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EVIDENCE THAT TICK ATTACHMENT TO THE HUMAN BODY ISN'T RANDOM

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ABSTRACT. An assumption that the attachment of tick to the human body is both non-random and painless was based on correlations discovered between different factors and the structure of the tick gnathosome receptor. Pearson correlation between data on tick attachment points (I) and data on confirmed cases of tick-borne encephalitis in the Russian Far East (1956–1985) after the single «bite» at the same points was 0.953. Correlation between I and data of number of biological active points (BAP) on the body, known as a painless for needle therapy was 0.997. Resistance of the points where *Ixodes persulcatus* attempted to attach (80–150 kΩ) was previously measured by the use of an Acupuncture Analyser (Fyodorova and Kosenko 2000). To verify the correctness of hypothesis that tick attachment points are not random and that tick «bite» is painless an experiment was performed with pathogen free hungry *Ixodes ricinus* females (F2, total number of tick 26). The torso of author, naked to the waist, was used to track, their length (measured by curvimeter) and speed of tick movement. The mean impedance value of the probing points was 172±3.2 kΩ. The female (track 12 cm, speed 12 cm/min) began to attach at a BAP (acupuncture point VB – 24 on the Thai-Vang meridian) with impedance 170 kΩ. This «bite» was painless. Only careful observation allowed the tick mouthparts to be seen plunging inside the skin. Long and speedy search of attachment points, many probing (max. 7) as well as absence of pain during attachment at one of BAP permitted the conclusion that the initial assumption was correct.

KEY WORDS: *Ixodes* tick, attachment point, human skin, indolence

INTRODUCTION

It is not clear why it often takes a long time before an ixodid tick finally gets attached to a certain spot of the human skin. Nor do we know properly why people usually do not feel the moment of tick attachment.

According to Ribeiro and Francischetti (2003), there are three main protective barriers against blood-suckers: hemostasis, inflammation and immunity. As Ixodid ticks take prolonged blood meals, it is essential that their attachment should be painless at least at the outset of an attachment to the tender human skin.

These authors emphasize that the tick saliva, which contains histamine- and serotonine-binding proteins, as well as kininase, which destroys bradikinine and most probably some other active components that seem to arise together with inflammation, suppresses the pain of a «bite». Their appearance, however, is not an instantaneous reaction. It comes as a result of the skin being cut by the chelicerae, and the insertion of the hypostome.

In principle, cutting the skin should be a painful process. So, though we are often able to feel a tick wandering over our body, why do we usually fail to notice the cheliceral blades at work? We know by experience that sometimes even a mosquito that is sucking our blood causes no pain whatever. This only means that, probably by pure chance, the mosquito must have inserted its needle-shaped laciniae in a spot on the skin that is free from pain receptors. Pain receptors on our dermis average 200 per one square cm (=100 mm^2) (Panov 1973). Thus, if they are evenly distributed, there is 1 receptor per 0.5 mm^2. The actual pain receptors are much smaller. The area covered a single of them near the skin surface is equal either 1.0 or 7.5 nm^2, so the total maximum area, which might be covered by them, is 1500 nm^2 or 1.5 mm^2 per cm^2 of skin surface. Hence the area between four neighboring receptors averages (100–1.5)/200/4 = 0.12 mm^2 (Osetrov, pers. comm.). Nobody knows perfectly how receptors are distributed on the different areas of the skin but if the maximum distance between two receptors is no more than 0.248 mm it seems impossible for a tick to attack not causing a pain.

We immediately feel a horse-fly cutting our skin by its mouthparts, which have large and rough blades. Mosquitoes, which also feed rapidly, can occasionally bite in-between pain receptors, but a tick must search for such places without fail. This would seem impossible, though, because the female hypostome of *Ixodes persulcatus* Schulze, 1930 can be as wide as 0.252 mm (Filippova 1985), whereas the male hypostome is even broader. It is noteworthy that males of this tick species are able to transmit tick-borne encephalitis (TBE) virus by getting attached to the human skin for some minutes, cutting it with their mouthparts to suck in very small amount of blood (Alekseev 1993). Ticks never become attached immediately after gaining access to a body. Both their negative geotaxis and an attractive smell of the vertebrate host stimulate rapid movement up the legs and then the body. Their main «prey» consists of ruminants and other relatively big quadrupeds. Ticks searching for the location of a potential source of blood frequently move not only up but also down man's skin, apparently looking for an appropriate place for attachment.

Fyodorova and Kosenko (2000 a, b) argue that locomotion over the human body of *I. persulcatus* and *Ixodes ricinus* (Linnaeus, 1758) followed specific lines termed meridians. According to these authors data ticks attached themselves only at specific biologically active points (BAP), where the values of ohmic resistance measured by a device called Power Channel Condition Analyzer were not less than 80 but not more than 150 kΩ. We have firstly tested the hypothesis that painless tick «bite» is caused by attaching them mainly (or exclusively) to BAP points of the skin by comparing the known spots of *Ixodes* tick attachment both with TBE morbidity data collected between 1956 and 1985 in Russian Far East (Okulova et al. 1989) and with the number and location of the points used in traditional Chinese acupuncture. The points used in needle therapy are well known to be painless (Chen Jing 1988; Luvsan 1991). The numbers of BAP along the meridians were calculated by us according to data in the above-cited authors' illustrations.

Second, to verify the correctness of hypothesis that tick attachment localities are not random and that its «bite» is painless an experiment was performed.

MATERIALS AND METHODS

Ticks

Pathogen free hungry *I. ricinus* females (F2) from the laboratory culture were used. Adults moulted in July 2005 from nymphs fed on white mice were kept in tubes with controlled humidity (Alekseev and Dubinina 1996) at 22.0±1.5° C and 20h light photoperiod.

Test object and experimental conditions

The torso of author naked to the waist was used to track tick locomotion. Places of stopping for probing (gnathosome bending) and attachment were marked and then the impedance was checked by the special device, analogue of Power Channel Condition Analyzer used by Fyodorova and Kosenko (2000 a or b) to demonstrate tick attachment at BAP («Acupuncture Analyser», made on plant «Spectr» in St. Petersburg, Russia. Patent 2158535, November 10 2000) The measurements to determine the BAP were taken using a very weak electric current (less then $1.0 \cdot 10^{-6}$ amp.) through the human body. The first electrode, a massive one was fixed to the subject's ankle. The second electrode, a thin copper wire, was inserted inside a fine capillary with a water-soaked cotton stopper at the end. This electrode was used for contacting the different points on the skin. Such a device permitted estimation of resistance on the display of device. The thin electrode was placed in contact with the skin where tick begins the process of attachment. The diameter of its end was comparable in size with the end of the tick gnathosome. In our study, when points of probing were on the right side of torso the second thin electrode was in the contact with the skin whereas first massive electrode was in the left fist and vice versa. The tick's track was plotted on a human body chart by the observer (second author). Later the length of the track was measured directly on the torso using curvimeter. The time of tick crawling was fixed at 25–30 min., allowing calculation of the speed.

All experiments were performed in the laboratory during 3 days (February 28, March 1–2 2006) at 20.0±1° C, RH 40–42% and light 400 lux.

RESULTS

We have firstly tested the hypothesis that painless tick «bite» occurs mainly to BAP points of the skin by comparing *Ixodes* tick attachment spots both with TBE morbidity data collected between 1956 and 1985 in Russian Far East (Okulova et al. 1989) and with the number and location of the points used in traditional Chinese acupuncture. The numbers of BAP along the meridians were calculated by us according to data in the above-cited authors' illustrations. As can be seen from Table 1, there is a very significant Pearson correlation between the parameters mentioned above.

In our experiment, tick placed on the right side of the torso (10 cm above haunch bone) either fell off (8 of 26) or began to crawl (Table 2). Some of them were slowly «travelled» only down and among 4 of these only one stopped to probe. At this point one of the minimal impedance values (37 kΩ) was detected. Most of the specimens (13) were active. They crawled up and down. Most of their tracks were on the upper parts of the torso; some stopped to probe on the neck, armpits, back and breasts, but no one went down to the waist (Fig. 1). The maximal length of track was more than 4.5 m and mean speed was 10 cm/min. Some of specimens probed 4 and even 7 times but no one of them attached. Impedance values varied from 30 to 452 kΩ. It is important to stress that mean impedance value of

Table 1.
Distribution of *Ixodes persulcatus* tick attachment points, biologically active points (BAP) and tick locations in tick-borne encephalitis patients

Regions		Attachment (%)* I	Number of BAP II	TBE cases (%)** III
Head or neck		55.0	302	39.2
Breast or back		18.6	91	16.7
Upper limbs or armpits		13.7	71	22.3
Lower body part or groin		8.8	61	15.6
No of checked cases		304	–	536
r, (Pearson)	I–II	0.997		
	I–III	0.953		

* Values of the attachment rates are given according to Fyodorova and Kosenko (2000a), data collected in the Veliky Novgorod Region, northwestern Russia between 1970–1999**. Data on TBE morbidity are given according to Okulova *et al*. (1989), who studied the casebooks of the patients with confirmed encephalitis, each after the «bite» of a single tick in the Primorye Region, Russian Far East between 1956 and 1985. In both studies, the percentages do not total 100.0, because there are no data on tick attachment to the lower limbs in the work of Fyodorova and Kosenko (2000a). BAP number was calculated from the sketches presented by (Chen Jing, 1988) and Luvsan (1991); BAP on the feet and hands are not included in the counts, because tick attachment on them either has never been checked or does not occur.

Table 2.
Behaviour of *Ixodes ricinus* females on the human body

Type of locomotion and No.of ticks*	Length of «travel» (cm)			Speed (cm/min)			No of stops (probing)			Impedance values (kΩ)		
	min	max	mean****	min	max	mean****	min	max	mean****	min	max	mean****
Refuse and fall (8)**	–	–	–	–	–	–	–	–	–	–	–	–
Only down (4)	24	32	29.2±1.8	1.6	6.0	3.2±0.98	0	1	1	–	37	37
Up and down (13)	104	460	244.7±29.9	5.2	18.4	10.3±0.6	1	7	3±0.4	30	452	172±3.2
Only up (1)***	–	12	12	–	12	12	–	1	1	–	170	170

* total no. tested – 26; ** in brackets no. of ticks showing of this type of behaviour; *** attaching during 15 min; **** mean with standard errors

thirty probes was 172.0±3.2 kΩ. One specimen, which moved only up quickly enough (12 cm/min) stopped very soon and began to attach at a BAP with impedance almost equal to the mean, 170 kΩ. The point of attachment practically coincided with acupuncture point VB — 24 on the Thai-Vang meridian (Chen Jing 1988). During 15 min the tick's mouthparts were inserted in the skin to the one third of their length, and then the tick was removed using miniature hook-forceps. A small achromatous papule with a miniature hole in the center was seen at the point of detachment. The tick «bite» was absolutely painless and only careful observation allowed the tick mouthparts to be seen plunging inside the skin.

DISCUSSSION

A very significant Pearson correlation was observed between data on tick attachment points and BAP (Fyodorova and Kosenko 2000) and between attachment points reported by Fyodova and Kosenko (2000) and attachment points of ticks to patients who developed tick-borne encephalitis (Okulova et al (1989). This strongly suggests that the processes of tick «bite» and attachment are not only non-random but they are definitely goal-seeking.

The preponderance of *Ixodes* tick attachments to the head or neck, which triggers the disease in children and teenagers [84.9 and 62.5% of cases, respectively (Okulova et al. 1989)], as well as the overall pattern of distribution of infective «bites» are most probably not accidental. The same holds true for the prevalence of tick attachment (Fig. 2) to young soldiers, usually 18–20 years old, who suffered from *Dermacentor nutalli* Olenev ticks. This species is known as vector of Omsk hemorrhagic fever in western Siberia, Russia. Military doctor

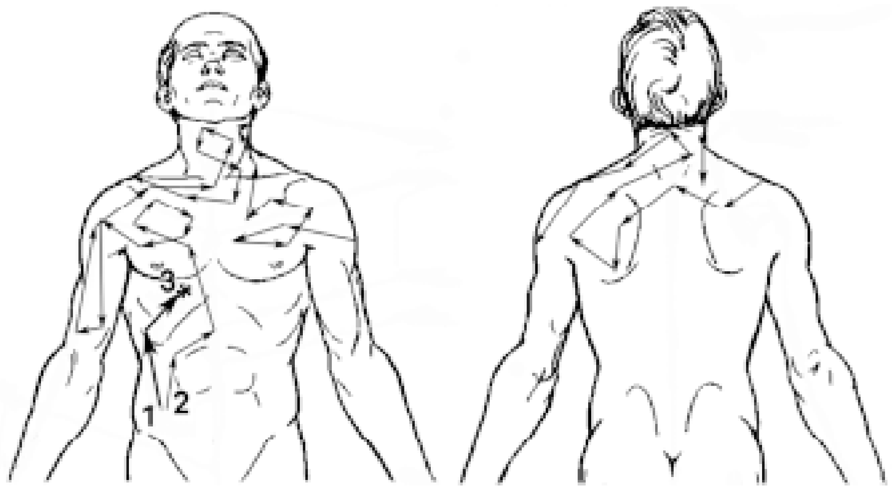

Fig. 1. Example of tracks of *Ixodes ricinus* movement on the human torso: 1 — only up; 2 — up and down; 3 — point of attachment.

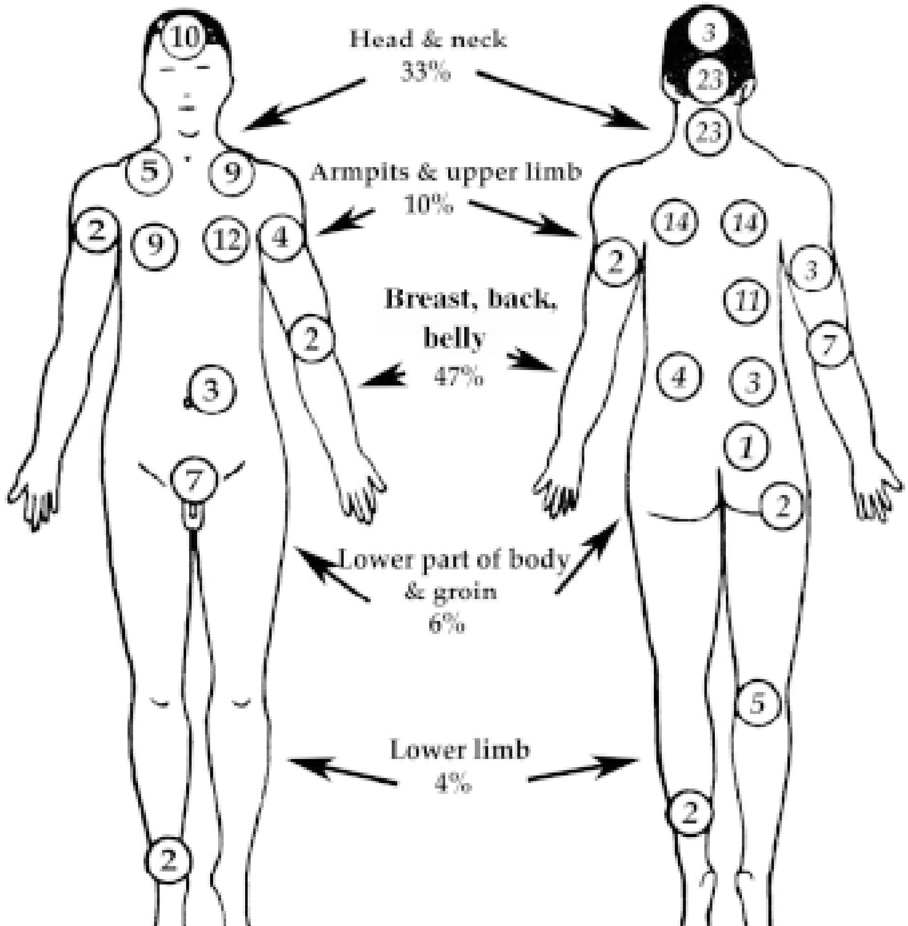

Fig. 2. Places of *Dermacentor nutalli* Olenev, 1929 females attachment to the human body (after Pavlovsky (1948), with changes).

Savul'kin (cited by Pavlovsky 1948) obtained these data while surveying young Red Army soldiers in the mid-1940s. Some peculiarities of *D. nutalli* distribution might well be associated with the special military clothes involved. Nevertheless, the differences are not very considerable and the correlation between the attachment spots and BAP is also positive, though less strong (r = 0.405). This means that *Dermacentor* attachment to the human body is also non-random, serving as one of the basic arguments that both vertebrate host and tick function as a complicated system.

In our experiment, we did not confirm the contention of Fyodorova and Kosenko (2000 a, b) that ticks crawl along the specific lines on the human body termed meridians. In our experiments they tracked up, down and around the torso, hands and neck but nevertheless they stopped and probed (bending their gnathosome) on different points of the body, however most of them correlated with full stops on the map of BAP. We observed that the single point of attachment had an impedance value (170 kΩ) just inside the interval of average impedance value (172 ± 3.2). This one female was lucky enough to detect it within the first minute of search on the lateral meridian of the body (point VB — 24 on the Thai-Vang meridian). As expected the attachment was absolutely painless despite of insertion of one third of hypostome in the skin. This process was observed for 15 minutes after which tick was carefully detached by miniature hook-forceps. The small achromatous papule with a miniature hole in the center disappeared on the third day of observation. The difference in intervals of impedance values revealed by us during observations of processes of probing and those described by Fyodorova and Kosenko (80–150 kΩ) can be explained by differences between the ticks used. The above-mentioned authors used specimens collected in nature at the very beginning of their seasonal activity whereas we used ticks developed in the laboratory in conditions, which did not replicate the natural ones and began to perform our experiments in winter.

According to the authors' own experience, as *Ixodes* ticks quest over the skin they repeatedly stop and probe for an appropriate place to begin the attachment process. During probing the gnathosome is bent down for the palpi to touch the surface of the skin. Similar behaviour is typical of Ixodidae when they are on a ruminant (Balashov et al. 1976). At the moment of probing, the palpi are still gathered up. It is very hard to measure exactly the distance between the centres of the palpal organs, which are situated on the fourth segment of the palp, but even very rough estimates show it to be 0.12±0.025 mm. Since the female hypostome of *I. persulcatus* can be as wide as 0.252 mm (Filippova 1985), not all of the BAP are available for the penetration of the tick's cheliceral blades inside the dermis but only some of them, in which the level of pain is low and at which (most probably) pain receptors are distributed unevenly.

Attachment is long known to depend on the results of palpal organ probing. This organ contains several types of receptor. In a crude experiment, El-

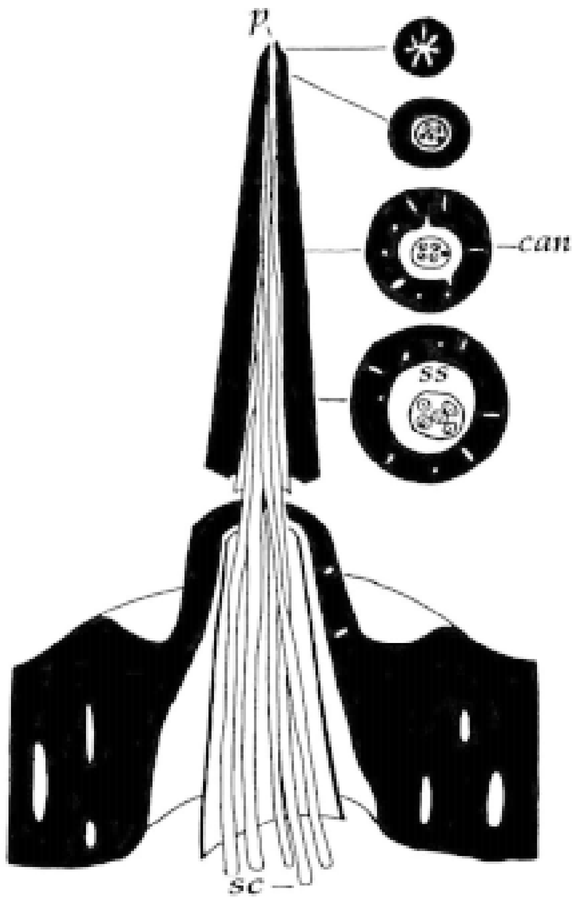

Fig. 3. Single pore — single walled (SP-SW) sensilla of the palpal organ of *Argas persicus* (Oken, 1818) [Courtesy of Leonovich (2005), with changes]. *p* — pore, *can* — channel, *ss* — scolopoid scheath; *sc* — sensor cilia.

Ziady (1958) demonstrated that ticks whose palpi had been removed were incapable of searching for suitable points of attachment. According to Balashov et al. (1976), two types of sensilla, *A* and *B*, are present on each palpal organ. The former were shown to react to certain chemical substances, and they also function as mechanoreceptors. Sensilla *B*, the so-called single (or upper) pore — single-walled (SP-SW), shows no reaction to any gustatory stimulus. Balashov et al. (1976) failed to demonstrate that SP-SW sensilla (Fig. 3) are mechanoreceptors and found difficulty in understanding of their function. Nevertheless Leonovich (2005) affirmed that the above authors actually meant that type *B* sensilla are responsible for the initiation of attachment. Taking into account all available evidence, it could be suggested that voltage and ohmic resistance are measured during probing, by a pair of «electrodes», probably the sensilla of type *B*, or SP-SW, placed on the pair of palpal organs. Such a hypothesis does not seem to be too controversial, because the sensitivity of arthropods to magnetic fields and other physical forces, to which a human organism is insensitive, is well documented. BAP have been used for acupuncture for many centuries but the tips of the needles used in acupuncture are thinner than a tick hypostome. So either the number of pain receptors per BAP is reduced or they are distributed less regularly within BAP than over the other parts of the human skin, or both. Most probably, the pain sensitivity thresholds of BAP differ between people and can even change over time in one and the same person. Then it becomes clear why ticks search for BAP, repeatedly probing and measuring the ohmic resistance by their palpal organs. This also explains why they may attach themselves differently to one and the same person on different days. Since suitable values of resistance, between 80 and 150 kΩ, can be present simultaneously in various BAP, the attachment of several ticks to one host is unsurprising.

In conclusion, an analysis of, and a clear-cut congruence between, the distribution of the places of tick attachment, the BAP distribution over the human body, the TBE infective «bites» and direct experiment showed dependence on the spots of tick bite, tick mouthparts size and conformation coupled with special sensilla. The latter are apparently responsible for the beginning of an attachment by measuring BAP ohmic resistance.

All this clearly shows that tick attachment to the human skin is not a random but a well-directed effort.

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