ISSN 0253-9756 Acta Pharmacol Sin 中国药理学报 1999 Sep; 20 (9): 769-777
http://www.simm.ac.cn; www.chinainfo.gov.cn/periodical

Review

Acupuncture: pain management coupled to immune stimulation

Randy L GOLLUB[1,2], Kathleen K S HUI[1] ([1]*Massachusettes General Hospital-NMR Center and* [2]*Department of Psychiatry, CNY 9109149 13th Street, Charlestown MA 02129, USA*); George B STEFANO[3] ([3]*Neuroscience Research Institute, State University of New York, Old Westbury NY 11568, USA*)

KEY WORDS acupuncture; peptide antibiotics; magnetic resonance imaging; methionine enkephalin

ABSTRACT

The phenomenon of acupuncture is both complex and dynamic. Recent information demonstrates that acupuncture may exert its actions on pain and immune processes. The coupling of these two systems occurs via common signaling molecules, ie, opioid peptides. In this regard, we surmise that opioid activation leads to the processing of opioid peptides from their precursor, proenkephalin, and the simultaneous release of antibacterial peptides contained within the precursor as well. Thus, central nervous system pain circuits may be coupled to immune enhancement. Furthermore, acupuncture needle manipulation elicited signal increases bilaterally in the region of the primary and secondary somatosensory corticies in human brain as determined by magnetic resonance imaging. The maps reveal marked signal decreases bilaterally in multiple limbic and deep gray structures including the nucleus accumbens, amygdala, hypothalamus, hippocampus, and ventral tegmental area. Taken together, we surmise a major central nervous system pathway as well as local pain and immune modulation during acupuncture.

INTRODUCTION

Acupuncture is an ancient medical practice of stimulating specific points on the body with needles. Over the past 2500 years the development of acupuncture has been based upon careful clinical observation and astute interpretation of the effects of treatment. According to traditional Chinese acupuncture, focal stimulation of the needle with the evocation of the *deqi* sensation is important in generating acupuncture effects. *Deqi* is a dull, aching sensation that develops at the site of acupuncture stimulation. However, the mechanisms by which acupuncture mediates its therapeutic effects are not yet understood. Traditionally, acupuncture practitioners have assumed that its beneficial effects are due to changes in the flow of life force, referred to as *Qi*. Advances in medical research have begun to identify the candidate chemical signaling molecules and neural circuitry that are the intermediary steps between acupuncture stimulation and improvements in health. For instance, there is substantial evidence that acupuncture analgesia is mediated at least in part by the release of the endogenous opioid peptides methionine enkephalin (ME) and β-endorphin[1,2]. Advances in the field of neuroimmunology have shown that these same endogenous opioid peptides are potent modulators of immune function[3-6], supporting a role for these signaling molecules in this phenomenon. In this review we develop a model for how acupuncture exerts its beneficial effects on immune function and pain suppression by actions on a common denominator, the endogenous opioids and opiates.

HOW COULD ACUPUNCTURE TREATMENT ENHANCE IMMUNE FUNCTION?

Immune function is compromised in people living with cancer. In addition to the intrinsic immunosuppressive action of the tumor, treatments for and

[3] Correspondence to Dr G B STEFANO.
Phn 1-516-876-2732. Fax 1-516-876-2727.
E-mail orgstefano@li.net
Accepted 1999-06-18

ISSN 0253-9756 Acta Pharmacol Sin 中国药理学报 1999 Sep; 20 (9)
E-mail aps@server.shcnc.ac.cn Phn/Fax 86-21-6474-2629

consequences of cancer have adverse effects on the normal functioning of the immune system. For instance, surgical trauma, irradiation, chemotherapy and malnutrition are all known to suppress immune function[7]. Decreased immune function is associated with greater susceptibility to infection and, in some cancers, with more rapid tumor progression. A major focus of immunology and oncology research is the development of effective methods to augment the immune system response to tumors[8,9].

Clinical and experimental data suggest that acupuncture may be a promising method to enhance immune function in people living with cancer[10-12]. Acupuncture has been shown to modulate both specific and non-specific immune functions. Indices of healthy immune function, such as levels of the cytokine interleukin-2, degree of natural killer cell (NKC) cytotoxicity, T-lymphocyte transformation rate and the ratio of helper T-cells to suppressor T-cells, are often suppressed in cancer patients. Studies have shown that treatment with acupuncture improves each of these indices[10,13-16]. Similar findings have been reproduced in animal models of cancer[17-19].

The presence of opioid peptide receptors on immunocytes and their role as modulators of the immune response with both enhancing and suppressing actions[3-67] suggest that these peptides may be a pathway by which acupuncture modulates immune function. Directly related to this is the finding that acupuncture augments endogenous opioid peptide levels in the cerebrospinal fluid and blood in both human and animal subjects[2].

In recent years a unique δ_2 opioid receptor subtype that is highly selective for ME[20] has been found on human immunocytes, suggesting that this opioid peptide is its signaling ligand. This observation was supported by the demonstration that ME can selectively increase the expression of the pro-inflammatory cytokine interleukin-6[21-23] as well as stimulate chemotaxis[20,22-29]. Furthermore, the plasma presence of ME-Arg-Phe, suggests that this naturally occurring opioid proenkephalin processed peptide may also serve this function[30] since it, as well as ME, is sensitive to naltrindole blockade[31]. Recently, these observations have been extended to include human endothelial cells that also express these delta receptor sites[32].

This concept of opioid peptide selectivity in the stimulation of immune activities has been enhanced by the finding that opiate alkaloids selectively suppress immune functions[3,33]. A novel μ opiate receptor subtype, designated μ_3, that is selective for opiate alkaloids but not endogenous or man made opioid peptides has also been demonstrated on these tissues. This dual expression of opioid receptor subtypes supports an opposing role for opiate alkaloids (whether naturally occurring morphine or exogenously administered opiate alkaloid drugs) in immune and vascular tissues[3,29,30,33-36]. Furthermore, naturally occurring morphine has been shown to antagonize the immune stimulating properties of opioid peptides[3,32]. In this regard, we have demonstrated that plasma levels of endogenous morphine increase many hours post-surgery in an attempt to restore neural, immune and vascular down regulation from the diffuse inflammatory response to surgery[3,21,22,37]. Thus, endogenous morphine serves to restore and limit immune and neural excitation, suggesting a protective function[38].

Based on these earlier studies, additional data has emerged demonstrating that various types of leukocytes (ie, macrophages) have the ability to make opioid peptides from the classical opioid precursors[39]. This ability appears to have originated in invertebrate organisms that also release opioid peptides (ie, ME) from their immunocytes[39-42]. Thus, in man, another source of these opioid peptides may be the skin itself[43,44], possibly released by macrophages located in the dermis. In this regard, acupuncture needles may stimulate this response, releasing these peptides that may not only affect pain perception but stimulate immune function as well (Fig 1).

In addition to providing opioid peptides that stimulate immune function, opioid precursors, ie, proenkephalin A, also contain antibacterial peptides bracketed by the same type of basic amino acids that surround ME. Once proteolytic processing of the proenkephalin molecule starts, both types of peptides are released[45-48]. This prompted us to suggest that opioid peptides may have originated within the context of immune function given this phenomenon's presence in animals that evolved 500 million years before man[49]. It is possible, therefore, that immune or neural signaling may lead to enhance proenkephalin proteolytic processing, freeing both opioid peptides and antibacterial peptides[49] (Fig 1).

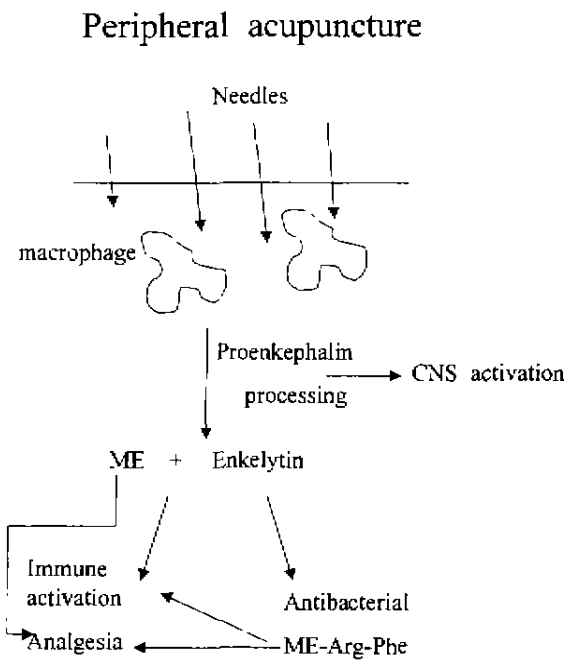

Fig 1. Hypothetical representation of methionine enkephalin (ME) mediated immune stimulatory and analgesia enhancing actions. LOCAL ACTIONS: Acupuncture needles inserted into the skin initiates immunocyte, ie, macrophage, ME release as well as proenkephalin processing. This results in the release of the opioid pentapeptide that then stimulates immunocyte activation and recruitment, causing an enhanced response reaching a more global level of signaling. Simultaneously to the release of ME is the release of enkelytin, a naturally occurring antibacterial peptide that also arises from proenkephalin by similar processing. This in turn protects the needle puncture area from microbial invasion. Enkelytin is then processed to ME-Arg-Phe, another opioid peptide with immunocyte activating and chemotaxic properties, enhancing the process once more. Arrows pointing to CNS indicates the network of neuronal activation produced by primary afferent fiber stimulation by the needle. Some of these actions are also mediated by the release of opioid peptides in the spinal cord and brain.

Incorporating this hypothesis within the phenomenon of acupuncture, we hypothesize that needle puncture or pressure on the skin may initiate the release of both types of peptides from resident immunocytes; opioid peptides stimulating additional immunocyte activation, chemotaxis and phagocytosis as well as the secretion of cytokines[3], and the antibacterial peptides attacking bacteria immediately (Fig 1). This process would allow time for the immunocyte-stimulating capabilities (ie, recruitment), of the opioid peptides to manifest themselves. In our model puncture of the skin causes activation of immunocytes and contributes to acupuncture analgesia and immunoregulation by virtue of the signaling through opioid peptides; however, this is unlikely to be the major pathway of acupuncture action in traditional Chinese acupuncture. Rather this initial stimulus activates neural processes that initiate CNS mediated effects including analgesia. We will now describe the evidence supporting a direct role of neuronal activity in mediating the therapeutic effects of acupuncture treatment, for both the immune stimulating and pain suppressing actions.

HOW COULD ACUPUNCTURE ENHANCE ENDOGENOUS ANALGESIA?

Current theory holds that the analgesic effects of acupuncture are mediated through descending inhibitory pathways localized in the brainstem[1,2,50]. Peripheral stimulation of sensory afferent fibers activates spinal neurons, which in turn activate multiple central brain regions including the brain stem, midbrain and hypothalamus. Acupuncture needle manipulation below the level of traumatic spinal cord damage does not generate *deqi* or therapeutic acupuncture effects. Clinically, in syringomyelia patients whose spinal pain and temperature afferent tracts are damaged, acupuncture stimulation beneath the level of damage will not produce any *deqi* sensation and acupuncture therapeutic effects are abolished or markedly reduced. Multiple investigators working with animal preparations have demonstrated that bilateral section of the ventrolateral fasciculus of the spinal cord abolishes acupuncture analgesia. These studies demonstrate unequivocally that the central nervous system is crucial to the analgesic actions of acupuncture.

Much work has been done to characterize the specific types of peripheral sensory neurons and the neurotransmitter receptors that they express in acupuncture points that are strongly analgesic. The analgesic action does not depend as much on the specificity of points and meridians as claimed by

ISSN 0253-9756 Acta Pharmacol Sin 中国药理学报 1999 Sep; 20 (9)
E-mail aps@server.shcnc.ac.cn Phn/Fax 86-21-6474-2629

traditional acupuncture theory, but more on the density and types of receptors and of conducting fibers at the site. "Among 10 different acupuncture points tested in human volunteers using potassium iontophoresis to induce experimental pain, Hegu (Large Intestine 4, LI 4) was found to be one of the most effective acupuncture points to produce a general analgesic effect" (Research Group of Acupuncture Anesthesia, 1973). This is likely due to the dense innervation of A-beta fibers in this area[51]. The gate control mechanism must be involved in acupuncture induced segmental analgesia, especially when acupuncture was given at a local tender point (the "Ashi" point or "Ah yes" point). The ancient acupuncturists recognized that the entire first interosseous space surrounding Hegu exerts acupuncture effects and named it the Hegu area.

Powerful emerging technology of functional neuroimaging is beginning to reveal the temporal and spatial details of how acupuncture needle manipulation modulates neuronal activity (Fig 2). A recent study by Cho and colleagues employed functional magnetic resonance imaging (fMRI) to study the effect of stimulation at acupuncture points that are indicated for treatment of eye disorders[52]. This experiment demonstrated that stimulation of these acupuncture points results in regionally specific activation in the occipital cortical areas by mediate vision[52]. Consistent with this report, Hui and colleagues have recently demonstrated that acupuncture needle manipulation at right or left LI 4 produces significant bilateral activation of somatosensory cortex across the territory subserving hand, face and upper body while signal decreases were observed in deep brain structures (Fig 2)[53,54].

Elevation of pain tolerance threshold by acupuncture is even more crucial for surgical analgesia than elevation of pain perception threshold[55]. This implies that the processing of affective information through limbic brain regions above the brainstem is important for the therapeutic efficacy of acupuncture. Consistent with this hypothesis is the demonstration that acupuncture needle manipulation at LI 4 decreases fMRI signals in the nucleus accumbens, amygdala, hippocampus, parahippocampus, hypothalamus, ventral tegmental area, cingulate gyrus, caudate, putamen, temporal pole, and insula in all subjects who experience acupuncture sensation (Fig 2)[53,54]. Importantly, puncturing the skin with the acupuncture needle and leaving it at rest for the same time period did not cause the fMRI signal decreases that occurred with needle manipulation; underscoring the importance of neuronal signaling to acupuncture effects. Nor did it produce the *deqi* sensation that is crucial to acupuncture efficacy.

Much evidence in animals and humans supports the hypothesis that opioid peptides acting at multiple brain regions including the brainstem, midbrain and hypothalamus are crucial to acupuncture analgesia. For instance, systemic or locally applied opiate receptor antagonists block acupuncture analgesia. Acupuncture augments endogenous opioid peptide levels in cerebrospinal fluid (CSF) and blood while decreases them in specific brain regions. And the acupuncture analgesia transmitted to a second animal by CSF transfer or cross-circulation can be reversed with naloxone[2]. Acupuncture-like stimulation has been shown to stimulate heterosegmental release of ME-like material in the rat spinal cord[56]. Injection of antibodies against ME and other opioid peptides into the periaqueductal gray matter of rabbits decreases the analgesic effect of electroacupuncture[57]. In humans with chronic pain acupuncture resulted in significant improvement of pain symptoms and this was associated with higher ME plasma levels[58]. Further evidence in support of the role of opioid peptides in the phenomenon of acupuncture analgesia is the demonstration that inhibition of enzymes that process these peptides into inactive products potentiates acupuncture analgesia in animals[59,60].

On the other hand there have been studies that demonstrate acupuncture stimulation significantly reduces plasma opioid-peptide-like levels[61]. However, it must be emphasized the timing of the sampling is quite important since plasma ME levels are subject to enzymatic regulation by neutral endopeptidase[39]. The activity of this enzyme is exquisitely sensitive to regulation by immune events. Therefore, future acupuncture studies must control for enzymatic processing of opioid peptides in the plasma[7, 62, 63].

Within the scope of acupuncture, we envisage a central modulating role for the opioid peptide ME. As noted earlier, this peptide could mediate the analgesic effect of acupuncture while simultaneously stimulating immune function[49]. A recent study using an animal

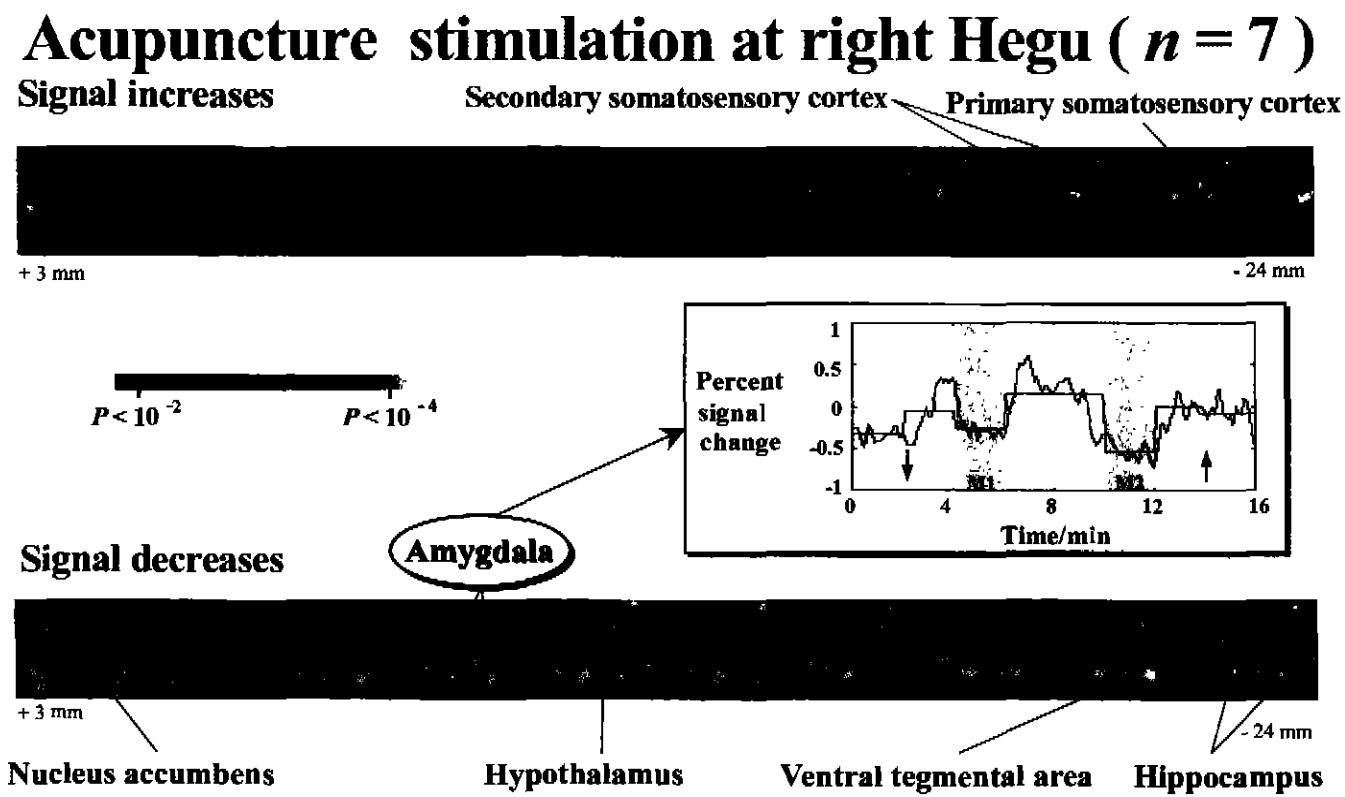

Fig 2. fMRI signal increases in somatosensory corticies and signal decreases in deep structures: results of acupuncture needle manipulation at **LI 4** of the right hand. The averaged result from a cohort of seven subjects is shown. Pseudocolor Kolmogorov-Smirnov statistical maps of signal increases (top row) and signal decreases (bottom row) are shown overlaid on the same high resolution anatomical scan in gray scale. The slice plane is indicated relative to the anterior commissure. Acupuncture needle manipulation elicited signal increases bilaterally in the region of the primary and secondary somatosensory corticies. The maps reveal marked signal decreases bilaterally in multiple limbic and deep gray structures including the nucleus accumbens, amygdala, hypothalamus, hippocampus and ventral tegmental area. The inset shows the time course of signal change from a representative region, the amygdala. The experimental paradigm was as follows. Scanning began 2 min prior to needle insertion. The needle was left at rest for 2 min before manipulation. Two epochs of needle manipulation (**M1, M2**) each lasting 2 min, were administered separated by a 4-min rest interval. The needle was removed 2 min after **M2**. Scanning was continued for an additional 2 min. For statistical analyses, the mean signal intensity of the 3 periods of needle at rest served as the baseline for changes in signal intensity during needling. Color bar shows the statistical significance, same color if signal increased or decreased. The horizontal line of each epoch represents the average signal intensity of that epoch. $n = 7$ replicates.

ISSN 0253-9756 Acta Pharmacol Sin 中国药理学报 1999 Sep; 20 (9)
E-mail aps@server.shcnc.ac.cn Phn/Fax 86-21-6474-2629

ISSN 0253-9756 Acta Pharmacol Sin 中国药理学报 1999 Sep; 20 (9)
E-mail aps@server.shcnc.ac.cn Phn/Fax 86-21-6474-2629

model supports the hypothesis of a dual role for the opioid peptides in modulating the function of the immune system and neural regulation of pain perception. Animals that demonstrated a significant degree of acupuncture analgesia also improved in NKC activity; the animals that did not have an analgesic response did not have a change in NKC activity[19]. Furthermore, when the analgesia non-responders were pretreated with *D*-phenylalanine, an opioid peptidase inhibitor, to enhance opioid peptide activity, the animals became responsive to acupuncture demonstrating both analgesia and improved NKC activity[19].

This model clarifies some important clinical observations. For instance, many of the commonly prescribed pain medications are known to interfere with normal immune system function[3,4]. This could be caused by opiate suppression of the immune system, mediated by μ_3 opiate alkaloid sensitive and opioid peptide insensitive receptors[25,32,64]. Furthermore, it is known that acupuncture analgesia not only reduces the requirement for these medications but can also attenuate the immunosuppressive side effect of opiates[12]. We surmise this may be due to the action of ME acting on immunocyte δ_2 opioid receptors.

WHY CONSIDER THE EFFECTS OF ACUPUNCTURE ON IMMUNE FUNCTION AND ANALGESIA TOGETHER?

The concept of pain is dynamically complex and we would refer the reader to a recent report since it more fully reflects our thinking on this phenomenon[49]. Briefly, "the association of antibacterial and opioid peptides reflects the fact that both types of molecules have evolved to fight the presence of microbes. Once this association was established organisms needed an early detection/surveillance system that could continuously monitor microbial penetration and growth. What is better alerting process than one that signaled attention by creating a noxious sensation such as pain? The following scenario might be envisaged: under serious situations commanding attention, the sensation of pain must subside momentarily (ie. analgesia) to allow for an appropriate response to the stimulus. While the organism is orienting itself to the stimulus, the simultaneous release of enkelytin, analgesic and immunocyte-stimulating opioids combats the pathogenic challenge of a bacterial presence. Once this is over, the pain-provoking process can emerge once more, possibly even at a stronger level, resulting in behaviors designed to alleviate the condition. Indeed, if pain evolved to fit this function, it evolved in association with immune processes. Furthermore, these opioid mediated activities were probably enhanced during evolution as the central nervous system (CNS) became closed off from the circulatory system by the blood-brain barrier, ultimately isolating the ganglionic neural activities that also required immune surveillance. However, regardless of the barrier, various immune cells (ie. those responsive to opioid peptides) were always allowed access to the isolated tissues, some taking up residence in the CNS as microglia. The reason for the evolving relationship between opioid neural and immune processes now appears quite simple, that is analgesic priority-setting activities associated with an anti-infectious/anti-inflammatory process. This combination would provide a high degree of survival benefit to any organism since it would ensure appropriate behavior to meet not only these non-cognitive challenges, but also cognitive ones".

Interestingly, as also noted above, the present hypothesis complements a large body of work that demonstrates a role for neural opioid and opiate processes in acupuncture[65-71]. These studies have implicated delta-opioid receptors in this phenomenon[65] as well as identified associated brain areas[66,67,69,70]. In addition, these works also document the interact ion with other neurotransmitter processes as well[67-71].

FUTURE STUDIES

One of the greatest challenges in acupuncture research has been the design of an adequate control condition. Few attempts have satisfied Western scientific standards[72,73]. The meridians and acupuncture points are derived from clinical experience; however, it has been reported that needling at non-classical acupuncture points can also have therapeutic effects[50,72-75]. The hypothetical model we are currently proposing could explain these results. Needle penetration of the skin, whether at the acupuncture

point or anywhere on the skin, could result in the release of ME that would have effects on the immune system and on the neuronal circuitry regulating pain perception. The analgesic actions of ME could be mediated both centrally and peripherally[2,75]. This could account for the high rate of placebo responders in studies employing "sham" acupuncture at non-traditional acupuncture points. Thus it is crucial that future studies are needed that purport to study acupuncture rigorously for these possibilities.

In conclusion, there is evidence that acupuncture suppresses pain perception and enhances immune function. Given the new information presented in this review about a possible mechanism of action, we feel we are moving closer to understanding this phenomenon. However, considerable effort is still required to reveal the secrets of how this ancient Chinese healing method works.

REFERENCES

1 Han JS, Terenius L. Neurochemical basis of acupuncture analgesia. Ann Rev Pharmacol Toxicol 1982; 22: 193 – 220.
2 Stux G, Pomeranz B. Basics of acupuncture. 4th ed. Berlin: Springer-Verlag; 1997. p322.
3 Stefano GB, Scharrer B, Smith EM, Hughes TK, Magazine HI, Bilfinger TV, *et al*. Opioid and opiate immunoregulatory processes. Crit Rev Immunol 1996; 16: 109 – 44.
4 Carr DJJ, Rogers TJ, Weber RJ. The relevance of opioids and opioid receptors on immunocompetence and immune homeostasis. Proc Soc Exp Biol Med 1996; 213: 248 – 57.
5 Peterson PK, Molitor TW, Chao CC. The opioid-cytokine connection. J Neuroimmunol 1998; 83: 63 – 9.
6 Li XY. Immunomodulating effects of methionine enkephalin. Acta Pharmacol Sin 1999; 19: 3 – 6.
7 Abbas AK, Lichtman AH, Pober JS. Congenital and acquired immunodeficiencies. 1997; 439 – 461.
8 Lanzavecchia A. Identifying strategies for immune intervention. Science 1993; 260: 937 – 44.
9 Boon T, Cerottini J, Van den Eynde B, Van der Bruggen P, Van Pel A. Tumor antigens recognized by T lymphocytes. Annu Rev Immunol 1994; 12: 337 – 65.
10 Wu B, Zhou RX, Zhou MS, Li M. Effect of acupuncture on interleukin-2 level and NK cell immunoactivity of peripheral blood of patients with malignant tumor. Chin J Integ Trad West Med 1994; 14: 537 – 9.
11 Bensoussan A, Meyers SP. Towards a safer choice. The practice of traditional Chinese medicine in Australia. Sydney: University of Western Sydney Press, 1996.
12 Zhang Y, Du L, Wu GC, Cao XD. Electro-acupuncture (EA) induced attenuation of immunosuppression appearing after epidural or intrathecal injection of morphine in patients and rats. Acupunct Electrother Res 1996; 21: 177 – 86.
13 Chen LL, Jie GY, Jiang KW, Gao JW, Zhang JM. Modulatory effect of acupuncture on immune function of lung cancer patients. Chin J Acupunct Moxibustion 1997; 17: 197 – 9.
14 Li H, Yu YC, Dai TC. Acupuncture treatment for side reactions of radiation and chemotherapy for cancer patients. Chin J Acupunct Moxibustion 1997; 17: 327 – 8.
15 Wu B, Zhou RX, Zhou MS. Effect of acupuncture on immunomodulation in patients with malignant tumors. Chin J Integ Trad West Med 1996; 16: 139 – 41.
16 Yuan JG, Zhou RX. Effect of acupuncture on T-lymphocyte and its subsets from the peripheral blood of patients with malignant neoplasm. Acupunct Electrother Res 1993; 18: 174 – 7.
17 Liu LJ, Guo CJ, Jian XM. Effect of acupuncture on immune function and histopathology of transplanted mammary cancer in mice. Chin J Integ Trad West Med 1995; 15: 615 – 7.
18 Liu HW, Fung XD, Pang B, Fung MQ, Fung MX. Effect of acupuncture on blood viscosity and tumor growth in tumor bearing mice. J Trad Chin Med 1997; 38: 327 – 8.
19 Fang JQ, Jie GY, Xu F, Chen PF, Lin WM. Influence of acupuncture analgesia on NK cytotoxicity of tumor-bearing rats induced by electromoxibustion. Acupunct Electrother Res 1998; 23: 117 – 21.
20 Stefano GB, Melchiorri P, Negri L, Hughes TK, Scharrer B. (D-Ala2)-Deltorphin I binding and pharmacological evidence for a special subtype of delta opioid receptor on human and invertebrate immune cells. Proc Natl Acad Sci USA 1992; 89: 9316 – 20.
21 Brix-Christensen V, Tonnesen E, Sorensen IJ, Bilfinger TV, Sanchez RG, Stefano GB. Effects of anesthesia based on high versus low doses of opioids on the cytokine and acute phase protein responses in patients undergoing cardiac surgery. Acta Anaesthesiol Scand 1998; 42: 63 – 70.
22 Tonnesen E, Brix-Christensen V, Bilfinger TV, Sanchez RG, Stefano GB. Endogenous morphine levels increase following cardiac surgery: decreasing proinflammatory cytokine levels and immunocyte activity. J Int Cardiol 1998; 62: 191 – 7.
23 Zhong F, Li XY, Yang S, Stefano GB, Fimiani C, Bilfinger TV. Methionine-enkephalin stimulates interleukin-6 mRNA expression: Human plasma levels in coronary artery bypass grafting. Int J Cardiol 1998; 64: S53 – S59.
24 Stefano GB, Cadet P, Scharrer B. Stimulatory effects of opioid neuropeptides on locomotory activity and conformational changes in invertebrate and human immunocytes: Evidence for a subtype of delta receptor. Proc Natl Acad Sci USA 1989; 86: 6307 – 11.
25 Stefano GB, Liu Y. Opiate antagonism of opioid actions

ISSN 0253-9756 Acta Pharmacol Sin 中国药理学报 1999 Sep; 20 (9)
E-mail aps@server.shcnc.ac.cn Phn/Fax 86-21-6474-2629

on immunocyte activation and nitric oxide release. Anim Biol 1996; 1: 11 – 6.

26 Bilfinger TV, Fricchione GL, Stefano GB. Neuroimmune implications of cardiopulmonary bypass. Adv Neuroimmunol 1993; 3: 277 – 88.

27 Bilfinger TV, Stefano GB. Evidence of immunocyte stimulatory molecules(s) in plasma of patients undergoing cardiopulmonary bypass.
J Cardiovasc Surg 1993; 34: 129 – 33.

28 Stefano GB, Bilfinger TV. Human neutrophil and macrophage chemokinesis induced by cardiopulmonary bypass; loss of DAME and IL-1 chemotaxis.
J Neuroimmunol 1993; 47: 189 – 98.

29 Stefano GB, Smith EM, Cadet P, Hughes TK. HIV GP120 alteration of DAMA and IL-1a induced chemotaxic responses in human and invertebrate immunocytes.
J Neuroimmunol 1993; 43: 177 – 84.

30 Stefano GB, Shipp MA, Scharrer B. A possible immunoregulatory function for [Met]-enkephalin-Arg6-Phe7 involving human and invertebrate granulocytes.
J Neuroimmunol 1991; 31: 97 – 103.

31 Stefano GB, Casares F, Liu U. Naltrindole sensitive δ_2 opioid receptor mediates invertebrate immunocyte activation. Acta Biol Hung 1995; 321 – 7.

32 Stefano GB, Salzet M, Hughes TK, Shao L, Wang Y, Bilfinger TV. δ_2 Opioid receptor subtype on human vascular endothelium uncouples morphine stimulated nitric oxide release. Int J Cardiol 1998; 64: S43 – S51.

33 Stefano GB, Scharrer B. Endogenous morphine and related opiates, a new class of chemical messengers.
Adv Neuroimmunol 1994; 4: 57 – 68.

34 Stefano GB, Digenis A, Spector S, Leung MK, Bilfinger TV, Makman MH, *et al*. Opiate-like substances in an invertebrate, a novel opiate receptor on invertebrate and human immunocytes, and a role in immunosuppression. Proc Natl Acad Sci USA 1993; 90: 11099 – 103.

35 Stefano GB, Leung MK, Bilfinger TV, Scharrer B. Effect of prolonged exposure to morphine on responsiveness of human and invertebrate immunocytes to stimulatory molecules. J Neuroimmunol 1995; 63: 175 – 81.

36 Stefano GB, Hartman A, Bilfinger TV, Magazine HI, Liu Y, Casares F, *et al*. Presence of the m3 opiate receptor in endothelial cells; coupling to nitric oxide production and vasodilation. J Biol Chem 1995; 270: 30290 – 3.

37 Stefano GB. Autoimmunovascular regulation; morphine and anandamide stimulated nitric oxide release.
J Neuroimmunol 1998; 83: 70 – 6.

38 Stefano GB. Substance abuse and HIV-gp120; are opiates protective? Arch Immunol Ther Exp (Warsz) 1999; 47: 99 – 106.

39 Stefano GB, Salzet M. Invertebrate opioid precursors; evolutionary conservation and the significance of enzymatic processing. Int Rev Cytol 1999; 187: 261 – 86.

40 Salzet M, Cocquerelle C, Verger-Bocquet M, Pryor SC, Rialas CM, Laurent V, Stefano GB. Leech immunocytes contain proopiomelanocortin; nitric oxide mediates hemolymph POMC processing. J Immunol 1997; 159: 5400 – 11.

41 Salzet M, Stefano GB. Invertebrate proenkephalin; delta opioid binding sites in leech ganglia and immunocytes. Brain Res 1997; 768: 224 – 232.

42 Salzet M, Stefano GB. Prodynorphin in invertebrates. Mol Brain Res 1997; 52: 46 – 52.

43 Nissen JB, Lund M, Stengaard-Pedersen K, Kragballe K. Enkephalin-like immunoreactivity in human skin is found selectively in a fraction of CD68-positive dermal cells; increase in enkephalin-positive cells in lesional psoriasis. Arch Dermatol Res 1997; 289: 265 – 71.

44 Schroder JM. Epithelial peptide antibiotics. Biochem Pharmacol 1999; 57: 121 – 34.

45 Goumon Y, Lugardon K, Kieffer B, Lefevre JF, Van Dorsselaer A, Aunis D, Metz-Boutigue MH. Characterization of antibacterial COOH-terminal proenkephalin-A-derived peptides (PEAP) in infectious fluids. Importance of enkelytin, the antibacterial PEAP209-237 secreted by stimulated chromaffin cells. J Biol Chem 1998; 273: 29847 – 56.

46 Goumon Y, Strub JM, Moniatte M, Nullans G, Poteur L, Hubert P, *et al*. The C-terminal biophosphorylated proenkephalin-A-(209 – 237)-peptide from adrenal medullary chromaffin granules possesses antibacterial activity. Eur J Biochem 1996; 235: 516 – 25.

47 Goumon Y, Strub JM, Moniatte M, Nullans G, Poteur L, Hubert P, *et al*. The C-terminal bisphosphorylated proenkephalin-A-(209 – 237)-peptide from adrenal medullary chromaffin granules possesses antibacterial activity [published erratum appears in Eur J Biochem 1996; 237: 883]. European Journal of Biochemistry 1996; 235: 516 – 25.

48 Metz-Boutigue MH, Goumon Y, Lugardon K, Strub JM, Aunis D. Antibacterial peptides are present in chromaffin cell secretory granules.
Cell Mol Neurobiol 1998; 18: 249 – 65.

49 Stefano GB, Salzet B, Fricchione GL. Enkelytin and opioid peptide association in invertebrates and vertebrates; immune activation and pain. Immunol Today 1998; 19: 265 – 8.

50 Melzack R. Folk medicine and the sensory modulation of pain. In: Wall PD, Melzack R, editors. Text book of pain. Edinburgh: Churchill Livingstone; 1989. p897 – 905.

51 Lu GW. Characteristics of afferent fiber innervation on acupuncture point Zusanli.
Am J Physiol 1983; 245: r606 – r612.

52 Cho ZH, Chung SC, Jones JP, Park JP, Park HJ, Lee HJ, *et al*. New findings of the correlation between acupoints and corresponding brain cortices using functional MRI. Proc Nat Acad Sci USA 1998; 95: 2670 – 3.

53 Hui KKS, Liu J, Chen AJW, Makris N, Kennedy DN, Moore CI, *et al*. Effects of acupuncture on human limbic system and basal ganglia measured by fMRI. Neuroimage 1997; 5: S226.

54 Hui KKS, Liu J, Chen AJW, Makris N, Gollub RL, Kennedy DN Jr, *et al*. Acupuncture modulates the limbic system and subcortical gray structures of the human brain-direct evidence by fMRI. Neuroimage 1999; 7: s441.

55 Cao XD, Xu SF, Lu WX. Inhibition of sympathetic nervous system by acupuncture. Acupunct Electrother Res 1983; 8: 25 - 35.

56 Bing Z, Cesselin F, Bourgoin S, Clot AM, Hamon M, Le Bars D. Acupuncture-like stimulation induces a hetero-segmental release of Met-enkephalin-like material in the rat spinal cord. Pain 1991; 47: 71 - 7.

57 Han JS, Xie GX, Zhou ZF, Folkesson R, Terenius L. Acupuncture mechanisms in rabbits studied with microinjection of antibodies against beta-endorphin, enkephalin and substance P. Neuropharmacology 1984; 23: 1 - 5.

58 Kiser RS, Khatami MJ, Gatchel RJ, Huang XY, Bhatia K, Altshuler KZ. Acupuncture relief of chronic pain syndrome correlates with increased plasma met-enkephalin concentrations. Lancet 1983; 2: 1394 - 6.

59 Ehrenpreis S. Pharmacology of enkephalinase inhibitors: animal and human studies. Acupunct Electrother Res 1985; 10: 203 - 8.

60 Kishioka S, Miyamoto Y, Fukunaga Y, Nishida S, Yamamoto H. Effects of a mixture of peptidase inhibitors (amastatin, captopril and phosphoramidon) on Met-enkephalin-, beta-endorphin-, dynorphin-(1 - 13)- and electroacupuncture-induced antinociception in rats. Jpn J Pharmacol 1994; 66: 337 - 45.

61 Szczudlik A, Lypka A. Plasma immunoreactive beta-endorphin and enkephalin concentration in healthy subjects before and after electroacupuncture. Acupunct Electrother Res 1983; 8: 127 - 37.

62 Shipp MA, Stefano GB, D'Adamio L, Switzer SN, Howard FD, Sinisterra JI, *et al*. Downregualtion of enkephalin-mediated inflammatory responses by CD10/neutral endopeptidase 24.11. Nature 1990; 347: 394 - 6.

63 Weissman D, Poli G, Fauci AS. IL-10 synergizes with multiple cytokines in enhancing HIV production in cells of monocytic lineage. J Acquir Immune Defic Syndr Hum Retrovirol 1995; 9: 442 - 9.

64 Prevot V, Rialas C, Croix D, Salzet M, Dupouy J-P, Puolain P, *et al*. Morphine and anandamide coupling to nitric oxide stimulated GnRH and CRF release from rat median eminence: neurovascular regulation. Brain Res 1998; 790: 236 - 44.

65 He XP, Cao XD. Effects of intrahippocampal delta-receptors on inhibition of electroconvulsive shock by electro-acupuncture. Acta Pharmacol Sin 1989; 10: 197 - 201.

66 Liu GJ, Wang S. Effects of nucleus raphe magnus and locus coeruleus in descending modulation of habenula on pain thresthold and acupuncture analgesia. Acta Pharmacol Sin 1988; 9: 18 - 22.

67 Pan JC, Zhang ZD, Yu LP, Wu AJ, Ding M, Jin GZ. Potentiation of norepinephrine on rat acupuncture analgesia antagonized by arcuate nucleus lesion or naloxone blocked on opiate receptors in periaqueductal gray. Acta Pharmacol Sin 1989; 10: 118 - 22.

68 He XP, Cao XD. Effects of etorphine and electro-acupuncture on electroconvulsive shock in rats. Acta Pharmacol Sin 1986; 7: 106 - 9.

69 Mo WY, Chou YT, Wang GN. Analgesia by injection of morphine and antagonism of electro-acupuncture analgesia by injection of naloxone into septal area of rabbits. Acta Pharmacol Sin 1982; 3: 150 - 4.

70 Cheng JS, Ou SP, He XP. Effect of electro-acupuncture on neurons of midbrain periaqueductal gray sensitive to morphine and 5-HT in rabbits. Acta Pharmacol Sin 1982; 3: 154 - 9.

71 Li SJ, Tang J, Han JS. Tolerance to 5-HT and its implication in electro-acupuncture tolerance and morphine tolerance. Acta Pharmacol Sin 1982; 3: 159 - 63.

72 Vincent CA, Richardson PH. The evaluation of therapeutic acupuncture: concepts and models. Pain 1986; 24: 1 - 13.

73 Richardson PH, Vincent PA. Acupuncture for the treatment of pain: a review of evaluative research. Pain 1986; 24: 15 - 40.

74 Margolin A, Chang P, Avants SK, Kosten TR. Effects of sham and real auricular needling: implications for trials of acupuncture for cocaine addiction. Am J Chin Med 1993; 21: 103 - 11.

75 Schafer M, Carter L, Stein C. Interleukin-1 beta and corticotropin-releasing factor inhibit pain by releasing opioids from immune cells in inflamed tissue. Proc Nat Acad Sci USA 1994; 91: 4219 - 22.

PP, 20(5) 768-777

针刺：痛觉调制与免疫激动耦连

Goll->PL Hui KKS R245-0

Randy L GOLLUB[1,2], Kathleen K S HUI[1]
([1]*Massachusettes General Hospital-NMR Center and* [2]*Department of Psychiatry, CNY 9109149 13th Street, Charlestown MA 02129, USA*); George B STEFANO[3]
([3]*Neuroscience Research Institute. State University of New York, Old Westbury NY 11568, USA*)

关键词 针刺；肽抗生素类；磁共振成像；甲硫氨酸脑啡肽

（责任编辑 朱倩蓉）