% effort, during which time heart rate was recorded at 5 minutes, 10 minutes, 15 minutes, and 1 minute after completion of the exercise.³⁴⁻³⁶ Heart rate recovery after exercise was calculated as the difference between heart rate at 15 minutes while exercising and 1 minute after exercise. Blood pressure was also recorded 1 minute after exercise to assess blood pressure response to exercise.

For heart rate variability analysis, length of adjacent QRS complexes (normal-to-normal [NN] intervals) were quantified from a continuous 5-minute EKG recording for each participant. Recordings were analyzed for mean interval length between adjacent QRS complexes (mean NN), SD of the mean interval (SDNN), and percentage of differences between adjacent NN intervals that were greater than 50 milliseconds (pNN50).³⁷

Osteopathic Assessment

An osteopathic physician (S.E.D. or J.H.) assessed each participant for the presence of anterior and posterior Chapman points related to the heart, adrenal glands, and ovaries, and spinal viscerosomatic reflexes at spinal levels of the heart (T1-5), adrenal glands (T8-10), and ovaries (T10-11) (**Figure 1**). These regions were selected because PCOS pathophysiology involves overproduction of androgens by the ovaries, and the adrenal glands have also been implicated in contributing to excessive androgen production in some patients. Tissue texture abnormalities palpated at the level of the corresponding organ were considered positive findings of increased sympathetic tones. These Chapman points were defined as small nodular masses, plaque-like changes, or stringiness of the tissue that were tender but nonradiating and remained attached to the deep tissues. The specific locations of the Chapman points were identified by referencing figures 52G-1 and 52G-2 in *Foundations of Osteopathic Medicine*.³³ Viscerosomatic reflexes were assessed by evaluating the paraspinal tissues in segmentally related somatic structures and were considered positive in areas that demonstrated muscle spasm, tenderness, or warmth and moisture.

Data Analysis

Power analysis was conducted with Statistical Decision Tree Power Calculator (QFAB Bioinformatics) to determine the sample size needed to detect a Pearson correlation coefficient of 0.6 with a power of at least .80 and a significance level of .05. GraphPad

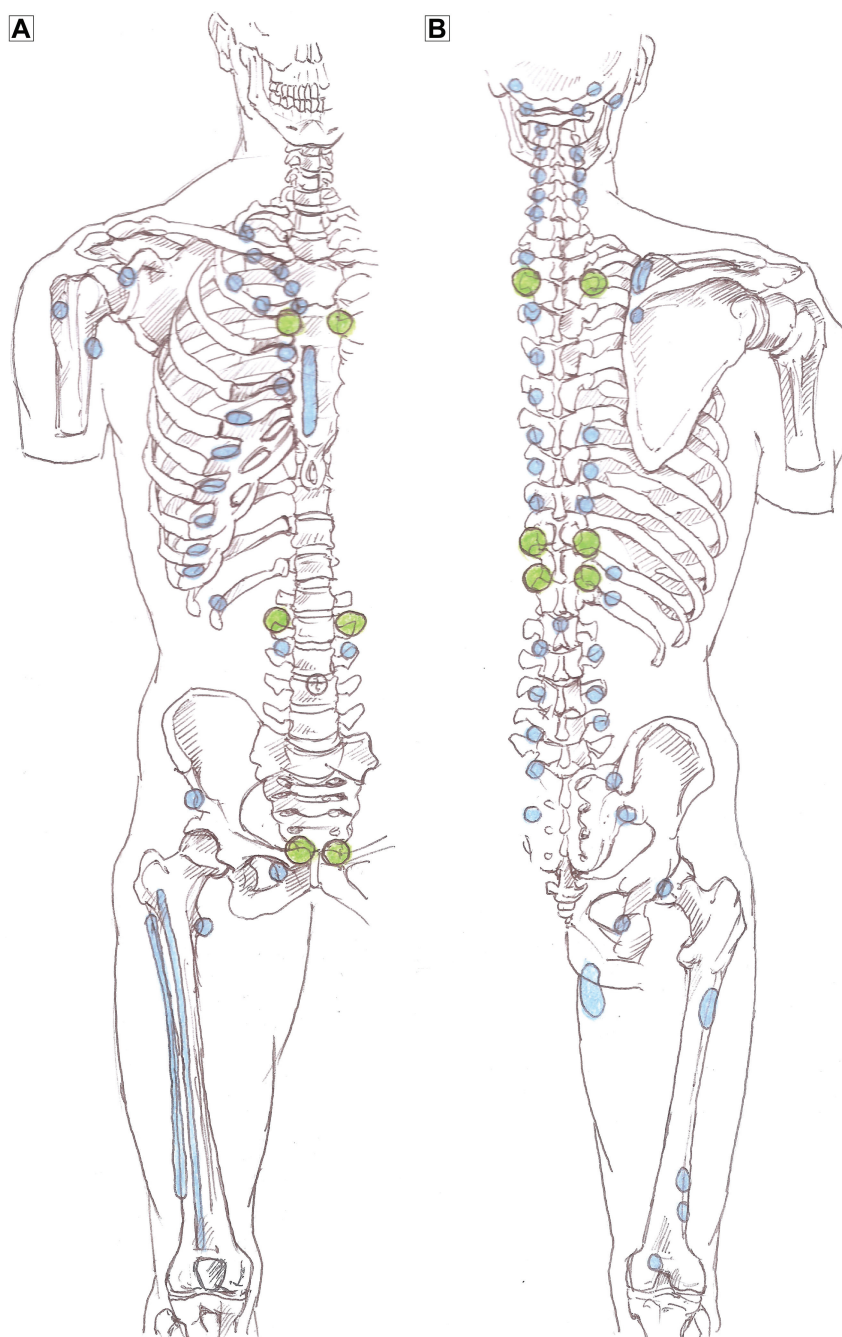


Figure 1.

Osteopathic structural examination of (A) anterior and (B) posterior Chapman points associated with the heart, adrenal glands, and ovaries. Palpation of tissue texture abnormality, asymmetry, restriction of motion, or tenderness associated with the respective spinal segments were considered positive findings for increased sympathetic tone of the corresponding organs. Blue markings indicate Chapman points; green markings indicate Chapman points that were assessed.

Prism 7.03 software was used for data analysis. Physiologic data were compared with reported normal values.^{34,38-41} Correlation between physiologic and osteopathic measurements was determined for each participant by comparing the number of physiologic findings indicative of increased sympathetic tone (defined as a value outside of the reported normal value) with the number of osteopathic findings indicative of increased sympathetic tone associated with the heart, adrenal glands, and ovaries (defined as the presence of a Chapman point or viscerosomatic reflex at these regions). Each measurement indicative of increased sympathetic tone was assigned 1 point. For physiologic measures, 1 point was assigned for each positive finding for any of the following: resting heart rate, resting blood pressure, mean NN by EKG, SDNN by EKG, pNN50 by EKG, postexercise blood pressure, and postexercise heart rate recovery, for a possible maximum of 7 points. For osteopathic measures, 1 point was assigned for each positive finding for any of the following: Chapman point(s) for the heart, adrenal glands, or ovaries, or a viscerosomatic reflex for the heart, adrenal glands, or ovaries, for a possible maximum of 6 points. A Pearson correlation test was used to determine correlation between the number of physiologic and osteopathic findings. Statistical significance was set at $P < .05$.

Results

Twenty-four women participated in the study (Table 1). Participants were aged 20 to 44 years with a confirmed diagnosis of PCOS.

Physiologic Measures of Increased Sympathetic Tone

Table 2 shows the mean physiologic measures for participants. The mean resting heart rate was normal, and the mean blood pressure was just above the normal range at 123/74 mm Hg. Twelve participants had systolic blood pressure greater than 120 mm Hg at rest, consistent with increased sympathetic tone. The

Table 1.
Demographics of Participants in a Study of the Correlation Between Physiologic and Osteopathic Measures of Sympathetic Activity in Women With Polycystic Ovary Syndrome (N=24)

Characteristic	Mean (SD)	Reference Range
Age, y	31.7 (6.9)	NA
Body mass index	34.3 (7.1)	18.5-24.9
Waist-hip ratio	0.83 (0.07)	<0.85
Average menstrual cycle length, d	>35	25-34
Free thyroxin, pg/mL	7.79 (5.8)	0.2-5.0

mean heart rate recovery after moderate-intensity exercise was normal, with 6 participants having a heart rate recovery of 12 seconds or less 1 minute after exercise. The mean postexercise blood pressure was slightly above the reported normal range of 133/77 mm Hg, with 13 participants exceeding a systolic blood pressure of 130 mm Hg. The results of the heart rate variability analysis indicated a slighter lower mean NN and SDNN compared with normal values, suggesting decreased heart rate variability in the study population and consistent with increased sympathetic tone. The mean pNN50 value was within reported normal range, although 10 participants (40%) had a pNN50 value below normal. Together, these findings indicate increased sympathetic tone by physiologic measures in this group of participants, with variability in positive parameters among all participants.

Osteopathic Measures of Increased Sympathetic Tone

Participants were found to have anterior and posterior Chapman points associated with the heart, adrenal glands, and ovaries, with the greatest number of positive findings for the heart and ovaries (Table 3). In addition, positive viscerosomatic reflexes were found in regions associated with the heart, adrenal glands, and

Table 2.
Physiologic Measures of Sympathetic Tone in
Participants With Polycystic Ovary Syndrome
(N=24)

Measure	Mean (SD)	Reference Range
Resting HR, beats/min	75.3 (11.3)	60-100
Resting SBP, mm Hg	122.8 (11.0)	≤120/80
Resting EKG, mean NN, ms	0.8306 (0.106)	0.875-0.977
Resting EKG, SDNN (ms)	0.050 (0.024)	0.072-0.107
Resting EKG, pNN50, %	23.1 (18)	12-23
SBP after exercise, mm Hg	132.8 (10.1)	≤130/85
HRR after exercise, s	24.7 (12.3)	>12

Abbreviations: EKG, electrocardiogram; HR, heart rate; HRR, heart rate recovery; NN, normal-to-normal intervals; pNN50, percentage of differences between adjacent NN intervals that were greater than 50 ms; SBP, systolic blood pressure; SDNN, SD of the mean interval.

ovaries (Figure 2). Together, these findings support osteopathic structural measures of increased sympathetic tone in women with PCOS.

Correlation Between Physiologic and Osteopathic Measures

Since the physiologic and osteopathic assessments each indicated the presence of increased sympathetic tone in the study population, we wanted to determine whether individual participants with physiologic measures of increased sympathetic tone also exhibited osteopathic measures of the same. For each participant, 1 point was assigned for each physiologic measure indicating increased sympathetic tone, and 1 point was assigned for each osteopathic measure indicating increased sympathetic tone.

We found that, although participants had both positive physiologic and osteopathic findings, there was no significant correlation between the number of osteopathic findings and the number of physiologic findings associated with increased sympathetic tone ($r=0.260$; $P=.110$). However, for most participants, the number of osteopathic findings associated with increased

sympathetic tone matched or exceeded the number of physiologic findings (Figure 3).

Discussion

Polycystic ovary syndrome is a common hormone disorder that affects many aspects of health. In addition to reproductive and endocrine issues, females with PCOS are reported to have increased sympathetic tone, which may exacerbate the associated symptoms and further stimulate the pathogenesis of the disorder, as well as increase the risk for cardiovascular sequelae, such as hypertension; endothelial dysfunction; increased arterial stiffness, basal heart rate, blood pressure, and mean arterial pressure; and decreased heart rate variability.^{15,17,18,22,42-44} Furthermore, increased ovarian sympathetic tone has been reported in females as well as animal models for PCOS, and the adrenal glands and heart are additional proxies for detecting increased sympathetic tone.^{15-19,27-32} Sympathetic tone is not often specifically assessed or addressed in this patient population. Physiologic methods to assess sympathetic tone include measuring blood pressure at rest as well as after exercise, measuring heart rate recovery after exercise, determining resting heart rate variability, measuring adrenergic metabolites in the serum or urine, and measuring muscle sympathetic nerve activity.^{17,20} However, as these methods can be time- and labor-intensive and are not always feasible during an office visit, we sought to determine whether we could instead use osteopathic tools to assess for increased sympathetic tone in females with PCOS.

We found physiologic evidence of increased sympathetic tone in our study population of women with PCOS, which is consistent with previous reports that females with PCOS have increased sympathetic tone.^{15,17,18,43-45}

We determined the presence of Chapman points and viscerosomatic reflexes associated with the heart, adrenal glands, and ovaries in the study participants. We focused on adrenal glands and ovaries because ovaries are the major site of excess androgen

Table 3.
Osteopathic Examination of Chapman Points in
Participants With Polycystic Ovary Syndrome
(N=24)

Organ	Chapman Points, No.		
	Anterior	Posterior	Total
Heart	9	7	16
Adrenal glands	8	3	11
Ovaries	15	7	22

production characteristic of PCOS, and adrenal glands may also contribute to hyperandrogenemia. We included regions associated with the heart because patients with PCOS are at increased risk for developing cardiovascular sequelae. Moreover, the adrenal glands and heart are proxies for detecting increased sympathetic tone. Increased ovarian sympathetic tone has been reported in females as well as animal models of PCOS. Of note, all of the participants had some evidence of increased sympathetic tone either by the presence of Chapman points (80% of participants), viscerosomatic reflexes (100% of participants), or both (80% of participants). The number of Chapman points associated with each of the organs or glands examined varied from participant to participant, as did the extent of the viscerosomatic reflexes. However, these findings indicate that women with PCOS may exhibit increased sympathetic tone that can be detected by osteopathic structural examination, perhaps even before systemic physiologic changes in sympathetic tone are detectable.

When comparing the physiologic findings with the osteopathic findings of increased sympathetic tone, we did not detect a significant correlation between the number of findings for each assessment method. One possible explanation is the physiologic measures of sympathetic tone used primarily relate to the cardiovascular effects of increased sympathetic tone, whereas the osteopathic measures relate to not only the heart but also to the adrenal glands and ovaries. A more in-depth assessment of both physiologic and osteopathic parameters associated with increased sympathetic tone

may reveal a significant correlation between these types of measures. Another possible explanation is that sympathetic hyperactivity was regional rather than generalized in our population. Perhaps it had not yet translated into measurable cardiovascular effects in some of these women, but it was palpable by structural examination of Chapman points and viscerosomatic reflexes. Interestingly, 3 women in our study had no or only 1 physiologic finding associated with increased sympathetic tone, yet they had multiple osteopathic findings associated with increased sympathetic tone. If osteopathic structural aberrations are detectable before physiologic evidence of sympathetic hyperactivity, this type of assessment may be quite valuable in detecting increased sympathetic tone before an impact on the cardiovascular system is physiologically evident. Furthermore, earlier detection of increased sympathetic tone, even before overt physiologic symptoms are present, could potentially represent an important window of time for intervention and possibly mitigate some symptoms and sequelae associated with PCOS. Indeed, somatic dysfunction leading to tissue texture changes could promote or further propagate the disease process and underscores the importance of

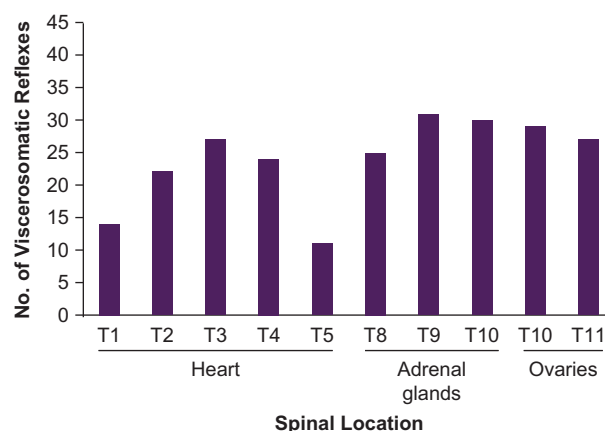


Figure 2.

Total number of viscerosomatic reflexes associated with various spinal levels for the heart, adrenal glands, and ovaries for all participants combined. They were assessed by evaluating the paraspinal tissues in segmentally related somatic structures and were considered positive in areas that demonstrated muscle spasm, tenderness, or warmth and moisture.

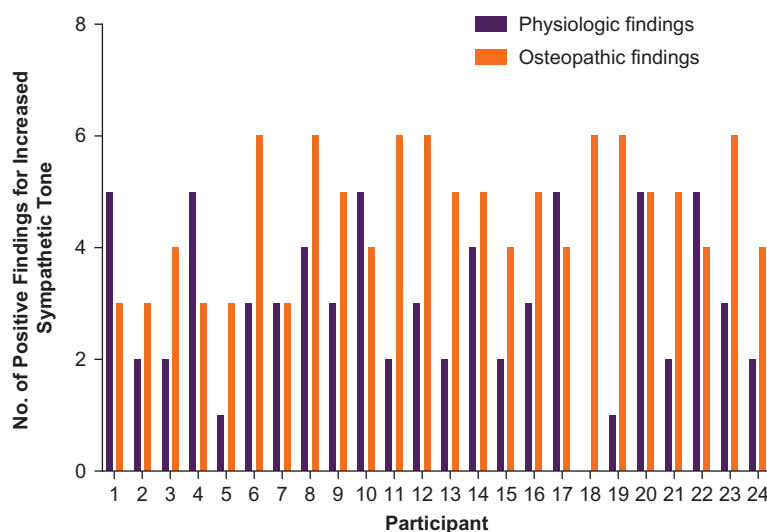


Figure 3.

Correlation between physiologic (heart rate, blood pressure, heart rate variability, heart rate recovery) and osteopathic (presence of a Chapman point or viscerosomatic reflex) findings indicative of increased sympathetic tone in participants with polycystic ovary syndrome. Each measurement indicative of increased sympathetic tone was assigned 1 point value, for a maximum total of 7 points possible for physiologic measures and 6 points possible for osteopathic measures.

detection with the possibility of intervention before more serious sequelae develop.⁴⁶ Manipulation of Chapman points, rib raising, and other forms of osteopathic manipulative treatment—and the effects of these interventions on sympathetic tone and other PCOS-related symptoms—will be investigated in a future study.

The relationship between PCOS and increased sympathetic tone has been well documented, though the specifics of how this relationship may contribute to the pathogenesis of the disorder continue to be investigated. Females with PCOS are reported to have both generalized as well as regional increases in sympathetic outflow. Evidence of generalized increased sympathetic tone includes greater levels of urinary catecholamines and their metabolites, attenuated heart rate variability, poorer heart rate recovery after exercise, hypertension, and greater sympathetic nerve activity in muscles.^{44,47–49} Females with PCOS and animal models of the disorder also indicate regional increases in sympathetic tone, including increased ovarian sympathetic outflow. A study⁵⁰ of rats with and without

PCOS undergoing in vitro fertilization showed that nerve growth factor (NGF), a sympathetic neurotrophin and marker for sympathetic nerve activity, was elevated in the follicular fluid and granulosa cell-conditioned medium in women with PCOS compared with women without the disorder. Rodent models of PCOS have demonstrated ovarian synthesis of NGF increases after sympathetic activation of the ovaries, and NGF can alter follicular activity and increase androgen synthesis in the ovaries, possibly contributing to the ovarian pathology associated with the disorder.^{19,27,28} Transection of the superior ovarian nerve in a rodent model restored estrous cyclicity and ovulation and attenuated androgen responses to human chorionic gonadotropin and a β -adrenergic receptor agonist.²⁷ Together, these findings underscore the role of the sympathetic nervous system in PCOS pathogenesis and highlight the importance of detecting increased sympathetic tone in patients with PCOS.

One limitation of our study was that we examined a population of women with PCOS but did not examine a control group of age- and women-matched by age

and body mass index (BMI). However, the normal values for the physiologic parameters examined herein are well documented,^{34,38-41} and the presence of any Chapman points or viscerosomatic reflexes is considered an abnormal finding.^{33,51} The presence of positive viscerosomatic reflexes in all patients may be explained by the overlap of spinal cord regions innervating the various visceral organs. For example, the sympathetic preganglionic neurons innervating the heart arise from T1-T5, and the regions innervating the pulmonary plexus and esophagus arise from T2-T7 and T2-T8, respectively. Another limitation is that the physiologic and osteopathic measurements were not made on the same day for all participants. The largest gap between measurements was 7 days for 3 of the participants, with most participant measurements occurring on the same day or consecutive days. Future studies will investigate whether the time between the physiologic and osteopathic assessments has an impact on the correlation between the 2 measures.

The participants were predominately overweight or obese, which is common in this patient population and can exacerbate sympathetic hyperactivity, as well as symptoms associated with PCOS.^{23,25,26,52-57} Metabolic dysfunction is common in women with PCOS, not only in terms of obesity but also in that they are more likely than age- and BMI-matched controls to exhibit insulin resistance and hyperinsulinemia. These women have a much greater risk of type 2 diabetes mellitus developing.¹² We did not specifically assess for Chapman points or viscerosomatic reflexes associated with the pancreas, which may have contributed to or yielded additional positive findings of increased sympathetic tone by osteopathic structural examination, as has previously been reported in patients with type 2 diabetes.⁵⁸ However, physiologic and osteopathic findings of increased sympathetic tone were noted even in the participants who had a normal BMI, in agreement with a previous report that sympathetic hyperactivity is a common finding in women with PCOS independent of BMI.¹⁸ Our findings underscore the importance of the osteopathic structural

examination to detect sympathetic dysfunction, as this assessment method may allow detection before physiologic manifestation of symptoms, and it is a relatively quick, inexpensive, and practical tool that can be used during an office visit.

Conclusion

An osteopathic structural examination provides a unique and valuable tool for detecting increased sympathetic tone in women with PCOS, even in the absence of overt physiologic signs of sympathetic dysfunction in this population. Information regarding sympathetic tone provides a more complete picture of a patient's well-being and allows physicians to better counsel and guide the patient toward improved health. Furthermore, this information may allow for earlier use of therapeutic options, such as osteopathic manipulative treatment, to improve sympathetic tone and overall patient health.

Author Contributions

Drs Davis, Hendryx, and Speelman provided substantial contributions to conception and design, acquisition of data, or analysis and interpretation of data; Drs Davis and Speelman drafted the article or revised it critically for important intellectual content; all authors gave final approval of the version of the article to be published; and all authors agree to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

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