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Protective efficacy of permethrin-treated trousers against tick infestation in forestry workers

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Abstract

Introduction and objective. Prevention of tick borne diseases in forestry workers is essentially based on the use of appropriate clothing. The objective of this pragmatic, randomized, controlled trial was to assess the potential benefit of permethrin-treated working trousers for the prevention of tick infestation during forestry work.

Materials and methods. N=164 male forestry workers were equipped for a period of 16 weeks with permethrin-treated (intervention group – I) or untreated work trousers (control group – II). Subgroups, according to the use of trousers with (I-1, II-1) or without cut protection lining (I-2, II-2) were constituted. Tick infestation (quantity of ticks on the body surface) was assessed by questionnaire after 16 workdays. Control and intervention groups were compared by calculating the infestation rate (percentage of subjects with ticks) and the average number of ticks per workday.

Results. The infestation rate was significantly lower in the intervention group than in the control group (36.6 vs. 63.4%, p=0.001; Fisher-test). Further analysis revealed a significant reduction of tick infestation by permethrin treatment only for subjects wearing trousers without the cut protection lining (I-2: 34.2 vs. II-2: 80.0%, p<0.001), while users of cut protection trousers did not benefit from such treated trousers (I-1: 38.6 vs. II-1: 47.6%, n.s.). Similar results were found for comparisons based on the average number of ticks per workday.

Conclusions. The use of permethrin-treated trousers does not completely prevent tick infestations. Improvement of tick protection has been shown only for some applications, but not in general. Additional prevention measures are therefore still indispensable.

Key words

Forestry worker, protective clothing, tick-borne diseases, borreliosis, permethrin, randomized control trial

INTRODUCTION

Due to their job, forestry and other outdoor workers are at risk of contracting tick-borne diseases, manifesting as, e.g. increased seroprevalence rates for tick-borne infections in respective workers [1, 2, 3, 4]. Personal protective measures, aimed at the prevention of tick infestation in general, are an essential line of defence to particularly avoid infections like borreliosis which, to date, cannot be prevented by vaccination [5].

In Germany, wearing long trousers and long-sleeved shirts, and regular application of repellents to exposed skin and/or clothing, are officially recommended as personal protection measures [6] and turned out to be effective in reducing the risk of borreliosis [7]. Tucking trousers into socks or boots to avoid unprotected skin areas and checking the body for ticks after outdoor activities to prevent a potential transfer of pathogens from attached ticks by early removal, are further recommendations [4, 8, 9, 10].

The protective effect of clothing against insects and mites can be basically improved by treatment with permethrin, a synthetic insecticide and acaraicide. Early field studies using clothing sprayed with or dipped in permethrin showed up to 100% protection against several American tick species, compared to untreated clothing [11, 12, 13, 14, 16]. However, if permethrin is applied in this way, the

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impregnation shows only low resistance towards washing, resulting in quick loss of protective efficacy and the need for repeated retreatment [15, 16]. However, factory-based and long-lasting permethrin impregnations have been developed recently to overcome this disadvantage. Initially made for military purposes, these impregnations exhibit superior resistance towards permethrin losses due to laundering and wearing, superseding any retreatment by the user. Laboratory experiments showed adequate protective activity of respective battle-dresses against ticks, even after long-term use or up to 100 wash cycles [17, 18, 19]. As a result of these findings, protective clothing for forestry workers has been developed by different producers.

To the best of our knowledge, the promised gain in protective efficacy associated with this clothing has not yet been evaluated in daily work routine. A respective evaluation, however, seems particularly necessary in view of the fact that wearing treated clothing usually entails a risk of permethrin absorption by the user [20, 21]. Although generally considered as a pesticide with low acute mammalian toxicity, the use of permethrin might raise some toxicological concerns since potential adverse health effects after long-term low dose exposure, including, for example, a mild carcinogenic potential, are still under debate [22, 23].

Objectives. A pragmatic, randomized, controlled trial was carried out to assess the potential effects on tick protection and the uptake of permethrin associated with the use of clothing treated with a long-lasting permethrin impregnation.

Results of a comparative survey on tick infestation in forestry workers wearing either trousers treated with

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permethrin or untreated trousers under regular working conditions are presented here. Trousers for different fields of application with respect to cut protection properties and two different producers were used, thus enabling a comparison of different groups of employees.

MATERIALS AND METHOD

Study design. A pragmatic, randomized, controlled trial following a parallel group design was conducted to assess tick infestation in forestry workers wearing either permethrin treated (intervention group – I) or untreated work trousers (control group – II). Data was collected in different regions of Germany during the tick seasons 2010 and 2011.

Due to differing job functions, subgroups wearing trousers with (subgroups I-1 and II-1) or without cut protection lining (subgroups I-2 and II-2) were formed. Further grouping was undertaken with respect to the supplier of the studied trousers.

The randomization procedure was restricted in two ways:

- The allocation into subgroups concerning cut protection properties was determined by job function of the particular participant.
- 2. Since the different study regions were likely to differ in the occurrence of ticks and the starting point of the study period differed in the particular regions, the participants were allocated into intervention and control group in an allocation ratio of 1:1 within each region, aiming at an even local distribution of different types of trousers.

Within the subgroups, trousers from different producers were allocated randomly. Subjects were aware of their group affiliation since the permethrin treated trousers were respectively labeled (Tab. 1).

Clothing. Factory-based permethrin treated trousers with or without cut protection lining were obtained from two

different producers (designated 'A' or 'B'). According to the suppliers' specifications, the initial permethrin content in all types of treated trousers was between $1,250 - 1,500 \text{ mg/m}^2$. A protective efficacy of the fabric for at least 50 launderings was stated.

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Control subjects wearing trousers with cut protection (group II-1) were equipped with untreated trousers, being equivalent to the permethrin treated trousers with respect to fashion and fabric. Control subjects wearing trousers without cut protection (group II-2) were asked to continue wearing their previously used, untreated work clothing, since the equivalent untreated trousers were not obtainable from both suppliers.

Subject recruitment. Study participation was voluntary. The recruitment was supported by several regional public forest administrations, the Federal German Forestry Administration and the responsible public sector accident insurers. Basic information about the study was disseminated to the employees via flyer. Persons interested in participating were asked to contact us and provide on a form personal data, data on job function, required type of trousers (with or without cut protection lining) and clothes size, as well as a self-evaluation of susceptibility towards ticks (rather high or rather low). Detailed written information about the study, a form for declaration of consent and a health questionnaire were sent to the interested parties. Subjects without medical contraindications were invited to visit a regional information event, which was held by staff of our institute. Visitors were informed once more, orally, about the contents of the study and had to undergo a short medical check-up by a physician. After completion of informed consent, the participants were issued their previously assigned study clothing (two identical trousers of specified type) and further study material (e.g. questionnaire forms and written instructions).

Data collection. The participants were asked to start wearing the study clothing on a Monday, as early as possible after

Table 1. Group classification and features of study subgroups (*participants in this group continued to wear their previously used and untreated trousers; n.n.: producer not specified)

Subgroup	Subgroup description	Permethrin treatment		Cut protection lining		Producer		
label		yes	no	yes	no	А	В	n.n.
I + II	total study group	х	x	х	х	x	х	х
1	intervention group	x		х	x	x	x	
I-1	intervention group, trousers with cut protection, producer A and B	х		х		х	х	
I-1A	intervention group, trousers with cut protection, only producer A	х		x		х		
I-1B	intervention group, trousers with cut protection, only producer B	x		х			x	
I-2	intervention group, trousers without cut protection, producer A and B	х			х	x	х	
I-2A	intervention group, trousers without cut protection, only producer A	х			x	х		
I-2B	intervention group, trousers without cut protection, only producer B	x			x		х	
	control group		x	x	x	x	x	x
II-1	control group, trousers with cut protection, producer A and B		х	х		х	х	
II-1A	control group, trousers with cut protection, only producer A		х	х		х		
II-1B	control group, trousers with cut protection, only producer B	-	x	х			х	-
II-2	control group, trousers without cut protection, producer not specified*		x		x			x



Figure 1. Data collection timeline

visiting the information event. Data collection was based on self-reports. The subjects were asked to fill in questionnaire forms on four consecutive workdays during four predefined weeks of a 16 week study period (Fig. 1).

The number and location (categorized in head/neck, upper body, arms/hands, upper legs/buttocks/groin, knees/lower legs/feet) of sucking or moving ticks (nymphal and adult) found on the skin after work were to be recorded daily, from Monday until Thursday, during each of the four survey weeks.

Participants were asked to send the completed questionnaires of each survey week via mail to the Institute of Occupational, Social, and Environmental Medicine, where conversion into an electronic format and analysis took place.

Statistical analysis. The average number of ticks on the body surface per workday (TN) and the infestation rate (IR) were used as dependent variables for subgroup comparisons. IR is the percentage of subjects reporting ticks on their body surface in a respective subgroup. TN was calculated individually for each subject by dividing the absolute number of ticks reported by the number of documented workdays. Group related means, maxima and standard deviations were calculated for TN. The calculation of relative measures was necessary since response rates and number of participants differed within the particular subgroups, and comparisons of absolute tick numbers would be prone to bias. Protective efficacies (expressed as percentage) were calculated by subtracting IRs (or TNs) for treated trousers from IRs (or TNs) for untreated trousers, and dividing these differences by the value for untreated trousers.

The non-parametric Mann-Whitney U test was used for statistical comparisons of TN, considering a skewed data distribution. Tick infestation rates and susceptibility to ticks in different subgroups were compared using Fisher's exact test. p < 0.05 was considered to indicate statistically significant differences between subgroups

Ethical aspects. The study protocol was approved by the Ethics Committee of the State Chamber of Physicians of Rhineland-Palatinate, Germany (Ref. No.: 837.497.09 (7003).

RESULTS

N=171 male participants were acquired for the presented study. 164 subjects (95.9%) provided data on tick infestation for at least one workday. 65.9% of the participants were employed as 'foresters', 20.1% as 'forest ranger/officer' (head of a particular forestry district), whereas 14.0% had other job functions (e.g. trainee, huntsman, tasks in plant nursing).

Table 2. Descriptive data of the study population and its subgroups

Subgroup label	n	Age [years] median (range)	Job functions [%]			Susceptibility to ticks [%]		
			forester	forest ranger	others	high	low	
I + II	164	44.5 (19–61)	65.9	20.1	14.0	62.9	37.1	
 	82	44.5 (19–61)	64.6	23.2	12.2	59.3	40.7	
I-1	44	42.5 (19–60)	90.9	2.3	6.8	50.0	50.0	
I-1A	22	42.5 (20–55)	95.5	0	4.5	52.4	47.6	
I-1B	22	42.5 (19–60)	86.4	4.5	9.1	47.8	52.2	
I-2	38	45.0 (24–61)	34.2	47.4	18.4	70.3	29.7	
I-2A	23	46.0 (24–61)	30.4	52.2	17.4	69.6	30.4	
I-2B	15	43.0 (30–56)	40.0	40.0	20.0	71.4	28.6	
	82	44.5 (19–58)	67.7	17.1	15.9	66.7	33.3	
II-1	42	43.0 (19–57)	97.6	0	2.4	58.5	41.5	
II-1A	23	43.0 (19–57)	95.7	0	4.3	59.1	40.9	
II-1B	19	42.0 (19–56)	100	0	0	57.9	42.1	
II-2	40	45.0 (20–58)	35.0	35.0	30	75.7	24.3	

Subjects wearing permethrin treated (group I) and untreated trousers (group II) were evenly distributed (n=82 in each group) among the respondents, and did not differ with respect to age and job function. Self-assessed tick susceptibility was the same in both groups with a majority of participants being 'rather highly susceptible' (p=0.794). Further details on the study population are given in Table 2.

Users of cut protection trousers were mainly (> 90%) foresters (I-1 and II-1), whereas trousers without cut protection were mostly used by forest rangers and others (I-2 and II-2). The distribution of job functions was similar in the intervention and control group for a particular type of trousers. Users of trousers without cut protection stated higher tick susceptibility (72.6 % rather highly susceptible in pooled analysis of group I-2 and II-2) than users of cut protection trousers (54.8% 'rather highly susceptible' in group I-1 and II-1, p=0.031). Tick susceptibility did not differ between the intervention and control groups for users of a particular type of trousers (p=0.515 for I-1 vs. II-1 and 0.794 for I-2 vs. II-2, respectively).

According to the study design, a maximum return of 2,736 questionnaires, each representing a respondent's tick infestation at the end of a particular workday, could be

Table 3. Tick infestation rates (IR) and average numbers of ticks per workday (TN) in subgroups wearing permethrin-treated and untreated trousers. (A and B: trousers from different producers; group II-2: respondents' own untreated trousers, producer not specified)

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Subgroups compared	l vs. ll	l-1 vs. ll-1	I-1A vs. II-1A	I-1B vs. II-1B	I-2 vs. II-2	I-2A vs. II-2	I-2B vs. II-2
intervention group (I)							
number of subjects (n)	82	44	22	22	38	23	15
number of considered work days	1126	593	289	304	533	301	232
number of reported ticks	127	45	31	14	82	54	28
IR (percentage of subjects with ticks)	36.6	38.6	45.6	31.8	34.2	34.8	33.3
TN – mean	0.13	0.10	0.14	0.06	0.16	0.17	0.14
TN– standard deviation	0.30	0.17	0.21	0.10	0.40	0.46	0.31
TN – maximum	1.85	0.64	0.64	0.40	1.85	1.85	1.17
control group (II)							
number of subjects (n)	82	42	23	19	40	40	40
number of considered work days	1024	464	268	196	560	560	560
number of reported ticks	520	112	59	53	408	408	408
IR (percentage of subjects with ticks)	63.4	47.6	52.2	42.1	80.0	80.0	80.0
TN – mean	0.44	0.22	0.21	0.24	0.67	0.67	0.67
TN – standard deviation	0.85	0.43	0.28	0.57	1.09	1.09	1.09
TN – maximum	5.31	2.38	0.81	2.38	5.31	5.31	5.31
inter-group comparison IR: p (Fisher-Test)	0.001	0.267	0.768	0.533	< 0.001	0.001	0.003
inter-group comparison TN:	< 0.001	0 237	0.420	0.405	< 0.001	< 0.001	0.003

expected. The effective response rate was of 78.6% (2,150 received questionnaires) with rates differing only slightly in the intervention (86.8%) and control groups (74.4%). Members of the intervention group (I) reported a total number of 127 ticks, based on data from 1,126 surveyed workdays. In the control group (II), 520 ticks were found in 1,024 considered workdays. Group-related results for the calculated parameters IR and TN are given in Table 3.

p (Mann-Whitney-U-Test)

Comparing all respondents, regardless of the particular type of trousers with respect to cut protection, statistically significantly differing (p=0.001) IRs of 36.6 and 63.4% were calculated for the intervention and control groups, respectively, yielding a protective efficacy of 42.2%. TN showed wide variation for both groups, resulting in large standard deviations. Inter-group differences were also statistically significant for this parameter (p< 0.001). Using the mean values of TN for respondents in the intervention (0.13) and control groups (0.44) for calculation, the protective efficacy was 70.5%.

Stratified analysis revealed considerable differences with respect to cut protection properties of the trousers. A moderate, statistically not significant, lowering of IR (from 47.6% to 36.6%) and TN (mean values 0.22 and 0.10) by permethrin treatment was found in users of cut protection trousers (I-1 vs. II-1), whereas the effect of permethrin treatment was more obvious in users of trousers without cut protection (I-2 vs. II-2). IR (mean TN) could be lowered from 80.0% (0.67) to 34.2% (0.16) in respective respondents (p<0.001, for both parameters). Calculated protective efficacies were 57.3% (IR) and 76.1% (TN), respectively. The effect of permethrin treatment as a function of the trousers' type is shown in Figure 2. This also illustrates marked differences within the control group (II-1 and II-2) with IR, and TN being



Figure 2. Frequency plot of average number of ticks per workday (TN) according to cut protection lining and permethrin treatment of pants used at work

significantly higher in control respondents wearing trousers without cut protection (II-2) than in subjects wearing trousers with cut protection lining (II-1) (IR: p= 0.003; TN: p<0.001).

Further analysis, stratified by the particular brand of permethrin treated trousers, yielded similar trends to the aforementioned pooled analysis. A statistically significant reduction of IR and TN was found only in respondents wearing trousers without cut protection lining, regardless of the producer of the trousers (I-2A vs. II-2 and I-2B vs. II-2, table 3)

Tick infestation was assessed with respect to particular body regions. Although the proportion of ticks found below the waistline was lower in the intervention group (45.7%,

Table 4. Locations of tick infestation in respondents wearing permethrintreated and untreated trousers

Total	100% (127)	100% (520)				
knees/lower legs/feet	27.6% (35)	waistline: 7.6% (35) 45.7% (58)		waistline: 60.6% (315)			
upper legs/buttocks/groin	18.1% (23)	below	26.2% (136)	below			
upper body	15.0% (19)	54.3% (69)	16.9% (88)	39.4% (205)			
arms/hands	34.6% (44)	waistline:	21.9% (114)	waistline:			
head/neck	4.7% (6)	above	0.6% (3)	above			
Body region	Intervent (gro	ion group oup I)	Control group (group II)				
	Percentage (number) of ticks found in a particular body region						

control group: 60.6%), participants still reported ticks in body regions covered by permethrin treated trousers (Tab. 4).

DISCUSSION

The protective efficacy of permethrin treated clothing against ticks has been evaluated previously in a number of field trials. Using pressurized sprays or dipping formulations for permethrin application, protective efficacies close to 100% were found immediately after treatment against several tick species [11, 12, 13, 14, 16]. A quick drop in the protection of these garments associated with use or washing [15, 16] was recently the starting point for the development of long-lasting factory-based permethrin impregnations. However, to date, only limited experience from field trials is available for this factory-treated clothing.

Faulde et al. found a protection rate of 95.5% when comparing infestation with *Ixodes ricinus* ticks, the most common ticks in Germany, on treated and untreated legs of trousers, each of them worn simultaneously by volunteers walking in known tick-infested habitats [24].

A reduction of 93% in the total incidence of tick bites was reported by Vaughn et al. [25] for a comparison of outdoor workers wearing permethrin treated clothing (including shirts, trousers, socks, hats, and boots), in addition to standard tick prevention measures (e.g. wearing long trousers and long sleeves, tucking trousers into boots or socks) with workers using only standard tick prevention measures.

In the latter study, tick bite frequencies were assessed under actual working conditions, whereas the aforementioned studies were based on more experimental settings with volunteers, as described, e.g. by Faulde et al [24].

Compared to reduction rates of > 90% reported in the literature, in the presented study, a lower protective efficacy (up to 76%) was found, depending on the particular subgroup in the study. Using clothing containing a nominal permethrin concentration ($\leq 1,250 \text{ mg/m}^2$), which is well comparable to other studies, the reasons for this deviation could be diverse. Apart from the survey by Vaughn et al. [25], the results obtained from more restrictive experimental settings are compared to results obtained under actual working conditions. In addition to differing tick exposure times, the assessment of tick infestation also differed between the studies. Tick counting was either performed solely on clothes [13, 16, 24], on clothes and skin [11, 12, 14], or only on the skin [25, this study]. It is probable that higher protection rates can be expected when referring to tick counts on the

surface clothing where direct contact between the tick and permethrin takes place, although in terms of prevention the number of ticks reaching the skin surface seems to be more informative. Apart from the study of Faulde et al. [24], a combination of permethrin-treated trousers and shirts was tested as protective clothing. In contrast, the participants in the current study were equipped with respective trousers only, taking poorer protection of the upper body into account. This was deliberate, since a consequence of the use of longsleeved shirts or jackets hardly represents actual working conditions, particularly in summer.

In addition, the respondents were asked not to change their general behaviour with respect to tick protection. Taping the cuffs or tucking them into the boots or socks was not requested, and treated shoes or socks to impede the tick's access to the lower legs were not provided, as in other studies (e.g. [25]). This might be a reason for lower protective efficacies and may also explain the considerable percentage of ticks on the legs of participants, even when wearing treated trousers. Taking into account the trouser leg ends as a portal of entry for ticks, also for treated trousers, following the rules of behavior mentioned above, an additional use of gaiters or an improvement of the trousers' design, e.g. by providing trouser-integrated gaiters, might further improve personal protection.

When comparing the control groups in the presented study, the risk of tick infestation was higher for respondents wearing trousers without cut protection. This is in line with self-assessment data gained from all participants before the start of the study, designating the subgroup of 'forest rangers/ officers' and 'others' as more vulnerable to tick infestation than 'foresters'. Differing job tasks, including tasks with a high risk of tick exposure, such as marking trees, maintenance of tree crops, or planting, in the former groups could be discussed as an explanation. However, this would be rather contradictory to the results of Cisak et al. [4], who found a higher risk of tick infestation for workers performing manual tasks (including particularly saw operators wearing cut protective trousers), compared to other forestry job categories.

An alternative explanation could be the use of inappropriate clothing which provided only poor protection against ticks by some members of the respective control group. This cannot be ruled out since the respondents in that particular group did not wear uniform (untreated) study trousers, due to a lack of availability at the suppliers. Whatever the case may be, the presented results show that protection against ticks for these employees can be improved by the use of uniform permethrin-treated trousers. Unfortunately, it cannot be designated which grade of protection can be achieved by wearing uniform trousers without further permethrin treatment from the presented data.

Since participation in this study was voluntary, a selection bias cannot be excluded. Respondents showing high tick susceptibility or a history of tick-borne diseases might be overrepresented in the presented study population. Generalizations of tick infestation rates may therefore be undertaken with caution. Nevertheless, the inter-group comparisons presented here may be considered as conclusive, since the participants did not know whether they would receive permethrin-treated or untreated trousers during the recruitment process. Furthermore, only groups of respondents stating comparable susceptibility towards tick infestation and job category have been compared.

CONCLUSION

Trousers with long-lasting permethrin impregnation can help to reduce tick infestation in forestry workers. However, potential benefit seems to be dependent on the particular type of clothing, with significantly enhanced protection only in particularly vulnerable employees using trousers without cut protection.

Considering this and calling to mind that the use of respective clothing might entail an uptake of permethrin by the user, a differentiated usage of respective clothing (e.g. for particular job tasks) would be preferable to its general use.

Since tick infestations have also been found when using permethrin-treated trousers, additional protection measures, such as checking the body for ticks after work or improving mechanical barrier functions of clothing, are still indispensable.

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