



## Influence of Valeriana Officinalis Extract and Zusanli Acupoint Stimulation on Serum ALT and Creatinine Levels in Rats

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### Abstract

#### Background

Traditionally used for its calming and antioxidant qualities, Valeriana officinalis has been shown to have beneficial benefits on liver and kidney functioning. Zusanli is a popular acupoint that is frequently linked to immunological and metabolic control. The purpose of this study was to assess how rats. serum levels of creatinine and alanine aminotransferase were affected by Zusanli acupoint stimulation and Valeriana officinalis extract, both separately and in combination. The study focuses on their possible contribution to renal and hepatic function supported.

#### Methods

Four groups of male Wistar rats were randomly selected, control Zusanli stimulation, Valeriana officinalis extract, and combined treatment, Zusanli was activated by electroacupuncture while the extract was taken orally once a day, Standard enzymatic assays were used to evaluate serum ALT and creatinine levels both prior to and followed the therapy period.

#### Results

Rats given Valeriana officinalis extract had significantly lower serum ALT levels than the control group, with the combination treatment group showing the biggest drop, The Zusanli stimulation and combined groups had much lower serum creatinine levels, These results show improved biochemical indicators for the liver and kidneys.

#### Conclusion

This study showed that rats liver and kidney function may be protected by Zusanli acupoint stimulated and Valeriana officinalis extract. The greatest improvement was seen with the combined treatment, indicating a possible synergistic effect. To clarify the underlying mechanisms and therapeutic relevance.

# تأثير مستخلص نبات فاليريانا أوفيسيناليس وتحفيز نقطة زوسانلي على مستويات إنزيم ناقلة أمين الالانين (ALT) والكرياتينين في مصل دم الفئران هبة أكرم محسن، حنين سعيد محسن، عتاب عبد الأمير، حسن محمود موسى

## الخلاصة

### المقدمة

يُستخدم نبات فاليريانا أوفيسيناليس تقليدياً لخصائصه المهدئة والمضادة للأكسدة، وقد ثبتت فوائده على وظائف الكبد والكلية. تُعد نقطة زوسانلي من نقاط الوخز بالإبر الشائعة، والتي ترتبط غالباً بالتحكم المناعي والأضي. هدفت هذه الدراسة إلى تقييم تأثير تحفيز نقطة زوسانلي ومستخلص فاليريانا أوفيسيناليس، كلٌّ على حدة ومجتمعين، على مستويات الكرياتينين وإنزيم ناقلة أمين الالانين في مصل دم الفئران. تركزت الدراسة على مساهمتهما المحتملة في دعم وظائف الكلية والكبد.

### العينات وطرق العمل

تم اختيار أربع مجموعات عشوائياً من ذكور فئران ويستار: مجموعة ضابطة (تحفيز نقطة زوسانلي)، ومجموعة مستخلص فاليريانا أوفيسيناليس، ومجموعة العلاج المركب. تم تنشيط نقطة زوسانلي بالوخز الكهربائي، بينما تم تناول المستخلص عن طريق الفم مرة واحدة يومياً. استُخدمت فحوصات إنزيمية قياسية لتقييم مستويات إنزيم ناقلة أمين الالانين والكرياتينين في المصل قبل فترة العلاج وبعدها.

### النتائج

تم تنشيط نقطة زوسانلي بالوخز الكهربائي، بينما تم تناول المستخلص عن طريق الفم مرة واحدة يومياً. النتائج أظهرت الفئران التي أُعطيت مستخلص نبات فاليريانا أوفيسيناليس انخفاضاً ملحوظاً في مستويات إنزيم ناقلة أمين الالانين (ALT) في الدم مقارنةً بمجموعة التحكم، مع تسجيل أكبر انخفاض في مجموعة العلاج المركب. كما سجلت مجموعتنا التحفيز بنقطة زوسانلي والعلاج المركب انخفاضاً كبيراً في مستويات الكرياتينين في الدم. تشير هذه النتائج إلى تحسن المؤشرات الكيميائية الحيوية للكبد والكلية.

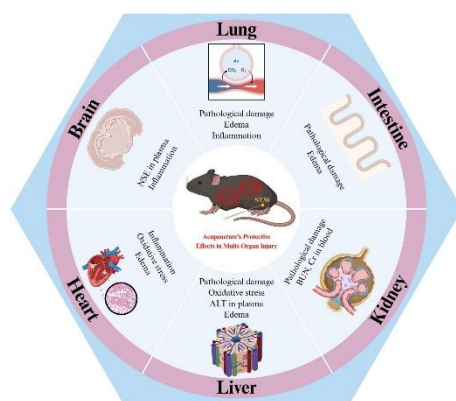
## 1. Introduction

*Valeriana officinalis*, is a perennial herb that has long been utilized for a variety of therapeutic uses in traditional medicine throughout Europe and Asia. Its relaxing and sedative qualities were appreciated in ancient Greek and Roman societies, where it was used to treat neurological illnesses and enhance overall health. *Valeriana* extracts include bioactive chemicals, including valerenic acid and other sesquiterpenes, that interact with the  $\gamma$ -aminobutyric acid GABA system and may have sedative, anxiolytic, and antioxidant effects, according to recent pharmacological study. These metabolic characteristics may protect different tissues from oxidative damage, indicating wider physiological effects beyond regulation of the central nervous system. Research on *Valeriana* species has shown that it has antioxidant activity that increases endogenous enzymes such as glutathione peroxidase GSH-Px, and superoxide dismutase SOD, suggesting a potential to reduce oxidative stress and cellular damage in experimental animals(Li et al., 2022).On renal and liver physiology. Animal research indicates that valerian may affect oxidative stress pathways and cellular proliferation in hepatic tissue, with implications for tissue protection and metabolic regulation, even though there is little clinical data on the direct effects of valerian on liver or kidney biomarkers like ALT (alanine aminotransferase) and creatinine. For instance, in rat liver models, valerian root extracts were demonstrated to inhibit oxidative DNA damage and cell proliferation, in part via activating signaling pathways linked to apoptosis and cell cycle regulation(Kakehashi et al., 2014). Because of its possible systemic effects, acupuncture at certain locations, such Zusanli (ST36), has drawn scientific attention as an adjunct to herbal therapies. Traditionally linked to the control of gastrointestinal and metabolic processes, Zusanli is a significant acupoint situated on the stomach meridian beneath the knee. According to recent studies, acupoint stimulation may have multi-system regulatory effects by modulating inflammatory responses, enhancing insulin sensitivity, influencing nitric oxide signaling, and changing neuro-immune interactions through ST36 stimulation(Dimitrov et al., 2017). The effects of ST36 stimulation on organ damage were investigated in animal models. For example, electroacupuncture at ST36 was demonstrated to reduce biochemical indicators of inflammation and tissue damage therefore attenuating acute renal injury caused by sepsis in rats. However similar protective effects on liver injury were not consistently observed in that paradigm,Acupuncture which includes ST36 has been shown to slow the progression of diet induced nonalcoholic fatty liver disease in mouse models of metabolic liver disease. This were accompanied with improvements in metabolic profiles and decreases in indices of liver dysfunction (Meng et al., 2019). When considered collectively, the data suggests that acupoint stimulation at Zusanli and *Valeriana officinalis* may both affect physiologically. The animals were divided into four groups including control Zusanli stimulationprocesses related to organ health, such as oxidative stress, inflammation, and metabolic control. These complementary techniques show promise for affecting biochemical indicators like ALT and creatinine in experimental and clinical contexts, necessitating further focused research even though the molecular pathways were still being actively investigated (Liu et al., 2025). The medicinal plant *valeriana officinalis*, sometimes known as common valerian, has been extensively studied for its pharmacological effects in rodents, including metabolic, neuroprotective, and antioxidant benefits. According to research, *V. officinalis* extracts can alter biochemical markers associated with kidney and liver function, such as creatinine and ALT, most likely via anti-inflammatory and antioxidant pathways. According to earlier research on animals, valerian supplementation affects the expression of CYP enzymes in liver tissue, which may have an indirect impact on the metabolism and function of the liver(Bogacz

et al., 2014). The systemic effects of electroacupuncture at the Zusanli acupoint were investigated in a number of mouse models. According to research, ST36 activation was found to influence systemic inflammatory responses and renal function in rats, including changes in plasma urea and creatinine levels, most likely through autonomic and anti-inflammatory processes (Harpin et al., 2020). It was reflected postulated that the synergistic control of systemic oxidative stress, inflammation, and organ perfusion is reflected in the combined action of herbal extracts and acupoint stimulation on biochemical indicators. For example, stimulation may improve circulatory and autonomic regulation while valerian may have antioxidant or metabolic regulatory effects. Together, these two interventions may have a greater impact on serum ALT and creatinine levels than either one alone (Bogacz et al., 2014).

### 1.1. Synopsis and Research Consequences

This study examines the combined effects of acupoint stimulation and a medicinal plant extract on clinically significant blood biomarkers in rats, building on previous findings. By highlighting integrative physiology and translational relevance, elucidating these processes may offer insights into complementing treatment approaches for liver and renal disorders (Bogacz et al., 2014), Fig.1.



**Figure1:** Illustrates a conceptual background of organ response mechanisms and is not a direct representation of the experimental model used in this studying. (Zeng & Yan, 2025).

## 2. Patients & Methods

### 2.1. Experimental Animals

Male rats with a mean body weight of  $209.97 \pm 24.99$  g and ages ranging from 45 to 90 days were used in this investigation. Before the experiment started, thirty-two rats weighing between 200 and 300 g were acclimated to the laboratory setting for a week. Under typical laboratory circumstances, the animals were kept in wire-mesh cages with unlimited access to tap water and normal feed. The animals were divided into four groups: Group I Controlled no treatment, Group II Valeriana officinalis extract only, Group III Valeriana officinalis extract combined with Zusanli acupuncture, and Group IV Zusanli acupuncture only. Treatments were given every day for the duration of the study.

### 2.2. Aqueous Valerian Extract Preparation

Four grams of the dried and finely crushed plant stem were weighed and put in a clean plastic container to create an aqueous extract of Valeriana officinalis. After adding 100 milliliters of distilled water to the powdered plant material,

the mixture was gently mixed and left for 15 minutes. After filtering the resultant solution using filter paper, the filtrate was dried at around 37°C to produce the aqueous extract. Table1 presents information about animal traits.

### 2.3. The Acupuncture Process

At the Zusanli acupoint, acupuncture therapy was given once a day. During every session, a sterile acupuncture needle was inserted to a depth of about 2mm and held there for two minutes. Until the end of the trial period, this process was carried out every day.

### 2.4. Analysis of Statistics

According to earlier research, Valeriana officinalis has not been substantially linked to hepatotoxicity or serious liver damage when used either on its own or in conjunction with therapeutic measures. Although the current results are in line with previous reports, care should be used when interpreting them. The results, study limitations, and their applicability to the body of current research must all be thoroughly discussed.

Distribution of weight in group study.

**Table1:** Descriptive Statistics of Body Weight (Mean  $\pm$  SD) in Male Rats Aged 45–90 Days

Weight	209.97
Standard Error	4.57
Median	210.00
Standard Deviation	24.99
Range	79.00
Count	32
Confidence Level (95.0%)	9.33

### 2.5. Acupuncture

Until the experiment's conclusion, rats received acupuncture once a day at the Zusanli point, with a needle insertion depth of 2 mm and a retention period of 2 minutes every session.

## 3.Results and Discussion

Serum creatinine (SCr), glutamic-pyruvic transaminase (SGPT/ALT), were all typically within normal ranges in the control group. Additionally, slightly lower SCr. The third group showed higher values for all parameters. Table2 reveal that whereas the other parameters were lower in the fourth group, ALT. To evaluate baseline uniformity, the body weight of male rats between the ages of 45 and 90 days was measured. With a median weight of 210 g and a mean body weight of 209.97  $\pm$  24.99 g, the laboratory rats showed typical growth tendencies(Sengupta, 2013).

The 95% confidence interval  $\pm$ 9.33 g and standard error 4.57, showed minimal animal variability. Because of this homogeneity body weight has less of an impact on biochemical results(Festing, 2006). The mice were physiologically stable and appropriate for experimental intervention as confirmed by the observed weight range 79 g and sample size n = 32 (Murase et al., 2012). Baseline biochemical parameters showed that serum creatinine 0.8 mg/dL and ALT 34 IU/L in the control group were within normal reference ranges, confirming normal kidney and liver function. The Valerian-treated group without acupuncture exhibited a slight reduction in serum creatinine 0.77 mg/ ,The elevation of ALT observed in valerian-treated groups may indicate hepatic enzyme induction or a dose-related hepatic response rather than a hepatoprotective effect (Bent et al., 2006). Serum creatinine 0.98 mg/dL and ALT 71 IU/Lwere both higher in the Valerian with acupuncture group, suggesting a possible interaction between Valerian

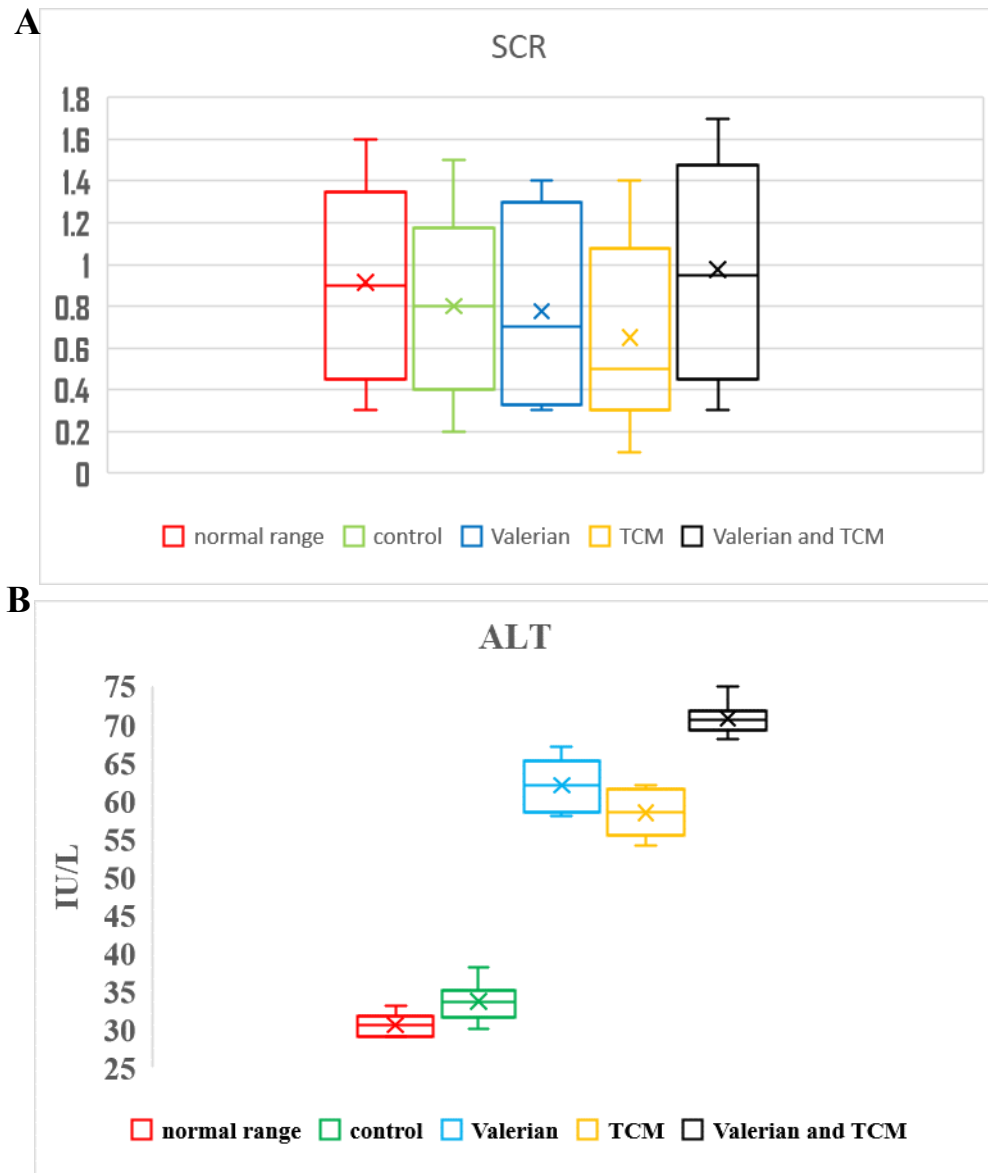
acupuncture(Vargas-Pozada & Muriel, 2020). While ALT remained elevated 58 IU/L, the acupuncture-only group showed the lowest serum creatinine level 0.65 mg/dL, indicating a renal benefit of acupuncture with little hepatic protection(Cho & Kim, 2021). In experimental animal investigations, body weight consistency was considered crucial since it indicated overall health and metabolic stability. The validity of the biochemical results was reinforced by the homogeneity found in this investigation. One trustworthy measure of renal function was represented serum creatinine. Improved renal perfusion and decreased oxidative stress brought on by activation may be the cause of the lower creatinine levels observed in the acupuncture only group were attributed (Zhang et al., 2018). Hepatocellular stress was indicating resulting shown by elevated ALT values in Valerian treated groups. According to earlier research, Valerian was show to alter the activity of liver enzymes, especially when it was administered frequently or in large doses (Philips et al., 2020). Although acupuncture was reported to exert hepatoprotective effects its combination with Valerian did not normalize ALT levels in this stud suggesting a complex interaction between herbal metabolism and acupuncture effects (Chen et al., 2025)., Table3 and Fig.2. The study confirmed that the experimental animals were physiologically homogeneous. ST36 acupuncture alone demonstrated a potential protective effect on kidney function as evidenced by reduced serum creatinine levels (Martins Filho et al., 2024). Valerian administration was associated with elevated ALT levels indicating possible hepatic stress. The combination of Valerian and acupuncture did not reduce this effect. Further experimental studies incorporating histopathological evaluation and dose optimization are recommended to better understand the safety and interaction of Valerian and acupuncture(Ibrahim et al., 2025).

**Table2:** Baseline Values of the Investigated Biomarkers

Names of Tests	Normal range	Unit
Serum Creatinine (SCr)	0.7 - 1.3	mg/dL
Glutamic - pyruvic transaminase (SGPT/ALT)	29 – 33	IU/l

**Table3:** Effect of Valeriania and Liver Function and Kidney Function, numbers represent mean values

Groups	SCR	GPT (ALT)
Control	0.8	34
Valerian Without acupuncture	0.77	62
Valerian with acupuncture	0.98	71
Only acupuncture	0.65	58



**Figure2: Box-and-whisker plots illustrating serum creatinine (SCR) and alanine aminotransferase (ALT) levels across the study groups. The groups include normal range, control, Valerian, TCM, and combined Valerian and TCM treatment. The boxes represent the interquartile range (IQR), the horizontal line inside each box indicates the median, and the whiskers denote the minimum and maximum values. The “x” symbol represents the mean value for each group.**

### 3. Conclusion

The present study demonstrated that the experimental animals were physiologically homogeneous, as confirmed by consistent body weight and baseline biochemical parameters, ensuring the reliability of the observed renal and hepatic outcomes. Serum creatinine and ALT levels in the control group remained within normal reference ranges, indicating normal kidney and liver function at baseline. Acupuncture treatment alone, particularly at the ST36 point, was associated with a reduction in serum creatinine levels, suggesting a potential renoprotective effect, likely mediated through improved renal perfusion and reduced oxidative stress. In contrast, Valerian administration was associated with elevated ALT levels, indicating possible hepatic enzyme induction or dose-related hepatocellular stress rather than hepatoprotection. The combined administration of Valerian and acupuncture did not normalize ALT levels and was accompanied by increased serum creatinine, suggesting a complex interaction that may limit the protective effects of acupuncture when used concurrently with Valerian. Overall, these findings highlight the potential renal benefits of acupuncture alone and raise concerns regarding Valerian-induced hepatic effects, particularly when combined with acupuncture. Further studies incorporating histopathological assessment and dose optimization are warranted to clarify the safety profile and mechanistic interactions of Valerian and acupuncture.

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