
ANTINOCICEPTIVE EFFECTS OF WHITE AND COLOR POLARIZED LIGHT IN ANIMAL FORMALIN TEST MODEL

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Introduction

Recently, incoherent P light application has become widespread in pain therapy. There are, practically, no experimental studies on animals that could provide an objective, quantitative assessment of the low-intensity P light antinociceptive effect on APs. It is well known that all animals have APs connected by meridians, whose topography, structure, and functional properties coincide with similar systems in humans (Schoen, 2001). Acupuncture is an officially recognized method of medical treatment in humans and animals. Needles, cold, heat, electrical current, laser, and other therapies may activate APs. Our recent researches suggest that low-intensity microwaves can also effectively stimulate APs (Limansky *et al.*, 1999). In the present work, we have demonstrated that exposure of APs or painful area to low-intensity P light evokes a statistically significant reduction of pain. Analgesia depends on exposure duration, action area, and P light wave-length (color).

Methods

Experiments were carried out on outbreed, adult male, albino mice. Tonic pain area was produced by subcutaneous injection of 5% formalin solution (dissolved in 0.9% NaCl solution) in dorsal surface of left hind foot (10 μ l/10 g of mass). Licking of the injected foot is a typical behavioral response (BR) to pain. Just after formalin injection mouse was exposed to P light. The control group received imitation of light application. A Bioptron Compact (Bioptron AG, Switzerland) was used as a light source. The power density of this device is 40 mW/cm²; light energy per minute is 2.4 J/cm², wave band 480-3400 nm, polarization up to 95%. We studied the dependence of the white P light effects on the duration of exposure (2, 6, and 10 min) and the zone (APs: E-36, V-56, V-60; zone without APs). Six color filters (Switzerland) permitted us to receive one of six color lights: red, orange, yellow, green, blue, and violet. Without color filters we had white light. Each color light was tested on two groups of mice: in one group the light was directed to AP E-36, in the other — to the painful area. The duration of pain BR for 60 min was calculated in control (n = 20) and experimental (n = 10) groups. The significant differences between the groups were determined with one-way ANOVA followed by a Student's t test. $P < 0.05$ was considered significant.

Skin surface temperature was measured in two groups of mice. AP E-36 was exposed to red or green P light for 10 min. The temperature was monitored every 60 s by a low — precision electronic thermometer.

Results and discussion

Influence of white polarized light on formalin-induced pain behavior in mice

Formalin-induced nociceptive response. Subcutaneous formalin solution injection into the left hind foot produced significant changes in mouse behavior (painful

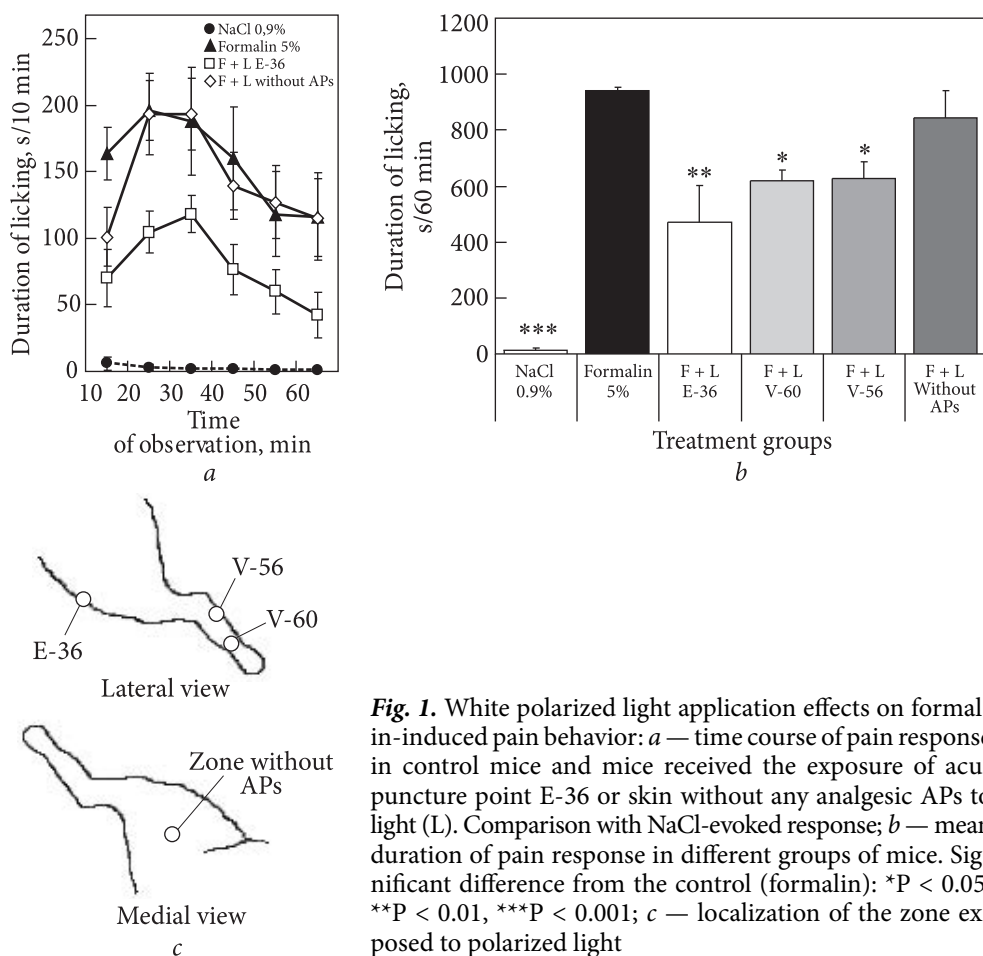


Fig. 1. White polarized light application effects on formalin-induced pain behavior: *a* — time course of pain response in control mice and mice received the exposure of acupuncture point E-36 or skin without any analgesic APs to light (L). Comparison with NaCl-evoked response; *b* — mean duration of pain response in different groups of mice. Significant difference from the control (formalin): * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$; *c* — localization of the zone exposed to polarized light

area licking) compared with the control injection of NaCl (Fig. 1). The formalin test was as a model of tonic pain in 1977 (Dubuisson, Dennis, 1977), and since then it has been used extensively in rats and mice. As during the first 10 min after formalin injection our animal movements were restricted by small plastic cages, we dealt only with the second tonic phase of formalin evoked pain response. Time course for this response is shown in Fig. 1A. The total licking time for 60 min in control mice was 942.1 ± 130.9 s.

Dependence of P light effects on the exposure duration. We tested 2 min, 6 min, and 10 min exposure of AP E-36 to P light. Mean values for the duration of licking the painful area differed significantly from the control values for 6 min ($t = 2.13$, $P < 0.05$) and 10 min ($t = 3.45$, $P < 0.01$) exposures to P light but not for 2 min exposure ($t = 0.49$). The most effective was 10 min exposure and it was used in all subsequent experiments.

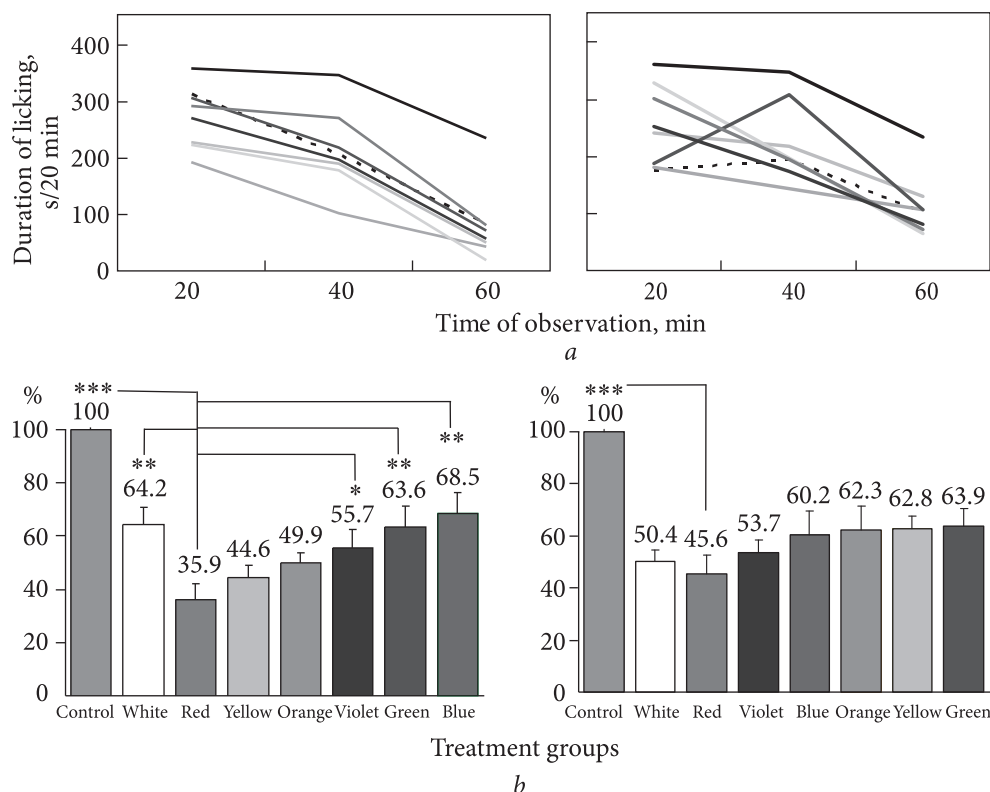


Fig. 2. Dependence of the formalin-induced pain behavioral response on the exposure of painful area or acupoint E-36 (AP E-36) to polarized (P) light of different colors: white, red, yellow, orange, violet, green, and blue: *a* — dynamics of pain responses in control mice and mice with the exposure to P light; *b* — mean duration of pain response in different mice groups. Significant difference from the red light group: * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$

Dependence of P light effects on the area of action. Formalin-induced pain was studied using mice exposed to P light on the three APs: E-36, V-60 and V-56. Analgesia made up 50% (E-36), 34.4% (V-60) and 33.2% (V-56), respectively. The same exposure of the skin without analgesic APs did not produce significant changes in pain response (Limansky *et al.*, 2006). AP E-36 appeared to be the most effective (Fig. 1). Our data show that the effect of P light does not depend on whether the AP E-36 is located on the same extremity as the painful area or contralateral to the painful area. Simultaneous illumination of both APs did not increase the antinociceptive effect. It is known that AP E-36 is one of the most effective pain treatment points in Chinese medicine. The injection of apitoxin into AP E-36 of rats with chronic arthritis evoked antinociceptive and antiinflammatory effects (Kwon *et al.*, 2001).

Effects of color polarized light on formalin-induced pain behavior

All tested colors evoked significant analgesic effects in either case when P light was applied to AP E-36 or to painful area (Tamarova *et al.*, 2009). Analgesia was 31.5-64.1 and 36.1-54.4%, respectively. The red light was the most effective to relieve pain. Analgesia from red light was 64.1% (painful area) and 54.4% (AP E-36). The effect from other types of color light was weaker. Exposure of the painful area to yellow, orange, violet, green or blue P light evokes analgesia up to 55.4%, 50.1%, 44.3%, 36.4% and 31.5%, respectively. Exposure of the AP E-36 to violet, blue, orange, yellow or green P light decreases the pain BR to 46.3, 39.8, 37.7, 37.2 and 36.1%, respectively (Fig. 2). As time and power of P light in all experiments were identical, it comes out that analgesic effect depends on the color of P light, in other words, on the wavelength. It is known that the depths of red wavelength penetration through the skin is deeper than those for yellow, green and blue colors (Ariane, Stuart, 1998; Jeffrey, 2005).

Mechanisms of low-intensity P light analgesic action are still poorly studied. There are evidences that antinociceptive effect of low-intensity laser on APs is produced via the opioidergic systems (Zhu *et al.*, 1990; Milchev *et al.*, 1992; Sing, Yang, 1997) and is accompanied by ATP and serotonin increasing release followed by reduction of the inflammatory process (Branco, Naeser, 1999).

Analgesia is not caused by thermal effect of P light

It is known that skin heating up to 43 °C is necessary to evoke an effect classified as thermopuncture (Chiba *et al.*, 1997). Our experiments have shown that after 10-min exposure of the AP E-36 to P light (2.4 J/cm²) the skin temperature rises from 35.3 °C (before light exposure) up to 37.4 °C (red light) or to 37 °C (green light). There are evidences that power density of light above 150 mW/cm² may give artifacts due to the effect of hyperthermia (Svaasand, 1985). We used much weaker P light (40 mW/cm²). Analgesic effect achieved by low-intensive P light application to formalin-evoked pain was not of thermal origin.

Conclusions

Exposure of painful area and AP to low-intensity white and color P light was followed by remarkable depression of formalin-induced pain response which was almost equal to that obtained with the sedative method of classical acupuncture, which requires prolonged exposure to needle (Han *et al.*, 1999). We found that the level of analgesia depends on the duration of exposure to P light, on the zone of exposure and on the color of light/wavelength. Color light therapy may be used as an effective noninvasive method to treat pain, and red light is the best wavelength selection to induce analgesia in clinical applications.

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