The American Journal of Chinese Medicine, Vol. 36, No. 1, 25–36

Institute for Advanced Research in Asian Science and Medicine

Acupuncture and Immunomodulation

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Abstract: Acupuncture is a well-known form of Asian medical treatment and it is used not only as an effective curative method but also to prevent illness and mai ntain health. It is used for the production of analgesic effect; stress related physical-mental disorders and homeostasis. Electroacupuncture (EA) stimulation, an application of electrical current on acupuncture needles, is one of the most popular types of this traditional therapy. In recent years, intensive studies have been carried out to explain the underlying mechanisms of the efficacy of acupuncture. An increase in the release of endogen opioid peptides is generally accepted to be a keystone pathway that affects the immune system after the acupuncture application. To understand the huge gap between specific skin point applications and immune responses, a vast number of accumulating data of experimental and clinical studies in the literature have been collected. This paper reviews the data to explain the updated mechanisms related to immune modulation via acupuncture therapy.

Keywords: Acupuncture; Immunomodulation.

Introduction

According to the 2002 National Health Interview Survey, acupuncture is performed to 2.1 million adults per year in the USA (Barnes *et al*., 2004). In the treatment of dysmenorrheal, osteoarthritis, fibromyositis, trigeminal neuralgia, post-operative ache, acupuncture has been used and different degrees of recovery have been observed (Creamer *et al*., 1999). Acupuncture may be used successfully in the treatment of anxiety and depression as the level of serotonin and encephalin increases in the central nervous system and plasma (Ullet *et al*., 1998). It has been reported that acupuncture application in obesity treatment is effective for weight loss (Ernst, 1997; Cabioglu and Ergene, 2005). Acupuncture has been applied in the rehabilitation of cases of hemiplegia (Wong *et al*., 1999).

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Clinical and experimental studies showed that EA applied to specific anatomical loci is successful in the treatment of stress induced physical disorders and immunodeficiency (Vasilenko *et al*., 1989). It increases the resistance to infection, alleviates the autoimmunity and hypersensitivity in clinics (Cheng, 1997). In various immune related disorders, such as cutaneous T cell lymphoma, delayed-type hypersensitivity reaction, rheumatoid arthritis, Hashimoto's thyroiditis, ulcerative colitis and some bacterial infection, acupuncture is effectively used (Hashiro *et al*., 1994; Tohya *et al*., 1989; Tang *et al*., 1992; Hu *et al*., 1993; Wu *et al*., 1997; Yamashita *et al*., 1998). Combined EA stimulation and moxibustion activates defense systems (Vasilenko *et al.*, 1989), enhances cellular immune function of patients with malignant tumors (Rogers *et al.*, 1992).

In general, immunomodulatory effects of acupuncture might be expressed under three categories: local, neuronal and neurohumoral immunomodulation. Since some of the mediators in analgesia are associated with the immune actions of acupuncture, analgesic mechanisms are reviewed simply with immunomodulation.

Local Immunomodulation

Histologic-Electrical Properties and Physiologic Effects of Acupuncture Points

There are increased structures of capillaries, symphatic nerve endings, dermal papillas and electrolyte embedded sites with condensed gap junctions in epidermis at the acupuncture points, so that they possess different electrical potentials than nearby sites. Some studies define acupuncture points in types according to their structures. Some acupuncture points are related to motor points, which produce maximum contraction in muscles. Some are the junctions for bilateral superficial nerves. Some are the points of high-density plexus and principle superficial nerves. Others are localized at the muscle-tendon junctions (Croley and Carlson, 1991; Looney, 2000). Histologic difference by receptor or effector types at the acupuncture points may convey different outcomes and may be useful as in various indications (Kothbauer and VanEngelburg, 1994; Rogers, 1992). There are electrical potential differences between acupuncture points and ordinary skin points (Shang, 1989). It is found that acupuncture points have low electrical resistance and various types of point detectors have been developed according to this property (Leonhardt, 1980). Also, enhanced nitric oxide (NO) concentrations and expression of nitric oxide synthesis (nNOS) in acupuncture points cause low electrical resistance (Ma, 2003). In addition, during needling, temperature difference and conductivity of the needle produces a low intensity galvanic current as a micro energy source to excite free nerve endings. Even after taking needle out, an electric potential field continues the stimuli for 72 hours due to high concentration of K^+ ions at the injury site and its chain reactions (Becher and Selden, 1985).

The skin, muscle tissue, nerves and sometimes ligaments or bones are affected by every kind of stimulation during the application. It has been determined that 70–80% of all acupuncture points are the same as the trigger points (Melzack *et al*., 1977) and that most of the acupuncture points are also the same as muscular motor points (Liao,

1975). The trigger points (painful points in skeletal muscle) and muscular motor points are inactivated during needling by spinal and supraspinal reflexes. Acupuncture points are also dense loci for nocireceptors (pain), golgi-tendon receptors, Meissner corpuscles, Krause's end-bulbs, glomus-bodies, smooth muscle and neuro-muscle junctional receptors (Kho and Robertson, 1997). When a needle is inserted into the Shangyang (LI 1), Zhongchong (P 9), and Shaoshang (L 11) points, it stimulates the receptors of touch and pressure, which are two sense receptors. When the needle is inserted into the Neiguan (P 6) and Yuji (L 10) points, it stimulates the muscle fibers; these points are located deep in the tissue, which has a lot of muscle fibers. Accordingly, when a needle is inserted into the Daling (P 7) point, it stimulates the receptor of the golgi tendon and/or pressure receptors (Wang and Liu, 1988). Besides, specific points have specific outcome according to the locus, such as HeGu point (LI 4) modifies intestinal movements and ZhiGou point (SJ 6) takes part in the treatment of ear diseases. Moreover, randomly selected sham acupuncture points have an analgesic effect on 28–35% of the patients whereas real acupuncture points have analgesia on 55–85% of the patients (Karavis, 1997). Electroacupuncture with a frequency of 2 Hz causes analgesia by methionine- encephalin, with 100 Hz mainly by A-dynorphine and with 15 Hz, quantities of both are equal. 60% of the patients with chronic myofascial pain of the lumbar portion of the spinal cord treated with warm compress (43–51°C) or ultrasound respond to analgesia for 90 min up to 7 days. However, for EA application to general acupuncture points, pain relief for 58% of the patients is for weeks up to 3 years. The long-term pain killing effect of acupuncture may be due to serotonin and met encephalin related mesolimbic descending pain inhibition system and spinal continuous inhibition of pain stimuli (Han's theory) (Baldry, 1993).

Inflammation and the Repair Command

Micro trauma occurring during needling initiates secretions of histamine, bradikinin, substance P, serotonin and proteases, which lead a local inflammation. Hageman factor (Factor-XII) secretion with coagulation system, plasminogen, kinins and complement system activation, prostoglandins take part in the case. At the stimulation point, mast cell of Lewis layer and cells around the needle secrete bradikinin, histamine-like substances, heparin, adrenocorticotropic hormone (ACTH), serotonin, and protease. Their secretions cause vasodilatation, increased local permeability and local reaction. Due to high concentration of nerve endings and capillaries, these local effects are exaggerated (Looney, 2000). After vasodilatation, local edema, migration of leucocytes and mast cells secretion of cytokines (tumor necrosis factor-α (TNF-α), interleukin-6 (IL-6), interleukin-1 (IL-1)) stimulate hypothalamus to secrete CRH (corticotrophin releasing hormone). The repair command happens by means of hypothalamus-pituitary-adrenal axis: CRH secretion from hypothalamus causes ACTH release from pituitary gland for adrenal gland secretion of glukocorticoids to regulate inflammation and healing. By CRH, lymphocytes secrete corticosteroids and anti-inflammation cytokines, which are IL-2, IL-4, IL-10, TGF-beta (Cho *et al*., 2006). Activated macrophages, local endothelial cells, fibroblasts and lymphocytes secrete colony-stimulating factors (CSF), such as GM-CSF, G-CSF, M-CSF, TNF and IL-1 (Guyton, 2001).

Neuronal Immunomodulation

Acupuncture Points Through Central Nervous System (*CNS*)

There are some theories to explain the perception of pain and transfer of stimuli through CNS. Simply, as the receptors (nerve endings) at the acupuncture points are evoked, the afferent nerves of either myelinated (Aδ) fibers of mechanoreceptors or non-myelinated (C) fibers of nociceptive receptors transfer the stimuli to the dorsolateral column of the medulla spinalis (dorsal reflex, primary afferent depolarization) with its pain-inhibiting complex. Activation of nociceptive afferent fibers (pain induction started by potassium bradykinin, acetylcholine, prostoglandins) and synthesis of substance-P by small cells of spinal ganglia and the gasserian ganglion lead the pain stimuli (Bonica, 1990). It is believed that secreted encephalin causes the presnaptic and postsynaptic inhibition at the places, where the C and Aδ type nerve fibers synapse in the dorsal horn (lamina-V). By neospinothalamic and paleospinothalamic tracts to thalamus and thalamocortical tracts to cortex, the stimuli are perceived. A pain controlling system is activated to inhibit the pain signals which come into the nervous system. This controlling system is called the analgesia system. When the acupuncture needle is inserted, it stimulates the pain receptors (nerve endings) and causes the secretion of endogen opioids. These play a role in pain control. When the pain controlling system is activated, the neurons which originate from mesensephalon, periacuductal gray substance in the periventricular region send their stimuli to the nuclei of rafe magnus and nucleus reticular paragigantocellularis (RPGC). Then, these stimuli go to the dorsolateral column of the medulla spinalis with its pain inhibiting complex. In the analgesia system, there are neurotransmitters like endorphin, encephalin and serotonin. Encephalin is secreted by most of the nerve fibers originating in periaqueductal gray substance and the nucleus of the periventricularis and terminate in the rafe magnus nucleus. Encephalin shows high affinity to opioid receptors delta and mü1 (Chen *et al*., 1996). Encephalin which is secreted by pain stimulation is connected to the mü1 receptors and creates supra-spinal analgesia. It is also connected to the delta receptors and creates spinal analgesia. The stimulus of pain causes secretion of serotonin from nerve fibers which originate in the rafe magnus nuclei and terminate in the dorsal horn of the medulla spinalis. It also causes the secretion of encephalin from local neurons of the medulla spinalis. It is believed that secreted encephalin cause the presnaptic and postsynaptic inhibition at the places where the C and Aδ type nerve fibers synapse in the dorsal horn (Guyton and Hall, 2001).

For the segmental control, the cutaneo-visceral, cutaneo-muscular and intersegmental (vegetative, stretch, polysynaptic) reflexes are important mechanisms related to 1-3 dermatomes for efficacy of acupuncture. In general, the cutaneo-visceral reflex is the irritation of a skin point which influences functionally the organs connected to the neurotomes. The somatosensory inputs from the skin or muscle are involved in the control of various autonomic functions (Jansson, 1969; Kehl, 1975; Koizumi *et al*., 1980). Acupuncture application on the back-shu points awakens the cutaneous-visceral reflexes and this causes a regulated effect on related organs. Back-shu points are used

in the treatment of visceral diseases such as dysfunction of gastric motility (Yu-Qing *et al*., 2006). On the other hand, acupuncture affects autonomic nervous system activity via the viscero-somatic reflexes. The skin and the related viscera have the same segmental innervations usually by dorsal roots, spinal nerves and nuclei. The nociceptive impulses from the affected viscera pass to the dorsal horn and then to anterior horn of spinal cord across interneurons. Visceral afferent nociceptors converge on the same pain projection neurons as the afferents from the skin and make a substantial mixing of information from these two sources of input. This cross-talk gives rise to the phenomenon of referred pain, where visceral nociceptor activation is perceived as a cutaneous sensation (Meyer *et al*., 1985; Bear *et al*., 2007). When there is a dysfunction on the visceral organs, the viscero-cutaneous reflexes are awakened and create a pain and sense on the back-shu points related to these organs**.** That distress situation creates a pain and irritation on the dermatomes of the related back-shu point (Youbang *et al*., 1989). For the supraspinal control, which is mentioned above, acupuncture affects cortical-sub cortical mechanisms (opioid, non-opioid, and sympathetic) in the treatment of psychiatric diseases, addictions and many pain syndromes (Omura, 1975; Looney, 2000). Concerning analgesic effect, EA application is more effective than traditional acupuncture (Wang *et al*., 1992).

CNS and Neuroendocrine System (NES) Effects

Acupuncture application effects cause changes in the concentrations of K^+, Na^+, Ca^{++} in the neurons (Deng, 1995), and EA application causes a great change in the action potential of nerve cells (Fu, 2000). Mediation of endogenous opioid peptides like beta endorphin (BE) and encephalin are widely believed to be the major mechanism for the action of acupuncture (Fu, 2000; Jin *et al.*, 1996). It has been determined that endomorphine-1, beta-endorphin, encephalin, and serotonin levels increase in plasma and brain tissue through acupuncture application (Jin *et al.*, 1996; Cabioglu and Ergene, 2005). Serotonin receptors [5-HT(1A)] activation and blockade have different effects on the immune response (Idova *et al.*, 2006). Serotonin associates the immune system through the expression of its receptor subtypes in the immune cells. In the patients of depression, the nervous and immune systems interact and might be related to the change in the expression or function of the serotonin transporters in lymphocytes (Lima and Urbina, 2002). EA application causes the secretion of endorphin, which plays a role in producing the analgesic effect of hypophysis and secretion of β-endorphin and ACTH from the anterior lob of the hypophysis (Takeshige *et al*., 1992; Pan *et al*., 1996). The endogenous opioids are connected to the opioid receptors, which are located in the central nervous system, and the surface membrane of nociceptors. They produce an analgesic effect, which is inhibited by naloxan (Pomeranz *et al*., 1977) and hypophysectomy (Takeshige *et al*., 1992). The action of acupuncture on immunomodulation, though it is still under investigation, is mainly related to the reticular formation of the brain and the associated mediators (CRH, ACTH, cholecystokinin, bombesin, encephalin, dynorphin and etc.) to be involved in feedback controls. The reticular formation controls consciousness, cardiopulmonary

rhythm, muscular-vessel tonus and are the junctions of sub cortical, autonomic and cortical functions. By the peripheral information or pain stimuli (EA, acupuncture, acupressure etc.), the reticular formation regulates consciousness at cortical level and alter peripheral organ functions via descending modulation systems.

Neurohumoral Immunomodulation

Modulation of Immune Molecules

Electroacupuncture or acupuncture stimulates the hypothamus to secrete β-endorphin and ACTH in equal amount. Endorphins pass through the blood-brain-barrier in the pituitary gland to act on peripheric cells and tissues. They also pass back to CSF (cerebrospinal fluid) for the opioid receptors in various centers in the CNS (Strauss, 1987). In the cells of the immune system, the existence of receptors of endogenous opioid peptides was established (Han *et al*., 1999). It has been determined that endogenous opioids increased in plasma and brain tissue through acupuncture application affects the levels of serum immunoglobulin (Jin *et al.*, 1996). EA to endocrine points on both ears in addition to body points Hegu (LI 4) and Xiaguan (St 7), saliva IgA levels significantly increased in the subjects who had previously lower levels of IgA, but decreased in those who previously had higher levels (Yang *et al*., 1989). Electroacupuncture to the Zusanli (St 36) on rats, the levels of interleucin-2, interferon γ and the activity of natural killer cells of the spleen and levels of beta endorphin and interferon γ in serum were increased with this application. When 10 mg/kg of naloxan was injected before electro acupuncture, it was noted that the increases in natural killer cell activity and interferon γ were less. It was concluded that electroacupuncture applications increased the spleen beta-endorphin secretions. As a result of this, natural killer cell activity and the levels of interferon γ were increased (Yu *et al*., 1997; 1998).

Kho and his colleagues (1990) investigated the patterns of adrenaline, noradrenalin, adrenocorticotropic hormone (ACTH), ß-endorphin, hydrocortisone, immunoglobulins (IgA, IgG and IgM) and total and differential leukocyte counts in the peripheral blood, during and 6 days after thyroid surgery. This study was performed in 20 patients under acupuncture and anesthesia by supplementing small doses of pethidine. In all cases, 4 ear points Shenmen, Jiaogan, Jing, Neifenbi, ipsilaterally to the thyroid nodules were selected for acupuncture. The needles were connected to a battery-powered stimulator and stimulation was applied (4 Hz, 20 mA and 20 volts). During surgery, an increase in the level of adrenaline, noradrenalin, ACTH, BE, hydrocortisone and a decrease in immunoglobulin were observed, whereas in leukocytes, a decreased percentage of eosinophils and a remarkably reduced percentage of neutrophils were demonstrated. In the postoperative phase, levels of noradrenalin and ß-endorphin remained elevated, whereas adrenaline, ACTH, hydrocortisone gradually returned to normal values. Immunoglobulin levels and eosinophils counts returned to the preliminary values within 24 hours and neutrophil and lymphocyte counts within 2 days. In the thyroid surgery under only general anesthesia, levels of immunoglobulin had not returned to normal values 3 days after the surgery

(Cohnen, 1972) whereas in this study, immunoglobulin levels returned to the preliminary values within 24 hours. The decrease in immunoglobulin during the surgery applied by acupuncture anesthesia returned to normal values in shorter time and BE values remained elevated.

Modulation of Immune Cells

The effect of acupuncture on the immune system is related to the effects of β-endorphin, methionine encephalin and leucine encephalin on this system. It has been noted that leukocyte has proopiomelanocortin mRNA. Because of this, leukocytes can synthesize ACTH and β-endorphin from promolecules. Besides, endogen opioid receptors have been found on B-lymphocytes, T-lymphocytes, natural killer cells, granulocytes, monocytes, platelets and complement terminal complex. There are also chemical and physical similarities between the opioid receptors in the neuroendocrin system and the opioid receptors in the immune system (Khansori *et al*., 1990).

It has been determined that α, β and γ endorphins have different immune functions. While α endorphin, like methionin encephalin and leusin encephalin, plays a role in the production of antibodies, γ-endorphins has no such effect (Jankovic, 1994). In this study, when methionin encephalin was applied into the cerebral cavity, it was observed that it had a stronger immunomodulator effect than periferic application. It was determined that methionin encephalin had a repairing effect upon the immune system of aged rats. When 5 mg/kg of methionin encephalin was injected into the cerebral cavity, there was a decrease in T helper lymphocytes. However, when 0.001 mg/kg methionin encephalin was injected into the same area, an increase in T helper lymphocytes was observed (Jankovic, 1994). Endorphin and encephalin increase the activity of natural killer cells, the generation of cytotoxic T lymphocyte, the chemotaxis of monocytes and the production of interferon-γ (IF-γ), IL-1, IL-2, IL-4 and IL-6. In the studies on this subject (Jankovic, 1994; Millar *et al*., 1990), it has been concluded that endogen opioids creates an immunomodulatory effect. The immunomodulatory effect of acupuncture application was connected with the increase in levels of endogen opioids.

Kho and his colleagues (1991) evaluated the changes of IgA, IgM and IgG levels and total and differential leukocyte counts in the peripheral blood in 29 male patients taken during and 6 days after the upper abdominal surgery, which was, performed under two different anesthetic techniques. Stimulation of ear and para vertebral points, supplemented in small doses of fentanyl was applied in Group 1 and moderate doses of fentanyl were applied in Group 2. After surgery, a decrease in the levels of immunoglobulin, in lymphocyte and eosinophil counts and an increase in leukocyte and neutrophil counts were notified in both groups. No recovery was observed until the last assessment by the sixth day after surgery in IgA, IgG, leukocyte, neutrophil and lymphocyte counts in both groups, whereas IgM and eosinophil counts were recovered by the 4th day. Acupuncture and transcutaneous stimulation analgesia performed for major abdominal surgery did not influence the immune system of the body either during or after surgery as measured by the concentrations of immunoglobulin and total and differential WBC counts.

It is known that endorphin and encephalin increase the activity of natural killer cells, the generation of cytotoxic T lymphocyte, the chemotaxis of monocytes and the production of IF-γ, IL-1, IL-2, IL-4 and IL-6. IL-2, IL-4 and IL-6 functions stimulate the proliferation of B-lymphocytes (Carosella *et al*., 1989; Nies *et al*., 2002; Bertolini and Benson**,** 1990). IF-γ stimulates the production of IgG subclasses that activate the complement pathway and promote opsonization (Estes *et al*., 1994). Electroacupuncture stimulates IF-γ, IL-2, IL-4 and IL-6 affect immunoglobulin by increasing B cell proliferation and antibody synthesis. Those endogen opioids create an immunomodulatory effect. It is thought that the immunomodulatory effect of electro acupuncture may result from the increase in levels of endogen opioids (Jankovic, 1994; Millar *et al*., 1990). EA enhances or restores the NK cell activity but it is suppressed by an anterior hypothalamic area lesion (Hahm *et al*., 2004). Additionally, electroacupuncture stimulation on ST-36 acupuncture points in mice increases IF-γ, BE and NK cell activity in spleen (Hisamitsu *et al*., 2002). EA preserve IL-2 production of spleen lymphocytes from immunosuppressively injured rats (Cheng *et al*., 1997). EA could suppress the increase of apoptosis and Fas protein expression in splenic lymphocytes induced by the surgical trauma stress. Electroacupuncture could decrease splenic lymphocytes apoptosis via inhibiting Fas protein expression; consequently prevent deleterious immunological changes in the post-operative state (Wang *et al*., 2005).

Conclusion

Over the last decades, treatments of either invasive applications or wide-radical surgeries have been changing into less-invasive techniques and “if possible” drugs possess fewer side effects by means of technical improvement. For the developing need of nil-side effects and non-invasive treatments, in the future, acupuncture treatment and some sort of complementary medicine modalities seem to be much more popular together with related clinical and experimental data support. By acupuncture application, both at CNS and plasma, an increase of levels of β-endorphin, met-encephalin, leu-encephalin, serotonin has been observed. These neurotransmitters have immunomodulator effects on the immune system. For all of these above effects, acupuncture can be applied for immune related diseases and reduce risks of infection and tissue repair.

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