DOI: 10.2428/ecea.2017.24(2)22 ECOL CHEM ENG A. 2017;24(2):269-275

Paula WRÓBLEWSKA-ŁUCZKA[1]*, Piotr ADAMCZUK[2]
and Jarogniew ŁUSZCZKI[2]

OCCUPATIONAL EXPOSURE AMONG FORESTERS TO DEET – SURVEY STUDIES, CHROMATOGRAPHIC ANALYSIS

NARAŻENIE ZAWODOWE LEŚNIKÓW NA DEET – BADANIA ANKIETOWE, ANALIZA CHROMATOGRAFICZNA

Abstract: Foresters, considering the character of their work, are exposed to the occurrence of many types of diseases. The main occupational risk among forestry workers is caused by infectious pathogens, the main reservoir of which are wild animals, and the vector are ticks. Forestry workers are exposed to the effect of repellents, including DEET (N,N-diethyl-m-toluamide) in association with protection against tick and mosquito bites. The goal of the project was to discover the amount of DEET identified in the samples of sweat and urine from the foresters who use chemical agents repelling insects. The study covered 22 foresters from the Janow Lubelski Forest District, and 10 individuals who constituted a control group. Questionnaires concerning the use of repellents, and urine samples were collected (2 from each person in the study), as well as sweat samples (4 each). Each collected biological sample was properly prepared and subjected to chromatographic analysis (GC/MS) for the identification of DEET. The season of repellents use is from March–October, or even November. The highest intensity of the use of repellents (as many as 5 days a week) is during the period from May–July. The chromatographic analyses performed did not show even the lowest content of DEET in the collected biological samples from both the control group and foresters. The absence of DEET in the foresters' urine may be explained by the late date of collection of the samples – in October, when they no longer use repellents, or use them very rarely. The studies conducted within this project allowed the observation that DEET is not accumulated, is subject to quick elimination from the body, which favourably affects the safety of its use. Exposure to chemical occupational hazards in forest areas is an underestimated problem of occupational medicine and public health, which has not been fully explored.

Keywords: DEET, occupational exposure, forestry workers

[1] Medical University of Lublin, Department of Pathophysiology, Jaczewskiego 8b, 20-090 Lublin, Poland, phone: +48 81 44 86 500.

[2] Institute of Rural Health, Jaczewskiego 2, 20-090 Lublin, Poland, phone: +48 81 71 84 488, +48 81 71 84 542.

* Corresponding author: wroblewska.paula88@gmail.com

Introduction

Foresters, considering the character of their work, are exposed to the occurrence of many types of diseases. The main occupational risk among forestry workers is caused by infectious pathogens, the main reservoir of which are wild animals, and the vector are ticks. High incidence of tick-borne diseases among foresters is related, among other things, with the lack of effective methods of ticks control in the natural environment. Reduction of the population of ticks by chemical methods is hindered, or impossible, considering the aspects of environment protection, and often unprofitable because it is poorly effective. In some countries, the phenomenon of resistance of ticks to acaricides has already been observed. These are miticides, which are chemical substances for the control of mites, arachnids, and moths, which are pests of cultivated plants and food products. The application of biological methods still remains at the stage of studies, with the lack of full studies in this direction [1, 2]. Therefore, the basic method of protection against tick bites is personal prophylaxis, such as proper clothes, inspection of the body after work in the forest, or the use of repellents which discourage ticks [3].

Objective

Forestry workers are exposed to the effect of repellents, including DEET (N,N-diethyl-m-toluamide) in association with protection against tick and mosquito bites. The use of repellents in this occupational group is related with everyday work, and the doses applied must be repeated (several times daily) in order to provide even better protection. Due to high daily doses of repellents with DEET this study group is most interesting considering exposure to this chemical agent.

The goal of the project was to discover the amount of DEET identified in the samples of sweat and urine from the foresters who use chemical agents repelling insects.

Materials and methods

The study covered 22 foresters from the Janow Lubelski Forest District, and 10 individuals who constituted a control group. The control group included healthy volunteers, who did not work in forestry, and did not have frequent contact with repellents. These were mainly office workers.

During 2 field visits (13 and 17 October 2016) questionnaires concerning the use of repellents, and urine samples were collected (2 from each person in the study), as well as sweat samples (4 each). Sweat samples were collected using special cotton patches of the size (6.5 × 3.8 cm) stuck on the lumbar part of the spine for the period of 10–12 hours. Two patches were used to collect sweat samples during the daytime, and 2 at night. Each participant of the study was thoroughly informed concerning its course and the method of collecting samples and their further management, for example, patches with sweat were closed in string bags in order to avoid the evaporation of sweat.

In addition, in accordance with the recommendations by the Bioethics Commission at the Institute of Rural Health, prior to the study, each examined person signed their consent to participate.

Each collected biological sample was properly prepared and subjected to chromatographic analysis (GC/MS) for the identification of DEET. The samples were extracted by means of diethyl ether in which the chemical compound DEET (N,N-diethyl-m--toluamide) is very well soluble. In order to create own library of spectra and for the assessment of the effectiveness of extraction, the DEET (Sigma-Aldrich) standard was prepared of the concentration of 250 g/dm^3, and a series of dilutions of DEET.

The method of extraction of DEET from biological tubes were based on the combination of: extraction methods known from forensic chemistry [4], and the methodology by Wu et al. with own modifications [5].

Chromatographic analysis was performed using a gas chromatograph Agilent 6890N connected with Agilent 5975B mass spectrometer.

Separation of compounds was achieved on a 30m HP-5MS 5% phenyl methyl siloxane capillary column. Helium at a flow rate of 0.001 dm^3/min was used as the carrier gas. The inlet temperature was set at 275^{o}C. The column temperature was initially held at 90^{o}C for 2 minutes and than programmed to 250^{o}C at a rate of 10^{o}C/min. The quadrupole was operated in the single ion monitoring mode. Chromatographic analysis was performed based on the method by Cherstniakova, Garcia, et al. 2006, with own modification of the SIM method (ions: 65, 91, 119, 190, 191) [6, 7].

Chromatographic analyses of DEET standards were performed, based on which the calibration curve and curve equation were plotted. Each of the collected samples of urine and sweat was also subjected to chromatographic analysis in order to determine the presence and content of DEET.

Results and discussion

According to the assumption, the individuals from the control group were mainly office workers, mean age 39. Only 3 persons (30%) declared the use of repellents in relation with occasional stay in a forest or a park. Then, they applied products such as 'OFF!' (15% DEET), or 'BROS' (up to 30% DEET).

The employees of the Janow Lubelski Forest District use repellents relatively regularly, the mean age in this group is 47. Among the foresters in the study 14 (64%) work exclusively in the field, while 8 (36%) perform mixed work, although their mean time spent in the field is 6.91 hours daily. The season of repellents use is from March–October, or even November. The highest intensity of the use of repellents (as many as 5 days a week) is during the period from May–July. The foresters apply repellents 4 times a week, twice daily, on average. The foresters in the study apply repellents on uncovered parts of the body (40%), or on uncovered parts of the body and on clothes (60%); mainly around the head area, sometimes on the whole body (Fig. 1).

The above-presented figure shows that 100% of the foresters in the study apply repellents around the head, while 45% additionally use repellents on the hands, and 5% on the whole body. All the foresters in the study (100%) use the product 'Ultrathon' (25% DEET),

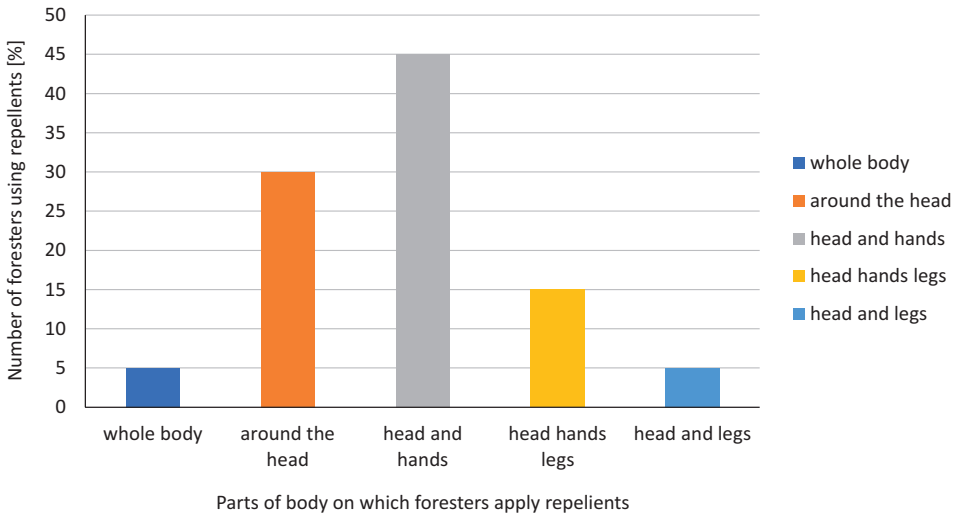

Fig. 1. Body parts on which foresters in the survey apply repellents

and 10% of them use the product 'Repel 100' (97% DEET). In addition, the foresters call repellents their 'deodorants' which shows the scale of the use of these products.

Chromatographic analyses of DEET standards were performed, based on which the calibration curve and curve equation were plotted (Fig. 2).

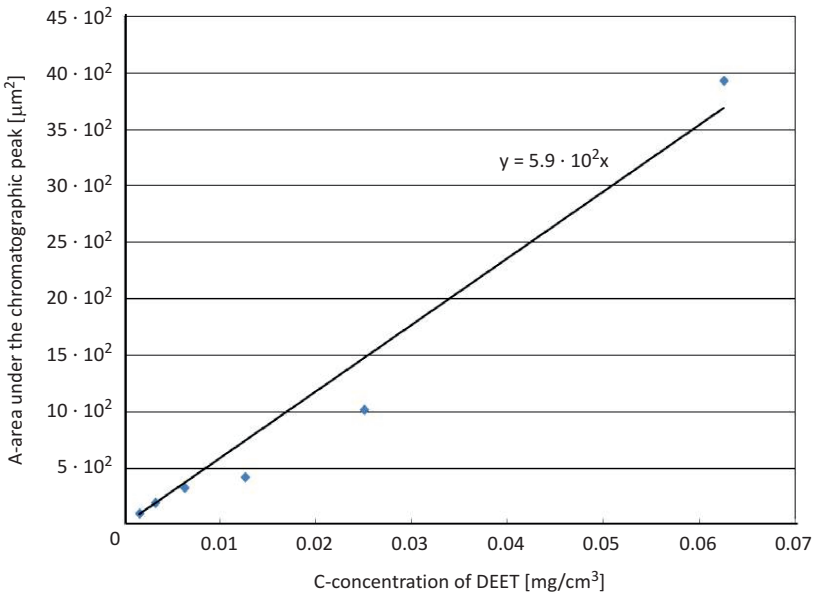

Fig. 2. Calibration curve which is the ratio between the area under the chromatographic peak and the examined concentration of DEET

The equation of the curve was obtained $y = 5.9 \cdot 10^9 \cdot x$, where y – is the area under the chromatographic peak, and x – the examined concentration of DEET. The lowest concentration of the identified DEET standard was the concentration 1.5625 μg/cm^3. The effectiveness of extraction of DEET from biological samples (where DEET was added in a known concentration as an internal standard) was 89%.

The chromatographic analyses performed did not show even the lowest content of DEET in the collected biological samples from both the control group and foresters. The absence of DEET in the foresters' urine may be explained by the late date of collection of the samples – in October, when they no longer use repellents, or use them very rarely. The results of the presented study and data available in literature allow the presumption that DEET, despite being excreted mainly in urine (as soon as 4 hours after application to the skin) [8], is not present in the body for a long time and is not accumulated.

DEET is a chemical substance which may have very hazardous effects, such as an irritating effect on the skin and eyes and may cause drowsiness or dizziness. In addition, it is dangerous for the water environment, it should not be inhaled, may cause eye damage, and irritation of the gastrointestinal tract [9]. While applying preparations containing DEET it is necessary to take special precautions, i.e. wash hands after spreading over the protected areas of the skin, when in the form of aerosols it should be used exclusively outdoors or in well-ventilated rooms, because – as any chemical compound – it may be toxic [10]. The occurrence of dermal (allergic) symptoms is also possible after the application of preparations containing DEET to the skin. DEET is considered safe for health; however, scientific studies conducted *in vitro* showed its apoptotic effects on hepatocytes (liver cells) [11]. Other studies conducted among pregnant women showed that low doses of DEET were safe for both the women and babies; nevertheless, after detailed studies, the presence of this substance in umbilical cord blood was noted [12]. Scientific studies show that physical activity exerts an effect on the duration of the protection time, decreasing the effectiveness of the preparations containing DEET [13]. However, on the other hand, DEET applied to the skin may disturb the production of sweat and normal evaporation, thus increasing the physiological burden while performing work requiring physical effort [14].

Among studies concerning the exposure to DEET, the examined groups of children should be mentioned (60 children aged 1–6), from agricultural families in North Carolina (USA). The samples of urine were collected from the children, and an interview was conducted among parents concerning plant protection products present in their environment of life. The urine samples were analyzed from the aspect of the content of many pesticides, herbicides, or repellents. DEET was identified in 10% of the urine samples from children. Children living in agricultural families are exposed to many sources of chemicals, which may remain in the environment for a long time [15].

Considering many side-effects of the use of DEET, including several cases of death due to a high amount absorbed through the skin, many studies are being conducted concerning the addition of new auxiliary substances to preparations containing DEET, in order to change their bio-distribution and decrease the absorption through the skin [16]. Studies are also conducted aimed at replacement of DEET by other alternative

natural components, e.g. essential oils [17]. The studies conducted within this project allowed the observation that DEET is not accumulated, is subject to quick elimination from the body, which favourably affects the safety of its use.

Discussion and conclusions

The problem of protection of humans against insect bites becomes an increasingly more important problem for both medicine and health protection of those exposed, as well as the problem of public health as a whole. The effective direction of efforts undertaken may be special textiles with protective functions against insects and ticks designed for clothing products. Special protective functions would predestine them for use in the production of working clothes designed for foresters, hunters, or employees of urban green areas. These materials would also be widely used in the production of clothes for those in whom exposure is associated with recreational stay in the forests or parks, including gardeners, mushroom pickers, and fishermen [18].

Another direction of activities is also the seeking for new active substances, the effect of which would be directed towards the repelling insects and ticks. These substances should be effective as well as safe for the user, and cause the least possible side-effects. It is also important that these substances should be safe for the environment.

Exposure to chemical occupational hazards in forest areas is an underestimated problem of occupational medicine and public health, which has not been fully explored.

Acknowledgements

The project implemented under Statutory Research for Young Scientists in Institute of Rural Health MN16003.

References

[1] Benelli G, Pavela R, Canale A, Mehlhorn H. Parasitol Res. 2016;115:2545-2560. DOI: 10.1007/s00436-016-5095-1.
[2] Żukiewicz-Sobczak W, Chmielewska-Badora J, Wróblewska P, Zwoliński J. Postep Derm Alergol. 2013;5:311-315. DOI: 10.5114/pdia.2013.38361.
[3] Rahlenbeck S, Fingerle V, Doggett S. Br J Gen Pract. 2016;66:492-494. DOI: 10.3399/bjgp16X687013.
[4] Jasiewicz B, Kowalczyk I, Kurek J. Chemia Sądowa (Forensic Chemistry). Poznań: Wyd. Uniwersytet Adama Mickiewicza; 2012. ISBN 978-83-63783-11-3.
[5] Wu A, Pearson ML, Shekoski DL, Soto RJ, Stewart RD. J Sep Sci. 1979;2(9):558-562. DOI: 10.1002/jhrc.1240020904.
[6] Cherstniakova SA, Garcia GE, Strong J, Bi D, Weitz J, Roy MJ, et al. J Anal Toxicol. 2006;30(1):21-26. DOI: 10.1093/jat/30.1.21.
[7] National Institute of Standards and Technology, http://webbook.nist.gov/cgi/cbook.cgi?ID=C134623&Mask=200.
[8] Selim S, Hartnagel Jr RE, Osimitz T, Gabriel KL, Schoenig GP. Fundam Appl Toxicol. 1995;25(1):95-100. DOI: 10.1006/faat.1995.1043.
[9] PubChem Open Chemistry Database, https://pubchem.ncbi.nlm.nih.gov/compound/4284#section=Hazards-Identification.

[10] Corbel V, Stankiewicz M, Pennetier C, Fournier D, Stojan J, Girard E, et al. BMC Biol. 2012;10:86. DOI: 10.1186/1741-7007-10-86.
[11] Das PC, Cao Y, Rose RL, Cherrington N, Hodgson E. Drug Metabol Drug Interact. 2008;23(3-4):237-260. DOI: 10.1515/DMDI.2008.23.3-4.237.
[12] McGready R, Hamilton KA, Simpson JA, Cho T, Luxemburger C, Edwards R, et al. Safety of the insect repellent N,N-diethyl-M-toluamide (DEET) in pregnancy. Am J Trop Med Hyg. 2001;65(4):285-289. http://www.ajtmh.org/docserver/fulltext/14761645/65/4/11693870.pdf?expires=1516907556&id=id&accname=guest&checksum=18E1E342095F528C8311170D7AA13BEA.
[13] Schofield S, Tepper M, Gadawski R. J Med Entomol. 2007;44(6):1026-1031. DOI: 10.1093/jmedent/44.6.1026.
[14] Kenefick RW, Cheuvront SN, Ely BR, Palombo LJ, Sawka MN. Eur J Appl Physiol. 2011;111(12):3061-3068. DOI: 10.1007/s00421-011-1932-1.
[15] Arcury TA, Grzywacz JG, Barr DB, Tapia J, Chen H, Quandt SA. Environ Health Perspect. 2007;115(8):1254-1260. DOI: 10.1289/ehp.9975.
[16] Qiu H, Jun HW, McCall JW. Pharmacokinetics, formulation, and safety of insect repellent N,N-diethyl-3-methylbenzamide (DEET): a review. J Am Mosq Control Assoc. 1998;14(1):12-27. https://archive.org/stream/cbarchive_103643_pharmacokineticsformulationand1998/JAMCA_V14_N1_P012-027_djvu.txt
[17] Ochoa SA, Sánchez-Torres LE, Nevárez-Moorillón GV, Camacho AD, Nogueda-Torres B. Biomedica. 2017; 37: 224-243. DOI: 10.7705/biomedica.v37i0.3475.
[18] Diaz JH. Wilderness Environ Med. 2016;27:153-163. DOI: 10.1016/j.wem.2015.11.007.

NARAŻENIE ZAWODOWE LEŚNIKÓW NA DEET – BADANIA ANKIETOWE, ANALIZA CHROMATOGRAFICZNA

Instytut Medycyny Wsi im. Witolda Chodźki, Lublin

Abstrakt: Leśnicy z uwagi na charakter wykonywanej pracy narażeni są na występowanie różnego typu chorób. Głównym zagrożeniem zawodowym dla pracowników leśnictwa są patogeny zakaźne, których głównym rezerwuarem są dzikie zwierzęta, a wektorem kleszcze. Pracownicy leśnictwa są narażeni na działanie repelentów w tym DEET (N,N-dietylo-m-toluamid) w związku z ochroną przed ukłuciami kleszczy i komarów. Celem projektu było poznanie, jaka ilość DEET zostanie zidentyfikowana w próbkach potu i moczu pochodzących od leśników, którzy stosują środki chemiczne odstraszające owady. W badaniach wzięło udział 22 leśników Nadleśnictwa Janów Lubelski oraz 10 osób stanowiących grupę kontrolną. Zostały zgromadzone ankiety dotyczące stosowania repelentów oraz zostały zebrane próbki moczu (po 2 od każdej osoby badanej) i próbki potu (po 4). Każda z zebranych próbek biologicznych została odpowiednio przygotowana i poddana analizie chromatograficznej (GC/MS) celem identyfikacji w nich DEET. Sezon stosowania repelentów przypada na okres od marca do października, a nawet listopada. Największe natężenie stosowania repelentów (bo aż 5 dni w tygodniu) przypada na okres od maja do lipca. Przeprowadzone badania chromatograficzne nie wykazały nawet najmniejszej zawartości DEET w zgromadzonych próbkach biologicznych pochodzących zarówno od grupy kontrolnej, jak i od leśników. Brak obecności DEET w moczu leśników należy tłumaczyć późnym terminem zbiorem próbek – w październiku, kiedy leśnicy nie stosują już repelentów lub stosują bardzo rzadko. Badania przeprowadzone w tym projekcie pozwoliły zaobserwować, że DEET nie jest akumulowany, podlega szybkiej eliminacji z organizmu, co wpływa korzystnie na bezpieczeństwo jego stosowania. Ekspozycja na chemiczne zagrożenia zawodowe na obszarach leśnych jest niedocenionym i nie do końca zbadanym problemem medycyny pracy i zdrowia publicznego.

Słowa kluczowe: DEET, narażenie zawodowe, leśnicy