Postprint of a Study on Electroacupuncture Combined with Shaoyao Gancao Tang Improving Synaptic Remodeling in Rats with Post-Stroke Limb Spasticity

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Abstract

Background Currently, the combination of acupuncture and medication demonstrates favorable efficacy in treating post-stroke limb spasticity, yet its underlying mechanism remains unclear.

Objective To investigate the effects of electroacupuncture combined with Shaoyao Gancao Decoction on motor function and synaptic remodeling in the M1 region of rats with post-stroke limb spasticity.

Methods From November 2022 to June 2023, 72 healthy male Sprague-Dawley (SD) rats, aged 7 weeks, were selected, with 60 rats ultimately included. The rats were randomly divided into blank group, model group, electroacupuncture group, baclofen group, traditional Chinese medicine group, and acupuncture-medicine combination group using a random number table method. Interventions were administered from day 1 to day 6 post-modeling. Rats in the electroacupuncture group received electroacupuncture at the affected side "Quchi" (LI11) and "Yanglingquan" (GB34) with dense wave at 100 Hz for 30 min per session, once daily. The traditional Chinese medicine group received Shaoyao Gancao Decoction gavage at 4.2 g/kg, once daily. The baclofen group received baclofen solution gavage at 5.25 mg/kg, once daily. The acupuncture-medicine combination group received electroacupuncture combined with traditional Chinese medicine gavage using the same methods and dosages as above, once daily. Catwalk gait analysis was performed before modeling and on days 1, 3, and 6 post-modeling. Following data collection on day 6, the cerebral cortex M1 region was harvested for analysis. Nissl staining was employed to observe neuronal damage in the M1 region. Fluorescence quantitative PCR and Western blotting were utilized to detect the expression levels of brain-derived neurotrophic factor

(BDNF), γ-aminobutyric acid (GABA), and neurotrophin-3 (NT3) in the M1 region. Electron microscopy was used to observe synaptic morphology in the M1 region.

Results On day 3 post-modeling, the average paw intensity in the acupuncture-medicine combination group was greater than that in the blank group, and on day 6 post-modeling, the percentage of four-paw support in the acupuncture-medicine combination group was higher than that in the blank group ($P<0.05$). Nissl staining revealed that neuronal damage in the acupuncture-medicine combination group was similar to that in the blank group. Fluorescence quantitative PCR results demonstrated that GABA expression level in the acupuncture-medicine combination group was lower than that in the blank group, while BDNF expression level was higher than that in the blank group ($P<0.05$). Western blotting results showed that relative protein contents of GABA, NT3, and BDNF in the model group, electroacupuncture group, and baclofen group were lower than those in the blank group ($P<0.05$); relative protein contents of GABA, NT3, and BDNF in the traditional Chinese medicine group and acupuncture-medicine combination group were higher than those in the model group ($P<0.001$). Electron microscopy revealed that synaptic morphology in the acupuncture-medicine combination group was relatively intact, with abundant vesicles, clear pre- and post-synaptic membrane boundaries, and longer contact boundaries, showing higher similarity to the blank group.

Conclusion Electroacupuncture combined with Shaoyao Gancao Decoction can improve motor function in rats with post-stroke limb spasticity, and its mechanism may involve enhancement of synaptic plasticity.

Full Text

Electroacupuncture Combined with Shaoyao Gancao Decoction Improves Synaptic Remodeling in Rats with Post-Stroke Limb Spasticity

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Abstract

Background: Currently, acupuncture combined with traditional Chinese medicine (TCM) herbs shows excellent outcomes in treating post-stroke spasticity, but the underlying mechanism remains unclear.

Objective: To observe the effects of Shaoyao Gancao Decoction combined with electroacupuncture on motor function and synaptic plasticity in the M1 cortex of rats with post-stroke limb spasticity.

Methods: Between November 2022 and June 2023, 72 healthy male Sprague-Dawley rats (7 weeks old) were purchased, with 60 rats ultimately included in the study. The animals were randomly divided into six groups (n=10 each): blank, model, electroacupuncture, baclofen, TCM herb, and electroacupuncture plus TCM herb. Interventions were administered from day 1 to day 6 post-modeling. The electroacupuncture group received electroacupuncture at "Quchi" (LI11) and "Yanglingquan" (GB34) on the affected side using dense wave at 100 Hz for 30 minutes daily. The TCM herb group received Shaoyao Gancao Decoction gavage at 4.2 g/kg daily. The baclofen group received baclofen solution at 5.25 mg/kg daily. The electroacupuncture plus TCM herb group received both electroacupuncture and herbal gavage as described above. Catwalk gait analysis was performed before modeling and on days 1, 3, and 6 post-modeling. After data collection on day 6, the M1 cortical region was harvested. Nissl staining was used to observe neuronal damage in the M1 region. Fluorescence quantitative PCR and Western blotting were employed to detect brain-derived neurotrophic factor (BDNF), γ-aminobutyric acid (GABA), and neurotrophin-3 (NT3) levels. Electron microscopy was used to observe synaptic growth in the M1 region.

Results: On day 3 post-modeling, the electroacupuncture plus TCM herb group showed greater average footprint intensity than the blank group; on day 6, this group demonstrated a higher percentage of four-paw support compared to the blank group ($P<0.05$). Nissl staining revealed that neuronal damage in the electroacupuncture plus TCM herb group was similar to that in the blank group. qPCR results showed that the electroacupuncture plus TCM herb group had lower GABA expression and higher BDNF expression than the blank group ($P<0.05$). Western blotting revealed that the model, electroacupuncture, and baclofen groups had lower relative protein levels of GABA, NT3, and BDNF compared to the blank group ($P<0.05$), while the TCM herb and electroacupuncture plus TCM herb groups had higher levels than the model group ($P<0.001$). Electron microscopy showed that the electroacupuncture plus TCM herb group exhibited relatively intact synaptic morphology with abundant vesicles, clear pre- and post-synaptic membrane boundaries, and long contact boundaries, closely resembling the blank group.

Conclusion: Electroacupuncture combined with Shaoyao Gancao Decoction can improve motor function in rats with post-stroke limb spasticity, possibly by enhancing synaptic plasticity.

Keywords: Stroke; Electroacupuncture; Shaoyao Gancao Decoction; Synapses; Limb spasticity

Introduction

Post-stroke limb spasticity commonly manifests as increased muscle tone in the biceps brachii and quadriceps femoris [?]. Without early intervention, it often leads to residual limb disability, imposing a tremendous burden on patients and society. The pathogenesis of post-stroke limb spasticity involves liver-kidney yin deficiency and wind-phlegm-stasis obstruction, with treatment typically focusing on supplementing qi, nourishing yin, extinguishing wind, and relaxing muscles to activate blood circulation. Consequently, contemporary physicians have explored decoctions such as Xiaoxuming Decoction, Buyang Huanwu Decoction, and Huangqi Guizhi Wuwu Decoction for their qi-supplementing and blood-activating effects. Although some researchers have investigated Shaoyao Gancao Decoction and Dihuang Yinzi Decoction, most have approached from the perspective of nourishing kidney and liver to soften tendons, with few studies exploring the scientific mechanisms in combination with electroacupuncture therapy [?].

Shaoyao Gancao Decoction first appeared in the "Treatise on Cold Damage Diseases" in the chapter "Differentiation and Treatment of Taiyang Disease," stating: "If the extremities become warm, administer Shaoyao Gancao Decoction, and the feet will extend." Clinically, it is commonly used to treat spasmodic pain, muscle tone disorders, and gastrointestinal diseases [?]. Studies have also found that Shaoyao Gancao Decoction can inhibit inflammatory responses after cerebral ischemia-reperfusion in rats and improve neuronal activity in the infarcted area [?]. The liver and gallbladder are interior-exteriorly related. Quchi is the He-sea point of the Large Intestine Meridian, while Yanglingquan is the He-sea point of the Gallbladder Meridian. Previous research has found that electroacupuncture at "Yanglingquan" and "Quchi" can upregulate glutamate and its receptor expression in the hippocampus, promote synaptic remodeling in the brain, and alleviate limb spasticity [?]. Li Chan in the Ming Dynasty's "Introduction to Medicine" stated: "The heart connects with the gallbladder, and the liver connects with the large intestine" [?]. Based on the theory of "organ interconnection," this study designed a protocol combining Shaoyao Gancao Decoction with electroacupuncture at "Quchi" and "Yanglingquan" to explore spasticity inhibition and synaptic plasticity-related indicators in the M1 motor cortex, comparing it with the commonly used Western medicine baclofen and employing the Catwalk gait analysis system for behavioral data collection.

1. Materials and Methods

1.1 Experimental Animals and Grouping From November 2022 to June 2023, 72 clean-grade healthy male Sprague-Dawley rats (7 weeks old, weight 270$±10g)werepurchasedfromGuangdongVitalRiverLaboratoryAnimalTechnologyCo., Ltd.[LicenseNo.SC 0063]andhousedattheSouthChinaResearchCenterforAcupunctureandMoxibustionofGuangzhouUniversity 12 h/12 h light-dark cycle) with free access to food and water. After one week of acclimatization and twice-weekly gait behavior training, 5 rats that could not continuously run through the data collection area were excluded. Using a random number table, rats were divided into a blank group and a modeling group. After modeling, Zea-Longa scoring was performed, and animals with scores of 2-3 were included, while 7 rats that died or were unqualified were excluded. The successfully modeled rats were then randomly divided into model, electroacupuncture, baclofen, electroacupuncture plus TCM herb, and TCM herb groups (n=10 each). All experimental procedures strictly followed animal ethics regulations (Animal Ethics Approval No. 00343960, Guangzhou University of Chinese Medicine).

1.3 Modeling Method Except for the blank group, the other five groups underwent a modified photothrombotic stroke plus anhydrous ethanol internal capsule injection method to establish a post-stroke limb spasticity rat model [?]. Rats were anesthetized with isoflurane (3-4% for induction, 1.5-2.5% for maintenance), shaved, disinfected, and draped. In the prone position on a stereotaxic apparatus, the bregma, left parietal bone, and parietal-frontal suture were exposed. The skull surface was cleaned with saline-soaked cotton swabs until clear. The skull and incisors were fixed, and the left cerebral cortex M1 region was accurately located according to the *Paxinos and Watson Rat Brain Atlas* (forelimb coordinates: 2.30 mm left of bregma, 1.00 mm anterior; hindlimb coordinates: 2.00 mm left of bregma, 0.00 mm anterior). A 0.80 mm diameter skull drill was used to create a hole without penetrating the dura mater. Rose bengal (15 mg/mL, 80 mg/kg) was injected intraperitoneally. A 532 nm fixed-wavelength laser was used to irradiate the drilled site (diameter 2 mm, distance 1 cm from dura mater, intensity 54 mW, duration 6 min). The position was then adjusted to the internal capsule (coordinates: 3.10 mm left of bregma, 2.00 mm caudal, 7.20 mm vertical). Another hole was drilled, and an ultra-fine needle penetrated the dura to a depth of 7.2 mm to inject 80 L anhydrous ethanol over 5 minutes. After 1 minute, the microinjector was removed, hemostasis was achieved, the hole was sealed with bone wax, the wound was cleaned with 0.9% saline, and sutured layer by layer.

1.4 Drug Preparation and Gavage Doses Traditional Chinese Medicine: Herbal slices of Shaoyao (peony) and Zhigancao (processed licorice) were prepared in a 3:1 ratio. The slices were soaked in distilled water for 1 hour, rapidly boiled with strong heat, then simmered for 2 hours and filtered through 4-layer gauze. The residue was re-decocted, filtered, and combined with the first extract, then concentrated by rotary evaporation to 1 g/mL crude drug concentration and stored at 4°C. Based on human-to-animal dose conversion coefficients, gavage began on day 1 post-modeling at 4.2 g/kg daily for 6 days [?].

Baclofen: Baclofen tablets were prepared as a 1 mg/mL solution. Based on dose conversion, gavage began on day 1 post-modeling at 5.25 mg/kg daily for 6 days [?].

1.5 Acupuncture Method According to the *Experimental Acupuncture* animal acupoint atlas [?], the affected side Yanglingquan (GB34) and Quchi (LI11) were located. Under isoflurane anesthesia (1.5-2.5%), acupoints were disinfected and needled with 0.5-inch acupuncture needles (Huatuo brand, 0.25$×$13 mm) using perpendicular insertion. Needles were rotated 90-180° at 60-90 rotations/minute for 1 minute. Electrodes were attached with parameters: dense wave, 100 Hz, current intensity 1-3 mA (adjusted to produce slight limb tremor). Treatment duration was 30 minutes daily for 6 days.

1.6 Intervention and Sampling **Interventions:** The blank and model groups received distilled water gavage (1 mL/d) and low-flow anesthesia for 30 min/d. The electroacupuncture group received distilled water gavage (1 mL/d) plus electroacupuncture. The baclofen group received baclofen solution gavage plus low-flow anesthesia for 30 min/d. The electroacupuncture plus TCM herb group received both electroacupuncture and herbal gavage. The TCM herb group received herbal gavage plus low-flow anesthesia for 30 min/d.

Sampling: After 6 days of continuous intervention, sampling was completed within 24 hours. Animals were euthanized with 10% chloral hydrate (4 mL/kg, i.p.), and brains were rapidly dissected on ice. Part of the left cortical M1 region (1 mm^3) was harvested and fixed in 2.5% glutaraldehyde. The remaining M1 tissue was snap-frozen in liquid nitrogen and stored at -80°C. Some whole brains were rinsed with pure water and fixed in 10% paraformaldehyde.

1.7 Index Collection

1.7.1 Catwalk Gait Behavior Testing Gait information was collected before modeling and on days 1, 3, and 6 post-modeling. Animals were placed in the experimental room 2 hours prior to ensure a quiet, odor-free environment. System parameters were adjusted to ensure animals moved continuously across the glass plate with their head forward, with at least 3 qualified data collections per session. The observation indices included average footprint intensity, maximum contact area, swing speed, three-paw support phase ratio, and four-paw support percentage [?].

1.7.2 Nissl Staining Paraffin sections were deparaffinized, stained with toluidine blue for 10 minutes, differentiated with 95% alcohol, dried, cleared with xylene for 5 minutes, and mounted with neutral resin [?].

1.7.3 Fluorescence Quantitative PCR Detection Tissues were lysed and RNA was extracted and reverse-transcribed. Primers (2 L) were prepared into a PCR reaction system according to kit instructions. The reaction sequence was: denaturation at 94°C for 2 min, followed by 45 cycles of 94°C for 5 s and 60°C for 30 s, then cooling. The 2^-ΔΔCt method was used to analyze target gene expression. Primer sequences are shown in Table 1 .

1.7.4 Western Blotting Detection Tissues were washed, ground, and protein was extracted. Protein concentration was determined by BCA assay. Gels were prepared according to instructions, and antibodies were diluted to appropriate concentrations (1:1000). Samples underwent gel electrophoresis at 100 V. PVDF membranes were activated in methanol for 1 minute, then transferred in ice-cold transfer buffer. Membranes were blocked in 5% skim milk for 30 minutes with shaking, incubated with primary antibodies (rabbit anti-GABA, NT-3, BDNF; Abcam) overnight at 4°C, washed 3 times with TBST (5 min each), incubated with HRP-labeled goat anti-rabbit IgG secondary antibody for 30 minutes, washed again, and developed with ECL solution. Image-Pro Plus software was used to analyze band optical density.

1.7.5 Electron Microscopy Observation of M1 Synaptic Growth M1 tissue fixed in 2.5% glutaraldehyde was post-fixed with osmium tetroxide, dehydrated, infiltrated, embedded, sectioned ultrathin (80 nm), double-stained with lead and uranium, dried overnight at room temperature, and imaged under transmission electron microscopy.

1.8 Statistical Methods SPSS 20.0 software was used for data analysis. Normally distributed data were analyzed by one-way ANOVA with LSD post-hoc test for pairwise comparisons. Non-normally distributed data were analyzed by rank-sum test. $P<0.05$ was considered statistically significant.

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2. Results

2.1 Catwalk Gait Analysis Significant differences were observed among the six groups in average footprint intensity and maximum contact area on day 3 post-modeling, and in swing speed, three-paw support phase ratio, and four-paw support percentage on day 6 ($P<0.05$). Specifically, the model, baclofen, and electroacupuncture plus TCM herb groups showed greater average footprint intensity than the blank group on day 3 ($P<0.05$). The model group exhibited larger maximum contact area than the blank group on day 3 ($P<0.05$). The model, electroacupuncture, and TCM herb groups showed lower swing speed than the blank group on day 6 ($P<0.05$). The electroacupuncture group had a lower three-paw support phase ratio than the blank group on day 6 ($P<0.05$). The electroacupuncture plus TCM herb group displayed a higher four-paw support percentage than the blank group on day 6 ($P<0.05$). No significant differences were found among groups in swing speed, three-paw support phase ratio, or four-paw support percentage on day 3, nor in footprint intensity or maximum contact area on day 6 ($P>0.05$). Detailed data are presented in Table 2 .

2.2 Nissl Staining In the blank group, neurons were intact with patchy or tigroid Nissl bodies visible in the cytoplasm and prominent nucleoli. The model group showed condensed and necrotic Nissl bodies with dissolved cytoplasm forming vacuolar structures. Compared with the model group, the electroacupuncture and baclofen groups had some intact cell bodies but obvious cellular edema. The TCM herb and electroacupuncture plus TCM herb groups showed few vacuolar structures, better neuronal morphology, clear nucleoli, and visible Nissl bodies, with the electroacupuncture plus TCM herb group having more neurons (Figure 1 [Figure 1: see original paper]).

2.3 qPCR Detection of GABA, NT3, and BDNF Expression Significant differences in GABA and BDNF expression levels were observed among the six groups ($P<0.05$). The baclofen group showed lower GABA expression than the blank group ($P<0.05$). The electroacupuncture plus TCM herb group exhibited lower GABA expression and higher BDNF expression compared to the blank group ($P<0.05$). No significant difference in NT3 expression was found among groups ($P>0.05$). Data are shown in Table 3 .

2.4 Western Blotting Detection of Protein Levels Significant differences in relative protein levels of GABA, NT3, and BDNF were observed among the six groups ($P<0.05$). The model, electroacupuncture, and baclofen groups showed lower protein levels than the blank group ($P<0.05$), while the TCM herb and electroacupuncture plus TCM herb groups had higher levels than the model group ($P<0.001$). Results are shown in Figure 2 [Figure 2: see original paper] and Table 4 .

2.5 Electron Microscopy Observation of M1 Synaptic Growth The blank group exhibited abundant synapses with numerous vesicles, intact and clear pre- and post-synaptic membrane boundaries, and long contact boundaries. The model group showed swollen synapses with few poorly shaped vesicles and fused membranes. Compared with the model group, the electroacupuncture group had clearer membrane boundaries, the baclofen group showed better synaptic morphology, and the TCM herb group had clearer boundaries with abundant vesicles and more synapses. The electroacupuncture plus TCM herb group demonstrated the most intact synaptic morphology with abundant vesicles, clear membrane boundaries, and long contact boundaries, most closely resembling the blank group (Figure 3 [Figure 3: see original paper]).

3. Discussion

Post-stroke limb spasticity often leads to residual disability, and early intervention can reduce motor impairment. The pathogenesis of stroke involves six factors: deficiency (yin/qi deficiency), fire (liver/heart fire), wind (liver wind), phlegm (wind-phlegm/damp-phlegm), qi (reversed qi), and blood (blood stasis) [?]. Ming Dynasty physician Li Chan proposed that "liver disease should be treated by dredging the large intestine" [?]. Quchi, as the He-sea point of the Large Intestine Meridian, is clinically used not only for Large Intestine Meridian disorders but also to relieve upper limb spasticity, inflammation, and pain [?]. Yanglingquan is the He-sea point of the Gallbladder Meridian and a commonly used acupoint for treating stroke and limb channel disorders [?]. Shaoyao Gancao Decoction, listed in the first batch of the "Catalogue of Ancient Classical Prescriptions" published by the National Administration of Traditional Chinese Medicine [?], has been preserved for its clinical efficacy. Analysis of ancient and modern literature on Shaoyao Gancao Decoction using Sankey diagrams reveals its main indications, ranked by frequency, are cold damage diseases, abdominal pain, and cough, with limb spasm after yin injury categorized under cold damage patterns [?]. Modern research also demonstrates that Shaoyao Gancao Decoction has significant anti-inflammatory, antispasmodic, analgesic, and hepatoprotective effects [?], and combined with exercise training [?] or acupuncture [?] can effectively reduce post-stroke muscle tone.

Therefore, based on previous clinical and mechanistic studies, this study designed a protocol combining Shaoyao Gancao Decoction with electroacupuncture at "Quchi" and "Yanglingquan" from the perspective of "organ interconnection" theory to explore its scientific mechanisms.

The Catwalk gait system, with its rich and intuitive data collection capabilities, is commonly used to evaluate motor function in neurological disorders [?]. This study selected velocity, support, and contact area-related indices for analysis. Average footprint intensity represents the mean pressure exerted by rat footprints. When one limb is spastic and cannot support body weight, the remaining three limbs must bear more weight, inevitably increasing pressure on the glass plate. Maximum contact area refers to the largest contact area between the paw and glass plate during a collection period. When a limb cannot support body weight, the proximal limb contacts the plate to compensate for insufficient distal support, dragging the distal part forward and reducing paw-plate contact area. Swing speed refers to the alternating limb movement speed during locomotion. When muscle groups are rigid, mechanical homeostasis is disrupted, motor coordination decreases, alternating swings are obstructed, and speed declines. The three-paw support phase ratio refers to the percentage of three-paw support time in the entire stride cycle. Quadrupeds normally walk with at least one limb in motion; three-paw support, as the most stable triangular structure, appears frequently during normal walking [?]. During muscle spasm, the affected limb may develop joint rigidity causing distal paw dragging, correspondingly reducing three-paw support time. The four-paw support percentage refers to the proportion of the cycle with all four paws simultaneously supporting the glass plate, commonly seen during static states or gait transition intervals. Frequent four-paw support during movement indicates poor limb coordination requiring frequent adjustment to maintain stability. In this study, the electroacupuncture plus TCM herb group showed lower three-paw support phase ratio and extremely high four-paw support percentage, but combined with other indices such as swing speed, this suggests that as limb spasm relieved and swing speed increased, stability decreased, leading to frequent gait adjustments. However, the ability to maintain body stability and complete the entire collection area indicates good overall motor adjustment effects in the electroacupuncture plus TCM herb group.

In the central nervous system, microglia-induced neuroinflammation accelerates neuronal apoptosis [?]. Nissl bodies, located in neuronal soma and dendrites and composed of rough endoplasmic reticulum and free ribosomes, are most sensitive to neuronal injury, showing dissolution and disappearance. Therefore, Nissl bodies are commonly used as markers for observing neuronal damage and regeneration [?]. GABA is the main inhibitory neurotransmitter in the cerebral cortex, primarily distributed in inhibitory synaptic vesicles and neuronal soma and dendrites, working with glutamate to maintain excitation-inhibition balance in the central nervous system. Abnormal GABA metabolism after stroke can cause synaptic dysfunction, reduced supraspinal inhibition, and abnormal lower-level neuronal excitation, representing a possible mechanism of post-stroke spasticity [?]. Microglia can balance the maturation and remodeling of inhibitory and excitatory synapses [?]. BDNF is one of the most widely distributed neurotrophic factors in the mammalian brain, playing important roles in synaptic development, growth, and plasticity, and influencing synaptic transmission efficacy [?]. Using transgenic technology to overexpress BDNF in microglia improved blood supply to the injured spinal cord, promoted neovascularization, and enhanced hindlimb motor function recovery in mice [?]. NT3 is an important member of the neurotrophin family, mediating glial cell growth along neuronal axons and promoting axonal regeneration [?]. NT3 injection into the affected limbs of stroke animals can induce surviving spinal cord neurons to grow across the midline toward the affected side [?]. NT3 can also inhibit immune responses in peripheral blood CD4+ and CD8+ T cells and reduce monocyte and T cell percentages during stress in acute stroke patients [?]. Synapses, as the primary sites of neural signal transmission, require structural integrity for normal function. Synaptic number, pre- and post-synaptic membrane contact area, postsynaptic density thickness, cleft width, and vesicle quantity are important indicators reflecting synaptic plasticity [?]. Microglia can regulate synaptic maturation and function and are closely related to inflammatory responses, exerting anti-inflammatory or pro-inflammatory effects in different environments [?]. Previous studies found that high-frequency electroacupuncture could increase postsynaptic density thickness and reduce synaptic cleft in Alzheimer' s disease rats [?]. Electroacupuncture at "Quchi" and "Yanglingquan" can regulate microglial polarization states [?]. Previous research indicated that Shaoyao Gancao

Decoction can promote interleukin-13 (IL-13) expression in rat cerebral cortex and striatum, thereby activating the JAK2/STAT6 signaling pathway, promoting microglial transition from M1 to M2 phenotype, and improving neurological function [?]. In this study, synaptic structure in the electroacupuncture, TCM herb, and electroacupuncture plus TCM herb groups all improved compared with the model group, with the electroacupuncture plus TCM herb group showing the best performance. We therefore hypothesize that in treating post-stroke limb spasticity, electroacupuncture combined with Shaoyao Gancao Decoction may exert synergistic effects by regulating inflammation and microglia from the central to peripheral nervous system, thereby influencing neuronal apoptosis and synaptic remodeling. Exploring the effects of active monomer components in Shaoyao Gancao Decoction on inflammatory responses and microglia in the ischemic penumbra will be a potential direction for future research.

In conclusion, electroacupuncture combined with Shaoyao Gancao Decoction can improve motor dysfunction in rats with post-stroke limb spasticity, possibly by enhancing synaptic remodeling.

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