final report

C-strain vaccination against Classical Swine Fever: effects on epidemic and final screening

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Op het moment komt Klassieke Varkenspest (KVP) niet voor in Nederland en er wordt ook niet preventief gevaccineerd tegen de ziekte. Een insleep van het KVP virus vormt echter een voortdurende bedreiging voor de Nederlandse varkensstapel. In het huidige beleidsdraaiboek is opgenomen dat bij een uitbraak van KVP bij voorkeur noodvaccinatie in 2 km ringen rondom een gedetecteerd bedrijf ingezet zal worden als beheersmaatregel. Hierbij is gekozen voor een markervaccin (E2-subunit vaccin) waardoor onderscheid gemaakt kan worden tussen gevaccineerde en geïnfecteerde dieren met de bijbehorende E_{rns} ELISA test.

Nadelen van deze aanpak zijn dat (a) het E2-subunit vaccin een langzame immuunrespons opwekt (gevaccineerd bedrijf beschermd in ongeveer 10 dagen), (b) de E_{rns} ELISA een lage sensitiviteit heeft en (c) er geen goede confirmatietest beschikbaar is. Als alternatief vaccin zou het conventionele C-stam vaccin gebruikt kunnen worden. Dit vaccin induceert een zeer snelle immuunrespons in gevaccineerde dieren (gevaccineerd bedrijf beschermd in ongeveer 3 dagen), maar mist de markereigenschappen. In de eindscreening zullen geïnfecteerde dieren dus niet meer serologisch gedetecteerd kunnen worden. Wel kan met PCR vastgesteld worden of dieren al dan niet viruspositief zijn.

In het epidemiologisch deel binnen dit onderzoek worden de verschillen tussen noodvaccinatie met het C-stam vaccin of het E2-subunit vaccin geëvalueerd in de effectiviteit van bestrijding en de eindscreening. Met behulp van een KVP transmissiemodel zijn hypothetische epidemieën gesimuleerd, onder verschillende scenario's. De bestudeerde bestrijdingsstrategieën zijn: EU maatregelen (minimaal vereist: ruimen van gedetecteerde bedrijven, instellen van beschermings- en toezichtsgebieden, transportmaatregelen en het traceren en screenen van contacten), preventief ruimen in 1 km rond gedetecteerde bedrijven en noodvaccinatie met het E2-subunit vaccin of het C-stam vaccin in 1 km, 2 km of 3 km ringen. Berekend zijn de grootte en de duur van de gesimuleerde epidemieën, als maat voor de effectiviteit van de beheersmaatregelen. Om inzicht te krijgen in de gevolgen voor de eindscreening is op de gesimuleerde epidemieën een eindscreening(model) toegepast.

De resultaten van deze studie tonen aan dat voor KVP epidemieën die starten in een gebied met hoge veedichtheid zoals De Peel, de minimale maatregelen die door de EU worden voorgeschreven niet voldoende zijn om de epidemie te beheersen (Tab. 6). Als aanvullende maatregel, verminderen zowel preventieve ruiming als ringvaccinatie de duur en de omvang van de epidemie. Preventieve ruiming in 1 km verkort de duur van de epidemie het meest, maar dit gaat ten koste van het grootste aantal geruimde bedrijven (Tab. 6). Een vergelijkbare effectiviteit door middel van noodvaccinatie wordt alleen bereikt bij vaccinatie in 3 km. Daarbij is het C-stam vaccin slechts enigszins effectiever dan het E2-subunit vaccin.

Vaccinatie voorkomt infectie niet meteen na toedienen van het vaccin, maar kan de virusverspreiding wel vertragen en daarna stoppen door de toenemende bescherming. Door het lage aantal besmette dieren op het bedrijf kunnen sommige geïnfecteerde bedrijven ongedetecteerd blijven als noodvaccinatie gebruikt wordt ter bestrijding van KVP. De aantallen niet-gedetecteerde dieren na de eindscreening in het land zijn vergelijkbaar voor E2-subunit en C-stam vaccinatie (Tab. 8)

Tijdens de eindscreening kunnen ook vals positieve uitslagen gevonden worden. Dit zijn bedrijven die niet geïnfecteerd zijn (geweest) maar waar wel dieren gevonden worden die positief testen in de eerste eindscreening. Voor de non-vaccinatie strategieën (EU maatregelen en preventieve ruiming) zijn die aantallen verwaarloosbaar. Voor de C-stam vaccinatiestrategieën worden ook geen vals positieven verwacht. E2-subunit vaccinatie zal echter enige tientallen vals positieve bedrijven opleveren. De reden zijn de grote aantallen te bemonsteren dieren en de relatief lage specificiteit van de E_{rns} ELISA (Tab. 9). Deze bedrijven zullen nogmaals bezocht moeten worden om de tonsillen van de positieve dieren in de PCR te testen (die vervolgens negatief zal zijn).

Economisch deel

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In het economisch deel van het onderzoek zijn de verschillen in economische effecten van noodvaccinatie bij een uitbraak van Klassieke Varkenspest (KVP) in een veedichtgebied in Nederland onderzocht. Hierbij is een levend vaccin gebaseerd op de C-stam vergeleken met het vaccin dat op het moment als voorkeursvaccin genoemd wordt in de bestrijdingsdraaiboeken (het E2-subunit vaccin).

De resultaten laten zien dat bij toepassing van een "vaccination-to-live" strategie[1] :

Bij een vaccinatiecirkel van 2 km rond geïnfecteerde bedrijven is de schade lager dan bij vaccinatie in een cirkel van 1 km of 3 km voor zowel C-stam als E2-subunit vaccin

- Er zijn geringe verschillen tussen de vaccinatiestrategie met C-stam of met E2-subunit vaccin. Dit geringe verschil in het voordeel van C stam geldt alleen als er aan een belangrijke voorwaarden rond de afzet van producten van gevaccineerde producten is voldaan. Deze rand voorwaarden zijn:
    - Aanvullende hittebehandeling van producten is niet vereist (de huidige regelgeving maakt op het ogenblik deze hittebehandeling wel noodzakelijk);
    - Er hoeft tijdens de eindscreening van de gevaccineerde dieren maar 1 dier per hok van 10 dieren onderzocht te worden met de relatief dure PCR techniek;
    - Afnemers in het buitenland accepteren (producten van) niet gevaccineerde dieren uit een land waar met C-stam gevaccineerd wordt.

Op het ogenblik is het onzeker of de beperkte economische voordelen opwegen tegen de vele onzekerheden en beperkingen die met de introductie van het C-stam vaccin gepaard gaan.

In tabel S1 zijn de belangrijkste resultaten van de berekeningen samengevat.

Bij de berekeningen zijn alleen de kosten die mogelijk verschillen tussen de onderzochte strategieën meegenomen. In deze berekeningen zijn de handhavingskosten en de gederfde export inkomsten niet meegenomen.

[1] De gevaccineerde dieren worden pas geslacht op het einde van hun normale productiecyclus en hun producten worden gekanaliseerd binnen Nederland afgezet.

Tabel S1 Kosten van de verschillende controle strategieën in het geval van een uitbraak van KVP in een varkensdicht gebied in Nederland (in Miljoen € per uitbraak)

	Controle kosten			Kosten veehouders			Totaal		
		Percentiel			Percentiel			Percentiel	
	Mediaan	**0,05**	**0,95**	**Mediaan**	**0,05**	**0,95**	**Mediaan**	**0,05**	**0,95**
vacE2_1km	35,63	12,13	74,10	12,77	2,57	29,80	48,40	15,40	103,82
vacE2_2km	27,59	11,18	50,33	17,64	4,93	35,36	45,23	16,56	84,85
vacE2_3km	25,02	11,05	44,68	23,12	7,65	44,60	48,15	19,66	87,85
vacC_1km*	32,40	11,91	62,05	10,99	2,63	25,16	43,40	14,70	85,90
vacC_2km*	26,75	11,43	48,32	15,93	4,44	31,03	42,68	16,73	77,96
vacC_3km*	25,21	11,14	42,20	21,03	6,90	39,14	46,24	19,31	80,07
vacC_1kmcul*	75,97	21,73	158,68	9,84	1,65	26,88	85,81	23,76	184,88
vacC_2kmcul*	108,15	34,20	204,48	11,80	2,32	28,51	119,95	36,76	231,53
vacC_3kmcul*	140,58	50,76	256,40	14,20	3,19	32,71	154,78	53,64	287,59

Summary

The Netherlands is currently free of Classical Swine Fever (CSF) without the use of preventive vaccination. However, an introduction of the CSF virus poses a continuous threat to the naive pig population. The Dutch contingency plan states that in the case of a CSF outbreak, emergency vaccination in 2 km rings around detected farms is preferred as additional control measure. Using a marker vaccine (E2-subunit vaccine), vaccinated animals can be distinguished from infected animals using the associated E_{rns} ELISA test.

Concerns regarding this approach are that (a) the E2-subunit vaccine induces a slower immune response in the animal than conventional vaccines, (b) the E_{rns} ELISA for vaccinated animals has a lower sensitivity than the E2 ELISA's for unvaccinated animals and (c) no proper confirmation test is available for E2-subunit vaccinated animals. As an alternative, the conventional C-strain vaccine has been proposed, that induces a faster immune response in the animal but lacks the marker properties. During the final screening, infected C-strain vaccinated animals cannot be detected serologically. Instead, they are tested for virus positivity using PCR.

In this project it is evaluated how the use of C-strain vaccine instead of E2-subunit vaccine will affect the effectiveness of controlling the epidemic. To this end a CSF transmission model was developed that describes virus transmission on three different levels: between animals, between pens and between herds. The results of transmission and vaccination experiments as well as the data from the 1997/1998 CSF epidemic in The Netherlands serve to parameterize the model. With the model hypothetical epidemics are simulated under different scenarios. The studied control strategies include EU measures (depopulation of detected herds, defining protection and surveillance zones, transport regulations, and tracing and screening of dangerous contacts), preemptive culling in 1 km around detected herds, and emergency vaccination with the E2-subunit or C-strain vaccine in 1 km, 2 km or 3 km rings. The size and duration of the simulated epidemics are a measure for the effectiveness of the control strategies. On the simulated epidemics a final screening (model) is applied, to assess how many herds and animals need to be tested, how many truly positive herds and animals will be detected during the final screeing and how many false positive results can be expected.

The results of this study show that for CSF epidemics starting in a densely populated livestock area such as De Peel, the minimal measures required by the EU are not sufficient to control the epidemic (Tab. 6). As additional measure, both preemptive culling and ring vaccination decrease the duration and size of the epidemic. Preemptive culling shortens the epidemic the most with the smallest control radius of 1 km, at the expense of the largest number of depopulated farms (Tab. 6). Similar effectiveness is only achieved by emergency vaccination when the control radius is 3 km. In this respect, C-strain vaccination is only slightly more effective than E2-subunit vaccination. Apparently the time scale of the immune response (3 or 10 days) is sufficiently small compared to the virus transmission within a herd (4 to 5 weeks) and between herds to effectively control the epidemic.

Vaccination does not prevent infection right after administering the vaccine, but it can slow down and eventually halt virus transmission in an infected herd due to the increasing protection. Because of the small number of infected animals, some infected herds may escape detection when vaccination is used to control CSF. E2-subunit vaccination is expected to yield larger numbers (medians of 26 - 38) of not-detected infected animals in the country after the epidemic than C-strain vaccination (medians of 19 - 28) (Tab. 8). During the final screening, infected E2-subunit vaccinated animals can be detected via the E_{rns} ELISA. Even though the test sensitivity is not very high, the sample sizes (1 animal per pen, or 10% of all animals) are sufficiently large to detect even a low seroprevalence. However, the subsequent confirmation test by PCR has a sensitivity that decreases over time: 80 days after infection only half of the animals will be PCR positive. For this reason, it is not uncommon that an infected herd is detected by E_{rns} ELISA, but then declared free of disease when the PCR turns out to be negative. The model results show that only 0, 1 or 2 infected herds are detected during the final screening. When C-strain vaccination is applied, another screening protocol is followed. Serological tests are not applicable, as they will always be positive for C-strain vaccinated animals. Instead, blood samples (of 1 animal per pen, or 10% of all animals) are tested by PCR. However, as virus will be present in the blood of C-strain vaccinated animals for a very short period after infection, none of them will be detected by PCR during the final screening. This is why after the final screening, the numbers of not-detected infected animals in the country are comparable for E2-subunit and C-strain vaccination (Tab. 8).

During the final screening test results can also be false positive. For the non-vaccination strategies (EU measures and preemptive culling) these numbers are negligible, because of the relatively small sample sizes from unvaccinated herds and the high test specificity of the (series of) serological tests. Also for the C-strain vaccination strategies, no false

positive results are expected using PCR testing. E2-subunit vaccination on the other hand will yield several dozens of false positive herds, because of the large sample sizes from vaccinated herds and the relatively low test specificity of the E_{rns} ELISA (Tab. 9). These herds will be visited again to collect the tonsils of the positively testes animals for PCR testing (which will yield negative results).

C-strain vaccination is less effective than was expected from the experimental results at individual level. The fast immune response induced by the C-strain vaccine will prevent most herds to be infected after vaccination. But when an already infected herd is vaccinated, the same fast immune response will soon halt the virus transmission and prevent detection. This is why C-strain vaccination will also yield undetected infected herds. The undetected infected animals in these herds cannot be detected during the final screening, as they cannot be distinguished from vaccinated animals.

1. Introduction

Classical Swine Fever (CSF) represents a continuous threat to pig populations that are free of disease without vaccination. When CSF virus is introduced, the minimal control strategy imposed by the EU is often insufficient to mitigate the epidemic. Preemptive culling as an additional control measure encounters ethical objections and has been abandoned in the updated Dutch contingency plan (Contingency plan Classical Swine Fever, 2007). Currently, the preferred additional measure is emergency vaccination using the E2-subunit vaccine in 2 km rings. Animals vaccinated with this marker vaccine can be distinguished from infected animals using the associated E_{rns} ELISA. The main concerns regarding this approach are that (a) the E2-subunit vaccine induces a slower immune response in the animal than conventional vaccines, (b) the E_{rns} ELISA for vaccinated animals has a lower sensitivity than the E2 ELISA's for unvaccinated animals and (c) no proper confirmation test is available for E2-subunit vaccinated animals. For these reasons, it has been proposed to use the conventional C-strain vaccine, that induces a fast immune response in the animal (Dewulf et al., 2004). Without the marker properties, infected C-strain vaccinated animals cannot be detected serologically, as is required for regaining the freedom of disease status (Council Directive 2001/89/EC). Instead, they are to be tested by PCR that can detect CSF virus fragments. Arguing that PCR negative animals will not pose a problem for re-emergence of the virus (even if they would have been infected), it might be accepted as a valid strategy to regain the freedom of disease status.

To determine whether C-strain vaccination is a valid alternative to E2-subunit vaccination, it must first be evaluated how the use of C-strain vaccine instead of E2-subunit vaccine will affect the effectiveness of control measures and the results of the final screening. Here we will answer these questions using a CSF transmission model to assess the effectiveness of several vaccination strategies, and a final screening model to assess the effect on the number of not-detected infected herds and animals.

2. Methods

2.a CSF transmission model

A stochastic individual-based model was previously developed describing virus transmission between animals, pens and farms (Bergevoet et al., 2007, Backer et al., 2009). Here the within-herd model is further adapted to include three pig types (finishers, piglets, sows; Tab. 1). The detection model is improved by imposing a detection limit on the number of infectious animals, that reproduces the detection times that were observed in the 1997/1998 CSF epidemic in the Netherlands (rather than drawing a random detection time). The between-herd model is altered to accommodate herds of varying population sizes and mixed farms containing more than one pig type.

The effect of the E2-subunit vaccine on the transmission of CSFV was modelled by separate effects on the susceptibility, infectiousness and infectious period of a vaccinated animal. For the C-strain vaccine however, infectiousness and infectious period were unaffected in vaccinated animals (based on limited experimental data), so only the effect on the susceptibility of vaccinated animals is modelled (Tab. 2 and Appendix A).

Table 1 Transmission parameters between animals and pens, for different herd types

parameter	finishing pigs	piglets	sows
latent period, T_{lat}	4 days	4 days	4 days
infectious period, $T_{inf,0}$	15 days (7 – 25)*	15 days (7 – 25)	15 days (7 – 25)
reproduction number, R_0	15.5	100	2.8
transmission rate parameter, β_0	1.03 day^{-1}	6.67 day^{-1}	0.187 day^{-1}
number of animals per pen, N_{pen}	10	10	1
total number of animals, N	variable	variable	variable
within-herd reproduction number, R	2.8	2.8	2.8
reduction factor between pens, ε	0.018	$2.8 \cdot 10^{-3}$	1
detection limit (# infectious animals)	12	23	26

* Between brackets the (5% - 95%) interval

Table 2 Effect of vaccination on CSFV transmission, as function of period τ between vaccination and infection (see Appendix A for figures).

effect of: / on:	E2-subunit vaccine	C-strain vaccine
susceptibility	Min[1, exp(-0.2(τ-6.4))]	Min[1,exp(-0.86 τ)]
infectiousness (day^{-1})	β_0 Min[1,exp(-0.5(τ-6.4))]	β_0
average infectious period (days)	$T_{inf,0}$ Min[1,(0.21(τ-6.4)+1)$^{-1}$]	$T_{inf,0}$

The transmission between herds was modelled as a function $k(r)$ of the distance r between source and destination herd: $k(r) = k_0 / (1+(r/r_0)^{\alpha})$ (Fig. 1). The transmission kernel parameters were estimated as r_0 = 1.0 km, α = 2.2, and k_0 = 0.0011 day^{-1} from the 1997/1998 CSF epidemic in the Netherlands (Boender et al., 2008, Backer et al., 2009).

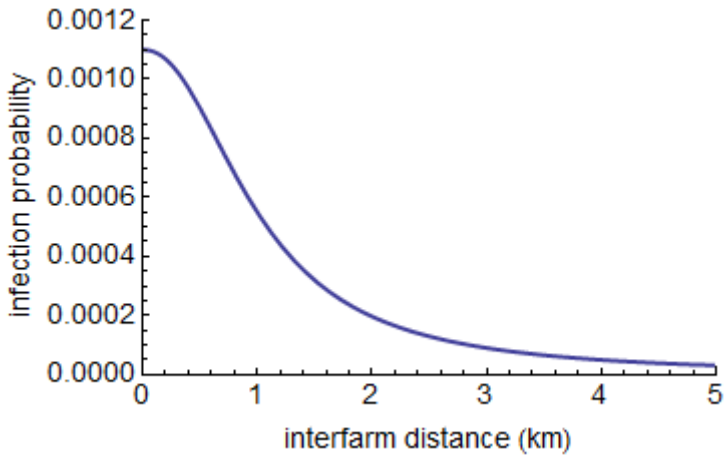

Figure 1 Transmission kernel describing the infection probability that a source herd will infect a susceptible herd as function of the interfarm distance.

2.b Final screening model

All farms in the protection zones of 3 km around detected herds, need to be tested during the final screening. Sample sizes are taken based on the type of herd and vaccination status. The final screening model calculates how many infected not-detected herds and animals can be expected after the final screening, how many cases (truly positive herds) are expected to be found during the final screening (setting back the freedom of disease status by at least 30 days) and how many herds are expected to be declared initially false positive in the final screening.

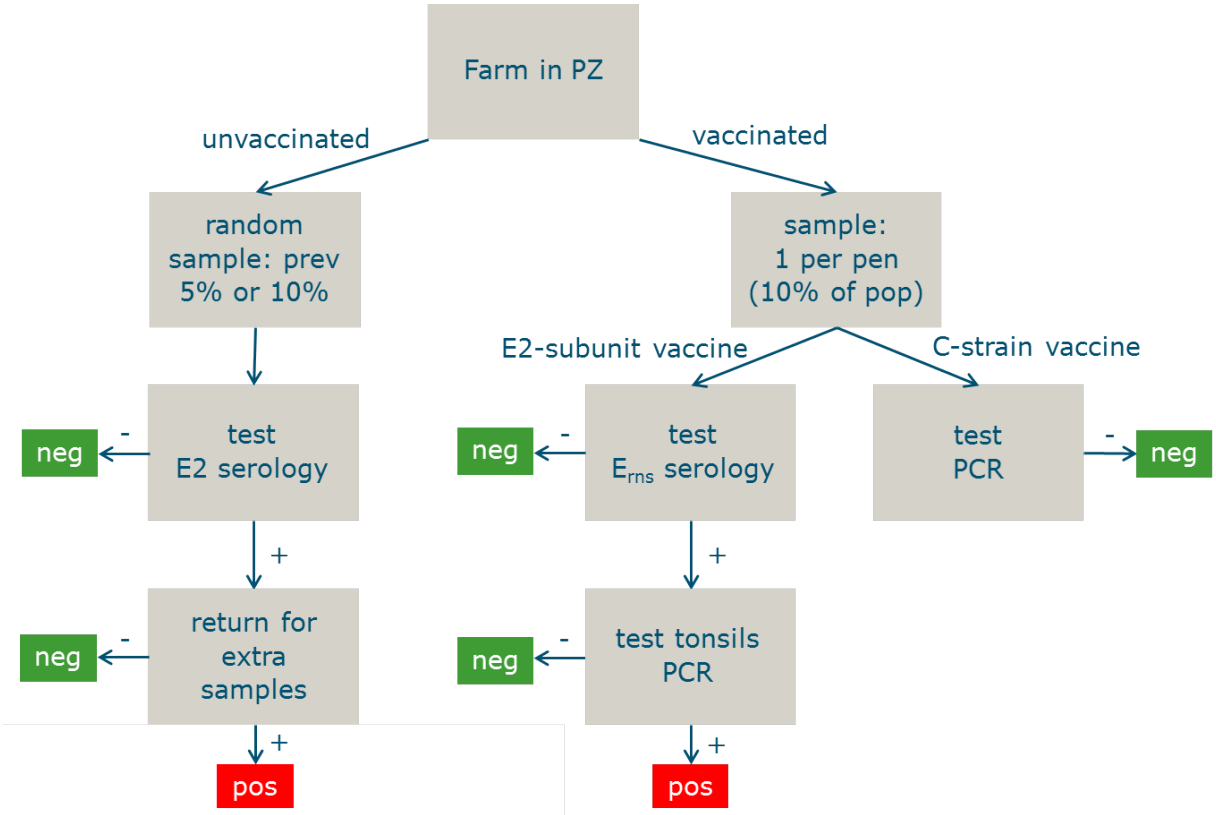

Figure 2 Schematic overview of final screening model (PZ = protection zone).

The final screening is modelled to take place after the simulated epidemic has ended (Fig. 2). All not-detected infected animals are assumed to have developed a serological response, and to be non-viraemic. Unvaccinated animals in the protection zone (3 km around IP's) will be sampled according to EU regulations and tested by E2 ELISA. Vaccinated animals will be more intensely screened and tested with either an E_{rns} ELISA (for E2-subunit vaccinated animals) or PCR (for C-strain vaccinated animals and as confirmation test for E2-subunit vaccinated animals). The test characteristics and sampling strategy vary for the different tests (Tab. 3 and Appendix B).

Table 3 Different tests, test characteristics at animal level and sampling strategies to be used in the final screening.

test	characteristics		applied to	sample size
	sens	spec		
E2 serology	92%	99.999%	unvaccinated finishers unvaccinated piglets unvaccinated sows	$n_{sample}(0.10,0.95)$ $n_{sample}(0.10,0.95)$ $n_{sample}(0.05,0.95)$
E_{rns} serology	73.4%	99.9%	E2-subunit vacc finishers E2-subunit vacc piglets	1 per pen 1 per pen
confirmation PCR	$se^{PCR}(\tau)$*	100%	E2-subunit vacc finishers E2-subunit vacc piglets	1 per pen 1 per pen
PCR	0%**	100%	C-strain vacc finishers C-strain vacc piglets	1 per pen 1 per pen

* PCR sensitivity in E2-subunit vaccinated animals decreases with time τ since infection.

** C-strain vaccinated animals are PCR negative (for the wild virus) soon after infection. At final screening none of them will be detected.

with:

$$n_{\text{sample}}\left(q, se_{\text{herd}}\right) = \min_{n}\left[1 - \sum_{i=0}^{\min[n,\lfloor qN \rfloor]}\left[\frac{\binom{\lfloor qN \rfloor}{i}\binom{N-\lfloor qN \rfloor}{n-i}}{\binom{N}{n}}(1-se)^{i}\right] \geq se_{\text{herd}}\right]$$

where q is the design prevalence, se_{herd} the desired herd sensitivity, N the herd size and se the test sensitivity. For instance, from an unvaccinated finishing herd of 1000 animals, 31 are randomly selected to be tested.

First, the final screening model calculates the probability p^{+}_{neg} of missing an infected herd, depending on whether a random sample (for E2 serology) or one animal per pen (for E_{rns} serology) is tested (Tab. 4).

Table 4 Calculation of final screening results per herd based on the probability p^{+}_{neg} of declaring an infected herd false negative, for different test regimes: the number S_{after} of infected not-detected animals after final screening, the probability p_{case} of finding a truly positive herd during the final screening and the probability p^{-}_{pos} of finding one false positive animal in a truly negative herd.

test	p^{+}_{neg}	S_{after}	p_{case}	p^{-}_{pos}
E2 serology	$\sum_{i=0}^{\min[n,S]}\left[\frac{\binom{S}{i}\binom{N-S}{n-i}}{\binom{N}{n}}(1-se)^{i}\right]$	$S\ p^{+}_{\text{neg}}$	$1-p^{+}_{\text{neg}}$ if S>1 0 if S=1	$1-sp^{n}$
E_{rns} serology	$\prod_{i=1}^{N/m}\left[\frac{m-s_i}{m}+\frac{s_i}{m}(1-se)\right]$	$S\ p^{+}_{\text{neg}}$ + $(S-S/m)$ $(1-p^{+}_{\text{neg}})$ $(1-se^{\text{PCR}}(\tau))$	$(1-p^{+}_{\text{neg}})$ $se^{\text{PCR}}(\tau)$	$1-sp^{n}$
PCR	1	0	0	0

with sample size n, herd size N, pen size m, total number of infected animals S in the herd, and their distribution s over the pens (i.e. $\Sigma s=S$), test sensitivity se, test specificity sp, and PCR sensitivity $se^{\text{PCR}}(\tau)$ depending on time τ since infection.

The expected number S_{after} of not-detected infected animals in the herd after the final screening, is found by multiplying p^{+}_{neg} by the number of infected animals S before final screening. When a truly positive herd is found, with probability $1-p^{+}_{neg}$, teams will return to the herd for additional sampling and testing. When in unvaccinated herds (using E2 serology) evidence of clustering of positive animals or spread is found, the herd will be declared a case. In the model this will happen when the total number of infected animals S is larger than one. It should be kept in mind that not-vaccinated not-detected infected herds outside the protection zone of 3 km, will not be tested and thus not detected during the final screening.

In E2-subunit vaccinated herds that were found positive in the E_{rns} ELISA, the additional sampling is done by euthanizing the positively tested animals and testing their tonsils in PCR. Only when virus fragments are detected, the herd will be declared a case. However, the chance of finding a positive PCR result decreases with time since infection (Appendix B). This is modelled by a time-dependent PCR sensitivity at animal level $se^{PCR}(\tau) = 1/(1+\exp(0.088\,\tau - 7.4))$, depending on time τ since infection. For simplicity we assume that only one animal (infected half way between the infection and clearance time of the herd) is positive when the E_{rns} ELISA is positive. When PCR results are negative, the infected animals in the sample (on average S/m) will have been euthanized for the PCR testing, but the remainder (on average $S-S/m$) will be left in the herd, which will add to the number S_{after} of not-detected infected animals after the final screening. Also, the probability of not-detecting the infected herd p^{+}_{neg} is increased with $(1-p^{+}_{neg})\,(1-se^{PCR}(\tau))$, i.e. the probability that the herd was positive in E_{rns} ELISA but negative in PCR.

The probability p^{-}_{pos} of testing a not-infected herd (falsely) positive depends on the test specificity and the number of sampled animals. All positive test results will be followed by additional sampling and testing, assumed to lead to a 100% specificity. This means that p^{-}_{pos} indicates the probability to *initially* test a herd false positive (and later to be found negative).

Summing S_{after} and p^{+}_{neg} ($+(1-p^{+}_{neg})\,(1-se^{PCR}(\tau))$ for E2-subunit vaccinated herds) over all infected herds gives the expected total number of not-detected infected animals and herds in the country after the final screening and summing p_{case} over all infected herds gives the expected number of truly positive herds found during the final screening. The expected total number of herds that are initially tested false positive is the summation of p^{-}_{pos} over all not-infected herds in the protection zones (3 km around IP's).

2.c Commercial pig farm data 2011

Dienst Regelingen (Ministry of Economic Affairs) provided pig transport data containing the number of pigs that were dispatched from each farm in 2011, in several pig categories. From these transport data, the average number of finishers, piglets and sows were calculated (Appendix C). Discarding the herds with less than 100 finishers, 100 piglets or 50 sows, the pig farm database contains 7018 farms (Tab. 5), distributed over The Netherlands (Fig. 3).

Table 5 Pig farm data for The Netherlands in year 2011 (calculated from animal transport data, Dienst Regelingen)

	# herds	# animals	herd size	
farms with		(x 1000)	median	(5% – 95%)
finishing pigs	5688	6479	1139	(136 – 3844)
piglets	2629	4517	1718	(211 – 4980)
sows	2425	945	390	(77 – 1087)
total	10742	11941		

The total number 10742 of herds with finishing pigs, piglets and sows is not the same as the total number of 7018 farms, because a farm can contain multiple herd types (e.g. sows and piglets).

The yearly survey (Land- en tuinbouwcijfers, 2012) in 2011 reported 12,4 million animals in 6526 pig farms. However, these numbers are based on available stable spaces rather than transports, and they reflect the situation at April 1st rather than the yearly average. For these reasons the reported numbers in the yearly survey and Table 5 are not the same.

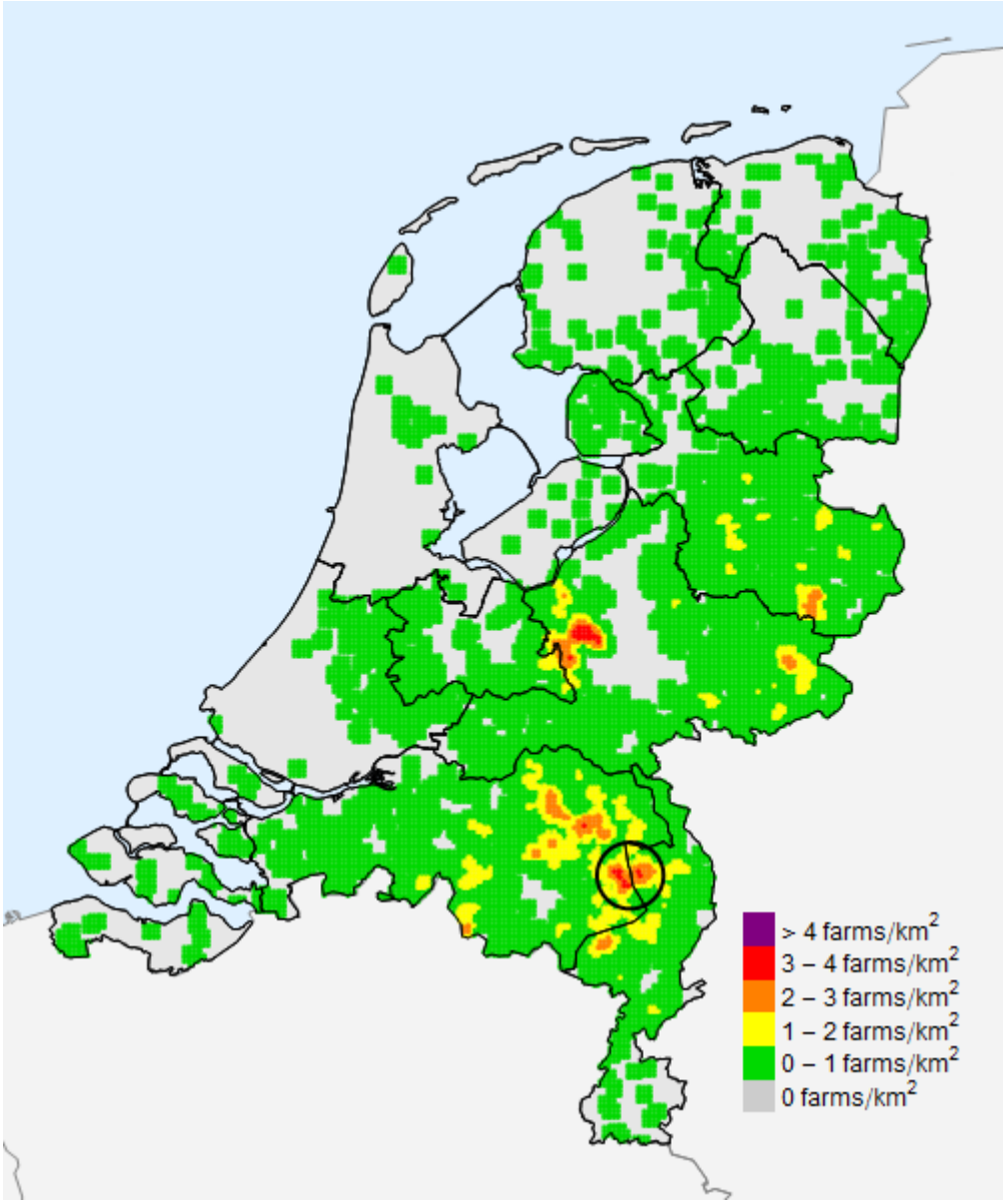

Figure 3 Map of commercial pig farm densities in The Netherlands (year 2011). The simulated epidemics start in De Peel (black circle), a densely populated livestock area.

2.d Simulations

To compare different control strategies, 1000 artificial High Risk Periods (HRP) were generated, by choosing an index herd in De Peel (black circle in Fig. 2), doubling the kernel height (Fig. 1), and requiring that:

- 5 farms are infected at the end of the HRP which lasts 5 weeks (default HRP), or
- 10 farms are infected at the end of the HRP which lasts 6 weeks (extreme HRP)

During the CSF epidemic in 1997/1998, it was estimated that 19 herds were infected at the moment the first case was detected (followed by a transmission boost before transport was officially prohibited, presumably infecting another 19 herds). This is generally considered an extreme HRP. With the increased hygiene, changes made in the production chain and decreasing farm numbers, we will choose 10 farms infected at the end of the HRP as the extreme scenario for the current situation. Half of this number – 5 farms infected at the end of the HRP – is considered to be the default scenario.

Upon detection of the first case, EU measures (strategy *EU*) will be applied: culling of infected premises (IP), transport regulations in protection (3 km around IP's) and surveillance (10 km around IP's) zones, as well as tracing and screening of dangerous contacts. When additional measures are applied, preemptive culling in 1 km around IP's will be applied during the first 5 days. After this period, 1 km preemptive culling either continues (strategy *cul_1km*) or is replaced by emergency vaccination in 1 km, 2 km or 3 km rings around IP's. For vaccination either the E2-subunit vaccine (strategies *vacE2_1km*, *vacE2_2km* and *vacE2_3km*) or the C-strain vaccine (strategies *vacC_1km*, *vacC_2km* and *vacC_3km*) is used.

The time between detection and culling of an IP is assumed to be one day. The time between culling of an IP and emergency vaccination or preemptive culling of herds in the control zone is also assumed to be one day. Culling and vaccination are assumed to be not limited by resources (i.e. vaccine doses, control teams, destruction capacity).

3. Results

Each control strategy is applied to each of the 1000 default and 1000 extreme HRP's. For each simulated epidemic the following characteristics are registered:

- duration of epidemic (time between first and last detection)
- number of detected farms
- number of preemptively culled farms
- number of vaccinated farms
- number of not-detected herds (before final screening)
- number of not-detected infected animals (before final screening)
- number of truly positive herds found (during final screening)
- number of not-detected herds (after final screening)
- number of not-detected infected animals (after final screening)
- number of tested herds (in 3 km protection zones)
- number of tested animals (in 3 km protection zones)
- number of initially false positive tested herds

The final screening model is only applied to the default HRP's.

3.a Effectiveness of control strategies for default HRP's

The effectiveness of the different control strategies is evaluated for the default HRP's where 5 farms are infected at the end of the HRP which lasts 5 weeks (Tab. 6).

The EU control strategy yields the longest and largest epidemics, although not nearly as long and large as the 1997/98 CSF epidemic in the Netherlands. Preemptive culling considerably shortens the epidemic but leads to a much larger number of depopulated farms. In the emergency vaccination strategies 8 (2-20) farms are preemptively culled during the first 5 days, after which farms are vaccinated in 1, 2, or 3 km rings around detected farms with the E2-subunit or C-strain vaccine. For both vaccines an increasing control circle decreases the epidemic duration and number of detected herds, with an accordingly increasing number of vaccinated herds. With identical control radius, the C-strain vaccination strategies are more effective than the E2-subunit vaccination strategies, as was expected from the higher effectiveness of the C-strain vaccine at individual level. But surprisingly, the differences are not very large: C-strain vaccination leads to around 2 detected farms less and ends the epidemic around 12 days earlier. Emergency vaccination (with either vaccine) in 3 km rings around detected farms is more or less as effective as 1 km preemptive culling.

The required depopulation capacity is highest for 1 km preemptive culling, at a median of 5 farms or 10000 animals per week (see Appendix D for figures), in the early stages of the epidemic. In the vaccination strategies (regardless of the vaccine), the 1 km strategies require a median of 5 farms or 10000 animals per week to be vaccinated, the 2 km strategies 10 farms of 30000 animals per week and the 3 km strategies 20 farms or 50000 animals per week (Appendices D and E). However, in the most extreme cases (95th percentiles) these numbers increase with a factor 5.

Table 6 Effectiveness of control strategies for default HRP's: epidemic duration, number of detected, preemptively culled and vaccinated farms and animals per epidemic; median values and (5% - 95%) interval between brackets.

control strategy	duration* (days)		number of detected farms		number of preemptively culled farms		number of vaccinated farms		number of culled animals (x1000)		number of vaccinated animals (x1000)	
EU	202	(65 - 475)	40	(10 - 128)	0	(0 - 0)	0	(0 - 0)	115	(27 - 363)	0	(0 - 0)
cul_1km	93	(35 - 199)	13	(5 - 27)	79	(26 - 173)	0	(0 - 0)	225	(72 - 488)	0	(0 - 0)
vacE2_1km	136	(45 - 310)	19	(6 - 52)	8	(2 - 20)	98	(20 - 254)	79	(30 - 166)	213	(42 - 562)
vacE2_2km	113	(37 - 236)	14	(5 - 32)	8	(2 - 20)	191	(56 - 414)	64	(25 - 123)	422	(114 - 884)
vacE2_3km	104	(34 - 203)	12	(5 - 26)	8	(2 - 20)	278	(95 - 588)	58	(25 - 113)	599	(203 - 1241)
vacC_1km	125	(42 - 264)	16	(5 - 41)	8	(2 - 20)	88	(19 - 223)	71	(28 - 145)	195	(42 - 483)
vacC_2km	101	(36 - 208)	12	(5 - 27)	8	(2 - 20)	175	(51 - 379)	59	(25 - 110)	388	(103 - 802)
vacC_3km	91	(31 - 185)	10	(4 - 22)	8	(2 - 20)	260	(83 - 515)	53	(22 - 100)	557	(174 - 1088)

* Duration of the epidemic is defined as the time between the first and the last detection.

3.b Effectiveness of control strategies for extreme HRP's

The effectiveness of the different control strategies is also evaluated in a more extreme starting situation where 10 farms are infected at the end of the HRP after 6 weeks (Tab. 7).

The more extreme starting situation leads to longer and larger epidemics than the default HRP starting situation (Tab. 6). The differences are however not large and the 95th percentiles are in fact comparable, showing that large epidemics are not only dependent on the starting situation, but can also develop during the control period due to stochasiticity. The comparison between the different control strategies is identical to the comparison made in the previous section for the default starting situations. So, the ranking of the strategies based on their effectiveness seems to be robust for the studied range.

Table 7 Effectiveness of control strategies for extreme HRP's: epidemic duration, number of detected, preemptively culled and vaccinated farms and animals per epidemic; median values and (5% - 95%) interval between brackets.

control strategy	duration* (days)		number of detected farms		number of preemptively culled farms		number of vaccinated farms		number of culled animals (x1000)		number of vaccinated animals (x1000)	
EU	226	(70 - 469)	51	(11 - 136)	0	(0 - 0)	0	(0 - 0)	143	(29 - 388)	0	(0 - 0)
cul_1km	110	(51 - 202)	22	(10 - 42)	134	(65 - 248)	0	(0 - 0)	382	(181 - 693)	0	(0 - 0)
vacE2_1km	160	(69 - 301)	34	(14 - 65)	11	(3 - 26)	165	(62 - 311)	128	(62 - 219)	362	(124 - 674)
vacE2_2km	127	(63 - 244)	25	(12 - 45)	11	(3 - 26)	310	(130 - 546)	104	(53 - 175)	670	(277 - 1157)
vacE2_3km	115	(56 - 212)	21	(11 - 36)	11	(3 - 26)	420	(208 - 735)	91	(50 - 146)	895	(451 - 1535)
vacC_1km	150	(71 - 287)	31	(13 - 60)	11	(3 - 26)	156	(58 - 298)	119	(59 - 203)	342	(118 - 654)
vacC_2km	120	(61 - 208)	22	(11 - 38)	11	(3 - 26)	284	(130 - 497)	93	(49 - 157)	616	(282 - 1061)
vacC_3km	103	(54 - 188)	18	(9 - 30)	11	(3 - 26)	395	(198 - 678)	82	(43 - 134)	839	(422 - 1424)

* Duration of the epidemic is defined as the time between the first and the last detection.

3.c Detection of infected herds during final screening

During the (simulated) epidemics some infected herds may not be detected, because the number of infectious animals stays under the detection limit, either by vaccination or by chance. The final screening that takes place at least 30 days after the last detection or vaccination event, is designed to detect the infected animals in these herds. The results of the final screening (Tab. 8) are obtained by applying the final screening model to the simulated epidemics with default HRP's (Tab. 6).

Table 8 Final screening of infected herds per control strategy (default HRP's): expected number of not-detected herds and animals before and after final screening and expected number of truly positive herds (cases) found during the final screening; median values and (5% - 95%) interval between brackets.

control strategy	number of not-detected herds per epidemic			number of not-detected animals per epidemic	
	before final screening	after final screening	declared as case during final scr.	before final screening	after final screening
EU	1 (0 - 4)	1 (0 - 3)	0 (0 – 0.4)	1 (0 - 12)	1 (0 - 10)
cul_1km	0 (0 - 1)	0 (0 - 1)	0 (0 - 0)	0 (0 - 2)	0 (0 - 2)
vacE2_1km	3 (0 - 10)	2 (0 - 9)	0.2 (0 – 1.6)	26 (0 - 124)	18 (0 - 101)
vacE2_2km	3 (0 - 10)	2 (0 - 9)	0.7 (0 – 2.3)	37 (0 - 129)	21 (0 - 106)
vacE2_3km	3 (0 - 10)	2 (0 - 9)	0.7 (0 – 2.5)	38 (0 - 127)	21 (0 - 99)
vacC_1km	3 (0 - 10)	3 (0 - 9)	0 (0 - 0)	19 (0 - 78)	19 (0 - 78)
vacC_2km	4 (0 - 10)	4 (0 - 10)	0 (0 - 0)	24 (0 - 77)	24 (0 - 77)
vacC_3km	4 (0 - 10)	4 (0 - 10)	0 (0 - 0)	28 (0 - 78)	28 (0 - 78)

The control strategies differ in how many infected herds escape detection. The non-vaccination strategies yield the least number of not-detected herds and animals. These numbers are greater for EU measures than for 1 km preemptive culling, because the epidemics under EU measures are much larger. Most not-detected not-vaccinated herds are sow herds with only one or a few infected animals. Because of this low seroprevalence, the sample sizes (designed to detect a seroprevalence of at least 5% in sow herds) are too small to detect these animals. Moreover, around 20% of the infected herds (for *EU*) are located outside the protection zone and not tested during the final

screening. For these reasons, the final screening has no (for *cul1*) or only limited (for *EU*) effect on the number of not-detected infected herds and animals after the final screening.

Before the final screening, the vaccination strategies yield considerable numbers of not-detected infected herds (0 to 10, with median of 3 or 4 herds), irrespective of the control radius or vaccine used. The C-strain vaccine may protect herds sooner, but when an infected herd is vaccinated, the C-strain vaccine will almost certainly prevent detection. The numbers of not-detected infected animals for the C-strain vaccination strategies are smaller than for the E2-subunit vaccination strategies, but they will not be detected during the final screening. This is because at the time of final screening, none of the infected C-strain vaccinated animals will be positive in the PCR test and there is no alternative test to distinguish them from not-infected C-strain vaccinated animals.

For the E2-subunit vaccinated animals, the E_{rns} ELISA does make a distinction between infected and non-infected animals. The large sample size taken on vaccinated farms (10% of all animals) compensates for the poor test sensitivity (of 73.4%, Tab. 3), leading to a reasonable chance of detection in the first ELISA stage. However, during the following confirmation stage with PCR, many herds – especially those infected early in the epidemic – will be tested negative by PCR and still escape detection. For this reason, the number of not-detected infected herds and animals is only moderately reduced by the final screening.

All in all, the final screening in the E2-subunit vaccination strategies reduces the numbers of not-detected animals to levels comparable to the C-strain vaccination strategies. The numbers of not-detected herds in the E2-subunit vaccination strategies are even brought below the levels of the C-strain vaccination strategies by the final screening.

3.d Number of tested and false positive herds during final screening

All farms in the protection zones of 3 km around detected herds, need to be tested during the final screening. Sample sizes are taken based on the type of herd and vaccination status. Most of the farms are not infected, but the test results can still point at a false positive result due to the non-perfect specificity of the ELISA's (Tab. 3).

The smallest number of herds and animals need to be tested in the 1 km preemptive culling strategy (Tab. 9), because of the small epidemics and small sample sizes in unvaccinated herds. The largest number of herds are tested for the *EU* control strategy because of the large epidemics affecting the largest geographical area. The number of animals to be tested though, is still moderate compared to the vaccination strategies.

Obviously, the number of vaccinated herds to be tested increases with increasing control radius, while the number of not-vaccinated herds to be tested decreases. For the 3 km strategies the vaccination zones and protection zones coincide, so the non-vaccinated herds that are tested are all sow herds. The number of animals to be tested in vaccinated herds is much larger than in not-vaccinated herds, because one animal per pen is tested (10% of the total population).

Finding a false positive in the non-vaccinated herds (using E2 serology) is very rare, because sample sizes are limited and the test specificity is high (99.999% at animal level). For C-strain vaccinated herds, false positive results do not occur because of the assumed 100% specificity of the PCR test. Considerable numbers of false positives are expected for the E2-subunit vaccinated herds that are tested by the E_{rns} ELISA. The large sample sizes per herd combined with the non-perfect specificity (of 99.9% at animal level), diminish the probability of finding a tested herd truly negative. For instance, a not-infected E2-subunit vaccinated herd of 5000 finishers of which 500 are tested, has a $1 - 0.999^{500} = 0.39$ probability of being falsely identified as infected. The subsequent PCR testing will confirm the herd to be negative, but this involves a considerable amount of extra work (returning to herd, euthanizing the positive reactors, taking tonsil samples, testing samples in PCR). It is expected that 15% (13%–17%) of the E2-subunit vaccinated herds are initially declared false positive.

Table 9 Number of tested herds and animals in the final screening and expected number of initially false positive herds per control strategy; median values and (5% - 95%) interval between brackets. Non-vaccinated herds are tested by E2 serology, E2-subunit vaccinated herds by E_{rns} serology (+ confirmation by PCR) and C-strain vaccinated herds by PCR.

control strategy	non-vaccinated						vaccinated					
	number of tested herds		number of tested animals (x1000)		number of initially false positive herds		number of tested herds		number of tested animals (x1000)		number of initially false positive herds	
EU	816	(312 - 1716)	31	(12 - 65)	0.3	(0.1 – 0.6)						
cul_1km	352	(135 - 665)	13	(5 - 25)	0.1	(0.1 – 0.3)						
vacE2_1km	445	(175 - 963)	18	(7 - 38)	0.2	(0.1 – 0.4)	116	(24 - 293)	20	(4 - 52)	17	(3 - 44)
vacE2_2km	274	(113 - 526)	12	(5 - 23)	0.1	(0.0 – 0.2)	228	(67 - 485)	41	(11 - 85)	34	(10 - 72)
vacE2_3km	128	(55 - 247)	7	(3 - 14)	0.1	(0.0 – 0.1)	336	(114 - 699)	59	(20 - 120)	50	(17 - 102)
vacC_1km	429	(159 - 872)	17	(6 - 35)	0.2	(0.1 – 0.3)	104	(23 - 262)	19	(4 - 46)	0	(0 - 0)
vacC_2km	260	(105 - 496)	11	(4 - 22)	0.1	(0.0 – 0.2)	212	(61 - 451)	37	(10 - 77)	0	(0 - 0)
vacC_3km	122	(51 - 223)	7	(3 - 13)	0.1	(0.0 – 0.1)	315	(100 - 624)	55	(17 - 107)	0	(0 - 0)

Discussion

The results of this study show that for CSF epidemics starting in a densely populated livestock area (DPLA) such as De Peel, the minimal measures required by the *EU* are not sufficient to control the epidemic. As additional measure, both preemptive culling and emergency vaccination decrease the duration and size of the epidemic. Preemptive culling shortens the epidemic the most with the smallest control radius of 1 km, at the expense of the largest number of depopulated farms. Similar effectiveness is only achieved by emergency vaccination when the control radius is 3 km. In this respect, C-strain vaccination is only slightly more effective than E2-subunit vaccination.

The non-vaccination strategies lead to the least number of not-detected infected herds and animals after final screening. E2-subunit vaccination is expected to yield considerable numbers (median around 20) of not-detected animals after final screening that are comparable to C-strain vaccination after final screening, but the former are distributed over less herds. For E2-subunit vaccination strategies an additional one or two truly positive herds are detected during the final screening, while some dozens of herds are identified falsely positive. The final screening results show that for the non-vaccination and C-strain vaccination strategies (almost) zero positive cases (true or false) are identified.

The effectiveness and final screening of E2-subunit vaccination have been assessed in a previous evaluation (Bergevoet et al., 2007, Backer et al., 2009). However, the results cannot be compared directly because several improvements have been made and input has changed: (a) a more recent commercial pig database of 2011 is used, (b) variable farm sizes and mixed farms are taken into account, (c) the HRP starts in a DPLA and generates a fixed number of 5 or 10 infected herds, and (d) the detection is based on a detection limit of infectious animals rather than on a randomly drawn detection time. Four points are worth noting though.

First, the 2011 pig farm database includes less farms than the previously used 2006 pig farm database. In the latter it was assumed that 1/3 of the farms contained a sow section, agreeing with a fraction of 35% now. The variable farm size in the current study does not have an effect on the detection times nor on the between-herd transmission in the model.

Second, in the previous study it was found that the effectiveness of 1 km preemptive culling was comparable to the effectiveness of 2 km vaccination, whereas in this study it

is found to be comparable to the effectiveness of 3 km vaccination. This is presumably caused by the condition that epidemics now start in a DPLA.

Third, a larger number of not-detected infected animals for non-vaccination strategies were predicted by the previous study. The new and more realistic method of detecting an infected farm at a certain number of infectious animals might be the cause of this. Regardless, the prediction of the previous study that the final screening brings the number of not-detected infected animals in vaccinated herds to the same level as not-vaccinated herds, cannot be upheld in this study.

Fourth, the final screening in the current study seems much less effective in detecting infected E2-subunit vaccinated herds. There are two reasons for this. First, the test sensitivity is now taken to be 73.4%, instead of 90% previously (based on the Bommeli (IDEXX) E_{rns} ELISA). This will affect the probablity of finding an infected herd in the first stage of the final screening. But the second reason has a larger effect: in the previous study a follow-up sensitivity of 100% was assumed. It was argued that more testing in herds that were positive in the E_{rns} ELISA would eventually eliminate all infected animals in that herd. In the current study, the confirmatory PCR testing is modelled in more detail, also taking the decreasing PCR sensitivity into account.

The poor performance of the final screening in E2-subunit vaccinated herds can be improved by more intensive testing in positive herds. When pen mates are additionally tested to search for evidence of virus spread (and – when positive – subsequently tested by PCR), a 100% follow-up specificitiy can be approached. However, this also means that all false positive herds need to be tested under this more intensive protocol and that the expected number of truly positive herds (cases) found during the final screening will increase. The performance of the E_{rns} screening could also be improved by starting the screening earlier during the epidemic, because it is more likely to find infected animals positive by PCR. The cases found in this way will not delay regaining the freedom-of-infection status. Of course this provisional screening will not cancel the obligation to test the herds again in the actual final screening, but it is less likely to find truly positive cases then.

Despite the faster immune response in animals that are C-strain vaccinated, the use of this vaccine is only slightly more effective in controlling the epidemic, compared to the E2-subunit vaccine. Apparently, the time scale of the E2-subunit vaccine induced immune response (approximately 10 days) is sufficiently small compared to the virus spread within a herd (4-5 weeks), to effectively halt the epidemic. Moreover, the C-strain vaccine does not prevent the occurrence of not-detected infected herds and animals. It is

impossible to detect these because they cannot be distinguished from vaccinated animals.

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Appendix A Effect of C-strain vaccine

To be able to assess control strategies with C-strain vaccination, the model needed to include the effect of the C-strain vaccine on the animal-to-animal transmission of CSFV. Experimental results were available to parameterize the model, but they were mostly limited to vaccination-challenge experiments in the presence of sentinel (unvaccinated) contact animals. For this reason, the C-strain vaccine model is simpler than the E2-subunit vaccine model that was developed previously (Bergevoet et al., 2007, Backer et al., 2009).

The C-strain vaccine model is based on 9 vaccination-challenge experiments, reported by Kaden et al. (2001) and Graham et al. (2012). These experiments were carried out with short time intervals between vaccination: at 0 days post vaccination (dpv), 1 dpv, 3 dpv and 5 dpv, with various challenge virus strains. At longer time intervals all vaccinated animals were fully protected at the time of challenge (Dewulf et al., 2004).

We will use the fraction of successful challenges as a measure of susceptibility (bearing in mind that the animals were inoculated, i.e. not naturally infected), and fitted an exponential decay function to the results (Fig. A1). The infectiousness of the vaccinated infected animals was assumed to be unaffected by the vaccine, because in each experiment (Kaden et al., 2001, Graham et al., 2012) with an infected vaccinated animal, all sentinel animals were contact infected. Furthermore, the period during which the vaccinated infected animals were PCR positive, was not shorter than for the unvaccinated control animals. For this reason, also the average infectious period of vaccinated infected animals was assumed to be unaffected by the vaccine.

The effect of the C-strain vaccine on the animal-to-animal transmission parameters is compared to the effect of the E2-subunit vaccine in Fig. A2. Note that parameters such as infectiousness at large vaccination-infection intervals bear no meaning for the C-strain vaccine, as vaccinated animals are already fully protected against infection at that time. This is also clear when comparing the effective reproduction number (in a fully vaccinated population) for the two vaccines (Fig. A2.d)

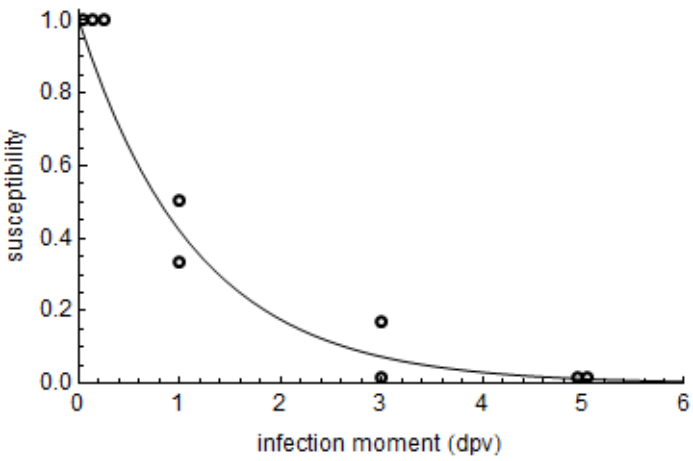

Figure A1 Effect of C-strain vaccine on susceptibility as function of the infection moment; the model (solid line) is based on experimental results (open circles) of Kaden et al., 2001 (infection moments at 0 dpv), and Graham et al., 2012 (infection moments >0 dpv); dpv: days post vaccination

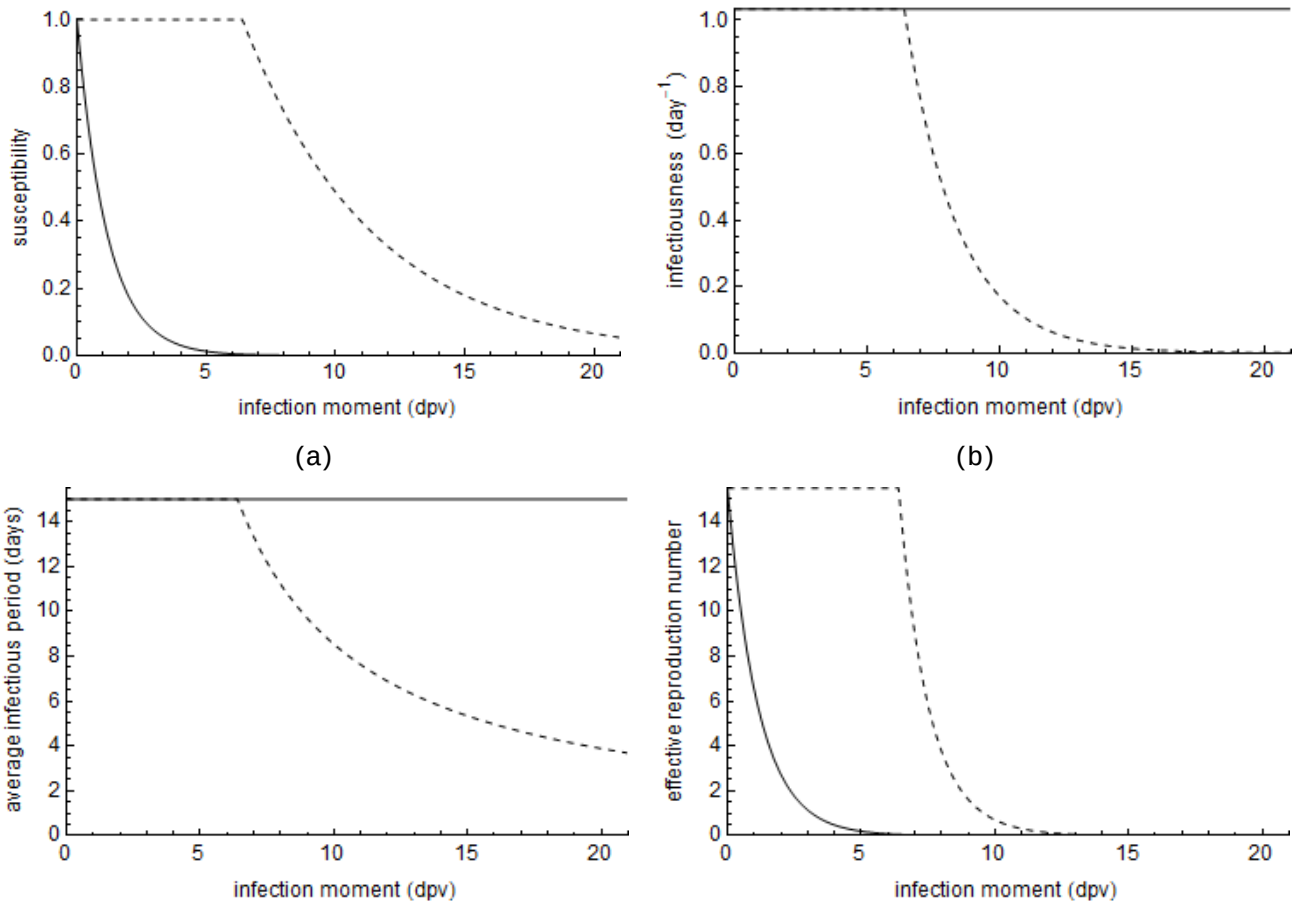

Figure A2 Effect of C-strain vaccine (solid line) and E2-subunit vaccine (dashed line) on (a) susceptibility, (b) infectiousness, (c) average infectious period and (d) effective reproduction number, as function of the infection moment (in dpv: days post vaccination)

Appendix B Test characteristics for final screening model

Table B1 Test characteristics at animal level for commercial CSF serology tests, determined at Central Veterinary Institute

test	characteristics		remarks
	sens	spec	
Prionics E2 ELISA	92%	98%	
IDEXX E2 ELISA	93%	99%	used by CVI since 2006
E2 VNT	94%	99.5%	specificity after correction for crossreaction with BD and BVD
Bommeli (IDEXX) E_{rns} ELISA	89.1%	99.6%	for finishing pigs
Prionics E_{rns} ELISA	73.4%	99.9%	for finishing pigs

E2-serology is used for unvaccinated animals. Samples are first tested with the Prionics E2 ELISA. When positive, they are tested in the IDEXX E2 ELISA. These tests combined have a sensitivity of 92% and a specificity of 99.98%. When samples are positive in both ELISA's, they are tested in the E2 VNT (virus neutralisation test). For the standard CSF surveillance programme, around 50000 samples are tested yearly in this three-step procedure, yielding 0 positives. This gives a lower bound of the overall specificity of 1-0.5/50000=0.99999 for an individual sample. The overall sensitivity is determined by the first ELISA that has the lowest sensitivity of 92%. Each of the ELISA's takes one day, and the VNT takes another 5 days, yielding a total period of 7 days for the whole procedure to finish.

E_{rns}-serology is used for E2-subunit vaccinated animals. The test characteristics of the E_{rns} ELISA's have been determined in evaluation at the CVI. A panel of 226 E2-subunit vaccinated and inoculated (more than 28 dpv) animals served to determine the test sensitivities and a panel of 1847 field sera of finishing pigs served to determine the test specificities. Schroeder et al. (2012) report the results of a ring trial on a smaller panel (84 samples of E2-subunit vaccinated and challenged animals), in which the Bommeli (IDEXX) test correctly identified 85% and the Prionics test 56% of the samples. These percentages are not identical to the sensitivities (as the same samples are tested multiple times and test results are thus not independent), but they do show the same relation between the two tests as found in the in-house evaluation. As the Bommeli (IDEXX) test has been taken out of production, the individual test characteristics in the final screening model are taken to be the test characteristics of the Prionics E_{rns} ELISA, i.e. 73.4%. Claims by the manufacturer that the sensitivity of this test has been

improved for E2-subunit vaccinated animals have not been validated yet in an appropriate panel.

When the E_{rns} ELISA is positive, tonsil samples are taken from the positively tested animals and tested by PCR, to determine whether the herd is declared positive or negative. The sensitivity of this PCR test decreases with time since infection, due to clearance of the virus. This means that even when a herd is tested truly positive in the ELISA, the PCR results can be negative, leading to a negatively declared herd.

To determine how the ability of the PCR to detect infected animals depends on the time since infection, experimental data are used (Loeffen, 2008). In total 38 Paderborn-infected animals (unvaccinated and E2-subunit vaccinated) that survived the acute phase were euthanized at various times since infection, and their tonsils PCR-tested. A logistic function was fitted through the results as $se^{PCR}(\tau) = 1/(1+\exp(0.088\,\tau - 7.4))$ (Fig. B1).

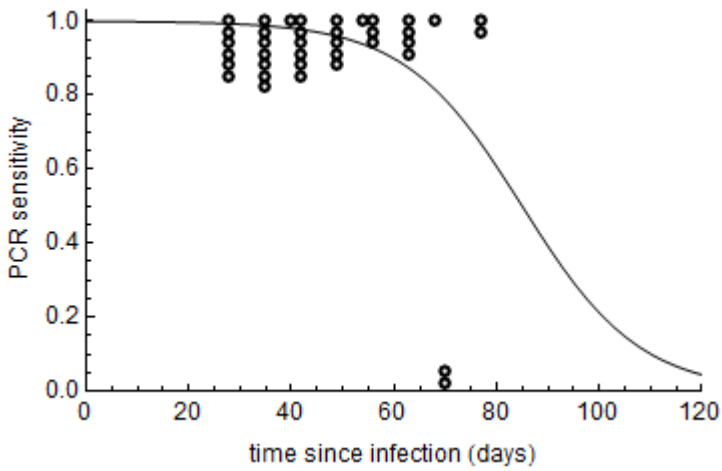

Figure B1 Sensitivity of PCR to detect infected animals as function of the time since infection; experimental data from Loeffen, 2008 (symbols near 1: positive PCR, symbols near 0: negative PCR), and fitted PCR sensitivity (solid line).

Appendix C Calculation of pig numbers per farm from pig transport data

Dienst Regelingen (Ministry of Economic Affairs) provided pig transport data containing the number of pigs that were dispatched from each UBN (unique farm identifier) in 2011, in the categories:

B: piglets

V: finishers

G1: sows and gilts

G2: gilts of 7 months of age

D: newly born piglets (ignored; these are dead piglets dispatched to Rendac)

O: other finishers (ignored; these are presumably sows that are to be replaced)

It is assumed that:

- suckling piglets are maximally 4 weeks of age,
- weaned piglets are maximally 9 weeks of age,
- finishers are maximally 26 weeks of age and
- gilts are maximally 35 weeks of age.

Finishers will be in the the finisher category for 26 – 9 = 17 weeks and gilts will be in the the gilt category for 35 – 9 = 26 weeks. In calculating the number of animals from the pig transport data, no distinction is made between breeding and multiplier farms nor between finishing and growing farms. They differ in the type farms they are allowed to dispatch their animals to, but this does not affect the population calculation. For each farm type (simplifying) assumptions are made to calculate the number of sows, the number of finishers and gilts, and the number of piglets that are present in a farm at a specific time (Tab. C1).

Table C1 Calculation of number of sows, finishers and piglets from pig transport data

Farm type	# sows	# finishers + # gilts	# piglets
Breeding farm (Fokbedrijf) - Sows have 27 piglets per year - No import of gilts or piglets	(B+V+G1+G2) /27	V 17/52 + (G1+G2) 26/52	(B+V+G1+G2) 9/52
Finishing farm (Mestbedrijf) - Piglets enter at 9 weeks of age - No sows or piglets present	0	V 17/52 + (G1+G2) 26/52	0
Growing farm (Opfokbedrijf) - Piglets enter at 9 weeks of age - No sows or piglets present	0	V 17/52 + (G1+G2) 26/52	0
Weaned piglets farms (E/F bedrijven) - Piglets enter at 4 weeks of age - Dispatched at 9 or 26 weeks of age	0	V 17/52 + (G1+G2) 26/52	(B+V+G1+G2) 5/52
Multiplier farm (Vermeerderaar) - Sows have 27 piglets per year - No import of gilts or piglets	(B+V+G1+G2) /27	V 17/52 + (G1+G2) 26/52	(B+V+G1+G2) 9/52

Appendix D Required depopulation and vaccination capacities

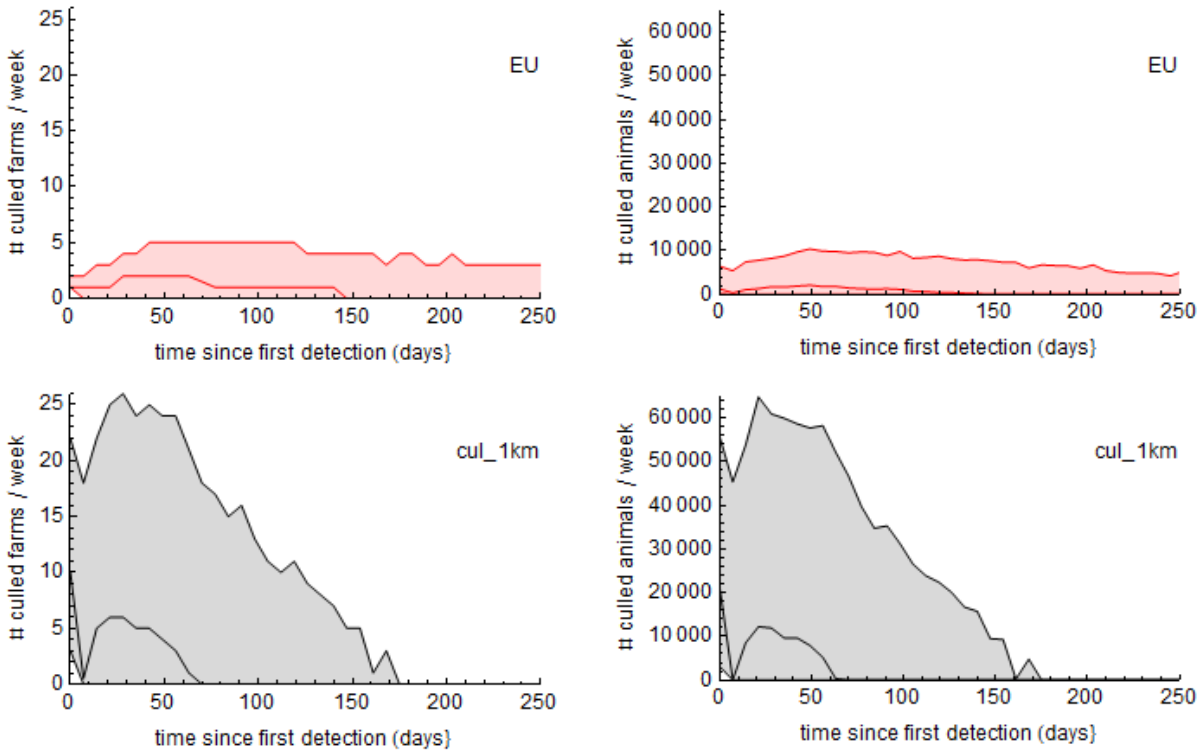

Figure D1 Required depopulation capacity for EU measures and 1 km preemptive culling, in number of farms and animals per week: median value (solid line) and 5%-95% interval (shaded area).

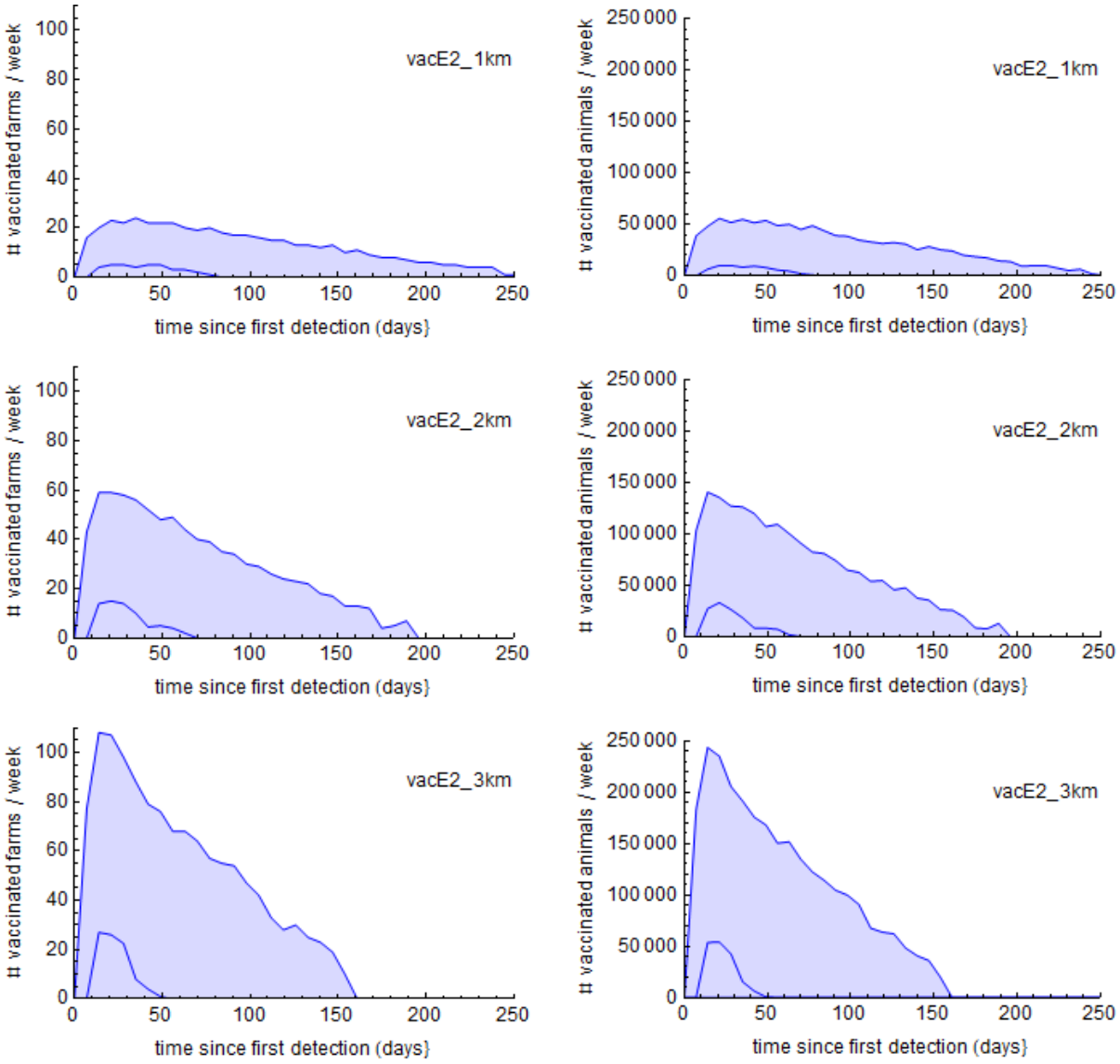

Figure D2 Required vaccination capacity for emergency vaccination with E2-subunit vaccine in 1 km, 2 km or 3 km around detected herds, in number of farms and animals per week: median value (solid line) and 5%-95% interval (shaded area).

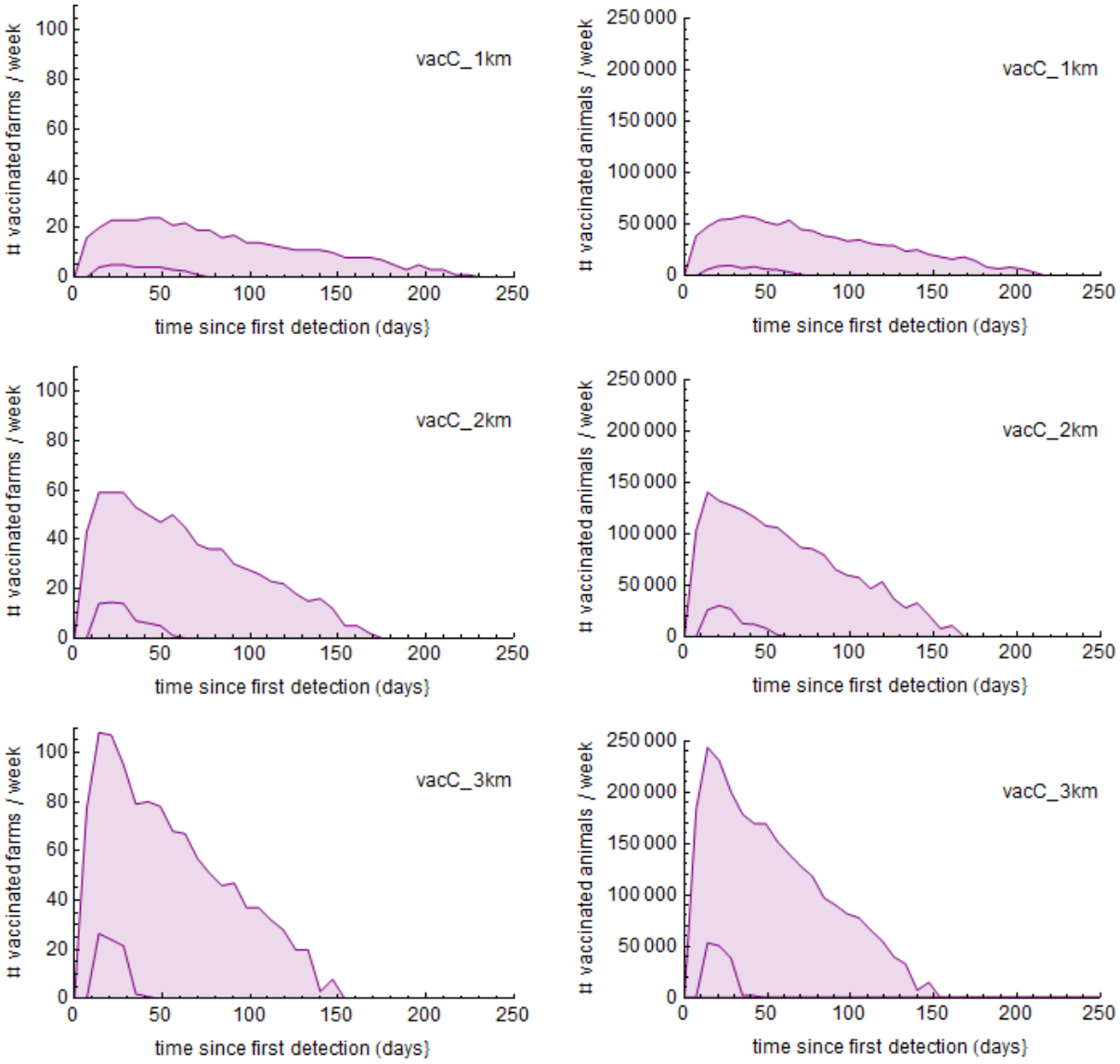

Figure D3 Required vaccination capacity for emergency vaccination with C-strain vaccine in 1 km, 2 km or 3 km around detected herds, in number of farms and animals per week: median value (solid line) and 5%-95% interval (shaded area).

Appendix E Required vaccination capacities – table

Table E1 Required number of farms and animals to be vaccinated using E2-subunit vaccination in 2 km (vacE2_2km) or 3 km (vacE2_3km) per week (week 1 is the first week of vaccination): median values and (5% - 95%) interval

week	vacE2_2km		vacE2_3km	
	number of farms	number of animals (x1000)	number of farms	number of animals (x1000)
0	0 (0 - 0)	0 (0 - 0)	0 (0 - 0)	0 (0 - 0)
1	0 (0 - 43)	0 (0 - 103)	0 (0 - 77)	0 (0 - 183)
2	14 (0 - 59)	27 (0 - 141)	27 (0 - 108)	54 (0 - 244)
3	15 (0 - 59)	33 (0 - 136)	26 (0 - 107)	54 (0 - 235)
4	14 (0 - 58)	27 (0 - 127)	22 (0 - 98)	43 (0 - 206)
5	10 (0 - 56)	18 (0 - 126)	8 (0 - 88)	16 (0 - 191)
6	4 (0 - 52)	8 (0 - 120)	4 (0 - 79)	6 (0 - 176)
7	5 (0 - 48)	8 (0 - 107)	1 (0 - 76)	1 (0 - 168)
8	4 (0 - 49)	7 (0 - 109)	0 (0 - 68)	0 (0 - 150)
9	2 (0 - 44)	2 (0 - 100)	0 (0 - 68)	0 (0 - 152)
10	0 (0 - 40)	0 (0 - 91)	0 (0 - 64)	0 (0 - 135)
11	0 (0 - 39)	0 (0 - 82)	0 (0 - 57)	0 (0 - 122)
12	0 (0 - 35)	0 (0 - 81)	0 (0 - 55)	0 (0 - 114)
13	0 (0 - 34)	0 (0 - 74)	0 (0 - 54)	0 (0 - 105)
14	0 (0 - 30)	0 (0 - 65)	0 (0 - 47)	0 (0 - 100)
15	0 (0 - 29)	0 (0 - 63)	0 (0 - 42)	0 (0 - 91)
16	0 (0 - 26)	0 (0 - 54)	0 (0 - 33)	0 (0 - 68)
17	0 (0 - 24)	0 (0 - 55)	0 (0 - 28)	0 (0 - 64)
18	0 (0 - 23)	0 (0 - 46)	0 (0 - 30)	0 (0 - 62)
19	0 (0 - 22)	0 (0 - 48)	0 (0 - 25)	0 (0 - 48)
20	0 (0 - 18)	0 (0 - 38)	0 (0 - 23)	0 (0 - 41)
21	0 (0 - 17)	0 (0 - 36)	0 (0 - 19)	0 (0 - 36)
22	0 (0 - 13)	0 (0 - 26)	0 (0 - 10)	0 (0 - 20)
23	0 (0 - 13)	0 (0 - 26)	0 (0 - 0)	0 (0 - 0)
24	0 (0 - 12)	0 (0 - 19)	0 (0 - 0)	0 (0 - 0)
25	0 (0 - 4)	0 (0 - 9)	0 (0 - 0)	0 (0 - 0)
26	0 (0 - 5)	0 (0 - 8)	0 (0 - 0)	0 (0 - 0)
27	0 (0 - 7)	0 (0 - 13)	0 (0 - 0)	0 (0 - 0)
28	0 (0 - 0)	0 (0 - 0)	0 (0 - 0)	0 (0 - 0)

Two remarks:

- For the first week of vaccination (week 1) the model predicts not too many farms to be vaccinated (median value of 0); however in practice detections will occur in this early stage of the epidemic due to the enhanced screening and tracing. This is not explicitly included in the model.
- Culling and vaccination capacities are not taken into account in the model; this will play a larger role for 3 km vaccination than for 2 km vaccination. For this reason the difference in epidemic duration and size for 2 and 3 km vaccination will in practice be smaller than predicted by the model. As a consequence, the difference between the required vaccination capacity between the two strategies will be larger