

ACUPOINT RENZHONG (JENCHUNG GV-26) IN THE HORSE. ANATOMICAL AND HISTOLOGICAL STUDY

Acupunto Renzhong en el caballo. Estudio anatómico e histológico.

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ABSTRACT

The purpose of this study was to document the anatomic and histologic characteristics of acupoint Renzhong GV-26 in ten Spanish horses. First, the electrical skin resistance was measured to locate the acupoint, and the needles were inserted to a depth of 1cm into the acupoint. Then, the tissue surrounding the acupoint was removed, sectioned serially, and processed by routine histologic techniques. The results show that the acupoint Renzhong GV-26 is situated in the intermediate area of the upper lip, represented by several structures of dense connective tissue, skeletal muscle tissue, nerve fibers and vascular structures. Therefore, numerous structures conform part of the acupoint Renzhong GV-26 in the horse.

Keywords: Acupuncture, Acupoint Renzhong GV-26, Horse, Histology, Anatomy.

RESUMEN

El propósito de este estudio es documentar las características histológicas y anatómicas del acupunto Renzhong GV-26 en diez caballos de raza española. Primero, se midió la resistencia eléctrica para la localización del acupunto, y posteriormente se insertó una aguja de acupuntura a una profundidad de 1 cm dentro del acupunto. Luego se obtuvieron muestras del acupunto y fueron procesadas por técnicas histológicas de rutina. Los resultados muestran que el acupunto Renzhong GV-26 está situado en el estrato intermedio del labio superior, representado por diferentes estructuras, como tejido conectivo denso, músculo estriado esquelético, fibras nerviosas y estructuras vasculares. Es decir, numerosas estructuras conforman el acupunto Renzhong GV-26 en el caballo.

Palabras clave: Acupuntura, Acupunto Renzhong GV-26, Caballo, Histología, Anatomía

INTRODUCTION

Acupuncture is a therapeutic system within traditional Chinese medicine characterized by inserting needles of different metals into specific parts of the body, called acupoints, in order to cure, relieve or eliminate pain. It is used in both human and veterinary medicine. In animals, these acupoints form currents of energy known as acupuncture meridians.

These meridians constitute the basis of acupuncture and their description originated due to the knowledge of the topography of the acupoints. They are situated symmetrically with respect to the sagittal axis of the body, and are found throughout the head, the trunk, the abdomen and the limbs, both in the anterior as well as in posterior part of the body, forming a closed system of circulation. Each one has its own distribution, a specific energy timetable, and corresponds to an organ (Becker and Rechmains, 1976).

The Renzhong acupoint (Jenchung, GV-26 or 26 Du in Du Mai Chinese) belongs to other governing meridian vessels (GV). In all species this meridian runs in the following directions: it begins in the coccyx and ascends the middle dorsal line to the neck. It then runs along the middle line in the head towards the forehead and the nose and finishes below the upper lip.

The Renzhong acupoint is found at the border of the middle and upper third of the distance between the nose and the upper lip in humans (Pomeranz and Stux, 2000) and in animals such as horses at the upper lip, at the midpoint of the *philtrum nasale* (Demontoy and Mailhac, 1980; Westermayer, 1985).

The Renzhong acupoint (Jenchung, GV-26 or 26 Du) is one of the most widely used in the emergency ward, in both animals and humans. A bibliography of more than 160 articles (1973-2001) documents the effects and clinical efficacy of the stimulation of this acupoint and leads one to speculate other physiological and clinical

applications. Very few studies exist on the microscopic structure of the acupoints in domestic animals. The main works refer to cattle and dogs (Kothbauer, 1961, 1986; Egerbacher, 1991, 1994, 1996; Ludewig, 1998). The structures found at the acupoints are not unique. Many of them are found in depressions situated in muscular zones (Helmes, 1995). These are areas of the skin containing a high concentration of nerve endings, nerve plexuses, mast cells, lymphocytes, capillaries and venules (Kendall, 1989). It has been suggested that the size of the acupoints is 1 mm². Histologically, they have their own structure characterized by a thinning of the skin due to a modification of its collagen's fibers of the dermis, this explains why they can be felt as a depression (She); they also present a spiral vascular network, surrounded by a dense network of collinergic-type amielinic nerve fibers (Niboyet, 1980). Heine (1988) revealed that 80% of acupoints correlated with perforations in the superficial fascia of human corpses. A nerve vessel bundle and vessels penetrated the skin through these orifices.

Studies carried out in cows (Kothbauer, 1961, 1986; Ludewig, 1998) and in dogs and cows (Egerbacher, 1994) have verified the correlation between acupoints and perforations in the superficial fascia by sensitive cutaneous nerves. Macroscopic examination of 27 points of the bladder meridian in a cow revealed very defined perforations in the thoracolumbar fascia through which nerve-vessel structures ran at 19 points. The remaining eight acupoints (B18-25) were situated at the entrance points of the middle cutaneous branch in the skin of the back of the animal. There were no perforations of the fascia, which was a new discovery. Fascia perforations with nerve-vessel structures were found at the five points of the gall bladder meridian, which were examined.

Histologic examination revealed a special disposition of the connective tissue, surrounding the nerve, with concentric laminas and loss of connective tissue composed of fine

fibers of collagen. A more compact sheath of connective tissue surrounded the nerve-vessel structure at the point where it perforates the fascia (Egerbacher, 1994). These results are valid for other points with reduced electrical resistance at the base of the teat (Ludewig, 1998). A high concentration of mast cells was also found in the area adjacent to the point (Zhai, 1998).

In reference to the vascular structures in the areas of the acupuncture, several authors stress the importance that they play in the regulation of local temperature. Talukdar *et al.* (1972) in a study performed on the skin of the horse describes special vascular structures in 43 areas of the skin. They consist of arterio-venous anastomosis in rings or spirals forming canals between arterio-venous, arterioles with epitheloid cells on the walls and glomus. These glomus might be related to the conservation of the heat and temperature regulation. Other authors focus the subject on the components of the connective tissue, lymphatic vessels, arterioles and veins at the acupoints, describing vascular structures surrounding collinergic amielinic nerve fibers.

Our study corresponds to the use of this acupoint in anaesthetic recovery in the horse. Considering that its anatomic and histologic characteristics have not been described, the purpose of this study is to report the morphologic characteristics of acupoint GV-26 in Spanish-bred horses.

MATERIALS AND METHOD

Ten Spanish-bred horses were used. Before sacrificing the horses, the localization of the Renzhong GV-26 acupoint was determined in the upper lip by means of an ohmiometre (WQ-IOC2R multiple Electronic Acupunctoscope. Made in China) and Chinese cartography. Acupunctoscope or «point finder» consist of a pencil-shaped metal sound connected by means of a cable to an ohmiometre. A second electrode

situated in a metallic cylinder completes the circuit. Some studies suggest that the resistance of the skin (impedance) at the acupoints is less than in the surrounding skin (Becker and Rechmains, 1976; Oleson *et al.*, 1980; Pomeranz and Stux, 2000).

Once the acupoint had been determined, the definite marking of it was performed by means of a disposable, surgical, stainless steel, philiform point and a depth of 3-cm (Hwato. Suzhou, China). The humane sacrifice of the animals was then performed using a 20-mg/Kg overdose of endovenous pentobarbital (Eutalender Lab. Normon S.A. Madrid, Spain). After the death of the animal, a sample of the pyramidal shaped upper lip was taken, corresponding to the acupuncture needle was situated, and two samples from adjacent areas, according to Egerbacher's protocol. By means of a scalpel and dissecting tweezers the boundary and depth of the sample was ascertained, approximately 1 cm wide by 3 cm long. The samples thus obtained were fixed in 10 % formaldehyde at room temperature and remained in the fixing liquid for a minimum of 10 days. Once fixed, they were treated in a tissue processor (TP 1050 LEICA, Germany), using usual procedures for their embedding on paraffin blocks. This was performed by means of a Tissue-Tek (Germany), which was fitted with disposable knives (Reichert, model 819, Germany). Sagittal cuts of a thickness of 10 mm were then placed on the surface of a double saucepan of water at 37°C to encourage stretching.

Next, they were placed on microscope slides (50 Microscope Slides, Menzel. Glaser, Germany) covered with Mayer's glycerinated albumin, remaining in the oven at 55°C for one hour until they were ready to be dyed histologically. The histologic techniques used were the hematoxylin-eosin (H-E) and Gallego's Trichromic methods. The dyed preparations were then viewed and photographed in a Leitz Dialux 20 R photomicroscope fitted with photographic equipment (Germany).

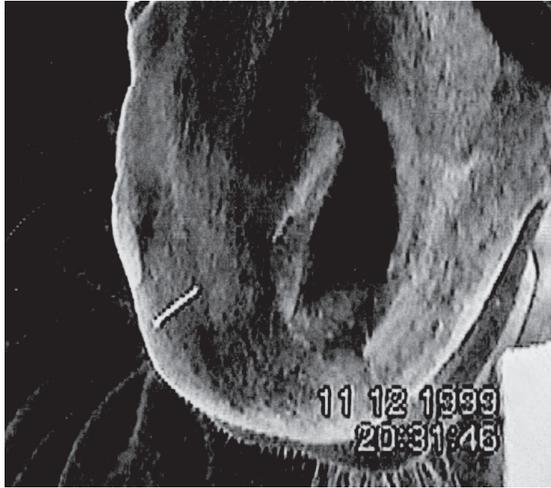


Figure 1. Renzhong GV-26 acupoint in horse. Needle at the midpoint of the philtrum nasale.



Figure 2. Renzhong GV-26 acupoint in horse. Anatomic section of the philtrum nasale.

RESULTS

In the animals under examination the area corresponding to Renzhong GV-26 acupoint is a prism whose base is parallel to the surface and is located in the middle area of the skin of the upper lip (figs 1, 2). The histological sections reveal predominant striated muscle bundles, which correspond to the fascicles of muscle *orbicularis oris*, *incisivus superior*, *levator nasolabialis*, *caninus* and *buccinator*. The study of the histologic sections corroborates that the muscle fibers are surrounded by the corresponding *endomysium* and grouped in fascicle of circular aspect, delimited by wide bands of *perimysium*.

A thick bundle of dense connective tissue runs through all the sections parallel to the surface of the skin and between the bundles of muscle fibers. This comes from the insertion of the

common sinew that both *m. levator labii superioris* offer in this species (fig. 3). In the depths of the prisms of the sections included in the connective tissue one can find an important number of vascular structures, which correspond to arterioles, venules and capillaries forming a small network of arteriovenous anastomosis. These vessels come from branches of the artery and upper labial vein. Sections of nervous fibers are visible amongst them and covered by a network of connective tissue with abundant cells (fig. 4, 5). In the adjacent areas, which were examined, a series of muscle bundles, connective tissue, adipose tissue and adenomers of the sublingual salivary glands were found. Also visible were a number of vascular and nervous structures. These findings were seen in the sections in an all between individual animals were observed.

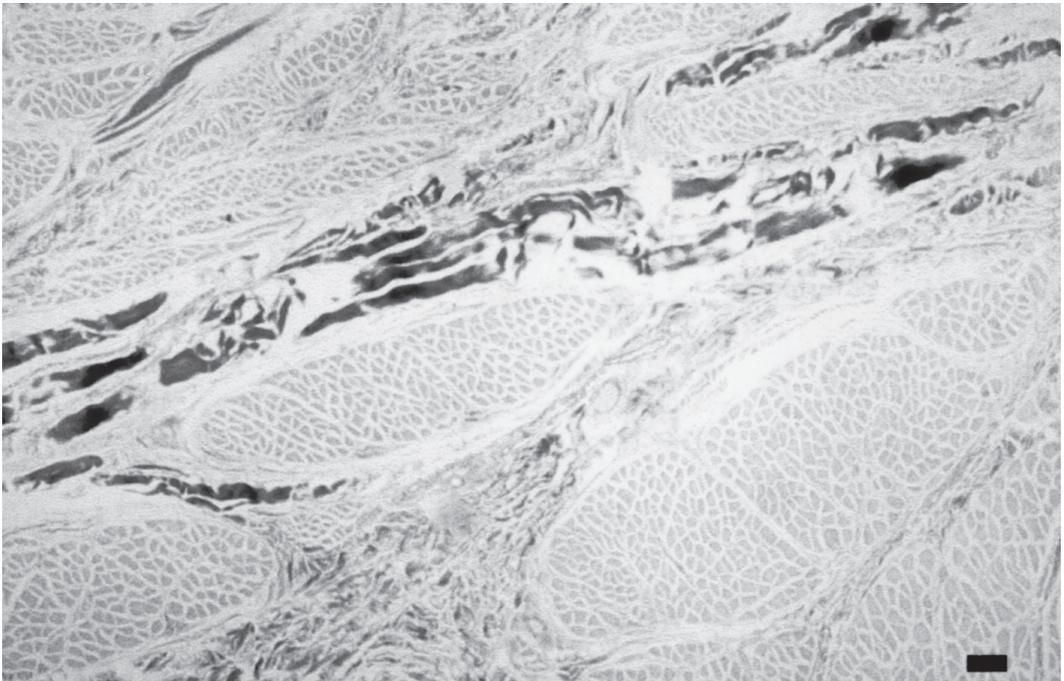


Figure 3. Skin. Acupoint Renzhong. Intermediate stratum. Muscle fibers and dense connective tissue. Trichromic staining of Gallego. Bar = 100 mm.

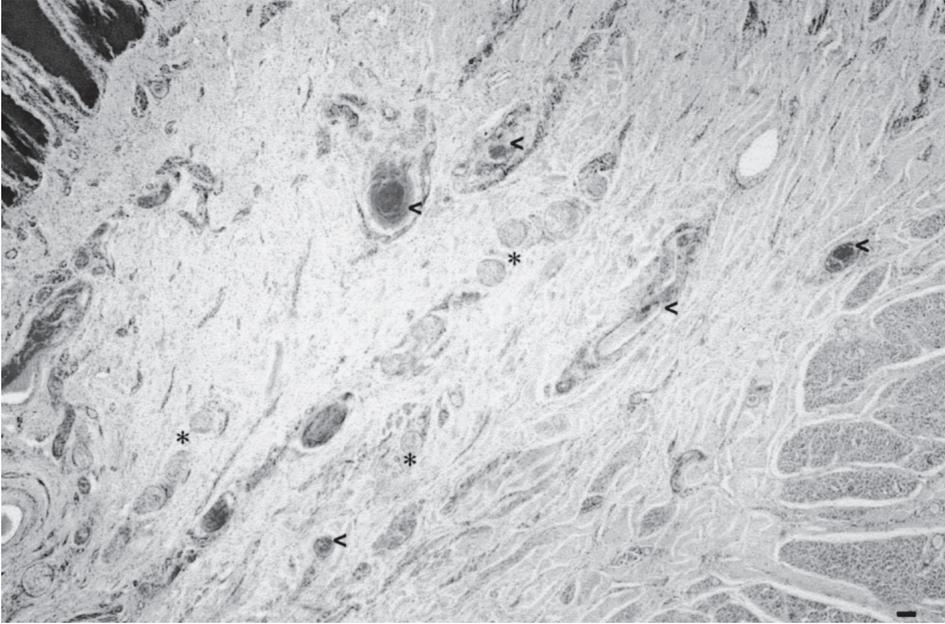


Figure 4. Skin. Acupoint Renzhong. Intermediate stratum. Court in different planes from nerves (\blacktriangleleft) and vessels structures ($*$). Hematoxilin-eosin. Bar = 100 mm.

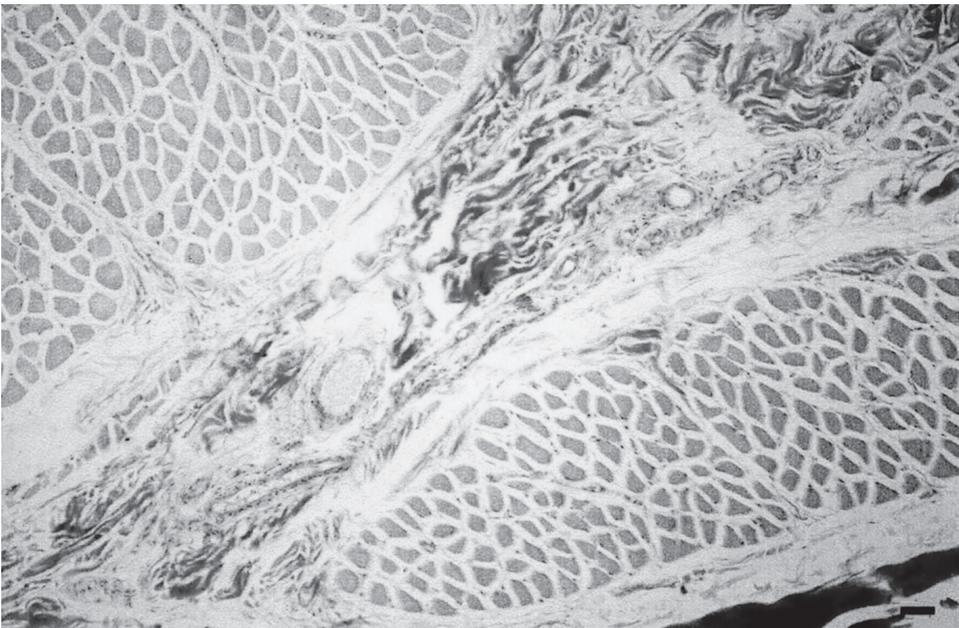


Figure 5. Skin. Acupoint Renzhong. Intermediate stratum. Court in different planes from vessels structures (\blacktriangleleft). Trichromic staining of Gallego. Bar = 50 mm.

DISCUSSION

This is the first description of the anatomical and histological structure of the Renzhong acupoint in the horse. From an anatomic point of view, it has been possible to examine dissections performed which reveal that once the acupuncture needle has gone through the skin layers (epidermis, dermis), it ends up in the area of the upper lip of the horse known as the «intermediate stratum» (Dyce *et al.* 1999). According to the same authors this is a part of the lip situated between the surface of the skin and the deep mucous, mainly made up of muscles, tendons, blood vessels and nerves.

The histologic sections performed, showed that the main components of this structure are skeletal striated muscle fibers, belonging to the group of muscles that pass into the upper lip of the horse (*m. orbicularis oris, incisivus superior, levator nasolabialis, caninus* and *buccinator*).

The characteristics of these findings corroborate the findings by Gunn *et al.* (1976) and Dung (1984) that point out the presence of muscle structures, together with gross bundles of connective tissue surrounding the acupoints. Blood capillaries and nerve endings (sensitive and motor) join all these structures to nourish and stimulate them. These sections are endings of the *trigeminus nervus* corresponding to bundles of the *infraorbitalis nervus* as described in the classical treaties of Veterinary Anatomy and also more recent ones (Climent *et al.* 1998).

Theories described by Lu and Liu (1988), Zheng *et al.* (1989), Chen *et al.* (1990) and Zhang *et al.* (1990) based on research in rabbits and referring to the path of the stimulus of the Renzhong GV-26 acupoint in the respiratory system, show that these nerve endings would be responsible for a fast afferent path to the brain.

In this study, it has not therefore been observed that the histological area corresponding to the Renzhong GV-26 acupoint in the horse is integrated by a single structure. These findings agree with other published studies by Kothbauer

(1961, 1986); Gunn (1976); Dung (1984); Heine (1987, 1988, 1996); Ludewig (1998); Egerbacher (1991, 1994, 1996) on other species, emphasizing the importance given to the special disposition of the connective tissue.

We think that we can rule out that special vascular structures may exist, like those referred to as glomus described on the skin by Talukdar *et al.* (1972). These are artery-venous structures in either rings or spirals and special intrarterial cushions in the structure to the dermis of the upper lip that may be related to thermoregulation.

The Renzhong GV-26 acupoint is located in the intermediate area of the superior lip of the horse. It is a complex area formed by diverse structures like muscular fibers, connective tissue, vascular structures and nerve fibers.

It is not known which structures are responsible for the communication channels. It is possible that the action at the mechanism may be due to a complex chain of tissue actions and not only to unique structure. The study of this type of structures and also more specific ones on nerves endings, inflammatory cells present in the acupoints and the special disposition of the connective tissue should be raised in future works.

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