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1 **TITLE:**

2 **Preventive vaccination contributes to control classical swine fever in wild boar (*Sus***
3 ***scrofa sp.*)**

4

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21

1 **ABSTRACT**

2

3

4 Over the last twenty years, oral vaccination implementing a live attenuated vaccine has been
5 experimented in Europe in order to control classical swine fever (CSF) in Wild Boar (*Sus*
6 *scrofa sp.*). This has generally led to an enhanced seroprevalence and a decreased
7 viro-prevalence at the scale of the whole vaccinated populations, but no quantitative analysis
8 has demonstrated the protective effect of preventive vaccination or intensive baiting. In the
9 present paper we conducted an retrospective analysis at the scale of the municipality, taking
10 into account the local dynamics and possible covariates of infection to test the effect of
11 preventive vaccination and of the baiting effort. To be efficient, vaccination was expected to
12 increase seroprevalence above the level considered as suitable for preventing disease invasion
13 (40-60%) independently of infection, to protect free areas from disease invasion or contribute
14 to control subsequent disease intensity and duration. We also hypothesized that a better
15 baiting effort would be correlated with an improvement of immunisation and disease control.
16 In uninfected municipalities, seroprevalence increased up to 40% after one year, i.e. three
17 vaccination campaigns. We observed a significant protective effect of preventive vaccination,
18 especially within municipalities that had been vaccinated at least one year before disease
19 emergence and where virus detection did not last more than one quarter. On the other hand,
20 we did not detect a significant effect of the baiting effort on local seroprevalence or disease
21 dynamics, suggesting that the baiting system could be improved. We discuss these results
22 regarding the improvement of management measures and further research perspective.

23

24 **Key-words : wild boar / classical swine fever / preventive vaccination**

25

1 1. INTRODUCTION

2

3

4 The control of classical swine fever (CSF) in the Wild Boar (*Sus scrofa sp.*) is required in the
5 European Union, since this species may act as a wild reservoir of the virus (Fritzmeier et al.,
6 2000, Anon 2001, Artois et al., 2002).

7 Mathematical models of this host-virus system have been developed and suggested that
8 preventive immunisation (40 to 60%) of susceptible populations may prevent disease
9 emergence (Hone et al., 1993, Guberti et al., 1998).

10 In the field, oral vaccination has been implemented using a Riems C-strain vaccine and baits
11 attractive to wild boars (Kaden et al., 2000). This process has been improved in Germany
12 during the 2000's to maximize individual antibody titre and seroprevalence (Kaden et al.,
13 2002, Kaden et al., 2003, Kaden et al., 2004). Retrospective analysis showed that high
14 seroprevalence (>60%) and low viro-prevalence (<0.1%) were maintained in vaccinated areas
15 (Kaden et al., 2002, Kaden et al., 2003, Louguet et al., 2005, Kaden et al., 2006, Rossi et al.,
16 2006a, Von Rden et al., 2008).

17 However, assessing the effect of vaccination on disease dynamics has been a difficult issue
18 because antibodies generated by the Riems C-strain vaccine and by the natural infection are
19 similar (Kaden et al., 2000). Furthermore, retrospective studies have often considered data
20 from a large infected area as a whole and have thus not taken into account the complex spatio-
21 temporal evolution of outbreaks. During the epizootic phase, the virus spreads over space and
22 locally decreases over time, moreover many other covariates interfere with disease dynamics
23 such as population size and landscape structure (Rossi et al., 2005a, Rossi et al., 2005b,
24 Kramer-Schadt et al., 2007). According to the principle of pseudo-mass action and critical
25 community size developed by modellers, the number of hosts is supposed to favour disease

1 invasion and persistence at both local and global level (Hone et al., 1993, Begon et al. 2002,
2 Rossi et al., 2005b, Kramer-Schadt et al., 2007). Forest continuity is known to influence the
3 probability of contacts between neighbouring wild boars and the resulting probability of
4 disease spreading and persistence at the scale of the whole area (Grenfell and Harwood 1997,
5 Rossi et al., 2005b, Kramer-Schadt et al., 2007, Kramer-Schadt *in press*). Lastly, even though
6 the baiting effort, *i.e.*, the number of baits delivered per wild boar, is strongly related to the
7 cost of vaccination, its effect has not been tested so far.

8

9 In the present paper we assess the effectiveness of preventive vaccination by considering data
10 at the municipality level, *i.e.*, at a spatial scale where wild boars experienced the same
11 epidemiological situation, where we could differentiate preventive from post-active
12 vaccination and avoid confounding effects of other covariates. We considered the data
13 collected in the Vosges mountains from 2003 to 2007. We targeted the following questions:

- 14 1. in uninfected areas, did vaccination increase seroprevalence above the level
15 considered as suitable for preventing disease invasion(40-60%)?
- 16 2. did preventive vaccination protect from disease invasion or contribute to control
17 subsequent disease intensity and duration?
- 18 3. was baiting effort correlated with a better immunisation and control of the outbreak?

19

20

2. MATERIALS AND METHODS

2.1. Study area, epidemiological context and vaccination process

The study area is located in France in the Vosges Mountains (48° 50' N and 7° 30' E); it covers 3,030 km² including 1,180 km² of forest land (Figure 1). Motorways, canals, rivers or towns constitute relative barriers to wildlife movements towards the west, east and south. Towards the north there is no physical barrier: the forest is uninterrupted between the Vosges and the Palatinate Forest (Germany) which constitute a large metapopulation (Rossi et al., 2005a).

In this area a previous outbreak of CSF occurred during the 90's and disappeared by 2000 (Rossi et al., 2005a). However, since April 2003 CSF has re-invaded the whole study area (Louquet et al., 2005, Pol et al., 2008). Oral vaccination has been implemented since August 2004 (Louquet et al., 2005) using a modified live vaccine (Riems C-strain) encapsulated into maize baits (Kaden et al., 2000). A double vaccination scheme has been adopted as recommended by Kaden et al., (2004), vaccination campaigns comprising two baits distributions have been performed three time a year: by February/March, June/July and August/September (2005-2006) or October/November (2007). On average, 40 baits were delivered per vaccination place each time, the area comprising on average 1.8 vaccination places per square kilometre (Kaden et al., 2002, Kaden et al., 2004, Kaden et al., 2006). Over the whole forested area, a total of 500 000 baits have been delivered each year; hunters buried each bait on feeding grounds where wild boars were attracted with maize and where hunting was temporarily interrupted to minimize the disturbance of animals (Louquet et al., 2005).

1 2.2. Sampling and diagnosis designs

2

3 Over the whole study area, blood and spleen were compulsorily sampled in all hunted wild
4 boars for serological and virological examinations (Louguet et al., 2005). Antibody detection
5 was performed using commercial ELISA kits according to the manufacturer's instructions
6 (Herdcheck CSFV Antibody test kit or CHEKIT CSF SERO Antibody, both distributed by
7 IDEXX® and of similar sensitivity). The genome of CSFV was amplified by real-time
8 polymerase chain reaction (r-RT-PCR) using a commercial r-RT-PCR kit according to the
9 manufacturer's instructions (TAQVET PPC® or ADIAVET CSF®). When positive or
10 doubtful r-RT-PCR results were observed, subsequent virus isolation was performed by the
11 French National Reference Laboratory for CSF (AFSSA) according to the EU-Diagnostic
12 Manual for CSF (Anon 2002). An animal was considered to be viropositive when either
13 CSFV could be isolated or the PCR threshold cycle was below 40 (Dewulf et al., 2004, Le
14 Potier et al., 2006, Depner et al., 2006, Le Dimna et al., 2008).

15

16 2.3. Covariates

17

18 • Vaccination

19 We considered the number of baits per shot wild boar as an indicator of the baiting effort
20 (Bait) that varied from 0.3 to 7 with an average of 4.4 baits delivered each time per shot wild
21 boar. The number of vaccination campaigns (T_V) conducted in each municipality was used as
22 an indicator of the vaccination effort over time.

23 Among the infected municipalities, we took into account the number of vaccination
24 campaigns since the first viropositive result (V_{acc}); we defined three different treatments:

- 1 i) Early vaccination, *i.e.*, when at least three campaigns had been performed before
2 the first viropositive result,
- 3 ii) Contemporaneous vaccination, *i.e.*, when one or two vaccination campaign(s) had
4 been performed before the first viropositive result,
- 5 iii) Late vaccination, *i.e.*, when the 1st PCR positive result had occurred before the
6 first vaccination campaign.

7 For the municipalities that were not yet infected at the beginning of vaccination, we estimated
8 the Euclidean distance (in Km) to the centroids of the closer infected municipalities (wave
9 front) (Dist). We considered that the municipalities located far from the wave front had been
10 preventively vaccinated as opposed to the others.

11

12 • **Other covariates**

13 For the infected municipalities, we defined a relative time scale (T_R) that was the sum of
14 quarters since the first viropositive result. We defined two seasons (Season): autumn/winter
15 from the first of October to the end of March and spring/summer from the first of April to the
16 end of September. Autumn/winter 2007 was considered as a third season type given that
17 vaccination was delayed to October-November in 2007 instead of August-September in 2005
18 and 2006.

19 We distinguished two ages classes according to the body mass (Age). The dressed carcasses
20 under 30kg were classified as juveniles, *i.e.*, less than one year old, whereas the others were
21 classified as more than one year old (Rossi et al., 2005a).

22 The hunting bag in 2006 was used as an indicator of the number of wild boars inhabiting the
23 municipality (Num). We took into account the proportion of forested areas (Forest) as an
24 index of connectivity between neighbouring municipalities. The number of roads separating

1 one municipality from its neighbours was calculated to measure a potential barrier effect of
2 roads (Road).

3

4 **2.4. Dependent variables and tested hypotheses**

5

6 We used data collected from April 2003 to December 2007 and defined four dependent
7 variables at the municipality level: the proportion of immune animals among unvaccinated
8 municipalities, the infectious status of municipalities, the proportion of viropositive animals
9 and the duration of infection.

10

11 • **Proportion of immune animals**

12 We considered that the municipalities exhibiting no viropositive result had not been infected.

13 In these municipalities, we studied the proportion of immune animals hypothesizing that
14 seropositive animals had been vaccinated and not infected.

15 The proportion of immune animals was likely to increase during the first vaccination
16 campaigns and no more subsequently due to a likely saturation of the vaccination process
17 (Louquet et al., 2005, Von Rden et al., 2008), we thus tested the effect of the number of
18 vaccination campaigns performed in each municipality (T_V). Given that wild boars less than
19 4.5 months old are poorly attracted by baits (Brauer et al., 2006, Rossi et al., *in prep.*) we
20 expected a lower immunity in juvenile wild boars compared to animals more than one year
21 old (Age). We also expected a difference between seasons (Season) (autumn/winter versus
22 spring/summer) since young wild boars grow during the year and since piglets born in spring
23 or summer enter the hunting bag each autumn, *i.e.*, when hunters start to hunt them.

24 We expected that to delay vaccination would increase the consumption of vaccine in autumn
25 because most of the wild boars would then be older than 4.5 months (Season). We finally

1 expected that the number of baits delivered per wild boar (Bait) would be positively
2 correlated with the proportion of immune animals.

3

4 • **Infectious status of municipalities**

5 The municipalities exhibiting at least one viropositive result were considered as infected
6 whereas the others were considered as uninfected. This status was examined in the
7 municipalities that were not yet infected when the vaccination started (August 2004). This
8 information was summarized as a binomial variable (0 for not infected / 1 for infected) to
9 study the factors influencing the probability of having been infected during the vaccination
10 period.

11 The probability of being infected was supposed to be influenced by the proportion of forest
12 (Forest), the number of roads (Road) and the number of wild boars (Num) since we assumed
13 that both forest continuity and a sufficient number of hosts were required for new areas to be
14 infected (Hone et al., 1993, Rossi et al., 2005b).

15 We expected a significant effect of the distance (Dist) from the wave front and of the baiting
16 effort (Bait) because we assumed preventive vaccination and intensive baiting could prevent
17 virus spread.

18

19 • **Proportion of viropositive animals**

20 At the level of each infected municipality, we analysed the virological results collected since
21 the first viropositive result. We considered the proportion of viropositive individuals as an
22 indicator of infection intensity.

23 The proportion of viropositive individuals was expected to be higher in juvenile wild boars
24 than in older individuals (Age) (Kern et al. 1999, Rossi et al. 2005b, Von Rden et al., 2008).

25 The proportion of viropositive individuals was also supposed to decrease over time (T_R)

1 (Kern et al., 1999, Rossi et al., 2005b, Von Rden et al., 2008). We expected that the number
2 of wild boars (Num), the proportion of forest (Forest) and the number of roads between
3 neighbouring municipalities (Road) would significantly influence the proportion of
4 viropositive animals because the number of hosts and the green corridors are supposed to
5 increase the probability of contacts between infected and susceptible animals (Begon et al.,
6 2002, Hone et al., 1993, Guberti et al., 1998, Rossi et al., 2005b).

7 Lastly, we expected that vaccination timing (Vacc) and baiting effort (Bait) would influence
8 the proportion of viropositive animals because we assumed that to apply preventive
9 vaccination and to intensify baiting could at least decrease the probability of virus
10 transmission (Hone et al., 1993, Guberti et al., 1998, Kaden et al., 2002).

11

12 • **Duration of infection**

13 We considered the sum of quarter(s) with viropositive results as an indicator of the local
14 duration of infection in the infected municipalities.

15 We expected that the number of wild boars (Num), the proportion of forest (Forest) and the
16 number of roads between neighbouring municipalities (Road) would significantly influence
17 the duration of infection. Indeed, at the level of the municipality, disease persistence is more
18 likely above a threshold population size and because at the level of a network of neighbouring
19 municipalities virus circulation is more likely if the forested areas are uninterrupted between
20 them (Grenfell and HarForest 1997, Rossi et al., 2005b, Kramer-Schadt *in press*).

21 We finally expected the duration of infection to be influenced by the vaccination timing
22 (Vacc) and the baiting effort (Bait) because we assumed that preventive vaccination and
23 intensive baiting would decrease the probability of a continuous chain of transmission over
24 time.

25

1 **2.5. Statistical analyses**

2

3 We used Poisson models to study the duration of infection and logistic models for the other
4 dependent variables (Dohoo et al., 2003). The sum of quarters with viropositive results
5 (Poisson model) was weighted by the sum of quarters with both viropositive and vironegative
6 results observed at the level of each infected municipality.

7 We adopted a step by step descendant process to test for the effect of each expected co-
8 variable. We used the Akaike Information Criterion (AIC) to select the best model. When the
9 difference in the AIC was less than 2, we selected the most parsimonious model, *i.e.*, the
10 model with the fewest parameters (Burnham and Anderson 1998). We tested for a significant
11 difference between specific parameters using Wald tests (Burnham and Anderson 1998). All
12 statistical analyses were performed using R 2.7.2 (the R foundation for statistical computing
13 2008).

14

15

1 3. RESULTS

2

3

4 3.1. Sampled animals and disease evolution in the whole study area

5

6 Within the 266 municipalities comprising the study area, 31,892 wild boars were sampled, of
7 which 30,097 exhibited a conclusive (positive/negative) virological result and 27,501
8 exhibited a conclusive serological result. At the level of the whole area, the proportion of
9 seropositive wild boars increased to 40% in juveniles and to 70% in older animals (Figure 2).

10 From 2003 to 2005 infection spread over the study area, and infection subsequently persisted
11 in a more enzootic way, the proportion of viropositive being less than 1% since 2006 (Figure
12 2).

13

14 3.2. Proportion of immune animals

15

16 Within the 168 municipalities with no viropositive result, 5,893 wild boars were sampled and
17 exhibited a conclusive serological result. The number of vaccination campaigns (T_v), age
18 (Age), season (Season) and an interaction between age and season (Age*Season) were
19 significantly correlated with the proportion of seropositive animals (table I). Contrary to our
20 assumptions, the number of baits delivered per wild boar (Bait) had no effect on
21 seroprevalence (table I). As expected, seroprevalence increased over the first three
22 vaccination campaigns and no more subsequently (Figure 3). As expected, seroprevalence
23 was lower in juveniles compared to older animals and this difference was even more
24 important during autumn/winter than in spring/summer (Figure 2). The proportion of juvenile
25 seropositive animals was slightly higher in autumn/winter 2007 (delayed vaccination) than in

1 the two previous years (figure 3) but this difference was not significant ($OR_{2007/other}=1.09$, P-
2 Wald=0.520) (table I).

3

4 **3.3. Infectious status of municipalities**

5

6 Within the 232 municipalities that were not yet infected when vaccination began, 5,908 wild
7 boars were sampled and exhibited a conclusive virological result. 91 municipalities among
8 232 exhibited at least one virological result and were considered as infected. The number of
9 wild boars (Num), the proportion of forest (Forest) and the distance from the wave front
10 (Dist) were significantly correlated with the status of the municipality (infected/not infected)
11 (table II). Contrary to our expectations, the number of roads (Road) and the number of baits
12 (Bait) had no effect on the status of the municipality (table II). As expected, the probability of
13 being infected was positively correlated with the number of wild boars ($OR_{1 \text{ shot wild boar}}=1.05$,
14 P-Wald= <0.001) and the proportion of forest ($OR_{1\% \text{ of Forest}}=6.03$, P-Wald=0.038) and
15 negatively correlated with the distance from the wave front ($OR_{1km}=0.88$, P-Wald= <0.001).

16

17 **3.4. Proportion of viropositive animals**

18

19 Within 91 infected municipalities, 2,065 wild boars exhibited a conclusive virological result
20 and were sampled after the observation of the first viropositive result. The number of wild
21 boars (Num), age (Age), time (T_R), the number of roads (Road), the vaccination treatment
22 (Vacc) and an interaction between time and vaccination treatment were significantly
23 correlated with the proportion of infected animals (table III). Contrary to our assumptions the
24 proportion of forest (Forest) and the number of baits (Bait) had no significant effect on the
25 proportion of infected animals (table III). As expected, the proportion of infected individuals

1 was higher in juveniles than in older animals ($OR_{\text{young/old}}=1.70$, $P\text{-Wald}<0.001$). As expected,
2 the proportion of infected animals was positively correlated with the number of wild boars
3 ($OR_{100 \text{ shot wild boar}}=1.35$, $P\text{-Wald}<0.001$) and slightly negatively correlated with the number of
4 roads separating neighbouring municipalities ($OR_{1 \text{ road}}=0.94$, $P\text{-Wald}=0.016$). The proportion
5 of infected animals decreased faster in the municipalities that had been vaccinated
6 contemporaneously with disease emergence compared to late vaccinated municipalities
7 (Figure 4). In municipalities vaccinated early, infection lasted no more than one quarter
8 (Figure 4).

9

10 **3.5. Duration of infection**

11

12 The number of wild boars (Num), the number of roads (Road), the vaccination treatment
13 (Vacc) and number of baits (Bait) had a significant effect on the duration of infection (table
14 IV). As expected, the duration of infection was positively correlated with the number of wild
15 boars ($OR_{100 \text{ shot wild boars}}=1.41$, $P\text{-Wald}<0.001$) and was slightly negatively correlated with the
16 number of roads between neighbouring municipalities ($OR_{1 \text{ road}}=0.94$, $P\text{-Wald}<0.001$). As
17 expected, the duration of infection was shorter in the municipalities vaccinated
18 contemporaneously with disease emergence compared to the municipalities vaccinated late
19 ($OR_{\text{contemporaneous/late}}=1.39$, $P\text{-Wald}<0.001$) and even shorter in the municipalities vaccinated
20 early ($OR_{\text{early/contemporaneous}}=1.54$, $P\text{-Wald}<0.001$). As assumed, the duration of infection and
21 the number of baits were slightly negatively correlated ($OR_{1 \text{ bait/shot wild boar}}=0.98$, $P\text{-}$
22 $Wald=0.016$).

23

24

1 4. DISCUSSION

2

3

4 As expected, the proportion of infected wild boars varied over time according to the
5 municipality; we thus confirm that in a study area comprising municipalities that were not
6 infected at the same time it is more appropriate to explore the effect of vaccination at the
7 municipality level than at the level of the whole study area. As expected wild boar abundance
8 and forest continuity had an important impact on infection emergence, intensity and
9 persistence, which is consistent with previous results in the same area (Rossi et al. 2005b) and
10 justifies the inclusion of these variables in the analysis.

11

12 As observed in previous field or experimental studies, juveniles were less efficiently
13 vaccinated and more frequently infected than older wild boars (Kern et al. 1999, Rossi et al.
14 2005b, Brauer et al., 2006, VonRüden et al. 2008). The delay of vaccination until
15 October/November slightly increased the proportion of juvenile seropositive animals in
16 comparison with August/September, possibly because a larger part of them were old enough
17 to be attracted by baits (Brauer et al., 2006, Rossi et al., 2006a); one may hypothesize that to
18 delay vaccination until winter could be even more effective. Obviously, duplicating the
19 experiment several times is required to test the effect of the different vaccination time-tables;
20 moreover, other factors must be considered: baits may be less attractive in October/November
21 owing to the seasonal oak mast production and competition may arise between juveniles and
22 adults even though baits are attractive to young wild boars (Rossi et al., 2006b, Von Rüden et
23 al. 2008). Our study does not properly address the problem of young animals but our results
24 confirm the interest of specific studies aiming to evaluate the role of young animals (Kaden et
25 al. 2006, Kramer-Schadt et al. in press, Rossi et al. in prep.) and to improve their vaccination

1 by developing more attractive baits (Bauer et al. 2006, Faust et al. 2007, Ballesteros et al. in
2 press).

3
4 In the municipalities where no viropositive result was detected, i.e., where we assumed that
5 the population was initially susceptible, the proportion of immune animals increased during
6 the first three vaccination campaigns before reaching the seroprevalence level required by
7 mathematical models (40-60%) for preventing disease emergence in a susceptible population
8 (Hone et al. 1993, Guberti et al. 1998). Accordingly, even though preventively vaccinated
9 municipalities became temporarily infected, we observed a lower intensity and duration of
10 infection in the municipalities where one to three vaccination campaigns had been performed
11 before disease emergence. These findings confirm the protective effect of preventive
12 vaccination and encourage the implementation of preventive rather than post-active
13 vaccination. During recent CSF outbreaks the wave front spread rate has been about 8km per
14 quarter over forested areas (von Rügen et al 2008, Rossi unpublished data); in such
15 conditions, i.e., an epizootic occurring in the absence of physical barriers such as open fields
16 or fenced motorways, preventive vaccination helped CSF control at a distance of 24km from
17 the wave front. Our results suggest that to vaccinate only part of a green corridor is not a
18 suitable option because CSF is likely to spread to free areas without any protective effect of
19 vaccination.

20
21 The number of baits delivered per wild boar neither affected the proportion of immune
22 animals nor the intensity of infection, which suggests that increasing the baiting effort did not
23 increase vaccination effectiveness. The slight negative correlation we observed between the
24 baiting effort and disease duration, although statistically significant, is difficult to interpret.
25 These surprising findings suggest that the baiting process was saturated; this possibly arises

1 because dominant wild boars had exclusive access to feeding grounds (Hebeisen 2007) and
2 consumed most of the baits delivered by hunters. Consequently, many baits might be wasted
3 and the cost-effectiveness of the system could be improved in the future by delivering fewer
4 baits or the same number of baits in a different way. To optimise the baiting effort will require
5 one to develop alternative delivery systems and to study the uptake pattern in detail. In this
6 respect, the use of biomarkers such as the iophenoxic-acid recently tested by Massei et al.
7 (2009) in this species would be helpful for exploring the bait uptake according to different
8 delivery systems.

9

10 Our retrospective analysis confirmed that preventive vaccination contributed to better control
11 CSF. However, to assess vaccination effectiveness in achieving disease eradication in the
12 areas still infected is a much more complex issue. In the Vosges/Palatinat forest, the CSF
13 genome was still detected by late 2008/early 2009, confirming the persistence of the virus in
14 this large metapopulation in spite of a huge vaccination effort
15 (http://ec.europa.eu/food/committees/regulatory/scfcah/animal_health/index_en.htm). In
16 another recently vaccinated area, CSF re-emerged after an apparent “silent phase” and was
17 still circulating by 2008 (Von Rügen et al., 2008,
18 http://ec.europa.eu/food/committees/regulatory/scfcah/animal_health/index_en.htm); on the
19 contrary, previous field observations suggested that vaccination lead to virus eradication
20 (Kaden et al., 2002, 2003). These different outcomes may possibly arise because the virulence
21 of recently observed strains was lower than in the past, owing to larger wild boar numbers, or
22 owing to the enhanced sampling effort and sensitivity of diagnostic techniques (r-RT-PCR).
23 Exploring this question requires specific theoretical approaches simulating different
24 vaccination scenarios and specifying the required conditions for CSF eradication (Kramer-
25 Schadt et al., 2007, Kramer-Schadt et al., in press).

26

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2

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8

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- 8

1 **Table I: Comparison of logistic models dealing with the proportion of immune animals**
 2 **observed in uninfected municipalities (the selected model is in bold and *italic*).**

3

Covariates	Number of parameters	AIC
Age*Season + Bait + Ln(T_v+1)	8	7014.2
Age + Season + Bait + Ln(T_v+1)	6	7022.0
<i>Age*Season + Ln(T_v+1)</i>	<i>7</i>	<i>7012.3</i>
Age**season + Bait	7	7641.1
Age + Season+ Ln(T_v+1)	4	7020.0
Age*Season	5	7645.7

4

5

1 **Table II: Comparison of logistic models dealing with the infectious status of**
 2 **municipalities (the selected model is in bold and *italic*).**

3

4

Covariates	Number of parameters	AIC
Forest + Road + Bait + Dist + Num	6	185.1
Forest + Road + Bait + Dist	5	205.6
Forest + Road + Bait + Num	5	199.0
Forest + Road + Dist + Num	5	183.1
Forest + Bait + Dist + "Num	5	186.6
Road + Bait + Dist + Num	5	189.1
Forest + Road + Dist	4	205.1
Forest + Road + Num	4	197.2
<i>Forest + Dist + Num</i>	4	184.8
Road + Dist + Num	4	187.1

5

6

1 **Table III: Comparison of logistic models dealing with the proportion of infected animals**
 2 **in infected municipalities (the selected model is in bold and *italic*).**

3

4

Covariates	Number of parameters	AIC
Age + T_R + Bait + Forest + Road + Num + Vacc+ Vacc* T_R	11	1201.8
Age + T_R + Bait + Forest + Road + Num + Vacc+ Vacc	9	1238.1
Age + T_R + Bait + Forest + Road + Vacc+ Vacc* T_R	10	1218.8
Age + T_R + Bait + Forest + Num + Vacc+ Vacc* T_R	10	1207.2
Age + T_R + Bait + Road + Num + Vacc+ Vacc* T_R	10	1201.5
Age + T_R + Forest + Road + Num + Vacc+ Vacc* T_R	10	1200.6
T_R + Bait + Forest + Road + Num + Vacc+ Vacc* T_R	10	1212.0
<i>Age + T_R + Road + Num + Vacc*T_R</i>	<i>9</i>	<i>1200.4</i>
Age + T_R + Road + Num + Vacc	7	1238.5
Age + T_R + Road + Vacc* T_R	8	1220.9
Age + T_R + Num + Vacc* T_R	8	1204.4
T_R + Road + Num + Vacc* T_R	8	1210.5

5

6

1 **Table IV: Comparison of Poisson models dealing with the duration of infection in**
 2 **infected municipalities (the selected model is in bold and *italic*).**

3

Covariates	Number of parameters	AIC
Num + Forest + Road + Bait + Vacc	7	411.7
Num + Forest + Road + Bait	5	480.5
Num + Forest + Road + Vacc	6	417.4
Num + Forest + Bait + Vacc	6	483.0
<i>Num + Road + Bait + Vacc</i>	6	411.3
Forest + Road + Bait + Vacc	6	684.4
Num + Road + Bait	4	478.4
Num + Road + Vacc	5	415.4
Num + Bait + Vacc	5	487.3
Road + Bait + Vacc	5	785.3

4

5

1 **Figure 1: The study area is located in north-eastern France at the border of the**
2 **Palatinate region (Germany).**

3

4 **Figure 2: Observed evolution of the proportion of immune and infected animals over the**
5 **whole study area.**

6

7 **Figure 3: Observed evolution of the proportion of immune animals over time (Seasons)**
8 **in uninfected municipalities.** Two seasons were considered spring/summer (Summer) and
9 autumn/winter (Winter).

10

11 **Figure 4: Predicted evolution of the proportion of infected animals over time according**
12 **to the three different vaccination treatments (early, contemporaneous, late).**

13

1 **CONFLICT OF INTEREST STATEMENT**

2

3 None

4

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