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LIVESTOCK PROTECTIVE FENCING (LPF) TO PROTECT DAIRY CATTLE AGAINST VECTORS IN SERBIA - PROJECT PROCESSES AND METHODOLOGY

Jean-Baptiste Rayaisse[1], Burkhard Bauer[2], Ivan Pavlović[3], Branislav Bingulac[4], Ljubiša Jovanović[5], Daniel Beltran-Alcrudo[6]

[1]Centre International de Recherche – Développement sur l'Elevage en zone Subhumide (CIRDES). 01 BP 4544 Bobo – Dioulasso 01, Burkina Faso
[2]Free University of Berlin, Institute for Parasitology and Tropical Veterinary Medicine, 14163 Berlin, Germany
[3]Scientific Veterinary Institute of Serbia, 11000 Belgrade, Serbia,
[4]Milcommerce Ltd.", 11080 Belgrade-Zemun, Serbia
[5]Serbian Dairy Association, 11000 Belgrade, Serbia
[6]FAO, Regional Office for Europe and Central Asia, 1068 Budapest, Hungary
Corresponding author: Ivan Pavlović, dripavlovic58@gmail.com
Short communication

Abstract: Lumpy skin disease (LSD) is among a number of vector-borne diseases (VBDs) threatening the Balkans and therefore, preventing feeding of insects on cattle would reduce the spread of VBDs. In order to test the efficiency of Livestock Protective Fences (LPF) in the protection of dairy cattle from insect bites, a case-control study was conducted, in the districts of Nišava, Pirot and Pčinja, Southern Serbia. It consisted in comparing the number of biting flies collected within time, between 10 farms protected with LPF and 10 non protected ones. The insects were collected using two types of traps; the monoconical Vavoua trap set outside in between forested areas or rivers and the actual farm, and the BG-sentinel trap baited with CO_2, placed in proximity of the cattle but outside the stable. Vectors were collected every 15 days for 48 hours from May to October 2018 and kept in vials containing 70% of ethanol. Catches per trap were separately stored and for each trap, insects were classified according to species and sex and then counted. Data on milk parameters were analyzed separately, on data collected within protected farms, before and after the LPF deployment, and on data without protection at all. It was not possible to detect a direct impact of LPF on vector densities but the number of bacteria colonies (CFU) values were reduced. Some corrections/adaption in the methodology used may lead to better impact.

Key words: Serbia, lumpy skin disease, vectors borne diseases, Livestock Protective Fence, milk quality

Introduction

Lumpy skin disease (LSD) is among a number of vector-borne diseases (VBDs) threatening the Balkans. This notifiable disease has dramatic effects on rural livelihoods and the effect at national level is also devastating due to strict trade restrictions (*Casal et al., 2018; Molla et al., 2017*). The situation worsens with the arrival of summer's higher temperatures, favouring insect multiplication and, hence, disease dissemination. Therefore, preventing feeding of insects on livestock would also reduce the spread of VBDs (*Bauer et al., 2006; Bauer et al., 2011*). Unfortunately, vector control strategies used so far are either costly, time consuming or environmentally unacceptable. The large-scale and indiscriminate use of insecticides, mostly pyrethroids, constitutes the mainstay of vector control efforts. Pyrethroids are all upsetting the function of the sodium channels. Resistance against one pyethroid will lead to resistance against the whole class. This has led to widespread pyrethroid insecticides resistance in target vectors insects like *Aedes sp., Culex sp.* or *Culicoides sp.* (*Laveissiere and Grebaut,1990; Caputo,2018; Pichler et al., 2018; Bengoa et al., 2017*) Stable flies (*Stomoxys* spp.) have found to be less or no longer susceptible, (*Reissert-Oppermann et al., 2019*). However, there is a highly effective technology, the use of insecticide-incorporated knitted textile screens (known as livestock protective fences – LPF), which has been very successfully used in sub-Saharan African countries for the control of several VBDs (*Heilmann et al., 2017; Maia et al., 2012; 2010; Bauer et al., 2011*). The fences are deployed in the vicinity of resting places near to cattle or alongside milking parlors, preventing insects from alighting on the animals.

The technology, never used in the Balkans, could be very useful in preventing VBDs. Another externality is stress reduction associated with insect bites of dairy cattle, an improvement of animal welfare and an increase in milk production (*Maia et al., 2010*). Moreover, significant reductions in mastitis cases have been recorded in the past, as insect vectors commonly transmit bacteria from animal to animal.

The current study aims to scientifically prove the efficiency of LPFs in the protection of dairy cattle from insect bites, thus reducing the transmission of VBD such as LSD, and reducing the stress of the animals due to insect bites.

If proven successful, the pilot study could be expanded within Serbia and to other affected and at-risk countries in the region (*Allepuz et al., 2018*).

Materials and Methods

Study location

Twenty farms were selected for a case-control study over one vector season, from mid-May to October 2018. Half of the farms were protected by LPF (case group), while the other half was considered as a control group, enabling to assess the trends of insect numbers and species. Half (50%) of the selected farms was located in the Nišava District with almost 100% of the case farms, and in Pirot and Pčinja districts, where control farms were mainly located (Figure 1). All farms were dairy production ones and were mostly maintained on a zero-grazing scheme, with cattle number not exceeding 40 in total.

The farms were characterized through a questionnaire covering the production system, dung management, insect control treatments, etc.

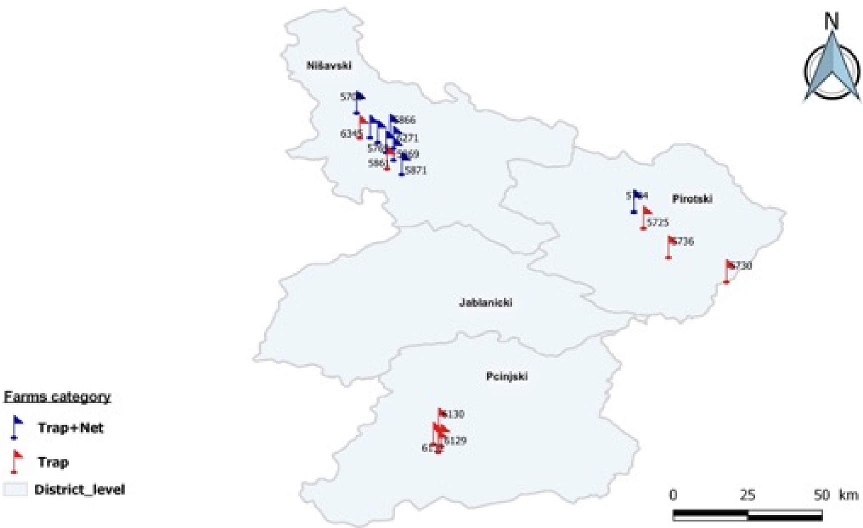

Figure 1. Location of the farms selected for the monitoring of the impact of LPF on the densities of LSD vectors

T0 data collection and monitoring of the LPF impact on vector densities

Once a farm was selected, and before the deployment of the LPF, two types of traps were used to estimate the presence of target insect species: Monoconical Vavoua (*Laveissière et al., 1990*) (Figure 2) and BG-sentinel trap (Figure 3). The Monoconical traps were set outside in between forested areas or rivers and the actual farm, while the BG-sentinel trap, baited with CO_2 (to enhance their efficacy) were placed in proximity of the cattle, but outside the stable, connected to a power source and attached to a CO_2 bottle. All trap locations were geo-referenced and visibly labelled. Throughout the intervention the positions of the traps were not supposed to be modified unless in a situation of emergency (change of pen location, bushfire, threat of theft). Continuous trapping would also allow assessing the LPF impact before and during intervention.

Figure 2. Monoconical Vavoua trap

Figure 3. BG-Sentinel trap

Likewise, data on milk parameters, notably, the CFU (colony-forming units; i.e. the number of viable bacterial/fungal cells/ml) and the number of somatic cells were collected from the dairy association, for comparison. These data related to milk production should allow to assess the effects of LPF on animal welfare and common dairy associated diseases such as mastitis.

Impregnation of LPF

Lambda-Cyhalothrin in a suspension concentrate (S.C.) in a concentration of 0.6% supplied from Changzhou Biochemical Co. Ltd, China, in 10 litre canisters was used for the impregnation. For each impregnation, the netting material (100 m *0.8m; 1-2 mm diameter holes) was soaked in 2.7 litres of solution of active ingredient and water, to thoroughly moisten the fabric with as little as possible relic solution remaining. After impregnation, the netting was first dried and then attached to horizontal pieces of wood to facilitate deployment (Figure 4).

In case farms, LPF were deployed covering all windows and openings in the stable, except for those needed for the operation of the farm, e.g. to get manure/feed in/out of the stable, and doors for animals and personnel to enter/exit/move around (Figure 5). In addition, LPF are also deployed around dung pits or ponds when not covered.

Figure 4. Netting preparation prior to deployment

Figure 5. Impregnated netting deployed around a farm to prevent insects from neighbouring forest

Vector collection and identification

Vectors were collected every 15 days for 48 hours from May to October 2018 by the farmers and then sent to the Scientific Veterinary Institute in Belgrade in vials containing 70% of ethanol. Any collected insect was duly labeled and identified based on morphological characteristics, using keys given (*Wall and Shearer, 1997; Cedric, 2005; Capinera 2008*). Catches per trap were separately stored and, for each trap, insects were divided/classified according to species and sex and then counted.

Data analysis

An ANOVA was first applied to the data, and for each treatment, (control or case), the period (month) was used to explain the variation of insect numbers random effects. Data were then submitted to a multiple comparison of means (Tukey contrast) using statistical package software available online (R Development Core Team, Vienna, Austria). Because (case and control) are very different in terms of geographical distribution, and so cannot be compared by pairs, this comparison was done within each type of treatment (case and control). The insects were first pooled by genus (Mosquitoes, Stomoxys, Tabanids, etc.) before being submitted to analysis.

For the data on milk parameters, analysis was made separately, on data collected within protected farms, before and after the LPF deployment, and on data without protection. As for cell numbers, CFU data were analyzed with a negative binomial generalized linear mixed model. The treatment was used as explanatory variable, and the farm ID and months were considered as random effects

Results

Insects collected

A total of 3,007 insects were collected during the 5-month monitoring period (Table 1), with up to 46% of *Musca spp* and 20% of *Ceratopogonidae*. Mosquitoes constitute up to 12% of the total insects caught, with a predominance of the genus *Culex* (54%)

Table 1. Total number of vectors collected during the 5 months monitoring

	Anopheles	*Culex*	*Aedes*	*Phlebo - tominae*	*Cerato-pogonidae*	*Tabani dae*	*Stomo-xyinae*	*Musca spp*	*Fan-niidae*
Mono	13	20	0	0	190	49	8	483	90
Mono net	30	11	0	0	157	33	7	376	80
BG	28	85	17	9	125	16	77	250	74
BG net	54	82	27	35	131	29	59	263	99
Total	**125**	**198**	**44**	**44**	**603**	**127**	**151**	**1372**	**343**

Aedes and *Phlebotominae* were only by BG traps (inside and closer to cattle) and none by the monoconical traps deployed outside and far from the cattle. Likewise, BG traps caught up 4-8 folds more *Culex* than monoconical traps.

Impact of LPF on vector densities

As indicated previously, this has been assessed in comparing the evolution of the densities within each type of treatment. Regardless of the natural variations, vector populations are expected to be decreasing within the time in farms where LPF was deployed. Numbers of different species of mosquitoes were pooled for this purpose.

For each of the species caught by both type of traps, no significant difference was observed between the two periods (before and after LPF deployment) in terms of insects' number.

Impact of LPF on milk quality parameters

In a first step, comparison was made within the 10 case farms before and after, showing a significant decrease of the number of CFU (a mean number of 319 to 122); p=0.028, figure 5.

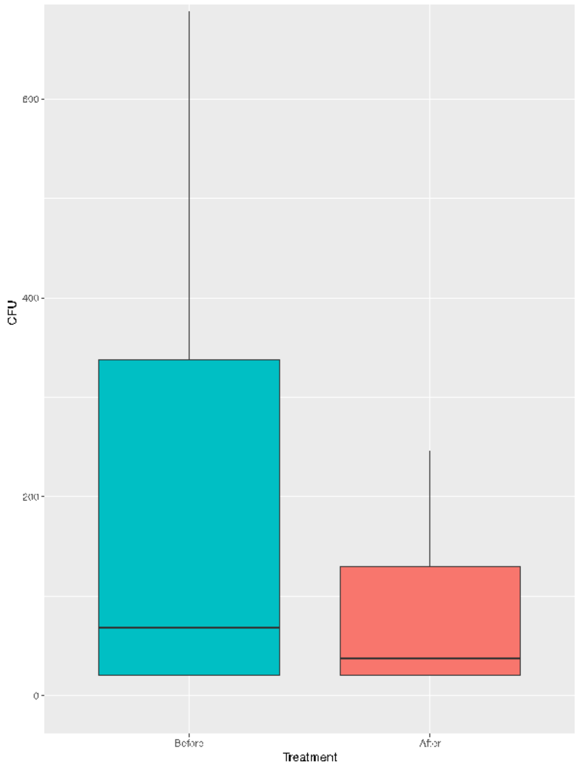

Figure 5. Mean number of CFU in protected farms, before and after the LPD deployment

For the somatic cells, however, the decrease is not significant; 241 *vs.* 200, p=0.519 (figure 6).

In a second step, the comparison was made within the 10 control farms, no significant evolution was observed for both CFU (275 *vs.* 181, p=0.795) and cells (253 *vs.* 296, p=0.442).

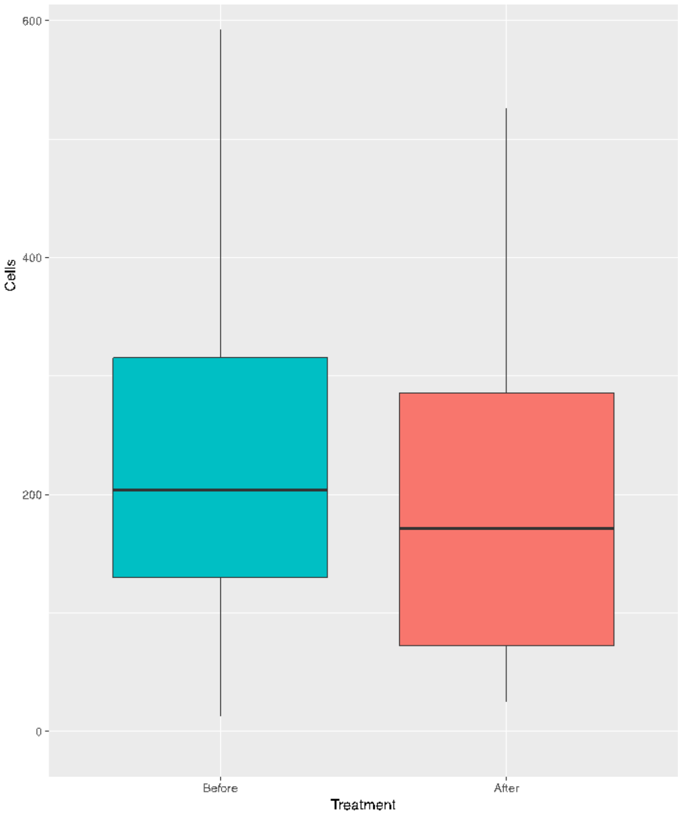

Figure 6. Mean number of cells in protected farms, before and after the LPD deployment

Discussion

Target insect densities were recorded and compared on 20 farms in southern Serbia. Ten farms served as controls and 10 farms benefited from a protection with insecticide-impregnated netting material that had been strategically deployed, closing windows and entrances. This approach was aiming at the control of putative vectors for LSD and other VBDs. Vector control may serve as a prevention measure against VBDs.

Insect densities were monitored by using a total of forty traps – i.e. two trap types per farm. Mono-conical ("Vavoua") traps were deployed outside the farm buildings but still within the premises of the respective farm. Mono-conical farms are particularly suited for catching horse flies and muscid flies – including stomoxyine biting flies, commonly known as stable flies.

BG sentinel traps are known for their efficacy of catching mosquito populations. Also, they have been used on a large scale for assessing other hematophagous vectors such as *ceratopogonidae* (biting midges) and *phlebotominae* (sandflies). BG traps were deployed in proximity of the cattle inside the farms. All traps were controlled at fortnightly intervals and all catches were transferred to Belgrade (Scientific Veterinary Institute) for identification and counting.

A cotton wool netting material was impregnated with lambda-cyhalothrin, a pyrethroid formulation. Previous work using this approach had shown great potential of controlling target insect populations in various African countries (*Bauer et al., 2011*) and in Germany. The ready-to-use fence was then attached to a wooden frame, reducing the space for entering target insect species. The trial lasted for five months during 2018.

As in previous studies, no impact on vector densities was found. Usually, it would take at least two years for any detectable reduction of target vector species (*Maia et al., 2010*). However, when controlling tsetse flies in a forest area of South-Eastern Ghana significant differences between protected and unprotected pens were detected (*Bauer et al., 2011*).

The year 2018 was particular in much of Europe with temperatures reaching or exceeding 40°C and a long, dry season. The climate may have had a negative impact on the survival rate of many insects. All the same, the initial figures of insect catches should have been high. However, at no time the numbers of collected insects corresponded to what has been recorded elsewhere. Indeed, during the Bluetongue outbreak in Germany, more than 1,000 Culicoides/trap/night were caught. It is puzzling and not well understood from our point of view why the total insect numbers were so low. For instance, it would be beyond any expectation from previous studies if a total of 603 biting midges were caught during five months with 40 traps. It is also puzzling to observe that the catches of midges with monoconical traps exceeded those ones recorded with BG sentinel traps. At the same time the catches of stable flies with mono-conical traps were widely below current expectations and cannot be explained at our level. It was also noted with surprise and cannot be explained why BG sentinel traps caught the vast majority of stable flies.

As already stated, any detectable impact on target vector populations would appear premature. However, it would be important and it is recommended to monitor and evaluate the efficacy of the netting material: immediately after impregnation and then at regular intervals up to the termination of this study.

During a study on dairy farms in Kenya, significant reductions of mastitis cases in milking cows due to their protection with LPF were shown. Similar assessment should be performed during an eventual follow-up study.

In view of the particular animal husbandry management system – all cattle were mostly kept inside the pens on a zero-grazing scheme, the putative vector populations can be narrowed down to stable flies, mosquito species, biting midges and sandflies. Arguably, the role of house flies should be considered as irrelevant in the transmission of LSD. Should there be a follow-up study, it might be worthwhile to record (with cameras) and compare defensive cattle movements on protected and control farms. Benefits would be more convincing if it were shown that the cattle well-being is greatly enhanced by a partial protection of their pens. Horse flies are exophile and exophagic. Their preference of feeding on horses and cattle while on pasture has been frequently observed. In reaction to this, it is observed for instance, that horses were trying to protect themselves by retreating under an open but roofed hut, whenever peak densities of horse flies were occurring. Stable flies, on the other hand readily feed on their hosts inside as well as outside the pens. Camera recordings of defensive movements would therefore go a long way in producing the required evidence for the benefits of protection.

Conclusion

The objective of this study was to explore the possibility of using LPF to protect cattle, in order to minimize risks of contracting VBD, notably LSD, and, also, to improve their wellbeing and milk quality. Although it was not possible to capture the direct impact of LPF on vector densities, the CFU values have improved. Some corrections/adaption in the methodology used may lead to better impact.

Zaštitna mreža za stoku (lpf) u zaštiti mlečnih krava od vektora prenosioca bolesti – projekat i metodologija

Jean-Baptiste Rayaisse, Burkhard Bauer, Ivan Pavlović, Branislav Bingulac, Ljubiša Jovanović, Daniel Beltran-Alcrudo

Rezime

Bolest kvrgave kože (LSD) spada među brojne bolesti koje se prenose vektorskim putem (VBD) koji prete Balkanu, pa bi sprečavanje hranjenja insekata stokom smanjilo širenje VBD-a. Da bi se testirala efikasnost stočnih zaštitnih mreža (LPF) u zaštiti mlečnih goveda od uboda insekata, sprovedena je studija njene efikasnosti. Ogledi su rađeni u Nišavskom, Pirotskom i Pčinjskom okrugu (južna Srbija). Sastojao se u poređenju broja hematofagih insekata prikupljenih u šestomesečnom periodu , na 10 farmi zaštićenih LPF i 10 nezaštićenih. Insekti su sakupljeni koristeći dve vrste zamki; monokonična zamka Vavoua koja je postavljena napolju između šumovitih područja ili reka i stvarne farme, i BG-sentinel zamka sa CO_2, smeštenih u blizini stoke, ali izvan štale. Vektori su prikupljeni svakih 15 dana tokom 48 sati od maja do oktobra 2018. godine i držali su se u bočicama sa 70% etanolom. Ulovi po zamci odvojeno su smešteni i za svaku zamku su insekti klasifikovani prema vrsti i polu, a zatim su brojani. Podaci o parametrima mleka analizirani su odvojeno. Podacima na zaštićenim farmama prikupljani su pre i posle primene LPF i sa farmi gde nije postavljena zaštita. Nije bilo moguće detektovati direktan uticaj LPF na gustinu vektora, ali je broj kolonija bakterija (CFU) u mleku smanjen. Neke korekcije / prilagođavanja u korišćenoj metodologiji mogu dovesti do boljeg uticaja primene LPF-a.

Ključne reči: Srbija, bolest kvrgave kože (LSD), vektorski prenosive bolesti, zaštitna mreža za stoku

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References

ALLEPUZ A., CASAL J., BELTRÁN-ALCRUDO D. (2018): Spatial analysis of lumpy skin disease (LSD) in Eurasia - Predicting areas at risk for further spread within the region. TBED 2018 Dec 6. doi: 10.1111/tbed.13090.
BAUER B., GITAU D., OLOO F.P., KARANJA S.M. (2016): Evaluation of a preliminary title to protect zero-grazed dairy cattle with insecticide-treated mosquito netting in western Kenya. Tropical animal health and production, 138(1), 29-34.
BAUER B., HOLZGREFE B., MAHAMA C.I., BAUMANN M.P., MEHLITZ D., CLAUSEN P.H. (2011): Managing tsetse transmitted trypanosomosis by insecticide treated nets--an affordable and sustainable method for resource poor pig farmers in Ghana. PLOS Neglected Tropical Diseases, 5(10), 1343. doi: 10.1371/journal.pntd.0001343. Epub 2011 Oct 11.BENGOA M., ERITJA R., DELACOUR S., MIRANDA M.Á., SUREDA A., LUCIENTES J. (2017): First Data on Resistance to Pyrethroids in Wild Populations of Aedes albopictus from Spain. Journal of the American Mosquito Control Association. 33(3), 246-249. doi: 10.2987/17-6636R.1.

CASAL J., ALLEPUZ A., MITEVA A., PITE L., TABAKOVSKY B., TERZIEVSKI D., ALEXANDROV T., BELTRÁN-ALCRUDO D. (2018): Economic cost of lumpy skin disease outbreaks in three Balkan countries: Albania, Bulgaria and the Former Yugoslav Republic of Macedonia (2016-2017). Transbound Emerg Diseases, 65(6), 1680-1688. doi: 10.1111/tbed.12926. Epub 2018 Jul 10.

CEDRIC G. (2005): Entomology, Springer, Berlin, Germany.

CAPINERA J. (2008): Encyclopedia of Entomology. Springer, Berlin, Germany.

HEILMANN M., ROESEL K., GRACE D., BAUER B., CLAUSEN P.H. (2017): The impact of insecticide-treated material to reduce flies among pork outlets in Kampala, Uganda.Parasitol Research, 116(6),1617-1626. doi: 10.1007/s00436-017-5450-x. Epub 2017 May 1.

LAVEISSIERE C., GREBAUT P. (1990): Recherches sur les pièges à glossines (*Diptera : Glossinidae*). Mise au point d'un modèle économique : le piège « Vavoua ». Tropical Medicine and Parasitology, 41**,** 185-192.

MAIA M.F., ABONUUSUM A., LORENZ LM., CLAUSEN P.H., BAUER B., GARMS R., KRUPPA T. (2012): The effect of deltamethrin-treated net fencing around cattle enclosures on outdoor-biting mosquitoes in Kumasi, Ghana. PLOS One, 7(9), e45794. doi: 10.1371/journal.pone.0045794. Epub 2012 Sep 19.

MAIA M., CLAUSEN P.H., MEHLITZ D., GARMS R., BAUER B. (2010): Protection of confined cattle against biting and nuisance flies (Muscidae: Diptera) with insecticide-treated nets in the Ghanaian forest zone at Kumasi. Parasitol Research, 106(6), 1307-1313. doi: 10.1007/s00436-010-1796-z. Epub 2010 Mar 9.

MOLLA W., DE JONG M.C.M., GARI G., FRANKENA K. (2017): Economic impact of lumpy skin disease and cost effectiveness of vaccination for the control of outbreaks in Ethiopia. Preventive Veterinary Medicine, 147, 100-107. doi: 10.1016/j.prevetmed.2017.09.003.

PICHLER V. BELLINI R., VERONESI R., ARNOLDI D., RIZZOLI A., LIA R.P., OTRANTO D., MONTARSI F., CARLIN S., BALLARDINI M., ANTOGNINI E., SALVEMINI M., BRIANTI E., GAGLIO G., MANICA M., COBRE P., SERINI P., VELO E., VONTAS J., KIOULOS I., PINTO J., DELLA TORRE A., CAPUTO B. (2018): First evidence of resistance to pyrethroid insecticides in Italian *Aedes albopictus* populations 26 years after invasion. Pest Management Science, 74(6), 1319-1327. doi: 10.1002/ps.4840. Epub 2018 Feb 21.

REISSERT-OPPERMANN S., BAUER B., STEUBER S., CLAUSEN P.H. (2019): Insecticide resistance in stable flies (Stomoxys calcitrans) on dairy farms in Germany. Parasitol Research, 118(9), 2499-2507. doi: 10.1007/s00436-019-06400-z.

WALL R., SHEARER D. (1997): Veterinary Ectoparasites: Biology, Pathology and Control, Blackwell Science Ltd., London.

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