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Imaging of biophoton emission from electrostimulated skin acupuncture point jg4: Effect of light enhancers

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Using an ultrasensitive CCD camera, an extremely low light intensity from the acupuncture-sensitive point JG4 at the left hand was recorded. As the intensity of the light was very weak and the time of electrostimulation exceeded the recommended period, the quality of biophoton images was poor. Chemiluminescent and fluorescent hydrophilic, hydrophobic and amphyphilic molecular probes were used to: (i) ensure penetration of probes into skin, (ii) enhance the intensity of BP emission, (iii) shorten time and (iv) obtain information about mechanisms of biophotons generation in EAP-sensitive points and channels. The results obtained partially fulfilled expectations and indicate on the necessity to elaborate special techniques of probes deposition on the skin.

Keywords: Biophotons, Electroacupuncture, Luminescent probes, Skin electrostimulation

Human body is known to emit ultra weak electromagnetic radiation (biophotons, BP) in the visible and near infrared spectral region. Correlations between parameters of BP and the functional state of the organism (stress, relaxation, meditation, pathological states, death) are now investigated[1-6].

It is expected that the acupuncture (AP), an old Chinese treatment of pain release may be also associated with BP emission. In a preliminary study using a modern version of AP, namely electroacupuncture (EAP), it was found that the electrostimulation of JG4 point at the hand results in the intensity and time duration of BP slightly different from BP of non stimulated skin[7]. Therefore in this study it was investigated whether the intensity of BP and electrostimulation time can be positively changed by application of chemiluminescent and fluorescent hydrophilic, hydrophobic and amphyphilic molecular probes overlayed on the skin. Moreover, the use of this kind of probes might deliver valuable information on the biochemical mechanism and energetics of BP generation in EAP active points.

Acupuncture (AP) produces analgesic effect and electroacupuncture (EP) is more effective than manual AP[1-3]. Electrical stimulation via skin path electrodes is as effective as AP. The prevention of EA-induced analgesia by naloxone and by antiserum against endorphins suggests that endorphins are involved. The release of endorphins into cerebrospinal fluid (CSF) was found after EA. Low frequency (2Hz) and high frequency (100Hz) of EA selectively induces the release of different types of endogenous types of opioids from the CNS – enkephalins and dynorphins in both animals and humans. Therefore EA produces an increase in pain threshold. A living system may be considered as an aggregation of matter organized to maintain energy flow with a guided purpose. It is also emitting and receiving focus of electromagnetic messages and constitutes an oscillating circuit that is capable to reach resonance with one of these waves i.e. corresponding to the frequency of the circuit. An ultra weak photon emission was recorded from electrostimulated acupuncture points using an ultrasensitive CCD camera[7]. In the present study an effort has been made to explore chemiluminescent and fluorescent hydrophilic, hydrophobic and amphiphilic molecular probes to: (i) enhance the intensity (ii) shorten time and (iii) obtain information about reactions of biophotons generation, energetics and possible mechanisms of energy transfer in electrostimulated acupuncture-sensitive points.

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Materials and Methods

Several types of chemiluminescent and luminescent light enhancers (molecular probes) have been tested: (i) Fluorescence photosensitizers (S) capable of effective transfer of the electronic energy from excited molecules (M^*) probably generated by EAP in the skin tissue. Hydrophillic, amphiphillic and hydrophobic strong fluorescers have been choosen to ensure penetration of enhancers into skin: riboflavine (vitamin B2), 7-hydroxycoumarin (umbelliferone) and 9,10-diphenyl anthracene; (ii) Chemiluminescent enhancers such as 3-amino-phthalhydrazide (luminol) and dimethyl diacridinium dinitrate (lucigenin) which react with reactive oxygen species (ROS) giving off a strong chemiluminescence. Reagents were of analytical purity from Sigma. Spectral characteristic of the used luminogenic probes is given in Table 1.

In the experiments a specific acupuncture point JG4 located at the left hand was used for electrostimulation. To obtain the control data, a non-acupuncture sensitive area (N) ca 4 cm left, at the metacarpus, between the left forefinger and middle finger was choosen. This area (N) does not give a signal indicating a low galvanic skin resistance.

For electrostimulation an Autopunkter TMPlusTest (InfTELMark) was applied. This device has the atest # 1511836 of the ministry of health. It produces the maximum output current 40 mA and pulses consisting of low and high frequency oscillations (patent # P-291121). Detailed description is given elsewhere[8].

The left hand with attached two active pin electrodes was inserted in the light-tight compartment of the CCD camera trough a light-tight sleeve, properly positioned and fixed to avoid any movement.

To monitor photon emission, a high sensitive 16 bit charge coupled device (CCD) imaging camera (512 × 512 pixels) Molecular Light Imager 981 "Night Owl" (EG&G Berthold) was used. The camera is sensitive in the 180-1100 nm wavelength range with the maximum sensitivity at around 650 nm and the quantum efficiency 70-80% at this wavelength. For the standardization procedure, a radioluminescent 63-Ni-porcelain-incorporated standard was used. Other details are given elsewhere[9].

Results and Discussion

Theoretically, an increase in the light output can also be achieved by using strongly fluorescing

Table 1— Spectral characteristic of the chemiluminescent and fluorescent enhancers[10]

Hydrophilic	Compound	Wavelength, nm		Quantum yield
		Excitation	Emission	
Enhancers	Riboflavine	360	500-600, max 530	0.85
	Lucigenine			
	Fluorescence	410	460	0.75
	Chemiluminescence	-	465	0.017
	Luminol			
	Fluorescence	280	425	0.87
	Chemiluminescence	-	430	0.02
Hydrophobic enhancers	9,10-diphenylanthracene			
	in hexane	250, 270, 320	380, 410, 486	0.55
	in oil	315	410	0.5
Amphiphilic enhancers	7-hydroxy-coumarin (umbelliferone) in water or *p*H 7-9 buffers			
	Fluorescence	360	400-550, max 470	0.87
	Chemiluminescence	-	410-560, max 470	

compounds, which are able to accept electronic excitation energy of reactive/excited primary products generated in electrochemical reactions (energy donors). Such an energy transfer process from the primary donors to acceptors (strongly fluorescing compounds) requires overlapping of the energy levels of donors and acceptors and can occur by the radiationless Foerster or Dexter mechanism as schematically shown in the Fig. 1.

The use of hydrophilic and hydrophobic luminescent enhancers may provide an additional information about the milieu of electrochemical and electrophysiological processes taking place during electroacupuncture treatment.

Table 1 gives a basic characterization of luminescent hydrophilic and hydrophobic properties of enhancers used in preliminary experiments[10].

From the images recorded (Figs 2 and 3) and quantitative (numerical) values of measurements of luminescence intensity, it appears that the electrostimulation of the JG4 acupuncture active point give weaker biophoton emission (1805 photons/mm^{2}s) than the stimulation of

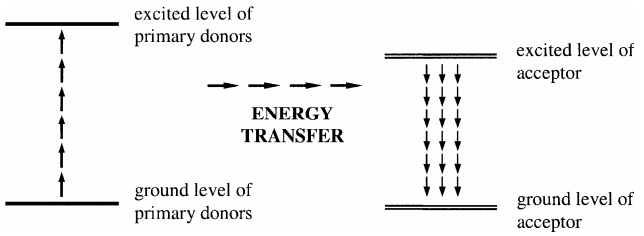

Fig. 1—Schematic diagram explaining the possible mechanism of light enhancement with the use of luminescent probes

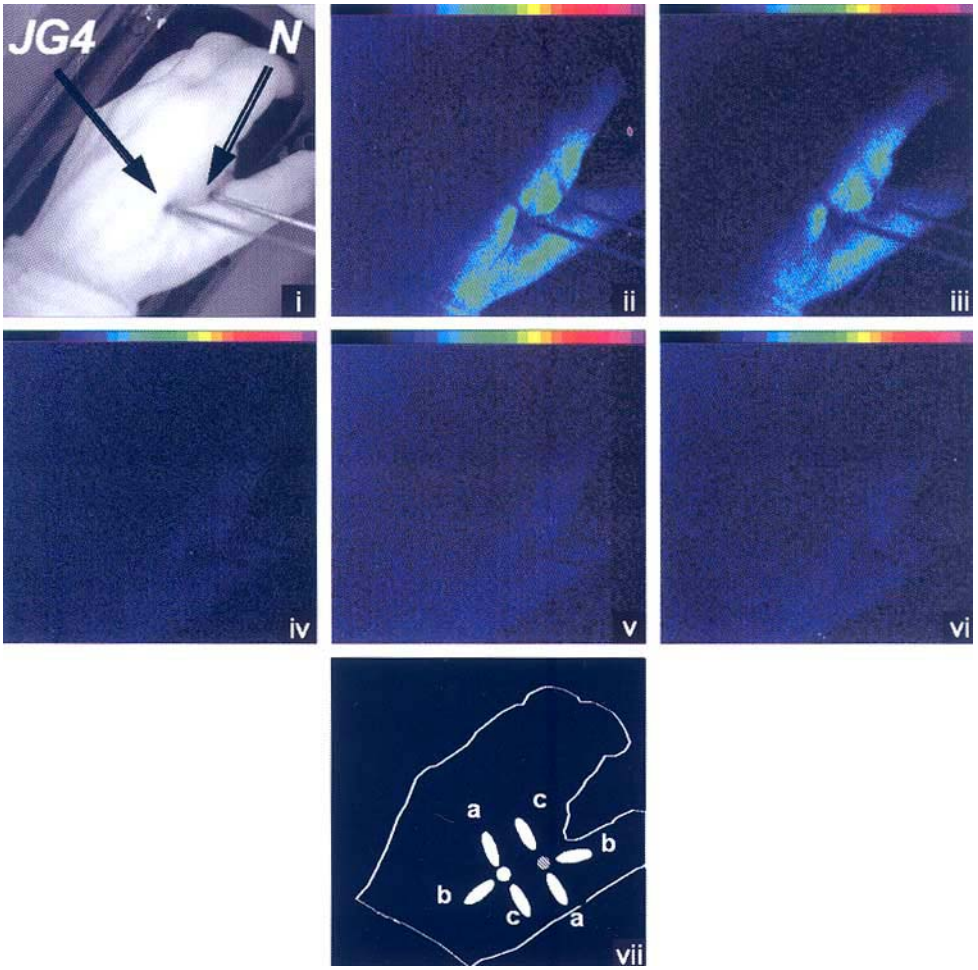

Fig. 2—Applied luminophores (compounds enhancing chemiluminescence and fluorescence). i: arrangement of electrodes on the left hand, ii: electrostimulation in JG4, iii: electrostimulation in N, iv-vi: luminescence decay after stimulation, vii: arrangement of luminophores on the left hand skin (a=riboflavin, b=lucigenin, c=luminol)

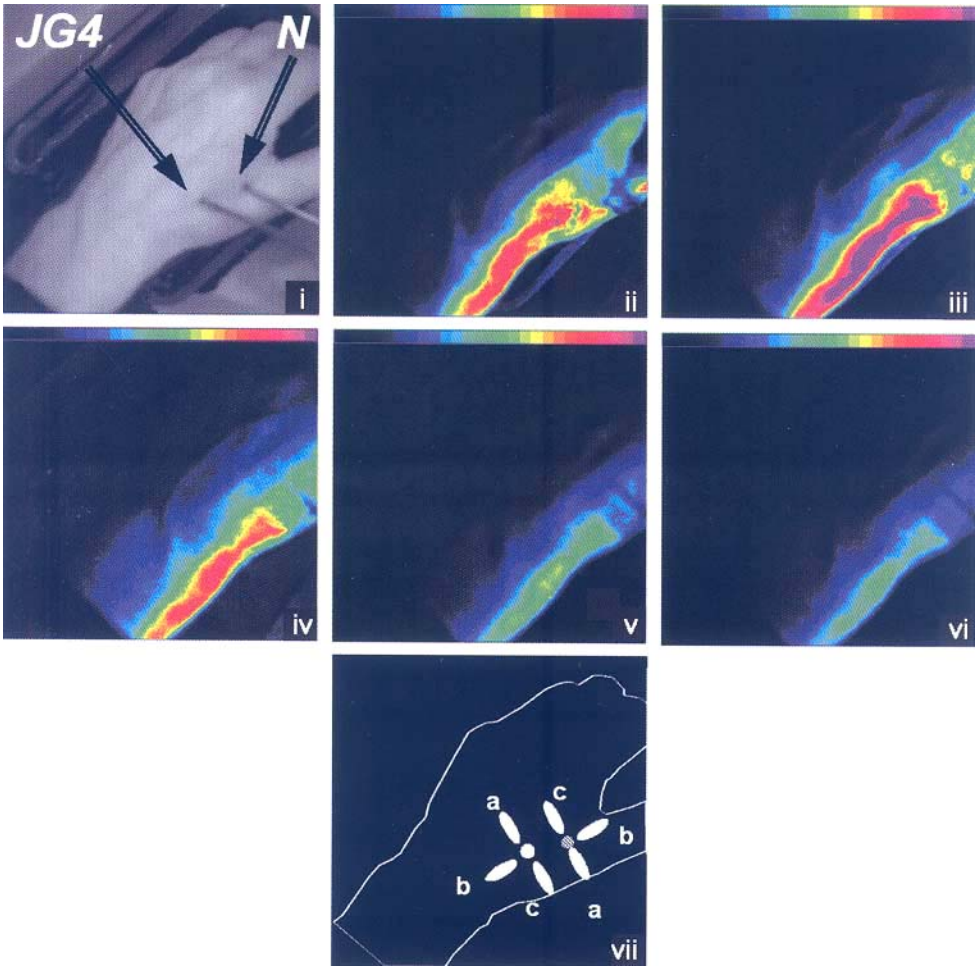

Fig. 3—Applied luminophores (compounds enhancing chemiluminescence and fluorescence). i: arrangement of electrodes on the left hand, ii: electrostimulation in JG4, iii: electrostimulation in N (neutral acupuncture area), iv-vi: luminescence decay after activation, vii: arrangement of luminophores (a=umbelliferone, b=9,10-diphenylanthracene hexane solution, c=9,10-diphenylanthracene oil solution)

neighbouring non active areas (2744 photons/mm^{2}s). It looks as if light-producing processes are channeled within acupuncture active points and less light is produced or transmitted outside. There is also a difference in the time decay of photon emission.

This study is a preliminary one and we did not find other works dealing with the use of luminescent enhancers- molecular probes for studying EAP. In this particular case many detailed problems regarding the preparation of molecular probes solutions, their introduction on or into the skin, time of experiment etc have to be studied.

Further experiments need to be performed with a close cooperation with dermatologists and experienced acupuncture specialists using other locations of acupuncture active and neutral points.

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